IN THE UNITED STATES DISTRICT COURT
FOR THE SOUTHERN DISTRICT OF WEST VIRGINIA
HUNTINGTON DIVISION

CHRISTOPHER FAIN; ZACHARY MARTELL; BRIAN MCNEMAR, SHAWN ANDERSON a/k/a SHAUNTAE ANDERSON and LEANN JAMES, individually and on behalf of all others similarly situated,

Plaintiffs,

vs.

WILLIAM CROUCH, in his official capacity as Cabinet Secretary of the West Virginia Department of Health and Human Resources; CYNTHIA BEANE, in her official capacity as Commissioner for the West Virginia Bureau for Medical Services; WEST VIRGINIA DEPARTMENT OF HEALTH AND HUMAN RESOURCES, BUREAU FOR MEDICAL SERVICES; JASON HAUGHT, in his official Capacity as Director of the West Virginia Public Employees Insurance Agency; and THE HEALTH PLAN OF WEST VIRGINIA, INC.,

Defendants.

Civil Action No.

3:20-cv-00740

REMOTE VIDEOTAPED DEPOSITION OF

JOHANNA OLSON-KENNEDY, M.D.

April 25, 2022

Reported By: Amy E. Simmons, CSR, RPR, CRR, CRC

REMOTE VIDEOTAPED DEPOSITION OF
JOHANNA OLSON-KENNEDY, M.D.

BE IT REMEMBERED that the remote videotaped deposition of JOHANNA OLSON-KENNEDY, M.D., was taken via videoconference by the Defendants before Veritext Legal Solutions, Amy E. Simmons, Court Reporter and Notary Public in and for the County of Ada, State of Idaho, on Monday, the 25th day of April, 2022, commencing at the hour of 8:39 a.m. Pacific Daylight Time in the above-entitled matter.

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Videographer: Jonathan Hernandez

Also Present: Michele Clanton-Lockhart

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THE VIDEOGRAPHER: Good morning. We are on the record. The time is 8:39 a.m., Pacific Standard Time. Today is April 25th, 2022.

My name is Jonathan Hernandez. I am a video technician with Veritext Legal Solutions located in Los Angeles, California. We are recording these proceedings over videoconference technology due to COVID-19.

This is Media 1 for the video deposition of Dr. Johanna Olson-Kennedy in the action entitled Christopher Fain, et al., vs. William Crouch, et al.

This deposition is being taken on behalf of the Defendants. The case number is 3:20-cv-00740.

Now would you all please identify yourself and who you represent, starting with the noticing attorney.

MR. DAVID: Caleb David on behalf of the Defendants.

MS. BORELLI: This is Tara Borelli with Lambda Legal on behalf of the Plaintiffs.

MS. SMITH-CARRINGTON: This is Avatara Smith-Carrington with Lambda Legal on behalf of

Plaintiffs.
THE WITNESS: My name is Johanna Olson-Kennedy. I am the expert witness here on behalf of the Plaintiffs.
THE VIDEOGRAPHER: Thank you. Now will the court reporter please administer the oath.

OHANNA OLSON-KENNEDY, M.D.,

a witness having been first duly sworn remotely to tell the truth, the whole truth and nothing but the truth, was examined and testified as follows:

MS. BORELLI: And before Mr. David begins his questions, we'd like to put a stipulation on the record.
The stipulation is that for purposes of this deposition, an objection to form will preserve all objections to form without needing to specifically state them.
Mr. David, is that agreeable to you?
MR. DAVID: That is agreeable to me.
MS. BORELLI: Thank you.
MR. DAVID: And, Tara, before I get going, do we want to note Walt's appearance? I just wanted to make sure.
MS. BORELLI: Sure. Walt, do you want to state your appearance?
MR. AUVIL: Walt Auvil for the Plaintiffs.

EXAMINATION

BY MR. DAVID:
Q. All right. Doctor, can you please state your name for the record.
A. My name is Johanna Olson-Kennedy.
Q. And, Doctor, we've met before the deposition began, but my name is Caleb David, and I represent the defendants in this lawsuit. We're here to take your deposition today.
And I know from your expert report that you have some experience with depositions.
So first, how many depositions have you given in the past?
MS. BORELLI: Objection; form.
THE WITNESS: I believe I've given four, more or less. I'd have to go back and look to know the exact number.
Q. (BY MR. DAVID) So you're familiar with the procedure and how things will work today, right?
A. I am.
MS. BORELLI: Objection; form.
Q. (BY MR. DAVID) All right. Doctor, how are you currently employed?
A. Currently, I am employed by USC Keck

School of Medicine.
Q. And what is your position there?
A. I am an associate professor of clinical pediatrics, which is my official title.
My acting role is as an attending physician at Children's Hospital Los Angeles.
Q. As an associate professor of clinical pediatrics, are you teaching in a classroom at all?
A. No.
Q. So are you, then, providing education to residents at the hospital?
A. I do provide education for trainees, including fellows, residents, medical students. And I do occasionally provide lectures for the medical school.
Q. When you have residents rotating through the hospital and they're under your supervision, what types of residencies or what specialties are they residents in?
A. The primary specialty is pediatrics. However, there are occasionally residents from other specialties that rotate through Children's Hospital.
Q. Are you aware of any current residencies

in gender medicine?
MS. BORELLI: Objection; form.
THE WITNESS: I don't think so. Not that I know of.
Q. (BY MR. DAVID) Let me ask you this: Do some of your residents have an interest in gender medicine?
MS. BORELLI: Objection; form.
THE WITNESS: Probably important to delineate that I do inpatient consultative services for adolescents, mostly not related to gender.
So the interaction that I have with the residents, the pediatric residents over at the hospital, are concerned with other aspects of adolescent care.
Q. (BY MR. DAVID) And I made some bad assumptions there. Let me ask you more about your practice.
So in your practice at the children's hospital, what conditions or what types of patients are you generally treating?
MS. BORELLI: Objection; form.
THE WITNESS: So in the center for Transyouth Health and Development, I am primarily

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meeting and taking care of folks who are coming in with issues related to gender.

However, I also cross-cover, and then in the hospital cover adolescents broadly, so all issues that impact adolescents.

Q. (BY MR. DAVID) So do you do any well visits for adolescents, or is it only at the hospital?

MS. BORELLI: Objection; form.

THE WITNESS: There are sites that occasionally I'm cross-covering where I'm doing well visits for adolescents.

Q. (BY MR. DAVID) Do you also provide prescription monitoring for things such as antidepressants and antianxiety medications?

MS. BORELLI: Objection; form.

THE WITNESS: What do you mean by prescription management?

Q. (BY MR. DAVID) Sure. Well, let me -- do you prescribe antidepressants and antianxiety medications?

MS. BORELLI: Objection; form.

THE WITNESS: I do occasionally prescribe those medications.

Q. (BY MR. DAVID) Do you prescribe insulin?

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A. I do not.

Q. Do you prescribe any medications for adolescents who are experiencing cardiac issues?

MS. BORELLI: Objection; form.

THE WITNESS: You'd have to be more specific about which cardiac issues that you mean.

Q. (BY MR. DAVID) What about hypertension?

MS. BORELLI: Objection; form.

THE WITNESS: I have prescribed medication for hypertension in the past.

Q. (BY MR. DAVID) What is the age range of your patient population?

MS. BORELLI: Objection; form.

THE WITNESS: In the work that I do over at the hospital, the majority of people are between the ages of 10, maybe, and 21 in the practice that I do in the ambulatory setting. In the Center for Transyouth Health and Development I have seen patients as young as 3 and up to 26.

Q. (BY MR. DAVID) And your practice in the hospital is generally focused on internal medicine conditions of individuals between the ranges -- age range of 10 to 21; is that correct?

MS. BORELLI: Objection; form.

THE WITNESS: Well, the practice that I

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am subspecialized in is called adolescent medicine, so it's not called internal medicine.

Q. (BY MR. DAVID) Is adolescent medicine a subspecialty of internal medicine?

MS. BORELLI: Objection; form.

THE WITNESS: No.

Q. (BY MR. DAVID) Is it a subspecialty of pediatrics?

MS. BORELLI: Objection; form.

THE WITNESS: Adolescent medicine is a subspecialty of pediatrics.

Q. (BY MR. DAVID) You don't perform surgery, correct?

MS. BORELLI: Objection; form.

THE WITNESS: I do not perform surgery.

Q. (BY MR. DAVID) Do you -- well, let me ask you this first: What is your role at the Center for Transyouth Health and Development at Children's Hospital Los Angeles?

A. I am the medical director of the program.

Q. And what are your general duties as the medical director of the program?

A. So as the medical director of the program, in addition to providing direct services for patients and providing referrals for patients

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and family members, I also oversee the activities of the center, coordinate between the different disciplines, coordinate aspects around assessing the necessary personnel. I sit in on the interviews and hiring of personnel. Those are some of the things.

Q. I think the first role that you described was directing services.

Can you explain what that means?

MS. BORELLI: Objection; form.

THE WITNESS: So we have a multidisciplinary team at the center, and so part of -- I need to go back and ask you -- can someone repeat my sentence about directing services? Because it feels out of context for me, so I'm not sure.

THE REPORTER: Did you want me to read that back, Counsel?

MR. DAVID: Yes, please.

(The following was read by the reporter:

"Question: So as the medical director of the program, in addition to providing direct services for patients and providing referrals for patients and family members, I also oversee the activities of the center, coordinate between the

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different disciplines, coordinate aspects around assessing the necessary personnel. I sit in on the interviews and hiring of personnel. Those are some of the things.")

Q. (BY MR. DAVID) So I wrote it down wrong. You said "providing direct services."

So can you explain what that means, please?

A. Yes. So I provide a range of services for families and patients that are coming in to see me. And those services range from explaining or giving information, educating people about the process of gender development, what we know about the experiences of young people.

Sometimes families come in who really are looking for other parents or parent support groups, how they can support their children; and sometimes people are coming in asking or seeking for medical interventions, including puberty blockers or gender-affirming hormones. Those are a few of the things that are part of my direct service provision.

Q. When you were providing education to patients and their families, do you provide them with any literature?

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MS. BORELLI: Objection; form.

THE WITNESS: Occasionally.

Q. (BY MR. DAVID) Are there specific pieces of literature that you normally will provide?

MS. BORELLI: Objection; form.

THE WITNESS: I don't know that I have something that I provide specifically each and every time, because the variety of things that people need necessitate different information.

Q. (BY MR. DAVID) Can you give me an example of one of the pieces of literature that you provide to patients or families?

MS. BORELLI: Objection; form.

THE WITNESS: Sure. So an example might be something that our center has created around safe binding practices that gives people information on how to safely bind in such a way that doesn't cause harm to their body.

Q. (BY MR. DAVID) Do you provide any literature to patients or families that is not created in-house?

MS. BORELLI: Objection; form.

THE WITNESS: Sometimes.

Q. (BY MR. DAVID) For instance, have you provided WPATH Standards of Care 7 to patients or their families?

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A. No.

Q. Have you provided the Endocrine Society Guidelines to patients or their families?

MS. BORELLI: Objection; form.

THE WITNESS: No.

Q. (BY MR. DAVID) So the literature that you're providing is -- sounds to me, at least, that it's more specific to the patient care aspects of what they're coming to see you for; is that right?

MS. BORELLI: Objection; form.

THE WITNESS: So some of the things that I'm providing have to do with that. Occasionally people will ask for information on other things or what's known in the field, what isn't known in the field, and I will provide them things.

It's hard to think of such things offhand, but the majority of what we're providing for people is pragmatic or practical things that they can use within their own home.

Q. (BY MR. DAVID) Okay. And I think that you mentioned that you see patients for medical intervention such as puberty blockers or gender-affirming hormones; is that correct?

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MS. BORELLI: Objection; form.

THE WITNESS: That is correct.

Q. (BY MR. DAVID) Are there any other medical interventions for gender dysphoria that you provide to patients?

MS. BORELLI: Objection; form.

THE WITNESS: Such as?

Q. (BY MR. DAVID) Anything.

MS. BORELLI: Same objection.

THE WITNESS: Gender dysphoria is complex, and not everybody needs the same thing. And therefore, there are times when people need -- for example, exactly what I was just talking about.

If somebody has gender dysphoria about their chest or about their genitals, we will provide some information on how to safely bind or how to safely tuck. Those are just a few examples of some of the things.

Q. (BY MR. DAVID) And I understand that you yourself don't perform surgery. We'll talk about surgery later on.

I'm just trying to find if there are any other medical therapies that you recommend or that you provide to patients for gender dysphoria.

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MS. BORELLI: Objection; form.
THE WITNESS: I will sometimes refer people for surgery, if that's what you mean. I may make recommendations for someone to see a surgeon in consultation regarding the specific surgery.
There are occasionally people who are seeking therapy or additional therapy services, so I'll make recommendations about that.
Q. (BY MR. DAVID) When you say people are seeking additional therapy recommendations, are you talking about psychiatric therapy?
MS. BORELLI: Objection; form.
THE WITNESS: I don't know what "psychiatric therapy" is.
There's the field of psychiatry where people engage in services related to medications. And some psychiatrists do talk therapy or cognitive behavioral therapy. But for the most part, cognitive behavioral therapy or dialectical behavorial therapy really are in the realm of psychologists or other types of mental health therapists. That's the way that the distinction is made in our program.
Q. (BY MR. DAVID) And I apologize. I'm

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going to unfortunately use some inexact terms sometimes, and I apologize. So thank you for correcting me.
So when you were mentioning therapy, were you talking about psychotherapy?
MS. BORELLI: Objection; form.
THE WITNESS: Sometimes people are -- the best place for them to be triaged to is to a psychiatrist. And sometimes the best place for people to be triaged to is a therapist.
Q. (BY MR. DAVID) So -- and setting aside surgery, but just services that you provide specifically for gender dysphoria is limited to puberty blockers and to gender-affirming hormones; is that correct?
MS. BORELLI: Objection; form.
THE WITNESS: Those are the medical interventions that I provide myself, that I pick up the pen and write prescriptions for, although nobody writes prescriptions with a pen. We mostly electronically send them in.
But yes, in addition to, I do a lot of education. Probably that takes up the majority of the visit time is educating people.
Q. (BY MR. DAVID) Okay. And again, we'll

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talk about -- well, I'll just mention it in this question.
So we've talked about puberty blockers, gender-affirming hormones, some psychological therapy and surgeries.
Are there any other treatment modalities that you're aware of for gender dysphorias?
MS. BORELLI: Objection; form.
THE WITNESS: So I should add in there another thing that I commonly do is prescribe oral contraceptive pills for the purpose of diminishing or induction of amenorrhea so somebody doesn't have a menstrual cycle. That's another one of the potential interventions for somebody with gender dysphoria.
Q. (BY MR. DAVID) Okay. With the addition of prescribing oral contraceptives, there's puberty blockers, gender-affirming hormones, surgery, and psychological therapy.
Is that the universe of treatment modalities for gender dysphoria?
MS. BORELLI: Objection; form.
THE WITNESS: Yes.
Q. (BY MR. DAVID) Okay. Do you yourself diagnose patients with gender dysphoria?

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MS. BORELLI: Objection; form.
THE WITNESS: I do.
Q. (BY MR. DAVID) Okay. At the Center for Transyouth Health and Development at Children's Hospital of Los Angeles, is your practice limited to gender medicine?
MS. BORELLI: Objection; form.
THE WITNESS: I'm not sure I understand. Within the center itself?
Q. (BY MR. DAVID) Yes. Within the center itself.
MS. BORELLI: Same objection.
THE WITNESS: So just -- I just want to be clear about what you mean by "gender medicine." People who have questions around their gender or are seeking interventions are the people that we see within the housing of the center.
Q. (BY MR. DAVID) And I think that that answers it.
Simply, do you see patients for well visits at the center?
MS. BORELLI: Objection; form.
THE WITNESS: No, we don't.
Q. (BY MR. DAVID) Okay. Have you ever participated in the drafting of health insurance

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guidelines for coverage?
A. No.
MS. BORELLI: Objection; form.
Q. (BY MR. DAVID) Have you yourself ever performed research relating to what health insurers must cover?
MS. BORELLI: Objection; form.
THE WITNESS: No.
Q. (BY MR. DAVID) In this case, have you reviewed any medical records of Christopher Fain?
MS. BORELLI: Objection; form.
THE WITNESS: No.
Q. (BY MR. DAVID) Have you reviewed any medical records of Shauntae Anderson?
MS. BORELLI: Objection; form.
THE WITNESS: No.
Q. (BY MR. DAVID) Have you spoken to either Christopher Fain or Shauntae Anderson?
A. No.
Q. So you are not going to be offering any opinions specific to Christopher Fain or Shauntae Anderson?
MS. BORELLI: Objection; form.
THE WITNESS: That's correct.
Q. (BY MR. DAVID) When you are diagnosing

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gender dysphoria in a patient, do you use the DSM-5 criteria for diagnosis?
MS. BORELLI: Objection; form.
THE WITNESS: Yes.
Q. (BY MR. DAVID) Are there any other criteria that you rely upon in making the diagnosis outside of the DSM-5?
MS. BORELLI: Objection; form.
THE WITNESS: I think that making the diagnosis in addition to what's in the DSM, I lean on my clinical experience, 16 years of doing this work, to facilitate that diagnosis.
Q. (BY MR. DAVID) Are there other diagnostic criteria other than what's listed in the DSM-5?
MS. BORELLI: Objection; form.
THE WITNESS: No. Those are the diagnostic criteria.
Q. (BY MR. DAVID) And I believe you just said that you have 16 years of clinical experience; is that correct?
A. That's correct.
Q. Over your 16 years of clinical experience, have you seen a shift in the patient population from primarily individuals who were

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assigned male at birth to now individuals who were assigned female at birth?
MS. BORELLI: Objection; form.
THE WITNESS: We have seen a shift in that ratio.
Q. (BY MR. DAVID) Do you have an explanation for why that shift is occurring?
MS. BORELLI: Objection; form.
THE WITNESS: I have thoughts about it, yes.
Q. (BY MR. DAVID) Okay. And I'll ask you your thoughts in a second.
Are you aware of any literature that has looked into that specific shift and determined why that shift has occurred?
MS. BORELLI: Objection; form.
THE WITNESS: No.
Q. (BY MR. DAVID) And now can you tell me your thoughts on why that shift has occurred?
MS. BORELLI: Objection; form.
THE WITNESS: I think that there are many things that have to be considered. The first is, you know, I work in a youth clinic. So I see people that primarily are accessing services at around the age of 16. And their experiences are

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of gender dysphoria -- their gender dysphoria is emerging around the time that they start puberty.
I think that it is critical to understand that the development of chest tissue is the first beginnings of puberty for people designated female at birth.
Because of that, it is likely that that change of puberty is the thing that is either exacerbating or creating the experience of gender dysphoria for them in a way that they can verbalize and talk about.
Q. (BY MR. DAVID) Okay. So would the rise in individuals who were assigned female at birth coming out as transgender in recent years compared to previously be more or less that that population has always existed, but now it's more acceptable from society's point of view to come out as transgender?
MS. BORELLI: Objection; form.
THE WITNESS: Yes, I believe so.
Q. (BY MR. DAVID) Okay. And so as a result of that, individuals who were assigned male at birth were, I guess, overrepresented in the ratio because those who were assigned female at birth were not comfortable coming out as transgender

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previously; is that right?
MS. BORELLI: Objection; form.
THE WITNESS: It's impossible for me to speak on behalf of the entire community. But I will say that there are things in our society that land harder on transfeminine people, on people designated male at birth, that are probably playing a role in the way that we see people and the timeline that we see people talking about their gender.
Q. (BY MR. DAVID) What sort of things are you referring to that land harder on transfeminine youth?
MS. BORELLI: Objection; form.
THE WITNESS: There are -- so, for example, going through a puberty as someone designated male at birth results in changes in one's body, some of which are impossible to walk back; some of which are impossible to change without surgical intervention.
And we live in a society that perceives somebody who -- if somebody has gone through a testosterone-dominant therapy, that person is likely to be perceived as a male who is dressing as a woman or acting like a woman. And that's a

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different -- there's a different response to somebody -- there always has been; this is historical as well -- than to somebody who may be perceived to be a little bit shorter or a little bit more effeminate as a man. There is a different societal response to that.
And because of that, it's just simply harder to be a trans woman who's gone through their first puberty.
Q. (BY MR. DAVID) I've probably gotten ahead of myself, and I tend to do that.
So I want to refer to your report.
Do you have that in front of you?
A. I do.
Q. Okay. And I'll refer to pages and to paragraph numbers so that we can all be on the same page and I don't have to keep flipping it on the screen, if that's okay with everyone.
MS. BORELLI: Yes. And, Caleb, can you also mark that as an exhibit so that it is in Exhibit Share for the participants to review as well?
MR. DAVID: Absolutely.
MS. BORELLI: Thank you.
MR. DAVID: And I will mark it as

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Exhibit 1. Let me make a note of it. I'll start my list.
(Deposition Exhibit No. 1 was marked.)
MS. BORELLI: And just to confirm, you plan to publish it to the "Marked Exhibits" folder in Exhibit Share, correct?
MR. DAVID: Yes, I do.
MS. BORELLI: Thank you.
Q. (BY MR. DAVID) Doctor, I want to move to page 5 of your report, and paragraph 18. And I'll go ahead and read the first couple sentences and then I'll have some questions for you.
MS. BORELLI: Actually, Caleb, sorry. But before we do questioning about the report, can you publish it to Exhibit Share --
MR. DAVID: Oh, sure. I'm sorry.
MS. BORELLI: -- so we can review it with you? Thank you.
MR. DAVID: I am sorry. I don't know if I'm -- let me see if I can't figure this out real quick.
I think it's there now.
MS. BORELLI: It is. Thank you.
MR. DAVID: Awesome. Thank you. I'm sorry. This is my first time using this system,

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so I'm learning on the fly.
MS. BORELLI: Understood.
Q. (BY MR. DAVID) Okay. Doctor, I'm looking at paragraph 18.
Are you with me?
A. Yes.
Q. Okay. "Gender identity, often simply termed 'gender,' is a distinct characteristic and is defined as one's internal sense of being male, female, both, neither, or some other gender identity."
So first, did I read that correctly?
A. Yes.
Q. Okay. And so to be clear, you're stating here that someone's gender identity can be female, correct?
MS. BORELLI: Objection; form.
THE WITNESS: Yes.
Q. (BY MR. DAVID) It can be male, correct?
MS. BORELLI: Objection; form.
Q. (BY MR. DAVID) I'm sorry. I didn't hear you.
A. Yes.
Q. Thank you. Can be both male and female, correct?

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MS. BORELLI: Objection; form.
THE WITNESS: Yes. Sorry.
Q. (BY MR. DAVID) Can be neither male nor female, correct?
MS. BORELLI: Objection; form.
THE WITNESS: Yes.
Q. (BY MR. DAVID) And can be some other gender identity, correct?
MS. BORELLI: Objection; form.
THE WITNESS: Yes.
Q. (BY MR. DAVID) Are there terms for some other gender identity?
MS. BORELLI: Objection; form.
THE WITNESS: Yes.
Q. (BY MR. DAVID) What are some of those terms?
MS. BORELLI: Objection; form.
THE WITNESS: There are many different ways that people describe their gender. I think that some of the things I've heard, a gender, third gender, hemiboy, demigirl. Those are just a few.
Q. (BY MR. DAVID) Are these ways that patients are describing their own gender identities to you?

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THE WITNESS: You're muted. Not you, Caleb.
MS. BORELLI: Thank you. I was attempting to lodge an objection.
Objection; form.
Thank you, Dr. Olson-Kennedy.
THE WITNESS: Yes. These are ways that patients describe their gender to me.
Q. (BY MR. DAVID) When someone is describing their gender identity as "third gender," what does that mean?
MS. BORELLI: Objection; form.
THE WITNESS: It doesn't mean the same thing to every person.
Q. (BY MR. DAVID) So is that simply someone's way of saying that they do not identify as either male or female?
MS. BORELLI: Objection; form.
THE WITNESS: Sometimes it might mean that.
Q. (BY MR. DAVID) What else would it mean?
MS. BORELLI: Objection; form.
THE WITNESS: Sometimes people use that to describe gender fluidity.
Q. (BY MR. DAVID) Can you explain "gender fluidity"?

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MS. BORELLI: Objection; form.
THE WITNESS: So sometimes people say that they feel differently or their gender feels stronger, more male on one period of time versus another period of time. That's extraordinarily rare that somebody describes their gender in this way, but I have had a handful of people describe their gender in that way.
Q. (BY MR. DAVID) In the cases where someone has described their gender that way at some point -- and I'll use this as an example, more male sometimes -- is there a specific event occurring or something that triggers that person to feel that their gender identity is more aligned with a traditional or stereotypical male identity?
MS. BORELLI: Objection; form.
THE WITNESS: No. Not that anyone has disclosed to me that there's a triggering event, no.
Q. (BY MR. DAVID) Is there some rhyme or reason as to when those individuals feel that their gender identity aligns more with a male or female gender identity?
MS. BORELLI: Objection; form.

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THE WITNESS: When you say "rhyme or reason," I guess I don't understand what you're asking.
Q. (BY MR. DAVID) And throughout this deposition, there will certainly be times that I ask bad questions. And if you don't understand, please let me know.
What I'm saying, is there a cycle? Is there a friend group that they're hanging out with more that makes them feel more aligned with a male gender identity or a female gender identity?
What are the reasons that they have provided to you that they feel more male aligned or more female aligned at certain times?
MS. BORELLI: Objection; form.
THE WITNESS: So again, I can't make a generalizable statement about every person, A, because it's very rare. And in the handful of people that over the course of my practice that have described this -- it sounds like you're asking me is there a rhythm or a reason that people have described to me. No.
Q. (BY MR. DAVID) Going back to your report, the next sentence in paragraph 18 is "It has a strong biological basis."

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Can you tell me what the biological basis is for gender identity?
MS. BORELLI: Objection; form.
THE WITNESS: So the studies that are looking at the morphology and connectiveness of the brain are giving us more and more indicators that gender identity lives in the brain, that it's determined by the way that our brain cells are organized and the way that they connect to each other.
This is certainly not my area of -- I'm not a neurobiologist. But these studies have been very helpful for us in understanding that the brain is playing a role in our gender, all of our gender identities, not just the gender identities of transgender people.
Q. (BY MR. DAVID) And if it's outside of your comfort zone because you're not a neurobiologist, please just let me know.
Is there some sort of structure of the brain that we could examine at birth that would tell us whether someone is more likely or less likely to have a transgender identity?
MS. BORELLI: Objection; form.
THE WITNESS: I haven't seen studies in

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infants or children yet in brain structures. These studies have only been carried out in adults as far as I know.
Q. (BY MR. DAVID) Okay. But in the adult population, is there some difference in brain structure that would allow us to identify whether someone is more or less likely to have a transgender identity?
MS. BORELLI: Objection; form.
THE WITNESS: I don't think that the science has taken us that far yet. I think that the information that we do have is moving us in that direction, but we don't have that yet.
Q. (BY MR. DAVID) Going back to your report, the next sentence in paragraph 18 is "Every person has a gender identity."
And my question is, we just talked about individuals who have neither a male nor female gender identity.
Do those people still have a gender identity?
MS. BORELLI: Objection; form.
THE WITNESS: Yes.
Q. (BY MR. DAVID) Is there a way to define that person's gender identity?

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MS. BORELLI: Objection; form.
THE WITNESS: I think that our lexicon is evolving around the way that people can describe their gender identity, but it's inadequate right now.
Q. (BY MR. DAVID) Are there studies that show that every person has a gender identity?
MS. BORELLI: Objection; form.
THE WITNESS: Not that I know of.
Q. (BY MR. DAVID) And such my question, what is your basis for stating that every person has a gender identity?
MS. BORELLI: Objection; form.
THE WITNESS: 16 years of working with people and talking about gender.
Q. (BY MR. DAVID) Are you referring to the time that you've spent at the Transyouth Health and Development Center?
MS. BORELLI: Objection; form.
THE WITNESS: Well, I've been working with people who have trans identities for 16 years.
But conversations with all people indicate that all people have a gender identity.
Q. (BY MR. DAVID) Back to your report, go

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to paragraph 20, the first sentence says: "While both gender identity and sex are often assumed and treated as binary and oppositional, they're more accurately experienced as along a spectrum."
First, did I read that correctly?
A. Yes.
Q. Can you explain how sex is experienced along a spectrum?
MS. BORELLI: Objection; form.
THE WITNESS: So I think further down in the paragraph I talk about this. That because sex is a multi-faceted part of our self-hood, there are many, many things that are contributing to it.
And we know, for example, that chromosomes don't only exist as XX and XY, that there are lots of different chromosomal variations.
And additionally, the pathway from chromosomes to reproductive tract genitalia, there are many, many different pathways of development in that trajectory.
And similarly, from the trajectory from genitalia and reproductive tract to brain development, there are also multiple pathways of development, hence why we have an LGBTQA group of

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humans.
Q. (BY MR. DAVID) Outside of -- well, first let me ask, those chromosomal variations that you discussed are incredibly rare in the world, correct?
MS. BORELLI: Objection; form.
THE WITNESS: One of the challenges about that is most people actually don't have an awareness of their chromosomes, of their karyotype. So I think our understanding of it is probably limited to folks who are coming in with various and related downstream issues from some of those chromosomal abnormalities.
So we actually don't have an understanding of the prevalence rate of chromosomal variations for sex chromosomes.
Q. (BY MR. DAVID) So is the literature not informed enough on the prevalence rate for variations in chromosomal makeup?
MS. BORELLI: Objection; form.
THE WITNESS: I don't know that there would be a reason for the literature to take up doing karyotypes on gobs and gobs of individuals.
So I don't know that I would say it's inadequate. I just think people don't do research

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on things that aren't necessarily informative.
My point is just that we make sometimes assumptions about people's karyotypes that may or may not be backed up by their actual karyotype is my point.
Q. (BY MR. DAVID) And in this paragraph you cite to the Endocrine Society Guidelines, correct?
A. Yes.
Q. Are you familiar with the Endocrine Society's Scientific Statement in 2021?
MS. BORELLI: Objection; form.
THE WITNESS: I would have to see it to know if I was familiar with it.
Q. (BY MR. DAVID) Let me see if I can make that happen. And I will move this over to the folder, assuming I know how to do that.
MS. BORELLI: Also, Caleb, we've been going for about an hour. Does it make sense to take a break while you're working with this exhibit, and we can pick that up after the break?
MR. DAVID: That works perfectly.
MS. BORELLI: Great. Dr. Olson-Kennedy, how much time would you like? Five to ten minutes?
THE WITNESS: Yeah, I need five minutes

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just to deal with a patient issue that just came on to my screen.
MR. DAVID: Take all the time you need for that, absolutely.
THE VIDEOGRAPHER: We're going off the record. The time is 9:29 a.m.
(Break taken from 9:29 a.m. to 9:39 a.m.)
THE VIDEOGRAPHER: We are back on the record. The time is 9:39 a.m.
(Deposition Exhibit No. 2 was marked.)
Q. (BY MR. DAVID) Doctor, before we went on a break, I was marking as Exhibit 2 the 2021 statement of the Endocrine Society that is titled "Considering Sex as a Biological Variable in Basic and Clinical Studies: An Endocrine Society Scientific Statement."
And first, I want to make sure that you and counsel are able to see the exhibit in the marked exhibits folder.
MS. BORELLI: Caleb, I am able to see it, but what I've just realized looking at both Exhibit 1 and Exhibit 2 is that they appear to have been published without exhibit stickers, unless it's just my view. But I think we may need the good folks at Veritext to ensure that exhibit

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stickers are affixed to these when the transcript is produced.
THE REPORTER: We can do that, no problem.
MS. BORELLI: Thank you so much.
Dr. Olson-Kennedy, are you able to see Exhibit 2?
THE WITNESS: I can.
MS. BORELLI: Great.
Q. (BY MR. DAVID) And, Doctor, I want to specifically draw your attention to the first -- I guess it's the first full paragraph of the article after the abstract. So it's on page 2.
Are you following me?
A. Yes.
Q. Okay. And I'll go ahead and read, and then I'll ask my question.
"Sex is an important biological variable that must be considered in the design and analysis of human and animal research. The term 'sex' and 'gender' should not be used interchangeably. Sex is dichotomous with sex determination in the fertilized zygote stemming from unequal expression of sex chromosomal genes."
First, did I read that correctly?

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A. Yes.
Q. Okay. And do you disagree with that statement?
MS. BORELLI: Objection; form.
THE WITNESS: I think that I would have to read through this entire article, because I feel like this entire article is talking about -- or it looks like this article or this statement -- is this a statement? What is this called? Yes. A scientific statement -- going on to talk about the myriad of ways that sex is actually not dichotomous.
So it seems at odds with itself, but I would need to read through the whole article in order to make an opinion about this particular statement.
Q. (BY MR. DAVID) Okay. Fair enough. Thank you.
So we'll move on from Exhibit 2 and go back to your report, which has been marked as Exhibit 1. And paragraph 21, which is on page 6, says, "As early as 1966, it has been understood that gender identity cannot be changed. Efforts to do so have been shown to be unsuccessful and harmful."

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First, did I read that correctly?
A. Yes.
Q. And I'm going to ask this, and it's going to be a terrible question, but -- so if you don't follow me, let me know.
At other points in your report you talk about that there are individuals who have been known to have a transgender identity at some point and to then have a cisgender identity at another point that you say is a very small number of individuals.
And my question is how does that interplay with the statement that gender identity cannot be changed?
MS. BORELLI: Objection; form.
THE WITNESS: Can you -- what's the paragraph that I say that in?
Q. (BY MR. DAVID) Sure. If you look at paragraph 69, you say in the middle of that paragraph, "The question is not 'should we provide access to medical interventions for people who had gender dysphoria in childhood that dissipated in adolescence?' because that population is not the population presenting for treatment, and medical care is not indicated for that population of

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children."
So does that not imply that there is a population of children who have gender dysphoria in childhood that dissipated in adolescence?
MS. BORELLI: Objection; form.
THE WITNESS: The quotation -- what I'm talking about here are people who, in childhood, prepubertal childhood, gender expression is outside of what we would typically expect for someone with their designated sex at birth.
And this comes from -- and again, the studies that look at children that are early studies really did not -- all of those people in those studies did not meet the criteria for a diagnosis of gender dysphoria.
And so without knowing those children, I can tell you that sometimes people's gender dysphoria dissipates. That doesn't actually talk about their gender identity changing.
Q. (BY MR. DAVID) So there are no individuals who ever desist from a gender identity?
MS. BORELLI: Objection; form.
THE WITNESS: How are you describing desistance?

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Q. (BY MR. DAVID) Someone who identifies as transgender and then later reverts and does not identify as transgender.
MS. BORELLI: Objection; form.
THE WITNESS: I think that there are a handful of individuals like that. I don't really have anyone like that in my practice, and I think it is a very complex situation.
Q. (BY MR. DAVID) And you've -- in the previous paragraph, paragraph 20, you cited to the Endocrine Society Guidelines.
And I'm sure that you're familiar that they state that 85 percent of children with a transgender identity desist into adolescence, right?
MS. BORELLI: Objection; form.
THE WITNESS: That's actually not what they say, and that's not what the studies they're referring to talk about either. They're talking about people who may or may not meet criteria for diagnosis of gender dysphoria, not have an identity that's a transgender identity.
Q. (BY MR. DAVID) Well, let's go ahead and look at that. I'll see if I can pull that up.
MR. DAVID: Okay. I have just moved over

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into the marked exhibits folder on Veritext what we'll mark as Exhibit 3 to your deposition. And that is the "Endocrine Treatment of Gender-Dysphoric/Gender-Incongruent Persons: An Endocrine Society Clinical Practice Guideline.

(Deposition Exhibit No. 3 was marked.)

THE WITNESS: Yes.

Q. (BY MR. DAVID) And if you will go to page 11 of that document --

A. They're not numbered like that in my pages.

Q. Oh, I'm sorry. The actual number in the top right is 3879.

A. Okay.

Q. And in the left-hand column, there is a paragraph that starts with a bold heading of "Evidence."

Are you with me?

A. Yes.

Q. Okay. And I'll go ahead and read it. "In most children diagnosed with GD/gender incongruence, it did not persist into adolescence. The percentages differed among studies, probably dependent on which versions of the DSM clinicians used, the patient's age, the recruitment criteria,

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and perhaps cultural factors. However, the large majority (about 85 percent) of prepubertal children with a childhood diagnosis did not remain GD/gender incongruent in adolescence."

First, did I read that correctly?

A. Yes.

Q. And the statement that a large majority, about 85 percent, of prepubertal children with a childhood diagnosis did not remain gender dysphoric or gender incongruent in adolescence does not mean that there are individuals who have a desistance in their transgender identity. It just means that they no longer meet the diagnostic criteria.

Is that what you're saying?

MS. BORELLI: Objection; form.

THE WITNESS: So I think that there is -- I would have to look at this article that they're referencing to understand the -- why they're saying that. Because my understanding of these studies is that these people were not diagnosed with gender incongruence. And so it would be really important to look at the source documentation for that. We could pull through it and find it.

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But I think that it is fair to say that people get a better understanding of their gender identity as they get older.

But this particular cohort of children that's being referred to is a really important one, and I think that understanding exactly what was happening in these studies matters to this conversation.

Q. (BY MR. DAVID) So back to the statement in your report that since 1966 it's been understood that gender identity cannot be changed, I'm sure that you are familiar with individuals who have come out and said that they transitioned, had surgery, and have since regretted that and have published about it widely on the internet or the Washington Post or the New York Times, correct?

MS. BORELLI: Objection; form.

THE WITNESS: Do you have a specific example?

Q. (BY MR. DAVID) I don't know that I have a specific example of which individual it was, but you've never seen an article published in any source that was published by someone who went through transition and then stated that they

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regretted it?

MS. BORELLI: Objection; form.

THE WITNESS: I don't remember the sources. I have seen reports from two such individuals.

Q. (BY MR. DAVID) Who are the individuals that you've seen those reports from?

MS. BORELLI: Objection; form.

THE WITNESS: Walter Heyer and Keira Bell.

Q. (BY MR. DAVID) And I'm not familiar with either of those individuals.

Just to make sure that we're all on the same page, those were not patients of yours, correct?

MS. BORELLI: Objection; form.

THE WITNESS: Those were not patients of mine.

Q. (BY MR. DAVID) Okay. And are you saying in paragraph 21 of your report that it's incredibly rare for gender identity to change, or that these people that you just mentioned were misdiagnosed or had an incorrect perception of their own identity?

MS. BORELLI: Objection; form.

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THE WITNESS: I don't know either of them personally, but what I'm referring to in that paragraph 21 is about efforts on behalf of professionals trying to change someone's gender identity.

Q. (BY MR. DAVID) Okay. So let me back up, then.

Are you saying that gender identity cannot be forced to change?

MS. BORELLI: Objection; form.

THE WITNESS: That's correct.

Q. (BY MR. DAVID) Okay. Thank you. Misunderstanding on my part.

In the next paragraph, paragraph 22, you referred to Dr. Levine, and you also mention conversion or reparative therapy.

And I wanted to ask whether you are saying that Dr. Levine engages in conversion therapy.

MS. BORELLI: Objection; form.

THE WITNESS: So I'm not saying that. From the reports that he seems to lean on, people should go to therapy to become comfortable with their -- the body that they have, which is a way to sort of talk people out of their experience.

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So I don't know if it falls under the rubric of conversion therapy or reparative therapy, but that seems to be what he leans on.

Q. (BY MR. DAVID) Is your understanding based solely upon the report that he filed in this case?

MS. BORELLI: Objection; form.

THE WITNESS: Yes.

Q. (BY MR. DAVID) Okay. Can a transgender identity emerge in adolescence without childhood distress?

MS. BORELLI: Objection; form.

THE WITNESS: So can people come to understand their gender more fully in adolescence? Yes.

Q. (BY MR. DAVID) And I guess what I'm trying to understand is once the person in their adolescence more fully understands their gender identity, is that the point in time when that individual will begin to experience distress from the incongruence between their gender identity and their sex assigned at birth?

MS. BORELLI: Objection; form.

THE WITNESS: Well, I think what's really important to understand is that everybody's process is individual. So that could be a possible trajectory for someone.

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Q. (BY MR. DAVID) And this might be a bad question: Is there a traditional presentation of gender dysphoria?

MS. BORELLI: Objection; form.

THE WITNESS: Well, gender dysphoria is a list of criteria. And so in that sense, that is sort of the -- I guess if you -- I don't know what the word "traditional" means in this context, but there are people who meet that diagnostic criteria and people who don't. So there's set criteria, I guess, is what I mean.

Q. (BY MR. DAVID) And again, that was a bad question on my part. I'll preface this so I'm making myself somewhat clear here, I guess, or at least trying to.

I deal with a lot of medical malpractice cases, and there are people who present to the emergency room with appendicitis. And a doctor will say, "That's a classic presentation of appendicitis."

And my question is, is there a classic presentation of gender dysphoria?

MS. BORELLI: Objection; form.

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THE WITNESS: So, yeah. The people who meet the criteria is outlined in the DSM-5. That is the definition of gender dysphoria.

Q. (BY MR. DAVID) And is there an age or an age range in which the presentation meeting those diagnostic criteria most often emerges?

MS. BORELLI: Objection; form.

THE WITNESS: Here is an important place to differentiate between the diagnostic criteria in children and the diagnostic criteria in adolescence, because they're different.

So in order to have that diagnosis in childhood, the criteria are different than the ones that are outlined for adolescents and adults.

Q. (BY MR. DAVID) And my question is, is there an age range where it is more prevalent for someone to first have a diagnosis of gender dysphoria?

MS. BORELLI: Objection; form.

THE WITNESS: No. People get this diagnosis at all different stages of development and age.

Q. (BY MR. DAVID) So is it the same amount of patients or the same percentage of patients diagnosed with gender dysphoria that's a third in

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childhood, a third in adolescence, and a third in adulthood?
MS. BORELLI: Objection; form.
THE WITNESS: So are you specifically asking about my practice or just the whole group of people with gender dysphoria?
Q. (BY MR. DAVID) My question was broader than your practice, but if you can only speak to your practice, that's perfectly fine.
MS. BORELLI: Objection; form.
THE WITNESS: So I see patients up to the age of 25, sometimes 26, and people access services all the way from age 3 up to age 26.
But I think I said this earlier, that the average age that people come to seek services is around 16. But that's in an adolescent/young adult clinic.
Q. (BY MR. DAVID) Sure. So in your clinic seeing patients between the ages of 3 and 25, is it fair to say that your patient population is primarily teenagers?
MS. BORELLI: Objection; form.
THE WITNESS: Yes.
Q. (BY MR. DAVID) Okay. And in the population we discussed earlier that there has

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been a shift in the ratio with more individuals presenting with -- who were assigned female at birth than previously were presenting --
MS. BORELLI: Objection; form.
I apologize, Caleb.
MR. DAVID: You're okay. I'm being a little clumsy with this, so I'll start over.
Q. (BY MR. DAVID) We previously talked at the beginning of your deposition about there is a shift in the ratio of your patient population from primarily those who were assigned male at birth to now a greater number who were assigned female at birth; is that right?
MS. BORELLI: Objection; form.
THE WITNESS: Well, let me clarify. There was not a time -- we -- there was not a time -- I'm going to go back because the historical context is important.
We've been providing services at our division of adolescent medicine since the '90s. But since we started tracking our new referrals, we -- in 2010 to 2015, there was an equal ratio.
And then in -- sorry, 2014-2015, we started getting a higher number of people designated female at birth new for consultation.

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Q. (BY MR. DAVID) Okay. Has that population that you've seen starting to shift from 2014-2015, has it continued to today?
MS. BORELLI: Objection; form.
THE WITNESS: We still have -- it evened out a little bit -- at our center it evened out a little bit over the last year or two years, but we still have -- more than 50 percent of the people seeking services are designated female at birth, but it has evened out a little bit more.
Q. (BY MR. DAVID) Okay. Has that cohort of patients that has shifted that ratio been involved in studies regarding the efficacy of the services that you specifically provide, puberty blockers and hormone therapy? And we'll leave out the oral contraceptives.
But for puberty blockers and for the hormone therapy, has that cohort of patients been studied?
MS. BORELLI: Objection; form.
THE WITNESS: In -- are you talking about just broadly speaking, or in our program?
Q. (BY MR. DAVID) Well, let's start broadly speaking.
MS. BORELLI: Same objection.

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THE WITNESS: So I -- I have to look at it to be intimately familiar, but I think that the Dutch did a study that looked at the characteristics of the folks that were relatively new into their program for consultation compared to the folks that they've seen longer ago. But again, I'd have to look at it to know the details.
Those are -- within my program and three other large programs across the United States, those young people are enrolled in the study that I'm the principal investigator on. So they are currently being studied.
Q. (BY MR. DAVID) Let's shift, then, to talk about your study.
First, when did your study begin?
MS. BORELLI: Objection; form.
THE WITNESS: 2015.
Q. (BY MR. DAVID) And is this the study that's mentioned in your report as being an NIH grant-funded study?
MS. BORELLI: Objection; form.
THE WITNESS: Yes.
Q. (BY MR. DAVID) How did you become aware that there was an NIH grant available?
MS. BORELLI: Objection; form.

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THE WITNESS: I am a researcher in addition to a clinician. And anytime that grants become broader, grants become available around subject matter that we're interested in, we are -- sometimes it's our division head or our assistant division head who will notify us as researchers that there are RFAs, or requests for application, around certain subject matter.

I don't remember exactly on this one. It was a long time ago that we started working on this grant, so I don't remember the exact details of how I came to know that that was available.

Q. (BY MR. DAVID) And I apologize. I'm completely unfamiliar with how this process works. So I guess what I'm asking, is there, like, a web page that has a list of requests for application?

A. Yes, there are those. And then sometimes institutions will send out, for example, like "Here's all the available grant funding," and they say everything that's available.

So there's a variety of ways that people become -- researchers become alerted to larger RFAS.

Q. And if you don't know the answer to this, please just tell me.

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Does there have to be legislation that provides the money to NIH for them to then provide grants?

MS. BORELLI: Objection; form.

THE WITNESS: I'm not really sure how the NIH budget gets determined. That's as confusing to me as possibly to you.

Q. (BY MR. DAVID) Okay. Do you know how it is determined that there will be a grant for a specific area of study?

MS. BORELLI: Objection; form.

THE WITNESS: No.

Q. (BY MR. DAVID) Okay. And what was the specific area of study for the grant that you were awarded?

MS. BORELLI: Objection; form.

THE WITNESS: The broad umbrella was to fund studies that were looking at the quality of life for LGBTQAI folks. That's the broad umbrella. I would have to go back and look at it to know exactly the specifics of it.

Q. (BY MR. DAVID) And did your study focus on the LGBT population as a whole, or was it specific to transgender individuals?

MS. BORELLI: Objection; form.

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THE WITNESS: So while our study is specifically looking at the impact of interventions for people with gender dysphoria, there are people also in our study that identify as lesbian, gay, or bisexual.

Q. (BY MR. DAVID) And as much as you can, can you explain to me what your study entailed?

MS. BORELLI: Objection; form.

THE WITNESS: So the study is still ongoing, and it is concerned with the -- both the physiologic and the psychosocial outcomes for people who either are new to puberty blockers or are new to gender-affirming hormones who sought that care related to gender dysphoria.

Q. (BY MR. DAVID) Can you explain what your methodology is for this study?

MS. BORELLI: Objection; form.

THE WITNESS: Sure. So we have four centers around the country who are specialized in providing gender-related care for adolescents and young adults.

And people who are eligible for the study are people who have a diagnosis of gender dysphoria who are going to initiate care with either puberty blockers or gender-affirming

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hormones.

And they, at baseline, will answer many, many, many, many -- so many questions.

And additionally, there are things that are abstracted from the charts about physiology, so baseline laboratory values, blood pressure, bone density, and then those same questions and same data is gathered as they move along in their treatment course.

Q. (BY MR. DAVID) So those lab values, how often are they drawn? How often are labs taken?

MS. BORELLI: Objection; form.

THE WITNESS: So because this is an observational study, we are observing what people are doing in practice related to how we would all monitor the safety of interventions within a clinical context.

The thing that makes it different are the questions about psychosocial and behavioral health.

Q. (BY MR. DAVID) Okay. So you're the principal investigator? Is that the term?

A. Yes.

Q. Okay. As the principal investigator, you're not directing the care at any of these four

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clinics that you're observing; is that right?
MS. BORELLI: Objection; form.
THE WITNESS: Just for clarity, there are people enrolled in the study at our site that are my clinical patients.
Q. (BY MR. DAVID) I guess I should have asked that first.
What are the four centers that you mention that are being observed, that the observations are taking place at?
MS. BORELLI: Objection; form.
THE WITNESS: So one of them is my own center at Children's Hospital Los Angeles.
There's also the Child & Adolescents Clinic at UCSF, Benioff Children's Hospital.
The third site is at Lurie Children's Hospital in Chicago.
And the fourth site is at Boston Children's Hospital in Massachusetts.
Q. (BY MR. DAVID) And if I'm understanding you correctly, when you're saying this is observational, you're not telling anyone at Boston Children's Hospital how they are to go about treating the patients that are involved in the study; is that correct?

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MS. BORELLI: Objection; form.
THE WITNESS: That's correct. And also important to note that this is what's called a multi PI or multi principal investigator. So there is a site PI, a PI at each of those other sites in addition to myself.
Q. (BY MR. DAVID) Okay. So the care that is being given at each of the four centers could vary depending on patient needs; is that right?
MS. BORELLI: Objection; form.
THE WITNESS: That's correct.
Q. (BY MR. DAVID) But are there specific intervals at which the patients complete the -- I don't remember the term that you used, but you said the questionnaire? Is that the term you used?
A. Yes, that's probably the broadest way to understand it, yes.
Q. Okay. So what are the intervals at which the patients complete those questionnaires?
MS. BORELLI: Objection; form.
THE WITNESS: So at baseline, before they start any interventions is the first time point. And then six months, a year, 18 months, 24 months, and then each subsequent year thereafter.

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Q. (BY MR. DAVID) You said that the study is still ongoing; is that correct?
A. That's correct.
Q. And I did read somewhere in your report -- I think you said that it was extended for five years; is that correct?
A. Yes.
Q. Okay. That's five years from what time frame?
A. NIH grants, these -- this specific mechanism is their five-year grants. And so they get renewed, if they get renewed, in five-year time frames.
Is that what you're asking? So this is the second five-year time frame.
Q. Okay. So did the first one start in 2014?
A. 2015 was when the study started getting funding, but that's not when enrollment started. Enrollment didn't start -- I'd have to go back and look at the exact dates. I think it's 2016.
Q. Okay. So when did the five-year -- when did the five-year extension begin?
A. Another complicated question. So it technically -- and the years are also squirrelly

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because the funding doesn't always start the same month.
So 2021 was when the second five years -- I have to go in and look at the exact dates, but 2021 is when the second five years started.
Q. Okay. And have there been any results of the first five years published?
MS. BORELLI: Objection; form.
THE WITNESS: So there have been some papers on the way that we created the network, on the things that we considered the protocol for the study, and baseline findings from the study have been published.
And there have been -- I have to go in and look at the exact number, but there have been a few papers published on the metabolic changes or the observed metabolic changes from the cohort after 12 months and two years.
Q. (BY MR. DAVID) You said that there have been some papers on creating the network.
Can you explain what that means?
MS. BORELLI: Objection; form.
THE WITNESS: So I wrote a manuscript about how the four sites came together, how we

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make decisions about what things to look at, what questions to ask, and how some of the -- it's been a long time since I wrote this paper, but some of the challenges of this kind of research.
Q. (BY MR. DAVID) So did the four principal investigators come together to determine how they would define the eligibility criteria for the study?
MS. BORELLI: Objection; form.
THE WITNESS: Yes.
Q. (BY MR. DAVID) Okay. And if I'm understanding correctly, the eligibility criteria is -- there has already been a diagnosis of gender dysphoria, but the patient has not yet started puberty blockers or cross-sex hormones; is that correct?
MS. BORELLI: Objection; form.
THE WITNESS: Just for clarity -- this is an important thing. So in the cohort of people who are going to start puberty blockers, they will have been naive to any treatment related to gender dysphoria.
For the people starting gender-affirming hormones, there are a small number of people that also had puberty blockers, but they were naive or

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new to hormone treatment.
Q. (BY MR. DAVID) Okay. So are there two different cohorts that you're following?
A. Yes.
Q. And so one is -- one cohort is patients who have not yet received puberty blockers, and another cohort is patients who have received puberty blockers but have not yet received hormones; is that correct?
MS. BORELLI: Objection; form.
THE WITNESS: No. There's one cohort of people who are going to initiate puberty blockers, and they received nothing -- no hormones, no puberty blockers, nothing related to gender dysphoria.
The second cohort are young people who are going to start taking gender-affirming hormones. And some of them have not -- the majority of them have not received any interventions related to gender. There is a small portion of those kids, of those people in that second cohort who had puberty blockers when they were younger.
Q. (BY MR. DAVID) Okay. So in the second cohort of individuals who are going to receive

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gender-affirming hormones, are there subcohorts between those who have received puberty blockers and those who haven't?
MS. BORELLI: Objection; form.
THE WITNESS: Yeah. So it's complicated in research, because when we are looking at different aspects of the study, we will stratify by subcohort maybe by designated sex at birth, maybe by people who experience puberty blockers, et cetera.
So it's -- I don't want to say that there are subcohorts. It's the ways that data are analyzed based on what outcomes you're looking at.
Q. (BY MR. DAVID) Okay. So has any literature been published to date that would show at baseline -- I think you said at baseline, 12 months, and 24 months the -- you said that there either has been some published at baseline, 12 months, and 24 months for metabolic changes; is that right?
MS. BORELLI: Objection; form.
THE WITNESS: That's correct.
Q. (BY MR. DAVID) Okay. And does that include any of the -- you also mentioned blood pressure and bone density.

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Has that also been published?
MS. BORELLI: Objection; form.
THE WITNESS: This is kind of complicated. I mean, I'm happy to send you a list of the publications, because they're separated out by different aspects of expected metabolic changes or tracking metabolic changes.
Q. (BY MR. DAVID) Okay. So if I were to go to PubMed or one of those, ResearchGate -- one of those research sites, is there literature that I could find from your study that tells me what the blood pressure was of a cohort of patients at baseline, 12 months, and 24 months?
MS. BORELLI: Objection; form.
THE WITNESS: Not exactly that, but there is a manuscript that talks about those changes over specified periods of time.
Q. (BY MR. DAVID) Okay.
A. And it doesn't give exact -- it doesn't give exact numbers of blood pressures for each participant. It talks about the changes, maybe, in blood pressure over time.
But again, I have to go back into that manuscript, and that was not the focus of the manuscript. So I believe that it's in there as an

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incidental, sort of parts of looking at metabolic changes with hormones specifically.
Q. And again, this might be just me not understanding things.
But in terms of taking a baseline blood pressure, do you take it multiple times over multiple days to kind of create a baseline?
Because my understanding is that, you know, my blood pressure might be high because I have something that's particularly stressful going on or something along those lines. Or it could be low just depending on what's going on that particular day. So I'm trying to understand how you determine a baseline blood pressure for your study.
MS. BORELLI: Objection; form.
THE WITNESS: So there are mechanisms in medicine when anyone has a high blood pressure, specifically if they have a high blood pressure that would make you pursue a clinical course where you would have repeated blood pressures. So...
Q. (BY MR. DAVID) Okay. So there's a way to control for their being outliers?
A. Yes.
Q. Okay. So same kind of series of

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questions as to bone density.
Is there somewhere that I could go in the internet and find literature from your study that says this is what someone's bone density -- or this is what the cohort's bone density was from baseline, 12 months, and 24 months?
MS. BORELLI: Objection; form.
THE WITNESS: Well, you can definitely go to literature and look at the paper about baseline bone density.
My knowledge of where the follow-up bone density paper is in its process of publishing is not 100 percent clear right now. I would have to go ask the primary author of that manuscript.
Q. (BY MR. DAVID) Okay. How many patients were in the cohort, both cohorts, at baseline?
MS. BORELLI: Objection; form.
THE WITNESS: Okay. So in the blocker cohort, there are 91 young people and 91 caretakers, because data is being collected from the parents or legal caretaker or guardian of the young person in the study.
And then in the older cohort, there are 314.
Q. (BY MR. DAVID) Are caretakers involved

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in the older cohort?
MS. BORELLI: Objection; form.
THE WITNESS: Data is not being collected on the caretakers in the older cohort, no.
Q. (BY MR. DAVID) As of the last time that you were aware, are all 91 of the patients in the first cohort, the younger cohort, still in the study?
MS. BORELLI: Objection; form.
THE WITNESS: So people, because of the way that grant re-funding works, people have to be re-consented to be enrolled in the second part of the study. And so we have not reenrolled everyone from that first cohort yet.
Q. (BY MR. DAVID) Okay. So at the end of the first five-year period, were there still 91 patients in the younger cohort?
MS. BORELLI: Objection; form.
THE WITNESS: No, there was some attrition, which we would expect. I think all us researchers struggle with COVID because we had to pivot the way our data was collected. So I don't have the exact numbers in front of me, but yes, there was some attrition from the study.
Q. (BY MR. DAVID) And was there attrition

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for the second cohort as well?
MS. BORELLI: Objection; form.
THE WITNESS: Yes.
Q. (BY MR. DAVID) Other than metabolic values, blood pressure, bone density, and the questionnaires that you have that we've discussed, are there other outcomes that you are measuring in your study?
MS. BORELLI: Objection; form.
THE WITNESS: I'm not sure I know what you mean. So within the questionnaires, there are hundreds of data variables that are assessing people's behavior and mental health and psychosocial well-being. So there are an enormous number of things that are being tracked.
Q. (BY MR. DAVID) Okay. I have seen studies that have tracked mental health visits before and after surgery or before and after hormone therapy.
Is that something that you're tracking?
MS. BORELLI: Objection; form.
THE WITNESS: So we are tracking service utilization. I think that falls in that category of what you're talking about.
Q. (BY MR. DAVID) I think that's exactly

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what I'm talking about. I just don't use the proper terms. I'm sorry.

Okay. Do you anticipate that your study will conclude at the end of this five-year extension?

MS. BORELLI: Objection; form.

THE WITNESS: I anticipate we will go for additional renewals as long as we can.

Q. (BY MR. DAVID) And again, this is just my ignorance of not knowing how this process works. It sounds like you are publishing -- I think you called them manuscripts -- as your research is developing and continuing.

A. I have to switch AirPods. Hang on one second.

Q. Can you hear us?

A. Hang on. Okay. Now I can.

Q. Okay. So if I'm understanding what's been happening correctly, as your study is ongoing, there have been manuscripts published that analyzed the results as they have happened so far; is that correct?

MS. BORELLI: Objection; form.

THE WITNESS: Well, there's a lot that's involved in publishing a manuscript. So data has

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to be analyzed and then conceptualized, written, and all of that.

So that's -- the idea is that we're doing that, but we are also still conducting, you know, and reenrolling new patients, so there's a lot happening all at once.

But yes, that is the idea is that, for example, right now there are two manuscripts in preparation about mental health outcomes after 24 months.

Q. (BY MR. DAVID) Okay. So just so that I'm clear, the study isn't being done so that at the end of ten years we can publish this one comprehensive document; it's something that's an ongoing process and there will be published articles and published literature as you're capable of doing it throughout the study?

MS. BORELLI: Objection; form.

THE WITNESS: Yes, that's correct.

Q. (BY MR. DAVID) Okay. You mentioned that the data has to be analyzed.

Is that something that you do as a principal investigator?

MS. BORELLI: Objection; form.

THE WITNESS: The research is done by a

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team, and so there are -- I am not personally doing the analysis of the data. That's a person whose specific area of expertise is in data management, data analysis, and bio statistics. So it's not me doing that.

Q. (BY MR. DAVID) Are you familiar with the evidence rating scale grade?

MS. BORELLI: Objection; form.

THE WITNESS: I am familiar with it. It's not an area of expertise, but I'm familiar with it.

Q. (BY MR. DAVID) Do any of your studies -- or excuse me. I keep saying the wrong terms.

Do any of the manuscripts that have been published so far out of your study, have they been -- has the evidence been graded under the grade scale?

MS. BORELLI: Objection; form.

THE WITNESS: I don't know the answer to that.

Q. (BY MR. DAVID) At least as it relates to the patients in the study that are at your clinic, how are -- I don't want to say recruit. I don't know if that's the right word, but how are they

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brought into the study?

MS. BORELLI: Objection; form.

THE WITNESS: So the way that people are recruited for the study is any -- it's complicated, because there are recruitment screening tools where, you know, if people are -- they meet the criteria for the study, the provider of their care -- I mean, there's more than one medical provider at all of these sites.

And so the provider of their medical care can fill out the screener or -- that happens in some places. The -- and sometimes the way it happens is the research coordinators will have -- get everybody scheduled for the day and they will talk to the provider, who is going to be eligible for the study.

That's -- so part of the criteria for inclusion is that someone must be seeking services at one of those four sites.

Q. (BY MR. DAVID) And what I guess I'm trying to fully understand is are these new patients coming into the clinic and they're being screened at that time? Or have they been there for a short period of time but have not yet started -- for instance, in the younger cohort,

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have not yet started puberty blockers?
MS. BORELLI: Objection; form.
THE WITNESS: They might be from either of those scenarios that you just described.
Q. (BY MR. DAVID) And you're not -- this isn't, like -- there isn't some public campaign seeking people to be a part of your study; is that right?
MS. BORELLI: Objection; form.
THE WITNESS: That's correct.
Q. (BY MR. DAVID) Okay. Are there providers outside of your clinic that refer patients for -- we'll just stick with the younger cohort.
Are there providers outside of your clinic that refer patients to you for medical treatment of gender dysphoria and also for potential enrollment in the study?
MS. BORELLI: Objection; form.
THE WITNESS: Do you mean that -- first of all, do you mean, like, do the providers refer people to our center because they want people with expertise with this care? It's not related to the study.
Q. (BY MR. DAVID) And that's -- so I assume

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that there are providers outside of your center who refer patients to you for care for gender dysphoria; is that right?
MS. BORELLI: Objection; form.
THE WITNESS: That's correct.
Q. (BY MR. DAVID) Okay. And what I'm asking is they're not also calling you and saying, "Doctor, I have a patient who I believe would benefit from a consultation with you for gender dysphoria and also might be -- might meet criteria for your study"?
MS. BORELLI: Objection; form.
THE WITNESS: No, that's never happened.
Q. (BY MR. DAVID) Okay.
A. At my site.
Q. Outside of your study, do you keep track of patient outcomes in any systematic way?
MS. BORELLI: Objection; form.
THE WITNESS: I mean, people who engage in care, that's part of -- it's embedded in the care. And I know that one of our providers does that, but it's -- that's part of clinical practice, is keeping track of people's clinical outcomes.
Q. (BY MR. DAVID) Sure. And again, bad

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question on my part.
There's a certain age range that you generally treat at your center, correct?
MS. BORELLI: Objection; form.
THE WITNESS: Yes. We treat people up to 25 or sometimes 26.
Q. (BY MR. DAVID) Okay. And what I'm asking is after those individuals leave your care, is there some mechanism in place for you to track their outcomes?
MS. BORELLI: Objection; form.
THE WITNESS: We don't have a mechanism. I mean, I happen to -- those people, we always invite them to keep in touch with us, let us know how they're doing or if they're struggling to engage in adult care. But we don't have a formal mechanism, as most people don't when people leave their practice.
Q. (BY MR. DAVID) Sure. Sure. I'm just trying to -- outside of your study that you're currently conducting, there isn't some other -- I don't want to say research, but some other way that you are tracking patient outcomes outside of your study; is that fair to say?
MS. BORELLI: Objection; form.

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THE WITNESS: Not after they leave our care.
Q. (BY MR. DAVID) Yes. Thank you for the clarification. That is what I meant.
Okay.
MR. DAVID: We've been going for about another hour, so if you all would like to take a break, I'm happy to do that.
THE WITNESS: That would be great.
MR. DAVID: Five minutes?
THE VIDEOGRAPHER: We're going off the record. The time is 10:42 a.m.
(Break taken from 10:42 a.m. to 10:50 a.m.)
THE VIDEOGRAPHER: We are back on the record. The time is 10:50 a.m.
Q. (BY MR. DAVID) Doctor, there is an article titled "Chest Reconstruction and Chest Dysphoria in Transmasculine Minors and Young Adults" that I believe you authored; is that correct?
A. That's correct.
Q. Did that article come out of the study, the NIH grant study that we've just been talking about?
MS. BORELLI: Objection; form.

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THE WITNESS: It did not.
Q. (BY MR. DAVID) Okay. I think that's one of the ones I couldn't find a PDF of.
Can you just generally explain what that article "Chest Reconstruction and Chest Dysphoria in Transmasculine Minors and Young Adults" was analyzing?
MS. BORELLI: Objection; form.
THE WITNESS: So the paper, the first part of the paper was creating a scale or measurement from some of the things that I'd been hearing over years and years of taking care of transmasculine patients with chest distress. And so we put together a scale of some of the more common things that we've heard from people.
But additionally, in that study, we really wanted to ask about some other aspects of chest surgery that I think are important. You know, did folks regret their decision, for example? Did they think it was a good decision? What kind of surgery did they have? What sort of potential complications did they have with the surgery?
So it was a way for us to understand what was happening around these things that people told

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us were happening around their chest. And then did those things change in the cohort of people or were those things different in the cohort of people who had already experienced chest surgery?
Q. (BY MR. DAVID) Probably not a fair question, so if it's not, just tell me.
Are you able to explain what the results of your study were?
MS. BORELLI: Objection; form.
THE WITNESS: Sure. I mean, in the young people who had not yet had surgery, their reporting of those particular elements of chest distress was very high.
And in the people who had experienced chest surgery, their report of those things very low.
And additionally -- I mean, I'd have to go in and look at it again to tell you about the reported side effects and things like that.
There was one person who said sometimes they regretted their decision to have surgery. That patient was my patient, and so I had worked with him for a very long time around this decision to undergo surgery.
And so it wasn't complicated by the fact

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that he didn't want a flat chest. He wanted a flat chest. He had feelings about the fact that he was going to carry a baby later and that he had mixed emotions about being able to feed the baby through his own chest.
So that's a summary of what was found.
Q. (BY MR. DAVID) And let me ask a foundational question first.
I assume that there was an informed consent process leading up to any surgery for the individuals that were included in this study; is that right?
MS. BORELLI: Objection; form.
THE WITNESS: Of course. There's informed consent for anyone who's undergoing surgery.
Q. (BY MR. DAVID) Okay. Were you a part of the informed consent discussions for the individuals who underwent surgery in this study?
MS. BORELLI: Objection; form.
THE WITNESS: Not for the surgical informed consent, no.
Q. (BY MR. DAVID) So did you separately provide risks, benefits, and alternatives to surgery for these individuals prior to them seeing

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a surgeon?
MS. BORELLI: Objection; form.
THE WITNESS: I think that it's important to mention here that all of those participants were not my personal patients. So that's probably an important distinction for my patients that we talk about the consultations for surgery.
I do talk with my patients about the many aspects of chest surgery that are really important, because I -- if I'm going to make a referral for someone to go to consultation with a surgeon, I feel like that's an important aspect of that care to discuss.
Q. (BY MR. DAVID) And is the loss of ability to breastfeed something that is discussed in that informed consent process prior to a consultation for chest surgery?
MS. BORELLI: Objection; form.
THE WITNESS: So for my patients, I have a very good understanding of where they are situated in relationship to fertility, future fertility as it pertains to potentially them carrying a child or them harvesting eggs for a biological child.
So, yes, that discussion around the

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ramifications of chest surgery are part of that discussion.
Q. (BY MR. DAVID) And for that particular patient who expressed some regret associated with -- well, let me ask, just so I'm clear.
If I'm understanding you correctly, the regret that was expressed by that patient was related to the ability to breastfeed; is that correct?
MS. BORELLI: Objection; form.
THE WITNESS: Yes. Well, yes. It was weighing, I hate my chest; I want it gone, but I also feel this obligation to this child that I might have in the future and that I won't be able to breastfeed this child. That was the ongoing dynamic of the concern that went on for a long, long time.
Q. (BY MR. DAVID) And following surgery, was it still a consideration that -- well, I'm asking a terrible question.
After the surgery, did the patient -- did that specific patient express a relief of the distress associated with his chest?
MS. BORELLI: Objection; form.
THE WITNESS: Yes.

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Q. (BY MR. DAVID) But that patient still had some regret as it related to the now inability to breastfeed a future child; is that correct?
MS. BORELLI: Objection; form.
THE WITNESS: That's correct.
Q. (BY MR. DAVID) Understanding that that was your patient, did you continue to follow that patient even after the study was concluded?
MS. BORELLI: Objection; form.
THE WITNESS: Yes. I mean, they were my patient. I'd have to go back into the medical record and look, but my memory is that that patient had surgery around '20 or '21, and they ended up moving to a different state, so I wasn't able.
But I continue to be in contact with them, so I did -- I provided medical care until he moved out of state.
Q. (BY MR. DAVID) Okay. And from your last communication with that patient, is it still sort of a mixed-emotions situation regarding the chest surgery?
MS. BORELLI: Objection; form.
THE WITNESS: So I don't recall if we talked about that specifically. I mean, his child

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is, like, way beyond the age of breastfeeding now, so I don't think it's a very relevant conversation anymore. So we haven't talked about that specifically.
Q. (BY MR. DAVID) So you mentioned that there were also complications that you were monitoring from these chest surgeries.
What sorts of complications were you monitoring?
MS. BORELLI: Objection; form.
THE WITNESS: So I think monitoring and capturing data are slightly different things, so it's important to say that.
These are well-known potential complications -- not complications -- not complications. Some of them are not complications but well-known side effects from surgery, like having some numbness in the chest area.
I think there were a handful of people who had hematomas or blood collection at the site of the surgery.
There were some people who reported the phenomenon called dog ears, and that has to do with the way that the tissue is connected on the side of the body. It happens for people who have

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a little bit more body fat when they have the procedure.
So there are a handful of different things. Some of those -- they call them -- the things like hematomas, those are all things that resolve. They were complications that happen in and around the time of surgery. And some of those things could take a longer time or don't resolve at all, like numbness.
Q. (BY MR. DAVID) So were there patients who underwent chest surgery and had a permanency to their numbness?
MS. BORELLI: Objection; form.
THE WITNESS: Yeah, I think that's a hard question to answer, because I'm only getting a cross-sectional. I don't know. I mean, permanent means it's there until you die, so I don't really know how to answer that question. There were some people that had -- that reported ongoing numbness of their chest, yes.
Q. (BY MR. DAVID) Okay. What was the time frame following surgery that the patients were followed?
MS. BORELLI: Objection; form.
THE WITNESS: I think that the longest

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one was five years from surgery, but I'd have to go back in. It's been a while since I wrote that paper to know the exact time frame.
Q. (BY MR. DAVID) Okay. Do you know whether there were any patients who after five years were still experiencing chest numbness?
MS. BORELLI: Objection; form.
THE WITNESS: Again, I'd have to go back into the data and look specifically.
Q. (BY MR. DAVID) Were there any patients who expressed dissatisfaction with their chest appearance following surgery?
MS. BORELLI: Objection; form.
THE WITNESS: I don't know if I asked that exact question. I don't believe that I did.
Q. (BY MR. DAVID) Now, I don't think I asked you this. If I did, I apologize.
What was the age range of the patient population for this study?
MS. BORELLI: Objection; form.
THE WITNESS: I think that the youngest -- so distinguishing age of surgery or age of taking the survey?
Q. (BY MR. DAVID) Let's start with age of taking the survey.

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MS. BORELLI: Same objection.
THE WITNESS: I'd have to go back in and look. I know that these ages are reported, but I want to say it was -- 14 might have been the -- on the young side. The youngest age of surgery was -- there was a 13-year-old in that cohort.
Q. (BY MR. DAVID) And what was the upper age?
A. Young 20s. 20s. I don't know exactly, but 23, maybe. I don't know. I'd have to go back in and look.
Q. If I understood your testimony from the beginning of your deposition correctly, you yourself do not perform surgeries, but you may refer patients for consultations for gender affirming surgery; is that correct?
MS. BORELLI: Objection; form.
THE WITNESS: That is correct.
Q. (BY MR. DAVID) How is it determined that you're going to refer a patient out for consultation for gender-affirming surgery?
MS. BORELLI: Objection; form.
THE WITNESS: Which surgery specifically?
Q. (BY MR. DAVID) We've been talking about chest surgeries. Let's stick with chest

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surgeries.
A. So again, I just want to emphasize the importance of individualized care, because everybody's process is different.
But people that get referred for chest surgery have chest distress; they have chest dysphoria. They indicate those things that are in my chest dysphoria scale.
They have all of -- perhaps a handful of, and they are -- it's impacting their quality of life. So people will talk about that in different kinds of ways, which is what I try to capture on that chest dysphoria scale.
For example, you know, "My life hasn't started yet because of my chest," or "I'm not" -- "I am not going to see doctors because of my chest," or "I don't want to bathe because of my chest," or "I can't participate in the things that other people my age are participating in," or "It's really impossible for me to have relationships."
The presence of a female chest contour is creating an issue with their functioning.
Q. (BY MR. DAVID) So is that how you determine whether a surgery is medically

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necessary?
MS. BORELLI: Objection; form.
THE WITNESS: The definition of "medical necessity" is healthcare services that are needed to, you know, diagnosis or treat a condition or its symptoms.
And so, you know, as related to the accepted standards of care and the standards of care for chest dysphoria and its disruption of function is chest surgery.
Q. (BY MR. DAVID) So if a patient presents with chest dysphoria, then -- let me start over.
If a patient presents to you and says that they're experiencing those issues like you mentioned, someone says that "I feel like my life hasn't started as a result of my chest," or some other expression of dysphoria with their chest, is there additional -- are there additional steps that you take prior to referring that patient for a consultation for chest surgery?
MS. BORELLI: Objection; form.
THE WITNESS: I think what gets lost in the translation is the relationship that I have with my patients. So for example, it's not like somebody comes in for their first visit with me on

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Tuesday and they're like, "I hate my chest," and I'm like, "Okay. Off to the surgeon you go."

I establish relationships with my patients. And sometimes people tell me about their chest distress when I first meet them, and sometimes that unfolds and as -- part of what my manuscript demonstrated was, you know, the length of time that people are taking testosterone.

So there's various and assorted things that contribute to people's growing discomfort or distress around their chest.

And so they have to see a therapist to get chest surgery, so they're either -- most of my patients are already engaged in therapy. If they're not, they -- you know, they get directed to see a therapist.

I think that it sort of short-shrifts the entire experience of creating the relationship and having the amount of time that I've had in this work, but also in the dialogue and ongoing conversations with people.

So I think that like many areas of medicine, we make these determinations about when to move forward with a consultation in the way that we would for anything that we might recommend

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that's more -- I'd say more advanced -- you know, advanced care or a more invasive procedure like a surgery.

Q. (BY MR. DAVID) So you're not going to make a referral for a surgical consultation the first time that you speak with a patient?

MS. BORELLI: Objection; form.

THE WITNESS: I don't think that I ever have. I suppose anything's possible, but that's not been something that's happened in my practice.

Q. (BY MR. DAVID) Okay. If a patient presents to you and is bringing up these feelings of chest dysphoria -- we'll just stick with chest because that's what we've been talking about.

If a patient presents to you and is discussing chest dysphoria with you and is describing the distress that they have associated with their chest, how is a surgical option brought up? Is it brought up by the patient? Is it brought up by you or another provider?

MS. BORELLI: Objection; form.

THE WITNESS: So I think one other group of people that we are overlooking are the parents. So many times -- so yes, any or all of those things.

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So I always talk with people about ways to mitigate chest dysphoria that aren't surgical. So binding, possibly K tape, other strategies for people to have a flat-appearing chest.

But the issue of surgery is sometimes brought up by parents; it's sometimes brought up -- most often by the patients themselves. All kinds of ways. Sometimes people will email me and ask about it.

But certainly when we talk about interventions, again, like over time, right? So people who are under 18 have a parent with them or parents or guardian that's going to be also responsible for signing off or going through that consent procedure. So I don't think it's always one way or another.

Q. (BY MR. DAVID) Sure. So do you have patients who express dysphoria both with their chest and with their vagina but only seek a chest surgery?

MS. BORELLI: Objection; form.

THE WITNESS: Sometimes.

Q. (BY MR. DAVID) In that situation, is it medically necessary both for a chest surgery and for a phalloplasty?

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MS. BORELLI: Objection; form.

THE WITNESS: Well, medical necessity is based on the individual, right? So it is important to understand where that person -- what that person is experiencing. And again, like, it's impossible to generalize and say every person that has genital dysphoria and chest dysphoria is going to do -- I mean, people are weighing that dysphoria with the intervention also.

So that's an important piece of the decision-making or the recommendations that come into play for providers.

Q. (BY MR. DAVID) I guess what I'm asking is if a patient has expressed similar levels of distress with both their chest and their genitals but is not ready to undergo a genital surgery, is the genital surgery still medically necessary?

MS. BORELLI: Objection; form.

THE WITNESS: I mean, they're medically necessary procedures, but if a person isn't ready to or they don't, these are -- we're talking about -- I mean, phalloplasty is a medically necessary procedure, but it also necessitates that somebody designate a large chunk of their time to undergoing those procedures.

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So I think what is important is thinking about what people are balancing when they make those decisions.

Q. (BY MR. DAVID) Is there a more significant post-surgical period of recovery for a genital surgery than there is for a chest surgery?

MS. BORELLI: Objection; form.

THE WITNESS: Generally speaking, absolutely.

Q. (BY MR. DAVID) And as a foundational question, I assume that when you are referring a patient for a surgical consultation for gender-affirming surgery, that is to treat gender dysphoria, correct?

MS. BORELLI: Objection; form.

THE WITNESS: That's correct in my practice, yes.

Q. (BY MR. DAVID) And so the goal would be to reduce the distress associated with -- sticking with chest surgery -- the stress associated with that person's chest; is that right?

MS. BORELLI: Objection; form.

THE WITNESS: That's correct.

Q. (BY MR. DAVID) Are there other risks that you are hoping that will be reduced as a

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result of surgery actually taking place?

MS. BORELLI: Objection; form.

THE WITNESS: Other risks? I'm not sure I know what you mean.

Q. (BY MR. DAVID) Another one of those bad questions.

Are you also hoping that there will be a reduction in the patient's level of depression or anxiety?

MS. BORELLI: Objection; form.

THE WITNESS: Yes. We always want people's body esteem and quality of life to go up with any interventions that we're recommending.

Q. (BY MR. DAVID) Okay. And are there long-term studies that have been done that have analyzed the long-term effects on -- we'll start with depression -- long-term effects on depression as a result of surgery?

MS. BORELLI: Objection; form.

THE WITNESS: What -- by "long-term," what's your time frame? What are you thinking of?

Q. (BY MR. DAVID) That's a good clarification. Let's say ten years.

A. I would have to go back into the literature to look exactly about that question of

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depression, as measured by depression.

I mean, I can tell you that from a clinical practice perspective, I've had people in my practice for longer than ten years, and people are very happy with their chest surgery. Most people consider it to be a major benchmark in their process.

I would have to look closer at the literature to be able to answer that question with absolute clarity. But there's been literature on -- there's been a lot of literature on chest surgery, but I'm not sure of the time frames.

Q. Are you aware of any literature on the long-term effects -- when I say "long-term," I'll go with ten years again -- on the long-term effects of antidepressants on depression?

MS. BORELLI: Objection; form.

THE WITNESS: Not my area of expertise, so I wouldn't know the answer to that.

Q. (BY MR. DAVID) Absolutely fair.

In your practice, if I remember correctly from the beginning of your deposition, you do prescribe antidepressants and antianxiety medications for some of your patients; is that correct?

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MS. BORELLI: Objection; form.

THE WITNESS: Yes, especially in the last handful of years, since the pandemic.

Q. (BY MR. DAVID) And that was something that I meant to ask you about at the beginning.

There has been a rise in -- especially from what I've read, at least -- teenagers who were assigned female at birth having depression and anxiety in the wake of the pandemic.

Is that your experience?

MS. BORELLI: Objection; form.

THE WITNESS: My experience has been across the board there has been an increase in not just adolescents but all age range of humans since the pandemic for sure.

Q. (BY MR. DAVID) And have you, in your clinical experience, seen that there have been helpful effects of prescribing antidepressants and antianxiety medications to those patients?

MS. BORELLI: Objection; form.

THE WITNESS: Some patients yes; some patients not as much. When patients are not having -- because psychiatry is not my primary area, when patients are not -- there's a variety of reasons why patients might not respond to

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medications, but I would usually bring our psychiatrist into their care.
And so, yes, some people get great relief; other people don't.
Q. (BY MR. DAVID) So there is a psychiatrist also at your center?
A. Yes. There are two, actually.
Q. Before a patient is sent for a surgical consultation, do one of your psychiatrists at the clinic perform a mental health evaluation?
MS. BORELLI: Objection; form.
THE WITNESS: What is a mental health evaluation, like specifically? Because I think people mean different things when they say that.
Q. (BY MR. DAVID) Sure. And I might be using the wrong terminology, but my understanding is that WPATH's standards of care require a mental health assessment, at least one mental health assessment prior to referring the patient for surgery; is that right?
MS. BORELLI: Objection; form.
THE WITNESS: That's correct. So that is why they go see the mental health provider prior to their consultation.
Q. (BY MR. DAVID) And is "mental health

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assessment" the right word?
A. That's what it's referred to as in the guidelines -- in the WPATH guidelines, yes.
Q. Okay. And I understand it's not your specialty, so again, if you don't know and I'm outside of your bounds, just let me know.
Do you know what the mental health assessment actually entails?
MS. BORELLI: Objection; form.
THE WITNESS: So I think that there's a couple of things, right? And I'm not certain that everybody does exactly the same thing exactly every time. So that's really hard because I don't know what happens inside people's offices.
But what I do know is that what comes out of that is a letter. And the letter sort of covers the things that are considered important by the WPATH. And it includes the diagnosis of gender dysphoria is met; it includes that somebody has the capacity to make informed consent about the procedure; it includes that somebody has an after care plan. There's a handful of other things. It includes, like, a little history of their experience.
So the letters encompass those elements

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that are considered -- I think are considered to be important in the assessment process.
But as to what happens in the room, don't know.
Q. (BY MR. DAVID) Are you aware of any other surgeries that first require a mental health assessment before the surgery?
MS. BORELLI: Objection; form.
THE WITNESS: I think that maybe, like, lap bands or things that are going to -- that are for weight loss, I think, also require that. I'm not sure if that's still the practice, but I know when it first started happening that those weight-reduction surgeries did require those similar things.
Q. (BY MR. DAVID) I know I'm jumping all over the place here, but if you don't mind to go back to your report, which was marked as Exhibit 1, and go to page 7, paragraph 24.
A. Yep. Got it.
Q. And read the first sentence. "Gender dysphoria is a serious medical condition characterized by distress due to a mismatch between assigned birth sex and a person's internal sense of gender."

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First, did I read that correctly?
A. Yes.
Q. And when you say that gender dysphoria is a serious medical condition, are you differentiating between a medical condition and a psychiatric condition?
MS. BORELLI: Objection; form.
THE WITNESS: All conditions that happen in all parts of the body are medical conditions.
Q. (BY MR. DAVID) Okay. And the reason I'm asking is because my understanding is that the diagnostic criteria to meet gender dysphoria comes from the DSM-5; is that right?
MS. BORELLI: Objection; form.
THE WITNESS: That's correct.
Q. (BY MR. DAVID) And the DSM-5 is a diagnostic manual of psychiatric conditions and their diagnostic criteria; is that right?
MS. BORELLI: Objection; form.
THE WITNESS: That's correct.
Q. (BY MR. DAVID) Okay. Are you aware of any other DSM-5 diagnoses that are treated with surgery?
A. There are some brain -- again, not my area of expertise, but there are some surgical

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interventions that can be recommended for people with very severe depression. And I think it's depression; I'd have to go back and look.

But yes. And they have a fancy name, so I can't remember what it's called.

Q. (BY MR. DAVID) Are those brain surgeries?

MS. BORELLI: Objection; form.

THE WITNESS: Yes.

Q. (BY MR. DAVID) And I understand it's not your area of expertise, but do you know how those surgeries are performed?

MS. BORELLI: Objection; form, outside the scope.

THE WITNESS: Yeah, I don't.

Q. (BY MR. DAVID) Okay. Do you refer patients for surgery for any DSM-5 condition other than gender dysphoria?

MS. BORELLI: Objection; form.

THE WITNESS: I do not.

MR. DAVID: I'm going to open up a new exhibit which we'll mark as Exhibit 4. And it is a gender dysphoria section of the DSM-5. And I've just dropped that into the marked exhibits folder.

(Deposition Exhibit No. 4 was marked.)

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THE WITNESS: Yep. Got it.

Q. (BY MR. DAVID) I would like to -- and we'll just -- we'll go to -- it's marked at the top left corner as 452.

Are you there?

A. Yep. Yes.

Q. And the -- this is the diagnostic criteria for -- and we'll focus on the first section there, the diagnostic criteria for gender dysphoria in children, correct?

A. Yes.

Q. And A says, "A marked incongruence between one's experienced/expressed gender and assigned gender, of at least six months' duration, as manifested by at least six of the following (one of which must be Criterion A1.)"

So first, did I read that correctly?

A. Yes.

Q. Okay. And Criteria A1 is required. You can't have the diagnosis without Criterion A1, correct?

MS. BORELLI: Objection; form.

THE WITNESS: Correct.

Q. (BY MR. DAVID) Okay. And the Criterion A1 is "A strong desire to be of the

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other gender or an insistence that one is the other gender (or some alternative gender different from one's assigned gender.)"

Did I read that correctly?

A. Yes.

Q. So that criterion includes individuals who are nonbinary or I think one of the terms you used was agender.

So that includes the whole spectrum of gender identity, correct?

MS. BORELLI: Objection; form.

THE WITNESS: Correct.

Q. (BY MR. DAVID) Okay. And then if my math is correct, you would need five of the next seven criteria to meet the definition of gender dysphoria; is that correct?

MS. BORELLI: Objection; form.

THE WITNESS: That's correct.

Q. (BY MR. DAVID) Okay. And some of these criteria, for instance, No. 3, "a strong preference for cross-gender roles in make-believe play or fantasy play," that's based off of our gender stereotypes of what is, for instance, stereotypical male make-believe play, correct?

MS. BORELLI: Objection; form.

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THE WITNESS: I didn't create these diagnostic criteria, but I imagine that's what this came from.

Q. (BY MR. DAVID) Well, and you apply these diagnostic criteria, correct?

MS. BORELLI: Objection; form.

THE WITNESS: I think again, importantly, these are the criteria for children or prepubertal children. And so decisions about medical interventions are not coming from these criteria.

But, yes, to diagnose someone who's -- a prepubertal child with gender dysphoria, we utilize these criteria.

Q. (BY MR. DAVID) And to be completely fair to you, you're not -- if someone meets the diagnostic criteria for gender dysphoria in children, you are not recommending any medical or surgical intervention for those people, correct?

MS. BORELLI: Objection; form.

THE WITNESS: That's correct.

Q. (BY MR. DAVID) It's when the patient meets the gender dysphoria in adolescents and adults criteria that you may recommend medical or surgical intervention?

MS. BORELLI: Objection; form.

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THE WITNESS: Correct.
Q. (BY MR. DAVID) Okay. So when you are diagnosing -- well, let me ask you this first: Do you diagnose gender dysphoria in children?
MS. BORELLI: Objection; form.
THE WITNESS: Yes.
Q. (BY MR. DAVID) Okay. And you use the DSM-5 diagnostic criteria to make that diagnosis, right?
A. Yes.
Q. Okay. So how do you interpret Criterion A3, "A strong preference for cross-gender roles in make-believe play or fantasy play"?
MS. BORELLI: Objection; form.
THE WITNESS: So I think the most culturally relevant way to check that off as part of your box is asking people about their avatars in their video games. So this is one place where we see people are maybe going to utilize an avatar of a different gender. That's one of the ways.
There are people who say, or their parents say, you know, "Every time my kid plays princess," or whatever, castle, whatever people call it, I don't really know. But when they're

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playing games that involve gender roles, they do inhabit the gender that you wouldn't necessarily expect them to inhabit.
So, "My kid always plays the daughter or the queen or the sister," or "My kid always says, 'Oh, I want to be the king,'" or "I want to be the prince."
So they're -- I'm happy to say there are still people who utilize their own imagination for names, and this is what they would endorse.
But I would say more commonly I will ask people about their avatars because I feel like more kids are doing virtual gaming than imaginative play.
Q. (BY MR. DAVID) And I guess the thought process is a child who is assigned male at birth will more likely than not choose an avatar that resembles a male.
Is that the thought process as to why that's something you can use for a diagnostic criterion?
MS. BORELLI: Objection; form.
THE WITNESS: So my experience in talking with kids who are asserting a different gender than their assumed one at birth, or their assigned

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sex at birth, will tell me, I will choose a -- you know, this avatar or that avatar.
I haven't really looked at data or I don't know if there is data about this, but this is -- it feels more relevant based on what people have told me.
And sometimes parents will tell me, oh, like in relationship to this kid's sister who always plays a girl or that -- that kind of thing.
Yes, I guess they're -- and again, I was not a part of making these diagnostic criteria. And so I think that there are, you know, things about this that probably should be updated.
But for this particular one, I have found that more people endorse in video games sort of asserting their gender as they might have in the past around imaginary games where gender roles are a part of it.
Q. (BY MR. DAVID) Do you find that that also applies to Criterion A4, which is "A strong preference for the toys, games, or activities stereo typically used or engaged in by the other gender"?
MS. BORELLI: Objection; form.
THE WITNESS: One of the things before

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prepubertal kids is that their thinking is more concrete. And so I think that there is -- that this criterion, probably more so than No. 3, is a very common one that's endorsed by patients. That, oh, my kid always wanted dolls, or my kid always wanted whatever the thing of the day is for girls.
Q. (BY MR. DAVID) When you say that for prepubertal children the thinking is more concrete, what do you mean by that?
MS. BORELLI: Objection; form.
THE WITNESS: I think that people's understanding of many things as they move into adolescence becomes much more abstract. And so it's not as easy for, say, a six-year-old to say this isn't a girl's toy or a boy's toy; this is just a toy. So for them, these kinds of criteria are, I think, more easily applied.
Q. (BY MR. DAVID) And can you explain what the difference is between Criterion A4 and Criterion A6?
MS. BORELLI: Objection; form.
THE WITNESS: I mean, I can give you what my interpretation of those things are.
Q. (BY MR. DAVID) Sure. Sure.

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A. So it comes holiday time and you are asking Santa Claus or whoever your person is that is responsible for your gifts, "I want a tiara and some nail polish," versus "I want a," you know, "basketball," or "I want" -- again, these things change over time.

And in Criterion A6, I feel like it is more like in the way that people play with their things.

And so, for example, a lot of my kids -- and again, it's not a large number, but a lot of my trans girls that come in when they're younger will say, "Yeah, no, I don't play basketball. I don't play sports like my brother does. I want to be over where the girls are playing with My Little Pony," and things like that.

Or for transmasculine kids, "Oh, yeah, I'm doing sports during recess. I'm not -- I don't want to be over with the girls doing the things that the girls do."

And again, so these ideas are very -- they're written in a time when I think that there was more differentiation between these things, and I think that there's less of that.

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But I still hear a lot of this, surprisingly, from a lot of people, that they have very clear ideas about what they like, what they don't like.

Q. (BY MR. DAVID) And today, is there -- or have you found that there is less differentiation between toys and games that are stereotypically associated with those who were assigned male at birth and those who were assigned female at birth?

MS. BORELLI: Objection; form.

THE WITNESS: I think there are, but it is still surprising how gendered things are.

Q. (BY MR. DAVID) Do you think that as -- well, let me ask you this: Do you think that as we go on, that things will become less gendered over time?

MS. BORELLI: Objection; form.

THE WITNESS: So you mean, like, as the years go by?

Q. (BY MR. DAVID) Yes.

A. As years go on? I really don't know the answer to that. It's very hard to know. It's very hard to know.

Q. If society progresses to a point where toys and games and occupations and the other aspects of society are less gendered, will that result in individuals experiencing less gender dysphoria?

MS. BORELLI: Objection; form.

THE WITNESS: I have no idea how to answer that question.

Q. (BY MR. DAVID) All right. Your report in paragraph 25 on page 7 mentions ICD-11, gender incongruence. And I wanted to ask you if that's something that you use in your practice.

MS. BORELLI: Objection; form.

THE WITNESS: It is not. We use ICD 10.

Q. (BY MR. DAVID) I don't know if this -- I don't know if this is the correct way to ask this, but are medical providers in America allowed to use ICD-11 codes?

MS. BORELLI: Objection; form.

THE WITNESS: I don't know how to answer that either.

Q. (BY MR. DAVID) Okay. Do you know why you use ICD 10 rather than 11?

MS. BORELLI: Objection; form.

THE WITNESS: My understanding is that we are at the mercy of the larger hospital structure.

Q. (BY MR. DAVID) And I guess it's to provide continuity of care between providers that

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want to make sure that everyone's working with the same deck of cards. Is that kind of the idea?

MS. BORELLI: Objection; form.

THE WITNESS: Yeah, I don't really know. I don't know who makes that decision.

Q. (BY MR. DAVID) Okay. Are you familiar with the ICD-11 definition of gender incongruence?

A. Not in detail. I mean, I know that it has changed over to the things that I wrote in my report about the changes, but I haven't delved in depth with it.

Q. It's my understanding that ICD 11's gender incongruence does not require clinically significant distress like that that is required in DSM-5; is that your understanding?

MS. BORELLI: Objection; form.

THE WITNESS: I think so. I think that's true.

Q. (BY MR. DAVID) Okay. And if a patient has gender incongruence but does not have clinically significant distress, is surgery -- gender-affirming surgery medically necessary, in your opinion?

MS. BORELLI: Objection; form.

THE WITNESS: I don't really know if I

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know how to answer that, to be honest. Because gender incongruence is a -- it seems to me that -- it seems that gender incongruence is a larger state of experience.

So I'm not sure how the ICD-11 is going to or does manage this, because I don't use it.

And so, for example, right now -- I don't know the intricacies of this, but I know that there is, like, a Z code in ICD 10 that's just transgender. And so it's sort of talking about this idea of someone being trans. Like, that's their experience.

And I think -- and then that's different than the coding for gender dysphoria. So I don't know if the -- the idea is because you can have gender incongruence and also have distress. Does that make sense?

So, for example, like, if somebody is experiencing distress around something that's happening in their life, you know, it may not necessarily be coded as, like, you know, divorce dysphoria, right? You can be the child of divorced parents and have, you know, adjustment reaction disorder or, like, issues of anxiety and things like that.

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So I just -- it's really hard to speculate on what the ICD-11 entails as far as the expectations around coding.

But my observation has been within the context of my practice that people do not seek surgery related to gender unless they have distress.

And so that's a weird hypothetical question to answer because I haven't had someone come in and say, "I haven't had distress about this, but I want surgery."

Q. (BY MR. DAVID) Okay. And I guess my real question is, is gender incongruence different than having a transgender identity?

MS. BORELLI: Objection; form.

THE WITNESS: As described by the ICD-11? I'd have to see exactly how they're describing it to answer that.

Q. (BY MR. DAVID) Okay. The only place that I know where that definition exists is in the draft for Standards of Care 8. So let's see if I can pull that up.

MR. DAVID: And, Tara, before I share this into the marked exhibits folder, the Standards of Care 8 draft that I have is 359 pages

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long.

Do you have any objection to me limiting it to the section that I'm going to talk about?

MS. BORELLI: I do not. And obviously if the witness needs to -- Dr. Olson-Kennedy needs to see a greater portion of the document in order to answer your questions, we can address it at that time. But for the moment, no objection here.

MR. DAVID: Okay. Great. I am now moving over to the marked exhibits folder what I marked as Exhibit 5.

And I will represent to you that this is the WPATH Standards of Care Version 8 draft that was circulated for public comment. And I've included kind of the cover letter and the table of contents and then the introductory statement.

(Deposition Exhibit No. 5 was marked.)

Q. (BY MR. DAVID) And I believe what I am looking for is in the first page of the introductory statement in the second paragraph.

And it says, "In the International Classification of Diseases Version 11, ICD-11, the diagnostic manual, the World Health Organization, the gender incongruence diagnosis is placed in a chapter on sexual health and focuses on the

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person's experienced identity and any desire for gender-affirming treatment that might stem from that identity. Such developments involving a depathologisation or, more precisely, a de-psychopathologisation of transgender identities are fundamentally important on a number of grounds. In the field of healthcare, they may have helped support a care model emphasizing a patient's active participation in decision-making about their own healthcare supported by a primary health care professionals. It is reasonable to suppose that these developments may also promote more socially inclusive policies, including legislative reform in gender recognition facilitating a rights-based approach without imposing requirements for diagnosis, hormone therapy, and/or surgery. Such developments may contribute greatly to the overall health and wellbeing of transgender and gender diverse people."

So first, did I read that very long section correctly?

A. Yes.

Q. Other than I may have messed up a couple of words there.

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And there might be a different portion of that that has the actual -- are you able to tell from that portion that I just read what the definition of gender incongruence under the ICD-11 is?

MS. BORELLI: Objection; form.

THE WITNESS: The thing that I take away from this is that it focuses on the person's identity and the desire for treatment that might stem from that identity.

Q. (BY MR. DAVID) And there's a different section that I thought I was showing you that I did not include. So let me quickly find that particular one.

MR. DAVID: Okay. So I'm about to drop a new exhibit -- we'll mark it as Exhibit 6 -- into the "Marked Exhibits" folder. I'll give you a second to pull that up.

THE WITNESS: Okay.

(Deposition Exhibit No. 6 was marked.)

Q. (BY MR. DAVID) Okay. So this is three pages from the WPATH Standards of Care 8 draft for public comment.

And on the second page of that -- see on the first page at the bottom where it's

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Statement 12A?

A. Yes.

Q. If you go to the second page, the second paragraph, there is a statement about three sentences in that says, "The most recent versions of these two systems, the DSM-5 and the ICD-11 respectively, reflect a long history of reconceptualizing and de-psychopathologizing gender-related diagnoses. Compared to the earlier version, the DSM-5 replaced gender identity disorder with gender dysphoria, acknowledging the distress experienced by some people stemming from the incongruence between experienced gender identify and sex assigned at birth. Compared to the ICD 10th edition, the gender incongruence classification was moved from the mental health chapter to a chapter 'Conditions Related to Sexual Health' in the ICD-11. One important reconceptualization in comparison to the DSM-5 gender dysphoria classification is that distress is not a required indicator of the ICD-11 gender incongruence classification."

First, did I read that correctly?

A. Yes.

Q. Okay. So my understanding from reading

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your report and from reading other things in this case is that a transgender identity is not a medical condition or mental health condition, correct?

MS. BORELLI: Objection; form.

THE WITNESS: Yes, that's correct.

Q. (BY MR. DAVID) Okay. I'm trying to understand if ICD-11's 's gender incongruence classification is something different than simply having a transgender identity.

MS. BORELLI: Objection; form.

Q. (BY MR. DAVID) Do you know the answer to that?

MS. BORELLI: Same objection.

THE WITNESS: I think that the -- it sounds like no, that's the point of moving away from it.

But the important part of that, I think, is that -- the interventions that are desired by the individual experiencing incongruence. Because they -- in ICD-11, they don't -- it's not a movement of saying gender incongruence is not a condition. It just moved it to a different place, right?

And so if we define medical necessity as

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healthcare services used to diagnose or treat an illness, injury, condition, disease, or symptoms that meet the standard of care, it still is a condition, I guess, is -- it's moving to a different section, but it's still considered a condition.

Q. (BY MR. DAVID) If there isn't any clinically significant distress from the incongruence, what are the medical and surgical interventions trying to relieve?

MS. BORELLI: Objection; form.

THE WITNESS: Yeah, so again, I go back to the statement of, like, people don't seek interventions if they aren't experiencing some distress about them.

Q. (BY MR. DAVID) So if a patient -- and I understand that this is hypothetical and that you have not experienced this in any way -- if a patient presented with gender incongruence without clinically significant distress and desired a surgical treatment, would that be medically necessary?

MS. BORELLI: Objection; form.

THE WITNESS: I guess I'll tell you if that happens.

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Q. (BY MR. DAVID) Okay. So it sounds to me that ICD-11 is making a transgender identity a medical condition.
MS. BORELLI: Objection; form.
Q. (BY MR. DAVID) Are you understanding that the same way that I am?
MS. BORELLI: Same objection.
THE WITNESS: Yeah. So this is a draft, so I actually -- I don't know. I'd have to see the document itself, but that's what it sounds like, yes.
Q. (BY MR. DAVID) Do you agree with that?
MS. BORELLI: Objection; form.
THE WITNESS: So this is, I think, very complicated. Because for me, I don't really understand why someone would be utilizing a medical code if someone came to them and they needed an intervention that was creating -- that -- whatever they were experiencing requiring that intervention was causing distress.
There's only two reasons that we would need a code like this. It would be if someone was seeking interventions because something was creating problems for them, or if you needed a

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code that would uncouple the gender marker from different various and assorted primary care screening things.
So those are the only two reasons that I think someone would need to use a medical code.
Q. (BY MR. DAVID) Okay. So -- and again, I understand that you're not using the ICD-11, but it could be as simple as a transgender woman needing some sort of a diagnostic code for a typical exam associated with someone assigned male at birth, such as a prostate exam or something like that, and needing to be able to reconcile the typically stereotypically gendered diagnostic codes?
MS. BORELLI: Objection; form.
THE WITNESS: Yes. So, exactly. Or similarly, if a person with a cervix needed a cervical screening. You would not want that to get fouled up in the system of coverage and billing or someone get denied that service related to their gender marker.
Q. (BY MR. DAVID) Is that your understanding based off of research that you've done? Or is that just logically you're thinking through it and that would be what makes sense?

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A. I mean, I have patients who some of their care has been denied because of that reason.
Q. And so do you know the specific reason for the change in the classification under ICD-11?
MS. BORELLI: Objection; form.
THE WITNESS: No.
MR. DAVID: Do you all want to take a break before I move to a completely different topic?
MS. BORELLI: Lets do that.
THE WITNESS: Sure. It's our lunchtime.
MR. DAVID: Oh, we can take a longer break if you'd like so that you can get something to eat.
THE WITNESS: That would be great.
MS. BORELLI: How long should we break? Would you like to break 20, 30 minutes, Dr. Olson-Kennedy?
THE WITNESS: How about 30 minutes?
MS. BORELLI: That sounds fantastic. So let's come back around 12:35 Pacific time.
THE VIDEOGRAPHER: We are going off the record. The time is 12:06 p.m.
(Break taken from 12:06 p.m. to 12:40 p.m.)
THE VIDEOGRAPHER: We are back on the

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record. The time is 12:40 p.m.
Q. (BY MR. DAVID) Doctor, before we went on a break, I was getting ready to start asking you some specific questions about surgical procedures. And because it's been a while, I'm probably going to re-ask some questions, unfortunately.
But to understand, you do make referrals for patients to have surgical consultations; is that correct?
A. Yes.
MS. BORELLI: Objection; form.
Q. (BY MR. DAVID) And when you make a referral for a surgical consultation, are you -- is your referral specific to a type of surgery? Or is it to refer the patient to discuss surgery with a surgeon?
MS. BORELLI: Objection; form.
THE WITNESS: When I make referrals, it's for consultation with a surgeon.
Q. (BY MR. DAVID) And again, bad question. Is it -- do you make specific referrals and say, "I'm making a referral to a surgeon for consultation for chest surgery"?
A. Yes.
MS. BORELLI: Objection; form.

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THE WITNESS: Yes.
Q. (BY MR. DAVID) Okay. So prior to the referral being made to a surgeon, you and the patient have discussed what type of surgery is going to -- is recommended; is that correct?
MS. BORELLI: Objection; form.
THE WITNESS: Well, there are things that I'm not -- it's not within my wheelhouse to make recommendations around, such as the type of approach or the specific way that the surgery is going to be done. But I can make broad categorizations about, like, masculinizing chest surgery, for example.
Q. (BY MR. DAVID) Okay. And so when you are making that referral for surgical consultation, is there -- there's already an idea of what general type of surgery is going to be done such as a chest masculinization surgery; is that right?
MS. BORELLI: Objection; form.
THE WITNESS: For the most part regarding chest surgery, that's true. Sometimes regarding other surgeries, that's not true. But for this particular surgery, yes.
Q. (BY MR. DAVID) Okay. So what other type

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of surgery is that not true for?
MS. BORELLI: Objection; form.
THE WITNESS: So for example, with genital surgery for transmasculine people, there are a number of different surgical things that people might be suited best for. And those are conversations that the patient should have with the surgeon so that they can go over all of the different types of interventions that are available.
Q. (BY MR. DAVID) So in the event of a genital surgery, you may refer a patient to a surgeon for consultation, but there isn't a full understanding at that point of what the -- what genital surgery is actually going to be performed; is that fair to say?
MS. BORELLI: Objection; form.
THE WITNESS: So before -- I mean, I definitely tell people about all of the surgeries that are available. And I think that the details of the ins and outs of those surgeries, recovery times, for example, what things will be able to come from those surgeries, what things won't, are gone over in more detail by the surgeon.
It's pretty rare that somebody doesn't

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know what they want, but because a lot of times there isn't, maybe, information available, it's really critical that they get that information from the surgeon.
Q. (BY MR. DAVID) Do you recommend patients or refer patients -- excuse me.
Do you refer patients for surgical consultations for facial feminization surgeries?
MS. BORELLI: Objection; form.
THE WITNESS: Yes, I have done that in the past. Not as often as the others.
Q. (BY MR. DAVID) Can that involve reshaping -- well, when you have made that referral for surgical consultation for facial feminization in the past, is it making a referral for facial feminization and then the surgeon talks about the particulars of the procedure? Or are you speaking with the patient about specific facial traits that they are desirous of changing?
MS. BORELLI: Objection; form.
THE WITNESS: It could be either of those things. So again, pulsing back to individual things -- so for example, if somebody wants information on this thing or "This part of my face is creating a lot of difficulty for me and it's

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creating a safety hazard for me," for example, then I'm not very versed at all in the different procedures around the face and the skull. So that would be something that I would look to my surgical colleagues to run through with the patient themselves.
Q. (BY MR. DAVID) When you say that some aspect of the person's face can be creating a safety hazard, what do you mean by that?
MS. BORELLI: Objection; form.
THE WITNESS: So what I mean by that is that when certain things happen to the face in a puberty that is dominated by testosterone if someone goes through an endogenous male puberty, there are things that develop that get that person potentially perceived as a trans woman, and trans women are targeted for the things that we previously talked about earlier in the day.
Q. (BY MR. DAVID) So have you had patients where you have specifically discussed referral to a surgeon for a chin reshaping?
MS. BORELLI: Objection; form.
THE WITNESS: Again, I -- that referral is much less common for me, so I'd have to, like, go back in and see if there was specifically a

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patient like that. But that's often a complaint that I hear a lot from people.
Q. (BY MR. DAVID) Are there specific aspects of facial feminization that you know that you have referred a patient for surgical consultation for?
MS. BORELLI: Objection; form.
THE WITNESS: Let me try to go back through my -- I mean, it really has not been a lot, but let me just go back and think about some of the patients.
Certainly, I think, brow is a big one. There are these places on the face that are different. So that there's, like, a wider gap between the bottom of the lip and the bottom of the chin in people who went through testosterone surgery.
The brow is more forward set in people who -- generally in men or people who have gone through a testosterone puberty that makes the eyes appear to be set back more.
Obviously Adam's apple is something that happens from testosterone.
Those are some of the things that I've talked about with patients around referrals for

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surgery.
Q. (BY MR. DAVID) And in those cases, how is it determined whether that's -- you mentioned brow, so I'll take that.
In those cases, do you make the determination as to whether a surgery to feminize the brow is medically necessary?
MS. BORELLI: Objection; form.
THE WITNESS: No. That decision is coming from the surgeon.
Q. (BY MR. DAVID) So is your referral, then, to allow for the surgeon to make a determination as to whether the surgeon believes it's medically necessary to feminize the patient's brow?
MS. BORELLI: Objection; form.
THE WITNESS: I think -- I mean, it's a bit more complicated. That feels black and white. It's not quite that black and white. I don't refer people for surgery unless I think it's medically necessary, but I'm not the ultimate determiner of that.
So my letter that accompanies that request for consultation puts the pieces of knowledge that I have from knowing that person

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into it to say, "These are the reasons that I think this surgery is medically necessary," but I'm not ultimately the person that is putting the request for the procedure in to the third-party payer.
Q. (BY MR. DAVID) And I think that that's a very fair clarification.
You don't order surgeons to perform surgery, correct?
MS. BORELLI: Objection; form.
THE WITNESS: Absolutely not.
Q. (BY MR. DAVID) Okay. So you can make a -- you can draft one of these referral letters, and the surgeon can say, "I'm not doing that"; is that fair?
MS. BORELLI: Objection; form.
THE WITNESS: Yes, that's correct.
Q. (BY MR. DAVID) Okay. But if you are writing a referral letter, then at least to you, you believe that it is medically necessary for the patient to undergo whatever specific type or general type of surgery you have put in that referral letter?
MS. BORELLI: Objection; form.
THE WITNESS: Yes.

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Q. (BY MR. DAVID) Okay. Have you made referrals for surgical consultation for transfeminine people who have developed small breasts as a result of hormone therapy and would like to have larger breasts?
MS. BORELLI: Objection; form.
THE WITNESS: I have only done that on two occasions. And it was -- they both were patients who, despite many -- more than five years of hormone therapy, had not progressed beyond about Tanner Stage 2, which is the very, very earliest stage of breast development, or maybe early Tanner Stage 3. But no, that's not something that I've done on a routine basis.
Q. (BY MR. DAVID) Is there a -- you mentioned Tanner Stage 2 and Tanner Stage 3.
First, can you explain what Tanner Stage 2 and Tanner Stage 3 means?
MS. BORELLI: Objection; form.
THE WITNESS: Sure. So the Tanner stages are named after Tanner because he's the person that wrote it down. But they describe sexual maturity rating so that everybody -- prepubertally everybody is at Tanner Stage 1 of development. And then Tanner Stage 2 is an indicator of the

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first stage of puberty.
So for -- when we're talking about breast development, Tanner Stage 2 is basically designated as when there is a difference between the nipple and the areola from the flat chest contour, from the flat chest wall.
And then Tanner Stage 3 -- and again, these aren't absolutes. These are fuzzier categories. But Tanner Stage 3 is when there's breast tissue that's differentiated from the nipple and the areola and from the flat chest contour.
And Tanner Stage 4 and 5 are a little bit more nebulous. Tanner Stage 5 is really like considered an adult breast shape. That's breast development.
Q. (BY MR. DAVID) Okay. And so in your transfeminine patients, is there a level of breast development in the terms of the Tanner stages where it would no longer be medically necessary for a breast augmentation?
MS. BORELLI: Objection; form.
THE WITNESS: I don't know if there's an exact answer to that. But I will say that two people that I sent for referrals have very, very

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minimal breast development, which is not uncommon for people who start hormone therapy later.
I work with young people, though, and younger people have more hormone receptors, so they generally tend to get better breast development because they're starting younger.
Q. (BY MR. DAVID) So in those two individuals, what specifically about their chest appearance made it medically necessary for them to have a surgical consultation for breast augmentation?
MS. BORELLI: Objection; form.
THE WITNESS: So similarly to a cisgender woman who did not develop breasts beyond Tanner Stage 2, their chest is not identifiable as an adult female chest. And that creates a lot of havoc for anyone who identifies as a woman. If a cisgender woman had a similar situation, I would also refer them for that procedure.
Q. (BY MR. DAVID) So if a cisgender woman had breast development that did not go beyond Tanner Stage 2, it would also be medically necessary for that woman to obtain breast augmentation to have female or stereotypically female or a feminizing chest appearance?

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MS. BORELLI: Objection; form.
THE WITNESS: If it was causing them distress, yes.
Q. (BY MR. DAVID) What is the medical or -- what is the medical condition for that for a cisgender woman?
MS. BORELLI: Objection; form.
THE WITNESS: I think it's called hypomastia. I have to double check on that, but I think that's technically what it's called.
Q. (BY MR. DAVID) I guess that makes sense to me that it would be hypomastia, yeah. Okay.
Do you have any knowledge one way or another whether West Virginia Medicaid covers surgery for hypomastia in cisgendered women?
MS. BORELLI: Objection; form.
THE WITNESS: No.
Q. (BY MR. DAVID) Do you know whether hypomastia requires clinically significant distress to meet the diagnostic criteria?
MS. BORELLI: Objection; form.
THE WITNESS: No.
Q. (BY MR. DAVID) I may have asked you this earlier, and if I did, I apologize.
Do you have patients that -- and since

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we've been talking about chest surgery, I'll use that as the leading example.
Do you have patients who have a diagnosis of gender dysphoria and want a chest surgery but not a genital surgery?
MS. BORELLI: Objection; form.
THE WITNESS: Are you talking specifically about, like, a transmasculine person or a transfeminine person or --
Q. (BY MR. DAVID) Let's say a transmasculine person.
MS. BORELLI: Same objection.
Q. (BY MR. DAVID) Do you have transmasculine patients who desire a chest surgery but not a genital surgery?
MS. BORELLI: Objection; form.
THE WITNESS: I do. And I want to add some clarification here, because the procedures that we have available to us right now for phalloplasty -- again, like I was talking about earlier, that's a really complex surgery, and it requires a lot of postoperative care and time.
And so I don't know that it's -- there are patients that say, you know, "I need chest surgery. I don't feel that way about my

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genitals."
But there's also, I know, people who say, "I need chest surgery, and if I could wake up and have a penis and testes and a scrotum, I would want that." But the mechanics of the procedure or the time or the things around it don't make it feasible.
So I think there's two different questions. That's why I'm adding that clarification.
Q. (BY MR. DAVID) Sure. And I appreciate the clarification.
So there are -- you have patients who are transmasculine who desire both a chest surgery and a genital surgery but do not have the capability to undergo genital surgery because of the significance of the postoperative care necessary and the support needed for the postoperative care for a genital surgery; is that correct?
MS. BORELLI: Objection; form.
THE WITNESS: Yes. And I think, like, the thing that is really important about the clarity is not that people want surgery; they want that body configuration, right? So that is a huge differentiation there, right? That it's one thing

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to -- I mean, nobody wants surgery. People don't want surgery. That's not a thing. They want the results of the surgery. So that's really important when we're talking about what people desire, what people want.
Q. (BY MR. DAVID) And so I'll rephrase the question.
So you have transmasculine patients who desire a masculine body configuration of the chest and of the genitalia, but are incapable of meeting the requirements of aftercare for genital surgery; is that correct?
MS. BORELLI: Objection; form.
THE WITNESS: I think, yeah, just for clarity, maybe it's not the right time for them or maybe they're in the middle of college or they don't have someone that can do aftercare for them. So there are -- yes, there are logistical issues that get in the way of them undergoing those procedures.
Q. (BY MR. DAVID) Do you also have transmasculine patients who desire a body configuration of a masculine chest but not a -- but not masculine genitals?
MS. BORELLI: Objection; form.

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THE WITNESS: Yes.
Q. (BY MR. DAVID) Do those patients express that they do not experience distress from their genitals?
MS. BORELLI: Objection; form.
THE WITNESS: Well, in our conversations, they -- the conversations don't -- first of all, again, everybody's different. But even in conversations around where people are -- where their physical or physiologic distress is coming from, it's very hard to separate out the logistical things that we're talking about from people's experience of their genitals.
But there are people who say, "I enjoy my genitals. I like how they are. I like how they function. I have no desire to change them."
Q. (BY MR. DAVID) Do you recommend or -- let me start that again.
Do you refer patients for surgical consultation for non-binary surgeries?
MS. BORELLI: Objection; form.
THE WITNESS: I don't know what a non-binary surgery is, but I have referred people who are non-binary for surgeries.
Q. (BY MR. DAVID) Okay. Have you ever

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referred a patient for surgical consultation for a penis-preserving phalloplasty? Or excuse me, a --
MS. BORELLI: Objection --
Q. (BY MR. DAVID) -- a penis-preserving vaginoplasty?
MS. BORELLI: Objection; form.
THE WITNESS: Yeah, that other thing would just be greedy.
But no, I haven't.
Q. (BY MR. DAVID) Okay. Have you ever referred a patient for surgical consultation for a nullification surgery?
MS. BORELLI: Objection; form.
THE WITNESS: I don't know what that is.
Q. (BY MR. DAVID) My understanding from reading literature is there are surgeries that maintain an intact urethra but would render the person without external genitalia.
A. Well, I've never had anyone ask for that. I have not referred anyone for that.
Q. Okay.
MS. BORELLI: And I'm sorry, I just want to interpose a late objection. I wasn't sure if there was a question pending, but to the extent that was a question, objection to the form.

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MR. DAVID: Sure.
Q. (BY MR. DAVID) When you refer patients for surgical consultation for transmasculine chest surgery, do you discuss with the patient alteration of nipple placement?
MS. BORELLI: Object to the form.
THE WITNESS: Yes. Sometimes when I'm reviewing the different kinds of surgeries that there are available for chest surgery, I will talk about nipple regrafting and resizing.
Q. (BY MR. DAVID) And is the alteration of nipple placement, nipple resizing, is that medically necessary for the patient who is -- you're referring for the surgery?
MS. BORELLI: Objection; form.
THE WITNESS: Well, the decision around the replacement of the nipples is determined by the surgeon. But there are typical ways that the surgeries are done that necessitate nipple resizing and replacement. So that's kind of what I'm talking about.
Q. (BY MR. DAVID) Okay. So in the event of a mammaplasty, is there a -- for lack of a better word, a surgical need for nipple resizing and nipple grafting?

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MS. BORELLI: Objection; form.
THE WITNESS: When somebody undergoes what is a bilateral incision procedure, yes, the nipples have to be removed and regrafted because of the way that the surgery is done.
Q. (BY MR. DAVID) And so I don't remember which one I asked you. Did I ask you about mammaplasty?
A. Yeah, you asked about mammaplasty, but I haven't really heard people use that language around this procedure.
But when people have a double-incision chest surgery, a chest procedure, they, just by nature of the way that the incisions are made, they have to have regrafting of the nipples, to be removed and regrafted.
Q. So whether it's for a chest feminization surgery or a chest masculinization surgery, nipple replacement and grafting is something that's done within the surgery as a matter of course?
MS. BORELLI: Objection; form.
THE WITNESS: For feminizing surgeries? I don't have an answer to that. I think probably because I refer much less often for that.
But within the context of chest

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masculinization, yes.
Q. (BY MR. DAVID) And I don't know if this would be something that would happen in your work at the transyouth center, but maybe it's something you come across at the hospital.
We talked a little bit about hypomastia. Do you treat patients with gynecomastia?
MS. BORELLI: Objection; form.
THE WITNESS: I have in the past, but that's not something that I commonly see or take care of within the scope of my practice.
Q. (BY MR. DAVID) Just to probably close that line of questioning, then, have you ever referred a patient for surgical consultation for gynecomastia?
MS. BORELLI: Objection; form.
THE WITNESS: One time. I think one time. But it's been a while.
Q. (BY MR. DAVID) When -- well, let me ask you, are there criteria that need to be met prior to referring a patient for surgical consultation for gynecomastia?
MS. BORELLI: Objection; form.
THE WITNESS: That's a great question. I don't know.

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Q. (BY MR. DAVID) Okay. Was the patient with gynecomastia expressing clinically significant distress? Is that the reason that the patient was referred for surgical consultation?
MS. BORELLI: Objection; form.
THE WITNESS: The patient had distress about the way that their chest looked, yes.
Q. (BY MR. DAVID) Did the patient experience any physical pain as a result of gynecomastia?
MS. BORELLI: Objection; form.
THE WITNESS: I don't remember. It's been more than ten years.
Q. (BY MR. DAVID) Okay. Are there presentations of gynecomastia that do cause physical pain?
MS. BORELLI: Objection; form.
THE WITNESS: I don't know the answer to that.
Q. (BY MR. DAVID) Okay. I want to go back to -- there was a brief discussion about Tanner Stage 2 and Tanner Stage 3 and the different Tanner stages.
At what age do patients generally enter Tanner Stage 2?

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MS. BORELLI: Objection; form.
THE WITNESS: So the beginning of puberty is a range for people with ovaries and people with testes. So in -- let's talk about cisgender kids because it's a little easier.
In cisgender girls, people with ovaries, they start their puberty sometime between 8 and 14. That's the typical age range. Outside of those age ranges we start to consider why they might be too early or too late.
For people with testes, their timeline of puberty is 9 to 16 when they would go into their first stages of puberty.
Q. (BY MR. DAVID) So for cisgender females, would anything before age 8 be considered precocious puberty?
MS. BORELLI: Objection; form.
THE WITNESS: Yes.
Q. (BY MR. DAVID) Okay. And if a patient were to begin puberty at age 6, puberty blockers might be used for that patient to allow for puberty to be started at the same time as the patient's peers; is that correct?
MS. BORELLI: Objection; form.
THE WITNESS: So I think that's the

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intent of it. From a pragmatic perspective, somebody would almost always stay on puberty blockers until they were 12. I think that's kind of an average age.
Q. (BY MR. DAVID) So if a patient did have precocious puberty, the normal process is to keep the patient on puberty blockers until age 12 and then take them off of that to allow them to go through their puberty at that time; is that correct?
MS. BORELLI: Objection; form.
THE WITNESS: So just for clarity, I'm not an endocrinologist, so I don't treat precocious puberty, but that's my understanding is they would go on puberty blockers until age 12.
Q. (BY MR. DAVID) Is that age range for -- at least to your understanding, is that age range for taking a patient with precocious puberty off of puberty blockers the same for cisgender boys?
MS. BORELLI: Objection; form.
THE WITNESS: I believe so.
Q. (BY MR. DAVID) Do you have an understanding of why age 12 is the time frame for when they're taken off of puberty blockers?

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MS. BORELLI: Objection; form.
THE WITNESS: I think it's because that's kind of an average age. I don't really know why that's the -- I don't know what the thinking was behind that recommendation, but my guess is that's why.
Q. (BY MR. DAVID) If you can go to your report, page 12, and to paragraph 40, this is where your report starts talking about puberty blockers, correct?
A. Yes.
Q. And is it your opinion that if a patient is started on puberty blockers in Tanner Stage 2, that gives the patient additional time to explore their gender identity prior to going through puberty?
MS. BORELLI: Objection; form.
THE WITNESS: I think in some cases that's absolutely true. That's not everybody's case.
Q. (BY MR. DAVID) Is there a time frame for -- and let me preface this first.
My understanding -- and I'll be honest, I don't remember if it was from your report or one of the other reports, but my understanding is that

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there's at least an opinion that if a patient is started on puberty blockers, that that's -- it's reversible. And once the patient is taken off of puberty blockers, that they can begin puberty as whatever their biological sex would dictate.
Is that a correct understanding of your opinion as to the use of puberty blockers?
MS. BORELLI: Objection; form.
THE WITNESS: I think what you mean is if somebody goes on puberty blockers and then they are not going to go on gender-affirming hormones, that their body will resume endogenous puberty.
Q. (BY MR. DAVID) That's exactly what I mean.
A. Yes.
Q. Okay. Is there an age or a -- my understanding is that once puberty blockers are started, Tanner stages are paused; is that right?
MS. BORELLI: Objection; form.
THE WITNESS: Yeah, progression through their endogenous puberty gets paused.
Q. (BY MR. DAVID) Okay. Is there a point in terms of age that a patient would need to have stopped puberty blockers to be able to go through an endogenous puberty?

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MS. BORELLI: Objection; form.
THE WITNESS: There hasn't been that so far. I mean, there hasn't been -- there are a handful of things that are considered when people are making decisions or we're making recommendations before care. But to date, there hasn't been someone whose wanted to stay on puberty blockers, for example, until they're 19, right? We just haven't had that clinical scenario, and so no. So far, no.
Q. (BY MR. DAVID) I think that that fully answer my questions, because I'm trying to -- if you've never actually seen that happen, then we don't know.
So what is the latest date or latest age that someone has stayed on puberty blockers in your clinical experience?
MS. BORELLI: Objection; form.
THE WITNESS: So I want to have clarification about this because I think people think about puberty blockers and gender-affirming hormones as sequential. But most people when they start gender-affirming hormones, if they start them, they're also going to be on blockers.
So I'm going to assume that what you're

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asking is what's the latest someone has stayed on puberty blockers as mono therapy without any endogenous hormones or gender-affirming hormones?
Q. (BY MR. DAVID) Yes, that is my question.
A. I think 16.
Q. And once that patient -- well, let me ask, did that patient go on to receive gender-affirming hormones?
MS. BORELLI: Objection; form.
THE WITNESS: That patient did.
Q. (BY MR. DAVID) Okay. Have you had a patient start on puberty blockers without gender-affirming hormones and stop puberty blockers and not go on to gender-affirming hormones as late as age 16?
MS. BORELLI: Objection; form.
THE WITNESS: I don't think so.
Q. (BY MR. DAVID) Have you had that situation happen with someone as late as age 16?
MS. BORELLI: Objection; form.
THE WITNESS: That is very hard. I've taken care of so many people, it would be very hard for me to know. I have had people go on blockers, then hormones, then stop both and go through their endogenous puberty as late as 15.

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Q. (BY MR. DAVID) Okay. Okay. Well, that works.
So in that instance where you've had someone start puberty blockers and then gender-affirming hormones and then stop both at age 15, did the patient have -- I don't want to use this word, but a normal endogenous puberty?
A. They did, and it was horrible, and then they had chest surgery and went back on hormones.
Q. So you have -- at least in your clinical experience, you have seen patients on puberty blockers as late as age 15, and once those puberty blockers are stopped, they have continued on through endogenous puberty?
MS. BORELLI: Objection; form.
THE WITNESS: I'd have to go to look at the exact ages, but I have had patients in my practice that have been on mono therapy, like their puberty got blocked in early puberty; they've been on puberty blockers for a handful of years, maybe three, four years; and then they stop and they go through their endogenous puberty as we would expect them to, yes.
Q. (BY MR. DAVID) Do you follow the developments in the use of puberty blockers in

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Europe?
MS. BORELLI: Objection; form.
THE WITNESS: Like, more specifically? I'm not sure I know what you're referring to specifically.
Q. (BY MR. DAVID) Sure. So it's my understanding that in Sweden, they have recently issued new -- I don't know if it's guidelines or recommendations, but have recently issued some statement that they do not recommend the use of hormone therapy in individuals under the age of 18.
Are you aware of that?
MS. BORELLI: Objection; form.
THE WITNESS: I'm confused because you asked me about blockers, but this sounds like it's about hormones.
Q. (BY MR. DAVID) And again, I'm sorry. I probably said the wrong term.
Are you familiar with a recommendation from Sweden that patients do not go through hormone therapy until they are at least age 18?
MS. BORELLI: Objection; form.
THE WITNESS: I have heard this, but I don't think I've seen that recommendation

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specifically.
Q. (BY MR. DAVID) And when -- I'm sorry, I'm going back to blockers.
When you are -- well, let me ask, do you yourself actually prescribe puberty blockers?
MS. BORELLI: Objection; form.
THE WITNESS: I do.
Q. (BY MR. DAVID) Okay. When you are considering prescribing puberty blockers to a patient, what does the informed consent process look like?
MS. BORELLI: Objection; form.
THE WITNESS: So the informed consent process includes -- so I can tell you in my practice specifically, I have -- it kind of sounds a little bit ridiculous when I say this, but I have a PowerPoint presentation that I show patients.
And it demonstrates how people's developmental processes occur, so how their Tanner stages occur, what the timeline is for those Tanner stages, and then what your body is doing during puberty, so how your brain is talking to your ovaries or your testes and what it's telling your ovaries and testes to do, and then what the

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gonads are making, the changes that happen in your body subsequently, and then how puberty blockers function and work in the body.
And then I talk about how the puberty blockers are delivered, like, if -- they come as an injection; they come as an implant.
And then we talk about what's going to happen with their body if they -- what their endogenous puberties would look like, where there body's going to get pause when their blockers start.
And then we talk about the impact on bone density. We talk about the potential impact on linear growth, and we talk about some of the other things that are important.
I give time for the patients or the parents or guardians to ask questions. So that's the initial part.
Then I send them a consent form, which has much of the same information on it. And they review it together as a family or with their therapist or whoever they want to bring in to the care team. And then we have another visit and we go through the consent form all line by line and then garner any questions, and then the patient

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and the parent both sign.
Q. (BY MR. DAVID) So there's a lot there. I'll try to unpack a little bit.
So you mentioned this PowerPoint presentation has a lot of information about the effects of the blockers. And one of the things that you said was the effect on bone density.
What is the effect on bone density?
MS. BORELLI: Objection; form.
THE WITNESS: So a lot of people just don't inherently understand how bone density works anyway, regardless if you were on puberty blocker or not.
So bone density is accruing even before you start puberty. But when you start puberty it accrues at a faster rate. So if you put someone on a puberty blocker, your bone density is only going to accrue at its pre-pubertal rate. So it's important that people know that, and it's kind of specific as to how we measure bone density in medicine.
So most people use what's called a DEXA scan, and with the DEXA scan, it gives them a number. But it's a Z score that's in relationship to other people of their same age.

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And so this is one of the reasons at our program where we use a different kind of bone density measurement, so that we can actually keep track of the quantitative number.
And so it is important that people understand that their bone density rate of accrual will most likely go down to the prepubertal rate again, which leads us to our recommendations that people do weightbearing exercise and that they supplement with calcium and vitamin D to try and preserve their bone density.
Q. (BY MR. DAVID) Okay. When a patient is started on gender-affirming hormones, does the patient's bone density accrue at the rate that would happen during endogenous puberty?
MS. BORELLI: Objection; form.
THE WITNESS: Well, that's not a question we can answer because they're not going through their endogenous puberty. So we don't have a comparison.
Q. (BY MR. DAVID) Okay. Are you saying that we don't know what the -- this particular patient's bone density accrual rate would be during endogenous puberty versus a puberty through gender-affirming hormones?

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MS. BORELLI: Objection; form.
THE WITNESS: It gets at the essence of it that there is a range of normal bone density, right, that is measured within -- you know to standard deviations of the normative number.
And what I can tell you is that the patients where we've gotten their bone density after they've been on hormones for some time have bone densities in that normal range.
But what I felt like I heard you asking was, like, is it the same as if they'd gone through endogenous puberty? Well, we don't know. You don't know what it would have been because they didn't go through it.
Q. (BY MR. DAVID) And again, it was an imprecise question on my part.
So if a patient is started on puberty blockers at Tanner Stage 2 and then later on has gender-affirming hormones, once they're through, I guess, a normal period of time for puberty, does that patient then have a statistically normal bone density?
MS. BORELLI: Objection; form.
THE WITNESS: So just one more point of clarification. Do you mean a person that goes

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through an exogenous puberty or a person that goes through their endogenous puberty?
Q. (BY MR. DAVID) I mean comparing the endogenous to the exogenous.
MS. BORELLI: Objection; form.
THE WITNESS: Again, because we can't -- we can get somebody's bone density before they start, before they start blockers. We can get their bone density while they're on blockers, which we do. We get their bone density after they've been on hormones for a while.
But what we see is that if people have been on hormones for a while and we check their bone density, that their bone density is within a normal range.
Q. (BY MR. DAVID) Is there a difference between the bone density of cisgender individuals -- cisgender males and cisgender females following endogenous puberty?
MS. BORELLI: Objection; form.
THE WITNESS: I think that I understand what you're saying. Like do girls and boys have different bone densities?
Q. (BY MR. DAVID) Right.
A. Yes. So they get -- when they read out a

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bone density Z score or comparative score, they will usually, unless we ask them to otherwise, do a Z score for persons with the same assigned sex at birth.
Q. Okay. And so when you are looking at bone density following an exogenous puberty, is the bone density comparable to the bone density of individuals who have gone through endogenous puberty who were assigned the same sex at birth?
MS. BORELLI: Objection; form.
THE WITNESS: Well, it depends which -- so there is a couple things. Remember that bone density is not just dependent on sex steroids. It's also dependent on weightbearing exercise. And again, this is from my clinical practice. So this isn't from a study, yet. It's part of what we're collecting.
So in general, let's just talk in terms of, like, trans girl, right? We have, in our study but also in other studies, at baseline, trans girls have lower bone density than cisgender boys. And there is speculation to why that is, but it may be related to lack of rough and tumble play. I mean, like, a lot of times kids feel very concerned about doing sports or physical activity

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because they're nervous about muscle development and things like that.
But anyway, across the board, they already have lower bone density, and this is before blockers, before any treatment. And so it's when they have gone through an exogenous puberty, feminizing hormones, their bone density is in the normal range, but they're getting measured on a girl's -- especially -- that's not always true. It depends on their gender marker, right, whether they get measured on a boy's or a girl's reference value.
But it's different for people who take testosterone, because in general their bone density is higher than it would have been otherwise. Because A, a lot of those kids are more active than their cisgender female peers, and so there's a lot that goes into what that means.
The concern for this work, the medical concern, is whether or not someone has osteoporosis. And in my practice, I've never had anyone with osteoporosis, regardless of their gender, regardless of their time on blockers, regardless of their use of gender-affirming hormones. And that is probably the most critical

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piece of this work is we do not want people to have osteoporosis, and we haven't had that happen.
Q. (BY MR. DAVID) Is there any research that you're aware of that has determined whether transgender individuals have a higher incidence of osteoporosis in their 40s, 50s, 60s, 70s, than cisgender individuals?
A. There might be. Not that I know of offhand.
Q. Okay. Something else that you mentioned that's a part of your PowerPoint presentation is linear growth.
Is that meaning simply height?
A. Yes.
Q. Okay. So is there a discussion that once puberty blockers are started, the rate of growth will slow? Is that what happens?
MS. BORELLI: Objection; form.
THE WITNESS: So it will be slower than it would have been if they had been going through endogenous puberty.
Q. (BY MR. DAVID) Okay. Something else that you mentioned was that puberty blockers could be administered through -- I think you said injection or implantation; is that right?

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A. That's correct.
Q. Okay. I assume that there aren't any pill forms; is that right?
A. Essentially blockers only come as injections. I'm not sure if there's a formulation anymore, but there used to be an intranasal form. But I don't -- it's not really routinely used, so I don't really even know if people do use that.
But central blockers only come as injectable or as the implant that gets placed in the arm.
Q. Can you describe to me what the implant is?
MS. BORELLI: Objection; form.
THE WITNESS: Sure. It's a little, flexible rod. And my understanding is that the medication is placed in layers so that it distributes a small amount every day.
Q. (BY MR. DAVID) Is that similar to an IUD?
MS. BORELLI: Objection; form.
THE WITNESS: No. An IUD gets placed through the cervix into the uterus. But it's similar to a Nexplanon, which is the same kind of implant that secretes a different kind of a

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hormone that also gets placed in the arm.
Q. (BY MR. DAVID) And I meant in terms of it being similar to an IUD, is it similar in terms of its release of medication in a -- I guess a daily or in some sort of a time frame over time?
MS. BORELLI: Objection; form.
THE WITNESS: Yeah. I mean, anything that is a sustained-release mechanism, from a pharmacologic perspective, is similar.
But it's also important to note that not all IUDs have medicine in them. There is another type of an IUD that doesn't do that. But essentially, yes, they're something that is putting out a little bit of medicine each day.
Q. (BY MR. DAVID) How long can the implant remain in someone's arm and continue to produce or to emit -- I don't know the proper word, I'm sorry -- but continue to provide medication?
MS. BORELLI: Objection; form.
THE WITNESS: So there's -- let me start by saying there's nothing -- let me start by saying this: It's marketed for a year. The pharmaceutical company that makes the implant markets it for a year. That's what they did the FDA trials for. We know, though, that they last

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longer for some people than a year.
Q. (BY MR. DAVID) Who is the company that makes the implant?
MS. BORELLI: Objection; form.
THE WITNESS: Endo Pharmaceuticals.
Q. (BY MR. DAVID) You mentioned that there are at least two ways or methods for measuring bone density; is that right?
MS. BORELLI: Objection; form.
THE WITNESS: Yes.
Q. (BY MR. DAVID) And at your center, you use -- I'm sorry, I don't remember the name of it.
There was one thing -- I think you said it was DEXA scan?
A. Yes.
Q. Okay. That's the one that you do not use, correct?
A. We do DEXA scans at Children's Hospital Los Angeles. But for my kids, I utilize a different mechanism called quantitative CT scan that gives an actual measurement rather than a Z score.
Q. So is that a CT scan, and you're able to determine, I guess, bone width and bone height?
MS. BORELLI: Objection; form.

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THE WITNESS: Well, it's specifically around bone density. So it's not measuring, like, the length or the circumference of bones. I guess is that what you're asking?

Q. (BY MR. DAVID) Yeah. So how does it measure the density I guess is my actual question.

A. I have no idea.

Q. Okay. In the study that you're doing with the NIH grant, you mentioned that bone density is something that's being measured.

Is that using the qualitative CT scan or the DEXA scan?

MS. BORELLI: Objection; form.

THE WITNESS: So it's quantitative, just so you know. But we're using both. So at my center, it's the quantitative CT scans; in other centers it's DEXA scans.

And there are certain times when patient's insurance doesn't want to -- they want someone to get a bone density scan at a different place, and so they might get a DEXA scan because of that reason.

Q. (BY MR. DAVID) Okay. So in the event that a patient goes to your center, builds a relationship with you, you go through this

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PowerPoint presentation with the patient; they're sent home with a consent form and they go over it with their family; they come back for another visit, and the patient does not feel comfortable starting puberty blockers at that time, are puberty blockers still medically necessary for that patient?

MS. BORELLI: Objection; form.

THE WITNESS: I think that that's more complicated than a yes-or-no answer. The -- and I can tell you from the experiences that I've had that the reason that would happen is if somebody had -- and I've talked a lot of people through this too -- have anxiety about what the implantation process is, that they have anxiety about injections.

If -- so it's not related to -- it's related to over -- when that situation happens, it's about fear that we have to help patients mitigate in order for them to get their blockers going.

So the necessity piece doesn't change. It's the -- whatever is happening for that young person around their experience or what they are anticipating the experience to be like.

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Q. (BY MR. DAVID) Have you had that situation occur?

MS. BORELLI: Objection; form.

THE WITNESS: Do you mean the one where someone's nervous about the -- yeah, I have.

Q. (BY MR. DAVID) And in that situation, I mean, can you walk me through generally what happens next?

MS. BORELLI: Objection; form.

THE WITNESS: Well, it's not always the same. I mean, this has happened on a couple of occasions. So we've addressed this in various and assorted ways.

So if somebody needs -- for example, there have been people who might -- they might get, like, laughing gas to get their implant placed so they don't have to be super conscious of it.

We may do additional things like, okay, let's have you bring in your iPad or bring in your phone and distract you while the procedure is taking place. So various and assorted ways.

There is a couple times where I've walked over to where the implants are being placed and kind of distracted someone and helped talk them

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through it. And so various and assorted things.

Sometimes people will go through, like, needle desensitization, which I don't have a lot of familiarity with, but if they're really struggling around needles, that might be something that we do to help them move through that.

Q. (BY MR. DAVID) So in those situations, the patient still desires the effects of pubertal suppression, but needs some other way to, I guess, distract them from the procedure itself? And is that correct?

MS. BORELLI: Objection; form.

THE WITNESS: Yes. That's been my experience of it. It goes back to a similar thing that we were talking about, that nobody wants surgery, right? Nobody wants something placed in their arm, but they want what comes from it, or the downstream impact.

Q. (BY MR. DAVID) Okay. If we can go back to your report on page 15.

And starting at paragraph 45, you discuss gender-affirming surgeries, correct?

A. Yes.

Q. Okay. And I'll read the paragraph 45. "Gender affirming surgeries: Some transgender

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individuals need surgical interventions to help bring their phenotype into alignment with their gender. Surgical interventions may include vaginoplasty, tracheal shave, liposuction, breast implants, and orchiectomy for transfeminine individuals, and chest reconstruction, hysterectomy, oophorectomy, salpingectomy, construction of neo-scrotum, and metoidioplasty or phalloplasty for transmasculine individuals."

I may have mispronounced some words, but did I read that correctly?

A. Yes.

Q. Okay. All of those surgical interventions that were listed in paragraph 45, have you made referrals to -- for surgical consultations for each of those surgeries?

MS. BORELLI: Objection; form.

THE WITNESS: I think the only one that I haven't is the liposuction. But everything else I think that I have sent people for.

Q. (BY MR. DAVID) Okay. So are there times that liposuction would be a needed surgical intervention to help bring someone's phenotype into alignment with their gender?

MS. BORELLI: Objection; form.

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THE WITNESS: I think there are. I haven't seen them. That may be a manifestation of the fact that I see younger clients. But this is something that people do talk about.

Q. (BY MR. DAVID) So the statement specific to liposuction, is that based upon your knowledge of literature or conversations with colleagues?

MS. BORELLI: Objection; form.

THE WITNESS: Yeah. So when I read literature about gender-affirming surgeries, it's part of it. I couldn't point to a very specific one right now, but yes.

Q. (BY MR. DAVID) Okay. Can you explain to me what a tracheal shave is?

MS. BORELLI: Objection; form.

THE WITNESS: A tracheal shave is a procedure in which the Adam's apple is reduced.

Q. (BY MR. DAVID) Does that have any effect on a patient's voice octave? I'll just leave it there.

Does that have any effect on the patient's voice?

MS. BORELLI: Objection; form.

THE WITNESS: No, I don't believe so. Again, not my area of expertise. I would defer to

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a surgeon for that. But my understanding is no, that's something to create a feminine neck contour.

Q. (BY MR. DAVID) I want to look at paragraph 46. And we'll break this down into separate sections here. But I'll read the first sentence.

"The current WPATH standards of care recommend that genital surgery -- i.e. surgery which may render the individual sterile -- not be carried out until the individual reaches the legal age of majority to give consent for medical procedures, while acknowledging that care is individualized."

First, did I read that correctly?

A. Yes.

Q. Okay. When you state that "while acknowledging that care is individualized," is that to say there may be certain circumstances where genital surgery is recommended under the age of majority?

MS. BORELLI: Objection; form.

THE WITNESS: Yes, possibly.

Q. (BY MR. DAVID) Under what circumstances would a genital surgery be recommended in a

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patient under the age of majority?

MS. BORELLI: Objection; form.

THE WITNESS: So if I can, I'll tell you some of the examples that I'm thinking of.

You know, I have a patient who is accepted to college overseas. And it just so happens that her 18th birthday is at the end of August. And so for her, in order to get surgery at age 18 or over, she is going to have to delay her entrance to college. This is incredibly disruptive to her life, when she could have had that surgery done three or four months before in summer before the school year starts. So this is really disruptive to her life for a pretty arbitrary reason. That's an example.

For her to have surgery the summer before she goes to college, she will be at home; she can do her recovery at home, and it won't disrupt her academic trajectory. So that would be an example.

Q. (BY MR. DAVID) So outside of, sort of, logistical concerns like that where the patient is not going to be in a position to have support through after care or there's a major life event like college coming up, are there times when a patient's distress is so significant that it

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requires surgery --
MS. BORELLI: Objection; form.
Q. (BY MR. DAVID) -- before the age of majority?
MS. BORELLI: Same objection.
THE WITNESS: So I -- my experience is that the distress is very severe. For the trans young women that I've had in my practice that have been transitioned for years and years, they are really left out of the things that young people do. They're navigating high school as girls with the wrong genitals.
And I really think that that is not something anyone can understand who is not in that situation. And the amount of distress that it causes for people is really quite overwhelming.
And I do have patients who transitioned young. They had puberty blockers; they went on to hormones; they are living their lives as themselves, and they aren't dating; they aren't doing sports; they're not hanging out with other cisgender girls who are their friends because of this, entirely because of this. It's really an enormous amount of distress.
Q. (BY MR. DAVID) Is there some way to

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quantify the distress that your patients are experiencing as a result of their genitals?
MS. BORELLI: Objection; form.
THE WITNESS: So very similar to chest dysphoria, we have talked about and considered creating a genital dysphoria scale or some kind of measure. But there hasn't been one to date that I know of.
So there isn't a way to quantify it in the sense of it's all going to become zeros and ones with means and statistical capacity. But I think there could be. That could be done.
Q. (BY MR. DAVID) So I guess maybe one of the other aspects of the statement that "while acknowledging the care is individualized" is that you have had a long-standing relationship with these patients and understand specifically how that patient is presenting and whether you see that the patient is experiencing more and more distress as time is going on.
Is that something that you're also referring to with that statement?
MS. BORELLI: Objection; form.
THE WITNESS: I think yes, absolutely. And I think it can be confusing to someone looking

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in on the outside saying, "But for all intents and purposes you're walking in the world as a girl; you're being perceived as a girl; why are you struggling so much?" and then sitting down and having a conversation with some of them young women and having them say, you know, "This is not something that is a shared experience for my friends. This is not something I can ask them about," you know.
"I'm not going to prom because of this."
"I'm not" -- you know, "What do I tell my partner? What if I'm dating a boy?" You know, "What do I say? How do I create language around this?"
"What do I tell people," you know, "when we're getting more serious?"
And it's -- it's hard to explain every, like, clinical scenario, but it's so disruptive to people's lives. It's unbelievably disruptive because, you know, high school and early college years are a time when sort of sexuality and sexual activity are at a premium. They're a big part of people's developing lives. And for a large majority of it, these young women are left out of that.

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Q. (BY MR. DAVID) Okay. The next sentence of paragraph 46 says, "In addition, the standards recommend that the other surgical interventions (e.g., chest surgery for transgender males and breast augmentation for transgender females) may occur earlier than the legal age of consent, preferably after ample time living in the desired gender role and after one year of hormone therapy."
First, did I read that correctly?
A. Yes.
Q. Okay. And so that's essentially what we just talked about, that there can be situations where an individual can undergo genital surgery while this talks about specifically chest surgery, but can undergo gender-affirming surgery prior to the age of majority?
MS. BORELLI: Objection; form.
THE WITNESS: I do think there are differences. This specific piece of the standard of care is really referring to a chest surgery.
Let me put it this way: This particular part is referring to not genital surgeries. And so I think that's important, because there is -- right now, the standards of care primarily

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are saying 18 or the age of consent in your country for genital surgeries.
Q. (BY MR. DAVID) And then the next sentence says, "The standards of care, however, further recognize that these are individual determinations and that 'different approaches may be more suitable, depending on an adolescent's specific clinical situation and goals for gender identity expression.'"
First, did I read that correctly?
A. Yes.
Q. And just to tie this up, that's essentially all -- what we were just talking about in terms of you knowing these patients and having a relationship with them and understanding their clinical presentations, correct?
MS. BORELLI: Objection; form.
THE WITNESS: Yes.
MR. DAVID: Lets go ahead and take a break. I'm pretty sure that I'm nearing the end here, but let's take a break and I'll regroup, and hopefully we'll be done soon.
THE WITNESS: Okay.
THE VIDEOGRAPHER: We're going off the record. The time is 2:05 p.m.

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(Break taken from 2:05 p.m. to 2:12 p.m.)
THE VIDEOGRAPHER: We are back on the record. The time is 2:12 p.m.
Q. (BY MR. DAVID) Doctor, earlier, before we took the break, I had asked you about recommendations from Sweden.
And I believe that you'd said that you heard something about it but weren't fully aware of or familiar with the specific recommendation; is that correct?
MS. BORELLI: Objection; form.
THE WITNESS: That's correct.
MR. DAVID: Okay. I'm going to go ahead and share this with everyone. And we'll mark this as Exhibit 7. I've dropped it now into the "Marked Exhibits" folder. And it is titled "Updated Recommendations for Hormone Therapy in Gender Dysphoria in Young People."
(Deposition Exhibit No. 7 was marked.)
Q. (BY MR. DAVID) Do I have that in front of you now?
A. I do.
Q. Okay. And I'll just read the first paragraph. "The National Board of Health and Welfare today publishes new recommendations

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regarding hormone therapy of young people under the age of 18 with gender dysphoria. Uncertain science and newly acquired knowledge mean that the National Board of Health and Welfare now recommends restraint when it comes to hormone therapy. At the same time, it is important that children and young people suffering from gender dysphoria are taken seriously, well treated, and offered accurate care measures."
So first, did I read that correctly?
A. Yes.
Q. Now that you've seen the specific document that I was talking about, were you aware of this?
MS. BORELLI: Objection; form.
THE WITNESS: Again, I think in this stratosphere of gender worlds I was aware of it, but not the specifics of it.
Q. (BY MR. DAVID) Okay. I assume that you disagree with that; is that correct?
MS. BORELLI: Objection; form.
THE WITNESS: I think that I would have to read the background, because I think that there's some fuzzy language. Like, what does "restraint" mean? Is there an assumption that

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people aren't doing this work with the level of, you know, detail that they would like people to be doing it with?
I don't really see anything in here about puberty blockers.
So I think it's -- I would just need more background to understand what they mean. Like, I think people should always practice with thoughtfulness.
Q. (BY MR. DAVID) Sure. If I gave you the opportunity to read -- it's only two pages, but this two-page document, would that be enough information? Or would you need additional understanding of the background that led to this outside of that two pages?
MS. BORELLI: Objection; form.
THE WITNESS: So just for clarity, you mean to say if I agree with it or -- I mean, I can read the two-page paper and tell you if I agree with it or not, but it seems like there probably was a little bit more than just what was presented here that went into their recommendation.
Q. (BY MR. DAVID) Okay. If you don't mind to go ahead and read the two-page paper and tell me what your thoughts were.

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A. Sure.
There's -- there's something wrong with the bottom of the first page.
Do you see that?
Q. Yeah. I can tell you what I believe that that says. It says, "currently outweigh the possible benefit for the group as a whole."
But if you can't read it, I understand.
A. I can read that part, but then is that the same -- the top of the second page the same as the bottom of the first?
Q. That's my belief is that --
A. Okay. Okay. Yeah, that makes sense. Okay.
So after reading this, one of the things that I think is really important in this, when I think of -- when I look at this document, what it says to me is, like, people should be doing this work with care.
And, I mean, hormonal treatments where they are deemed justified, I don't really know who's recommending hormone treatments when they're not justified. That's frustrating for me.
This isn't necessarily a physician paper on a change. It just feels like it's calling

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attention to, you know -- I don't know. Is there a perception that the work wasn't being done with caution? But to me what they're recommending is something that certainly in our practice we do but I hope that people providing these services overseas are doing as well, which is, you know, justified medical interventions, you know.
And I don't think anybody practices this work without, you know, psychosocial, psychiatric, mental health therapy as a part of that practice.
So it's -- it feels a little soft to me. So if they came out with a hard recommendation and said, "No one should have hormones before age 18," I would disagree with that. But it doesn't really seem like that's what they're saying.
Q. So you just -- I believe you just said that you're not aware of anyone who is recommending treatment without mental health assessments being done; is that right?
MS. BORELLI: Objection; form.
THE WITNESS: No. I don't know anyone who is not doing this work thoughtfully. And I don't know anyone who's recommending hormones that aren't -- what is the language that they used? Justified.

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Q. (BY MR. DAVID) Okay. So you were saying that in your practice, at least, and your knowledge of other people's practices, they're already meeting the criteria or the recommendation that's listed in this document because they are going through a process to ensure that it is justified that patients are receiving these hormones under the age of 18?
MS. BORELLI: Objection; form.
THE WITNESS: Yes.
Q. (BY MR. DAVID) Okay. Have you done any research to determine what brought on this recommendation from Sweden?
MS. BORELLI: Objection; form.
THE WITNESS: No.
MR. DAVID: Okay. I wanted to ask you some questions about an article that you were one of the coauthors of titled "Physiologic Response to Gender-Affirming Hormones Among Transgender Youth."
And I will put that into the "Marked Exhibits" folder as Exhibit 8.
(Deposition Exhibit No. 8 was marked.)
Q. (BY MR. DAVID) That should be there now.
A. Yes.

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Q. Okay. And the -- in the abstract, it says that "The purpose of this study was to examine the physiologic impact of hormones on youth with gender dysphoria. These data represent follow-up data in youth ages 12 to 23 years over a two-year time period of hormone administration."
First, did I read that correctly?
A. Yes.
Q. Okay. Well, let me ask this first: Was this developed out of your research from the NIH grant?
MS. BORELLI: Objection; form.
THE WITNESS: Yeah. Oh, sorry.
MS. BORELLI: That's okay. Objection; form.
THE WITNESS: No. This was a study that I did to collect preliminary data for that NIH application.
Q. (BY MR. DAVID) And the method -- it says, "This prospective, longitudinal study initially enrolled 101 youth with gender dysphoria at baseline from those presenting consecutively for care between February 2011 and June 2013. Physiologic data at baseline follow-up were abstracted from medical charts. Data were

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analyzed by descriptive statistics."
First, did I read that correctly?
A. Yes.
Q. Okay. The physiologic data at baseline follow-up were abstracted from medical charts.
My question is, are these your patients that you were treating or are these different patients from other centers that you were following?
MS. BORELLI: Objection; form.
THE WITNESS: These were all patients that were receiving services at our center.
Q. (BY MR. DAVID) Okay.
A. Just for clarity, they weren't all my personal patients, but they were all patients of our center that one of our providers was following.
Q. Okay. And now looking at the list of authors, it looks like all of them have some affiliation with the Division of Adolescent Medicine, Children's Hospital of Los Angeles which is where your center is, correct?
A. That's correct.
Q. Okay. So they may have been patients of the other coauthors or your patients?

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A. The only other coauthor on there that's a medical doctor is Dr. Marvin Belzer, and he would have been the other person providing medical services at that time. We were the only two.
Q. Okay. And in the "Results" section of the abstract, it says, "Of the initial 101 participants, 59 youth had follow-up physiologic data collected between 21 and 31 months after initiation of hormones available for analysis. Metabolic parameters changes were not clinically significant with the exception of sex steroid levels intended to be the target of intervention."
First, did I read that correctly?
A. Yes.
Q. Okay. So does that mean that the -- there weren't metabolic -- and I'm sorry if I ask another bad question.
Does that mean that there weren't metabolic changes that were seen as a result of the use of gender-affirming hormones?
MS. BORELLI: Objection; form.
THE WITNESS: So that means that there were not statistically significant changes in the metabolic parameters except for the sex steroid levels.

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Q. (BY MR. DAVID) Which were something that you expected as a result of using gender-affirming hormones, correct?
A. Correct.
Q. Okay. So in terms of the effects of gender-affirming hormones on patients, in layman's terms, what were the results of your study?
A. So the results of the study indicate that over this approximate -- it's 21 to 31 months, so it was around-ish two years of treatment, that from a metabolic perspective, hormone use, exogenous hormone use was safe.
Q. Now, it says that 59 youth had follow-up physiologic data out of the initial 101 participants.
Does that mean that 42 patients refused to answer a survey or no longer were using gender-affirming hormones? What does that mean?
MS. BORELLI: Objection; form.
THE WITNESS: So there were no surveys involved in this study. This is just going back to the chart. So it means they were no longer in care -- that's the most common reason. Because of the age range, people are moving out and going to college and things like that.

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Just interestingly, I did a five- to seven-year follow-up with the same cohort, and I actually found 65 of them, which was amazing because the majority of them were actually out of services at that point.
Q. (BY MR. DAVID) When you did that longer-term follow-up, were you doing it just to check in on them? Or were you obtaining information for research purposes?
MS. BORELLI: Objection; form.
THE WITNESS: Sorry. I was doing a research project. It was part of my research. It was part of this study to do the follow-up with them.
Q. (BY MR. DAVID) Okay. What information did you gather from them after that five- to seven-year period?
A. So -- you know what, I just recalled that I did have them answer questions in the first round. I totally forgot about that. So the five- to seven-year follow-up, I pivoted all of those questions into REDCap. I don't know if you know what that is, but it's a mechanism of getting data where people can participate from afar, from a remote space.

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And I mean, there's a lot of stuff. I couldn't go through all of it, but what I really -- I didn't have any metabolic data because most of those patients were out of care, so there was no chart abstraction to be had.

But I really wanted to find out about patients' wellbeing, their depression, their thriving, and all of those things were very positive.

Q. (BY MR. DAVID) Okay. Now, in your conclusions on the abstract, it says, "Although the impact of hormones on some historically concerning physiologic parameters, including lipids, potassium, hemoglobin, and prolactin, were statistically significant, clinical significance was not observed. Hormone levels physiologic concordant with gender of identity were achieved with feminizing and masculinizing medication regimens. Extensive and frequent laboratory examination and transgender adolescents may be unnecessary. The use of hormones in transgender youth appears to be safe over a treatment course of approximately two years."

First, did I read that correctly?

A. Yes.

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Q. Okay. Does that mean that there were statistically significant changes in lipids, potassium, hemoglobin, and prolactin, but it did not have any clinical effect on the patients?

MS. BORELLI: Objection; form.

THE WITNESS: I have to go and check each one of those parameters, but in essence, that's what it means, that there were some statistically significant changes, but that they didn't have clinical implications.

Q. (BY MR. DAVID) Okay. So in terms of potassium, a patient may have experienced a change in potassium levels, but it wasn't to an extent that they were hyperkalemic and in need of hospitalization?

MS. BORELLI: Objection; form.

THE WITNESS: Right. So in other words -- I'll talk about potassium specifically.

So in our transfeminine youth, in that study, the mean capacity at baseline was 4.25, and at follow-up it was 4.47. The normal range of potassium is 3.8 to 5.1.

So even though that's a statistically significant change, it's not a change of clinical importance.

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Q. (BY MR. DAVID) Okay. So it wasn't so significant that it moved a patient from a normal range into either an abnormally low or abnormally high range?

A. Correct.

Q. And I guess for the lipids -- I don't know how that's measured. How are the lipids measured?

MS. BORELLI: Objection; form.

THE WITNESS: Well, they're -- sorry. Lipids include cholesterol, the breakdown of cholesterol and triglycerides.

Q. (BY MR. DAVID) Okay. So no patients developed any chronic high cholesterol or anything like that?

A. So in our transfeminine patients, their total cholesterol went from 168.9 to 166.1. So they showed a slight improvement in their cholesterol.

And in our transmasculine, it went from 163.6 to 164.5.

Q. Okay. In the patient population that you had for this study for 101 participants, were they all previously on or concurrently on puberty blockers?

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A. No.

Q. So the age range was 12 to 23 years. So was there a cohort of patients that were on puberty blockers at the time of this study?

A. I would not call it a cohort. I want to say there was maybe one or two. I'd have to go back through and look, because I talk about it somewhere, but I don't know where. It was small. Maybe one or two.

Q. Okay. Do you know whether or not those one or two patients who were on puberty blockers were among the 42 patients who did not complete the study?

MS. BORELLI: Objection; form.

THE WITNESS: I could go back -- there was one transfeminine person and two transmasculine people. And I don't know if they were part of that cohort or not. I'd have to go back and look.

The likelihood, though, is low, because they most likely were younger patients, and so they would have been in care with me for a longer period of time.

Q. (BY MR. DAVID) Something I wanted to ask you about, the NIH grant study that you're doing.

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Is there a group of gender dysphoric youths that are not receiving interventions, not from your study but that you're aware of outside of your study that you're able to compare the differences between those not receiving treatment and those in your study who are receiving treatment?
MS. BORELLI: Objection; form.
THE WITNESS: Do you mean an untreated control group?
Q. (BY MR. DAVID) I am -- my understanding, at least, is that you can't have a -- you can't say, okay, we're going to have 30 kids that are going to not be treated as a part of this.
My understanding from reading your -- I believe it was your report and others is that that would be unethical; is that right?
MS. BORELLI: Objection; form.
THE WITNESS: That's correct.
Q. (BY MR. DAVID) Okay. But not every gender dysphoric youth is receiving treatment -- gender-affirming treatment, correct?
MS. BORELLI: Objection; form.
Q. (BY MR. DAVID) I'm not saying within your study. I'm saying outside of your study.
MS. BORELLI: Same objection.

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THE WITNESS: Yeah. I get what you're saying. And my sort of thought process around that is our treatment group at baseline acts as a proxy for an untreated group.
Q. (BY MR. DAVID) So how are you able to determine -- so let's say that you have your group of -- I believe you said that the younger cohort are individuals who are receiving puberty blockers only at this point, and you are following them.
How are you able to determine whether someone who goes through two years of puberty blockers has better or more optimal health outcomes than someone who has been gender dysphoric that whole time and is not undergoing puberty blockers?
MS. BORELLI: Objection; form.
THE WITNESS: That's a great question, and I will tell you.
So within our bigger group of people who -- the 314 who are going on gender-affirming hormones, 24 of those kids had their puberty blocked early, or they were late to puberty, so they never had experienced -- let's use the marker as people who never experienced a lot of their endogenous puberty, because that's how we're

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defining it.
And so I don't remember if we talked about this, but interestingly, at baseline -- at baseline, the cohort of young people who had had their puberty blocked or did not experience their endogenous puberty were, across the board, psychologically in better shape than the rest of the cohort by a very significant number.
Let's take depression, for example. So we measure depression on the Beck Depression Inventory, which is the pretty standard measure of depression. And those young people who had been blocked or had not experienced a lot of their endogenous puberty, their depression scores were, on average, 9 percent better than the rest of the cohort.
So we do have a natural comparison in there.
And additionally, the blocker cohort, many of those young people will be going on to gender-affirming hormones, and so we will have -- we have them to follow over time as well.
So there's ways to do comparisons between these groups and the just natural exposures that they've had because of when they entered care or

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when they didn't enter care to make that exact -- to answer that exact question.
Q. (BY MR. DAVID) So if I'm understanding correctly, part of the older cohort had experienced endogenous puberty up until baseline; is that correct?
MS. BORELLI: Objection; form.
THE WITNESS: The majority of the cohort except for those 24 young people had experienced their puberty, yes. Either completed it or finished most of it, yes.
Q. (BY MR. DAVID) Okay. And so you're able to look at their -- I don't know if it's scores, but you're able to look at how they are responding to the questionnaire at baseline and compare that to how the younger cohort answers the questionnaire after X number of months or X number of years of puberty suppression?
MS. BORELLI: Objection; form.
THE WITNESS: So it's not a younger cohort. There are people -- so it's not a younger cohort. It's just the subcohort of people who had their puberty blocked. And then now they're starting hormones versus the people who did not have their puberty blocked and now they're

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starting hormones.
Q. (BY MR. DAVID) Okay. So I think I was misunderstanding.
So you're talking about the 24 who did have their puberty blocked compared to 290 who did not have their puberty blocked?
A. Correct.
Q. Okay. Are you also looking at the -- my understanding is that there is a younger cohort separate from the gender-affirming cohort.
Are you also looking at what their responses are to the questionnaire once they have reached a similar age to those in -- the 290 in the older cohort?
MS. BORELLI: Objection; form.
THE WITNESS: Yes. So it's more complicated than that because measures and scales often change a little bit as people age. But we will have the capacity to -- what we do is we shift. When they get to a certain age, they will get the appropriate measure for that age, and then they will be comparable.
So we'll have, in addition to those 24 people, will have the other 91 and will have them answering those questions as well as the 24. So

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we'll have a larger number of people who -- that we can look at who had an opportunity to get their puberty blocked.
Q. (BY MR. DAVID) I'm going to bounce around here, and I'm trying to finish up.
I believe you testified earlier that going through an endogenous puberty can have irreversible effects that are only corrected with -- potentially with surgery, and some may not even be correct with a surgery; is that correct?
MS. BORELLI: Objection; form.
THE WITNESS: That's correct.
Q. (BY MR. DAVID) Do gender-affirming hormones carry the same possibility of irreversible changes?
MS. BORELLI: Objection; form.
THE WITNESS: Yes. Both -- because it's the same hormones. I just want to be clear that the hormones we use for exogenous phenotype change are the bioidentical hormones to what your body is making.
So in the same way, yes, there are some permanent changes of both endogenous hormones and exogenous hormones, because they're the same hormones.

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Q. (BY MR. DAVID) Okay. This was a long time ago, but a long time ago, we discussed some of the side effects of gender-affirming surgeries, and you mentioned something called dog ears.
Do you recall that?
A. I do.
Q. Okay. Are there procedures to correct the dog ears?
MS. BORELLI: Objection; form.
THE WITNESS: Yeah, there are. There are surgical procedures that can correct it after the swelling comes down and you can finally see what the final contour is after the healing has happened.
Q. (BY MR. DAVID) And is the surgery to correct the appearance of dog ears medically necessary?
MS. BORELLI: Objection; form.
THE WITNESS: I think so. The cases where I have referred for those revisions have been. I think I've only made that referral two times.
Q. (BY MR. DAVID) And what is the basis for the medical necessity of removal of the dog ears?
MS. BORELLI: Objection; form.

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THE WITNESS: Sorry. It's similar to the original reason, need for the surgery, right? Is the appearance of a masculine chest that matches does not include something that has physiologic parentheses -- I guess that's the best way I can think about it is like -- you can't see my whole torso so I can't really show you, but it's called dog ears because there are these little triangular, like, portions where the surgical incision meets the rest of the body is the best way I can describe it.
Q. (BY MR. DAVID) Is it essentially scar tissue?
MS. BORELLI: Objection; form.
THE WITNESS: No. It's not scar tissue. It has to do with because of the -- it has to do with the procedure that flattens the chest tissue but then it meets the rest of the tissue. And so there is an irregularity -- it's really hard to describe without showing it to you, but it has to do with that. It's not scar tissue.
Q. (BY MR. DAVID) Okay. Have you reviewed any specific provisions of West Virginia Medicaid's policy regarding gender-affirming care?
MS. BORELLI: Objection; form.

THE WITNESS: I have not.
MR. DAVID: Doctor, I think those are all the questions that I have for you. Thank you very much.
THE WITNESS: You're so welcome.
MS. BORELLI: And the plaintiffs have no questions for Dr. Olson-Kennedy today. And we will read and sign.
THE VIDEOGRAPHER: This is the end of Media No. 1. We're going off the record. The time is 2:51 p.m.

(The remote videotaped deposition concluded at 2:51 p.m.)

* * *

(Signature was requested.)

REPORTER'S CERTIFICATE

STATE OF IDAHO)
) ss.
COUNTY OF ADA)

I, AMY E. SIMMONS, Certified Shorthand Reporter and Notary Public in and for the State of Idaho, do hereby certify:

That prior to being examined, the witness named in the foregoing deposition was by me duly sworn remotely to testify to the truth, the whole truth and nothing but the truth;

That said deposition was taken down by me in shorthand at the time and place therein named and thereafter reduced to typewriting under my direction, and that the foregoing transcript contains a full, true and verbatim record of said deposition.

I further certify that I have no interest in the event of the action.

WITNESS my hand and seal this 10th day of May, 2022.

AMY E. SIMMONS
CSR, RPR, CRR, CRC and Notary Public in and for the State of Idaho.

My Commission Expires: 06-13-2022

Veritext Legal Solutions
1100 Superior Ave
Suite 1820
Cleveland, Ohio 44114
Phone: 216-523-1313

May 11, 2022

To: Tara Borelli, Esq.

Case Name: Fain, Christopher, et al. v. Crouch, William, et al.

Veritext Reference Number: 5200240

Witness: Johanna Olson-Kennedy, M.D. Deposition Date: 4/25/2022

Dear Sir/Madam:

Enclosed please find a deposition transcript. Please have the witness review the transcript and note any changes or corrections on the included errata sheet, indicating the page, line number, change, and the reason for the change. Have the witness' signature notarized and forward the completed page(s) back to us at the Production address shown above, or email to production-midwest@veritext.com.

If the errata is not returned within thirty days of your receipt of this letter, the reading and signing will be deemed waived.

Sincerely,
Production Department

NO NOTARY REQUIRED IN CA

DEPOSITION REVIEW
CERTIFICATION OF WITNESS

ASSIGNMENT REFERENCE NO: 5200240
CASE NAME: Fain, Christopher, et al. v. Crouch, William, et al.
DATE OF DEPOSITION: 4/25/2022
WITNESS' NAME: Johanna Olson-Kennedy, M.D.

In accordance with the Rules of Civil Procedure, I have read the entire transcript of my testimony or it has been read to me.

I have made no changes to the testimony as transcribed by the court reporter.

_____ _____
Date Johanna Olson-Kennedy, M.D.

Sworn to and subscribed before me, a Notary Public in and for the State and County, the referenced witness did personally appear and acknowledge that:

They have read the transcript;
They signed the foregoing Sworn Statement; and
Their execution of this Statement is of their free act and deed.

I have affixed my name and official seal this _____ day of_____, 20____.

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Notary Public

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Commission Expiration Date

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I have listed my changes on the attached Errata Sheet, listing page and line numbers as well as the reason(s) for the change(s).

I request that these changes be entered as part of the record of my testimony.

I have executed the Errata Sheet, as well as this Certificate, and request and authorize that both be appended to the transcript of my testimony and be incorporated therein.

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Date Johanna Olson-Kennedy, M.D.

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ERRATA SHEET
VERITEXT LEGAL SOLUTIONS MIDWEST
ASSIGNMENT NO: 5200240

PAGE/LINE(S) / CHANGE /REASON

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_____ _____
Date Johanna Olson-Kennedy, M.D.

SUBSCRIBED AND SWORN TO BEFORE ME THIS _____
DAY OF _____, 20_____ .

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Notary Public

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Commission Expiration Date

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q

Federal Rules of Civil Procedure

Rule 30

(e) Review By the Witness; Changes.

(1) Review; Statement of Changes. On request by the deponent or a party before the deposition is completed, the deponent must be allowed 30 days after being notified by the officer that the transcript or recording is available in which:

(A) to review the transcript or recording; and

(B) if there are changes in form or substance, to sign a statement listing the changes and the reasons for making them.

(2) Changes Indicated in the Officer's Certificate. The officer must note in the certificate prescribed by Rule 30(f)(1) whether a review was requested and, if so, must attach any changes the deponent makes during the 30-day period.

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VERITEXT LEGAL SOLUTIONS
COMPANY CERTIFICATE AND DISCLOSURE STATEMENT

Veritext Legal Solutions represents that the foregoing transcript is a true, correct and complete transcript of the colloquies, questions and answers as submitted by the court reporter. Veritext Legal Solutions further represents that the attached exhibits, if any, are true, correct and complete documents as submitted by the court reporter and/or attorneys in relation to this deposition and that the documents were processed in accordance with our litigation support and production standards.

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Veritext Legal Solutions
1100 Superior Ave
Suite 1820
Cleveland, Ohio 44114
Phone: 216-523-1313

May 11, 2022

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DEPOSITION REVIEW
CERTIFICATION OF WITNESS

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I request that these changes be entered as part of the record of my testimony.

I have executed the Errata Sheet, as well as this Certificate, and request and authorize that both be appended to the transcript of my testimony and be incorporated therein.

May 21, 2022 _____
Date Johanna Olson-Kennedy, M.D.

Sworn to and subscribed before me, a Notary Public in and for the State and County, the referenced witness did personally appear and acknowledge that:

They have read the transcript;
They have listed all of their corrections in the appended Errata Sheet;
They signed the foregoing Sworn Statement; and
Their execution of this Statement is of their free act and deed.

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Notary Public

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Commission Expiration Date

ERRATA SHEET

VERITEXT LEGAL SOLUTIONS MIDWEST

ASSIGNMENT NO: 5200240

PAGE/LINE(S) /	CHANGE	/REASON
P. 6 / line 6	"OHANNA" to "JOHANNA"	Transcription error
P. 26 / line 3	"therapy" to "puberty"	Transcription error
P. 30 / line 20	"a gender" to "agender"	Transcription error
P. 34 / line 5	"connectiveness" to "connectivity"	Transcription error
P. 58 / line 3	strike "become broader, grants"	Transcription error
P. 111 / line 10	"names" to "games"	Transcription error
P. 112 / line 25	"before" to "about"	Transcription error
P. 134 / line 17	"surgery" to "puberty"	Transcription error
P. 195 / line 20	remove the word "capacity"	Transcription error
P. 200 / line 15	"percent" to "points"	Transcription error

May 21, 2022 _____

Date Johanna Olson-Kennedy, M.D.

SUBSCRIBED AND SWORN TO BEFORE ME THIS _____

DAY OF _____, 20_____ .

_____

Notary Public

_____

Commission Expiration Date

IN THE UNITED STATES DISTRICT COURT
FOR THE SOUTHERN DISTRICT OF WEST VIRGINIA
HUNTINGTON DIVISION

EXHIBIT 1

CHRISTOPHER FAIN, *et al.*, individually and on behalf of all others similarly situated, *Plaintiffs,* v. WILLIAM CROUCH, *et al.*, *Defendants.*	CIVIL ACTION NO. 3:20-cv-00740 HON. ROBERT C. CHAMBERS

CERTIFICATE OF SERVICE

I hereby certify that the EXPERT REBUTTAL REPORT OF DR. JOHANNA OLSON-KENNEDY, M.D., M.S. was served electronically on the 18th day of March, 2022 on the following counsel for Defendants in the above-captioned case:

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Roberta F. Green (WVSB #6598)
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Dated: March 18, 2022

Respectfully submitted,

s/ Walt Auvil

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IN THE UNITED STATES DISTRICT COURT
FOR THE SOUTHERN DISTRICT OF WEST VIRGINIA
HUNTINGTON DIVISION

CHRISTOPHER FAIN, *et al.*, individually and on behalf of all others similarly situated,

Plaintiffs,

v.

WILLIAM CROUCH, *et al.*,

Defendants.

CIVIL ACTION NO. 3:20-cv-00740

HON. ROBERT C. CHAMBERS, JUDGE

EXPERT REBUTTAL REPORT OF DR. JOHANNA OLSON-KENNEDY, M.D., M.S.

I, Johanna Olson-Kennedy, M.D., M.S., declare as follows:

1. My name is Johanna Olson-Kennedy. I have been retained by counsel for Plaintiffs as an expert in connection with the above-captioned litigation.

2. I have been asked by Plaintiffs' counsel to provide my expert opinion on gender identity, the treatment and diagnosis of gender dysphoria, particularly as it pertains to children and adolescents, and to respond to, rebut, and provide my expert opinion regarding the report by Dr. Stephen B. Levine in this case ("Levine Report").

3. I have actual knowledge of the matters stated herein. If called to testify in this matter, I would testify truthfully and based on my expert opinion.

I. BACKGROUND AND QUALIFICATIONS

4. I received my Doctor of Medicine (M.D.) degree from the Chicago Medical School in 1997. In 2000, I completed my residency in pediatrics at the Children's Hospital of

Orange County, California, and from 2000 to 2003, I was a Fellow in adolescent medicine at the Children's Hospital of Los Angeles.

5. I have been a licensed physician in California since 2000 and am Double Board Certified by the American Board of Pediatrics in Pediatrics and in Adolescent Medicine. I specialize in the care of transgender youth and gender diverse children, and am currently the Medical Director of the Center for Transyouth Health and Development, in the Division of Adolescent Medicine at the Children's Hospital in Los Angeles, California. The Center is one of the largest clinics in the United States for transgender youth and provides gender diverse youth with both medical and mental health services, including consultation for families with gender diverse children and routine use of medications to suppress puberty in peri-pubertal youth (i.e., youth at the onset of puberty), gender affirming hormone use for masculinization and feminization as well as surgical referrals. Under my direction, the Center conducts rigorous research aimed at understanding the experience of gender diversity and gender dysphoria from childhood through early adulthood.

6. Over the course of my work with this population during the past 16 years, I have provided services for approximately 1,000 young people and their families, and currently have an active panel of around 650 patients of varying ages, up to 25 years old.

7. I have been awarded research grants to examine the impact of early interventions, including puberty-delaying treatment and gender affirming hormones, on the physiological and psychosocial development of gender diverse and transgender youth. I have lectured extensively on the treatment and care of gender diverse children and transgender adolescents, including topics such as pubertal suppression, gender affirming hormone therapy, transitioning teens and the adolescent experience, age considerations in administering hormones, and the needs, risks,

and outcomes of hormonal treatments. I have published numerous articles and chapters, both peer reviewed and non-peer reviewed, on transgender health-related issues.

8. I am currently the principal investigator on a multisite NIH grant which recently received funding to continue, for an additional 5 years, an ongoing study examining the impact of gender affirming medical care for transgender youth on physiologic and psychological health and well-being. The first five years have already been completed. This is the first study of its kind in the U.S. to determine longitudinal outcomes among this population of vulnerable youth. The study to date has yielded approximately 26 manuscripts.

9. I am an Associate Professor at the Keck School of Medicine at the University of Southern California and attending physician at Children's Hospital of Los Angeles. I have been a member of the World Professional Association for Transgender Health (WPATH) since 2010, and a Board Member of the U.S. Professional Association for Transgender Health (USPATH) since 2017. I am also a member of the Society for Adolescent Health and Medicine and the American Academy of Pediatrics. In addition, I am a member of the LGBT Special Interest Group of the Society for Adolescent Health and Development.

10. I am the 2014 Recognition Awardee for the Southern California Regional Chapter of the Society for Adolescent Health and Medicine.

11. In 2019, I was invited by the University of Bristol as a Benjamin Meaker visiting professor, the purpose of which is to bring distinguished researchers from overseas to Bristol in order to enhance the research activity of the university.

12. In preparing this report, I have relied on my training and years of research and clinical experience, as set out in my curriculum vitae, and on the materials listed therein. A true and accurate copy of my curriculum vitae is attached hereto as Exhibit A. It documents my

education, training, research, and years of experience in this field and includes a list of publications.

13. I have also reviewed the materials listed in the attached bibliography (Exhibit B). The sources cited therein are authoritative, scientific peer-reviewed publications. I generally rely on these materials when I provide expert testimony, and they include the documents specifically cited as supportive examples in particular sections of this declaration.

14. In addition, I have reviewed the First Amended Complaint in this case and the report by Dr. Levine.

15. The materials I have relied upon in preparing this report are the same types of materials that experts in my field of study regularly rely upon when forming opinions on the subject. I reserve the right to revise and supplement the opinions expressed in this report or the bases for them if any new information becomes available in the future, including as a result of new scientific research or publications or in response to statements and issues that may arise in my area of expertise.

Prior Testimony

16. In the last four years, I have testified as an expert at trial or by deposition in the following cases: *Kadel v. Folwell*, Case No. 1:19-cv-00272-LCB-LPA (M.D.N.C.); *In the interest of JA.D.Y. and JU.D.Y.*, Children, Case No. DF-15-09887 (255th Jud. District Ct., Dallas Cty., Tex.); and *Paul E. v. Courtney F.*, No. FC2010-051045 (Superior Ct., Maricopa Cty., Ariz.).

Compensation

17. I am being compensated for my work on this matter at a rate of $200.00 per hour for preparation of declarations and expert reports, as well as any pre-deposition and/or pre-trial

preparation and any deposition testimony or trial testimony. My compensation does not depend on the outcome of this litigation, the opinions I express, or the testimony I may provide.

II. EXPERT OPINIONS[1]

A. Gender Identity

18. Gender identity, often simply termed "gender," is a distinct characteristic and is defined as one's internal sense of being male, female, both, neither, or some other gender identity. It has a strong biological basis. Every person has a gender identity. The term cisgender refers to a person whose gender identity matches their sex assigned at birth. The term transgender refers to a person whose gender identity does not match their sex assigned at birth.

19. Historically, "gender" was equated with a person's sex assigned at birth, which refers to the sex assigned to a person when they are born, generally based on external genitalia. However, a more contemporary understanding of gender shows that one's gender identity may differ from one's sex assigned at birth.

20. While both gender identity and sex are often assumed and treated as binary and oppositional, they are more accurately experienced as along a spectrum. For example, there are multiple sex characteristics, such as genitalia, chromosomal makeup, hormones, and variations in brain structure and function. For some of these characteristics there is significant variance as reflected by the dozens of intersex mechanisms and varying gender identities. Additionally, not all sex characteristics, including gender identity, are always in alignment. Accordingly, the Endocrine Society Guidelines, state that, "As these may not be in line with each other (*e.g.*, a

[1] Subsections A and B of this report explain several concepts and provide some necessary background information that is necessary to understand the more specific critiques of the report by Dr. Levine that I lay out in subsection C.

person with XY chromosomes may have female-appearing genitalia), the terms biological sex and biological male or female are imprecise and should be avoided."

21. As early as 1966 it has been understood that gender identity cannot be changed. Efforts to do so have been shown to be unsuccessful and harmful.

22. "Conversion" or "reparative" therapy refers to the practice of attempting to change an individual's sexual orientation and attractions from members of the same sex to those of the other sex. A similar model of therapy for individuals with a transgender identity or experience has historically been an approach promoted by some individuals, such as Dr. Levine, notwithstanding its ineffectiveness and harmful effects. Accordingly, 20 states and the District of Columbia have banned reparative therapy for youth, and major medical organizations have issued statements deeming the practice to be unethical.

23. A Williams Institute report published in 2018 estimates that just under 700,000 LGBT individuals in the United States have undergone "conversion therapy" at some point in their lifetime, about half of those during adolescence. Because some psychiatrists and sexologists working in the 1960's and 70's perpetuated the idea that being transgender was likely the result of a pathological early childhood experience, many professionals and lay community members continue to believe that gender is malleable. Tactics have ranged from simple redirection, thought pattern alteration or hypnosis to aversion techniques including induction of vomiting, nausea, paralysis, or electric shocks to change the expression, behavior, and assertion of one's authentic gender. (Mallory, et al., 2019). However, multiple studies show that gender identity has a strong biological basis and cannot be changed. As such, reparative therapy is both ineffective and harmful for transgender and gender diverse youth.

B. Gender Dysphoria and its Treatment

24. Gender Dysphoria (GD) is a serious medical condition characterized by distress due to a mismatch between assigned birth sex and a person's internal sense of their gender. By definition this diagnosis applies to transgender people, not cisgender people. GD was formerly categorized as Gender Identity Disorder (GID) but the condition was renamed in May 2013, with the release by the American Psychiatric Association (APA)'s fifth edition of the Diagnostic and Statistical Manual of Mental Disorders (DSM-V). In announcing this change, the APA explained that in addition to the name change, the criteria for the diagnosis were revised "to better characterize the experiences of affected children, adolescents, and adults." The APA further stressed that "gender nonconformity is not in itself a mental disorder. The critical element of gender dysphoria is the presence of clinically significant distress associated with the condition."

25. On May 25, 2019, the World Health Assembly approved International Classification of Diseases (ICD) version 11 that had been published by the World Health Organization in 2018. In this newest version of the ICD, all transgender-related diagnostic codes were removed from the chapter "Mental and Behavioral Disorders," and the code "Gender incongruence" was included in a new chapter "Conditions related to sexual health." These codes replaced the outdated "Gender Identity Disorder of childhood" (F64.2), "Gender Identity Disorder not otherwise specified" (F64.9), "transsexualism" (F64.0), and "Dual-role transvestism" (F64.1) codes which perpetuated the idea that patients seeking and undergoing medical interventions for this medical condition are mentally ill. (Suess Schwend, 2020).

26. For a person to be diagnosed with GD, there must be a marked difference between the individual's expressed/experienced gender and their assigned sex at birth , present for at least

six months. In children, the desire to be of the other gender must be present and verbalized.[2] The condition must cause clinically significant distress or impairment in social, occupational, or other important areas of functioning.

27. The World Professional Association of Transgender Health (WPATH) has clear recommendations for the health of transgender and gender non-conforming people in what is now the Standards of Care version 7. The SOC are based on the best available science and expert professional consensus. They are currently under revision to create an updated version 8. The WPATH Standards of Care have been endorsed and cited as authoritative by most major medical associations in the United States, including the American Medical Association, the American Psychiatric Association, the American Psychological Association, the Endocrine Society, the Pediatric Endocrine Society, the American College of Physicians, and the American Academy of Family Physicians, among others.

28. The UCSF Center for Excellence in Transgender Care as well as the Endocrine Society have both published comprehensive guidelines for the care of transgender and non-binary individuals that are largely consistent with the WPATH Standards of Care.

29. There are a significant number of *pre-pubertal* children who demonstrate an interest or preference for clothing, toys, and games that are stereotypically of interest to members of the "other" gender. Some of these children are transgender and some are not. It is the study of such *pre-pubertal* children that has created confusion about the persistence of gender dysphoria into adolescence and adulthood. Specifically, the *pre-pubertal* children who were the

[2] Notably, the DSM-IV included a separate diagnosis for GID in children, which required the child to display a number of behaviors stereotypical of the non-natal gender. That diagnosis, and its list of behavioral requirements, have been deleted from the DSM-V and replaced by updated and more precise diagnostic criteria.

subject of research endeavors in the late 20th century included both of these groups of children, those that would have met current criteria for a diagnosis of "Gender Dysphoria in Children" and those who would be considered "sub-threshold" for this diagnosis. At the time of these studies, criteria B had not yet been added to the diagnosis, which is "the presence of clinically significant distress associated with the condition." In addition, the criteria for then-used "gender identity disorder in children" diagnosis did not require a child to have "a strong desire to be of the other gender or an insistence that one is the other gender (or some alternative gender different from one's assigned gender)," which the current "gender dysphoria in children" diagnosis does. Thus, given the broader criteria used at the time, it is unsurprising that some of the research undertaken toward the end of the 20th century demonstrated that most children who exhibited gender-nonconforming behavior did not go on to have a transgender identity in adolescence. Yet, notwithstanding its inapplicability and faulty underpinnings, this "evidence" has been used to argue against gender affirmation for children and adolescents.

30. In any event, these arguments are wholly irrelevant to the question of coverage and provision of medical care as treatment for GD. That is because the majority of desistance research to date is among gender non-conforming pre-pubescent children, and my clinical experience has been that if gender dysphoria and gender identification with a gender other than that recorded at birth persists into adolescence, then desistance is incredibly rare. No medical or surgical treatments are recommended for *pre-pubertal* children.

31. Additionally, no studies have ever demonstrated that gender affirmation in childhood "leads to" a child being transgender who otherwise might not have been. Studies have demonstrated that the majority of youth whose GD and cross-gender identity continue to be present in adolescence, or those whose GD emerges in adolescence, are highly unlikely to

identify and live as cisgender individuals. Youth with GD, particularly those who are unaffirmed and denied care, are at high risk for depression, anxiety, isolation, self-harm and suicidality at the onset of puberty-related changes that feel wrong to them.

32. In his report, Dr. Levine discusses a number of approaches to care, though he fails to properly describe them and to discuss their limitations.

33. One of the approaches discussed by Dr. Levine is also known as "reparative" or "corrective" therapy. As discussed above, this so-called "therapy" has proven to be ineffective at best, and harmful at worst, and has been deemed to be unethical.

34. **"Redirection"** – Under this approach, advocated by people like Dr. Levine, a mental health therapist would encourage caregivers to use positive reinforcement to try to "redirect" children toward behavior that is more typical of their birth-designated sex or less gender specific. Underlying this approach is the assumption that a child's gender identity is malleable through social interventions. The goal of redirection is thus to eliminate gender-diverse desires and expressions over time, and to try to prevent the transgender child from being transgender. This approach is not recommended because negative reinforcement (e.g., shaming the child for gender diverse expression) has substantial negative mental and social health consequences. (Turban and Ehrensaft, 2018; Ehrensaft, 2017). It also ignores that gender identity is innate and cannot be involuntarily changed.

35. **Wait-and-see** – The wait-and-see approach (also called watchful waiting) involves waiting to see if the child's gender identity will change as the child gets older. This approach typically recommends that caregivers prohibit a prepubertal social transition but may allow cross-gender play and clothing within the home or support both masculine and feminine activities as the child explores their interests in other social settings. The wait-and-see approach

assumes that gender is binary and becomes fixed at a certain age; it pathologizes gender diversity. It is distinguished from following the child's lead, an affirming approach that allows the child to present in the gender role that feels correct and moves at a pace that is largely directed by the child. This watchful waiting approach ignores evidence that young children thrive when given permission to live in the gender that is most authentic to them and are at risk for symptomatic behaviors if prevented from doing so. (Ehrensaft, 2017).

36. **Affirmation** – The affirmative approach considers no gender identity outcome: transgender, cisgender, or otherwise, to be preferable. (Turban and Ehrensaft, 2018). It permits a child to explore gender development and self-definition within a safe setting. A fundamental concept of this approach is that gender diversity is not a mental illness. The gender affirmative model is defined as a method of therapeutic care that includes allowing children to speak for themselves about their self-experienced gender identity and expressions and providing support for them to evolve into their authentic gender selves, no matter at what age. Under this model, a child's self-report is embedded within a collaborative model with the child as subject and the collaborative team including the child, parents, and professionals. Support is not characterized by "encouraging" children or youth to be transgender or not, but rather by allowing children who express a desire to undergo a social transition (which may include changing names, pronouns, clothing, hairstyles, etc.) to do so. **For children who have not yet reached puberty, medical intervention is unnecessary and unwarranted.** After the onset of puberty medical interventions such as puberty blockers, and later hormones and surgery may be appropriate.

37. While some argue that gender affirmation leads a child or adolescent down a path of inevitable transgender identity, no such evidence exists, either in the scientific or the clinical

setting. To the contrary, studies show that gender identification does not meaningfully differ before and after social transition. (Rae, et al., 2019).

38. Under both the "wait and see" and affirmative care models, as understood in the scientific literature, medical care is recommended following the onset of puberty. (Ehrensaft, 2017).

39. The most effective treatment for adolescents and young adults with GD, in terms of both their mental and medical health, contemplates an approach allowing each patient to access care based on their particular need. Medical and surgical treatment interventions are determined by the care team (usually a medical and mental health professional) in collaboration with the patient, and the patient's family. These medical decisions are made by the care team in conjunction with the patient and the patient's family and consider the patient's social situation, the level of gender dysphoria, developmental stage, chronologic age, existing medical conditions, and other relevant factors. Sometimes treatment begins with puberty delaying medications (also referred to as puberty blockers), later followed by gender affirming hormones. Most youth accessing treatment are already well into or have completed puberty. Gender-affirming genital surgeries are generally sought after hormone treatment has commenced.

40. *Puberty blockers:* The beginning signs of puberty in transgender youth (the development of breast buds in birth-assigned females and increased testicular volume in birth-assigned males) is often a painful and sometimes traumatic experience that brings increased body dysphoria and the potential development of a host of comorbidities including depression, anxiety, substance abuse, self-harming behaviors, social isolation, high-risk sexual behaviors, and increased suicidality. Puberty blocking, which involves the administration of gonadotrophin-releasing hormone analogues (GnRH), essentially pauses puberty, thereby

allowing the young person an opportunity to explore gender without having to experience the anxiety and distress associated with developing the undesired secondary sexual characteristics. In addition, for parents/guardians uneducated about gender diversity and/or who have only recently become aware of their child's transgender identity, puberty blockers provide additional time and opportunity to integrate this new information into their own experience and to develop skills to support their child. Puberty suppression also has the benefit of potentially rendering obsolete some gender-affirming surgeries down the line, such as male chest reconstruction, tracheal shave, facial feminization, and vocal cord alteration, which otherwise would be required to correct the initial "incorrect" puberty.

41. Puberty suppression has been used safely for decades in children with other medical conditions and is a reversible intervention. If the medication is discontinued, the young person continues their endogenous puberty several months after puberty suppression is discontinued. The "Dutch protocol," developed from a study conducted in the Netherlands and published in 2006, calls for the commencement of puberty blockers for appropriately diagnosed and assessed gender dysphoric youth as early as 12 years of age. (de Vries, et al., 2014). Both the Endocrine Society and the WPATH's Standards of Care, however, recommend initiation of puberty suppression at the earliest stages of puberty (usually, Tanner 2, assuming someone is engaged in services before or around this time), regardless of chronological age, in order to avoid the stress and trauma associated with developing secondary sex characteristics of the natal sex.

42. A growing body of evidence that demonstrates the positive impact of pubertal suppression in youth with GD on psychological functioning including a decrease in behavioral and emotional problems, a decrease in depressive symptoms, and improvement in general functioning. (Turban, et al., 2020; de Vries, et al., 2014).

43. Puberty-delaying treatment, thus, affords youth the opportunity to undergo a single, correct pubertal process and avoid many of the surgical interventions previously necessary for assimilation into an authentic gender role. It is a simple reversible intervention that has the capacity to improve health outcomes and save lives. Over the course of my work in the past sixteen years with gender diverse and transgender youth, I have prescribed hormone suppression for over 300 patients. All of those patients have benefitted from putting their endogenous puberty process on pause, even the small handful who discontinued GnRH analogues and went through their endogenous puberty. Many of these young people were able to matriculate back into school environments, begin appropriate peer relationships, participate meaningfully in therapy, and family functions. Children who had contemplated or attempted suicide or self-harm (including cutting and burning) associated with monthly menstruation or the anxiety about their voice dropping were offered respite from those dark places of despair. GnRH analogues for puberty suppression are, in my opinion, a sentinel event in the history of transgender medicine, and have changed the landscape almost as much as the development of synthetic hormones.

44. *Gender affirming hormones:* Cross-gender or gender affirming hormone therapy involves administering steroids of the experienced sex (i.e., their gender identity) (estrogen for transfeminine individuals and testosterone for transmasculine individuals). The purpose of this treatment is to attain the appropriate masculinization or feminization of the transgender person to achieve a gender phenotype that matches as closely as possible to their gender identity. Gender affirming hormone therapy is a partially reversible treatment in that some of the effects produced by the hormones are reversible (e.g., changes in body fat composition, decrease in facial and body hair) while others are irreversible (e.g., deepening of the voice, breast development).

Eligibility and medical necessity should be determined case-by-case, based on an assessment of the youth's unique cognitive and emotional maturation and ability to provide knowing and informed consent. The decision would be made only after a careful review with the youth and parents/guardians of the potential risks and benefits of hormone therapy. The youth's primary care provider, therapist, or another experienced mental health professional can help document and confirm the patient's history of gender dysphoria, the medical necessity of the intervention, and the youth's readiness to transition medically.

45. *Gender-affirming surgeries:* Some transgender individuals need surgical interventions to help bring their phenotype into alignment with their gender. Surgical interventions may include vaginoplasty, tracheal shave, liposuction, breast implants, and orchiectomy for transfeminine individuals and chest reconstruction, hysterectomy, oophorectomy, salpingectomy, construction of neo-scrotum, and metoidioplasty or phalloplasty for transmasculine individuals.

46. The current WPATH Standards of Care recommend that genital surgery – i.e., surgery which may render the individual sterile – not be carried out until the individual reaches the legal age of majority to give consent for medical procedures, while acknowledging that care is individualized. In addition, the Standards recommend that the other surgical interventions (e.g., chest surgery for transgender males and breast augmentation for transgender females) may occur earlier than the legal age of consent, preferably after ample time living in the desired gender role and after one year of hormone therapy. The Standards of Care, however, further recognize that these are individual determinations and that "different approaches may be more suitable, depending on an adolescent's specific clinical situation and goals for gender identity expression."

47. Gender affirming medical interventions are considered medically necessary and are recognized as such by many major professional organizations. The denial of this care results in negative health consequences.

48. There are those, like Dr. Levine, who would make the argument that the recent uptick in youth presenting for services related to GD is the result of "social contagion." But if "social contagion" applied to gender and gender identity, there would be zero transgender people because of the consistent exposure to an overwhelming majority of cisgender people. The social contagion argument that is posited by some confuses the relationship between one's recognition of their gender and their exposure to gender related information and community – particularly with regard to internet activity – asserting that youth are declaring themselves to be transgender or gender diverse because they were exposed to this online, or they have multiple friends who are also experiencing GD. But adolescent development includes finding like groups of peers, which extends to finding friend groups who are also gender diverse. Finally, attributing GD to "social contagion" is a simplistic perspective that ignores that the process of seeking care is complex and difficult and involves parental consent for minors.

49. There is no scientific evidence that one develops gender dysphoria from being exposed to people with GD. To the contrary, most evidence shows that gender identity has a biological basis (Korpaisarn, et al., 2019; Saraswat, et al., 2015) and is affixed by early childhood (Slaby, et al., 1975).

C. Specific Critiques

50. Overall, Dr. Levine shows a lack of familiarity and understanding regarding the existing research about gender identity and gender dysphoria, as well as the clinical experience surrounding the treatment of gender dysphoria, particularly regarding transgender youth. This

lack of familiarity and understanding makes sense, as Dr. Levine appears to have very limited experience working with transgender youth and has not been a member of WPATH for decades.

51. Dr. Levine has critiqued and opposed the provision of gender affirming care as treatment for gender dysphoria for decades. Yet, in all of these years, he has not undertaken the research he calls for to answer the questions he raises. Rather it seems his primary goal is opposing affirming care for transgender people, instead of finding answers to questions and providing the best care for transgender people suffering from gender dysphoria.

52. Below I outline additional, more specific critiques regarding Dr. Levine's report.

53. Dr. Levine's claim that treatment for gender dysphoria is experimental and unproven is simply a statement of opinion, and not fact. *See, e.g.*, Levine Report ¶ 23. We have decades of research and clinical experience on gender dysphoria and its care. To be sure, as with all medical care, there is a range of quality in the existing data regarding the treatment of gender dysphoria (see UCSF Guidelines), and there is certainly a need for additional studies of a longitudinal nature. But again, that is true with most medical care.

54. One of the intrinsic elements of rating the quality of evidence is the study design. Randomized controlled studies are considered the highest quality in the grading of evidence. But given the length of time that gender affirming medical interventions have been around and vast amount of clinical knowledge about their efficacy, having untreated control groups of patients with gender dysphoria is unethical. For that reason, the majority of studies investigating the impact of gender affirming medical interventions are observational. This is not uncommon. For example, "Despite GnRH analogue treatment being used in precocious puberty for more than 20 years, there are no randomized controlled trials to evaluate the effect of GnRHa on a final height compared with untreated controls." (Mul, et al., 2008). However, there are several studies

which demonstrate the safety and positive impact of gender affirming medical interventions. Additionally, larger longitudinal studies are currently underway to help substantiate the significant existing data we have. (de Vries, et al, 2021; Weinand, 2015).

55. Additionally, although it is not possible to ethically conduct randomized control trials for gender-affirming care, we have a large de facto group of untreated individuals with gender dysphoria who experience significant psychiatric symptoms because of widespread barriers to access to care. Clinicians who are competent in the care of transgender individuals practice according to a "first do no harm" ethic which understands that doing nothing is not a neutral option for those with gender dysphoria. Multiple studies have demonstrated the safety of gender affirming hormones, and a growing body of evidence does the same with regards to the safety of GnRH analogs. (Kuper, et al., 2020; Chew, et al., 2018; Colton-Meier, et al., 2011). The same is true with regards to surgery. (Marano, et al., 2021; Olson-Kennedy, et al., 2018; Murad, et al., 2010; Smith, et al., 2005; Pfafflin & Junge, 1998).

56. Dr. Levine inaccurately suggests that diagnosis of gender dysphoria is done solely through a patient's self-diagnosis. Levine Report ¶ 148. His critique demonstrates a fundamental misunderstanding of how gender affirming care is provided. While we have continued to attain a greater understanding about the etiology of gender incongruence, patients do not "self-diagnose," as Dr. Levine suggests. However, it is not unusual or extraordinary in medicine for a provider to consider patients' reports of their symptoms as part of the medical assessment. Much like the diagnosis of many clinical conditions, providers rely on self-report to ascertain accurate diagnoses. Consider the diagnosis of chronic fatigue. The diagnostic criteria for this diagnosis include the following: fatigue so severe that it interferes with the ability to engage in pre-illness activities; of new or definite onset (not lifelong); not substantially alleviated

by rest; worsened by physical, mental or emotional exertion. Like gender dysphoria, these diagnostic criteria are a subjective telling of an individual's personal experience. It is incumbent upon providers of gender affirming care to acquire skills that help them ascertain many details about their patient's gender experience including but not limited to the history, developmental trajectory and expectations regarding treatment options.

57. Dr. Levine also discusses the increase in numbers of youth presenting for care related to GD in recent years. Levine Report ¶ 59. For one, varying estimates of prevalence are the result of inconsistent measures of transgender populations. Some studies have assessed the fraction of a population which had received the DSM-IV diagnosis of GID or the ICD 10 diagnosis of transsexualism, both of which were limited to clinical populations who sought a binary transition (male-to-female or female-to-male). For example, the prevalence reported in DSM-5 (0.005–0.014% for birth-assigned males; 0.002–0.003% for birth-assigned females) are based on people who received a diagnosis of GID or transsexualism and were seeking hormone treatment and surgery from gender specialty clinics, and, therefore, do not reflect the prevalence of all individuals with gender dysphoria or who identify as transgender. Other studies have reported on those who self-identified as transgender or gender incongruent and found that measuring self-identity yields much higher numbers. In 2016, data from the Center for Disease Control's Behavioral Risk Factor Surveillance System suggested that 0.6% of U.S. adults identify as transgender, double the estimate utilizing data from the previous decade. (Byne, et al., 2018). Ultimately, there is nothing surprising about the fact that more transgender people have begun identifying themselves to others as societal stigma has started to abate.

58. Dr. Levine further suggests that after "self-diagnosis" transgender patients will receive "rapid approval" for medical interventions. Levine Report ¶ 148. Self-reporting of

symptoms, as discussed above, is considered by the medical community to be an important aspect of history taking to assist professionals in the process of providing a diagnosis, but it is only part of the process. While many patients may have an acute understanding that they are experiencing gender dysphoria, providers in this field rely on their own understanding and clinical experience in working with patients with GD in order to exercise professional judgment while making this diagnosis and providing recommendations for care. Rather than providing hasty approval as Dr. Levine suggests, the process is careful, thoughtful, and considered. If anything, historically , unnecessarily long periods of psychiatric evaluation were required prior to initiating any medical intervention because gender incongruence was considered a psychopathologic condition.

59. Dr. Levine claims that there is a lack of consensus among psychiatrists and psychotherapists about the cause of, and therapeutic response to, gender dysphoria and because of this, the field is experimental. The entire field of medicine is dynamic, growing as more information becomes available. This does not preclude professionals from providing interventions and necessary care. For example, in the field of cancer care a more complete understanding of how cancer is acquired, spread, and contained leads to improvement in chemotherapy, as well as other modalities for intervention. Whether or not individuals consider the field of cancer to be experimental or not is irrelevant and does not preclude practitioners from providing available treatment.

60. In his discussion about "biology," Dr. Levine makes several assertions that bear examination. First, Dr. Levine references that no matter how many endocrinological or surgical procedures an individual undergoes, they can never be made a "complete man" or "complete woman," reserving that label to those who possess the germinal cells of ovaries or testes and can

reproduce. Levine Report ¶ 18. Terms like "complete man" or "complete woman" are not scientific, as even recognized by the work of Magnus Hirschfield over a century ago and ignore the current scientific understanding of sex. Note that, as described above, there are multiple sex characteristics. Indeed, aside from its offensiveness, Dr. Levine's opinion would mean that people born with differences in sex development (DSD) conditions could not also be considered a "complete man" or "complete woman."

61. Dr. Levine also references "rapid-onset gender dysphoria," and critiques WPATH for not discussing it in WPATH's upcoming eighth version of the Standards of Care. Levine Report ¶ 79. This is a fabricated name for a fabricated entity that arose out of a deeply flawed research endeavor that gathered parents of youth with gender dysphoria from distinctly anti-gender affirming websites. (Littman 2021.) Investigators and clinicians who practice in this area of expertise do not utilize this terminology.

62. Dr. Levine asserts that a disproportionate number of children from communities of color are diagnosed with gender dysphoria. Levine Report ¶ 156. This is patently untrue across the United States. In fact, the opposite is true. The youth seeking puberty suppression do not deviate significantly from general demographics; indeed, the preponderance of youth seeking puberty suppression are of European descent. In any event, this is irrelevant. Many medical conditions impact some communities more than others. (Manton, et al., 1997). That is not a reason to deny medically necessary health care. Additionally, nothing in the science supports withholding medically necessary care from patients simply because they are neuro-diverse, and the predominant recommendation is certainly not that individuals with ASD be denied care related to their gender dysphoria simply for being neuro-atypical.

63. Dr. Levine distorts the literature to suggest that gender-affirming care does not lower suicidality, and indeed insinuates that such care may contribute to suicidality. *See, e.g.*, Levine Report ¶ 95. Dr. Levine misuses the data, specifically, the Cecilia Djhene manuscript about suicidality among transgender women who underwent genital surgery compared to the entire population statistics. This research has been consistently misused, much to the dismay of the first author, whom I have communicated with about this very issue. The Dhejne study specifically states that, "For the purpose of evaluating whether sex reassignment is an effective treatment for gender dysphoria, it is reasonable to compare reported gender dysphoria pre and post treatment. Such studies have been conducted either prospectively or retrospectively and suggest that sex reassignment of transsexual persons improves quality of life and gender dysphoria." Dr. Levine's characterization of the Dhejne research is misleading, because the two comparison groups were transgender women who underwent surgery and aged matched individuals from the general population of Sweden. It is well known that transgender individuals have a higher suicide rate than cisgender individuals. That is explained by the fact that transgender people, even after obtaining gender affirming care, suffer from large and disproportionate rates of discrimination, harassment, family rejection, and violence, all of which could contribute to larger suicidality rates when compared to the general population. Additionally, the data in the Dhejne study was gathered from patients seeking surgery between 1973 and 2003. The political and cultural context is vastly different in 2021 and the surgical techniques are improved.

64. Dr. Levine further equates participants who are lost to follow up as a potential indicator of desistance and/or who regretted undergoing medical interventions, Levine Report ¶ 92, but he provides no support for his assertions. A recent study confirms that the majority of

people who detransition do so because of external factors such as pressure from family and societal stigma. (Turban, et al., 2021). In addition, studies show that regret rates for those who have undertaken gender affirming care are extremely low. (Narayan, et al., 2021; Wiepjes, et al., 2018).

65. Dr. Levine and others who espouse similar "concern" about gender affirming medical procedures have had decades to test their own recommendations about how gender dysphoria should be managed. Nothing scientific has come from those efforts, except several accountings of the negative sequelae experienced by many who underwent conversion therapy. Dr. Levine also assumes that gender-affirming care focuses only on moving youth down a transgender pathway, without spending any time or effort addressing the young person's mental health. Both of these claims are false. Mental health practitioners who are practicing an affirming model of care are providing a safe space in which mental health symptoms or issues can be identified and addressed. Conversion therapy is not supported by any scientific evidence or rigorous data.

66. It is concerning that Dr. Levine consistently misrepresents the affirmative model of care, particularly in pre-pubertal children. Affirming approaches promote exploration of gender development and self-definition within a safe setting. A fundamental concept of this approach is that gender diversity is not a mental illness. The gender affirmative model is defined as a method of therapeutic care that includes allowing children to speak for themselves about their gender identity and expressions and providing support for them to evolve into their authentic gender selves. Support is not characterized by "encouraging" children or youth to be transgender or not. Interventions may include social transition, the changing of one's

presentation to more closely align with one's gender, as well as later medical interventions after the onset of puberty, such as puberty blockers, hormones, or surgery.

67. Dr. Levine asserts that there is a growing body of evidence that suggests that affirmation of gender diverse children results in a higher likelihood of persistence of gender incongruence. He cites an article entitled "The myth of persistence: Response to 'A critical commentary on follow-up studies and 'desistance' theories about transgender and gender non-conforming children'" by Temple Newhook et al. (2018) written by Ken Zucker. This is not a research article. It simply provides a rebuttal by Dr. Zucker to a previous manuscript. In it Dr. Zucker reviews some of the existing literature about persistence and desistance of gender incongruence among children over time. As previously noted, though, the studies upon which Dr. Zucker relies were based on the now obsolete and overly broad categorizations contained in the diagnosis for "Gender Identity Disorder in Children." None of the studies use the current DSM-5 "Gender Dysphoria in Children" diagnosis. Thus, the desistance rates of which Dr. Levine speaks include children who did not identify as transgender to begin with or would be considered "sub-threshold" for a Gender Dysphoria diagnosis. In addition, research shows that children who identify as transgender into adolescence, which is when any medical treatment begins, persist in their transgender identity. (de Vries, et al., 2011).

68. Dr. Levine attempts to create a causal relationship by asserting that gender affirmation (social transition specifically) in childhood causes children to continue to assert a gender incongruent with the sex they assigned at birth and that they would not have done so had they not undergone social transition. There is a failure to consider the clinical observation that children who end up socially transitioning are often experiencing the greatest distress about their gender incongruence, a discussed predictor of persistence. He presents an argument against

affirmation, social transition in particular, in light of the data that suggests the majority of children with gender non-conforming behaviors in childhood grow out of those behaviors and feelings as they move into adolescence. However, research shows that that gender identification does not meaningfully differ before and after social transition. (Rae, et al., 2019).

69. Even if we fully embrace the idea that most children who are gender non-conforming in childhood do not go on to assert gender incongruence in adolescence, it has no relevance to the medical treatment of adolescents and adults who do have gender incongruence, which is the subject of this case. The question is not "should we provide access to medical interventions for people who had GD in childhood that dissipated in adolescence?" because that population is not the population presenting for treatment and medical care is not indicated for that population of children. Transgender adolescents and adults with gender dysphoria are the patients we are discussing.

70. Dr. Levine goes on to discuss the purported lack of quality evidence regarding the impact of gender affirming interventions. Like all areas of medicine, clinical care often outpaces the science in various respects, as is the case with transgender youth care. But the current evidence base for treatment of transgender youth is commensurate with the evidence base for many other types of treatment for adolescents. Additionally, the increase in younger patients seeking services is being paralleled by the increase in data collection, with the promise of the creation of a rich database to better answer some of the still unanswered questions. Nothing about that is unique to gender dysphoria. Additionally, many existing studies have small numbers of participants because transgender experience is uncommon, there exist multiple barriers to accessing services, and there is a historical mistrust of medical institutions based on

an unimaginable amount of harm that such institutions have perpetuated upon this vulnerable community. (Sharman, 2016).

71. Dr. Levine also "wonder[s]" whether using medications off label (i.e., without formal approval by the U.S. Food and Drug Administration) is supportable. Levine Report ¶ 103. But it is common for medications to be used "off label" across all domains of medicine. In addition to there being fewer studies in children and adolescents, pharmaceutical companies often do not want to spend the money to get an FDA indication for use in a very small population.

72. Dr. Levine critiques the Endocrine Society's guidelines for the treatment of gender dysphoria, Levine Report ¶ 104, observing that the guidelines grade the evidence supporting hormone interventions for adolescents as low quality. Dr. Levine fails to understand that this is typical of clinical guidelines for many widely accepted types of care. In fact, within the Endocrine Society Guidelines for the clinical care of pediatric obesity, 48% of the recommendations are graded as very low-quality evidence. By contrast, within the Endocrine Society Gender Dysphoria/Gender Incongruence guidelines, only 23% of the recommendations are graded as very low-quality evidence. It is unlikely that Dr. Levine would suggest we don't treat pediatric obesity because the recommendations are based on low quality evidence. This is another example of applying a different standard to gender affirming care than to other areas of medicine.

73. Dr. Levine asserts that there is no data to suggest that affirmation will lower suicide deaths more than a psychotherapeutic model or watchful waiting, *see e.g.*, Levine Report ¶ 95, while also admitting that his preferred model of providing psychotherapy and withholding other medical interventions is "lacking in long-term evidence" and "quality evidence." Levine

Report ¶¶ 37, 160. However, there is evidence that youth who have been exposed to both a psychotherapeutic model and/or a watchful waiting approach, which deny affirmation and withhold medical care from adolescents, have died by suicide. It does not seem logical to keep employing a method that has been unsuccessful in preventing such deaths. His criticism of Jack Turban's manuscript includes that there was a high level of suicidality (both ideation and attempts) among both those who wanted and received blockers and those who wanted and did not receive blockers. Levine Report ¶ 114. That was not the thrust of the article. The article was demonstrating a decrease of suicidal ideation among the cohort that got blockers. While it is true that the raw data indicates that a higher percentage of the group that had access to puberty blockers had hospitalizations related to suicide attempts (n=5), this difference was not statistically significant, whereas lifetime suicidal ideation was statistically significantly lower in that cohort.

74. Dr. Levine opines about what he believes are a series of health risks related to gender-affirming care. Levine Report ¶¶ 118-144. As a psychiatrist, Dr. Levine is not qualified to offer opinions on several of these topics, including the intricacies of gender-affirming surgery. Nonetheless, I respond to several of his claims here.

75. **Disease and mortality:** Dr. Levine suggests that a series of health effects are associated with gender-affirming care, Levine Report ¶¶ 119-123, but his claims simply demonstrate a lack of familiarity with how these medical interventions are provided. For example, he cites risks involved with hormonal interventions, Levine Report ¶¶ 119, 122-123, but seems unfamiliar with the fact that newer evidence is demonstrating that the cardiovascular risk from gender affirming hormones is actually much lower than previously thought, as even one of his own sources show. As with every area of medicine, the risks and benefits of treatment

are discussed with the patient, and patients are monitored to ensure that their risk profile remains within the normal range.

76. Dr. Levine also refers briefly to puberty-delaying treatment as affecting height. Levine Report ¶ 134. The use of puberty blockers may impact height, but primarily providing an opportunity for transmasculine youth to grow taller, which is generally a desirable feature. Additionally, Dr. Levine makes the conclusory allegation that "[s]hortened life expectancy has been repeatedly documented." Levine Report ¶ 120. To the extent he suggests that gender-affirming care reduces one's life expectancy, no data support that exclusion—in fact, as discussed above they support the opposite conclusion.

77. **Fertility:** Dr. Levine expresses concerns about fertility from surgery and cross-sex hormone therapy, and claims (inaccurately) that puberty blockers may cause infertility as well. Levine Report ¶¶ 127-128. Aside from this being an overly simplistic perspective about a significantly long and complicated process, it is wholly divorced from the reality of care for transgender people. First, like all health care, gender affirming care for every transgender person is individualized. There simply is no one specific route. Second, there is no evidence that affirmation of pre-pubertal children in their identity or the provision of puberty blockers lead to sterility. Indeed, the effects of puberty blockers are reversible. To the extent a person desires and needs hormone treatment or surgery, such care is not provided until well into maturity and after discussion of the effects of such. In addition, patients who may need a procedure or treatment that will result in the side-effect of sterility are informed of such consequences and are provided with alternative options such as fertility preservation before initiating such care. (Chen, et al., 2017).

78. **Sexual function:** Dr. Levine asserts, without support, that puberty blockers may contribute to lack of sexual function. When data are lacking, we rely on clinical experience gathered from patient care and conversations as well as existing data on extrapolatable cases. There is a body of research on the capacity of minors for sexual arousal and orgasm, and there is no data to support the idea that gender affirmation diminishes sexual capacity. More commonly youth with GD experience dysphoria from the act of masturbation, and often even the possibility or thoughts around sexual intimacy. In fact, there is data that demonstrates improved satisfaction with sexual intimacy after gender affirming interventions.

79. Informed consent is the legal embodiment of the concept that each individual has the right to make decisions affecting their health. Physicians engaged in patient-physician relationships involving medical informed consent have a moral responsibility to identify the best treatments for each patient on the basis of available medical evidence and to discuss with patients the hoped-for benefits and the potential risks. Physicians must allow for patients' questions about the proposed treatments, benefits, and risks and must answer those questions from the available medical literature and their professional experience. This exchange of information and ideas is the foundation of the patient-physician partnership and promotes informed decision making in the most complex medical situations. (Paterick, et al., 2008). As noted above, speaking from my own clinical experience, at our center we strive to ensure that we are obtaining informed consent from every patient (and their parent/guardian) throughout the course of treatment.

80. **Psychosocial effects:** Dr. Levine expresses concern that puberty-delaying medication will "halt" maturation, and cause transgender adolescents to "undergo[] puberty at a substantially older age." Levine Report ¶¶ 134-135. Dr. Levine's assertion that puberty

suppression for a limited time has adverse effects on cognition is not supported by evidence within the realm of transgender youth care. Additionally, puberty suppression does not impact somatic growth, or emotional maturation. Dr. Levine fails to point out that experiencing the changes of a puberty that does not align with one's gender identity creates significant problems for transgender and nonbinary youth, including an exacerbation of anxiety, depression, isolation and sometimes poor coping mechanisms including self-harm and substance abuse. Youth going through an endogenous puberty that does not align with their gender express that it is difficult for them to participate in school, therapy, family and social activities. Most youth who utilize puberty blockers will likely go on to add exogenous hormones so that they do undergo puberty on the somewhat older side of normal range, but still well within normal range.

III. CONCLUSION

81. In conclusion, I do not disagree that, as with every field of medicine, there is more to learn in the field of transgender youth care. That is why I became an investigator. However, there is room to provide gender affirming medical interventions in a thoughtful manner that extrapolates from relevant fields of science and medicine, existing data, and clinical expertise while simultaneously carrying out further investigations. The denial of much needed care only serves to harm transgender people.

I declare under penalty of perjury under the laws of the United States of America that the foregoing is true and correct. Executed this 17 day of March, 2022.

Johanna Olson-Kennedy (Mar 17, 2022 10:26 PDT)

Johanna Olson-Kennedy, M.D., M.S.

Subscribed and sworn before me, a Notary Public in and for the County of Norfolk, State of Virginia, this 17 day of March, 2022.

KETSIA MCCLEASE
Electronic Notary Public
Commonwealth of Virginia
Registration No. 327724
My Commission Expires Apr 30, 2023

Ketsia McClease
Signature of Notary

This notarial act was performed online by way of two-way audio/video communication technology.

1647520704-olson-kennedy-rebuttal-report_final

Final Audit Report 2022-03-17

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Exhibit A

CURRICULUM VITAE
JOHANNA OLSON-KENNEDY MS, MD
MAY 8, 2021

EDUCATION AND PROFESSIONAL APPOINTMENTS

EDUCATION:

Year	*Degree, Field, Institution, City*
1992	BA, Mammalian Physiology, UC San Diego, San Diego
1993	MS, Animal Physiology, The Chicago Medical School, Chicago
1997	MD, Medical Doctor, The Chicago Medical Shool, Chicago
2015	MS, Clinical and Biomedical Investigations in Translational Science, USC, Los Angeles

POST-GRADUATE TRAINING:

Year-Year	*Training Type, Field, Mentor, Department, Institution, City*
1997-1998	Internship, Pediatrics, Children's Hospital Orange County, Orange
1998-2000	Residency, Pediatrics, Antonio Arrieta, Children's Hosptial Orange County, Orange
2000-2003	Fellowship, Adolescent Medicine, Children's Hospital Los Angeles, Los Angeles
2012-2015	Master's Degree, Clinical and Biomedical Investigations in Translational Science, USC

ACADEMIC APPOINTMENTS:

Year-Year	*Appointment*	*Department, Institution, City, Country*
2012-present	Medical Director	The Center for Transyouth Health and Development, Division of Adolescent Medicine, Children's Hospital Los Angeles, Los Angeles, USA
2008-2012	Fellowship Director	Division of Adolescent Medicine, Children's Hospital Los Angeles, Los Angeles, USA
2006-2016	Assistant Professor of Clinical Pediatrics	Division of Adolescent Medicine, Children's Hospital Los Angeles/USC Keck School of Medicine, Los Angeles, USA
2016 - Present	Associate Professor of Clinical Pediatrics	Division of Adolescent Medicine, Children's Hospital Los Angeles/USC Keck School of Medicine, Los Angeles, USA

LICENSURE, CERTIFICATIONS

LICENSURE:

Year	*License number, State, Status*
2000	A-67352, California, Active

BOARD CERTIFICATION OR ELIGIBILITY:

Year	*Board, State, Status*
2001, 2009, 2015	Pediatrics, California, active

SPECIALTY CERTIFICATION:

Year	*Specialty Certification, Status*
2003, 2013	Adolescent Medicine, California, active

HONORS, AWARDS:

Year	*Description*	*Awarding agency, address, city*
2019	Benjamin Meaker Visiting Professorship	University of Bristol, Bristol UK
2015	The Champion Award	The Division of Adolescent Medicine; CHAMPION FUND 5000 Sunset Blvd. Los Angeles
2014	Recognition Award for Outstandin Compassionate and Innovative Service	SoCal Society for Adolescent Health and Medicine Regional Chapter, Los Angeles
2014	Anne Marie Staas Ally Award	Stonewall Democratic Club; 1049 Havenhurst Drive #325, West Hollywood
2012	Extraordinary Service Award	Equality California, 202 W 1st St., Suite 3-0130 Los Angeles
2010	Clinical Research Academic Caree Development Award	Saban Research Center TSRI Program: Community Health Outcomes and Intervention, Los Angeles
2009	Health Care Advocacy Champion	Democratic Advocates for Disability Issues, Los Angeles

TEACHING

UNDERGRADUATE, GRADUATE AND MEDICAL STUDENT (OR OTHER) MENTORSHIP:

Year-Year	*Trainee Name*	*Trainee Type*	*Dissertation/Thesis/Project Title*
2020-Present	Richard Mateo Mora	MD	Fertility Preservation Among Transgender Women
2019-2021	Laer Streeter	MD	Comparison of Histrelin Implants
2016-Present	Jonathan Warus	MD/KL2	Affecting Pre-Exposure Prophylaxis (PreP) Decision Making to Improve Youth Engagement in HIV Prevention Services
2015-2020	Shannon Dunlap	PhD	Developmental Aspects of Gender Non-Conformong Youth
2015-2016	David Lyons	MD	Transgender Youth Clinical Clerkship
2014-2015	Michael Haymer	MD	Transgender Youth Clinical Clerkship

POSTGRADUATE MENTORSHIP:

Year-Year	*Trainee Name*	*If past trainee, current position and location*
2020-Present	Marianela Gomez-Rincon	Adolescent Medicine Fellow
2015-2018	Jonathan Warus, MD	Faculty, CHLA/USC Keck School of Medicine
2015-2017	Patrick Shepherd, MD	CHLA Endocrinology Fellow
2014	Julie Spencer, MD	Adolescent Medicine Provider Kaiser Hospital
2013	Shelley Aggarwal, MD	Clinical Instructor – StanfordUniversity School of Medicine
2012-2013	Lisa Simons, MD	Clinical Instructor – Lurie Children's Hospital

SERVICE

DEPARTMENT SERVICE:

Year-Year	*Position, Committee*	*Organization/Institution*
2010-2015	Secretary, The CHAMPION Fund Executive Board	The Division of Adolescent Medicine, Children's Hospital Los Angeles

PROFESSIONAL SERVICE:

Year-Year	*Position, Committee*	*Organization/Institution*
2012-present	Member, LGBT Special Interest Group	Society for Adolescent Health and Medicine

CONSULTANTSHIPS AND ADVISORY BOARDS:

Year	*Position, Board*	*Organization/Hospital/School, Institution*
2017 - Present	Board Member	US Professional Association of Transgen Health
2021	Member, Advisory Board	The National LGBTQIA+ Health Education Center
2010-2017	Member, Advisory Board	Transyouth Family Allies
2017-present	Member, National Medical Committee	Planned Parenthood

PROFESSIONAL SOCIETY MEMBERSHIPS:

Year- Year	*Society*
2017 - present	US Professional Association for Transgender Health
2014-present	Society for Pediatric Research
2010-present	World Professional Association for Transgender Health
2006-2011	Los Angeles Pediatric Society (Past president 2010)
2005-2012	American Academy of Pediatrics
2003-present	Society for Adolescent Health and Medicine

MAJOR LEADERSHIP POSITIONS: (E.G., DEAN, CHAIR, INSTITUTE DIRECTOR, HOSPITAL ADMINISTRATION, ETC.)

RESEARCH AND SCHOLARSHIP

EDITORSHIPS AND EDITORIAL BOARDS:

Year-Year	*Position*	*Journal/Board Name*
2015-present	Associate Editor	Journal of Transgender Health

MANUSCRIPT REVIEW:

Year-Year	*Journal*
2018-present	Journal of Transgender Health
2018 - present	Clinical Child Psychology and Psychiatry
2018 - present	Journal of Sexual Medicine
2015-present	Journal of Transgender Health
2014-present	International Journal of Transgenderism
2014-present	LGBT Health
2014-present	Journal of Adolescent Health
2014-present	Pediatrics

MAJOR AREAS OF RESEARCH INTEREST

Research Areas
1. Gender diverse children, transgender youth and young adults
2. HIV medication adherence

GRANT SUPPORT - CURRENT:

Grant No. (PI)2R01HD082554-06A1	*Dates of Award: 2021-2026*
Agency: NICHD	*Percent Effort 25%*
Title: The Impact of Early Medical Treatment in Transgender Youth	
Description: This is the continuations of a multicenter study, the first of its kind in the U.S. to evaluate the long-term outcomes of medical treatment for transgender youth. This study will provide essential, evidence-based information on the physiological and psychosocial impact, as well as safety, of hormone blockers and cross-sex hormones use in this population.	
Role: Principle Investigator	
*Total Direct Costs: $*4,918,586	

Grant No. (PI) 1R01HD097122-01	*Dates of Award: 2019-2024*
Agency: NICHD	*Percent Effort 10%*
Title: A Longitudinal Study of Gender Nonconformity in Prepubescent Children	
Description: The purpose of this study is to establish a national cohort of prepubertal transgender/gender nonconforming (TGNC) children (and their parents), and longitudinally observe this cohort to expand the body of empirical knowledge pertaining to gender development and cognition in TGNC children, their mental health symptomology and functioning over time, and how family-initiated social gender transition may predict or alleviate mental health symptoms and/or diagnoses.	
Role:Co-Investigator	
Total Direct Costs: $2,884,950	

GRANT SUPPORT - PAST:

Grant No. (PI) 1RO1HD082554-01A1	*Dates of Award: 2015-2020*
Agency: NICHD	*Percent Effort 45%*
Title: The Impact of Early Medical Treatment in Transgender Youth	
Description: This is a multicenter study, the first of its kind in the U.S. to evaluate the long-term outcomes of medical treatment for transgender youth. This study will provide essential, evidence-based information on the physiological and psychosocial impact, as well as safety, of hormone blockers and cross-sex hormones use in this population.	
Role: Principle Investigator	
*Total Direct Costs: $*4,631,970	
Grant No. (COI) R01AI128796-01	Dates of Award: 2/24/17-1/31/18
Agency: NIAID	Percent Effort: 5%
Title: Maturation, Infectibility and Trauma Contributes to HIV Susceptibility in Adolescents	
Description: This proposal explores the overarching hypothesis that fluctuations in sex steroid levels and mucosal trauma (sexual activity) are key determinants of mucosal immune activation and epithelial integrity, and that microbial communities are central to these processes. We will pursue this hypothesis by examining longitudinal changes in the anogenital microbiome as well as protein expression at these mucosal sites during sexual maturation (cisgender youth) and in hormonally-controlled sexual maturation (transgender youth). Associations between sex steroid levels, microbial community composition, mucosal trauma, and vaginal proteins will be determined and modeled.	
Role: Co-Investigator	
Total Direct Costs: $44,816	

Grant No. (PI) U01HD040463	*Dates of Award* 2006 – 2016
Agency: NIH/NICHD	*Percent Effort: 10%*
Title: Adolescent Medicine Trials Network for HIV/AIDS	
Description: Adolescent Medicine Trials Network for HIV/AIDS	
Role: Co-Investigator	
Total Direct Costs: 2,225,674	

Grant No. (PI) SC CTSI 8KL2TR000131	*Dates of Award: 2012-2014*

Agency: KL2 Mentored Career Research Development Program of the Center for Education, Training and Career Development	*Percent Effort: 37.5%*
Title: The Impact of Hormone Blockers on the Physiologic and Psychosocial Development of Gender Non-Conforming Peri-Pubertal Youth	
Description: This study aimed to understand the impact of puberty blocking medications on mental health and physiolgic parameters in peri-pubertal transgender youth.	
Role: Principal Investigator	
Total Direct Costs: 191,525	

Invited Lectures, Symposia, keynote addresses

Year	*Type*	*Title, Location*
2021	Invited Lecture	Approach to the Care of Gender Diverse Children and Transgender Youth, USC Keck Medical School, Virtual Lecture
2021	Invited Lecture	Caring for Gender Diverse and Transgender Youth. SLO Acceptance, Cal Poly, Virtual Presentation
2020	Symposium	Trans Youth Care, Chico Transgender Week, Virtual Presentation
2020	Invited Lecture	Gender Nonconforming and Transgender Youth, Novartis, Virtual Presentation
2020	Invited Lecture	Advanced Hormones; More than Just T and E, CHLA, Virtual Presentation
2020	Invited Lecture	Video Telehealth and Transgender Youth, Telehealth Best Practices for the Trans Community, The Central Texas Transgender Health Coalition, Virtual Presentation
2020	Invited Lecture	Caring for Gender Diverse and Transgender Youth , Center for Juvenile Justice Reform Supporting the Well-Being of LGBTQ Youth Certificate Program, Virtual Presentation
2020	Invited Lecture	Gear Talk, Transforming Families, Virtual Lecture
2020	Invited Lecture	Tips for Parenting a Trans or Gender Diverse Youth, Models of Pride, Virtual Presentation
2020	Invited Lecture	Caring for Gender Diverse and Transgender Youth, LGBTQ+ Clinical Academy, Palo Alto University, Virtual presentation
2020	Invited Lecture	Approach to the Care of Gender Non-conforming Children and Transgender Youth, USC Medical School, Los Angeles, CA
2020	Invited Lecture	Medical Interventions for transgender youth, Cal State Los Angeles, Los Angeles
2020	Plenary Session	Understanding Issues Involving Gender Non-Conforming and Transgender Individuals Coming to a Courtroom Near You, Mid-Winter Workshop for Judges of the Ninth Circuit, Palm Springs, CA
2019	Keynote	Transgender Youth Care, SickKids, Toronto, Canada

2019	Symposium	The Care of Trans and Gender Non-Conforming Youth and Young Adults, Cal State Los Angeles, California
2019	Symposium	The Care of Trans and Gender Non-Conforming Youth and Young Adults, Claremont Colleges, California
2019	Symposium	TransYouth Care; Flagstaff, AZ
2019	Keynote	Future Directions, USPATH, Washington DC
2019	Invited Lecture	Just a Boy, Just a Girl, Gender Odyssey San Diego, San Diego, CA
2019	Invited Lecture	Hormonas que Affirman el Genero pasa Juventud y Adultos Menores Trans, Transformando Desde el Amor y Las Familias, Colombia
2019	Invited Lecture	Infancia Trans y da Genero Diverso, Transformando Desde el Amor y Las Familias, Colombia
2019	Invited Lecture	Gender Dysphoria; A Deeper Dive Beyond the Diagnosis, Keynote address, Inaugaral LGBTQ summit, Santa Clara CA
2019	Invited Lecture	Transgender and Gender Non-conforming Youth, Ascend Residential Treatment, Utah
2019	Invited Lecture	Gender Diverse and Transgender Youth; What Pediatricians Should Know, Common Problems in Pediatrics Conference, Utah AAP, Utah
2019	Invited Lecture	Gender Diverse and Transgender Youth; What Pediatricians Should Know, Common Problems in Pediatrics Conference, Utah AAP, Utah
2019	Invited Lecture	Caring for Gender Diverse and Transgender Youth, Grand Rounds, UCLA Olive View, CA
2019	Invited Lecture	Caring for Gender Diverse and Transgender Youth, Grand Rounds, Good Samaritan, CA
2019	*Invited Lecture*	Gender Dysphoria; A Deeper Dive Beyond the Diagnosis, Advance LA Conference, California
2019	Invited Lecture	Puberty Suppression and Hormones; Medical Interventions for Transgender Youth, USC Keck School of Medicine Reproductive Health Section. Los Angeles, CA
2019	Invited Lecture	Transgender Youth: Medical and Mental Health Needs, Bristol, United Kingdom
2019	Invited Lecture	Rethinking Gender, University of Bristol, United Kingdom
2019	Invited Lecture	Puberty Suppression in Youth with Gender Dysphoria, Fenway Trans Health Program, Boston
2019	Invited Lecture	Recognizing the Needs of Transgender Youth, California Department of Corrections And Rehabilitation, Ventura, CA
2019	Invited Lecture	Gender Dysphoria; Beyond the Diagnosis, Gender Education Demystification Symposium, GA
2019	Invited Lecture	Caring for Gender Nonconforming and Transgender Youth, Los Angeles Superior Court/Los Angeles Bar Association Training, CA
2019	Invited Lecture	Supporting Gender Diverse and Transgender Youth; A Deeper Look at Gender Dysphoria, Oakwood School, CA

2018	Invited Lecture	Chest Reconstruction and Chest Dysphoria in Transmasculine Adolescents and Young Adults: Comparison of Nonsurgical and Postsurgical Cohorts, Buenos Aires, Argentina
2018	Invited Lecture	Transyouth Care – An NIH Multisite Study About the Impact of Early Medical Treatment in Transgender Youth in the US, Buenos Aires, Argentina
2018	Invited Lecture	Transgender Youth and Gender Affirming Hormones; A 6-8 year follow-up, Buenos Aires, Argentina
2018	Invited Lecture	Supporting Gender Diverse and Transgender Youth: A Deeper Look at Gender Dysphoria, Studio City, CA
2018	Invited Lecture	Gender Dysphoria: Beyond the Diagnosis, Washington DC
2018	Invited Lecture	Uso de Hormonas Reaffirmantes de Genero en Adolescentes Transgenero, Trans Amor Congreso Nacional de Transexualidad Juvenil y Infantos, Monterey, Mexico
2018	Invited Lecture	Bloquedores de la Pubertad, Trans Amor Congreso Nacional de Transexualidad Juvenil y Infantos, Monterey, Mexico
2018	Invited Lecture	Working with Trans and Gender Non-Conforming Youth, Children's Hospital Orange County, CA
2018	Invited Lecture	Caring for gender Non-conforming and Transgender Youth and Young Adults, Ascend Residential, Encino CA
2018	Invited Lecture	Gender Dysphoria; Beyond the Diagnosis; Midwest LGBTQ Health Symposium, Chicago, IL
2018	Invited Lecture	Caring for gender Non-conforming and Transgender Youth and Young Adults, California State University Northridge, Northridge, CA
2018	Invited Lecture	Puberty Suppression and Gender Affirming Hormones, Gender Fest, Las Vegas, NV
2018	Invited Lecture	Gender Google; Gender Odyssey Family Conference, Seattle WA
2018	Invited Lecture	Gender Dysphoria; Beyond the Diagnosis, Gender Odyssey Family Conference, Seattle WA
2018	Invited Lecture	Puberty Suppression: What, When, and How, Gender Odyssey Family Conference, Seattle WA
2018	Invited Lecture	Gender Dysphoria; School Nurse Organization of Idaho Annual Conference, Idaho
2018	Invited Lecture	Understanding Gender Dysphoria, Gender Spectrum Family Conference, Moraga, CA
2018	Invited Lecture	Puberty Suppression and Gender Affirming Hormones, Gender Odyssey Family, Los Angeles, CA
2018	Invited Lecture	Gender Dysphoria – Beyond the Diagnosis, Gender Odyssey Family, Los Angeles, CA
2018	Invited Lecture	Gender and What You Should Know, Archer School for Girls, Brentwood, CA
2018	Symposium	Caring for Gender Non-Conforming and Transgender Youth, TransYouth Care, Oceanside, CA

2018	Invited Lecture	Gender Dysphoria: Beyond the Diagnosis, Advance LA, Los Angeles, CA
2018	Invited Lecture	Caring for Gender Non-Conforming and Transgender Youth, Andrology Society of America Clinical Symposium, Portland, OR
2018	Symposium	Caring for Gender Non-Conforming and Transgender Youth, TransYouth Care, Los Angeles, CA
2018	Invited Lecture	Caring for Gender Non-Conforming and Transgender Youth, Center for Early Education, Los Angeles, CA
2017	Symposium	Caring for Gender Non-Conforming and Transgender Youth, TransYouth Care, Santa Barbara, CA
2017	Invited Lecture	Gender Dysphoria, Beyond the Diagnosis, Pink Competency, Oslo Norway
2017	Invited Lecture	"Just a Boy, Just a Girl" Gender Infinity, Houston TX
2017	Invited Lecture	Caring for Gender Non-Conforming Children and Transgender Adolescents:
		A United States Perspective, Pink Competency, Oslo Norway
2017	Invited Lecture	Gender Dysphoria; Beyond the Diagnosis, Models of Pride, Los Angeles, CA
2017	Invited Lecture	Puberty Delay and Cross Hormones for Trans* Youth, Models of Pride, Los Angeles, CA
2017	Invited Lecture	Healthcare for TGNC Youth, Expanding Competency for LGBT Youth in the System, Washington DC
2017	Invited Lecture	Gender Non-conforming and Transgender Children and Youth; Center for Early Education, West Hollywood, CA
2017	Invited Lecture	Rethinking Gender, University of Massachusetts
		Annual Convocation Welcome Luncheon, Worcester, MA
2017	Invited Lecture	Puberty Delay and Cross Hormones for Trans* Youth, Gender Odyssey Family Conference, Seattle, WA
2017	Invited Lecture	Puberty Suppression; What, When and How, Gender Odyssey Family Conference, Seattle, WA
2017	Invited Lecture	Just a Boy, Just a Girl, Gender Odyssey, Los Angeles, California
2017	Invited Lecture	Puberty Blockers and Cross Sex Hormones, Gender Odyssey, Los Angeles, California
2017	Invited Lecture	Caring for Gender Non-conforming and Transgender youth and Young Adults, Diverse Families Forum: The Importance of Family Support In The Trans And LGBT Children, Organized by COPRED and The International Association Of Families For Diversity (FDS), Mexico City, Mexico
2017	Invited Lecture	Gender Non-Conforming Children and Transgender Youth, Board of Behavioral Sciences, Orange, CA
2017	Invited Lecture	Puberty Suppression and Hormones; Medical Interventions for Transgender Youth, Santa Monica Rape Treatment Center, Santa

		Monica, CA
2017	Invited Lecture	Gender Nonconforming and Transgender Youth, CSU Fullerton, Fullerton, CA
2017	Invited Lecture	Rethinking Gender, Chico TransGNC Week, Chico, California
2017	Invited Lecture	Caring for Gender Non-Conforming and Transgender Youth, Chico TransGNC Week, Chico, California
2017	Invited Lecture	Transgender Youth Care in the New Millennium, USC Law and Global Health Initiative, Los Angeles, CA

Invited Grand Rounds, CME Lectures

Year	***Type***	***Title, Location***
2021		
2020	CME Lecture	Histrelin Implants for Suppression of Puberty in Youth with Gender Dysphoria: a Comparison of 50 mcg/day (Vantas) and 65 mcg/day (SupprelinLA), WPATH Conference, Virtual Presentation
2020	CME Lecture	Chest Reconstruction and Chest Dysphoria in Transmasculine Adolescents and Young Adults, Comparison of Post-surgical and Non-surgical Cohorts, WPATH Conference, Virtual Presentation
2020	CME Lecture	Gender Affirmation Through a Social Justice Lens, SAHM Conference, Virtual Presentation
2020	CME Lecture	Introduction to the Care of Gender Diverse and Transgender Youth, AAP Conference, Virtual Lecture
2020	CME Lecture	Conversations with LGBTQ youth; the role of the pediatrician, AAP Conference, Virtual Lecture
2020	Grand Rounds	Creating Affirming Environments for Trans and Gender Diverse Patients, USC OB/Gyn Grand Rounds, Virtual Presentation
2020	CME Lecture	Introduction to the Care of Gender Diverse and Transgender Youth, Resident Lecture, CHLA
2020	CME Lecture	Introduction to the Care of Gender Diverse and Transgender Youth, Facey Medical Group, Los Angeles, CA
2020	Plenary Lecture	Reframing Gender Dysphoria, LEAH Conference, Los Angeles, CA
2020	CME Lecture	Gender Affirming Care for Pre and Peri-pubertal Trans and Gender Diverse Youth, LEAH Conference, Los Angeles, CA
2020	CME Lecture	Introduction to the Care of Gender Diverse and Transgender Youth, Division of Endocrinology, USC, Los Angeles , CA
2020	Plenary Session	Understanding Issues Involving Gender Non-Conforming and Transgender Individuals Coming to a Courtroom Near You, Mid-Winter Workshop for Judges of the Ninth Circuit, Palm Springs, CA
2019	Symposium	Recognizing the Needs of Transgender Youth, California Department of Corrections and Rehabilitation, Stockton, CA

2019	Keynote	Transgender Youth Care, SickKids, Toronto, Canada
2019	Symposium	The Care of Trans and Gender Non-Conforming Youth and Young Adults, Cal State Los Angeles, California
2019	Symposium	The Care of Trans and Gender Non-Conforming Youth and Young Adults, Claremont Colleges, California
2019	CME Lecture	Gender Diverse and Transgender Youth, Harbor UCLA Medical Center Grand Rounds, Torrance, CA
2019	CME Lecture	Gender Dysphoria – Beyond the Diagnosis, Gender Odyssey San Diego, San Diego, CA
2019	CME Lecture	Hormones 201 – Beyond T and E, Gender Odyssey San Diego, San Diego, CA
2019	*Grand Rounds*	Transgender Youth; What's New in 2019?, Children's Hospital Los Angeles, CA
2019	Oral Presentation	Male Chest Reconstruction and Chest Dysphoria in Transmasculine Adolescents and Young Adults, European Professional Association of Transgender Health, Rome Italy
2019	Oral Presentation	Transgender Youth and Gender Affirming Hormones; 5-7 Year Follow Up, European Professional Association of Transgender Health, Rome Italy
2019	CME Educational Lecture	Gender Dysphoria; Beyond the Diagnosis, European Professional Association of Transgender Health, Rome Italy
2019	CME Symposium	Caring for Gender Nonconforming and Transgender Youth, Children's Hospital Orange County, CA
2019	CME Symposium	Caring for Gender Nonconforming and Transgender Youth, Stanislaus County Behavioral Health and Recovery Services, CA
2019	CME Eduational Lecture	Rethinking Gender, Olive View Medical Center Grand Rounds, CA
2018	CME Symposium	Caring for Gender Nonconforming and Transgender Youth, Glendale Unified School District, CA
2018	CME Educational Lecture	Caring for Gender Non-Conforming Children and Transgender Youth, CME by the Sea, CA
2018	CME Symposium	Caring for Gender Non-Conforming and Transgender Youth, TransYouth Care, Austin, TX
2018	CME Educational Lecture	Gender Affirming Hormone Therapy for Transmasculine Adolescents and Young Adults, Gender Infinity, Houston, Texas
2018	CME Educational Lecture	Outside of the Binary; Care for Non-Binary Adolescents and Young Adults, Gender Infinity, Houston, Texas
2018	CME Educational Lecture	Chest Dysphoria and the Impact of Chest Reconstruction, Gender Infinity, Houston, Texas
2018	CME Educational Lecture	Just a Girl, Just a Boy, Gender Infinity, Houston, Texas
2018	CME Educational Lecture	Hormones 201: More than Testosterone and Estrogen, Gender Odyssey Professional Symposium, WA
2018	CME Educational Lecture	Male Chest Reconstruction and Chest Dysphoria in Transmasculine Minors and Young Adults, Gender Odyssey Professional Symposium,

		WA
2018	CME Educational Lecture	Chest Surgery, Gender Spectrum, Moraga, CA
2018	CME Educational Lecture	Understanding Gender Dysphoria, Gender Spectrum, Moraga, CA
2018	CME Educational Lecture	Puberty Suppression and Gender Affirming Hormones, Gender Odyssey, Los Angeles, CA
2018	CME Educational Lecture	Gender Dysphoria – Beyond the Diagnosis, Gender Odyssey, Los Angeles, CA
2018	CME Educational Lecture	Approach to the Care of Gender Non-Conforming Children and Transgender Youth, Desert Oasis Healthcare, Palm Desert, CA
2018	CME Educational Lecture	Puberty Blockers and Gender Affirming Hormones for Transgender Youth: What Do We Know, and What Have We Learned, Pediatric Academic Societies, Toronto, Canada
2018	CME Workshop	Mental and Medical Healthcare for Transgender Adolescents, California Association of Marriage and Family Therapists, Garden Grove, CA
2018	CME Educational Lecture	Approach to the Care of Gender Non-Conforming Children and Transgender Youth, Keck School of Medicine, Los Angeles, CA
2018	Grand Rounds	Caring for Gender Non-Conforming Children and Transgender Adolescents, Primary Children's Hospital, Salt Lake City, UT
2018	CME Educational Lecture	Caring for Transgender Youth, Chico Trans Week, Chico, CA
2018	CME Educational Lecture	Rethinking Gender, UCSD Medical School, San Diego, CA
2018	CME Educational Lecture	Rethinking Gender, UCLA Medical School, Los Angeles, CA
2018	CME Educational Lecture	Transyouth Care – Self-reflection On Personal Biases and Their Impact On Care, Society for Adolescent Health and Medicine, Seattle WA
2018	CME Educational Lecture	Rethinking Gender, Society for Adolescent Health and Medicine, Seattle WA
2018	CME Educational Lecture	Providing 360 degree transgender hormone therapy: beyond the protocols, Medical Directors Council (MeDC) 14th Annual Clinical Update in Reproductive Health and Medical Leadership, Snowbird, Utah
2018	CME Educational Lecture	Gender Dysphoria: Beyond the Diagnosis, Gender Education and deMystification Symposium, Salt Lake City, Utah
2018	CME Educational Lecture - Keynote	Rethinking Gender, SoCal LGBTQIA health conference, Los Angeles, CA
2017	CME Educational Seminar	The Care of Gender Non-Conforming children and Transgender Youth; Orange County Health Care Agency, Orange County, CA
2017	CME Educational Lecture	Rethinking Gender, Adolescent Grand Rounds, Children's Hospital Los Angeles, Los Angeles, CA
2017	CME Educational Lecture	"Just a Boy, Just a Girl" Gender Infinity, Houston TX
2017	CME Educational Lecture	Chest Dysphoria – The Impact of Male Chest Reconstruction, Gender Infinity, Houston TX

2017	CME Educational Lecture	Outside of the Binary; Care for Non-Binary Adolescents and Young Adults, Gender Infinity, Houston TX
2017	CME Educational Lecture	Puberty Blockers; What, When and How, Gender Infinity, Houston TX
2017	CME Educational Lecture	Gender Non-Conforming Children and Transgender Youth, Pasadena CA
2017	CME Educational Lecture	Gender Non-Conforming Children and Transgender Youth; Integrated Care Conference, Los Angeles, CA
2017	CME Educational Lecture	Gender Non-Conforming and Transgender Children and Adolescents; A Multidisciplinary Approach, California Psychiatric Association Annual Conference, Yosemite, CA
2017	CME Educational Lecture	Gender Non-Conforming and Transgender Children and Adolescents, Developmental Pediatrics continuing education lecture, Children's Hospital Los Angeles, CA
2017	CME Educational Lecture	Masculinizing Hormones, Central Texas Transgender Health Conference, Austin, TX
2017	CME Educational Lecture	Children, Youth, Families and Hormone Blockers, Central Texas Transgender Health Conference, Austin, TX
2017	CME Educational Lecture	Chest Dysphoria – The Impact of Male Chest Reconstruction, Gender Odyssey Professional Symposium, Seattle, WA
2017	CME Educational Lecture	Puberty Delay and Cross Hormones for Transyouth, Gender Odyssey Professional Symposium, Seattle, WA
2017	CME Invited Lecture	Just a Girl, Just a Boy, Gender Odyssey Professional Symposium, Seattle, WA
2017	CME Educational Lecture	Gender Dysphoria, Gender Spectrum Family Conference, Moraga, CA
2017	CME Educational Lecture	Care of Gender Non-Conforming Children and Transgender Adolescents, Lopez Family Foundation Educational Lecture, Los Angeles, CA
2017	CME Educational Lecture	Puberty Suppression and Hormones; Medical Interventions for Transgender Youth, USC Keck School of Medicine Reproductive Health, Los Angeles, CA
2017	CME Educational Seminar	Caring for Gender Non-Conforming and Transgender Youth, TransYouth Care, San Diego, CA
2017	CME Educational Lecture	Puberty Suppression in the United States; practice models, lessons learned, and unanswered questions, US Professional Association of Transgender Health, Los Angeles, CA
2017	CME Educational Lecture	The Impact of Male Chest Reconstruction on Chest Dysphoria in Transmasculine Adolescents and Young Men; A Preliminary Study, US Professional Association of Transgender Health, Los Angeles, CA
2017	CME Educational Lecture	Outside of the Binary; Care for Non-Binary Adolescents and Young Adults, US Professional Association of Transgender Health, Los Angeles, CA

PUBLICATIONS:

*** INDICATES TRAINEES**
**** INDICATE YOURSELF AS CO-FIRST OR CO-CORRESPONDING OR SENIOR AUTHORS**

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Olson-Kennedy J,** Cohen-Kettenis P. T., Kreukels B.P.C, Meyer-Bahlburg H.F.L, Garofalo R, Meyer W, Rosenthal S.M., Research Priorities for Gender Nonconforming/Transgender Youth: Gender Identity Development and Biopsychosocial Outcomes, Curr Opin Endocrinol Diabetes Obes. 2016 Jan 27. [Epub ahead of print]: Role: coordinated information from all authors, wrote all drafts, and finalized manuscript; first author

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Health Effects of Transitioning in Teen Years Remain Unknown, NPR

STUDY: Being Young and Trans Is Not the Result of a Hormonal Imbalance

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Parenting My Transgender Teen: Britt Rubenstein, Mom-Momstampblog

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Exhibit B

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EXHIBIT 2

Endocrine Reviews, 2021, Vol. XX, No. XX, 1–40
doi:10.1210/endrev/bnaa034
Scientific Statement

Scientific Statement

Considering Sex as a Biological Variable in Basic and Clinical Studies: An Endocrine Society Scientific Statement

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Abbreviations: ACTH, adrenocorticotropic hormone; AT_2R, angiotensin type 2 receptor; BMI, body mass index; cAMP, cyclic adenosine monophosphate; CKD, chronic kidney disease; CRF, corticotropin-releasing factor; CVD, cardiovascular disease; dMRI, diffusion magnetic resonance imaging; fMRI, functional magnetic resonance imaging; FCG, Four Core Genotypes (model); GMV, gray matter volume; GPCR, G-protein coupled receptor; HPA, hypothalamic-pituitary-adrenal; KYN, kynurenine; LC, locus coeruleus; MIH, Müllerian inhibitory hormone; PAR, pseudoautosomal region; PKA, protein kinase A; PTSD, posttraumatic stress disorder; RAAS, renin-angiotensin-aldosterone system; rs-fMRI, resting state functional magnetic resonance imaging; sMRI, structural magnetic resonance imaging; UCN, urocortin.

Received: 22 December 2020; First Published Online: 11 March 2021; Corrected and Typeset: 11 March 2021.

Abstract

In May 2014, the National Institutes of Health (NIH) stated its intent to "require applicants to consider sex as a biological variable (SABV) in the design and analysis of NIH-funded research involving animals and cells." Since then, proposed research plans that include animals routinely state that both sexes/genders will be used; however, in many instances, researchers and reviewers are at a loss about the issue of sex differences. Moreover, the terms *sex* and *gender* are used interchangeably by many researchers,

ISSN Print: 0163-769X
ISSN Online: 1945-7189
Printed: in USA

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further complicating the issue. In addition, the sex or gender of the researcher might influence study outcomes, especially those concerning behavioral studies, in both animals and humans. The act of observation may change the outcome (the "observer effect") and any experimental manipulation, no matter how well-controlled, is subject to it. This is nowhere more applicable than in physiology and behavior. The sex of established cultured cell lines is another issue, in addition to aneuploidy; chromosomal numbers can change as cells are passaged. Additionally, culture medium contains steroids, growth hormone, and insulin that might influence expression of various genes. These issues often are not taken into account, determined, or even considered. Issues pertaining to the "sex" of cultured cells are beyond the scope of this Statement. However, we will discuss the factors that influence sex and gender in both basic research (that using animal models) and clinical research (that involving human subjects), as well as in some areas of science where sex differences are routinely studied. Sex differences in baseline physiology and associated mechanisms form the foundation for understanding sex differences in diseases pathology, treatments, and outcomes. The purpose of this Statement is to highlight lessons learned, caveats, and what to consider when evaluating data pertaining to sex differences, using 3 areas of research as examples; it is not intended to serve as a guideline for research design.

Key Words: brain-gut, cardiovascular disease, chromosome complement, gender, sex differences, steroid hormones

Sex is an important biological variable that must be considered in the design and analysis of human and animal research. The terms *sex* and *gender* should not be used interchangeably. Sex is dichotomous, with sex determination in the fertilized zygote stemming from unequal expression of sex chromosomal genes. By contrast, gender includes perception of the individual as male, female, or other, both by the individual and by society; both humans and animals have sex, but only humans have gender. Both sexes produce estrogens, androgens, and progestins; there are no male- or female-specific sex hormones, *per se*, although these steroids are present in substantially different levels in males and females. Sex differences are caused by 3 major factors—sex hormones, genes, and environment. To understand disease mechanisms and exploit sex differences in protection or exacerbation of diseases, one needs to determine the relative contribution of factors, including observer effect (1), causing sex differences. Here—using 3 broad research areas as examples—the roles of sex differences in brain anatomy, brain-gut axis, and cardiovascular disease are discussed. Contemporary brain imaging methods show age- and sex-related differences in brain size, global and regional gray matter volume, white matter connectivity, and neuroanatomic regulation of appetite and satiety; while these differences are seen in large population-based studies, there is tremendous individual overlap, but such group-level findings do not inform findings, physiology, or pathology at the individual level. Sex differences in disorders of the brain-gut axis, obesity, type 2 diabetes, and metabolic syndrome are caused by differential actions of brain-gut peptide and steroid hormones. The activation, signaling, and pharmacotherapy responses of the components of the hypothalamic-pituitary-adrenal (HPA) axis differ between the sexes. Heart and kidney functions are linked. Age, hormones, and sex biases seen in cardiovascular and chronic kidney diseases also differentially influence pharmacologic responses in men and women. Thus, sex differences pervade biology and medicine, and while not discussed in this Statement, must be considered in virtually all areas of biomedical research.

Section I

Sex Versus Gender

Much of the American public is surprisingly prudish about the word *sex*; it has now become commonplace to use the seemingly more genteel term *gender* when one really means *sex*. In *Moritz v Commissioner of Internal Revenue* (469 F. 2d 466 [1972]), Ruth Bader Ginsburg (subsequently, The Honorable Ruth Bader Ginsburg) argued against discrimination "on the basis of sex" not "on the basis of gender," thus clearly, knowledgeably, and presciently understanding that "sex" does not equal "gender." In a decision 48 years later (*Bostock v Clayton County*, 590 US, decided June 15, 2020), the United States Supreme Court separately ruled against discrimination on the basis of gender. *Gender* is often misused as a synonym for *sex*—for example, when filling out forms for various activities, we are routinely

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asked to check a box labeled "gender," but the only available options are boxes labeled "M" and "F." But *sex* is not the same thing as *gender* and using these terms as equivalents obfuscates differences that are real and important in society in general and biomedical research in particular.

Biological Sex: The Definition of Male and Female

Sex is a biological concept. Asexual reproduction (cloning) is routine in microorganisms and some plants, but most vertebrates and all mammals have 2 distinct sexes. Even single-cell organisms have "mating types" to facilitate sexual reproduction. Only cells belonging to different mating types can fuse together to reproduce sexually (2, 3). Sexual reproduction allows for exchange of genetic information and promotes genetic diversity. The classical biological definition of the **2 sexes** is that females have ovaries and make larger female gametes (eggs), whereas males have testes and make smaller male gametes (sperm); the 2 gametes fertilize to form the zygote, which has the potential to become a new individual. The advantage of this simple definition is first that it can be applied universally to any species of sexually reproducing organism. Second, it is a bedrock concept of evolution, because selection of traits may differ in the 2 sexes. Thirdly, the definition can be extended to the ovaries and testes, and in this way the categories—female and male—can be applied also to individuals who have gonads but do not make gametes.

In mammals, numerous sexual traits (gonads, genitalia, etc) that typically differ in males and females are tightly linked to each other because one characteristic leads to sex differences in other traits. The type of gonads is controlled by the presence of XX or XY chromosomes, and gonadal secretions in turn regulate formation of female or male reproductive tissues, and characteristics that differ in typical males or females. These characteristics include external genitalia, uterus and oviducts, sperm ducts, and secondary sexual characteristics such as facial hair and pitch of voice. However, many people cannot make either eggs or sperm, yet are recognized as female or male based on other physical characteristics; people who do not have either ovaries or testes are rare. For individuals that possess a combination of male- and female-typical characteristics, these clusters of traits are sufficient to classify most individuals as either biologically male or female. For example, a person with testes and a penis, who cannot make sperm, is usually classified as a biological male, as long as the person does not possess female features such as a vagina, ovaries, or uterus. Based on evidence presented, to define male and female individuals in general society, we expand the defining characteristics of sex to include nongonadal traits, as well as classical gonadal traits.

A simple biological definition of male and female, satisfactory to all people, is elusive. In human societies, the terms *female* and *male* can have several meanings, as they refer both to a person's biological sex and to their social roles. Most people learn to discriminate males and females from an early age, but often not based on biological traits (4). For example, behaviors such as pair-bonding, sexual activity, offspring defense and care, and mate/partner selection (5) involve complex interplay between sex steroid hormones and peptide hormones (oxytocin and arginine vasopressin); these behaviors are encouraged differently in women and men, which influences their role in the society and culture in which they live to behave as "females" or "males." While these factors have little impact on their biological sex, they can have profoundly different outcomes in the behavior and health of an individual. Biological sex is dichotomous because of the different roles of each sex in reproduction. For scientific research, it is important to define biological sex and distinguish it from other meanings.

Sex Chromosomes and Biological Sex Determination

Among mammals and many other taxa, males are characterized as the heterogametic sex (6), having 2 different sex chromosomes, X and Y, whereas females are homogametic (XX). By contrast birds, many reptiles, and some other organisms have Z and W chromosomes (7). In these organisms, the female is the heterogametic sex (ZW) and males are homogametic (ZZ). Some adult fish and reptiles can also change sex in response to environmental factors (8, 9), and even the adult mouse gonad can undergo partial sex reversal when specific genes are deleted (10, 11). Human biological sex is often assessed by examining the individual's complement of sex chromosomes as determined by karyotypic analysis: males are XY and females are XX. Karyotypic sex is actually a surrogate for genetic sex, determined by the presence of the *SRY* gene on the Y chromosome (12, 13). However, karyotypic analysis may be misleading, as there are well-described 46,XX males (with testes). Most of these individuals carry a short segment of the Y chromosome that includes *SRY* transferred to an X chromosome, but up to 10% lack an *SRY* gene (14, 15). Similarly, there are 46,XY females, who have *SRY* but also have a duplication of *DAX1* (*d*osage-sensitive sex reversal, *a*drenal hypoplasia critical region, on chromosome *X*, gene 1) (16).

Sex Determination and Sex Differentiation

In mammals, sex determination begins with the inheritance of XX or XY chromosomes, which are the only factors that are different in XX and XY zygotes. Thus, all phenotypic sex differences, including gonadal development, stem originally from the unequal effects of XX and XY

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sex chromosomes. Phenotypic sex differences develop in XX and XY embryos as soon as transcription begins. The categories of X and Y genes that are unequally represented or expressed in male and female mammalian zygotes, which could cause phenotypic sex differences, fall into 3 main categories (17).

1. *Y genes causing male-specific effects*. These Y-linked genes do not have homologous genes on the X chromosome. The most important Y-linked gene is *SRY*, the testis-determining gene, which encodes the SRY transcription factor expressed during embryonic life in the bipotential gonadal ridge; SRY activates downstream autosomal genes such as *SOX9* to cause formation of a testis (18). In the absence of *SRY,* autosomal and X chromosome genes (*WNT-4*, *DAX-1*, *FOXL2, COUP-TFII*, and *RSPO1)* are activated to cause formation of an ovary (19-22). Both testicular and ovarian development are subject to active genetic regulation (12, 13, 16). Pathways downstream of *SRY* inhibit ovary-determining pathways, and ovary-determining pathways also inhibit pathways for testis development. Once the testes form, they secrete sex hormones that act widely throughout the body to cause male differentiation of nongonadal tissues. Other Y genes also have male-specific effects (for example, those required for spermatogenesis) (23, 24).
2. *X gene dosage or parental imprint*. Because XX nongermline cells inactivate one X chromosome (25, 26), it was long thought that both XX and XY cells have only one active X chromosome, with little inherent difference in expression related to the number of X chromosomes. The inactivated regions of the X chromosome are "coated" with large noncoding RNA transcribed from the X-inactive specific transcript (*XIST*) gene, part of the XIC (X inactivation center) located on Xq13 (27, 28). But some genes escape X inactivation (termed as *X escapees*), and therefore are expressed more in XX than XY cells, resulting in imbalance or incomplete dosage compensation (29). About 23% of human X-linked genes are more abundantly expressed in XX cells than XY cells in many tissues (30, 31). Recent evidence from mouse studies suggests that the inherent male-female difference in expression of X genes leads to significant sex differences in disease phenotypes. For example, sex differences in placental *Ogt* expression are associated with sex differences in prenatal vulnerability to stress (32). X escapee *Kdm6a*, a histone demethylase, contributes to sex differences in mouse models of bladder cancer (33), autoimmune disease (34), and Alzheimer disease (35). Similarly, variations in human *KDM6A* are associated with prognosis of bladder cancer or cognitive decline in female patients (33). The dose of another X escapee histone demethylase, *Kdm5c*, contributes to sex differences in adiposity and body weight in mice, and variations in *KDM5C* in humans are associated with body mass (36).

   Sex differences may also arise from genes in the pseudoautosomal regions (PARs) of the sex chromosomes, small regions of sequence similarity on the X and Y chromosomes that allow for X and Y chromosome pairing during meiosis. Both XX and XY cells have 2 PARs, implying equivalent effects of XX and XY PARs. Paradoxically, the process of X inactivation appears to spill over into the PAR and reduce expression on one X chromosome only in XX cells, leading to greater expression of PAR genes in XY cells compared to XX cells in the human transcriptome (30). A third potential source of X-linked imbalance stems from parentally imprinted genes in XX cells, which have one X chromosome from each parent and thus are influenced by any imprint on X genes from either parent. XY cells only receive imprints from the mother, and thus differ phenotypically from XX cells (37).
3. *XX mosaicism*. Female mammals are a mosaic of cells of 2 types: those expressing the X chromosome from the father (Xp), or from the mother (Xm) because of X inactivation (25). In contrast, XY individuals will lack this diversity within cell types in each organ because only one X (Xm) chromosome and only the maternal imprint of X genes will be expressed in each cell. The mosaicism in females means that in genetically diverse populations, the effects of disease-promoting X-linked alleles, inherited from one parent, will be muted in XX cells because half of the cells will have a different allele (38), and genomic imprints from each parent will only be expressed in half of the cells. In general, XX tissues are thought to have less extreme phenotypes than XY tissues, because the effects of extremely deleterious or beneficial alleles or imprints are buffered by the diversity of X alleles and imprints. For example, hemophilia A and hemophilia B (clotting factor VIII and IX deficiencies, respectively), are X-linked diseases that affect men, whereas most women are asymptomatic carriers.

Sexual Differentiation Caused by Gonadal and Nongonadal Hormones

In mammals, the process of reproductive system development requires the action of hormones (peptide/gonadotropins and steroids) from the pituitary gland, the adrenal cortex, and the gonads. Testicular development leads to secretion of Müllerian inhibitory hormone (MIH, also termed anti-Müllerian hormone, AMH), a glycopeptide, and testosterone, which affects many sex differences in nongonadal tissues (39). In contrast to the fetal testis, the fetal ovary makes minimal steroid hormones

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(40), and ovarian function is not needed for development of the female reproductive system, as evidenced by the normal female anatomy of individuals with Turner syndrome, who have 45,X gonadal dysgenesis. The pioneering work of Alfred Jost suggested that 2 classes of testicular hormones are involved in sexual differentiation. First, testicular androgens drive the differentiation of the fetal external genitalia from female morphology to that of the male and are required for the differentiation of embryonic Wolffian ducts into male internal reproductive structures (41, 42). Androgens, secreted by Leydig cells, are required for the differentiation of embryonic Wolffian ducts into male internal reproductive structures (epididymis, vas deferens, ejaculatory ducts, prostate, and seminal vesicles), and drive the differentiation of the undifferentiated external genitalia toward male morphology. Second, the testis produces locally acting MIH that causes involution of the Müllerian ducts, which would otherwise develop into the fallopian tubes, uterus, and cervix (43, 44).

It was long thought that only the involution of the Müllerian ducts was an active process, with the Wolffian ducts simply involuting in the absence of androgens. Recent evidence from mice indicates that Wolffian involution is also an active process controlled by the transcription factor COUP-TFII (22, 45), but the nature of any factors stimulating COUP-TFII remains unknown (22). Some aspects of gonadal differentiation are active throughout life, preventing ovarian follicle cells from transdifferentiating into "testis-like" cells (11). MIH is secreted by Sertoli cells and androgenic steroid hormones, usually testosterone, are secreted by Leydig cells. Testosterone and its more potent derivative dihydrotestosterone are responsible for the development of the male external genitalia (46). Androgens from adrenal glands and alternative pathway androgen biosynthesis in the human placenta can influence virilization of the developing fetus (47, 48). The adrenals of adult primates also produce abundant androgens, profoundly influencing phenotypes, so that not all sex steroids are gonadal (see Boxes 1 and 2). Although the term *sexual differentiation* is usually applied to the development of sex differences in genitalia and other organs such as the brain in the growing fetus; sex differences also occur later in life during the mini-puberty of infancy (49), puberty, the female menstrual cycle, menopause in women, and andropause in men. The actions of gonadal and nongonadal hormones as well as sex and autosomal chromosome gene products in adult people causes many sex differences in health and disease.

Influence of Gonadal Steroid Hormones and Nongonadal Hormones in Brain Development

Differentiation of the brain by gonadal hormones is implemented during a restricted critical window, which is operationally defined by the onset of copious androgen

Box 1. Steroidogenesis in gonadal and nongonadal tissues

All biologically active sex steroids, whether gonadal or nongonadal in origin, are derived from cholesterol by the process of steroidogenesis. Two steroidogenic steps must be considered (for details see (50)). First, the cholesterol side-chain cleavage enzyme, P450scc (CYP11A1) initiates steroidogenesis by converting cholesterol to pregnenolone; expression of P450scc renders a tissue "steroidogenic," that is, able to make steroids de novo (51). The gonads, adrenals, and placenta express abundant P450scc and produce the familiar circulating endocrine steroids, but the brain, skin, and some other organs also express low levels of P450scc and produce steroids involved in paracrine actions. Brain steroidogenesis has been studied mainly in fetal rodents, with little information in other systems (52). Many nonsteroidogenic tissues (liver, kidney, fat, breast, heart) do not express P450scc but express other steroidogenic enzymes that modify steroids taken up from the circulation. Fat and breast express CYP19A1 (aromatase), permitting local production of estradiol from circulating 19-carbon (C19) steroids; this estradiol is important in breast cancer but is not a gonadal steroid. Similarly, prostate and genital skin express several enzymes leading to dihydrotestosterone, accounting for the failure of "androgen deprivation therapy" by gonadectomy in prostate cancer. Not all gonadal steroids are sex steroids, as both the ovary and testis secrete some "upstream" steroids that are precursors of the classic sex steroids. For example, dehydroepiandrosterone (DHEA) does not bind to sex steroid receptors, but it can be converted into testosterone and estrone. Second, synthesis of all sex steroids requires P450c17 (CYP17A1), which catalyzes 17α-hydroxylation and the 17,20 lyase activity that changes 21-carbon steroids to C19 precursors of androgens and estrogens. P450c17 is abundantly expressed in the gonads of all vertebrates and in the adrenals of most vertebrates other than rodents, but the rodent *Cyp17A1* gene is silenced by tissue-specific methylation (53). Consequently, rodents make only miniscule amounts of adrenal C19 steroids and also use corticosterone instead of cortisol as their glucocorticoid. In most mammals, P450c17 has low 17,20 lyase activity, so that their adrenals produce rather small amounts of C19 steroids, but primate P450c17 has abundant 17,20 lyase activity, generating abundant C19 androgen precursors (DHEA, DHEA-sulfate, androstenedione) (47, 48). Furthermore, production of these C19 steroids proceeds by different pathways in rodents and primates: primates favor the "Δ5 pathway," through DHEA, whereas rodents favor the "Δ4 pathway" through 17OH-progesterone (17OHP) (50). Primate adrenals also produce a true androgen, 11-keto-testosterone (54), profoundly influencing phenotypes (apocrine odor; female sexual hair). Thus, not all sex steroids are gonadal: ~ 50% of the circulating androgens in adult women are of adrenal origin.

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Box 2. Gonadectomy and sex steroids

Many animal studies employ gonadectomy to eliminate the actions of sex steroids (estrogens, androgens, progestins). If using this approach, the investigator must consider whether nongonadal tissues will produce sufficient sex steroids to influence the study. The gonads produce most but not all circulating sex steroids; furthermore, some tissues produce steroids that act locally and do not enter the circulation, hence absence of a measurable steroids in blood does not ensure absence of its action in the target tissue. Both sexes produce all steroids and their metabolites, hence there are no male- or female-specific sex hormones, *per se*. In male mammals, testosterone release is highly pulsatile in nature (49, 55) and in laboratory mice, strain-dependent variations in androgen levels are reported (56). In female rodents, circulating levels of estradiol, testosterone, and DHT are highest in proestrus phase; a comprehensive analyses of sex steroids in intact and gonadectomized rodents can be found elsewhere (57). Circulating concentrations of testosterone in adult women are similar to those of boys in early puberty, and estradiol concentrations in men are similar to those in mid-cycle women, but the tenfold higher concentrations of testosterone obscure its effects. Rodents are widely used in research, but they differ from primates in several important aspects of steroidogenesis (see Box 1), and hence must be used with caution in studies seeking to model aspects of human physiology that might be influenced by steroids. These differences include: (i) In humans, substantial amounts of circulating sex steroids are bound to sex hormone–binding globulin (SHBG), whereas this carrier protein is not present in rodent circulation (58). (ii) Dehydroepiandrosterone (DHEA) and androstenedione, 19-carbon (C19) precursors for testosterone and estrone, that do not bind to sex steroid receptors, are secreted from the adrenal glands, the ovary and testis in humans, but not rodents (59). Thus, not all gonadal steroids are sex steroids. (iii) The rodent ovarian corpus luteum produces progesterone throughout pregnancy but in human pregnancy the corpus luteum involutes early in the second trimester, after which the placenta produces the progesterone needed to suppress uterine contractility, permitting term pregnancy. (iv) Adrenal-specific methylation of rodent *Cyp17A1* prohibits their adrenal synthesis of C19 precursors of sex steroids; however, changes in methylation status can occur under conditions of pathology. (v) As a further consequence of adrenal *Cyp17A1* methylation, rodents utilize corticosterone as their glucocorticoid, whereas almost all other vertebrates use cortisol. (vi) Rodent adrenals use high-density lipoproteins (HDL) taken up via scavenger receptor B1 (SRB1), as their principal source of cholesterol for steroidogenesis, whereas primates use low-density lipoproteins (LDL) taken up by receptor-mediated endocytosis. (vii) Several genes encoding steroidogenic enzymes are duplicated; rodents and primates differ in which copy(ies) of these genes are expressed: *CYP21*; *HSD3B*, *HSD17B, AKR1-3*. Such differences may affect laboratory results in unanticipated fashions. (viii) In rodents, nonsteroidogenic tissues such as the gut, liver, kidney, fat, breast, heart, thymus, skin, and the placenta have all been shown to make steroids. Thus, gonadectomy may eliminate most, but not all, circulating sex steroids, depending on the species being studied and may not reveal much about the paracrine effects of sex steroids present in the tissue(s) under investigation. Nonetheless, gonadectomy is an invaluable research tool that helps unequivocally confirm the influence of gonadal hormones in sex differences.

production from the fetal testis. Human fetal androgen production begins at 8 to 10 weeks postconception and in rodents is closer to parturition, at embryonic days 16 to 18, with birth following 2 to 4 days later. An important effect of this androgen surge is to masculinize the rodent brain. Steady but pulsatile release of the gonadotropins luteinizing hormone and follicle stimulating hormone from the pituitary gland support continuous steroidogenesis and production of sperm (60). In female rodents, the feminization of the brain proceeds in the absence of exposure to high levels of androgens or their aromatized byproducts, estrogens, a developmental strategy highly analogous to that used for masculinization of the gonads, reproductive tract, and secondary sexual characteristics, with the exception that estrogens are actively downregulated in male rodents. In human females, gonadotropins from the pituitary gland regulate ova development, induction of ovulation, and stimulation of estradiol and progesterone from the ovaries (49). An important feature of this developmental strategy is the existence of a sensitive period in female rodents (61). Male rodents must be exposed to high levels of androgens during the critical period; if exposure occurs too early or too late it will be ineffective at inducing masculinization. However, females are also sensitive to androgens during a restricted period of development, hence a sensitive period in rodents. In males, the critical period closes shortly after androgen exposure because the cellular and molecular processes of masculinization have been initiated and cannot be reversed; the train has left the station. In both primates and rodents this process is largely prenatal, but female rodents remain sensitive to androgen exposure into the first postnatal week. Injecting a newborn female rodent with androgens will initiate the process of masculinization, thus she is still sensitive. After the first week, the feminization process cannot be overridden by androgens and thus the sensitive period has closed. The existence of the sensitive period in females is useful as a research tool—it is important in understanding the potential impact of exposure to endocrine-disrupting compounds or other cellular agents of masculinization that act in an analogous manner to androgen exposure in modulating female brain development. There is evidence for a later sensitive

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period for brain feminization mediated by small increases in estrogens (62); this topic warrants further investigation. The closing of the sensitive period in primates, especially humans, remains poorly understood, but it appears to end prenatally, similar to the critical period in rodents. The sources of androgens that females can be exposed to during the sensitive period include from: (i) experimental interventions; (ii) male littermates in animals; (iii) or human adrenals carrying genetic mutations in the steroidogenic pathway (as in congenital adrenal hyperplasia).

Given that the critical and sensitive periods for sexual differentiation are defined by the production and response to gonadal steroids, it is not surprising that steroids are the primary drivers of developmental origins of sex differences in brain (and probably other tissues) and behavior. But how do steroids achieve this? The first step in any investigation is often is to identify the active steroid metabolite(s). In rodents, circulating fetal testicular testosterone enters the fetal brain where it can serve as a direct precursor for estradiol synthesis via aromatase (*Cyp19A1*) (see Box 1). Fetal and adult neurons can aromatize testosterone to estradiol in a nonrandom distribution: neurons of the hypothalamus, preoptic area, and amygdala are particularly active for local estradiol synthesis, whereas the hippocampus and parts of the cortex, midbrain, and spinal cord are also active at a lower level (63). For most reproductive endpoints, it is the local actions of estradiol that drive neural phenotype toward masculinization, which to some seems counterintuitive, given that estradiol is so often referred to as a "female" hormone (64), and further highlights that it is impossible to completely eliminate the effects of sex steroids, especially in the brain, by simple gonadectomy (see Box 2). Developing rodent embryos sequester maternal estrogens by binding to circulating alpha-fetoprotein, which is present only during the critical/sensitive period; when it is genetically deleted, all the offspring are masculinized (65). However, in humans, sex hormone–binding protein, not alpha-fetoprotein, is the major serum glycoprotein that binds androgens and estrogens with an undetermined role in fetal sexual development (66, 67).

In rodents, there is abundant evidence that gonadal androgens are metabolized to estrogens in the brain and mediate "masculinizing" effects on the brain; similar evidence in primates is limited. In primates, the principal masculinizing agents are androgens, not estrogens, and although there is alpha-fetoprotein present in fetal circulation, it has a weak binding affinity for estradiol (68), and instead it plays a much broader role in brain and body development (69). The conclusion of no strong role for estrogens in humans is based on individuals with dysfunctional aromatase or androgen receptors. Males lacking aromatase still identify as men, while XY individuals with complete androgen insensitivity identify as women (70). The disparity between the principal differentiating hormones in primates versus rodents suggests that findings may not be easily extrapolated, and it is important to specify both the hormone and species under investigation. To discern whether the biological basis of sexual differentiation of brain and behavior differs between primates and rodents, one needs to identify mechanisms by which steroids transduce signals to modify the trajectory of the nervous system. While those mechanisms are incompletely understood, a few general principles are clear. First, there is no unified mechanism that applies broadly across the brain, with the exception that androgens and estrogens are the primary drivers of masculinization during a restricted developmental window. Similar masculinizing effects of testicular androgens may also occur during puberty (71). Second, all aspects of neural development are capable of being "organized" or programmed by sex steroids. This includes cell genesis, migration, myelination, dendritic and axonal growth and branching, synapse formation, synapse elimination, and neurochemical differentiation. Effects are not limited to neurons, with both astrocytes and microglia also exhibiting morphological sex differences. Third, each discrete brain region, nucleus, or subnucleus appears to have unique mechanisms of cellular masculinization. In some brain regions, such as the preoptic area, there are multiple separate mechanisms at play simultaneously. Sex steroids act in both paracrine and endocrine manners to influence structural development and function (72, 73).

Biological Basis of Diversity in Sexual/Gender Development and Orientation

Given the complexities of the biology of sexual determination and differentiation, it is not surprising that there are dozens of examples of variations or errors in these pathways associated with genetic mutations that are now well known to endocrinologists and geneticists (74); in medicine, these situations are generally termed *disorders of sexual development* (DSD) or *differences in sexual development* (75). DSD includes genetic disorders in the sexual determination pathway (76), disorders of steroidogenesis (50, 77), disorders of steroid hormone action, especially androgen insensitivity syndrome (78), and less well-defined "developmental field defects" (79), such as Mayer–Rokitansky–Küster–Hauser syndrome (80). The study of genes and factors underlying DSD and the diagnosis and management of the various forms of DSD is a complex and rapidly evolving area of endocrinology: clinical management is complex (81) and requires both contemporary molecular genetics (82) and well-integrated interdisciplinary care (83).

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Gender includes perception of the individual as male, female, or other, both by the individual and by society. *Gender identity* is a psychological concept that refers to an individual's self-perception; while associations between gender identity, neuroanatomic, genetic, and hormone levels exist, a clear causative biological underpinning of gender identity remains to be demonstrated. Both animals and human beings have biological sex, but only humans have evident self-awareness that allows them to express gender; self-awareness in animals has not been investigated in this context. Gender also includes differences that males and females experience in their social and physical environments, which can have differentiating effects on the sexes. Human social environments are poorly modeled in laboratory animals and thus animal studies are usually limited to addressing sex differences. For centuries, the concept of male and female did not distinguish between biological sex differences and those caused by consistent differences in the environments. Thus *sex differences* are those caused by biological factors, whereas *gender differences* reflect a complex interplay of psychological, environmental, cultural, and biological factors (Fig. 1).

At birth, individuals are assigned a sex or gender ("natal gender"), almost always based on the appearance of the external genitalia. In most individuals, the various biological determinants of sex are consistent with one another, and this biological sex is also consistent with the individual's self-perception—the sex and gender are concordant. However, a substantial minority of people who do not have DSD have some degree of variation in their self-perception of their gender, which may differ from their biological sex; this is usually termed *gender incongruence* (84). The term *gender disorder* has been replaced with the term *gender dysphoria* which describes the distress that an individual might feel as a consequence of having gender incongruence. *Transgender* (often called *trans*) refers to individuals who do not identify themselves as being of their natal gender, whereas *cisgender* (*cis*) people do not experience gender incongruence (85). Readers are also referred to Endocrine Society's 2017 Clinical Practice Guideline and Transgender Health Fact Sheet (84). Estimates of the prevalence of male-to-female transgender individuals among general populations range from 0.5% to 1.3% and estimates for female-to-male transgender individuals range from 0.4% to 1.2% (85). State level population-based surveys indicate that 0.6 % of US adults (25-64 years of age) and 0.7% of adolescents and young adults (13-24 years of age) identify as transgender. Other studies of US high school

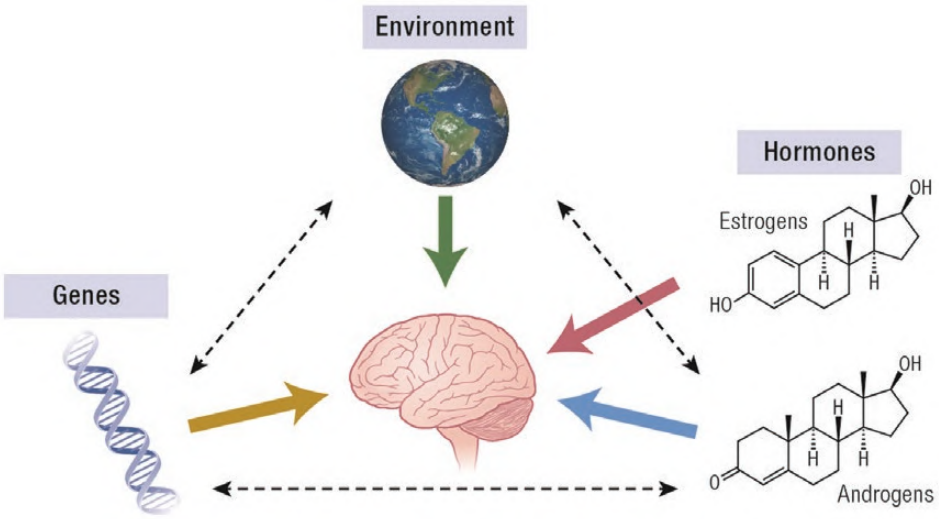

Figure 1. Simplified view of the factors influencing sex differences in the brain. Three broad groups of factors influence the sexually dimorphic brain, as indicated by the broad, colored arrows. 1) Genes and genetic factors that influence the brain include both those on sex chromosomes and autosomes, and include both the DNA itself (represented by the classic double helix) but also chemical modification of DNA (eg, methylation) and modifications of proteins associated with DNA to form chromatin, including histones, and also changes in proteins that bind to DNA. 2) Hormones clearly influence sexual dimorphism in the brain; these are represented by the principal sex steroids, estradiol and testosterone, but also include other steroid and protein hormones (progestins, MIH, oxytocin, prolactin, etc). 3) The environment includes a wide spectrum of influences, including perinatal nutrition and familial support, socioeconomic and demographic factors, intrinsic factors of brain development, age, and gender, and larger environmental factors, such as education, profession, and societal expectations (the "gendered environment"). In addition to each class of factor influencing the brain (bold arrows), the human brain also reciprocally influences each of these groups of factors. Furthermore, each group of factors influences the other, as represented by the dotted arrows. Some examples include: the environment influences genes via epigenomics and genes influence the environment by population sizes and domains; the environment influences hormones by seasonal variations and the actions of xenobiotics, and hormones influence the environment by promoting reproduction and consumption of foodstuffs; genes directly influence hormones by regulating their production and action, and many hormones, including all steroid hormones, regulate gene transcription.

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students suggest a prevalence of 1.8% to 2.7% of being gender nonconforming or transgender (86-88). However, several factors may influence reported prevalence of gender dysphoria: (i) small sample sizes; (ii) differences in assessment techniques leading to incomplete ascertainment of gender dysphoric individuals; (iii) unwillingness of some individuals to respond fully and honestly, especially in older studies or studies deriving from locales where gender incongruence is a social taboo; (iv) differences in the subjects ages. *Sexual orientation,* not to be confused with gender identity, refers to the group of persons to whom an individual is sexually attracted; both cisgender and transgender individuals may be hetero-, homo-, or bi-sexual (89).

Although gender is strongly influenced by environmental and cultural forces, it is unknown if the choice to function in society in male, female, or other role(s) is also affected by biological factors (89-91). A general issue is that the association of sex, gender, or sexual orientation with specific brain structures, or with other biological variables, does not establish whether the biological variables are causes or consequences or noncausal correlates of the behavioral characteristics or function of the individuals studied. Three areas of biological difference have been studied fairly extensively: neuroanatomy, genetics, and hormones. Studies have reported differences in the hypothalamic INAH3 nucleus in men vs women and in homosexual vs heterosexual men (92, 93). Although initially controversial, others have confirmed sex differences in INAH3 numbers, not in size or densities, whereas no evidence for sexual dimorphism of any other INAH structures are reported (94). Studies in people with gender dysphoria found that the phenotypes of specific brain structures, such as the bed nucleus of the stria terminalis, of transgender women and transgender men differ from cisgender men and women, with partial, but incomplete sex reversal of sexually dimorphic structures (95). Brain networks involved in one's body perception, (pregenual anterior cingulate cortex, temporo-parietal junction, and fusiform body area) differ in individuals with gender dysphoria compared with cisgender individuals (96-98). Neuroimaging shows that testosterone treatment resulted in functional and structural changes in brain areas associated with self-referential and own body perception (99). Transgender men have thicker medial prefrontal cortex than cis men. Testosterone treatment does not change prefrontal cortex thickness in transgender men, but it has other effects on cortical thickness, connectivity, and fractional anisotropy (99).

Genetics may play a role in gender identity (100): monozygotic twins have 39% concordance for gender dysphoria (101). Attempts to identify specific genes governing gender identity have been plagued by small numbers of subjects and low statistical significance; no specific gene has been reproducibly identified. However, such studies have suggested associations with genes encoding steroidogenic enzymes and sex steroid receptors, and it is generally agreed that androgens play an important but not determinative role. For example, many 46,XX individuals with severe virilizing congenital adrenal hyperplasia (steroid 21-hydroxylase deficiency) are exposed to intrauterine testosterone concentrations typical of those in normal male fetuses and consequently have severely virilized external genitalia; nevertheless, most have a female gender identity, but about 5% to 10% of such individuals have gender dysphoria, an atypical gender identity (89, 102, 103), or atypical sexual orientation and gender behavior (104, 105). Similarly, about half of 46,XY individuals with defects in androgen synthesis who were raised as females revert to a male gender role (106). The biological underpinnings of sexual orientation and gender identity are apparently related but are not the same (107). Thus, there is ample but incomplete evidence for biological substrates—neuroanatomic, genetic, and hormonal—for gender orientation, making this an important area of ongoing research.

Hormonal Versus Sex Chromosome Effects

Sex differences are caused by 3 major factors—sex hormones, genes on sex chromosomes/autosomes, and environment (Fig. 1). To understand disease mechanisms in both sexes and exploit sex differences in protection or exacerbation of diseases, it is important to determine the relative contribution of each of these factors in causing sex differences (17). Many sex differences caused by gonadal hormones have been discovered by measurements of sex steroids and gonadotropins during human development, and in animals by similar measurements or by interventional methods, such as gonadectomy, hormone administration, or the expression of synthetic enzymes or receptors in transgenic mice. Sex steroids play an integral part in many physiological processes (Box 1). Whereas the gonads are the major site of sex steroid synthesis, the adrenals, placenta, brain, and skin can also initiate steroidogenesis, and steroid-modifying enzymes are found elsewhere, especially in liver and fat, permitting synthesis of sex steroid hormones in multiple other sites (50). Thus, animal gonadectomy may provide information about endocrine effects of gonadal steroid hormones but cannot address tissue-specific paracrine effects (Box 2). Moreover, gonadectomy cannot mimic low pre-pubertal levels or physiological conditions in which hormone levels decrease, such as aging or menopause. Manipulations of human gonadal hormones are routinely used in contraception and in the management of sex steroid–dependent cancers (eg, breast, prostate). When

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a sex difference is discovered in human disease, and modeled in animals, the investigation of possible hormonal causation of the sex difference is usually the first option considered.

To detect effects of sex chromosomes that cause sex differences, one can compare people who have differences in their sex chromosomes, revealing effects of X or Y chromosome number (108-110). These results strongly suggest direct sex chromosomal contributions to sex differences in cell function. Comparison of brains of XY patients with complete androgen insensitivity (who are phenotypically female), with brains of control XY males and XX females, suggests that cortical thickness and functional connectivity between the limbic regions and the cortex are influenced not only by testosterone actions, but by sex chromosome factors as well (111). However, changes in the sex chromosome ploidy also alter gonadal hormones, so it can be difficult to isolate sex chromosome effects not mediated by gonadal hormone effects. Circulating human embryonic/fetal sex steroid concentrations are poorly characterized, and the tissue concentrations are almost totally unknown. Another approach is to use mice to identify genes on the X or Y chromosome that act outside of the gonads to cause sex differences, and then seek evidence that the orthologous human genes cause human sex differences. Controlled experiments are possible in which XX or XY mice with comparable gonadal hormones can be compared. A frequently used model is the Four Core Genotypes (FCG) model, in which the testis-determining mouse *Sry* gene is deleted from the Y chromosome (creating the Y^- or "Y minus" chromosome) and inserted as a transgene on chromosome 3 (*Sry+)* (Fig. 2 and Box 3) (112). The utility and limitations of these models have been extensively discussed (113, 114).

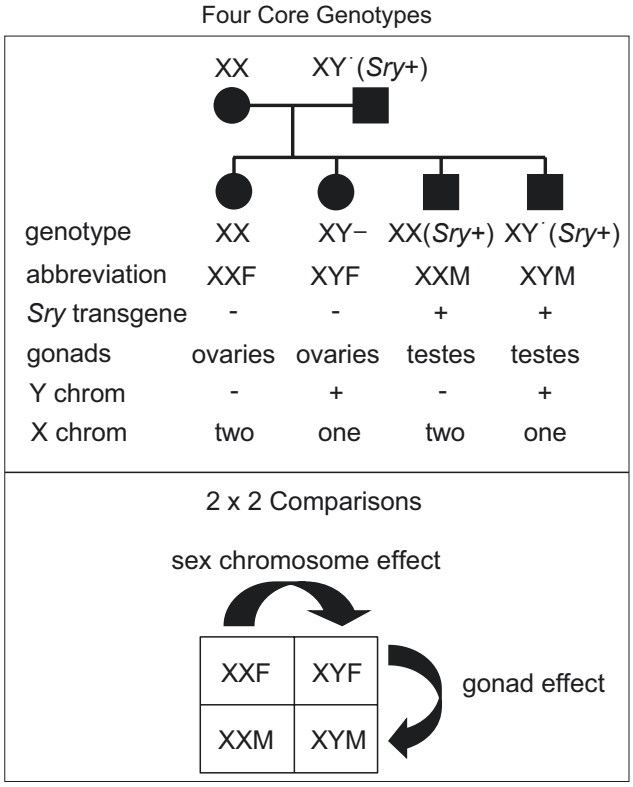

Figure 2. Schematic diagram of the Four Core Genotypes mouse model. The testis-determining gene *Sry* is deleted from the Y chromosome, producing the Y^- chromosome. An *Sry* transgene is inserted onto chromosome 3. Thus, the type of gonad is no longer linked to the sex chromosomes. The model produces XX and XY mice with *Sry* and testes, and XX and XY mice without *Sry*, with ovaries. Sex differences in phenotype can be attributed to an effect of gonadal hormones, comparing mice with ovaries and testes, or to an effect of sex chromosomes, comparing XX and XY mice with the same type of gonad. [Modified with permission from Arnold AP & Chen X. *Front Neuroendocrinol,* 2009; 30(1) © Elsevier Inc. (112)].

Considering Sex and/or Gender as Variables in Health and Disease

Women and men differ in many physiological and psychological variables. It is important to establish the mechanisms causing such differences in health and disease, and to consider sex-related variables in studies of human health and disease. These variables include, but are not limited to, sex- and gender-related factors. The inability to control all variables in human studies means that it may be impossible to determine the relative roles of environment and biology in causing a difference between women and men, when both types of variable can influence the trait. Furthermore, while "gender expression/behavior" can be observed, "gender identity" can only be known by what an individual states. Thus, gender identity, *per se*, cannot be studied in animals. In human studies, it is unethical to selectively manipulate specific biological and environmental variables, and most currently available data derive from studies comparing groups of men with groups of women. It is therefore difficult to disentangle the specific contribution of sex-related genes, hormones, gender-related variables, and other variables that contribute to being female or male. Because sex has long been defined by gonadal type, the list of sex-influencing factors has been primarily associated with gonadal hormones, especially estrogens, progestins, and androgens (121). However, some phenotypic sex differences develop before the gonads differentiate as testes or ovaries (122), so other factors also contribute to sex differences (123) but are seldom considered.

Sex is an essential part of vertebrate biology, but gender is a human phenomenon; sex often influences gender, but gender cannot influence sex. Studies of animal physiology must consider sex as a variable (124), with sex steroids (of both gonadal and nongonadal origins), sex chromosomes, and other factors contributing to sex differences in many physiologic processes. Similarly, studies of human physiology and disease must also consider sex for the same reason (125) and its disorders must

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Box 3. Investigating sex chromosome complement versus gonadal hormones in health and disease: the four core genotypes (FCG) model

The FCG model allows for discriminating hormonal vs sex chromosome effects in animals. Gonadal males (XY^-(*Sry+*)), bred to XX gonadal females, produce 4 types of offspring: XY^- and XX mice with the *Sry* transgene and testes, and XY^- and XX gonadal females lacking the *Sry* gene (Fig. 2). Thus, it is possible to compare XX and XY mice with the same type of gonad, in 2 separate comparisons. Differences between XX and XY are attributed to effects of sex chromosome genes acting on nongonadal tissues. To determine if this sex chromosome effect is caused by X or Y genes, a second model is studied, the XY* model (113, 114). This model produces genotypes that are similar to XO, XX, XY, and XXY. An effect of number of X chromosomes is discovered by comparing XO and XX, or XY and XXY. An effect of the Y chromosome genes is discovered by comparing XO and XY, or XX and XXY. These mouse models have been used to demonstrate sex chromosome effects causing sex differences in a wide variety of phenotypes and disease models, including brain and behavioral phenotypes, metabolism, autoimmune, cardiovascular and pulmonary diseases, Alzheimer disease, aging, and cancer (35, 113, 115). These models have facilitated discovery of several disease phenotypes in which the number of X chromosomes contributes to sex differences (116), and a smaller number of sex-biasing effects of Y genes (117). Sex chromosome effects occur in the same disease systems alongside sex-biasing effects of gonadal hormones, such that the 2 effects can synergize to increase the amount of sex difference, or counterbalance each other to reduce a sex difference. Moreover, genes encoded on the Y chromosome can have gene-specific effects, and/or effects that overlap with those of X genes (118). In the cardiovascular system and associated physiological/disease states, sex chromosomes and gonadal hormones can have opposing effects. Estrogens generally protect from cardiac ischemia/reperfusion injury and other cardiovascular diseases, reducing disease in female relative to male mice. However, studies of ischemia/reperfusion injury in gonadectomized FCG mice reveal that the XX sex chromosome complement is associated with worse outcomes, relative to XY (119). In another study, sex chromosome effects in angiotensin II-induced hypertension showed that arterial pressure was greater in gonadectomized XX mice than in gonadectomized XY mice (120). Sex chromosome complement also influences the development of abdominal aortic aneurysms, fat metabolism and adiposity, plasma lipids and lipoprotein levels (particularly HDL-C) (115)).

also consider gender. However, human gender is a spectrum from feminine to gender-neutral to masculine, and also likely includes individuals who do not fit readily on a simple linear continuum (84). Studies addressing the endocrine care of transgender youth during the time of their potential gender transition (84, 89) find that they have a higher prevalence of stress-associated mental health disorders such as depression and anxiety, which can be ameliorated by gender-affirming endocrine treatment (126). It is essential to recognize these sex and gender differences as our health care systems endeavor to develop "individualized medicine."

Despite the fact that biological sex is such a fundamental source of intraspecific variation in anatomy and physiology, much basic and clinical science has tended to focus studies on one sex (typically male). Few studies have done side-by-side testing for sex differences at baseline and in experimental models of human diseases (127-129). Studies in laboratory animals that manipulate biological (eg, genes and hormones) and environmental variables (eg, housing conditions, diet, physical activity, etc) demonstrate that many variables can affect sex-related aspects of an animal's physiology. However, laboratory rodents may show male-female differences caused by different housing conditions, which could be misinterpreted as being caused directly by biological differences without environmental mediation. In studies concerning animal behavior, the sex and gender of the researcher conducting behavioral measures may also influence outcomes (130). Thus, for reproducibility and proper interpretation of the data, at the minimum, it is important to state the precise housing conditions, anesthetics, analgesics (different effects in sexes), doses, surgical manipulations, diet, sex, strain, species, and age of animals used, as well as sex/gender of the researcher(s) performing experiments.

Having laid the foundation for several factors that contribute to sex versus gender, this Statement will use 3 areas of research as examples (not as a literature review) where human and animal sex differences are well known. First, sex differences in specific brain regions of healthy men and women are increasingly being documented along with differences in brain connectomes; these will be discussed in detail in Section II. Second, stress-related pathophysiologies are known to affect twice as many women as men. However, few studies systematically include study designs to ascertain function or mechanisms that may be similar or different between males and females. Hormones and signaling pathways that contribute to sex-specific differences in stress-based pathophysiologies will be discussed in Section III. Similarly, sex differences in manifestation of cardiovascular and renal diseases are well recognized and will be discussed in Section IV.

Section II

Developmental Origins of Sex Differences in Brain Anatomy, Function, and Behavior

Sex differences in the human brain are a topic of intense popular and scientific interest. Several scientific observations motivate the search for sex differences in brain structure

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and function. First, the act of sexual reproduction requires that the male and female animals show qualitatively different reproductive behaviors. The stereotyped emergence of these reproductively critical and sexually differentiated behavior reflects biologically programmed (or "innate") sex differences in the organization of those brain circuits that support the motivational and consummatory phases of copulatory behavior (131). Second, the fact that males and females make different biological investments in reproduction—eg, the risks of pregnancy in mammals are borne entirely by the female—sets up sex differences in the behavioral strategies that optimize reproductive fitness (132). Sexual selection based on sex-biased behavioral strategies is predicted to drive the evolution of sex differences in those brain circuits that are responsible for sexually selected behaviors. Third, males and females can show consistent sex biases in broader behavioral domains beyond those that directly relate to reproductive strategies. In our own species for example, there are highly consistent sex differences in the prevalence of physical aggression and violence (both male-biased) (133), as well as extensively documented sex differences in risk for different mental disorders (134).

In this section, we will first describe the main neuroimaging techniques commonly used in comparisons of brain anatomy, connectivity, function, and subnetwork organizations. We then review the key aspects of sex-biased brain anatomy and connectivity that have been revealed by these techniques; sex differences in stimulus-based or task-based functional magnetic resonance imaging (fMRI) studies are not addressed here. Next, we discuss specific disease states that appear to have different outcomes in the 2 sexes due to baseline differences in the "connectome" and animal models used in neuroimaging. Finally, we will address some important caveats and controversies in the field of brain imaging.

Brain Imaging Techniques

Modern neuroimaging methods make it possible to characterize diverse aspects of brain structure, function, and connectivity in vivo. This large toolbox of methods has been used to examine sex differences in brain organization at several levels of analysis. These techniques aim to analyze, map, and visualize regional and inter-regional (connectomic) features of the brain at macroscopic (systems-level) and mesoscopic (neural circuit architecture) levels in order to illuminate brain organization in health and disease (135). Of note, cellular-level details are beyond the resolution of most in vivo brain imaging techniques.

Sex differences in global and regional brain anatomy can be measured in vivo using structural magnetic resonance imaging (sMRI). Several considerations have made sMRI an especially popular technique in the study of brain sex differences in humans. First, sMRI allows a quick and spatially comprehensive screen of the entire brain that can quantify thousands of morphometric properties simultaneously in vivo across a large number of individuals. These characteristics not only facilitate testing for sex differences outside defined regions of interest, but also allow longitudinal measurements that can track the emergence of brain sex differences over development (136, 137). Second, because sMRI considers structure rather than function, it can leverage evolutionary conservation of the basic mammalian brain plan (138), and it is therefore particularly well-suited for cross-species investigation of sex differences in humans and animals. Thus, a critical role for sMRI research in the study of brain sex differences is to screen for brain regions that can then be prioritized for closer analysis using more resource-intensive assays that are typically applied in a regionally selective manner.

Complimenting sMRI, other in vivo neuroimaging techniques such as diffusion MRI (dMRI), resting state functional MRI (rs-fMRI), and fMRI provide unprecedented insights into tissue microstructure and brain connectivity. fMRI maps brain circuitry based on stimulus- or task-based brain functional responses. In contrast, rs-fMRI, by measuring changes in blood flow in the brain generated by signals dependent on blood-oxygen-levels, helps explore the brain's functional organization by providing insights into intrinsic brain activity without requiring participants to be trained in specific tasks, thereby eliminating task performance as a confounder (139, 140). dMRI measures the differential patterns of water diffusivity in biological tissue revealing details of tissue microstructure, especially in white matter (141). Fiber tractography on dMRI enables mapping the fiber architecture of the brain, and subsequently, the network organization of the brain through structural connectomes (142-144). A brain connectome is an extensive map of the white matter structural or functional connections of the brain, created using dMRI or rs-fMRI (145). Modeling efforts, such as the Human Connectome Project, and the use of connectome-based predictive modeling, have provided an integrative, in-depth, and multilevel understanding of the structural and functional connectivity (regions that get coactivated) of the neuronal networks (146, 147).

Sex Differences in Global and Regional Brain Anatomy

It is well established that men have an average total brain volume that is approximately 10% greater than that of women (148, 149). A similar sex difference in average

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human brain volume (~8%) appears to be present at birth (150) and is sustained throughout childhood and adolescence (151). The sex differences for total brain volume also hold for the 2 main subdivisions of brain tissue—gray matter and white matter—despite these 2 brain compartments following very different developmental trajectories (151, 152) (Fig. 3).

The robust sex difference in brain volume identified through human sMRI research cannot be fully explained by the fact that brain volume is positively correlated with height (average height is greater in men that in women). Statistical control for body size diminishes, but does not remove, sex differences in total brain volume (149), and boys also show greater average brain volume than girls during early adolescent development, at a time when girls are taller than boys (153). Thus, available literature supports a consistent picture in which there is overlap between the distribution of brain size in men and women, but the mean of this distribution is significantly greater in men than women. The medium effect size of sex on brain volume exists above and beyond sex differences in stature. However, it is important to note that no known functional sex differences associate with the sex difference in overall brain size. Sex differences in overall brain size, and their developmental timing, are both theoretically and methodologically important when considering: (i) whether neuroanatomical sex differences are conserved across species; (ii) whether there are sex differences in regional brain anatomy above and beyond sex differences in overall brain size; and (iii) whether there is concordance between sex differences in brain size and any observed associations between brain size and putative biological causes of sex differences, such as gonadal or sex chromosome status (see below).

The patterning of sex differences in behavior and mental illness risk across the lifespan suggest that sex differences in human brain organization are likely to vary across different brain sub-systems or regions, and potentially also across different developmental periods. Structures in human gray matter compartments mediate neural computation and information processing—in contrast to axon-rich white matter compartments that are primarily involved in connectivity between different brain regions (see "Sex Differences in Brain Network Organization: The Brain Connectome," below). Here, we focus on sMRI studies that have tested for sex differences in regional gray matter volume (regional GMV) after controlling for sex differences in overall brain size. Regional GMV sex differences that survive statistical correction for total brain volume variation are of special interest because they exist beyond global sex differences in brain size. We emphasize GMV rather than other morphometric properties of the brain such as cortical thickness, sulcation, or the shape of subcortical structures (144, 154), because GMV provides a common metric that can be examined across cortical and subcortical structures, with equal applicability to humans and mice. Independent large-scale human sMRI studies in biobanks have identified a reproducible pattern of sex differences in regional GMV using sample sizes that are

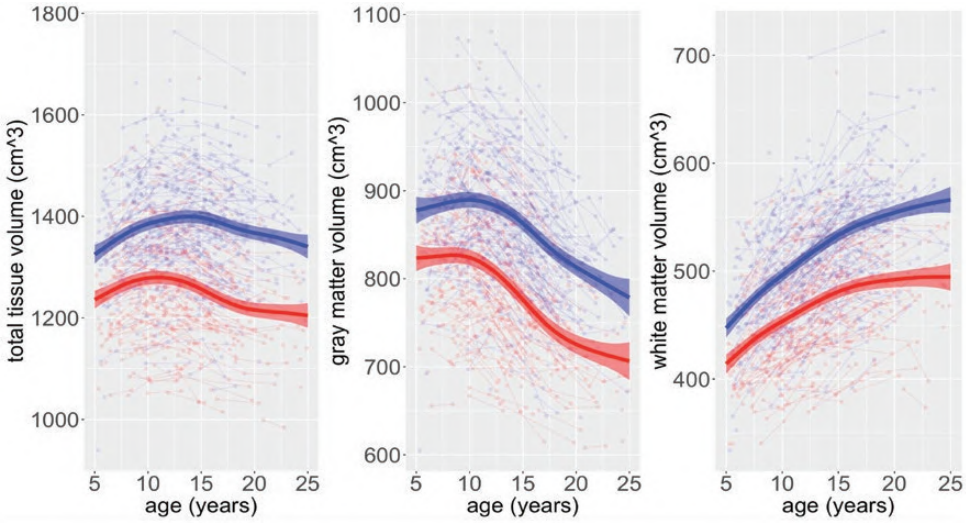

Figure 3. Developmental trajectories for total brain tissue volume, gray matter volume, and white matter volume in men and women over Development. Person-level data are shown for women (red) and men (blue) as points, with lines linking measures from the same person over time. Note the large interindividual variation in volumes within each sex, and the overlap of these distributions, between the sexes. Superimposed on these person-level data are group-level best fit volume trajectories (bold lines with shaded 95% confidence intervals). The developmental window covered is 5 to 25 years of age. For all plots, there are statistically significant sex differences in both trajectory shape (ie, sex differences in the tempo of volume change, $P < 0.00001$), and trajectory "height" (ie, sex differences in absolute volume across ages, $P < 0.00001$). [Adapted with permission from Giedd JN et al. *Neuropsychopharmacology,* 2015; 40 © Springer Nature (153)].

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significantly larger than those used in earlier work (148, 149, 155). A structural neuroimaging study involving >2000 individuals demonstrated that higher regional expression of sex-linked genes was coupled with greater GMV in men relative to women (155). These studies, by different laboratories, using different datasets and different techniques for sMRI analysis, find a largely overlapping regional pattern of GMV sex differences after correction for sex differences in total brain volume. These independent replications of regional sex differences in GMV are also in agreement with meta-analytic studies (156). Together, these studies show that, in adulthood, regional GMV is (on average): (i) greater in women than men within superior parietal, dorsolateral frontal, and anterior cingulate cortices; and (ii) greater in men than women within occipital, fusiform, and parahippocampal cortices as well as the amygdala and putamen. Furthermore, while these studies lack temporally resolved developmental maps of male-female differences in regional GMV throughout the brain, there is extensive evidence from focused studies of particular structures that neuroanatomical sex differences can vary dynamically over development, such as observed with amygdala volume and shape (156).

The rapidly expanding body of sMRI research on regional GMV sex differences in the murine brain shows important overlaps and differences with findings from human studies (137, 157). These murine sMRI studies—which are most commonly conducted ex vivo at a spatial resolution of <100 μm throughout the whole brain—have been able to confirm the identification of all classically sexually dimorphic nuclei of male-biased volume from prior histological research, including the bed nucleus of the stria terminalis and medial amygdala (137, 157). These brain regions play a predominant role in modulating social and goal-directed behaviors, pain, and cardiovascular control, all of which are conserved among mammalian species and subject to sexually dimorphic outcomes. By allowing a full-brain screen, murine sMRI has also newly identified a reproducible set of regions with greater GMV in females, including the cerebellar cortex, ventral thalamus, and somatosensory cortex (137, 157). Furthermore, a longitudinal sMRI study in mice found that the set of regions with male-biased GMV can be detected by early postnatal life (with some accentuating over puberty), whereas regions of female-biased GMV in murine adulthood appear to emerge in adolescence (137). To date, there are no studies that formally seek to compare the spatiotemporal patterning of regional GMV sex differences in humans and mice, although existing work already suggests some potential homologies, including foci of greater cerebellar cortex GMV in females vs males by adulthood (137, 148) and the adolescent accentuation of male-biased amygdala volume (158, 159). An important technical challenge in assessing the degree of anatomical homology between regions of sex-biased brain anatomy in humans and mice is that most of the best-established and histologically validated foci of sex-biased brain volume in mice (eg, bed nucleus stria terminalis, medial preoptic nucleus of the hypothalamus) are hard to image in humans due to their small size and intrinsic tissue contrast properties.

Sex Differences in Brain Network Organization: The Brain Connectome

The structural or functional brain network is represented by a "connectome," wherein the structural or functional connectivity between coactivated regions is encoded either through fiber tracts or functional co-activations (160). These connectomes can be studied at the level of subnetworks like visuospatial, auditory, cognitive control, or macro-scale level through global measures of network segregation, integration, and efficiency, to obtain functional associations (161).

A study of 949 individuals (aged 8-22 years; 428 males and 521 females) showed that on average, there are significant differences between the sexes in their structural connectomes (Fig. 4) (162). On average, men had greater within-hemispheric connectivity, as well as enhanced network segregation, whereas between-hemispheric connectivity and network integration predominated in women (Fig. 4A), but these differences were most prominent during adolescence (Fig. 4B-4D). However, an opposite trend was seen for cerebellar connections, which developed differently between human males and females in adolescence and adulthood. The structural connectivity findings were consistent with a behavioral study conducted on the parent cohort (the above-mentioned imaging study was performed on a subset of participants), with women outperforming men on attention, word and face memory, and social cognition tasks, and men performing better on spatial processing and motor and sensorimotor speed tasks (163). An analysis of the Human Connectome Project rs-fMRI data identified age and sex as independent variables that contributed to differences in functional connectivity (164). In brains of men, functional connectivity was more clustered locally in all lobes, except in the cerebellum, whereas the brains of women showed a higher clustering coefficient at the whole-brain level. Thus, brains of men were classified as more segregated and brains of women as more integrated, which agrees with the structural connectivity findings (162). In connectomes, the identification of subnetwork properties (165) can reveal how the complex functional and behavioral repertoire emerges from the simultaneous processes of segregated neuronal clusters and their

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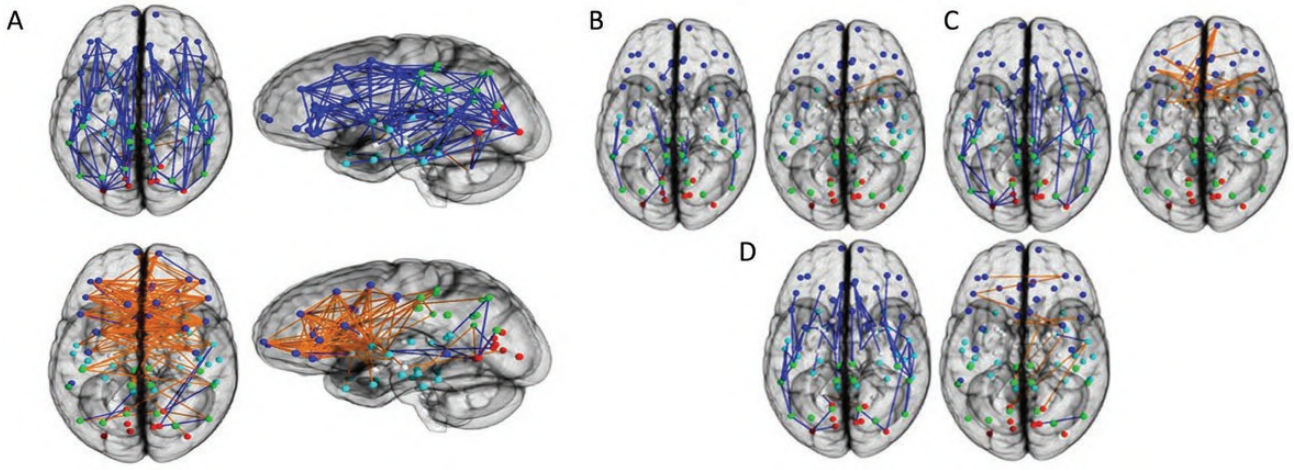

Figure 4. Sex differences in structural connectomes across development. Connectomes representing the white matter structural connectivity in the brain, with nodes indicating the brain regions and edges between the nodes representing the structural connectivity between the nodes. Node colors representing respective brain regions are as follows: dark blue, frontal; cyan, temporal; green, parietal; red, occipital; white, subcortical. The depicted edges shown are those that survived permutation testing at $P = 0.05$. **A,** shows increased intrahemispheric connectivity in men (Upper, in blue) and increased inter-hemispheric connectivity in women (Lower, in orange) on average. **B-D:** Connectivity differences shown in A separated by age groups are shown: **B,** under 13 years, **C,** adolescent (13-18 years), and **D,** young adults (18-22 years). Left image: Men/Boys; Right image: Women/Girls. [Adapted with permission from Ingalhalikar M et al. *Proc Natl Acad Sci U S A,* 2014; 111(2) © National Academy of Sciences (163)].

integration during complicated cognitive tasks (166, 167). Consistent with the behavioral findings on sex differences, men had increased connectivity between motor and sensory (auditory) systems, along with increased connectivity in the fronto-parietal and cingulo-opercular systems that are traditionally associated with complex reasoning and control, whereas women had higher connectivity between reward, memory, and sensory (auditory) systems (163, 168). Better spatial skills in men and improved memory and social cognition skills in women have been reported in behavioral literature (169, 170).

It is important to point out that observed group-level differences in brain structure, function, or connectivity in men and women may reflect the influence of several extraneous factors. For example, in a set of elegant studies, brains of men were imaged to ascertain the contribution of performing complex spatial navigation tasks as part of their daily work on gray matter volume. These studies found that posterior hippocampi of London taxi drivers were significantly larger compared with controls (171), although the work did not address sex differences. Driving a taxi in London before the era of digital maps/navigation systems required extensive training and learning to navigate complex routes before being given a license to operate. In a subsequent study, comparison between London taxi drivers and bus drivers matched and controlled for age, education, intellectual, and stress levels, as well as years of driving experience, showed that taxi drivers had greater GMV in the posterior and less volume in the anterior hippocampi compared with bus drivers (172). Interestingly, years of navigation experience associated with hippocampal volume in taxi drivers alone, but they were significantly worse at acquiring or retrieving novel visuo-spatial information than bus drivers. Importantly, no differences in other GMV, including the caudate nucleus, were found between the taxi and bus drivers; the caudate nucleus is associated with a myriad of cognitive and emotional functions. These studies illustrate brain plasticity and that professional work and years of performing certain tasks can result in brain structural, volume, and connectivity differences that may have little to do with sex or gender per se, but more with training, social environments, and behaviors. In other studies, GMV changes were greater in professional musicians, or after induced training (juggling for 3 months), and in early bilinguals, and white matter volume changes were found in adults learning a second language, irrespective of sex, when reported (173-176). These findings suggest that brain structure retains its plasticity and controlling for factors other than sex or gender are key in interpreting data on structural volumes and associated functions.

The above-mentioned existing datasets did not collect the requisite information on self-report of gender, thereby precluding retrospective analysis of gender in these cases. As identifying correspondence between behavioral scores and the regions that are involved in the manifestation of that behavior remains challenging, analyses of subnetworks pertaining to functional and behavioral domains can help elucidate a brain-behavior correspondence. The detailed description of sex differences in brain organization at the group level, and concerted efforts to specify

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the role of sex-biased biological factors in shaping such sex differences, is of fundamental importance (177) and also provides a crucial adjunct for indispensable studies on environmental and wider societal contributions to sex-biased brain development. Such studies should be undertaken jointly using structural and functional connectivity. These studies elucidate the various ways in which sex differences in brain microstructure and connectivity can be investigated.

Sex Differences in Structural and Functional Brain Regions in Obesity

The hypothalamus has long been known as the "center" where peripheral and neural signals converge in the regulation of food intake and energy homeostasis in both sexes. Advances in neuroimaging studies have helped identify activation of several distinct brain regions comprising brain networks in response to eating in men and women. Behavioral and sociocultural factors may play a role in the observed sex differences in ingestive behaviors, appetite, and cravings related to obesity (178). Women report higher prevalence of maladaptive ingestive behaviors such as binge eating, food cravings, and "food addiction," and the lifetime prevalence of disordered eating behaviors are about 3 times higher in women than in men (179, 180). Women also experience episodes of food cravings of greater intensity (181, 182), and greater frequency (183-185), and are less able to suppress food cravings than men (184, 186). Despite the wealth of data indicating that women experience disproportionately higher rates of food cravings, stress eating, and eating disorders than men, the reasons for these differences are incompletely understood (184, 187).

Regulation of food intake entails both homeostatic and nonhomeostatic factors (188). Homeostatic regulation balances energy needs with energy consumption, whereas nonhomeostatic regulation—in particular hedonic regulation and food addiction—involves reward-seeking behaviors that drive humans and animals to consume food beyond their metabolic needs, leading to the development of obesity (189-191). These findings have directed attention toward the extended reward system in obesity-related research, which consists mainly of basal ganglia regions and is involved in dopamine signaling and addiction-like behaviors (192). The extended reward system is composed of 6 interconnected brain networks—salience, central autonomic, basal ganglia, somatosensory, executive control, and emotional regulation (192).

Functional MRI studies have found that, in response to food images, obese individuals show greater activation than normal-weight individuals in regions associated with reward anticipation, dopamine signaling, and addiction-like behaviors (193-196). Greater activity in brain regions of the extended reward network may drive obesity-related behaviors, such as greater responses to food odors and food consumption (197-199). Recent meta-analyses have further supported the role of the brain in disrupting the balance between energy consumption and expenditure. This combination of increased activity in regions associated with reward-driven behaviors and decreased activity in regions moderating top-down control of appetite may lead to consumption of excess calories (188).

Furthermore, sex-specific activations in response to food intake have been observed in cognitive, emotional, and reward-related regions (200-202). For example, obese men had greater activation than obese women in the supplementary motor area, precentral gyrus, fusiform gyrus, and inferior parietal lobule, which are associated with motor control, visuospatial attention, and responding to salient new or alerting stimuli (203). In this same study, obese women showed greater activation than obese men in the caudate and parahippocampal gyrus, regions implicated in reward processing and memory (203). Using graph theory to define the underlying architecture of brain structural connectivity obtained from diffusion tensor imaging, sex differences were observed in the topological measures of centrality (which determine the degree of information flow in specific brain regions) in regions of reward and salience networks in women, and in reward and sensorimotor networks in men (204). Resting state fMRI studies have found sex differences and commonalities in body mass index (BMI)-related connectivity associated with specific defined regions of interest in the reward network (205). For example, women had increased associations between BMI and increased connectivity in the in right globus pallidus and bilateral putamen. In men, BMI was associated with increased connectivity in the medial frontal cortex. A study of sex differences in response to visual and auditory food cues found that women experience greater activation in lateral and dorsolateral prefrontal and parietal cortical regions involved in cognitive planning and executive guidance and evaluation of behavior, compared with men (202). When viewed together, these studies highlight the importance of investigating sex differences in obesity-related alterations in the core and extended reward networks.

Although many single-sex studies of fMRI and obesity have been published, with the majority having all-female subjects, few studies have specifically investigated sex differences in brain function and structure in obesity. Despite the literature supporting sex differences in the brain, including in regions implicated in reward behaviors and energy homeostasis, few comprehensive reviews of sexually dimorphic brain signatures related to obesity have

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been performed. A recent meta-analysis using an activation likelihood estimation approach to evaluate comparisons in functional responses to stimuli by obesity and by sex revealed differential sex- and BMI-related activations in reward anticipation and response, in shaping food-related memories, and in generating top-down control of appetitive processes. Together, these findings have important implications for sex-specific obesity treatments.

Models to Study Sex Differences in Normal Brain Structure and During Pathophysiology

Studies of sex differences offer important considerations for personalized medicine. The prevalence, clinical presentation, and symptomatic progression of many neurological and psychiatric disorders are remarkably different between the sexes. In addition to common X-lined mental retardation syndromes, men have a greater prevalence of neuropsychiatric disorders such as autism, attention-deficit/hyperactivity disorder (ADHD), and Tourette syndrome (206), whereas women have a greater prevalence of mood and eating disorders (207, 208). From the perspective of developmental disorders, the differences in the developmental trajectories of the sexes perhaps represent different vulnerabilities of maturing brain circuitry, leading to differences in symptoms, onset, and severity of neurological disorders. There are also sex differences in the risk factors, average age of onset, and prevalence of late-life dementias, as well as cerebrovascular disease (209). Additionally, in traumatic brain injuries, where the network organization of the brain is affected by the injury, such as the corpus callosum region, sex differences in inter-hemispheric connectivity and brain subnetworks may influence the impact of injury, and hence subsequent recovery. Thus, sex differences in brain connections are crucial to identify, as they may elucidate mechanisms in disease risk and potential treatment and recovery (210).

Most models of sex-biased mammalian brain development are based on experimental data from rodents (now largely from mice, but previously also from guinea pigs and rats). One of the most systematic dissociations of gonadal and chromosomal contributions to sex-biased anatomical brain organization in mammals is provided by a recent sMRI study of adult mice from the FCG model (112, 211). By combining sMRI with behavioral assays, these studies determined the contribution of sex chromosomes and gonads to adult mouse brain structure and function (211). This study revealed: (i) an effect of sex chromosomes on regional GMV in the cerebellar cortex and olfactory bulb; and (ii) an effect of gonads on regional GMV in the parietotemporal cortex and the bed nucleus of the stria terminalis. Some of these effects overlapped with regions of normal sex differences in murine GMV (eg, cerebellar cortex and bed nucleus of the stria terminalis), and some brain regions were anatomically sensitive to both effects (basal forebrain and periaqueductal gray matter). Sex-chromosome effects on regional gray matter anatomy have also been reported by complementary sets of sMRI studies in both mice and humans that compare groups of euploid individuals with groups carrying X-chromosome aneuploidy (157, 212). Finally, in both mice (137) and humans (155), the spatial patterning of sex differences in regional GMV in adulthood appears to be preferentially aligned with the spatial patterning of sex-chromosome gene expression—which points toward a potential role of sex-linked genes in the establishment of maintenance of regional GMV sex differences. These studies emphasize the need for integrative models that view biological contribution to sex-biased brain development as a developmental dance of coordinated influences from both gonads and sex chromosomes.

Caveats and Critiques Relating to Neuroimaging of Brain Sex Differences

While several sMRI studies apparently establish that there are highly reproducible male-female differences in regional gray matter volume after controlling for variation in total brain size in humans, this conclusion should be considered in the light of several important caveats and critiques to avoid misinterpretation. First, all sMRI phenotypes that show reproducible and statistically significant sex differences also show a considerable overlap between men and women. This overlap is illustrated by total brain volume: total brain volume averages 10% greater in men than women, but many women have a total brain volume above the 30th centile for male brain volume, and many men have a total brain volume below the 30th centile for female brain volume (149). Sex differences in brain structure and organization are present across the lifespan and vary based on age, so inferences should be drawn cautiously. Thus, while total brain size shows a robust mean difference between men and women, an individual's total brain volume is a weak predictor of biological sex. These 2 facts arise because biological sex is only one source of variation in brain size (149), and other factors/variables that influence total brain size are unknown and/or hard to model statistically (Fig. 1). By extension, because sources of anatomical variation can differ between brain regions—the same individual can have GMV values that appear to be "sex-typical" in one region, but "sex-atypical" in another (when typical and atypical are defined by an individual's percentile position relative to the distribution of population-level trait variation in each sex) (213). This interpretation offers one

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potential explanation for the observation that an individual brain can show varying degrees of GMV "sex-typicality" in different brain regions (relative to the population distribution). Alternative explanations have been proposed, including regional variations in programs of sex-biased development such that one individual's brain may be considered a "mosaic" of male and female parts regardless of their chromosomal and/or gonadal sex (213).

Second, although sex differences in regional GMV are highly reproducible in humans and mice, these mesoanatomical sex differences *cannot* be assumed to correlate with behavioral sex differences. The functional relevance of neuroanatomical sex differences is hard to establish experimentally in humans, but correlations between anatomical and behavioral sex differences could be modeled in humans using several feasible study designs. To date, however, very few studies have directly tested for such structure-function correlations in humans (161), and this is an important priority area for future research. Several other challenges will need to be addressed in future work for any given sex-biased sMRI phenotype, including which aspects of behavior to measure and how to consider properly all possible configurations of brain-behavior association in 2 groups (eg, varying intercepts and/or regression slopes across groups). Moreover, some sex-biased sMRI phenotypes, such as trajectories of anatomical change, can only be estimated from group-level data, which complicates comparisons with interindividual variation in behavior. More fundamentally, however, regional GMV sex differences may be useful for understanding the brain basis for sex-biased behavior without GMV variation itself being the behaviorally relevant marker. For example, sex differences in mean regional GMV may help to define brain circuits that subserve sex-biased behaviors through their molecular, cellular, or connectivity features rather than through their volume *per se*. It is also important to entertain the possibility that sex differences in the anatomical organization of a given brain system may actually serve to equilibrate function between the sexes despite each sex having a categorically different genetic starting point.

Third, in addition to the functional considerations above, full understanding of a given sex bias in regional brain anatomy requires a mechanistic account that can link observed anatomical sex differences back to specific genetic and/or environmental factors that differ between men and women. It is usually impossible to disentangle biological sex differences from those which could be the result of environmental influences during development, differences in gender, and in sexual orientation (Fig. 1). Strict causal tests for mechanistic models of sex-biased brain development are very hard to achieve in humans, although several informative approaches have been pursued including: (i) modeling sMRI data using normative variation in hypothalamic-pituitary-gonadal axis maturation or function (214); (ii) applying sMRI methods to cohorts undergoing gender-reassignment (215); and (iii) studying how sMRI features differ between typically developing groups and those affected by medical disorders involving the sex chromosomes (eg, sex chromosome aneuploidies) or sex steroids (eg, androgen insensitivity, congenital adrenal hyperplasia) (215, 216). However, the opportunistic and correlational nature of these approaches places considerable limits on the inferential power of mechanistic studies of human sex-biased brain development. Moreover, as challenging as it is to study chromosomal or gonadal factors in humans, it is even harder to address empirically the many plausible hypotheses about the potential for experiential and societal influences to differentially shape brain development in both sexes (121) or genders.

Section III

Sex Differences in Molecular Mechanisms Underlying Brain-Gut Disorders

The brain and the gut communicate with each other in a bidirectional way through parallel and interacting channels, involving immune, endocrine, and neural signaling mechanisms (217). The brain is able to modulate gut permeability, motility, intestinal transit, and microbial function via the autonomic nervous system (217), and the gut in turn sends signals to the brain to modulate behavior, in rodents (218). This brain-gut communication is especially critical in mediating stress responses and in stress-based disorders. In psychiatric and other neurological diseases, there are notable sex differences that point to different underlying neurobiological mechanisms in men vs women (219-221). Despite their clear documentation, these sex differences have largely been ignored, in order to develop broadly applicable pharmacotherapies that come at a considerable cost, especially for women's health (222, 223). Sex biases in psychiatric risk are particularly instructive as they are developmentally patterned in a manner that is highly reproducible across different cultural settings and historical epochs: early-onset neurodevelopmental and gut disorders are more prevalent in boys than girls, while the opposite sex-bias is seen for adolescent-emergent mood disorders (134, 224). Brain-gut disorders are more prevalent in women than men, but this may be due to underreporting by men due to social stigma associated with several of these

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disorders. The etiologies and risk factors for several brain-gut disorders differ between the sexes, yet study designs include predominantly male sex. In this section, we discuss the possibilities that shared and distinct mechanisms operate in males and females resulting in similar as well as distinct manifestation of symptoms for a given disease/disorder.

Sex-Related Differences in Obesity

Although prevalence rates for obesity are at unprecedented levels in all ages (225) and are almost equal in men and women (except when stratified by race or ethnicity) (226), recent surveys indicate an increase in the incidence of obesity in adults and sex differences in the associations between weight, physical health, and psychosocial functions (227, 228). Sex differences in body fat distribution have also been observed (178, 229), with women showing an increased propensity to gain total body fat, especially subcutaneous abdominal fat, whereas men tend to have more visceral adipose fat (230), which is associated with higher risks of type 2 diabetes, hypertension, dyslipidemia, and cardiovascular disease (231). Most clinical trials do not report sex differences related to health outcomes or treatment responses, but a few existing reports suggest women are less likely to complete treatment, tend to lose less weight than men, have a greater number of unsuccessful attempts to maintain weight loss resulting in the well-known "yoyo" diet phenomenon, and have limited responses to pharmacological treatments (225). Obesity-related studies in humans and rodents have expanded in scope to not only focus on structural and functional brain differences between obese and lean male and females, but also include investigations into the bidirectional signaling associated with the brain-gut microbiome axis (232, 233). In obese individuals, changes in the relative abundance and gut microbial diversity have been linked to changes in metabolism, insulin resistance, inflammation, and fat deposition (234). The importance of the intestinal microbiome to human health has been of interest over the past few decades, with multiple studies now linking the microbiome to energy homeostasis, immune function, and development of obesity and metabolic syndrome (235-237), even though few studies have addressed causality.

Not only does the brain-gut axis demonstrate changes in obese individuals, but evidence also highlights differences in the microbiota based on sex hormones (238). More recently, the effect of sex hormones on the composition of the gut microbiota has been explored, with differences seen in the microbiota between men and women during various stages of human development and maturation (238). These sexually dimorphic microbiome signatures are likely to contribute to differences in susceptibility to autoimmune and metabolic diseases between the sexes. Studies performed in immunocompromised mouse models have shown delayed onset and lessened severity of type 1 diabetes in female mice who receive male microbiota transplants; testosterone activity and androgen receptor signaling was essential for this protection (239, 240).

These sex-specific differences in the microbial communities persist throughout adult development, with murine models demonstrating the role of testosterone in orchestrating these divergences in host selection of microbial communities (240). In rodents, males exhibit lower microbiome variability relative to females, likely due to the pulsatile nature of estrogens (240). Human studies comparing the microbiome of twins also revealed more divergences in microbial composition in opposite-sex versus same-sex twins (241). When the cecal contents from adult male mice is transferred into female mice, metabolomic profile changes and masculinization of the hormonal profile results, suggesting the gut microbiota's influence on sex-specific metabolic and behavioral phenotypes (239, 242).

Circulating estrogens in the body are metabolized by the liver and undergo methylation, hydroxylation, and conjugation reactions to produce metabolites that affect host metabolism (243). Certain metabolites are excreted through the bile and are further processed by microbial enzymes in the distal small and large intestine. Certain microbial species secrete beta-glucuronidase, an enzyme that deconjugates biliary estrogen metabolites and allows for its reabsorption into the bloodstream to act on distal sites through binding of estrogen receptors (244). Dysbiosis and decreased microbial diversity result in decreased production of absorbable estrogen metabolites. This mechanism has been implicated in pathologies associated with low circulating estrogens, such as obesity, metabolic syndrome, cardiovascular disease, and cognitive decline in women (245, 246); however, estrogen replacement therapy does not reverse these conditions (247). Growth hormone similarly contributes to sexually dimorphic responses in the above-mentioned diseases (248). In addition, estrogens modulate inflammatory pathways driving disease processes such as nonalcoholic fatty liver disease (NAFLD) and type 2 diabetes (249, 250). More specifically, estrogens regulate adipokines and lipopolysaccharides, which respectively are adipocyte-derived hormones and endotoxins that have been associated with type 2 diabetes (251). Adipokines play a role in metabolic homeostasis as well as in mediating the beneficial and detrimental effects of inflammation (252). The androgen- and estrogen-dependent regulation of adipokines, including leptin, resistin, adiponectin, and visfatin, provides a possible mechanistic link between metabolic disorders (obesity,

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atherosclerosis, insulin resistance) and autoimmune dysfunction. The estrogen-microbiome axis can provide a potential avenue for a sex-specific approach to combating metabolic disorders and highlights the bidirectional interaction of estrogens and microbial communities in the pathogenesis of disease processes.

Although the exact signaling mechanisms underlying the communication within the brain-gut-microbiome axis remain incompletely understood, tryptophan metabolites have been implicated as important signaling molecules (253). The most extensively studied tryptophan metabolite is serotonin (5-HT), a molecule with diverse roles in both the gastrointestinal tract (ie, peristalsis, secretion, and absorption) and the central nervous system (ie, mood, pain modulation, behavior, sleep, and ingestive and cognitive functions) (254). Tryptophan also acts as a precursor to the kynurenine (KYN) family of molecules (255). In obesity, the KYN pathway is preferentially activated and may contribute to immune-mediated inflammation, which may drive inflammation-associated changes to the extended reward network described in previous brain studies, particularly changes involving the amygdala and lateral orbitofrontal cortex (256-259). KYN may also modulate signaling within the brain-gut-microbiome axis through downstream neuroactive metabolites, such as kynurenic acid and quinolinic acid, functioning as N-methyl-D-aspartate (NMDA) antagonists and NMDA excitotoxins, respectively (260). Sex differences have been reported in these metabolite products in obese individuals, with lower tryptophan levels but elevated KYN and KYN/tryptophan ratios in women with high BMI compared to men with high BMI (256, 261, 262).

Sex Differences in Stress-Based (Patho) Physiologies

Epidemiological data reveal that the majority of psychiatric disorders occur at different rates in men and women. For example, men are more likely to suffer from attention-deficit/hyperactivity disorder (ADHD), whereas women are more likely to suffer from major depression and posttraumatic stress disorder (PTSD) (219, 263-265). Even when the rates of disorders are similar, their presentations can differ. Schizophrenia, for example, is only slightly more common in men than women, but men develop schizophrenia at an earlier age and present with more negative symptoms, such as social withdrawal and lack of motivation. (224). In the case of bipolar disorder, rates are similar between the sexes, but women more often have more rapid cycling and mixed episodes and they report higher comorbidity with eating disorders and PTSD, whereas men report higher comorbidity with alcoholism (266). Not only does the risk and presentation of psychiatric disorders vary between men and women, but there are differences in treatment responses. For example, the efficacy of antidepressants differs between the sexes: men respond better to tricyclic antidepressants, whereas women respond better to selective serotonin reuptake inhibitors (267, 268). These findings implicate neurobiological sex differences in contributing to disease. In support of this idea, recent studies using animal models are beginning to uncover molecular processes that can bias males and females toward different pathology. Findings from some of these basic research studies will be highlighted here as examples of how including sex as a biological variable can inform our understanding of the etiology of stress-based disorders, as well as guide the development of better treatments.

While there are sex differences in rodent studies in the structure and the size of certain brain regions that can contribute to sex differences in behavior (211), imaging studies that focused on sex differences in cortical thickness and gyration suggest a role for these brain regions in humans as well. In adolescent girls, cortical thinning in the right temporal regions, the left temporoparietal junction and the left orbitofrontal cortex is faster than in boys (154). In contrast, changes in cortical folding were only found in one cluster of the right prefrontal region, suggesting that the mechanisms underlying changes in cortical thickness and gyrification in adolescents are distinct. Sexual dimorphism in the developmental course of the cortical maturation, which coincides with the onset of puberty, might explain sex differences in the age of onset and clinical presentation of many psychiatric disorders (154). Recent evidence has revealed that molecular sex differences in the brain are more widespread than initially thought and such seemingly small-scale differences can have a large impact on physiology and behavior (269). Neurons typically communicate with each other via neurotransmitters and neuropeptides, which are released from a presynaptic neuron and travel across a synapse to bind to receptors on the postsynaptic neuron to exert downstream cellular effects. There are sex differences in production and release of many neurotransmitters and neuropeptides that can result in behavioral changes. In other instances, sex differences in these systems are compensatory, leading to similar behavior endpoints via different mechanisms. For example, both male and female juvenile rats play, but the release of the inhibitory neurotransmitter gamma-aminobutyric acid (GABA) into the lateral septum mediates juvenile play only in female rats (270). There are also sex differences in receptors that can influence how these neurochemicals affect their downstream targets. For instance, dopamine 1 (D1) receptors, which belong to the family of G protein-coupled receptors (GPCRs), in the nucleus accumbens, are necessary for social

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withdrawal in female but not male California mice (271). The function of GPCRs is often complex and they can induce different downstream effects depending on their conformation and location. Sex differences can occur at each level of receptor function, in some cases altering physiology differently in male vs female rodents. Sex differences in GPCR signaling are particularly important to consider, especially given that GPCRs are the most studied drug target family for a myriad of indications; in fact, 34% of all US Food and Drug Administration (FDA)-approved drugs are targets of GPCRs (272). As an example of the myriad of sex differences that can be mediated by receptors, we will use the corticotropin-releasing factor 1 and 2 (CRF_1 and CRF_2, respectively) receptors that facilitate responses to stress, exhibit sexually dimorphic expression pattern, are modulated by both estrogens and androgens, and have been relatively well characterized in both sexes (273, 274).

Upon perception of stress or perturbation of homeostasis, CRF is synthesized in the paraventricular nucleus and released from the median eminence of the hypothalamus into the pituitary portal circulation, which in turn stimulates the synthesis and secretion of adrenocorticotrophic hormone (ACTH) from the anterior pituitary into the general circulation. ACTH acts on the adrenal cortex to stimulate the synthesis and release of glucocorticoids and other steroids. This activation of the HPA axis in the classic "flight or fight" response by the CRF system is present in all mammals. The mammalian CRF family comprises 4 agonists, CRF and 3 urocortins (UCN1-3); and 2 known class B GPCRs, CRF_1 and CRF_2. While CRF_1 and CRF_2 share ~68% identity at the amino acid level (275), they perform distinct functions; CRF binding to CRF_1 initiates stress responses by activating the HPA axis, whereas UCN1-3 binding to CRF_2 brings systems back to homeostasis (274). Not surprisingly, perturbations in the components of the CRF family impact several organs and lead to brain-gut disorders, type 2 diabetes, metabolic syndrome, cardiovascular, and reproductive diseases, among others (274). There are sex differences in CRF's endocrine effects. In female rats, higher levels of CRF mRNA in the paraventricular nucleus are reported that associate with the estrous cycle (276, 277) and are reviewed elsewhere (274). Perhaps as a compensatory response, CRF binding protein, an endogenous protein that sequesters CRF thus preventing its bioavailability, is expressed at higher levels in the pituitary of female compared with male mice (278). In humans, there is evidence for increased CRF receptor sensitivity at the level of the pituitary of women relative to men, because peripherally administered CRF, which acts at the pituitary, increases ACTH to a greater degree in women (279).

During stress, CRF is also released centrally into many brain regions, where its neuromodulatory effects coordinate cognitive and behavioral changes to promote stress coping (280). There are sex differences in the way these brain regions respond to CRF that are largely due to sex differences in CRF receptor signaling (274). For example, there is greater CRF_1 receptor binding in the basolateral amygdala in female rats (281). In contrast, binding of the CRF_2 receptor subtype, which is involved in stress recovery, is greater in the central nucleus of the amygdala in male rats (281). It is unknown precisely how these sex differences affect behavior, but given that the amygdala is critically involved in fear, it is likely that these receptor sex differences differently alter fear processing in males and females. In the brain, CRF_2 is most abundant in the bed nucleus of the stria terminalis, a region that regulates sexual behavior and stress-related functions (282, 283). Promoters in genes for CRF_1 and CRF_2 receptors harbor estrogen and androgen responsive elements and show tissue-specific modulation by sex hormones (284, 285). The sexually dimorphic expression pattern of these receptors at normal physiological states and during stress or disease pathology are summarized in a recent review (274).

Sex differences in CRF_1 receptor signaling have been identified in the noradrenergic-containing nucleus of the locus coeruleus (LC) and these differences have important implications for understanding disease vulnerability (273). The LC-noradrenergic system regulates levels of arousal such that higher levels of norepinephrin are associated with greater levels of arousal (286-289). Stressor exposure causes CRF to be released into the LC, which speeds up LC neuronal firing, increasing norepinephrin release (290, 291). Activation of this system during an acute or moderate stressor is thought to be adaptive, because it is important to be alert during a stressful event. However, if this system is activated inappropriately or persistently it can lead to hyperarousal that contributes to agitation, restlessness, impaired concentration, and sleep disturbance. Hyperarousal is a key feature of PTSD and reported in a subset of depressed patients (292, 293). Similar sex differences in spatiotemporal expression of CRF_2 and its ligands are found in humans with gut disorders, where they could contribute to differences between males and females in vulnerability to brain-gut disorders (127, 294).

There are sex differences in CRF_1 receptor signaling in the LC that increase female sensitivity to CRF. In the LC, CRF receptors primarily couple to Gs to initiate signaling through the cyclic adenosine monophosphate (cAMP)-protein kinase A (PKA) signaling pathway (295-297). Sex differences in CRF_1-induced cAMP-PKA signaling are linked to greater coupling of the CRF_1 receptor to Gs in females compared to males (298). This sex difference in

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coupling of Gs may indicate that the CRF_1 receptor has a different conformation or binding partner in females vs. males, permitting different proteins to preferentially bind in each sex. Further support for this idea comes from studies demonstrating that, in male rats, acute swim stress increases the binding of a different protein, β-arrestin2, to the CRF_1 receptor, and this effect is not observed in female rats (298). The increased β-arrestin2 in male rats likely contributes to the greater CRF_1 receptor internalization in stressed males (298). When taken together, these findings suggest that CRF_1 receptors preferentially signal through different pathways in males (small GTPases) and females (cAMP-PKA) (299). This difference in signaling could alter physiology and disease risk. In fact, sex differences in CRF_1 receptor signaling in cortex were linked to increased Alzheimer-related pathology, including increased tau phosphorylation and amyloid β signaling in female compared with male mice (300). Few studies investigate sex differences in GPCR signaling, but it is likely that sex differences in GPCRs are also found in receptors other than CRF and that these differences could confer vulnerability and resilience to many diseases.

In human studies, single nucleotide polymorphisms in the CRF receptor gene (*CRHR2*) are associated with negative emotions in patients with irritable bowel syndrome (IBS) (301). Immune cells secrete CRF_2 in extracellular vesicles that circulate in the plasma and associate negatively with disease severity scores in IBS-diarrhea patients (294). Single nucleotide polymorphisms in *CRHR2* are also associated with lifetime PTSD in women (302) and with type 2 diabetes (303). The prevalence of type 2 diabetes and insulin resistance is greater in men (304). Epidemiological studies have shown that men with high levels of self-reported perceived stress have a 1.4 higher odds ratio of developing type 2 diabetes during a 10-year follow-up period and are at 2-fold higher risk of developing diabetes than women with similar levels of reported stress (305). In agreement with human data, male mice lacking functional stress receptors (*Crhr2*$^{-/-}$) and haploinsufficient (*Crhr2*$^{+/}$) mice have worse glucose and insulin tolerance, microvesicular hepatic steatosis, and dyslipidemia than female *Crhr2*$^{-/-}$ or C57BL/6 male and female mice in a high-fat diet–induced model of diabetes (129). Female *Crhr2*$^{-/-}$ mice had significantly greater brown adipose fat mass on high-fat diet than C57BL/6 female or male mice of either genotype, suggesting greater thermogenic responses that might be protective. However, the mouse study did not address whether steroid hormones contributed to changes in adipose mass or function. Thermogenesis in brown adipose tissue in humans in response to a meal or cold stress suggests that women have greater thermogenic responses than men and that these responses correlate positively with progesterone levels, but negatively with cortisol levels (306). Thus, analyzing data from both sexes provides insights into sex-specific mechanisms that regulate physiological processes in both sexes.

In colonic tissues of pediatric patients with Crohn's disease, subcellular localization of CRF_2 differs between boys and girls (127). Furthermore, lack of CRF_2 revealed several sex-specific signaling pathways and differential degree of inflammatory responses in male and female mice (127). Treatment with UCN1, a high-affinity agonist for both CRF receptors, rescued *Crhr2*$^{-/-}$ male mice from colitis-induced mortality, whereas UCN1 treatment increased mortality in *Crhr2*$^{-/-}$ female mice (127). Both diabetes and Crohn's disease show sex differences in disease prevalence and outcomes, yet most animal studies use male sex to delineate mechanisms. Analysis of the data by segregating the 2 sexes can reveal significant insights into distinct and shared mechanisms and factors that exist at baseline and during disease. For example, sex differences exist in the etiology of pancreatitis: alcohol and tobacco predominate in men, whereas idiopathic and obstructive etiologies predominate in women (307), yet to date only a few studies have used both sexes to study mechanisms involved in pancreatitis. While both males and females develop pancreatitis in animal models, when administered identical doses of the pancreatic stressor caerulein, C57BL6 female mice show less severe pancreatitis and histological damage than male mice (128). Lack of CRF_2 rendered female mice more susceptible to caerulein-induced pancreatitis compared with male *Crhr2*$^{-/-}$ mice (128), with both male and female *Crhr2*$^{-/-}$ mice exhibiting similar levels of total histological damage (128). Detailed analysis of components contributing to histopathological damage showed that female C57BL6J mice have less necrosis, zymogen granules, and vacuolization than male mice with pancreatitis, but they have similar levels of edema and neutrophil infiltration as male mice (128). This data segregation allowed isolation of factors that differentially contribute to histological damage, which otherwise would be lost, if grouped together in this analysis. Taken together, these data support a role for the CRF receptors, product of an autosomal gene and regulated by steroid hormones to bring about sex-specific cellular signaling and function.

Sex Differences in Pharmacotherapy of Stress-Based Diseases

Sex differences in GPCR signaling are also relevant for pharmacology. Biased ligands can shift signaling toward

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β-arrestin pathways and away from G-protein-mediated pathways based on how they bind to the GPCR (308). These biased ligands are being designed with the hope of providing more targeted therapies with fewer side effects (308, 309). Understanding sex differences in signaling and how such differences contribute to changes in physiology can inform the development of these biased ligands. For example, a CRF_1 receptor ligand that biases signaling through β-arrestin pathways may be useful for treating hyperarousal symptoms or reducing the progression of Alzheimer disease, especially in women. An idea for such a compound would never have come about if women were excluded from preclinical and clinical studies on CRF_1 receptor function.

The idea of using CRF_1 antagonists to treat depression, PTSD, and irritable bowel syndrome has been around for decades, but these compounds were ineffective in several clinical trials (222, 310). Sex differences in CRF_1 and CRF_2 receptor signaling may also explain the failure of different selective CRF_1 antagonists as treatments for these disorders. While there are likely many reasons for their failure, critical ones could be sex differences in their target, association of CRF receptors with different binding partners in female versus male cells, or heteromerization of CRF receptors (311-313), all of which can result in altered signaling. The consistent efficacy of CRF_1 antagonists in reducing anxiety-like and depressive-like behavior in rodents and nonhuman primates was established in studies primarily conducted in male animals (222, 314-317). In a study in which females were included, local blockade of CRF_1 receptors in the dorsal raphe with an antagonist reduced anxiety in male but not female mice, highlighting sex differences in efficacy (318). Yet these compounds developed primarily in male rodents were tested in clinical trials with participants of both sexes or only in women. Notably the only CRF_1 antagonist study that had success in reducing depressive symptoms, NBI-34041, was conducted only in men (222, 319). The approach of developing compounds in male animal models is not unique to CRF_1 antagonists and has been common practice (222). Collectively, these studies suggest that a failure of certain therapeutics may result from ignoring sex differences in their targets. Sex differences in targets are not well known because most preclinical studies use only male rodents (320, 321). Excluding females in the drug development stage particularly impacts women's health. Indeed, it is likely that some compounds deemed ineffective in male rodents would work in females, yet such compounds never would have a chance to make it to market, because of testing exclusively in male subjects. Moreover, the fact that most drugs are designed using males also likely contributes to the higher rates of adverse drug reactions in women compared to men (322).

Including both sexes in mechanistic studies is critical for developing drugs that work efficaciously in both sexes (see Box 4). Latent sex differences can also impact drug development: a compound targeting a mechanism in men may not work in women. As the field moves forward, we may find that sex-specific therapeutics based on understanding latent sex differences are required to truly improve patient outcomes. In sum, there are observable sex differences in behavior that extend beyond reproductive function. Molecular sex differences in several organs, such as the gut and the central nervous system, play a key role in driving these functional and behavioral differences. Moreover, even when function and behavior are consistent between the sexes, the underlying processes can differ. Thus, including both sexes in preclinical molecular studies guiding drug development is key for improving the health of men and women.

Section IV

Sex Differences in the Cardiovascular-Renal System

Cardiovascular disease (CVD) is the major cause of premature death in both sexes worldwide, although women generally develop CVD 10 years later than men (328). In 2016, ~18 million people died from CVD, representing ~30% of all deaths worldwide (329). There are marked sex differences in CVD and renal disease. For example, women are protected from heart disease during the reproductive years but are more likely to die in the first year following a cardiovascular event than males (330). Most heart conditions, including myocardial infarction, Takotsubo syndrome, and cardiac arrythmia, exhibit sex differences in symptoms and severity (331). Chronic kidney disease (CKD) is more prevalent in women but, once established, progresses more rapidly in men (332). However, this female advantage is lost after menopause. These sex differences in cardiovascular and renal disease have long been overlooked and underappreciated. The clinical presentation, the response to pharmacotherapies, standard care practices, and the underlying pathophysiological mechanisms differ in women compared to men. Furthermore, lack of understanding of sex differences in mechanisms underpinning cardiovascular and renal disease has led to poorer outcomes in women than in men. A major problem is that mechanistic preclinical studies in animal models have largely been conducted in males (333). Yet, it has become increasingly clear that sex differences

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Box 4. Sex differences in pharmacokinetics and pharmacodynamics of drugs

Thalidomide, a sedative that was prescribed to many pregnant women to relieve pregnancy-associated nausea, was first sold in Germany (without a prescription) in 1957; it had been tested in animals and in men, but not in women. It was soon noted to cause multiple birth defects, most notably phocomelia (arrested limb development) and postnatal deaths. Fortunately, it was never approved in the United States, but thousands of children were affected around the world. In 1962, the US Congress passed the Kefauver-Harris Drug Amendments Act requiring manufacturers to prove a drug is both safe and effective (323). Consequently, the US Food and Drug Administration (FDA) recommended against drug testing on women, particularly those of child-bearing age, until the early 1990s. To date, most treatment guidelines are based on results from clinical trials conducted on middle-aged men. Dosage, pharmacokinetics, and pharmacodynamics data for women (and children) are lacking for most drugs. Activities of cytochrome P450 (CYP) enzymes show significant sex differences in drug metabolism in Phase I clinical trials (324). Gastric enzymes involved in oxidative degradation such as alcohol and aldehyde dehydrogenases are significantly more active in men than in women resulting in higher bioavailability of ethanol in women versus men. In Phase II trials, glucuronidating enzymes and some efflux transporters have been shown to be more active in men then in women. Together with estrogens and androgen that alter transmembrane transporters, these processes contribute to efficacy of metabolism in both Phase I and II. Drugs used for treatment of cardiovascular disease, such as angiotensin-converting enzyme inhibitors (ACE inhibitors), angiotensin II receptor blockers, diuretics, the aldosterone blocker eplerenone, antiplatelet agents, and oral antithrombotic medications, all show sex differences in efficacy and safety (325, 326). Over-the-counter nonsteroidal anti-inflammatory drugs (NSAIDs) such as ibuprofen and naproxen are more effective in men than women; there is more liver toxicity with acetaminophen use in women, whereas opioids and benzodiazepine work better in women. While some sex differences in metabolic clearance for statins and beta-blockers are known for these frequently prescribed drugs, dosing and adverse event monitoring in routine clinical practice is inadequate. Alosetron, a serotonin receptor 3 antagonist, is approved for treatment of severe irritable bowel syndrome–diarrhea symptoms in women, as it is largely ineffective in men (327). These findings emphasize that women and men take divergent routes (molecular mechanisms and signaling pathways) to reach the same destination (normal function or diseased state), with paths often intersecting. In the era of personalized medicine, there is no one-size-fits-all therapy, and considering sex-specific outcomes in pharmacokinetics and pharmacodynamics of drugs as well as clinical guidelines is warranted to ensure efficacy and safety of medications.

are apparent in all endocrine systems, which are modified by sex chromosomes and sex hormones, with temporal actions across the lifespan.

Blood Pressure Links Cardiovascular and Renal Diseases

Cardiovascular and renal diseases are linked by the relationship of each to arterial pressure (Fig. 5). The cardiovascular system determines arterial pressure, with the heart generating cardiac output and the blood vessels determining total peripheral resistance. The kidneys contribute by regulating extracellular and intravascular fluid volume, and hence blood volume, and venous return. It is established that CVD leads to chronic kidney disease (CKD) and that CKD leads to the development of CVD. For example, following a myocardial infarct, cardiac output declines and arterial pressure falls causing the kidney to vasoconstrict and retain extracellular fluid, with the effect to increase venous return and normalize cardiac output. However, this has the unwanted effect of placing further stress on the failing heart. Conversely, kidney failure causes fluid retention and hypertension (334). Thus, cardiovascular and kidney function are intertwined, as are the endocrine systems that regulate organ function; including the renin-angiotensin-aldosterone system

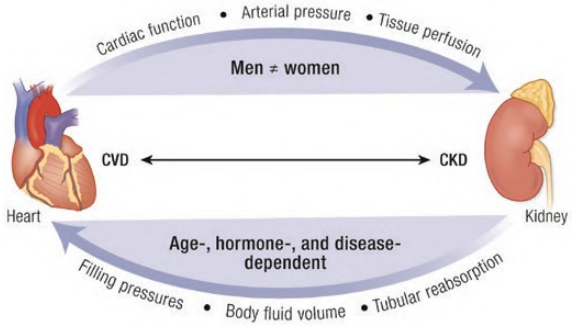

Figure 5. Heart and kidney functions are linked. Sex differences exist in many aspects of heart and kidney function at baseline and in CVD and CKD, as shown. Both organs feed-forward and influence each other's function. Genes, hormones, and age are some known factors that modulate this relationship in a sex-specific manner. Abbreviations: CKD, chronic kidney disease; CVD, cardiovascular disease.

(RAAS), the endothelin system, atrial natriuretic peptides, vasopressin, and glucocorticoid and mineralocorticoid hormones. There is an increasing recognition that there are fundamental sex differences in each of these systems. For example, aldosterone contributes to obesity-induced CVD with a greater impact in females than males (335). However, further research is required to fully elucidate the sex differences present in each endocrine system and how these impact disease development and progression.

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Sex Differences in Arterial Pressure and Hypertension

Hypertension is a major risk factor for cardiovascular and renal disease. Over the lifespan there are age- and sex-related differences in arterial pressure. The majority of the data are derived from cross-sectional studies, but a few powerful studies have tracked arterial pressure over decades within a population (332, 336-339). Arterial pressure increases in both men and women with age, although the slope of the relationship is different between men and women. Sex differences in arterial pressure emerge during adolescence and are maintained throughout adulthood until women reach menopause (336, 337, 339). Arterial pressure is ~5 to 10 mmHg greater in men than age-matched women during the reproductive years (340-342). Postmenopause arterial pressure rises steeply in women regardless of race, ethnicity, or country of origin (340-342). One of the most striking characteristics of hypertension is that the prevalence and severity is lower in premenopausal women than in age-matched men. The prevalence of hypertension is ~10% in young premenopausal women, ~50% in postmenopausal women and by the age of 75 years almost ~80% of women are hypertensive (342-344).

Nonhuman mammalian species also display sex differences in arterial pressure. Arterial pressure in adult females is lower in normotensive dogs, sheep, rabbits, rats, and mice as compared with adult males (338, 345). Furthermore, in rodents, rabbits, and sheep, females of reproductive age are protected against the development of hypertension, such that arterial pressure increases significantly less in females than in males, in settings of disease (338). Thus, sex differences are present in the pathophysiology of cardiovascular and renal diseases. Yet, the mechanisms underlying the sexual dimorphism of arterial pressure in men and women as they age are poorly understood. However, extensive evidence indicates that sex hormones likely contribute to the regulation of arterial pressure through their actions on endocrine systems.

Sex Differences in Endocrine Control of Arterial Pressure and Kidney Function

There are subtle differences in most endocrine actions between men and women. It is not the maximal response of each system but rather the slope of the response that is altered. In this manner, a system responds maximally in a hemodynamic crisis (eg, hemorrhage) but in a sex-specific manner to lesser challenges. For example, a greater dose of the vasoconstrictor angiotensin II is required to increase arterial pressure in female than male mice (346). Consistent with this finding, the same dose of angiotensin II caused a greater reduction in renal blood flow in men than women, with the suggestion that this was an angiotensin type 2 receptor (AT_2R) mediated effect (347). In rodents, females of reproductive age have a greater AT_2R to angiotensin type 1 receptor (AT_1R) ratio than males, which contributes to the reduced pressor response to angiotensin II (348). This has been indirectly demonstrated in women, in studies examining forearm vascular resistance responses to AT_2R blockade (349). The AT_2R also mediates a leftward shift in the pressure natriuresis-diuresis relationship, an effect that is greater in female than male mice (350). In women, indirect evidence also indicates a more pronounced role for the AT_2R in the regulation of renal blood flow responses to angiotensin II (347). This is linked to differential expression of components of the RAAS in males and females, which have been demonstrated in most mammalian species, including humans (351). In the context of the above example, estrogen interacts with the glucocorticoid response element on the X-linked *AGTR2* gene, to increase AT_2R expression in females (352). In addition, there are sex differences in human aminopeptidase A, aminopeptidase N, and angiotensin-converting enzyme 2 levels, responsible for generation of the angiotensin peptide fragments, angiotensin III, and angiotensin-(1–7), which have a high affinity for the vasodilatory AT_2R and Mas receptors, respectively (353-356). Lastly, there are marked and important sex differences in the production and function of aldosterone, although this has only recently been started to be examined (335). Thus, in females the RAAS is balanced toward the protective depressor RAAS arm, which at the lower physiological range may prevent arterial pressure increasing to the same extent as in males. However, this delicate balance may be lost in women after menopause and in the situation of metabolic syndrome.

Other vasoconstrictor systems also have sexually dimorphic actions. Endothelin-1 causes vasoconstriction via the endothelin type A receptor (ET_AR), and vasodilation and sodium excretion via the ET_BR. Testosterone increases ET_AR and estrogen increases ET_BR expression, which contributes to the differential control of arterial blood pressure and renal function between the sexes (357). Vasopressin, with important roles in circulatory and water homeostasis, is affected by age and sex. Urinary concentrating ability declines with age, but more steeply in women. Young men produce more concentrated urine than women, in part due higher plasma arginine vasopressin levels and greater vasopressin type 2 receptor expression in the collecting ducts of the kidney in males (358, 359). Renal vasopressin type 2 receptor expression declines with age in association with a reduction in maximal urine concentrating ability (358, 359). Interestingly, aldosterone signaling via mineralocorticoid receptors is associated with increased CVD risk and is

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enhanced in obese women (another example of how the RAAS is differentially modulated in females), which has been linked to leptin-induced endothelial dysfunction (360, 361). Moreover, evidence in rodents indicates that sodium reabsorption along the length of the renal tubule is sexually dimorphic, with reabsorption shifted to the later segments in females compared to males. This was associated with greater sodium epithelial channel expression, under the control of aldosterone, in the collecting duct, which could also contribute to the increased cardiovascular and renal risk associated with aldosterone in females (362). Finally, oxytocin, relaxin, and prolactin, which are traditionally known for their roles in pregnancy, have differential cardiovascular and renal actions in nonpregnant female and male rodents (348, 363, 364). Thus, evidence points to sex differences in endocrine control of extracellular fluid homeostasis and vascular function, which likely contribute to age- and sex-related disparities in renal and cardiovascular disease risk. Further studies are warranted to understand this complex issue more fully. In particular, it is important to take into account the subtle effects within the physiological range that counterbalance function of each hormonal system, rather than examine the impact of pharmacological doses which can mask sex differences in responses.

Cardioprotective Mechanisms in Women Sustain a Healthy Pregnancy

The cardioprotective mechanisms that predominate in women during the reproductive years enable the extensive hemodynamic adaptations required to meet the metabolic demands of the developing fetus and a successful pregnancy. During a normotensive pregnancy, blood volume increases and cardiac output increase by ~30% to 50%, but arterial pressure declines due to marked peripheral vasodilatation (365, 366). The associated renal vasodilation accommodates an increase in glomerular filtration rate to process the additional blood volume, but an increase in vasopressin type 2 receptor expression enables increased tubule reabsorption of sodium and water. However, in women with preeclampsia, a pregnancy-induced form of hypertension, these cardiovascular adaptations are perturbed. Accumulating evidence now indicates that women with a history of pregnancy-associated hypertension have a 2- to 5-fold increased risk of CVD in later life (367). Understanding the mechanisms underpinning this dysregulation of vascular function in pregnancy-related hypertension may lead to the identification of new therapeutic targets for the treatment of cardiovascular disease in both sexes. For example, relaxin, which is known best for its role in pregnancy but is also produced in males, plays roles in the regulation of renal function, blood pressure, and tissue fibrosis (363). Thus, it is a mistake to assign hormonal systems a specific role as most have wide-ranging tissue-specific pleiotropic effects.

Sex Hormones and Sex Chromosome Complement in CVD

Sex hormones contribute to sexual dimorphism in endocrine control of the cardiovascular system, with evidence suggesting that there is a “sweet spot” for both testosterone and estradiol, as unusually high or low levels of either promote disease (368-370). This has been the cause of apparent discrepancies in the literature. In particular, this remains a problem in animal studies in which the dose of estrogen used to study the impact of estrogen replacement in aged or gonadectomized models varies widely (~1000-fold), as does the route or length of administration; none of which accurately reflect the cyclic pattern of in vivo production. This lack of rigor into investigation of the effects of sex hormones in preclinical models likely contributes to the controversy that surrounds hormone replacement therapy for the prevention of CVD risk. Despite extensive evidence that hormone replacement therapy is cardioprotective, the negative results of the Women’s Health Initiative Trial effectively halted the use of hormone replacement therapy (371). Certainly, high-dose estrogen can increase blood pressure and cardiovascular risk in women (372). However, continued investigation supports the use of hormone replacement therapy in subsets of women, and further work in this area is required (373). In contrast, in men with low testosterone, beneficial cardiovascular effects are seen with testosterone replacement (374). In women with polycystic ovary syndrome, high testosterone levels are associated with elevated blood pressure (374). Dose-ranging studies are required to delineate these effects.

The sex chromosomes may have a direct impact on sex differences in the physiology and pathophysiology of the cardiovascular system and cardiovascular risk, independent of sex hormones. Human sex chromosome aneuploidies, such as Turner and Klinefelter syndromes, suggest that sex chromosome abnormalities can carry an increased risk of CVD. Women with Turner syndrome have around a 3-fold greater mortality and reduced life expectancy relative to the general population (375-377). CVD is a leading cause of increased mortality in Turner syndrome (375-377). Congenital cardiac anomalies, hypertension, coarctation of the aorta, diabetes, ischemic heart disease, and stroke are commonly associated with this condition (378). Similarly, men with Klinefelter syndrome have a high cardiovascular risk profile (379, 380), and an increased risk of

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mortality from cardiovascular disease (381, 382). However, observations from studies in individuals with sex chromosome aneuploidies are complicated by confounding factors, including abnormal gonadal sex hormone levels associated with gonadal failure. Thus, it is very difficult to distinguish between hormonal versus genetic mechanisms and cardiovascular risk in these human conditions.

Experimental approaches, such as the FCG mouse model discussed in "Section I," and Box 3 can discriminate between hormonal and sex chromosome effects in cardiovascular disease (115). Beyond genes on the sex chromosomes, there are sex differences in autosomal gene expression, which can be both organ or cell specific (383). In the kidney and the heart, hundreds of rat and human genes are regulated differently between the sexes (384-386). This disparate expression is triggered by sex hormones in ~30% of cases, with the other 70% linked to sex chromosome and microRNA dimorphisms (384, 385). For example, sex differences have been reported in the expression of nitric oxide synthase, tyrosine hydroxylase, and sodium channels in the rodent heart and kidney (332). However, few studies to date have compared gene expression and the effect on the proteome between the human sexes, and further studies are required.

Sex Differences in Pharmacotherapy for Cardiovascular and Renal Disease

Men and women respond to disease differently: kidney diseases progress faster in men than women, kidney transplants from women to men tend to fail more frequently than the reverse, and the effects of diabetes on the kidney differ between the sexes (387-392). Furthermore, symptoms and mechanisms of heart failure differ between the sexes (393). This suggests that sex-specific treatments for CKD and CVD could be required. There is currently little evidence to suggest that men and women respond differently to current treatments for hypertension (394). In large part, this is because clinical trials have lacked statistical power to take this into account. It will be difficult to achieve such an outcome for drugs that have already received FDA approval. However, some treatments are more frequently prescribed, without any basis in evidence (395). There are also marked differences in pharmacokinetics and pharmacodynamics (see Box 4), leading to more frequent adverse drug reactions in women, related to differences in drug clearance and breakdown (396). Therefore, sex should be taken in account for new treatments seeking approval in the future. When women are considered, important and unexpected sex differences are observed in almost every aspect of cardiovascular and renal function in health and disease. Further research is required to fully understand these differences, and in turn to guide the development of sex-specific treatment guidelines for CVD and CKD.

Section V

Challenges for the Future of Sex Differences Research—Areas Requiring Special Attention

Sex differences exist in anatomy, behavior, and physiology across the animal taxa. By extension, because of these innate differences, sex differences exist at molecular and cellular levels in mechanisms that underlie these processes. Despite concerted efforts by the Office of Research on Women's Health and the Organization for the Study of Sex Differences in educating researchers about the distinction between sex versus gender, the indiscriminate use of the word "gender" continues to pervade scientific literature. The sex of established cultured cell lines is another issue; in addition to aneuploidy, chromosomal numbers change as cells are passaged and are dependent upon the tissue of origin (397, 398), but this aspect is beyond the scope of this Statement. Not surprisingly, sex differences are seen in etiology, prevalence, and outcomes in a myriad of human diseases that range from psychological and autoimmune to gastrointestinal, cardiovascular, renal, and reproductive; SARS-CoV-2 causes more severe COVID-19 disease in men than in women despite similar infection rates (399-401). Besides genetic makeup (predisposition), extraneous factors, such as the socioeconomics, demographics, education level, profession, age, and the environment, greatly influence an individual's health; COVID-19 disease outcomes especially highlight the contribution of these extraneous factors in health disparities. Factors such as the endocrine-disruptive chemicals can disproportionately affect one sex over the other; regardless, whether favorable or adverse effects are present in one or both sexes, the effects would impact trans and cisgender persons, and hence these sex-specific effects should not be overlooked or underestimated (402). Some human studies addressing sex differences take these factors into account, whereas others are more selective. Many studies of disease pathways are sensitive to levels of gonadal steroid hormones, which contribute to sex differences. In human studies, unless gender information is explicitly collected or available, the study deals with biological sex, not gender. Use of sex and gender interchangeably deemphasizes the importance of studying gender as an independent variable.

In animals or experimental models of human diseases, effects of estrogens have been investigated more often than effects of progestins and androgens, which should

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be corrected. Paradoxically, female sex is often excluded from experimental design on the basis that: (i) the estrus cycle will interfere with data interpretation; (ii) mechanisms that operate in the male sex will operate in the female sex and thus only need to be confirmed in females; (iii) metabolic demands are similar between the sexes; (iv) the X chromosome in males and females is subject to similar regulation; and (v) autosomal genes will be subject to equal variance between the sexes. The same studies often ignore the diurnal cycling nature of testosterone in males; testosterone levels in male rodents can show more day-to-day variability than estrogen and progesterone levels in females. Other steroid hormones, such as glucocorticoids, that show circadian rhythm and whose levels differ between the sexes also influence gene expression and function. In rodents but not primates, sex differences in secretion of growth hormone result in sexually dimorphic hepatic metabolism of drugs and xenobiotics (403). In rodents, endocrine disruption can have transgenerational effects on male and female reproductive systems (404). Since changes in hormone levels and gene expression are dynamic, can be localized, and are spatiotemporally distinct, no one study design or condition can be used as a gold standard. Animal housing and handling conditions can also create sex differences, and thus any experimental design and data interpretation should take these variables into account. If sex-segregated data does not differ for the aspects under study, then data can be pooled from the 2 sexes and reported accordingly.

Studies in animal models have just begun to uncover unequal effects of the sex chromosomes in XX vs XY cells, so we expect further discoveries about such effects in the future. Once genes that cause sex differences are discovered in animals, the findings generate new hypotheses and rationalize human studies to determine whether the same gene also creates sex differences in humans. That question can be studied by the methods of human genetics, relating genetic variation to disease incidence and outcome. Without the animal studies, however, it is difficult to understand detailed molecular mechanisms. It is also important to remember that no single rodent or animal model can capture the complexity of any human disease, but each model provides valuable insights into one or another major aspect of disease. If different etiologies of a given disease share mechanisms, then mimicking the precise conditions that initiate human disease may not be critical.

The study of sex chromosome effects is in its infancy and has focused on proving that sex chromosomes play a role and finding the genes responsible for the effects. So far there has been little effort to understand how these factors interact with steroid hormones to cause sex differences. If both types of factors cause differences in disease incidence, are they affecting the same or different downstream pathways? Do their effects converge, or do they independently affect different mechanisms that each influence a complex disease? Do male-biased factors (hormones, Y-chromosome genes) act synergistically to induce a male-specific state, or do they counteract each other to reduce the difference between males and females (123, 405)? Are the diverse sex-biasing factors changing in their effects across the lifespan, leading to changes in the type or amount of sex difference at different ages?

When studying sex differences in animal models of human diseases, it is important to first understand and elucidate differences at baseline in gonadally intact animals. As pointed out earlier, steroidogenic enzymes are also present in nongonadal tissues, especially the brain, thus it is not entirely possible to eliminate effects of sex steroids from all tissues. Moreover, tamoxifen-inducible *Cre* recombinase used to routinely perform lineage tracing and gene inactivation studies in mice has its own problems (406, 407) that are largely ignored and can further confound sex-specific data analysis; tamoxifen antagonizes actions of estrogen receptor-β and inhibits expression of over 70 genes (408), but the contribution of these tamoxifen-regulated genes on study results and outcomes is never accounted for and requires careful consideration. Before mechanisms behind sex differences in physiology and disease can be elucidated, a fundamental understanding of sex differences that exist at baseline, is needed.

Acknowledgments

The authors thank Stephen M. Rosenthal and Robert M. Carey for critically reading the manuscript.

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Disclosures: The authors have nothing to disclose.

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Endocrine Treatment of Gender-Dysphoric/ Gender-Incongruent Persons: An Endocrine Society* Clinical Practice Guideline

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*Cosponsoring Associations: American Association of Clinical Endocrinologists, American Society of Andrology, European Society for Pediatric Endocrinology, European Society of Endocrinology, Pediatric Endocrine Society, and World Professional Association for Transgender Health.

Objective: To update the "Endocrine Treatment of Transsexual Persons: An Endocrine Society Clinical Practice Guideline," published by the Endocrine Society in 2009.

Participants: The participants include an Endocrine Society–appointed task force of nine experts, a methodologist, and a medical writer.

Evidence: This evidence-based guideline was developed using the Grading of Recommendations, Assessment, Development, and Evaluation approach to describe the strength of recommendations and the quality of evidence. The task force commissioned two systematic reviews and used the best available evidence from other published systematic reviews and individual studies.

Consensus Process: Group meetings, conference calls, and e-mail communications enabled consensus. Endocrine Society committees, members and cosponsoring organizations reviewed and commented on preliminary drafts of the guidelines.

Conclusion: Gender affirmation is multidisciplinary treatment in which endocrinologists play an important role. Gender-dysphoric/gender-incongruent persons seek and/or are referred to endocrinologists to develop the physical characteristics of the affirmed gender. They require a safe and effective hormone regimen that will (1) suppress endogenous sex hormone secretion determined by the person's genetic/gonadal sex and (2) maintain sex hormone levels within the normal range for the person's affirmed gender. Hormone treatment is not recommended for prepubertal gender-dysphoric/gender-incongruent persons. Those clinicians who recommend gender-affirming endocrine treatments—appropriately trained diagnosing clinicians (required), a mental health provider for adolescents (required) and mental health

ISSN Print 0021-972X ISSN Online 1945-7197
Printed in USA
Copyright © 2017 Endocrine Society
Received 24 July 2017. Accepted 24 August 2017.
First Published Online 13 September 2017

Abbreviations: BMD, bone mineral density; DSD, disorder/difference of sex development; DSM, Diagnostic and Statistical Manual of Mental Disorders; GD, gender dysphoria; GnRH, gonadotropin-releasing hormone; ICD, International Statistical Classification of Diseases and Related Health Problems; MHP, mental health professional; VTE, venous thromboembolism.

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professional for adults (recommended)—should be knowledgeable about the diagnostic criteria and criteria for gender-affirming treatment, have sufficient training and experience in assessing psychopathology, and be willing to participate in the ongoing care throughout the endocrine transition. We recommend treating gender-dysphoric/gender-incongruent adolescents who have entered puberty at Tanner Stage G2/B2 by suppression with gonadotropin-releasing hormone agonists. Clinicians may add gender-affirming hormones after a multidisciplinary team has confirmed the persistence of gender dysphoria/gender incongruence and sufficient mental capacity to give informed consent to this partially irreversible treatment. Most adolescents have this capacity by age 16 years old. We recognize that there may be compelling reasons to initiate sex hormone treatment prior to age 16 years, although there is minimal published experience treating prior to 13.5 to 14 years of age. For the care of peripubertal youths and older adolescents, we recommend that an expert multidisciplinary team comprised of medical professionals and mental health professionals manage this treatment. The treating physician must confirm the criteria for treatment used by the referring mental health practitioner and collaborate with them in decisions about gender-affirming surgery in older adolescents. For adult gender-dysphoric/gender-incongruent persons, the treating clinicians (collectively) should have expertise in transgender-specific diagnostic criteria, mental health, primary care, hormone treatment, and surgery, as needed by the patient. We suggest maintaining physiologic levels of gender-appropriate hormones and monitoring for known risks and complications. When high doses of sex steroids are required to suppress endogenous sex steroids and/or in advanced age, clinicians may consider surgically removing natal gonads along with reducing sex steroid treatment. Clinicians should monitor both transgender males (female to male) and transgender females (male to female) for reproductive organ cancer risk when surgical removal is incomplete. Additionally, clinicians should persistently monitor adverse effects of sex steroids. For gender-affirming surgeries in adults, the treating physician must collaborate with and confirm the criteria for treatment used by the referring physician. Clinicians should avoid harming individuals (via hormone treatment) who have conditions other than gender dysphoria/gender incongruence and who may not benefit from the physical changes associated with this treatment. ***(J Clin Endocrinol Metab* 102: 3869–3903, 2017)**

Summary of Recommendations

1.0 Evaluation of youth and adults

1.1. We advise that only trained mental health professionals (MHPs) who meet the following criteria should diagnose gender dysphoria (GD)/gender incongruence in adults: (1) competence in using the Diagnostic and Statistical Manual of Mental Disorders (DSM) and/or the International Statistical Classification of Diseases and Related Health Problems (ICD) for diagnostic purposes, (2) the ability to diagnose GD/gender incongruence and make a distinction between GD/gender incongruence and conditions that have similar features (*e.g.*, body dysmorphic disorder), (3) training in diagnosing psychiatric conditions, (4) the ability to undertake or refer for appropriate treatment, (5) the ability to psychosocially assess the person's understanding, mental health, and social conditions that can impact gender-affirming hormone therapy, and (6) a practice of regularly attending relevant professional meetings. (Ungraded Good Practice Statement)

1.2. We advise that only MHPs who meet the following criteria should diagnose GD/gender incongruence in children and adolescents: (1) training in child and adolescent developmental psychology and psychopathology, (2) competence in using the DSM and/or the ICD for diagnostic purposes, (3) the ability to make a distinction between GD/gender incongruence and conditions that have similar features (*e.g.*, body dysmorphic disorder), (4) training in diagnosing psychiatric conditions, (5) the ability to undertake or refer for appropriate treatment, (6) the ability to psychosocially assess the person's understanding and social conditions that can impact gender-affirming hormone therapy, (7) a practice of regularly attending relevant professional meetings, and (8) knowledge of the criteria for puberty blocking and gender-affirming hormone treatment in adolescents. (Ungraded Good Practice Statement)

1.3. We advise that decisions regarding the social transition of prepubertal youths with GD/gender incongruence are made with the assistance of an MHP or another experienced professional. (Ungraded Good Practice Statement).

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doi: 10.1210/jc.2017-01658

1.4. We recommend against puberty blocking and gender-affirming hormone treatment in prepubertal children with GD/gender incongruence. (1 |⊕⊕◯◯)

1.5. We recommend that clinicians inform and counsel all individuals seeking gender-affirming medical treatment regarding options for fertility preservation prior to initiating puberty suppression in adolescents and prior to treating with hormonal therapy of the affirmed gender in both adolescents and adults. (1 |⊕⊕⊕◯)

2.0 Treatment of adolescents

2.1. We suggest that adolescents who meet diagnostic criteria for GD/gender incongruence, fulfill criteria for treatment, and are requesting treatment should initially undergo treatment to suppress pubertal development. (2 |⊕⊕◯◯)

2.2. We suggest that clinicians begin pubertal hormone suppression after girls and boys first exhibit physical changes of puberty. (2 |⊕⊕◯◯)

2.3. We recommend that, where indicated, GnRH analogues are used to suppress pubertal hormones. (1 |⊕⊕◯◯)

2.4. In adolescents who request sex hormone treatment (given this is a partly irreversible treatment), we recommend initiating treatment using a gradually increasing dose schedule after a multidisciplinary team of medical and MHPs has confirmed the persistence of GD/gender incongruence and sufficient mental capacity to give informed consent, which most adolescents have by age 16 years. (1 |⊕⊕◯◯).

2.5. We recognize that there may be compelling reasons to initiate sex hormone treatment prior to the age of 16 years in some adolescents with GD/gender incongruence, even though there are minimal published studies of gender-affirming hormone treatments administered before age 13.5 to 14 years. As with the care of adolescents ≥16 years of age, we recommend that an expert multidisciplinary team of medical and MHPs manage this treatment. (1 |⊕◯◯◯)

2.6. We suggest monitoring clinical pubertal development every 3 to 6 months and laboratory parameters every 6 to 12 months during sex hormone treatment. (2 |⊕⊕◯◯)

3.0 Hormonal therapy for transgender adults

3.1. We recommend that clinicians confirm the diagnostic criteria of GD/gender incongruence and the criteria for the endocrine phase of gender transition before beginning treatment. (1 |⊕⊕⊕◯)

3.2. We recommend that clinicians evaluate and address medical conditions that can be exacerbated by hormone depletion and treatment with sex hormones of the affirmed gender before beginning treatment. (1 |⊕⊕⊕◯)

3.3. We suggest that clinicians measure hormone levels during treatment to ensure that endogenous sex steroids are suppressed and administered sex steroids are maintained in the normal physiologic range for the affirmed gender. (2 |⊕⊕◯◯)

3.4. We suggest that endocrinologists provide education to transgender individuals undergoing treatment about the onset and time course of physical changes induced by sex hormone treatment. (2 |⊕◯◯◯)

4.0 Adverse outcome prevention and long-term care

4.1. We suggest regular clinical evaluation for physical changes and potential adverse changes in response to sex steroid hormones and laboratory monitoring of sex steroid hormone levels every 3 months during the first year of hormone therapy for transgender males and females and then once or twice yearly. (2 |⊕⊕◯◯)

4.2. We suggest periodically monitoring prolactin levels in transgender females treated with estrogens. (2 |⊕⊕◯◯)

4.3. We suggest that clinicians evaluate transgender persons treated with hormones for cardiovascular risk factors using fasting lipid profiles, diabetes screening, and/or other diagnostic tools. (2 |⊕⊕◯◯)

4.4. We recommend that clinicians obtain bone mineral density (BMD) measurements when risk factors for osteoporosis exist, specifically in those who stop sex hormone therapy after gonadectomy. (1 |⊕⊕◯◯)

4.5. We suggest that transgender females with no known increased risk of breast cancer follow breast-screening guidelines recommended for non-transgender females. (2 |⊕⊕◯◯)

4.6. We suggest that transgender females treated with estrogens follow individualized screening according to personal risk for prostatic disease and prostate cancer. (2 |⊕◯◯◯)

4.7. We advise that clinicians determine the medical necessity of including a total hysterectomy and oophorectomy as part of gender-affirming surgery. (Ungraded Good Practice Statement)

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5.0 Surgery for sex reassignment and gender confirmation

5.1. We recommend that a patient pursue genital gender-affirming surgery only after the MHP and the clinician responsible for endocrine transition therapy both agree that surgery is medically necessary and would benefit the patient's overall health and/or well-being. (1 ⊕⊕◯◯)

5.2. We advise that clinicians approve genital gender-affirming surgery only after completion of at least 1 year of consistent and compliant hormone treatment, unless hormone therapy is not desired or medically contraindicated. (Ungraded Good Practice Statement)

5.3. We advise that the clinician responsible for endocrine treatment and the primary care provider ensure appropriate medical clearance of transgender individuals for genital gender-affirming surgery and collaborate with the surgeon regarding hormone use during and after surgery. (Ungraded Good Practice Statement)

5.4. We recommend that clinicians refer hormone-treated transgender individuals for genital surgery when: (1) the individual has had a satisfactory social role change, (2) the individual is satisfied about the hormonal effects, and (3) the individual desires definitive surgical changes. (1 ⊕◯◯◯)

5.5. We suggest that clinicians delay gender-affirming genital surgery involving gonadectomy and/or hysterectomy until the patient is at least 18 years old or legal age of majority in his or her country. (2 ⊕⊕◯◯).

5.6. We suggest that clinicians determine the timing of breast surgery for transgender males based upon the physical and mental health status of the individual. There is insufficient evidence to recommend a specific age requirement. (2 ⊕◯◯◯)

Changes Since the Previous Guideline

Both the current guideline and the one published in 2009 contain similar sections. Listed here are the sections contained in the current guideline and the corresponding number of recommendations: Introduction, Evaluation of Youth and Adults (5), Treatment of Adolescents (6), Hormonal Therapy for Transgender Adults (4), Adverse Outcomes Prevention and Long-term Care (7), and Surgery for Sex Reassignment and Gender Confirmation (6). The current introduction updates the diagnostic classification of "gender dysphoria/gender incongruence." It also reviews the development of "gender identity" and summarizes its natural development. The section on clinical evaluation of both youth and adults, defines in detail the professional qualifications required of those who diagnose and treat both adolescents and adults. We advise that decisions regarding the social transition of prepubertal youth are made with the assistance of a mental health professional or similarly experienced professional. We recommend against puberty blocking followed by gender-affirming hormone treatment of prepubertal children. Clinicians should inform pubertal children, adolescents, and adults seeking gender-confirming treatment of their options for fertility preservation. Prior to treatment, clinicians should evaluate the presence of medical conditions that may be worsened by hormone depletion and/or treatment. A multidisciplinary team, preferably composed of medical and mental health professionals, should monitor treatments. Clinicians evaluating transgender adults for endocrine treatment should confirm the diagnosis of persistent gender dysphoria/gender incongruence. Physicians should educate transgender persons regarding the time course of steroid-induced physical changes. Treatment should include periodic monitoring of hormone levels and metabolic parameters, as well as assessments of bone density and the impact upon prostate, gonads, and uterus. We also make recommendations for transgender persons who plan genital gender-affirming surgery.

Method of Development of Evidence-Based Clinical Practice Guidelines

The Clinical Guidelines Subcommittee (CGS) of the Endocrine Society deemed the diagnosis and treatment of individuals with GD/gender incongruence a priority area for revision and appointed a task force to formulate evidence-based recommendations. The task force followed the approach recommended by the Grading of Recommendations, Assessment, Development, and Evaluation group, an international group with expertise in the development and implementation of evidence-based guidelines (1). A detailed description of the grading scheme has been published elsewhere (2). The task force used the best available research evidence to develop the recommendations. The task force also used consistent language and graphical descriptions of both the strength of a recommendation and the quality of evidence. In terms of the strength of the recommendation, strong recommendations use the phrase "we recommend" and the number 1, and weak recommendations use the phrase "we suggest" and the number 2. Cross-filled circles indicate the quality of the evidence, such that ⊕◯◯◯ denotes very low–quality evidence; ⊕⊕◯◯, low quality; ⊕⊕⊕◯, moderate quality; and ⊕⊕⊕⊕, high quality. The task force has confidence that persons who receive care according to the strong recommendations will derive, on average, more benefit than harm. Weak recommendations require more careful consideration of the person's circumstances, values, and preferences to determine the best course of action. Linked to each recommendation is a description of the evidence and the

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doi: 10.1210/jc.2017-01658

values that the task force considered in making the recommendation. In some instances, there are remarks in which the task force offers technical suggestions for testing conditions, dosing, and monitoring. These technical comments reflect the best available evidence applied to a typical person being treated. Often this evidence comes from the unsystematic observations of the task force and their preferences; therefore, one should consider these remarks as suggestions.

In this guideline, the task force made several statements to emphasize the importance of shared decision-making, general preventive care measures, and basic principles of the treatment of transgender persons. They labeled these "Ungraded Good Practice Statement." Direct evidence for these statements was either unavailable or not systematically appraised and considered out of the scope of this guideline. The intention of these statements is to draw attention to these principles.

The Endocrine Society maintains a rigorous conflict-of-interest review process for developing clinical practice guidelines. All task force members must declare any potential conflicts of interest by completing a conflict-of-interest form. The CGS reviews all conflicts of interest before the Society's Council approves the members to participate on the task force and periodically during the development of the guideline. All others participating in the guideline's development must also disclose any conflicts of interest in the matter under study, and most of these participants must be without any conflicts of interest. The CGS and the task force have reviewed all disclosures for this guideline and resolved or managed all identified conflicts of interest.

Conflicts of interest are defined as remuneration in any amount from commercial interests; grants; research support; consulting fees; salary; ownership interests [*e.g.*, stocks and stock options (excluding diversified mutual funds)]; honoraria and other payments for participation in speakers' bureaus, advisory boards, or boards of directors; and all other financial benefits. Completed forms are available through the Endocrine Society office.

The Endocrine Society provided the funding for this guideline; the task force received no funding or remuneration from commercial or other entities.

Commissioned Systematic Review

The task force commissioned two systematic reviews to support this guideline. The first one aimed to summarize the available evidence on the effect of sex steroid use in transgender individuals on lipids and cardiovascular outcomes. The review identified 29 eligible studies at moderate risk of bias. In transgender males (female to male), sex steroid therapy was associated with a statistically significant increase in serum triglycerides and low-density lipoprotein cholesterol levels. High-density lipoprotein cholesterol levels decreased significantly across all follow-up time periods. In transgender females (male to female), serum triglycerides were significantly higher without any changes in other parameters. Few myocardial infarction, stroke, venous thromboembolism (VTE), and death events were reported. These events were more frequent in transgender females. However, the quality of the evidence was low. The second review summarized the available evidence regarding the effect of sex steroids on bone health in transgender individuals and identified 13 studies. In transgender males, there was no statistically significant difference in the lumbar spine, femoral neck, or total hip BMD at 12 and 24 months compared with baseline values before initiating masculinizing hormone therapy. In transgender females, there was a statistically significant increase in lumbar spine BMD at 12 months and 24 months compared with baseline values before initiation of feminizing hormone therapy. There was minimal information on fracture rates. The quality of evidence was also low.

Introduction

Throughout recorded history (in the absence of an endocrine disorder) some men and women have experienced confusion and anguish resulting from rigid, forced conformity to sexual dimorphism. In modern history, there have been numerous ongoing biological, psychological, cultural, political, and sociological debates over various aspects of gender variance. The 20th century marked the emergence of a social awakening for men and women with the belief that they are "trapped" in the wrong body (3). Magnus Hirschfeld and Harry Benjamin, among others, pioneered the medical responses to those who sought relief from and a resolution to their profound discomfort. Although the term transsexual became widely known after Benjamin wrote "The Transsexual Phenomenon" (4), it was Hirschfeld who coined the term "transsexual" in 1923 to describe people who want to live a life that corresponds with their experienced gender vs their designated gender (5). Magnus Hirschfeld (6) and others (4, 7) have described other types of trans phenomena besides transsexualism. These early researchers proposed that the gender identity of these people was located somewhere along a unidimensional continuum. This continuum ranged from all male through "something in between" to all female. Yet such a classification does not take into account that people may have gender identities outside this continuum. For instance, some experience themselves as having both a male and female gender identity, whereas others completely renounce any gender classification (8, 9). There are also reports of individuals experiencing a continuous and rapid involuntary alternation between a male and female identity (10) or men who do not experience themselves as men but do not want to live as women (11, 12). In some countries, (*e.g.*, Nepal, Bangladesh, and Australia), these nonmale or nonfemale genders are officially recognized (13). Specific treatment protocols, however, have not yet been developed for these groups.

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Instead of the term transsexualism, the current classification system of the American Psychiatric Association uses the term gender dysphoria in its diagnosis of persons who are not satisfied with their designated gender (14). The current version of the World Health Organization's ICD-10 still uses the term transsexualism when diagnosing adolescents and adults. However, for the ICD-11, the World Health Organization has proposed using the term "gender incongruence" (15).

Treating persons with GD/gender incongruence (15) was previously limited to relatively ineffective elixirs or creams. However, more effective endocrinology-based treatments became possible with the availability of testosterone in 1935 and diethylstilbestrol in 1938. Reports of individuals with GD/gender incongruence who were treated with hormones and gender-affirming surgery appeared in the press during the second half of the 20th century. The Harry Benjamin International Gender Dysphoria Association was founded in September 1979 and is now called the World Professional Association for Transgender Health (WPATH). WPATH published its first Standards of Care in 1979. These standards have since been regularly updated, providing guidance for treating persons with GD/gender incongruence (16).

Prior to 1975, few peer-reviewed articles were published concerning endocrine treatment of transgender persons. Since then, more than two thousand articles about various aspects of transgender care have appeared.

It is the purpose of this guideline to make detailed recommendations and suggestions, based on existing medical literature and clinical experience, that will enable treating physicians to maximize benefit and minimize risk when caring for individuals diagnosed with GD/gender incongruence.

In the future, we need more rigorous evaluations of the effectiveness and safety of endocrine and surgical protocols. Specifically, endocrine treatment protocols for GD/gender incongruence should include the careful assessment of the following: (1) the effects of prolonged delay of puberty in adolescents on bone health, gonadal function, and the brain (including effects on cognitive, emotional, social, and sexual development); (2) the effects of treatment in adults on sex hormone levels; (3) the requirement for and the effects of progestins and other agents used to suppress endogenous sex steroids during treatment; and (4) the risks and benefits of gender-affirming hormone treatment in older transgender people.

To successfully establish and enact these protocols, a commitment of mental health and endocrine investigators is required to collaborate in long-term, large-scale studies across countries that use the same diagnostic and inclusion criteria, medications, assay methods, and response assessment tools (*e.g.*, the European Network for the Investigation of Gender Incongruence) (17, 18).

Terminology and its use vary and continue to evolve. Table 1 contains the definitions of terms as they are used throughout this guideline.

Biological Determinants of Gender Identity Development

One's self-awareness as male or female changes gradually during infant life and childhood. This process of cognitive and affective learning evolves with interactions with parents, peers, and environment. A fairly accurate timetable exists outlining the steps in this process (19). Normative psychological literature, however, does not address if and when gender identity becomes crystallized and what factors contribute to the development of a gender identity that is not congruent with the gender of rearing. Results of studies from a variety of biomedical disciplines—genetic, endocrine, and neuroanatomic—support the concept that gender identity and/or gender expression (20) likely reflect a complex interplay of biological, environmental, and cultural factors (21, 22).

With respect to endocrine considerations, studies have failed to find differences in circulating levels of sex steroids between transgender and nontransgender individuals (23). However, studies in individuals with a disorder/difference of sex development (DSD) have informed our understanding of the role that hormones may play in gender identity outcome, even though most persons with GD/gender incongruence do not have a DSD. For example, although most 46,XX adult individuals with virilizing congenital adrenal hyperplasia caused by mutations in *CYP21A2* reported a female gender identity, the prevalence of GD/gender incongruence was much greater in this group than in the general population without a DSD. This supports the concept that there is a role for prenatal/postnatal androgens in gender development (24–26), although some studies indicate that prenatal androgens are more likely to affect gender behavior and sexual orientation rather than gender identity *per se* (27, 28).

Researchers have made similar observations regarding the potential role of androgens in the development of gender identity in other individuals with DSD. For example, a review of two groups of 46,XY persons, each with androgen synthesis deficiencies and female raised, reported transgender male (female-to-male) gender role changes in 56% to 63% and 39% to 64% of patients, respectively (29). Also, in 46,XY female-raised individuals with cloacal

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doi: 10.1210/jc.2017-01658

Table 1. Definitions of Terms Used in This Guideline

Biological sex, biological male or female: These terms refer to physical aspects of maleness and femaleness. As these may not be in line with each other (*e.g.*, a person with XY chromosomes may have female-appearing genitalia), the terms biological sex and biological male or female are imprecise and should be avoided.
Cisgender: This means not transgender. An alternative way to describe individuals who are not transgender is "non-transgender people."
Gender-affirming (hormone) treatment: See "gender reassignment"
Gender dysphoria: This is the distress and unease experienced if gender identity and designated gender are not completely congruent (see Table 2). In 2013, the American Psychiatric Association released the fifth edition of the DSM-5, which replaced "gender identity disorder" with "gender dysphoria" and changed the criteria for diagnosis.
Gender expression: This refers to external manifestations of gender, expressed through one's name, pronouns, clothing, haircut, behavior, voice, or body characteristics. Typically, transgender people seek to make their gender expression align with their gender identity, rather than their designated gender.
Gender identity/experienced gender: This refers to one's internal, deeply held sense of gender. For transgender people, their gender identity does not match their sex designated at birth. Most people have a gender identity of man or woman (or boy or girl). For some people, their gender identity does not fit neatly into one of those two choices. Unlike gender expression (see below), gender identity is not visible to others.
Gender identity disorder: This is the term used for GD/gender incongruence in previous versions of DSM (see "gender dysphoria"). The ICD-10 still uses the term for diagnosing child diagnoses, but the upcoming ICD-11 has proposed using "gender incongruence of childhood."
Gender incongruence: This is an umbrella term used when the gender identity and/or gender expression differs from what is typically associated with the designated gender. Gender incongruence is also the proposed name of the gender identity–related diagnoses in ICD-11. Not all individuals with gender incongruence have gender dysphoria or seek treatment.
Gender variance: See "gender incongruence"
Gender reassignment: This refers to the treatment procedure for those who want to adapt their bodies to the experienced gender by means of hormones and/or surgery. This is also called gender-confirming or gender-affirming treatment.
Gender-reassignment surgery (gender-confirming/gender-affirming surgery): These terms refer only to the surgical part of gender-confirming/gender-affirming treatment.
Gender role: This refers to behaviors, attitudes, and personality traits that a society (in a given culture and historical period) designates as masculine or feminine and/or that society associates with or considers typical of the social role of men or women.
Sex designated at birth: This refers to sex assigned at birth, usually based on genital anatomy.
Sex: This refers to attributes that characterize biological maleness or femaleness. The best known attributes include the sex-determining genes, the sex chromosomes, the H-Y antigen, the gonads, sex hormones, internal and external genitalia, and secondary sex characteristics.
Sexual orientation: This term describes an individual's enduring physical and emotional attraction to another person. Gender identity and sexual orientation are not the same. Irrespective of their gender identity, transgender people may be attracted to women (gynephilic), attracted to men (androphilic), bisexual, asexual, or queer.
Transgender: This is an umbrella term for people whose gender identity and/or gender expression differs from what is typically associated with their sex designated at birth. Not all transgender individuals seek treatment.
Transgender male (also: trans man, female-to-male, transgender male): This refers to individuals assigned female at birth but who identify and live as men.
Transgender woman (also: trans woman, male-to female, transgender female): This refers to individuals assigned male at birth but who identify and live as women.
Transition: This refers to the process during which transgender persons change their physical, social, and/or legal characteristics consistent with the affirmed gender identity. Prepubertal children may choose to transition socially.
Transsexual: This is an older term that originated in the medical and psychological communities to refer to individuals who have permanently transitioned through medical interventions or desired to do so.

exstrophy and penile agenesis, the occurrence of transgender male changes was significantly more prevalent than in the general population (30, 31). However, the fact that a high percentage of individuals with the same conditions did not change gender suggests that cultural factors may play a role as well.

With respect to genetics and gender identity, several studies have suggested heritability of GD/gender incongruence (32, 33). In particular, a study by Heylens *et al.* (33) demonstrated a 39.1% concordance rate for gender identity disorder (based on the DSM-IV criteria) in 23 monozygotic twin pairs but no concordance in 21 same-sex dizygotic or seven opposite-sex twin pairs. Although numerous investigators have sought to identify specific genes associated with GD/gender incongruence, such studies have been inconsistent and without strong statistical significance (34–38).

Studies focusing on brain structure suggest that the brain phenotypes of people with GD/gender incongruence differ in various ways from control males and females, but that there is not a complete sex reversal in brain structures (39).

In summary, although there is much that is still unknown with respect to gender identity and its expression, compelling studies support the concept that biologic factors, in addition to environmental factors, contribute to this fundamental aspect of human development.

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Natural History of Children With GD/Gender Incongruence

With current knowledge, we cannot predict the psychosexual outcome for any specific child. Prospective follow-up studies show that childhood GD/gender incongruence does not invariably persist into adolescence and adulthood (so-called "desisters"). Combining all outcome studies to date, the GD/gender incongruence of a minority of prepubertal children appears to persist in adolescence (20, 40). In adolescence, a significant number of these desisters identify as homosexual or bisexual. It may be that children who only showed some gender nonconforming characteristics have been included in the follow-up studies, because the DSM-IV text revision criteria for a diagnosis were rather broad. However, the persistence of GD/gender incongruence into adolescence is more likely if it had been extreme in childhood (41, 42). With the newer, stricter criteria of the DSM-5 (Table 2), persistence rates may well be different in future studies.

1.0 Evaluation of Youth and Adults

Gender-affirming treatment is a multidisciplinary effort. After evaluation, education, and diagnosis, treatment may include mental health care, hormone therapy, and/or surgical therapy. Together with an MHP, hormone-prescribing clinicians should examine the psychosocial impact of the potential changes on people's lives, including mental health, friends, family, jobs, and their role in society. Transgender individuals should be encouraged to experience living in the new gender role and assess whether this improves their quality of life. Although the focus of this guideline is gender-affirming hormone therapy, collaboration with appropriate professionals responsible for each aspect of treatment maximizes a successful outcome.

Diagnostic assessment and mental health care

GD/gender incongruence may be accompanied with psychological or psychiatric problems (43–51). It is therefore necessary that clinicians who prescribe hormones and are involved in diagnosis and psychosocial assessment meet the following criteria: (1) are competent in using the DSM and/or the ICD for diagnostic purposes, (2) are able to diagnose GD/gender incongruence and make a distinction between GD/gender incongruence and conditions that have similar features (*e.g.*, body dysmorphic disorder), (3) are trained in diagnosing psychiatric conditions, (4) undertake or refer for appropriate treatment, (5) are able to do a psychosocial assessment of the patient's understanding, mental health, and social conditions that can impact gender-affirming hormone therapy, and (6) regularly attend relevant professional meetings.

Because of the psychological vulnerability of many individuals with GD/gender incongruence, it is important that mental health care is available before, during, and sometimes also after transitioning. For children and adolescents, an MHP who has training/experience in child and adolescent gender development (as well as child and adolescent psychopathology) should make the diagnosis, because assessing GD/gender incongruence in children and adolescents is often extremely complex.

During assessment, the clinician obtains information from the individual seeking gender-affirming treatment. In the case

Table 2. DSM-5 Criteria for Gender Dysphoria in Adolescents and Adults

A. A marked incongruence between one's experienced/expressed gender and natal gender of at least 6 mo in duration, as manifested by at least two of the following:
   1. A marked incongruence between one's experienced/expressed gender and primary and/or secondary sex characteristics (or in young adolescents, the anticipated secondary sex characteristics)
   2. A strong desire to be rid of one's primary and/or secondary sex characteristics because of a marked incongruence with one's experienced/expressed gender (or in young adolescents, a desire to prevent the development of the anticipated secondary sex characteristics)
   3. A strong desire for the primary and/or secondary sex characteristics of the other gender
   4. A strong desire to be of the other gender (or some alternative gender different from one's designated gender)
   5. A strong desire to be treated as the other gender (or some alternative gender different from one's designated gender)
   6. A strong conviction that one has the typical feelings and reactions of the other gender (or some alternative gender different from one's designated gender)

B. The condition is associated with clinically significant distress or impairment in social, occupational, or other important areas of functioning.

Specify if:
   1. The condition exists with a disorder of sex development.
   2. The condition is posttransitional, in that the individual has transitioned to full-time living in the desired gender (with or without legalization of gender change) and has undergone (or is preparing to have) at least one sex-related medical procedure or treatment regimen—namely, regular sex hormone treatment or gender reassignment surgery confirming the desired gender (*e.g.*, penectomy, vaginoplasty in natal males; mastectomy or phalloplasty in natal females).

Reference: American Psychiatric Association (14).

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doi: 10.1210/jc.2017-01658

of adolescents, the clinician also obtains information from the parents or guardians regarding various aspects of the child's general and psychosexual development and current functioning. On the basis of this information, the clinician:

- decides whether the individual fulfills criteria for treatment (see Tables 2 and 3) for GD/gender incongruence (DSM-5) or transsexualism (DSM-5 and/or ICD-10);
- informs the individual about the possibilities and limitations of various kinds of treatment (hormonal/surgical and nonhormonal), and if medical treatment is desired, provides correct information to prevent unrealistically high expectations;
- assesses whether medical interventions may result in unfavorable psychological and social outcomes.

In cases in which severe psychopathology, circumstances, or both seriously interfere with the diagnostic work or make satisfactory treatment unlikely, clinicians should assist the adolescent in managing these other issues. Literature on postoperative regret suggests that besides poor quality of surgery, severe psychiatric comorbidity and lack of support may interfere with positive outcomes (52–56).

For adolescents, the diagnostic procedure usually includes a complete psychodiagnostic assessment (57) and an assessment of the decision-making capability of the youth. An evaluation to assess the family's ability to endure stress, give support, and deal with the complexities of the adolescent's situation should be part of the diagnostic phase (58).

Social transitioning

A change in gender expression and role (which may involve living part time or full time in another gender role that is consistent with one's gender identity) may test the person's resolve, the capacity to function in the affirmed gender, and the adequacy of social, economic, and psychological supports. It assists both the individual and the clinician in their judgments about how to proceed (16). During social transitioning, the person's feelings about the social transformation (including coping with the responses of others) is a major focus of the counseling. The optimal timing for social transitioning may differ between individuals. Sometimes people wait until they start gender-affirming hormone treatment to make social transitioning easier, but individuals increasingly start social transitioning long before they receive medically supervised, gender-affirming hormone treatment.

Criteria

Adolescents and adults seeking gender-affirming hormone treatment and surgery should satisfy certain criteria before proceeding (16). Criteria for gender-affirming hormone therapy for adults are in Table 4, and criteria for gender-affirming hormone therapy for adolescents are in Table 5. Follow-up studies in adults meeting these criteria indicate a high satisfaction rate with treatment (59). However, the quality of evidence is usually low. A few follow-up studies on adolescents who fulfilled these criteria also indicated good treatment results (60–63).

Recommendations for Those Involved in the Gender-Affirming Hormone Treatment of Individuals With GD/Gender Incongruence

1.1. We advise that only trained MHPs who meet the following criteria should diagnose GD/gender incongruence in adults: (1) competence in using the DSM and/or the ICD for diagnostic purposes, (2) the ability to diagnose GD/gender incongruence and make a distinction between GD/gender incongruence and conditions that have similar features (*e.g.*, body dysmorphic disorder), (3) training in diagnosing psychiatric conditions, (4) the ability to undertake or refer for appropriate treatment, (5) the ability to psychosocially assess the person's understanding, mental health, and social conditions that can impact gender-affirming hormone therapy, and (6) a practice of regularly attending relevant professional meetings. (Ungraded Good Practice Statement)

1.2. We advise that only MHPs who meet the following criteria should diagnose GD/gender incongruence in children and adolescents: (1) training in child and adolescent developmental psychology and psychopathology, (2) competence in using the DSM and/or ICD for diagnostic

Table 3. ICD-10 Criteria for Transsexualism

Transsexualism (F64.0) has three criteria:
1. The desire to live and be accepted as a member of the opposite sex, usually accompanied by the wish to make his or her body as congruent as possible with the preferred sex through surgery and hormone treatments.
2. The transsexual identity has been present persistently for at least 2 y.
3. The disorder is not a symptom of another mental disorder or a genetic, DSD, or chromosomal abnormality.

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Table 4. Criteria for Gender-Affirming Hormone Therapy for Adults

1. Persistent, well-documented gender dysphoria/gender incongruence
2. The capacity to make a fully informed decision and to consent for treatment
3. The age of majority in a given country (if younger, follow the criteria for adolescents)
4. Mental health concerns, if present, must be reasonably well controlled

Reproduced from World Professional Association for Transgender Health (16).

purposes, (3) the ability to make a distinction between GD/gender incongruence and conditions that have similar features (*e.g.*, body dysmorphic disorder), (4) training in diagnosing psychiatric conditions, (5) the ability to undertake or refer for appropriate treatment, (6) the ability to psychosocially assess the person's understanding and social conditions that can impact gender-affirming hormone therapy, (7) a practice of regularly attending relevant professional meetings, and (8) knowledge of the criteria for puberty blocking and gender-affirming hormone treatment in adolescents. (Ungraded Good Practice Statement)

Evidence

Individuals with gender identity issues may have psychological or psychiatric problems (43–48, 50, 51, 64, 65). It is therefore necessary that clinicians making the diagnosis are able to make a distinction between GD/gender incongruence and conditions that have similar features. Examples of conditions with similar features are body dysmorphic disorder, body identity integrity disorder (a condition in which individuals have a sense that their anatomical configuration as an able-bodied person is somehow wrong or inappropriate) (66), or certain forms of eunuchism (in which a person is preoccupied with or engages in castration and/or penectomy for

Table 5. Criteria for Gender-Affirming Hormone Therapy for Adolescents

Adolescents are eligible for GnRH agonist treatment if:

1. A qualified MHP has confirmed that:
- the adolescent has demonstrated a long-lasting and intense pattern of gender nonconformity or gender dysphoria (whether suppressed or expressed),
- gender dysphoria worsened with the onset of puberty,
- any coexisting psychological, medical, or social problems that could interfere with treatment (*e.g.*, that may compromise treatment adherence) have been addressed, such that the adolescent's situation and functioning are stable enough to start treatment,
- the adolescent has sufficient mental capacity to give informed consent to this (reversible) treatment,

2. And the adolescent:
- has been informed of the effects and side effects of treatment (including potential loss of fertility if the individual subsequently continues with sex hormone treatment) and options to preserve fertility,
- has given informed consent and (particularly when the adolescent has not reached the age of legal medical consent, depending on applicable legislation) the parents or other caretakers or guardians have consented to the treatment and are involved in supporting the adolescent throughout the treatment process,

3. And a pediatric endocrinologist or other clinician experienced in pubertal assessment
- agrees with the indication for GnRH agonist treatment,
- has confirmed that puberty has started in the adolescent (Tanner stage ≥G2/B2),
- has confirmed that there are no medical contraindications to GnRH agonist treatment.

Adolescents are eligible for subsequent sex hormone treatment if:

1. A qualified MHP has confirmed:
- the persistence of gender dysphoria,
- any coexisting psychological, medical, or social problems that could interfere with treatment (*e.g.*, that may compromise treatment adherence) have been addressed, such that the adolescent's situation and functioning are stable enough to start sex hormone treatment,
- the adolescent has sufficient mental capacity (which most adolescents have by age 16 years) to estimate the consequences of this (partly) irreversible treatment, weigh the benefits and risks, and give informed consent to this (partly) irreversible treatment,

2. And the adolescent:
- has been informed of the (irreversible) effects and side effects of treatment (including potential loss of fertility and options to preserve fertility),
- has given informed consent and (particularly when the adolescent has not reached the age of legal medical consent, depending on applicable legislation) the parents or other caretakers or guardians have consented to the treatment and are involved in supporting the adolescent throughout the treatment process,

3. And a pediatric endocrinologist or other clinician experienced in pubertal induction:
- agrees with the indication for sex hormone treatment,
- has confirmed that there are no medical contraindications to sex hormone treatment.

Reproduced from World Professional Association for Transgender Health (16).

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doi: 10.1210/jc.2017-01658

reasons that are not gender identity related) (11). Clinicians should also be able to diagnose psychiatric conditions accurately and ensure that these conditions are treated appropriately, particularly when the conditions may complicate treatment, affect the outcome of gender-affirming treatment, or be affected by hormone use.

Values and preferences

The task force placed a very high value on avoiding harm from hormone treatment in individuals who have conditions other than GD/gender incongruence and who may not benefit from the physical changes associated with this treatment and placed a low value on any potential benefit these persons believe they may derive from hormone treatment. This justifies the good practice statement.

1.3. We advise that decisions regarding the social transition of prepubertal youths with GD/gender incongruence are made with the assistance of an MHP or another experienced professional. (Ungraded Good Practice Statement).

1.4. We recommend against puberty blocking and gender-affirming hormone treatment in prepubertal children with GD/gender incongruence. (1 |⊕⊕◯◯)

Evidence

In most children diagnosed with GD/gender incongruence, it did not persist into adolescence. The percentages differed among studies, probably dependent on which version of the DSM clinicians used, the patient's age, the recruitment criteria, and perhaps cultural factors. However, the large majority (about 85%) of prepubertal children with a childhood diagnosis did not remain GD/gender incongruent in adolescence (20). If children have completely socially transitioned, they may have great difficulty in returning to the original gender role upon entering puberty (40). Social transition is associated with the persistence of GD/gender incongruence as a child progresses into adolescence. It may be that the presence of GD/gender incongruence in prepubertal children is the earliest sign that a child is destined to be transgender as an adolescent/adult (20). However, social transition (in addition to GD/gender incongruence) has been found to contribute to the likelihood of persistence.

This recommendation, however, does not imply that children should be discouraged from showing gender-variant behaviors or should be punished for exhibiting such behaviors. In individual cases, an early complete social transition may result in a more favorable outcome, but there are currently no criteria to identify the GD/gender-incongruent children to whom this applies. At the present time, clinical experience suggests that persistence of GD/gender incongruence can only be reliably assessed after the first signs of puberty.

Values and preferences

The task force placed a high value on avoiding harm with gender-affirming hormone therapy in prepubertal children with GD/gender incongruence. This justifies the strong recommendation in the face of low-quality evidence.

1.5. We recommend that clinicians inform and counsel all individuals seeking gender-affirming medical treatment regarding options for fertility preservation prior to initiating puberty suppression in adolescents and prior to treating with hormonal therapy of the affirmed gender in both adolescents and adults. (1 |⊕⊕⊕◯)

Remarks

Persons considering hormone use for gender affirmation need adequate information about this treatment in general and about fertility effects of hormone treatment in particular to make an informed and balanced decision (67, 68). Because young adolescents may not feel qualified to make decisions about fertility and may not fully understand the potential effects of hormonal interventions, consent and protocol education should include parents, the referring MHP(s), and other members of the adolescent's support group. To our knowledge, there are no formally evaluated decision aids available to assist in the discussion and decision regarding the future fertility of adolescents or adults beginning gender-affirming treatment.

Treating early pubertal youth with GnRH analogs will temporarily impair spermatogenesis and oocyte maturation. Given that an increasing number of transgender youth want to preserve fertility potential, delaying or temporarily discontinuing GnRH analogs to promote gamete maturation is an option. This option is often not preferred, because mature sperm production is associated with later stages of puberty and with the significant development of secondary sex characteristics.

For those designated male at birth with GD/gender incongruence and who are in early puberty, sperm production and the development of the reproductive tract are insufficient for the cryopreservation of sperm. However, prolonged pubertal suppression using GnRH analogs is reversible and clinicians should inform these individuals that sperm production can be initiated following prolonged gonadotropin suppression. This can be accomplished by spontaneous gonadotropin recovery after

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cessation of GnRH analogs or by gonadotropin treatment and will probably be associated with physical manifestations of testosterone production, as stated above. Note that there are no data in this population concerning the time required for sufficient spermatogenesis to collect enough sperm for later fertility. In males treated for precocious puberty, spermarche was reported 0.7 to 3 years after cessation of GnRH analogs (69). In adult men with gonadotropin deficiency, sperm are noted in seminal fluid by 6 to 12 months of gonadotropin treatment. However, sperm numbers when partners of these patients conceive are far below the "normal range" (70, 71).

In girls, no studies have reported long-term, adverse effects of pubertal suppression on ovarian function after treatment cessation (72, 73). Clinicians should inform adolescents that no data are available regarding either time to spontaneous ovulation after cessation of GnRH analogs or the response to ovulation induction following prolonged gonadotropin suppression.

In males with GD/gender incongruence, when medical treatment is started in a later phase of puberty or in adulthood, spermatogenesis is sufficient for cryopreservation and storage of sperm. *In vitro* spermatogenesis is currently under investigation. Restoration of spermatogenesis after prolonged estrogen treatment has not been studied.

In females with GD/gender incongruence, the effect of prolonged treatment with exogenous testosterone on ovarian function is uncertain. There have been reports of an increased incidence of polycystic ovaries in transgender males, both prior to and as a result of androgen treatment (74–77), although these reports were not confirmed by others (78). Pregnancy has been reported in transgender males who have had prolonged androgen treatment and have discontinued testosterone but have not had genital surgery (79, 80). A reproductive endocrine gynecologist can counsel patients before gender-affirming hormone treatment or surgery regarding potential fertility options (81). Techniques for cryopreservation of oocytes, embryos, and ovarian tissue continue to improve, and oocyte maturation of immature tissue is being studied (82).

2.0 Treatment of Adolescents

During the past decade, clinicians have progressively acknowledged the suffering of young adolescents with GD/gender incongruence. In some forms of GD/gender incongruence, psychological interventions may be useful and sufficient. However, for many adolescents with GD/gender incongruence, the pubertal physical changes are unbearable. As early medical intervention may prevent psychological harm, various clinics have decided to start treating young adolescents with GD/gender incongruence with puberty-suppressing medication (a GnRH analog). As compared with starting gender-affirming treatment long after the first phases of puberty, a benefit of pubertal suppression at early puberty may be a better psychological and physical outcome.

In girls, the first physical sign of puberty is the budding of the breasts followed by an increase in breast and fat tissue. Breast development is also associated with the pubertal growth spurt, and menarche occurs ~2 years later. In boys, the first physical change is testicular growth. A testicular volume ≥4 mL is seen as consistent with the initiation of physical puberty. At the beginning of puberty, estradiol and testosterone levels are still low and are best measured in the early morning with an ultrasensitive assay. From a testicular volume of 10 mL, daytime testosterone levels increase, leading to virilization (83). Note that pubic hair and/or axillary hair/odor may not reflect the onset of gonadarche; instead, it may reflect adrenarche alone.

2.1. We suggest that adolescents who meet diagnostic criteria for GD/gender incongruence, fulfill criteria for treatment (Table 5), and are requesting treatment should initially undergo treatment to suppress pubertal development. (2 ⊕⊕◯◯)

2.2. We suggest that clinicians begin pubertal hormone suppression after girls and boys first exhibit physical changes of puberty (Tanner stages G2/B2). (2 ⊕⊕◯◯)

Evidence

Pubertal suppression can expand the diagnostic phase by a long period, giving the subject more time to explore options and to live in the experienced gender before making a decision to proceed with gender-affirming sex hormone treatments and/or surgery, some of which is irreversible (84, 85). Pubertal suppression is fully reversible, enabling full pubertal development in the natal gender, after cessation of treatment, if appropriate. The experience of full endogenous puberty is an undesirable condition for the GD/gender-incongruent individual and may seriously interfere with healthy psychological functioning and well-being. Treating GD/gender-incongruent adolescents entering puberty with GnRH analogs has been shown to improve psychological functioning in several domains (86).

Another reason to start blocking pubertal hormones early in puberty is that the physical outcome is improved compared with initiating physical transition after puberty has been completed (60, 62). Looking like a man or woman when living as the opposite sex creates difficult

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doi: 10.1210/jc.2017-01658

barriers with enormous life-long disadvantages. We therefore advise starting suppression in early puberty to prevent the irreversible development of undesirable secondary sex characteristics. However, adolescents with GD/gender incongruence should experience the first changes of their endogenous spontaneous puberty, because their emotional reaction to these first physical changes has diagnostic value in establishing the persistence of GD/gender incongruence (85). Thus, Tanner stage 2 is the optimal time to start pubertal suppression. However, pubertal suppression treatment in early puberty will limit the growth of the penis and scrotum, which will have a potential effect on future surgical treatments (87).

Clinicians can also use pubertal suppression in adolescents in later pubertal stages to stop menses in transgender males and prevent facial hair growth in transgender females. However, in contrast to the effects in early pubertal adolescents, physical sex characteristics (such as more advanced breast development in transgender boys and lowering of the voice and outgrowth of the jaw and brow in transgender girls) are not reversible.

Values and preferences

These recommendations place a high value on avoiding an unsatisfactory physical outcome when secondary sex characteristics have become manifest and irreversible, a higher value on psychological well-being, and a lower value on avoiding potential harm from early pubertal suppression.

Remarks

Table 6 lists the Tanner stages of breast and male genital development. Careful documentation of hallmarks of pubertal development will ensure precise timing when initiating pubertal suppression once puberty has started. Clinicians can use pubertal LH and sex steroid levels to confirm that puberty has progressed sufficiently before starting pubertal suppression (88). Reference ranges for sex steroids by Tanner stage may vary depending on the assay used. Ultrasensitive sex steroid and gonadotropin assays will help clinicians document early pubertal changes.

Irreversible and, for GD/gender-incongruent adolescents, undesirable sex characteristics in female puberty are breasts, female body habitus, and, in some cases, relative short stature. In male puberty, they are a prominent Adam's apple; low voice; male bone configuration, such as a large jaw, big feet and hands, and tall stature; and male hair pattern on the face and extremities.

2.3. We recommend that, where indicated, GnRH analogues are used to suppress pubertal hormones. (1 l⊕⊕○○)

Evidence

Clinicians can suppress pubertal development and gonadal function most effectively via gonadotropin suppression using GnRH analogs. GnRH analogs are long-acting agonists that suppress gonadotropins by GnRH receptor desensitization after an initial increase of gonadotropins during ~10 days after the first and (to a lesser degree) the second injection (89). Antagonists immediately suppress pituitary gonadotropin secretion (90, 91). Long-acting GnRH analogs are the currently preferred treatment option. Clinicians may consider long-acting GnRH antagonists when evidence on their safety and efficacy in adolescents becomes available.

During GnRH analog treatment, slight development of secondary sex characteristics may regress, and in a later phase of pubertal development, it will stop. In girls, breast tissue will become atrophic, and menses will stop. In boys, virilization will stop, and testicular volume may decrease (92).

An advantage of using GnRH analogs is the reversibility of the intervention. If, after extensive exploration of his/her transition wish, the individual no longer desires transition, they can discontinue pubertal suppression. In subjects with

Table 6. Tanner Stages of Breast Development and Male External Genitalia

The description of Tanner stages for breast development:
1. Prepubertal
2. Breast and papilla elevated as small mound; areolar diameter increased
3. Breast and areola enlarged, no contour separation
4. Areola and papilla form secondary mound
5. Mature; nipple projects, areola part of general breast contour

For penis and testes:
1. Prepubertal, testicular volume <4 mL
2. Slight enlargement of penis; enlarged scrotum, pink, texture altered, testes 4–6 mL
3. Penis longer, testes larger (8–12 mL)
4. Penis and glans larger, including increase in breadth; testes larger (12–15 mL), scrotum dark
5. Penis adult size; testicular volume > 15 ml

Adapted from Lawrence (56).

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precocious puberty, spontaneous pubertal development has been shown to resume after patients discontinue taking GnRH analogs (93).

Recommendations 2.1 to 2.3 are supported by a prospective follow-up study from The Netherlands. This report assessed mental health outcomes in 55 transgender adolescents/young adults (22 transgender females and 33 transgender males) at three time points: (1) before the start of GnRH agonist (average age of 14.8 years at start of treatment), (2) at initiation of gender-affirming hormones (average age of 16.7 years at start of treatment), and (3) 1 year after "gender-reassignment surgery" (average age of 20.7 years) (63). Despite a decrease in depression and an improvement in general mental health functioning, GD/gender incongruence persisted through pubertal suppression, as previously reported (86). However, following sex hormone treatment and gender-reassignment surgery, GD/gender incongruence was resolved and psychological functioning steadily improved (63). Furthermore, well-being was similar to or better than that reported by age-matched young adults from the general population, and none of the study participants regretted treatment. This study represents the first long-term follow-up of individuals managed according to currently existing clinical practice guidelines for transgender youth, and it underscores the benefit of the multidisciplinary approach pioneered in The Netherlands; however, further studies are needed.

Side effects

The primary risks of pubertal suppression in GD/gender-incongruent adolescents may include adverse effects on bone mineralization (which can theoretically be reversed with sex hormone treatment), compromised fertility if the person subsequently is treated with sex hormones, and unknown effects on brain development. Few data are available on the effect of GnRH analogs on BMD in adolescents with GD/gender incongruence. Initial data in GD/gender-incongruent subjects demonstrated no change of absolute areal BMD during 2 years of GnRH analog therapy but a decrease in BMD *z* scores (85). A recent study also suggested suboptimal bone mineral accrual during GnRH analog treatment. The study reported a decrease in areal BMD *z* scores and of bone mineral apparent density *z* scores (which takes the size of the bone into account) in 19 transgender males treated with GnRH analogs from a mean age of 15.0 years (standard deviation = 2.0 years) for a median duration of 1.5 years (0.3 to 5.2 years) and in 15 transgender females treated from 14.9 (±1.9) years for 1.3 years (0.5 to 3.8 years), although not all changes were statistically significant (94). There was incomplete catch-up at age 22 years after sex hormone treatment from age 16.6 (±1.4) years for a median duration of 5.8 years (3.0 to 8.0 years) in transgender females and from age 16.4 (±2.3) years for 5.4 years (2.8 to 7.8 years) in transgender males. Little is known about more prolonged use of GnRH analogs. Researchers reported normal BMD *z* scores at age 35 years in one individual who used GnRH analogs from age 13.7 years until age 18.6 years before initiating sex hormone treatment (65).

Additional data are available from individuals with late puberty or GnRH analog treatment of other indications. Some studies reported that men with constitutionally delayed puberty have decreased BMD in adulthood (95). However, other studies reported that these men have normal BMD (96, 97). Treating adults with GnRH analogs results in a decrease of BMD (98). In children with central precocious puberty, treatment with GnRH analogs has been found to result in a decrease of BMD during treatment by some (99) but not others (100). Studies have reported normal BMD after discontinuing therapy (69, 72, 73, 101, 102). In adolescents treated with growth hormone who are small for gestational age and have normal pubertal timing, 2-year GnRH analog treatments did not adversely affect BMD (103). Calcium supplementation may be beneficial in optimizing bone health in GnRH analog–treated individuals (104). There are no studies of vitamin D supplementation in this context, but clinicians should offer supplements to vitamin D–deficient adolescents. Physical activity, especially during growth, is important for bone mass in healthy individuals (103) and is therefore likely to be beneficial for bone health in GnRH analog–treated subjects.

GnRH analogs did not induce a change in body mass index standard deviation score in GD/gender-incongruent adolescents (94) but caused an increase in fat mass and decrease in lean body mass percentage (92). Studies in girls treated for precocious puberty also reported a stable body mass index standard deviation score during treatment (72) and body mass index and body composition comparable to controls after treatment (73).

Arterial hypertension has been reported as an adverse effect in a few girls treated with GnRH analogs for precocious/early puberty (105, 106). Blood pressure monitoring before and during treatment is recommended.

Individuals may also experience hot flashes, fatigue, and mood alterations as a consequence of pubertal suppression. There is no consensus on treatment of these side effects in this context.

It is recommended that any use of pubertal blockers (and subsequent use of sex hormones, as detailed below) include a discussion about implications for fertility (see recommendation 1.3). Transgender adolescents may

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doi: 10.1210/jc.2017-01658

want to preserve fertility, which may be otherwise compromised if puberty is suppressed at an early stage and the individual completes phenotypic transition with the use of sex hormones.

Limited data are available regarding the effects of GnRH analogs on brain development. A single cross-sectional study demonstrated no compromise of executive function (107), but animal data suggest there may be an effect of GnRH analogs on cognitive function (108).

Values and preferences

Our recommendation of GnRH analogs places a higher value on the superior efficacy, safety, and reversibility of the pubertal hormone suppression achieved (as compared with the alternatives) and a relatively lower value on limiting the cost of therapy. Of the available alternatives, depot and oral progestin preparations are effective. Experience with this treatment dates back prior to the emergence of GnRH analogs for treating precocious puberty in papers from the 1960s and early 1970s (109–112). These compounds are usually safe, but some side effects have been reported (113–115). Only two recent studies involved transgender youth (116, 117). One of these studies described the use of oral lynestrenol monotherapy followed by the addition of testosterone treatment in transgender boys who were at Tanner stage B4 or further at the start of treatment (117). They found lynestrenol safe, but gonadotropins were not fully suppressed. The study reported metrorrhagia in approximately half of the individuals, mainly in the first 6 months. Acne, headache, hot flashes, and fatigue were other frequent side effects. Another progestin that has been studied in the United States is medroxyprogesterone. This agent is not as effective as GnRH analogs in lowering endogenous sex hormones either and may be associated with other side effects (116). Progestin preparations may be an acceptable treatment for persons without access to GnRH analogs or with a needle phobia. If GnRH analog treatment is not available (insurance denial, prohibitive cost, or other reasons), postpubertal, transgender female adolescents may be treated with an antiandrogen that directly suppresses androgen synthesis or action (see adult section).

Remarks

Measurements of gonadotropin and sex steroid levels give precise information about gonadal axis suppression, although there is insufficient evidence for any specific short-term monitoring scheme in children treated with GnRH analogs (88). If the gonadal axis is not completely suppressed—as evidenced by (for example) menses, erections, or progressive hair growth—the interval of GnRH analog treatment can be shortened or the dose increased. During treatment, adolescents should be monitored for negative effects of delaying puberty, including a halted growth spurt and impaired bone mineral accretion. Table 7 illustrates a suggested clinical protocol.

Anthropometric measurements and X-rays of the left hand to monitor bone age are informative for evaluating growth. To assess BMD, clinicians can perform dual-energy X-ray absorptiometry scans.

2.4. In adolescents who request sex hormone treatment (given this is a partly irreversible treatment), we recommend initiating treatment using a gradually increasing dose schedule (see Table 8) after a multidisciplinary team of medical and MHPs has confirmed the persistence of GD/gender incongruence and sufficient mental capacity to give informed consent, which most adolescents have by age 16 years (Table 5). (1 |⊕⊕◯◯)

2.5. We recognize that there may be compelling reasons to initiate sex hormone treatment prior to the age of 16 years in some adolescents with GD/gender incongruence, even though there are minimal published studies of gender-affirming hormone treatments administered before age 13.5 to 14 years. As with the care of adolescents ≥16 years of age, we recommend that an expert multidisciplinary team of medical and MHPs manage this treatment. (1 |⊕◯◯◯)

2.6. We suggest monitoring clinical pubertal development every 3 to 6 months and laboratory parameters every 6 to 12 months during sex hormone treatment (Table 9). (2 |⊕⊕◯◯)

Table 7. Baseline and Follow-Up Protocol During Suppression of Puberty

Every 3–6 mo
Anthropometry: height, weight, sitting height, blood pressure, Tanner stages
Every 6–12 mo
Laboratory: LH, FSH, E2/T, 25OH vitamin D
Every 1–2 y
Bone density using DXA
Bone age on X-ray of the left hand (if clinically indicated)

Adapted from Hembree *et al.* (118).

Abbreviations: DXA, dual-energy X-ray absorptiometry; E2, estradiol; FSH, follicle stimulating hormone; LH, luteinizing hormone; T, testosterone;

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Table 8. Protocol Induction of Puberty

Induction of female puberty with oral 17β-estradiol, increasing the dose every 6 mo:
- 5 μg/kg/d
- 10 μg/kg/d
- 15 μg/kg/d
- 20 μg/kg/d
- Adult dose = 2–6 mg/d
- *In postpubertal transgender female adolescents, the dose of 17β-estradiol can be increased more rapidly:*
  - 1 mg/d for 6 mo
  - 2 mg/d

Induction of female puberty with transdermal 17β-estradiol, increasing the dose every 6 mo (new patch is placed every 3.5 d):
- 6.25–12.5 μg/24 h (cut 25-μg patch into quarters, then halves)
- 25 μg/24 h
- 37.5 μg/24 h
- Adult dose = 50–200 μg/24 h
- *For alternatives once at adult dose, see Table 11.*
- *Adjust maintenance dose to mimic physiological estradiol levels (see Table 15).*

Induction of male puberty with testosterone esters increasing the dose every 6 mo (IM or SC):
- 25 mg/m^2/2 wk (or alternatively, half this dose weekly, or double the dose every 4 wk)
- 50 mg/m^2/2 wk
- 75 mg/m^2/2 wk
- 100 mg/m^2/2 wk
- Adult dose = 100–200 mg every 2 wk
- *In postpubertal transgender male adolescents the dose of testosterone esters can be increased more rapidly:*
  - 75 mg/2 wk for 6 mo
  - 125 mg/2 wk
- *For alternatives once at adult dose, see Table 11.*
- *Adjust maintenance dose to mimic physiological testosterone levels (see Table 14).*

Adapted from Hembree *et al.* (118).

Abbreviations: IM, intramuscularly; SC, subcutaneously.

Evidence

Adolescents develop competence in decision making at their own pace. Ideally, the supervising medical professionals should individually assess this competence, although no objective tools to make such an assessment are currently available.

Many adolescents have achieved a reasonable level of competence by age 15 to 16 years (119), and in many countries 16-year-olds are legally competent with regard to medical decision making (120). However, others believe that although some capacities are generally achieved before age 16 years, other abilities (such as good risk assessment) do not develop until well after 18 years (121). They suggest that health care procedures should be divided along a matrix of relative risk, so that younger adolescents can be allowed to decide about low-risk procedures, such as most diagnostic tests and common therapies, but not about high-risk procedures, such as most surgical procedures (121).

Currently available data from transgender adolescents support treatment with sex hormones starting at age 16 years (63, 122). However, some patients may incur potential risks by waiting until age 16 years. These include the potential risk to bone health if puberty is suppressed

Table 9. Baseline and Follow-up Protocol During Induction of Puberty

Every 3–6 mo
- •Anthropometry: height, weight, sitting height, blood pressure, Tanner stages

Every 6–12 mo
- •In transgender males: hemoglobin/hematocrit, lipids, testosterone, 25OH vitamin D
- •In transgender females: prolactin, estradiol, 25OH vitamin D

Every 1–2 y
- •BMD using DXA
- •Bone age on X-ray of the left hand (if clinically indicated)

BMD should be monitored into adulthood (until the age of 25–30 y or until peak bone mass has been reached).
For recommendations on monitoring once pubertal induction has been completed, see Tables 14 and 15.

Adapted from Hembree *et al.* (118).

Abbreviation: DXA, dual-energy X-ray absorptiometry.

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doi: 10.1210/jc.2017-01658

for 6 to 7 years before initiating sex hormones (*e.g.*, if someone reached Tanner stage 2 at age 9-10 years old). Additionally, there may be concerns about inappropriate height and potential harm to mental health (emotional and social isolation) if initiation of secondary sex characteristics must wait until the person has reached 16 years of age. However, only minimal data supporting earlier use of gender-affirming hormones in transgender adolescents currently exist (63). Clearly, long-term studies are needed to determine the optimal age of sex hormone treatment in GD/gender-incongruent adolescents.

The MHP who has followed the adolescent during GnRH analog treatment plays an essential role in assessing whether the adolescent is eligible to start sex hormone therapy and capable of consenting to this treatment (Table 5). Support of the family/environment is essential. Prior to the start of sex hormones, clinicians should discuss the implications for fertility (see recommendation 1.5). Throughout pubertal induction, an MHP and a pediatric endocrinologist (or other clinician competent in the evaluation and induction of pubertal development) should monitor the adolescent. In addition to monitoring therapy, it is also important to pay attention to general adolescent health issues, including healthy life style choices, such as not smoking, contraception, and appropriate vaccinations (*e.g.*, human papillomavirus).

For the induction of puberty, clinicians can use a similar dose scheme for hypogonadal adolescents with GD/gender incongruence as they use in other individuals with hypogonadism, carefully monitoring for desired and undesired effects (Table 8). In transgender female adolescents, transdermal 17β-estradiol may be an alternative for oral 17β-estradiol. It is increasingly used for pubertal induction in hypogonadal females. However, the absence of low-dose estrogen patches may be a problem. As a result, individuals may need to cut patches to size themselves to achieve appropriate dosing (123). In transgender male adolescents, clinicians can give testosterone injections intramuscularly or subcutaneously (124, 125).

When puberty is initiated with a gradually increasing schedule of sex steroid doses, the initial levels will not be high enough to suppress endogenous sex steroid secretion. Gonadotropin secretion and endogenous production of testosterone may resume and interfere with the effectiveness of estrogen treatment, in transgender female adolescents (126, 127). Therefore, continuation of GnRH analog treatment is advised until gonadectomy. Given that GD/gender-incongruent adolescents may opt not to have gonadectomy, long-term studies are necessary to examine the potential risks of prolonged GnRH analog treatment. Alternatively, in transgender male adolescents, GnRH analog treatment can be discontinued once an adult dose of testosterone has been reached and the individual is well virilized. If uterine bleeding occurs, a progestin can be added. However, the combined use of a GnRH analog (for ovarian suppression) and testosterone may enable phenotypic transition with a lower dose of testosterone in comparison with testosterone alone. If there is a wish or need to discontinue GnRH analog treatment in transgender female adolescents, they may be treated with an antiandrogen that directly suppresses androgen synthesis or action (see section 3.0 "Hormonal Therapy for Transgender Adults").

Values and preferences

The recommendation to initiate pubertal induction only when the individual has sufficient mental capacity (roughly age 16 years) to give informed consent for this partly irreversible treatment places a higher value on the ability of the adolescent to fully understand and oversee the partially irreversible consequences of sex hormone treatment and to give informed consent. It places a lower value on the possible negative effects of delayed puberty. We may not currently have the means to weigh adequately the potential benefits of waiting until around age 16 years to initiate sex hormones vs the potential risks/harm to BMD and the sense of social isolation from having the timing of puberty be so out of sync with peers (128).

Remarks

Before starting sex hormone treatment, effects on fertility and options for fertility preservation should be discussed. Adult height may be a concern in transgender adolescents. In a transgender female adolescent, clinicians may consider higher doses of estrogen or a more rapid tempo of dose escalation during pubertal induction. There are no established treatments yet to augment adult height in a transgender male adolescent with open epiphyses during pubertal induction. It is not uncommon for transgender adolescents to present for clinical services after having completed or nearly completed puberty. In such cases, induction of puberty with sex hormones can be done more rapidly (see Table 8). Additionally, an adult dose of testosterone in transgender male adolescents may suffice to suppress the gonadal axis without the need to use a separate agent. At the appropriate time, the multidisciplinary team should adequately prepare the adolescent for transition to adult care.

3.0 Hormonal Therapy for Transgender Adults

The two major goals of hormonal therapy are (1) to reduce endogenous sex hormone levels, and thus reduce

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the secondary sex characteristics of the individual's designated gender, and (2) to replace endogenous sex hormone levels consistent with the individual's gender identity by using the principles of hormone replacement treatment of hypogonadal patients. The timing of these two goals and the age at which to begin treatment with the sex hormones of the chosen gender is codetermined in collaboration with both the person pursuing transition and the health care providers. The treatment team should include a medical provider knowledgeable in transgender hormone therapy, an MHP knowledgeable in GD/gender incongruence and the mental health concerns of transition, and a primary care provider able to provide care appropriate for transgender individuals. The physical changes induced by this sex hormone transition are usually accompanied by an improvement in mental well-being (129, 130).

3.1. We recommend that clinicians confirm the diagnostic criteria of GD/gender incongruence and the criteria for the endocrine phase of gender transition before beginning treatment. (1 ⊕⊕⊕◯)

3.2. We recommend that clinicians evaluate and address medical conditions that can be exacerbated by hormone depletion and treatment with sex hormones of the affirmed gender before beginning treatment (Table 10). (1 ⊕⊕⊕◯)

3.3. We suggest that clinicians measure hormone levels during treatment to ensure that endogenous sex steroids are suppressed and administered sex steroids are maintained in the normal physiologic range for the affirmed gender. (2 ⊕⊕◯◯)

Evidence

It is the responsibility of the treating clinician to confirm that the person fulfills criteria for treatment. The treating clinician should become familiar with the terms and criteria presented in Tables 1–5 and take a thorough history from the patient in collaboration with the other members of the treatment team. The treating clinician must ensure that the desire for transition is appropriate; the consequences, risks, and benefits of treatment are well understood; and the desire for transition persists. They also need to discuss fertility preservation options (see recommendation 1.3) (67, 68).

Transgender males

Clinical studies have demonstrated the efficacy of several different androgen preparations to induce masculinization in transgender males (Appendix A) (113, 114, 131–134). Regimens to change secondary sex characteristics follow the general principle of hormone replacement treatment of male hypogonadism (135). Clinicians can use either parenteral or transdermal preparations to achieve testosterone values in the normal male range (this is dependent on the specific assay, but is typically 320 to 1000 ng/dL) (Table 11) (136). Sustained supraphysiologic levels of testosterone increase the risk of adverse reactions (see section 4.0 "Adverse Outcome Prevention and Long-Term Care") and should be avoided.

Similar to androgen therapy in hypogonadal men, testosterone treatment in transgender males results in increased muscle mass and decreased fat mass, increased facial hair and acne, male pattern baldness in those genetically predisposed, and increased sexual desire (137).

Table 10. Medical Risks Associated With Sex Hormone Therapy

Transgender female: estrogen
Very high risk of adverse outcomes:
•Thromboembolic disease
Moderate risk of adverse outcomes:
•Macroprolactinoma
•Breast cancer
•Coronary artery disease
•Cerebrovascular disease
•Cholelithiasis
•Hypertriglyceridemia
Transgender male: testosterone
Very high risk of adverse outcomes:
•Erythrocytosis (hematocrit > 50%)
Moderate risk of adverse outcomes:
•Severe liver dysfunction (transaminases > threefold upper limit of normal)
•Coronary artery disease
•Cerebrovascular disease
•Hypertension
•Breast or uterine cancer

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doi: 10.1210/jc.2017-01658

Table 11. Hormone Regimens in Transgender Persons

Transgender females[a]	
Estrogen	
Oral	
Estradiol	2.0–6.0 mg/d
Transdermal	
Estradiol transdermal patch (New patch placed every 3–5 d)	0.025–0.2 mg/d
Parenteral	
Estradiol valerate or cypionate	5–30 mg IM every 2 wk 2–10 mg IM every week
Anti-androgens	
Spironolactone	100–300 mg/d
Cyproterone acetate[b]	25–50 mg/d
GnRH agonist	3.75 mg SQ (SC) monthly 11.25 mg SQ (SC) 3-monthly
Transgender males	
Testosterone	
Parenteral testosterone	
Testosterone enanthate or cypionate	100–200 mg SQ (IM) every 2 wk or SQ (SC) 50% per week
Testosterone undecanoate[c]	1000 mg every 12 wk
Transdermal testosterone	
Testosterone gel 1.6%[d]	50–100 mg/d
Testosterone transdermal patch	2.5–7.5 mg/d

Abbreviations: IM, intramuscularly; SQ, sequentially; SC, subcutaneously.

[a]Estrogens used with or without antiandrogens or GnRH agonist.

[b]Not available in the United States.

[c]One thousand milligrams initially followed by an injection at 6 wk then at 12-wk intervals.

[d]Avoid cutaneous transfer to other individuals.

In transgender males, testosterone will result in clitoromegaly, temporary or permanent decreased fertility, deepening of the voice, cessation of menses (usually), and a significant increase in body hair, particularly on the face, chest, and abdomen. Cessation of menses may occur within a few months with testosterone treatment alone, although high doses of testosterone may be required. If uterine bleeding continues, clinicians may consider the addition of a progestational agent or endometrial ablation (138). Clinicians may also administer GnRH analogs or depot medroxyprogesterone to stop menses prior to testosterone treatment.

Transgender females

The hormone regimen for transgender females is more complex than the transgender male regimen (Appendix B). Treatment with physiologic doses of estrogen alone is insufficient to suppress testosterone levels into the normal range for females (139). Most published clinical studies report the need for adjunctive therapy to achieve testosterone levels in the female range (21, 113, 114, 132–134, 139, 140).

Multiple adjunctive medications are available, such as progestins with antiandrogen activity and GnRH agonists (141). Spironolactone works by directly blocking androgens during their interaction with the androgen receptor (114, 133, 142). It may also have estrogenic activity (143). Cyproterone acetate, a progestational compound with antiandrogenic properties (113, 132, 144), is widely used in Europe. 5α-Reductase inhibitors do not reduce testosterone levels and have adverse effects (145).

Dittrich *et al.* (141) reported that monthly doses of the GnRH agonist goserelin acetate in combination with estrogen were effective in reducing testosterone levels with a low incidence of adverse reactions in 60 transgender females. Leuprolide and transdermal estrogen were as effective as cyproterone and transdermal estrogen in a comparative retrospective study (146).

Patients can take estrogen as oral conjugated estrogens, oral 17β-estradiol, or transdermal 17β-estradiol. Among estrogen options, the increased risk of thromboembolic events associated with estrogens in general seems most concerning with ethinyl estradiol specifically (134, 140, 141), which is why we specifically suggest that it not be used in any transgender treatment plan. Data distinguishing among other estrogen options are less well established although there is some thought that oral routes of administration are more thrombogenic due to the "first pass effect" than are transdermal and parenteral routes, and that the risk of thromboembolic events is dose-dependent. Injectable estrogen and sublingual

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estrogen may benefit from avoiding the first pass effect, but they can result in more rapid peaks with greater overall periodicity and thus are more difficult to monitor (147, 148). However, there are no data demonstrating that increased periodicity is harmful otherwise.

Clinicians can use serum estradiol levels to monitor oral, transdermal, and intramuscular estradiol. Blood tests cannot monitor conjugated estrogens or synthetic estrogen use. Clinicians should measure serum estradiol and serum testosterone and maintain them at the level for premenopausal females (100 to 200 pg/mL and <50 ng/dL, respectively). The transdermal preparations and injectable estradiol cypionate or valerate preparations may confer an advantage in older transgender females who may be at higher risk for thromboembolic disease (149).

Values

Our recommendation to maintain levels of gender-affirming hormones in the normal adult range places a high value on the avoidance of the long-term complications of pharmacologic doses. Those patients receiving endocrine treatment who have relative contraindications to hormones should have an in-depth discussion with their physician to balance the risks and benefits of therapy.

Remarks

Clinicians should inform all endocrine-treated individuals of all risks and benefits of gender-affirming hormones prior to initiating therapy. Clinicians should strongly encourage tobacco use cessation in transgender females to avoid increased risk of VTE and cardiovascular complications. We strongly discourage the unsupervised use of hormone therapy (150).

Not all individuals with GD/gender incongruence seek treatment as described (*e.g.*, male-to-eunuchs and individuals seeking partial transition). Tailoring current protocols to the individual may be done within the context of accepted safety guidelines using a multidisciplinary approach including mental health. No evidence-based protocols are available for these groups (151). We need prospective studies to better understand treatment options for these persons.

3.4. We suggest that endocrinologists provide education to transgender individuals undergoing treatment about the onset and time course of physical changes induced by sex hormone treatment. (2 |⊕○○○)

Evidence

Transgender males

Physical changes that are expected to occur during the first 1 to 6 months of testosterone therapy include cessation of menses, increased sexual desire, increased facial and body hair, increased oiliness of skin, increased muscle, and redistribution of fat mass. Changes that occur within the first year of testosterone therapy include deepening of the voice (152, 153), clitoromegaly, and male pattern hair loss (in some cases) (114, 144, 154, 155) (Table 12).

Transgender females

Physical changes that may occur in transgender females in the first 3 to 12 months of estrogen and antiandrogen therapy include decreased sexual desire, decreased spontaneous erections, decreased facial and body hair (usually mild), decreased oiliness of skin, increased breast tissue growth, and redistribution of fat mass (114, 139, 149, 154, 155, 161) (Table 13). Breast development is generally maximal at 2 years after initiating hormones (114, 139, 149, 155). Over a long period of time, the prostate gland and testicles will undergo atrophy.

Although the time course of breast development in transgender females has been studied (150), precise information about other changes induced by sex hormones is lacking (141). There is a great deal of variability among individuals, as evidenced during pubertal development. We all know that a major concern for transgender females is breast development. If we work with estrogens, the result will be often not what the transgender female expects.

Alternatively, there are transgender females who report an anecdotal improved breast development, mood, or sexual desire with the use of progestogens. However, there have been no well-designed studies of the role of progestogens in feminizing hormone regimens, so the question is still open.

Our knowledge concerning the natural history and effects of different cross-sex hormone therapies on breast

Table 12. Masculinizing Effects in Transgender Males

Effect	Onset	Maximum
Skin oiliness/acne	1–6 mo	1–2 y
Facial/body hair growth	6–12 mo	4–5 y
Scalp hair loss	6–12 mo	—[a]
Increased muscle mass/strength	6–12 mo	2–5 y
Fat redistribution	1–6 mo	2–5 y
Cessation of menses	1–6 mo	—[b]
Clitoral enlargement	1–6 mo	1–2 y
Vaginal atrophy	1–6 mo	1–2 y
Deepening of voice	6–12 mo	1–2 y

Estimates represent clinical observations: Toorians *et al.* (149), Asscheman *et al.* (156), Gooren *et al.* (157), Wierckx *et al.* (158).

[a] Prevention and treatment as recommended for biological men.

[b] Menorrhagia requires diagnosis and treatment by a gynecologist.

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doi: 10.1210/jc.2017-01658

Table 13. Feminizing Effects in Transgender Females

Effect	Onset	Maximum
Redistribution of body fat	3–6 mo	2–3 y
Decrease in muscle mass and strength	3–6 mo	1–2 y
Softening of skin/decreased oiliness	3–6 mo	Unknown
Decreased sexual desire	1–3 mo	3–6 mo
Decreased spontaneous erections	1–3 mo	3–6 mo
Male sexual dysfunction	Variable	Variable
Breast growth	3–6 mo	2–3 y
Decreased testicular volume	3–6 mo	2–3 y
Decreased sperm production	Unknown	>3 y
Decreased terminal hair growth	6–12 mo	>3 y[a]
Scalp hair	Variable	—[b]
Voice changes	None	—[c]

Estimates represent clinical observations: Toorians *et al.* (149), Asscheman *et al.* (156), Gooren *et al.* (157).

[a]Complete removal of male sexual hair requires electrolysis or laser treatment or both.

[b]Familial scalp hair loss may occur if estrogens are stopped.

[c]Treatment by speech pathologists for voice training is most effective.

development in transgender females is extremely sparse and based on the low quality of evidence. Current evidence does not indicate that progestogens enhance breast development in transgender females, nor does evidence prove the absence of such an effect. This prevents us from drawing any firm conclusion at this moment and demonstrates the need for further research to clarify these important clinical questions (162).

Values and preferences

Transgender persons have very high expectations regarding the physical changes of hormone treatment and are aware that body changes can be enhanced by surgical procedures (*e.g.*, breast, face, and body habitus). Clear expectations for the extent and timing of sex hormone–induced changes may prevent the potential harm and expense of unnecessary procedures.

4.0 Adverse Outcome Prevention and Long-Term Care

Hormone therapy for transgender males and females confers many of the same risks associated with sex hormone replacement therapy in nontransgender persons. The risks arise from and are worsened by inadvertent or intentional use of supraphysiologic doses of sex hormones, as well as use of inadequate doses of sex hormones to maintain normal physiology (131, 139).

4.1. We suggest regular clinical evaluation for physical changes and potential adverse changes in response to sex steroid hormones and laboratory monitoring of sex steroid hormone levels every 3 months during the first year of hormone therapy for transgender males and females and then once or twice yearly. (2 ⊕⊕◯◯)

Evidence

Pretreatment screening and appropriate regular medical monitoring are recommended for both transgender males and females during the endocrine transition and periodically thereafter (26, 155). Clinicians should monitor weight and blood pressure, conduct physical exams, and assess routine health questions, such as tobacco use, symptoms of depression, and risk of adverse events such as deep vein thrombosis/pulmonary embolism and other adverse effects of sex steroids.

Transgender males

Table 14 contains a standard monitoring plan for transgender males on testosterone therapy (154, 159). Key issues include maintaining testosterone levels in the physiologic normal male range and avoiding adverse events resulting from excess testosterone therapy, particularly erythrocytosis, sleep apnea, hypertension, excessive weight gain, salt retention, lipid changes, and excessive or cystic acne (135).

Because oral 17-alkylated testosterone is not recommended, serious hepatic toxicity is not anticipated with parenteral or transdermal testosterone use (163, 164). Past concerns regarding liver toxicity with testosterone have been alleviated with subsequent reports that indicate the risk of serious liver disease is minimal (144, 165, 166).

Transgender females

Table 15 contains a standard monitoring plan for transgender females on estrogens, gonadotropin suppression, or antiandrogens (160). Key issues include avoiding supraphysiologic doses or blood levels of estrogen that may lead to increased risk for thromboembolic disease, liver dysfunction, and hypertension. Clinicians should monitor serum estradiol levels using laboratories participating in external quality control, as measurements of estradiol in blood can be very challenging (167).

VTE may be a serious complication. A study reported a 20-fold increase in venous thromboembolic disease in a large cohort of Dutch transgender subjects (161). This increase may have been associated with the use of the synthetic estrogen, ethinyl estradiol (149). The incidence decreased when clinicians stopped administering ethinyl estradiol (161). Thus, the use of synthetic estrogens and conjugated estrogens is undesirable because of the inability to regulate doses by measuring serum levels and the risk of thromboembolic disease. In a German gender clinic, deep vein thrombosis occurred in 1 of 60 of transgender females treated with a GnRH analog and oral

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Table 14. Monitoring of Transgender Persons on Gender-Affirming Hormone Therapy: Transgender Male

1. Evaluate patient every 3 mo in the first year and then one to two times per year to monitor for appropriate signs of virilization and for development of adverse reactions.
2. Measure serum testosterone every 3 mo until levels are in the normal physiologic male range:[a]
   a. For testosterone enanthate/cypionate injections, the testosterone level should be measured midway between injections. The target level is 400–700 ng/dL to 400 ng/dL. Alternatively, measure peak and trough levels to ensure levels remain in the normal male range.
   b. For parenteral testosterone undecanoate, testosterone should be measured just before the following injection. If the level is <400 ng/dL, adjust dosing interval.
   c. For transdermal testosterone, the testosterone level can be measured no sooner than after 1 wk of daily application (at least 2 h after application).
3. Measure hematocrit or hemoglobin at baseline and every 3 mo for the first year and then one to two times a year. Monitor weight, blood pressure, and lipids at regular intervals.
4. Screening for osteoporosis should be conducted in those who stop testosterone treatment, are not compliant with hormone therapy, or who develop risks for bone loss.
5. If cervical tissue is present, monitoring as recommended by the American College of Obstetricians and Gynecologists.
6. Ovariectomy can be considered after completion of hormone transition.
7. Conduct sub- and periareolar annual breast examinations if mastectomy performed. If mastectomy is not performed, then consider mammograms as recommended by the American Cancer Society.

[a]Adapted from Lapauw *et al.* (154) and Ott *et al.* (159).

estradiol (141). The patient who developed a deep vein thrombosis was found to have a homozygous C677 T mutation in the methylenetetrahydrofolate reductase gene. In an Austrian gender clinic, administering gender-affirming hormones to 162 transgender females and 89 transgender males was not associated with VTE, despite an 8.0% and 5.6% incidence of thrombophilia (159). A more recent multinational study reported only 10 cases of VTE from a cohort of 1073 subjects (168). Thrombophilia screening of transgender persons initiating hormone treatment should be restricted to those with a personal or family history of VTE (159). Monitoring D-dimer levels during treatment is not recommended (169).

4.2. We suggest periodically monitoring prolactin levels in transgender females treated with estrogens. (2 l⊕⊕◯◯)

Evidence

Estrogen therapy can increase the growth of pituitary lactrotroph cells. There have been several reports of prolactinomas occurring after long-term, high-dose estrogen therapy (170–173). Up to 20% of transgender females treated with estrogens may have elevations in prolactin levels associated with enlargement of the pituitary gland (156). In most cases, the serum prolactin levels will return to the normal range with a reduction or discontinuation of the estrogen therapy or discontinuation of cyproterone acetate (157, 174, 175).

The onset and time course of hyperprolactinemia during estrogen treatment are not known. Clinicians should measure prolactin levels at baseline and then at least annually during the transition period and every 2 years thereafter. Given that only a few case studies reported prolactinomas, and prolactinomas were not reported in large cohorts of estrogen-treated persons, the risk is likely to be very low. Because the major presenting findings of microprolactinomas (hypogonadism and sometimes gynecomastia) are not apparent in transgender females, clinicians may perform radiologic examinations of the pituitary in those patients whose prolactin levels persistently increase despite stable or reduced estrogen levels. Some transgender individuals receive psychotropic medications that can increase prolactin levels (174).

Table 15. Monitoring of Transgender Persons on Gender-Affirming Hormone Therapy: Transgender Female

1. Evaluate patient every 3 mo in the first year and then one to two times per year to monitor for appropriate signs of feminization and for development of adverse reactions.
2. Measure serum testosterone and estradiol every 3 mo.
   a. Serum testosterone levels should be <50 ng/dL.
   b. Serum estradiol should not exceed the peak physiologic range: 100–200 pg/mL.
3. For individuals on spironolactone, serum electrolytes, particularly potassium, should be monitored every 3 mo in the first year and annually thereafter.
4. Routine cancer screening is recommended, as in nontransgender individuals (all tissues present).
5. Consider BMD testing at baseline (160). In individuals at low risk, screening for osteoporosis should be conducted at age 60 years or in those who are not compliant with hormone therapy.

This table presents strong recommendations and does not include lower level recommendations.

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doi: 10.1210/jc.2017-01658

4.3. We suggest that clinicians evaluate transgender persons treated with hormones for cardiovascular risk factors using fasting lipid profiles, diabetes screening, and/or other diagnostic tools. (2 ⊕⊕◯◯)

Evidence

Transgender males

Administering testosterone to transgender males results in a more atherogenic lipid profile with lowered high-density lipoprotein cholesterol and higher triglyceride and low-density lipoprotein cholesterol values (176–179). Studies of the effect of testosterone on insulin sensitivity have mixed results (178, 180). A randomized, open-label uncontrolled safety study of transgender males treated with testosterone undecanoate demonstrated no insulin resistance after 1 year (181, 182). Numerous studies have demonstrated the effects of sex hormone treatment on the cardiovascular system (160, 179, 183, 184). Long-term studies from The Netherlands found no increased risk for cardiovascular mortality (161). Likewise, a meta-analysis of 19 randomized trials in nontransgender males on testosterone replacement showed no increased incidence of cardiovascular events (185). A systematic review of the literature found that data were insufficient (due to very low–quality evidence) to allow a meaningful assessment of patient-important outcomes, such as death, stroke, myocardial infarction, or VTE in transgender males (176). Future research is needed to ascertain the potential harm of hormonal therapies (176). Clinicians should manage cardiovascular risk factors as they emerge according to established guidelines (186).

Transgender females

A prospective study of transgender females found favorable changes in lipid parameters with increased high-density lipoprotein and decreased low-density lipoprotein concentrations (178). However, increased weight, blood pressure, and markers of insulin resistance attenuated these favorable lipid changes. In a meta-analysis, only serum triglycerides were higher at ≥24 months without changes in other parameters (187). The largest cohort of transgender females (mean age 41 years, followed for a mean of 10 years) showed no increase in cardiovascular mortality despite a 32% rate of tobacco use (161).

Thus, there is limited evidence to determine whether estrogen is protective or detrimental on lipid and glucose metabolism in transgender females (176). With aging, there is usually an increase of body weight. Therefore, as with nontransgender individuals, clinicians should monitor and manage glucose and lipid metabolism and blood pressure regularly according to established guidelines (186).

4.4. We recommend that clinicians obtain BMD measurements when risk factors for osteoporosis exist, specifically in those who stop sex hormone therapy after gonadectomy. (1 ⊕⊕◯◯)

Evidence

Transgender males

Baseline bone mineral measurements in transgender males are generally in the expected range for their pretreatment gender (188). However, adequate dosing of testosterone is important to maintain bone mass in transgender males (189, 190). In one study (190), serum LH levels were inversely related to BMD, suggesting that low levels of sex hormones were associated with bone loss. Thus, LH levels in the normal range may serve as an indicator of the adequacy of sex steroid administration to preserve bone mass. The protective effect of testosterone may be mediated by peripheral conversion to estradiol, both systemically and locally in the bone.

Transgender females

A baseline study of BMD reported T scores less than −2.5 in 16% of transgender females (191). In aging males, studies suggest that serum estradiol more positively correlates with BMD than does testosterone (192, 193) and is more important for peak bone mass (194). Estrogen preserves BMD in transgender females who continue on estrogen and antiandrogen therapies (188, 190, 191, 195, 196).

Fracture data in transgender males and females are not available. Transgender persons who have undergone gonadectomy may choose not to continue consistent sex steroid treatment after hormonal and surgical sex reassignment, thereby becoming at risk for bone loss. There have been no studies to determine whether clinicians should use the sex assigned at birth or affirmed gender for assessing osteoporosis (*e.g.*, when using the FRAX tool). Although some researchers use the sex assigned at birth (with the assumption that bone mass has usually peaked for transgender people who initiate hormones in early adulthood), this should be assessed on a case-by-case basis until there are more data available. This assumption will be further complicated by the increasing prevalence of transgender people who undergo hormonal transition at a pubertal age or soon after puberty. Sex for comparison within risk assessment tools may be based on the age at which hormones were initiated and the length of exposure to hormones. In some cases, it may be

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reasonable to assess risk using both the male and female calculators and using an intermediate value. Because all subjects underwent normal pubertal development, with known effects on bone size, reference values for birth sex were used for all participants (154).

4.5. We suggest that transgender females with no known increased risk of breast cancer follow breast-screening guidelines recommended for those designated female at birth. (2 ⊕⊕◯◯)

4.6. We suggest that transgender females treated with estrogens follow individualized screening according to personal risk for prostatic disease and prostate cancer. (2 ⊕◯◯◯)

Evidence

Studies have reported a few cases of breast cancer in transgender females (197–200). A Dutch study of 1800 transgender females followed for a mean of 15 years (range of 1 30 years) found one case of breast cancer. The Women's Health Initiative study reported that females taking conjugated equine estrogen without progesterone for 7 years did not have an increased risk of breast cancer as compared with females taking placebo (137).

In transgender males, a large retrospective study conducted at the U.S. Veterans Affairs medical health system identified seven breast cancers (194). The authors reported that this was not above the expected rate of breast cancers in cisgender females in this cohort. Furthermore, they did report one breast cancer that developed in a transgender male patient after mastectomy, supporting the fact that breast cancer can occur even after mastectomy. Indeed, there have been case reports of breast cancer developing in subareolar tissue in transgender males, which occurred after mastectomy (201, 202).

Women with primary hypogonadism (Turner syndrome) treated with estrogen replacement exhibited a significantly decreased incidence of breast cancer as compared with national standardized incidence ratios (203, 204). These studies suggest that estrogen therapy does not increase the risk of breast cancer in the short term (<20 to 30 years). We need long-term studies to determine the actual risk, as well as the role of screening mammograms. Regular examinations and gynecologic advice should determine monitoring for breast cancer.

Prostate cancer is very rare before the age of 40, especially with androgen deprivation therapy (205). Childhood or pubertal castration results in regression of the prostate and adult castration reverses benign prostate hypertrophy (206). Although van Kesteren *et al.* (207) reported that estrogen therapy does not induce hypertrophy or premalignant changes in the prostates of transgender females, studies have reported cases of benign prostatic hyperplasia in transgender females treated with estrogens for 20 to 25 years (208, 209). Studies have also reported a few cases of prostate carcinoma in transgender females (210–214).

Transgender females may feel uncomfortable scheduling regular prostate examinations. Gynecologists are not trained to screen for prostate cancer or to monitor prostate growth. Thus, it may be reasonable for transgender females who transitioned after age 20 years to have annual screening digital rectal examinations after age 50 years and prostate-specific antigen tests consistent with U.S. Preventive Services Task Force Guidelines (215).

4.7. We advise that clinicians determine the medical necessity of including a total hysterectomy and oophorectomy as part of gender-affirming surgery. (Ungraded Good Practice Statement)

Evidence

Although aromatization of testosterone to estradiol in transgender males has been suggested as a risk factor for endometrial cancer (216), no cases have been reported. When transgender males undergo hysterectomy, the uterus is small and there is endometrial atrophy (217, 218). Studies have reported cases of ovarian cancer (219, 220). Although there is limited evidence for increased risk of reproductive tract cancers in transgender males, health care providers should determine the medical necessity of a laparoscopic total hysterectomy as part of a gender-affirming surgery to prevent reproductive tract cancer (221).

Values

Given the discomfort that transgender males experience accessing gynecologic care, our recommendation for the medical necessity of total hysterectomy and oophorectomy places a high value on eliminating the risks of female reproductive tract disease and cancer and a lower value on avoiding the risks of these surgical procedures (related to the surgery and to the potential undesirable health consequences of oophorectomy) and their associated costs.

Remarks

The sexual orientation and type of sexual practices will determine the need and types of gynecologic care required following transition. Additionally, in certain countries, the approval required to change the sex in a birth certificate for transgender males may be dependent on having a complete hysterectomy. Clinicians should help patients research nonmedical administrative criteria and

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doi: 10.1210/jc.2017-01658

provide counseling. If individuals decide not to undergo hysterectomy, screening for cervical cancer is the same as all other females.

5.0 Surgery for Sex Reassignment and Gender Confirmation

For many transgender adults, genital gender-affirming surgery may be the necessary step toward achieving their ultimate goal of living successfully in their desired gender role. The type of surgery falls into two main categories: (1) those that directly affect fertility and (2) those that do not. Those that change fertility (previously called sex reassignment surgery) include genital surgery to remove the penis and gonads in the male and removal of the uterus and gonads in the female. The surgeries that effect fertility are often governed by the legal system of the state or country in which they are performed. Other gender-conforming surgeries that do not directly affect fertility are not so tightly governed.

Gender-affirming surgical techniques have improved markedly during the past 10 years. Reconstructive genital surgery that preserves neurologic sensation is now the standard. The satisfaction rate with surgical reassignment of sex is now very high (187). Additionally, the mental health of the individual seems to be improved by participating in a treatment program that defines a pathway of gender-affirming treatment that includes hormones and surgery (130, 144) (Table 16).

Surgery that affects fertility is irreversible. The World Professional Association for Transgender Health Standards of Care (222) emphasizes that the "threshold of 18 should not be seen as an indication in itself for active intervention." If the social transition has not been satisfactory, if the person is not satisfied with or is ambivalent about the effects of sex hormone treatment, or if the person is ambivalent about surgery then the individual should not be referred for surgery (223, 224).

Gender-affirming genital surgeries for transgender females that affect fertility include gonadectomy, penectomy, and creation of a neovagina (225, 226). Surgeons often invert the skin of the penis to form the wall of the vagina, and several literatures reviews have reported on outcomes (227). Sometimes there is inadequate tissue to form a full neovagina, so clinicians have revisited using intestine and found it to be successful (87, 228, 229). Some newer vaginoplasty techniques may involve autologuous oral epithelial cells (230, 231).

The scrotum becomes the labia majora. Surgeons use reconstructive surgery to fashion the clitoris and its hood, preserving the neurovascular bundle at the tip of the penis as the neurosensory supply to the clitoris. Some surgeons are also creating a sensate pedicled-spot adding a G spot to the neovagina to increase sensation (232). Most recently, plastic surgeons have developed techniques to fashion labia minora. To further complete the feminization, uterine transplants have been proposed and even attempted (233).

Neovaginal prolapse, rectovaginal fistula, delayed healing, vaginal stenosis, and other complications do sometimes occur (234, 235). Clinicians should strongly remind the transgender person to use their dilators to maintain the depth and width of the vagina throughout the postoperative period. Genital sexual responsivity and other aspects of sexual function are usually preserved following genital gender-affirming surgery (236, 237).

Ancillary surgeries for more feminine or masculine appearance are not within the scope of this guideline. Voice therapy by a speech language pathologist is available to transform speech patterns to the affirmed gender (148). Spontaneous voice deepening occurs during testosterone treatment of transgender males (152, 238). No studies have compared the effectiveness of speech therapy, laryngeal surgery, or combined treatment.

Breast surgery is a good example of gender-confirming surgery that does not affect fertility. In all females, breast size exhibits a very broad spectrum. For transgender females to make the best informed decision, clinicians should delay breast augmentation surgery until the patient has completed at least 2 years of estrogen therapy, because the breasts continue to grow during that time (141, 155).

Another major procedure is the removal of facial and masculine-appearing body hair using either electrolysis or

Table 16. Criteria for Gender-Affirming Surgery, Which Affects Fertility

1. Persistent, well-documented gender dysphoria
2. Legal age of majority in the given country
3. Having continuously and responsibly used gender-affirming hormones for 12 mo (if there is no medical contraindication to receiving such therapy)
4. Successful continuous full-time living in the new gender role for 12 mo
5. If significant medical or mental health concerns are present, they must be well controlled
6. Demonstrable knowledge of all practical aspects of surgery (*e.g.*, cost, required lengths of hospitalizations, likely complications, postsurgical rehabilitation)

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laser treatments. Other feminizing surgeries, such as that to feminize the face, are now becoming more popular (239–241).

In transgender males, clinicians usually delay gender-affirming genital surgeries until after a few years of androgen therapy. Those surgeries that affect fertility in this group include oophorectomy, vaginectomy, and complete hysterectomy. Surgeons can safely perform them vaginally with laparoscopy. These are sometimes done in conjunction with the creation of a neopenis. The cosmetic appearance of a neopenis is now very good, but the surgery is multistage and very expensive (242, 243). Radial forearm flap seems to be the most satisfactory procedure (228, 244). Other flaps also exist (245). Surgeons can make neopenile erections possible by reinervation of the flap and subsequent contraction of the muscle, leading to stiffening of the neopenis (246, 247), but results are inconsistent (248). Surgeons can also stiffen the penis by imbedding some mechanical device (*e.g.*, a rod or some inflatable apparatus) (249, 250). Because of these limitations, the creation of a neopenis has often been less than satisfactory. Recently, penis transplants are being proposed (233).

In fact, most transgender males do not have any external genital surgery because of the lack of access, high cost, and significant potential complications. Some choose a metaoidioplasty that brings forward the clitoris, thereby allowing them to void in a standing position without wetting themselves (251, 252). Surgeons can create the scrotum from the labia majora with good cosmetic effect and can implant testicular prostheses (253).

The most important masculinizing surgery for the transgender male is mastectomy, and it does not affect fertility. Breast size only partially regresses with androgen therapy (155). In adults, discussions about mastectomy usually take place after androgen therapy has started. Because some transgender male adolescents present after significant breast development has occurred, they may also consider mastectomy 2 years after they begin androgen therapy and before age 18 years. Clinicians should individualize treatment based on the physical and mental health status of the individual. There are now newer approaches to mastectomy with better outcomes (254, 255). These often involve chest contouring (256). Mastectomy is often necessary for living comfortably in the new gender (256).

5.1. We recommend that a patient pursue genital gender-affirming surgery only after the MHP and the clinician responsible for endocrine transition therapy both agree that surgery is medically necessary and would benefit the patient's overall health and/or well-being. (1 ⊕⊕◯◯)

5.2. We advise that clinicians approve genital gender-affirming surgery only after completion of at least 1 year of consistent and compliant hormone treatment, unless hormone therapy is not desired or medically contraindicated. (Ungraded Good Practice Statement)

5.3. We advise that the clinician responsible for endocrine treatment and the primary care provider ensure appropriate medical clearance of transgender individuals for genital gender-affirming surgery and collaborate with the surgeon regarding hormone use during and after surgery. (Ungraded Good Practice Statement)

5.4. We recommend that clinicians refer hormone-treated transgender individuals for genital surgery when: (1) the individual has had a satisfactory social role change, (2) the individual is satisfied about the hormonal effects, and (3) the individual desires definitive surgical changes. (1 ⊕◯◯◯)

5.5. We suggest that clinicians delay gender-affirming genital surgery involving gonadectomy and/or hysterectomy until the patient is at least 18 years old or legal age of majority in his or her country. (2 ⊕⊕◯◯).

5.6. We suggest that clinicians determine the timing of breast surgery for transgender males based upon the physical and mental health status of the individual. There is insufficient evidence to recommend a specific age requirement. (2 ⊕◯◯◯)

Evidence

Owing to the lack of controlled studies, incomplete follow-up, and lack of valid assessment measures, evaluating various surgical approaches and techniques is difficult. However, one systematic review including a large numbers of studies reported satisfactory cosmetic and functional results for vaginoplasty/neovagina construction (257). For transgender males, the outcomes are less certain. However, the problems are now better understood (258). Several postoperative studies report significant long-term psychological and psychiatric pathology (259–261). One study showed satisfaction with breasts, genitals, and femininity increased significantly and showed the importance of surgical treatment as a key therapeutic option for transgender females (262). Another analysis demonstrated that, despite the young average age at death following surgery and the relatively larger number of individuals with somatic morbidity, the study does not allow for determination of

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doi: 10.1210/jc.2017-01658

causal relationships between, for example, specific types of hormonal or surgical treatment received and somatic morbidity and mortality (263). Reversal surgery in regretful male-to-female transsexuals after sexual reassignment surgery represents a complex, multistage procedure with satisfactory outcomes. Further insight into the characteristics of persons who regret their decision postoperatively would facilitate better future selection of applicants eligible for sexual reassignment surgery. We need more studies with appropriate controls that examine long-term quality of life, psychosocial outcomes, and psychiatric outcomes to determine the long-term benefits of surgical treatment.

When a transgender individual decides to have gender-affirming surgery, both the hormone prescribing clinician and the MHP must certify that the patient satisfies criteria for gender-affirming surgery (Table 16).

There is some concern that estrogen therapy may cause an increased risk for venous thrombosis during or following surgery (176). For this reason, the surgeon and the hormone-prescribing clinician should collaborate in making a decision about the use of hormones before and following surgery. One study suggests that preoperative factors (such as compliance) are less important for patient satisfaction than are the physical postoperative results (56). However, other studies and clinical experience dictate that individuals who do not follow medical instructions and do not work with their physicians toward a common goal do not achieve treatment goals (264) and experience higher rates of postoperative infections and other complications (265, 266). It is also important that the person requesting surgery feels comfortable with the anatomical changes that have occurred during hormone therapy. Dissatisfaction with social and physical outcomes during the hormone transition may be a contraindication to surgery (223).

An endocrinologist or experienced medical provider should monitor transgender individuals after surgery. Those who undergo gonadectomy will require hormone replacement therapy, surveillance, or both to prevent adverse effects of chronic hormone deficiency.

Financial Disclosures of the Task Force*

Wylie C. Hembree (chair)—financial or business/organizational interests: none declared, significant financial interest or leadership position: none declared. **Peggy T. Cohen-Kettenis**—financial or business/organizational interests: none declared, significant financial interest or leadership position: none declared. **Louis Gooren**—financial or business/organizational interests: none declared, significant financial interest or leadership position: none declared. **Sabine E. Hannema**—financial or business/organizational interests: none declared, significant financial interest or leadership position: Ferring Pharmaceuticals Inc. (lecture/conference), Pfizer (lecture). **Walter J. Meyer**—financial or business/organizational interests: none declared, significant financial interest or leadership position: none declared. **M. Hassan Murad****—financial or business/organizational interests: Mayo Clinic, Evidence-based Practice Center, significant financial interest or leadership position: none declared. **Stephen M. Rosenthal**—financial or business/organizational interests: AbbVie (consultant), National Institutes of Health (grantee), significant financial interest or leadership position: Pediatric Endocrine Society (immediate past president). **Joshua D. Safer, FACP**—financial or business/organizational interests: none declared, significant financial interest or leadership position: none declared. **Vin Tangpricha**—financial or business/organizational interests: Cystic Fibrosis Foundation (grantee), National Institutes of Health (grantee), significant financial interest or leadership position, Elsevier *Journal of Clinical and Translational Endocrinology* (editor). **Guy G. T'Sjoen**—financial or business/organizational interests: none declared, significant financial interest or leadership position: none declared.* Financial, business, and organizational disclosures of the task force cover the year prior to publication. Disclosures prior to this time period are archived.**Evidence-based reviews for this guideline were prepared under contract with the Endocrine Society.

Acknowledgments

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Disclosure Summary: See Financial Disclosures.

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Gender Dysphoria

In this chapter, there is one overarching diagnosis of gender dysphoria, with separate developmentally appropriate criteria sets for children and for adolescents and adults. The area of sex and gender is highly controversial and has led to a proliferation of terms whose meanings vary over time and within and between disciplines. An additional source of confusion is that in English "sex" connotes both male/female and sexuality. This chapter employs constructs and terms as they are widely used by clinicians from various disciplines with specialization in this area. In this chapter, *sex* and *sexual* refer to the biological indicators of male and female (understood in the context of reproductive capacity), such as in sex chromosomes, gonads, sex hormones, and nonambiguous internal and external genitalia. Disorders of sex development denote conditions of inborn somatic deviations of the reproductive tract from the norm and/or discrepancies among the biological indicators of male and female. *Cross-sex* hormone treatment denotes the use of feminizing hormones in an individual assigned male at birth based on traditional biological indicators or the use of masculinizing hormones in an individual assigned female at birth.

The need to introduce the term *gender* arose with the realization that for individuals with conflicting or ambiguous biological indicators of sex (i.e., "intersex"), the lived role in society and/or the identification as male or female could not be uniformly associated with or predicted from the biological indicators and, later, that some individuals develop an identity as female or male at variance with their uniform set of classical biological indicators. Thus, *gender* is used to denote the public (and usually legally recognized) lived role as boy or girl, man or woman, but, in contrast to certain social constructionist theories, biological factors are seen as contributing, in interaction with social and psychological factors, to gender development. *Gender assignment* refers to the initial assignment as male or female. This occurs usually at birth and, thereby, yields the "natal gender." *Gender-atypical* refers to somatic features or behaviors that are not typical (in a statistical sense) of individuals with the same assigned gender in a given society and historical era; for behavior, *gender-nonconforming* is an alternative descriptive term. *Gender reassignment* denotes an official (and usually legal) change of gender. *Gender identity* is a category of social identity and refers to an individual's identification as male, female, or, occasionally, some category other than male or female. *Gender dysphoria* as a general descriptive term refers to an individual's affective/cognitive discontent with the assigned gender but is more specifically defined when used as a diagnostic category. *Transgender* refers to the broad spectrum of individuals who transiently or persistently identify with a gender different from their natal gender. *Transsexual* denotes an individual who seeks, or has undergone, a social transition from male to female or female to male, which in many, but not all, cases also involves a somatic transition by cross-sex hormone treatment and genital surgery (*sex reassignment surgery*).

Gender dysphoria refers to the distress that may accompany the incongruence between one's experienced or expressed gender and one's assigned gender. Although not all individuals will experience distress as a result of such incongruence, many are distressed if the desired physical interventions by means of hormones and/or surgery are not available. The current term is more descriptive than the previous DSM-IV term *gender identity disorder* and focuses on dysphoria as the clinical problem, not identity per se.

Gender Dysphoria

Diagnostic Criteria

Gender Dysphoria in Children 302.6 (F64.2)

A. A marked incongruence between one's experienced/expressed gender and assigned gender, of at least 6 months' duration, as manifested by at least six of the following (one of which must be Criterion A1):

1. A strong desire to be of the other gender or an insistence that one is the other gender (or some alternative gender different from one's assigned gender).
2. In boys (assigned gender), a strong preference for cross-dressing or simulating female attire; or in girls (assigned gender), a strong preference for wearing only typical masculine clothing and a strong resistance to the wearing of typical feminine clothing.
3. A strong preference for cross-gender roles in make-believe play or fantasy play.
4. A strong preference for the toys, games, or activities stereotypically used or engaged in by the other gender.
5. A strong preference for playmates of the other gender.
6. In boys (assigned gender), a strong rejection of typically masculine toys, games, and activities and a strong avoidance of rough-and-tumble play; or in girls (assigned gender), a strong rejection of typically feminine toys, games, and activities.
7. A strong dislike of one's sexual anatomy.
8. A strong desire for the primary and/or secondary sex characteristics that match one's experienced gender.

B. The condition is associated with clinically significant distress or impairment in social, school, or other important areas of functioning.

Specify if:

With a disorder of sex development (e.g., a congenital adrenogenital disorder such as 255.2 [E25.0] congenital adrenal hyperplasia or 259.50 [E34.50] androgen insensitivity syndrome).

Coding note: Code the disorder of sex development as well as gender dysphoria.

Gender Dysphoria in Adolescents and Adults 302.85 (F64.1)

A. A marked incongruence between one's experienced/expressed gender and assigned gender, of at least 6 months' duration, as manifested by at least two of the following:

1. A marked incongruence between one's experienced/expressed gender and primary and/or secondary sex characteristics (or in young adolescents, the anticipated secondary sex characteristics).
2. A strong desire to be rid of one's primary and/or secondary sex characteristics because of a marked incongruence with one's experienced/expressed gender (or in young adolescents, a desire to prevent the development of the anticipated secondary sex characteristics).
3. A strong desire for the primary and/or secondary sex characteristics of the other gender.
4. A strong desire to be of the other gender (or some alternative gender different from one's assigned gender).
5. A strong desire to be treated as the other gender (or some alternative gender different from one's assigned gender).
6. A strong conviction that one has the typical feelings and reactions of the other gender (or some alternative gender different from one's assigned gender).

B. The condition is associated with clinically significant distress or impairment in social, occupational, or other important areas of functioning.

Specify if:

With a disorder of sex development (e.g., a congenital adrenogenital disorder such as 255.2 [E25.0] congenital adrenal hyperplasia or 259.50 [E34.50] androgen insensitivity syndrome).

Coding note: Code the disorder of sex development as well as gender dysphoria.

Specify if:

Posttransition: The individual has transitioned to full-time living in the desired gender (with or without legalization of gender change) and has undergone (or is preparing to have) at least one cross-sex medical procedure or treatment regimen—namely, regular cross-sex hormone treatment or gender reassignment surgery confirming the desired gender (e.g., penectomy, vaginoplasty in a natal male; mastectomy or phalloplasty in a natal female).

Specifiers

The posttransition specifier may be used in the context of continuing treatment procedures that serve to support the new gender assignment.

Diagnostic Features

Individuals with gender dysphoria have a marked incongruence between the gender they have been assigned to (usually at birth, referred to as *natal gender*) and their experienced/expressed gender. This discrepancy is the core component of the diagnosis. There must also be evidence of distress about this incongruence. Experienced gender may include alternative gender identities beyond binary stereotypes. Consequently, the distress is not limited to a desire to simply be of the other gender, but may include a desire to be of an alternative gender, provided that it differs from the individual's assigned gender.

Gender dysphoria manifests itself differently in different age groups. Prepubertal natal girls with gender dysphoria may express the wish to be a boy, assert they are a boy, or assert they will grow up to be a man. They prefer boys' clothing and hairstyles, are often perceived by strangers as boys, and may ask to be called by a boy's name. Usually, they display intense negative reactions to parental attempts to have them wear dresses or other feminine attire. Some may refuse to attend school or social events where such clothes are required. These girls may demonstrate marked cross-gender identification in role-playing, dreams, and fantasies. Contact sports, rough-and-tumble play, traditional boyhood games, and boys as playmates are most often preferred. They show little interest in stereotypically feminine toys (e.g., dolls) or activities (e.g., feminine dress-up or role-play). Occasionally, they refuse to urinate in a sitting position. Some natal girls may express a desire to have a penis or claim to have a penis or that they will grow one when older. They may also state that they do not want to develop breasts or menstruate.

Prepubertal natal boys with gender dysphoria may express the wish to be a girl or assert they are a girl or that they will grow up to be a woman. They have a preference for dressing in girls' or women's clothes or may improvise clothing from available materials (e.g., using towels, aprons, and scarves for long hair or skirts). These children may role-play female figures (e.g., playing "mother") and often are intensely interested in female fantasy figures. Traditional feminine activities, stereotypical games, and pastimes (e.g., "playing house"; drawing feminine pictures; watching television or videos of favorite female characters) are most often preferred. Stereotypical female-type dolls (e.g., Barbie) are often favorite toys, and girls are their preferred playmates. They avoid rough-and-tumble play and competitive sports and have little interest in stereotypically masculine toys (e.g., cars, trucks). Some may pretend not to have a penis and insist on sitting to urinate. More

rarely, they may state that they find their penis or testes disgusting, that they wish them removed, or that they have, or wish to have, a vagina.

In young adolescents with gender dysphoria, clinical features may resemble those of children or adults with the condition, depending on developmental level. As secondary sex characteristics of young adolescents are not yet fully developed, these individuals may not state dislike of them, but they are concerned about imminent physical changes.

In adults with gender dysphoria, the discrepancy between experienced gender and physical sex characteristics is often, but not always, accompanied by a desire to be rid of primary and/or secondary sex characteristics and/or a strong desire to acquire some primary and/or secondary sex characteristics of the other gender. To varying degrees, adults with gender dysphoria may adopt the behavior, clothing, and mannerisms of the experienced gender. They feel uncomfortable being regarded by others, or functioning in society, as members of their assigned gender. Some adults may have a strong desire to be of a different gender and treated as such, and they may have an inner certainty to feel and respond as the experienced gender without seeking medical treatment to alter body characteristics. They may find other ways to resolve the incongruence between experienced/expressed and assigned gender by partially living in the desired role or by adopting a gender role neither conventionally male nor conventionally female.

Associated Features Supporting Diagnosis

When visible signs of puberty develop, natal boys may shave their legs at the first signs of hair growth. They sometimes bind their genitals to make erections less visible. Girls may bind their breasts, walk with a stoop, or use loose sweaters to make breasts less visible. Increasingly, adolescents request, or may obtain without medical prescription and supervision, hormonal suppressors ("blockers") of gonadal steroids (e.g., gonadotropin-releasing hormone [GnRH] analog, spironolactone). Clinically referred adolescents often want hormone treatment and many also wish for gender reassignment surgery. Adolescents living in an accepting environment may openly express the desire to be and be treated as the experienced gender and dress partly or completely as the experienced gender, have a hairstyle typical of the experienced gender, preferentially seek friendships with peers of the other gender, and/or adopt a new first name consistent with the experienced gender. Older adolescents, when sexually active, usually do not show or allow partners to touch their sexual organs. For adults with an aversion toward their genitals, sexual activity is constrained by the preference that their genitals not be seen or touched by their partners. Some adults may seek hormone treatment (sometimes without medical prescription and supervision) and gender reassignment surgery. Others are satisfied with either hormone treatment or surgery alone.

Adolescents and adults with gender dysphoria before gender reassignment are at increased risk for suicidal ideation, suicide attempts, and suicides. After gender reassignment, adjustment may vary, and suicide risk may persist.

Prevaience

For natal adult males, prevalence ranges from 0.005% to 0.014%, and for natal females, from 0.002% to 0.003%. Since not all adults seeking hormone treatment and surgical reassignment attend specialty clinics, these rates are likely modest underestimates. Sex differences in rate of referrals to specialty clinics vary by age group. In children, sex ratios of natal boys to girls range from 2:1 to 4.5:1. In adolescents, the sex ratio is close to parity; in adults, the sex ratio favors natal males, with ratios ranging from 1:1 to 6.1:1. In two countries, the sex ratio appears to favor natal females (Japan: 2.2:1; Poland: 3.4:1).

Development and Course

Because expression of gender dysphoria varies with age, there are separate criteria sets for children versus adolescents and adults. Criteria for children are defined in a more con-

crete, behavioral manner than those for adolescents and adults. Many of the core criteria draw on well-documented behavioral gender differences between typically developing boys and girls. Young children are less likely than older children, adolescents, and adults to express extreme and persistent anatomic dysphoria. In adolescents and adults, incongruence between experienced gender and somatic sex is a central feature of the diagnosis. Factors related to distress and impairment also vary with age. A very young child may show signs of distress (e.g., intense crying) only when parents tell the child that he or she is "really" not a member of the other gender but only "desires" to be. Distress may not be manifest in social environments supportive of the child's desire to live in the role of the other gender and may emerge only if the desire is interfered with. In adolescents and adults, distress may manifest because of strong incongruence between experienced gender and somatic sex. Such distress may, however, be mitigated by supportive environments and knowledge that biomedical treatments exist to reduce incongruence. Impairment (e.g., school refusal, development of depression, anxiety, and substance abuse) may be a consequence of gender dysphoria.

Gender dysphoria without a disorder of sex development. For clinic-referred children, onset of cross-gender behaviors is usually between ages 2 and 4 years. This corresponds to the developmental time period in which most typically developing children begin expressing gendered behaviors and interests. For some preschool-age children, both pervasive cross-gender behaviors and the expressed desire to be the other gender may be present, or, more rarely, labeling oneself as a member of the other gender may occur. In some cases, the expressed desire to be the other gender appears later, usually at entry into elementary school. A small minority of children express discomfort with their sexual anatomy or will state the desire to have a sexual anatomy corresponding to the experienced gender ("anatomic dysphoria"). Expressions of anatomic dysphoria become more common as children with gender dysphoria approach and anticipate puberty.

Rates of persistence of gender dysphoria from childhood into adolescence or adulthood vary. In natal males, persistence has ranged from 2.2% to 30%. In natal females, persistence has ranged from 12% to 50%. Persistence of gender dysphoria is modestly correlated with dimensional measures of severity ascertained at the time of a childhood baseline assessment. In one sample of natal males, lower socioeconomic background was also modestly correlated with persistence. It is unclear if particular therapeutic approaches to gender dysphoria in children are related to rates of long-term persistence. Extant follow-up samples consisted of children receiving no formal therapeutic intervention or receiving therapeutic interventions of various types, ranging from active efforts to reduce gender dysphoria to a more neutral, "watchful waiting" approach. It is unclear if children "encouraged" or supported to live socially in the desired gender will show higher rates of persistence, since such children have not yet been followed longitudinally in a systematic manner. For both natal male and female children showing persistence, almost all are sexually attracted to individuals of their natal sex. For natal male children whose gender dysphoria does not persist, the majority are *androphilic* (sexually attracted to males) and often self-identify as gay or homosexual (ranging from 63% to 100%). In natal female children whose gender dysphoria does not persist, the percentage who are *gynephilic* (sexually attracted to females) and self-identify as lesbian is lower (ranging from 32% to 50%).

In both adolescent and adult natal males, there are two broad trajectories for development of gender dysphoria: early onset and late onset. *Early-onset gender dysphoria* starts in childhood and continues into adolescence and adulthood; or, there is an intermittent period in which the gender dysphoria desists and these individuals self-identify as gay or homosexual, followed by recurrence of gender dysphoria. *Late-onset gender dysphoria* occurs around puberty or much later in life. Some of these individuals report having had a desire to be of the other gender in childhood that was not expressed verbally to others. Others do not recall any signs of childhood gender dysphoria. For adolescent males with late-onset gender dysphoria, parents often report surprise because they did not see signs of gender

dysphoria during childhood. Expressions of anatomic dysphoria are more common and salient in adolescents and adults once secondary sex characteristics have developed.

Adolescent and adult natal males with early-onset gender dysphoria are almost always sexually attracted to men (androphilic). Adolescents and adults with late-onset gender dysphoria frequently engage in transvestic behavior with sexual excitement. The majority of these individuals are gynephilic or sexually attracted to other posttransition natal males with late-onset gender dysphoria. A substantial percentage of adult males with late-onset gender dysphoria cohabit with or are married to natal females. After gender transition, many self-identify as lesbian. Among adult natal males with gender dysphoria, the early-onset group seeks out clinical care for hormone treatment and reassignment surgery at an earlier age than does the late-onset group. The late-onset group may have more fluctuations in the degree of gender dysphoria and be more ambivalent about and less likely satisfied after gender reassignment surgery.

In both adolescent and adult natal females, the most common course is the early-onset form of gender dysphoria. The late-onset form is much less common in natal females compared with natal males. As in natal males with gender dysphoria, there may have been a period in which the gender dysphoria desisted and these individuals self-identified as lesbian; however, with recurrence of gender dysphoria, clinical consultation is sought, often with the desire for hormone treatment and reassignment surgery. Parents of natal adolescent females with the late-onset form also report surprise, as no signs of childhood gender dysphoria were evident. Expressions of anatomic dysphoria are much more common and salient in adolescents and adults than in children.

Adolescent and adult natal females with early-onset gender dysphoria are almost always gynephilic. Adolescents and adults with the late-onset form of gender dysphoria are usually androphilic and after gender transition self-identify as gay men. Natal females with the late-onset form do not have co-occurring transvestic behavior with sexual excitement.

Gender dysphoria in association with a disorder of sex development. Most individuals with a disorder of sex development who develop gender dysphoria have already come to medical attention at an early age. For many, starting at birth, issues of gender assignment were raised by physicians and parents. Moreover, as infertility is quite common for this group, physicians are more willing to perform cross-sex hormone treatments and genital surgery before adulthood.

Disorders of sex development in general are frequently associated with gender-atypical behavior starting in early childhood. However, in the majority of cases, this does not lead to gender dysphoria. As individuals with a disorder of sex development become aware of their medical history and condition, many experience uncertainty about their gender, as opposed to developing a firm conviction that they are another gender. However, most do not progress to gender transition. Gender dysphoria and gender transition may vary considerably as a function of a disorder of sex development, its severity, and assigned gender.

Risk and Prognostic Factors

Temperamental. For individuals with gender dysphoria without a disorder of sex development, atypical gender behavior among individuals with early-onset gender dysphoria develops in early preschool age, and it is possible that a high degree of atypicality makes the development of gender dysphoria and its persistence into adolescence and adulthood more likely.

Environmental. Among individuals with gender dysphoria without a disorder of sex development, males with gender dysphoria (in both childhood and adolescence) more commonly have older brothers than do males without the condition. Additional predisposing

factors under consideration, especially in individuals with late-onset gender dysphoria (adolescence, adulthood), include habitual fetishistic transvestism developing into autogynephilia (i.e., sexual arousal associated with the thought or image of oneself as a woman) and other forms of more general social, psychological, or developmental problems.

Genetic and physiological. For individuals with gender dysphoria without a disorder of sex development, some genetic contribution is suggested by evidence for (weak) familiality of transsexualism among nontwin siblings, increased concordance for transsexualism in monozygotic compared with dizygotic same-sex twins, and some degree of heritability of gender dysphoria. As to endocrine findings, no endogenous systemic abnormalities in sex-hormone levels have been found in 46,XY individuals, whereas there appear to be increased androgen levels (in the range found in hirsute women but far below normal male levels) in 46,XX individuals. Overall, current evidence is insufficient to label gender dysphoria without a disorder of sex development as a form of intersexuality limited to the central nervous system.

In gender dysphoria associated with a disorder of sex development, the likelihood of later gender dysphoria is increased if prenatal production and utilization (via receptor sensitivity) of androgens are grossly atypical relative to what is usually seen in individuals with the same assigned gender. Examples include 46,XY individuals with a history of normal male prenatal hormone milieu but inborn nonhormonal genital defects (as in cloacal bladder exstrophy or penile agenesis) and who have been assigned to the female gender. The likelihood of gender dysphoria is further enhanced by additional, prolonged, highly gender-atypical postnatal androgen exposure with somatic virilization as may occur in female-raised and noncastrated 46,XY individuals with 5-alpha reductase-2 deficiency or 17-beta-hydroxysteroid dehydrogenase-3 deficiency or in female-raised 46,XX individuals with classical congenital adrenal hyperplasia with prolonged periods of non-adherence to glucocorticoid replacement therapy. However, the prenatal androgen milieu is more closely related to gendered behavior than to gender identity. Many individuals with disorders of sex development and markedly gender-atypical behavior do not develop gender dysphoria. Thus, gender-atypical behavior by itself should not be interpreted as an indicator of current or future gender dysphoria. There appears to be a higher rate of gender dysphoria and patient-initiated gender change from assigned female to male than from assigned male to female in 46,XY individuals with a disorder of sex development.

Culture-Related Diagnostic Issues

Individuals with gender dysphoria have been reported across many countries and cultures. The equivalent of gender dysphoria has also been reported in individuals living in cultures with institutionalized gender categories other than male or female. It is unclear whether with these individuals the diagnostic criteria for gender dysphoria would be met.

Diagnostic Markers

Individuals with a somatic disorder of sex development show some correlation of final gender identity outcome with the degree of prenatal androgen production and utilization. However, the correlation is not robust enough for the biological factor, where ascertainable, to replace a detailed and comprehensive diagnostic interview evaluation for gender dysphoria.

Functional Consequences of Gender Dysphoria

Preoccupation with cross-gender wishes may develop at all ages after the first 2–3 years of childhood and often interfere with daily activities. In older children, failure to develop age-typical same-sex peer relationships and skills may lead to isolation from peer groups and to distress. Some children may refuse to attend school because of teasing and harass-

ment or pressure to dress in attire associated with their assigned sex. Also in adolescents and adults, preoccupation with cross-gender wishes often interferes with daily activities. Relationship difficulties, including sexual relationship problems, are common, and functioning at school or at work may be impaired. Gender dysphoria, along with atypical gender expression, is associated with high levels of stigmatization, discrimination, and victimization, leading to negative self-concept, increased rates of mental disorder comorbidity, school dropout, and economic marginalization, including unemployment, with attendant social and mental health risks, especially in individuals from resource-poor family backgrounds. In addition, these individuals' access to health services and mental health services may be impeded by structural barriers, such as institutional discomfort or inexperience in working with this patient population.

Differential Diagnosis

Nonconformity to gender roles. Gender dysphoria should be distinguished from simple nonconformity to stereotypical gender role behavior by the strong desire to be of another gender than the assigned one and by the extent and pervasiveness of gender-variant activities and interests. The diagnosis is not meant to merely describe nonconformity to stereotypical gender role behavior (e.g., "tomboyism" in girls, "girly-boy" behavior in boys, occasional cross-dressing in adult men). Given the increased openness of atypical gender expressions by individuals across the entire range of the transgender spectrum, it is important that the clinical diagnosis be limited to those individuals whose distress and impairment meet the specified criteria.

Transvestic disorder. Transvestic disorder occurs in heterosexual (or bisexual) adolescent and adult males (rarely in females) for whom cross-dressing behavior generates sexual excitement and causes distress and/or impairment without drawing their primary gender into question. It is occasionally accompanied by gender dysphoria. An individual with transvestic disorder who also has clinically significant gender dysphoria can be given both diagnoses. In many cases of late-onset gender dysphoria in gynephilic natal males, transvestic behavior with sexual excitement is a precursor.

Body dysmorphic disorder. An individual with body dysmorphic disorder focuses on the alteration or removal of a specific body part because it is perceived as abnormally formed, not because it represents a repudiated assigned gender. When an individual's presentation meets criteria for both gender dysphoria and body dysmorphic disorder, both diagnoses can be given. Individuals wishing to have a healthy limb amputated (termed by some *body integrity identity disorder*) because it makes them feel more "complete" usually do not wish to change gender, but rather desire to live as an amputee or a disabled person.

Schizophrenia and other psychotic disorders. In schizophrenia, there may rarely be delusions of belonging to some other gender. In the absence of psychotic symptoms, insistence by an individual with gender dysphoria that he or she is of some other gender is not considered a delusion. Schizophrenia (or other psychotic disorders) and gender dysphoria may co-occur.

Other clinical presentations. Some individuals with an emasculinization desire who develop an alternative, nonmale/nonfemale gender identity do have a presentation that meets criteria for gender dysphoria. However, some males seek castration and/or penectomy for aesthetic reasons or to remove psychological effects of androgens without changing male identity; in these cases, the criteria for gender dysphoria are not met.

Comorbidity

Clinically referred children with gender dysphoria show elevated levels of emotional and behavioral problems—most commonly, anxiety, disruptive and impulse-control, and de-

pressive disorders. In prepubertal children, increasing age is associated with having more behavioral or emotional problems; this is related to the increasing non-acceptance of gender-variant behavior by others. In older children, gender-variant behavior often leads to peer ostracism, which may lead to more behavioral problems. The prevalence of mental health problems differs among cultures; these differences may also be related to differences in attitudes toward gender variance in children. However, also in some non-Western cultures, anxiety has been found to be relatively common in individuals with gender dysphoria, even in cultures with accepting attitudes toward gender-variant behavior. Autism spectrum disorder is more prevalent in clinically referred children with gender dysphoria than in the general population. Clinically referred adolescents with gender dysphoria appear to have comorbid mental disorders, with anxiety and depressive disorders being the most common. As in children, autism spectrum disorder is more prevalent in clinically referred adolescents with gender dysphoria than in the general population. Clinically referred adults with gender dysphoria may have coexisting mental health problems, most commonly anxiety and depressive disorders.

Other Specified Gender Dysphoria

302.6 (F64.8)

This category applies to presentations in which symptoms characteristic of gender dysphoria that cause clinically significant distress or impairment in social, occupational, or other important areas of functioning predominate but do not meet the full criteria for gender dysphoria. The other specified gender dysphoria category is used in situations in which the clinician chooses to communicate the specific reason that the presentation does not meet the criteria for gender dysphoria. This is done by recording "other specified gender dysphoria" followed by the specific reason (e.g., "brief gender dysphoria").

An example of a presentation that can be specified using the "other specified" designation is the following:

The current disturbance meets symptom criteria for gender dysphoria, but the duration is iess than 6 months.

Unspecified Gender Dysphoria

302.6 (F64.9)

This category applies to presentations in which symptoms characteristic of gender dysphoria that cause clinically significant distress or impairment in social, occupational, or other important areas of functioning predominate but do not meet the full criteria for gender dysphoria. The unspecified gender dysphoria category is used in situations in which the clinician chooses *not* to specify the reason that the criteria are not met for gender dysphoria, and includes presentations in which there is insufficient information to make a more specific diagnosis.

EXHIBIT 5

WPATH
WORLD PROFESSIONAL ASSOCIATION FOR TRANSGENDER HEALTH

December 2, 2021

Dear All:

On behalf of the Standards of Care Version 8 Committee, we are pleased to present the DRAFT Version on the Standards of Care Version 8, now **available through Thursday, December 16, 2021, at 11:59pm eastern time**, for public comment.

Please note that this document is WPATH property and is being disseminated for public comment only, it is not to be copied or distributed. The final document will include an introduction, methodology section, several appendices, and supplementary information. More information is available on the SOC8 revision process on the WPATH website at **https://www.wpath.org/soc8.**

Your comments will be reviewed to shape the SOC8. Please note that all statements have been developed based on the available literature and clinical expertise. Once developed they have been approved by every member (120+) of the SOC8 (approval required 75% acceptance rate). While statements likely cannot be changed, there is more opportunity to make edits to the explications of the statements. Please include any comments to the draft of the SOC8 in general or to the statements and these will be carefully considered.

Please note that the titles of each chapter have not been finalised.

By clicking the links below, you will be taken to the survey for each chapter, within the preamble of each survey is the link to the draft version of that chapter. Return to this document to review other chapters and follow the same process.

Chapter Name	Survey Monkey Link
Adolescent	https://www.surveymonkey.com/r/85PD33R
Assessment	https://www.surveymonkey.com/r/LQL3528
Child	https://www.surveymonkey.com/r/RPSP59G
Education	https://www.surveymonkey.com/r/KWYYQSR
Epidemiology	https://www.surveymonkey.com/r/WH9Q2GR
Ethics	https://www.surveymonkey.com/r/5FV262F
Eunuch	https://www.surveymonkey.com/r/LK7T2MZ
Global	https://www.surveymonkey.com/r/KQZZHXL
Hormone Therapy	https://www.surveymonkey.com/r/LKSSGJZ
Institutions	https://www.surveymonkey.com/r/LLCTGHK
Intersex	https://www.surveymonkey.com/r/WYJB9L6
Mental Health	https://www.surveymonkey.com/r/6ZTH5VK
Nonbinary	https://www.surveymonkey.com/r/KFTQ9YM

Primary Care	https://www.surveymonkey.com/r/3LS8GJ9
Reproductive Health	https://www.surveymonkey.com/r/85NBN57
Sexual Health	https://www.surveymonkey.com/r/KF7PK9N
Surgery	https://www.surveymonkey.com/r/LSMPJRR
Terminology	https://www.surveymonkey.com/r/RBKLRWL
Voice	https://www.surveymonkey.com/r/5FWYJLF

It is very important to understand how your comments relate to specific statements, please be sure your comments relate to the statement. Of course, there is no need to make comments for every single chapter and statement.

Finally, it is important to know that reference style, grammatical and spelling issues will be corrected/reviewed as the last stage before publication by an independent editor, hence there is no need to add comments regarding grammar, spelling or related to reference style.

We look forward to receiving your comments and finishing the Standards of Care Version 8.

Please note that we may not be able to respond individually to each comment but will try our best to consider each comment carefully.

We look forward to reviewing your comments received by **Thursday, December 16, 2021, at 11:59pm eastern time.**

Kind regards,

Eli Coleman (Chair)
Asa Radix (Co-Chair)
Jon Arcelus (Co-Chair)

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SOC8 CHAPTERS

CHAPTERS

The Guideline Steering Committee, in discussion with chapter members, determined the chapters for inclusion in the Standards of Care, based on the previous editions of the SOC. Four new chapters were added. The chapters in the Standards of Care 8th Version are:

1. Global Applicability of the Standards of Care
2. Terminology – Diagnostic Criteria
3. Epidemiologic Considerations
4. ~~Overview of Therapeutic Approaches for Gender Health~~
5. Assessment, Support and Therapeutic Approaches for Children
6. Assessment, Support and Therapeutic Approaches for Adolescents with Gender Variance/Dysphoria **NEW**
7. Assessment for Adults with Gender Variance/Dysphoria
8. Assessment, Support and Therapeutic Approaches for Non-Binary Individuals **NEW**
9. Managing Mental and Behavioral Health Conditions in Adults
10. The Role of Primary Care in Gender Health
11. Hormone Therapy for Adolescents and Adults
12. Sexual Health Across the Lifespan **NEW**
13. Reproductive Health for Adolescents and Adults
14. Voice and Communication Therapy
15. Surgery for Adolescents and Adults and Postoperative Care and Follow-Up
16. Applicability of the Standards of Care to People Living in Institutional Environments
17. Applicability of the Standards of Care to People with Intersex Conditions
18. Applicability of the Standards of Care to Eunuchs **NEW**
19. Competency, Training, Education **NEW**
20. Ethics **NEW**

https://www.wpath.org/soc8/chapters

Global

Introduction

Transgender and gender diverse people are a highly diverse population (both in terms of their identities and healthcare needs) and many experience stigma and consequent marginalisation throughout their lives. Seen from a global perspective, violence against transgender and gender diverse people is widespread, diverse in nature (emotional, sexual and physical), and involves a range of perpetrators (including State actors). Worldwide, statistics on murder are alarming, with over 4000 documented killings between January 2008 and September 2021; a number widely regarded as under-reported (TGEU, 2020). Experiences such as these (and the anticipation or fear of encountering such experiences) lead to what Meyer has described as minority stress (Meyer, 2003), and are associated with poor health outcomes; both physical (e.g., Rich et al, 2020) and psychological (e.g., Scandurra et al, 2017; Shipherd et al, 2019, Tan et al, 2021).

Since the publication of the Standards of Care, Version 7 (SOC-7) there have been dramatic changes in perspectives on transgender and gender diverse people and their healthcare. Mainstream global medicine no longer classifies transgender and gender diverse identities as a mental disorder. In the Diagnostic and Statistical Manual, Version 5 (DSM-5) from the American Psychiatric Association (APA, 2013), the diagnosis of *Gender Dysphoria* focuses on any distress and discomfort that accompanies being transgender and gender diverse, rather than on the gender identity itself. In the International Classification of Diseases, Version 11 (ICD-11), the diagnostic manual of the World Health Organisation (WHO, 2019b), the *Gender Incongruence* diagnosis is placed in a chapter on sexual health, and focuses on the person's experienced identity, and any desire for gender affirming treatment that might stem from that identity. Such developments, involving a depathologisation (or more precisely a de-psychopathologisation) of transgender identities, are fundamentally important on a number of grounds. In the field of healthcare, they may have helped support a care model emphasising patients' active participation in decision-making about their own healthcare, supported by primary healthcare professionals (Baleige et al, 2021). It is reasonable to suppose that these developments may also promote more socially inclusive policies, including legislative reform in gender recognition facilitating a rights-based approach without imposing requirements for diagnosis, hormone therapy and/or surgery. Such developments may contribute greatly to the overall health and wellbeing of transgender and gender diverse people (Aristegui et al, 2017).

Previous editions of the SOC have revealed that much of the recorded clinical experience and knowledge in this area is derived from North American and Western European sources. They have focused on gender-affirming healthcare in high income countries enjoying relatively well-resourced healthcare systems (including with trained mental health providers, endocrinologists, surgeons and other specialists), where services are often funded publicly or (at least for some patients) by way of private insurance. For many countries such healthcare provision is aspirational. Few if any health professionals (primary or specialist) may exist, and even fewer may be competent to work with transgender and gender diverse people. Psychological, hormonal, and surgical healthcare may not be available and training options limited (e.g. Martins et al, 2020). Funding for gender-affirming healthcare may be absent and patients often bear the full costs of whatever healthcare they access.

Accessing gender-affirming healthcare options for this population can also be challenging. Across much of the world resourcing in this area is non-existent or limited. Healthcare is often unavailable, inappropriate, difficult to access and/or unaffordable. Healthcare providers

often lack clinical and/or cultural competence, or opportunities for training. As already noted, mainstream 'Western' medicine historically viewed transgender and gender diverse people as mentally disordered; a perspective that has only recently changed. For all these reasons, transgender and gender diverse people have commonly found themselves disempowered as consumers of whatever healthcare is available. Healthcare providers have found that the relevant literature is largely North American and European, presenting particular challenges for persons working in healthcare systems that are even less well resourced. Recent initiatives, often involving transgender and gender diverse stakeholders as partners, are changing this situation somewhat, providing a body of knowledge about how to provide effective transgender and gender diverse healthcare in low- and middle-income countries outside the Global North.

Within the field a wide range of valuable healthcare resources have been developed in recent years. Dahlen et al (2021) review clinical guidelines intended to be international in scope; over half those reviewed originate from professional bodies based in North America (e.g., Hembree et al, 2017) or Europe (e.g., T'Sjoen et al, 2020). These have informed numerous healthcare resources including those developed for global use (WHO 2014; UNDP et al, 2016), and for use in specific countries or regions outside North America and Europe. Regional examples can be found in Asia and the Pacific (Health Policy Project et al, 2015, APTN, 2021), the Caribbean (PAHO, 2014), Thailand (Center for Excellence in Transgender Health, 2021a,b), Australia (Telfer et al, 2020) and Aotearoa New Zealand (Oliphant et al, 2018), and are commonly created through the initiatives of, or in partnership with, transgender and gender diverse communities locally or internationally. These resources may be of particular value to those planning, organising and delivering services, including in low-income, low-resource countries of the Global South. There are likely to be other resources published in languages other than English of which we are unaware.

Globally, transgender and gender diverse identities may be associated with differing conceptual frameworks of sex, gender and sexuality, and exist in widely diverse cultural contexts and histories. Considering the complex relationships between social and cultural factors, the law, and the demand for and provisions of gender-affirming healthcare, the SOC-8 should be interpreted through a lens that is appropriate for and within the context of each health professional's individual practice while maintaining alignment to the core principles that underscore it (APTN and UNDP, 2012; PAHO, 2014; Health Policy Project et al, 2015).
It is in this context, and by drawing broadly on the experiences of transgender and gender diverse people and healthcare providers internationally, that we consider the global applicability of SOC-8 within this chapter. We set out key considerations for health professionals and conclude by recommending core principles and practices fundamental to contemporary healthcare for transgender and gender diverse people, regardless of where they live or the resources available to those who seek to provide such healthcare.

Summary of Recommendations

Statement 1: We recommend that health professionals and other users of the Standards of Care, Version 8 (SOC-8) should apply the recommendations in ways that meet the needs of local transgender and gender diverse communities, by being sensitive to the cultures they work with and the realities of the countries they are practising in.

Statement 2: We recommend that healthcare providers understand the impact of social attitudes, laws, economic circumstances and health systems on the lived experiences of transgender and gender diverse people worldwide.

Statement 3: We recommend that translations of the SOC focus on cross-cultural, conceptual and literal equivalence to ensure alignment with the core principles that underpin the SOC-8.

Statement 4: We recommend that health professionals and policymakers always apply the SOC-8 core principles to their work with transgender and gender diverse people to ensure respect for human rights and access to appropriate and competent healthcare, including:

General principles

- Be empowering and inclusive. Work to reduce stigma and facilitate access to appropriate healthcare, for all who seek it;
- Respect diversity. Respect all clients, and all gender identities. Do not pathologize differences in gender identity or expression;
- Respect universal human rights including the right to bodily and mental integrity, autonomy and self-determination; freedom from discrimination and the right to the highest attainable standard of health.

Principles around developing and implementing appropriate services and accessible healthcare

- Involve transgender and gender diverse people in the development and implementation of services;
- Become aware of social, cultural, economic and legal factors that might impact the health (and healthcare needs) of your client, as well as the willingness and capacity of the person to access your services;
- Provide healthcare (or refer to knowledgeable colleagues) that affirms clients' gender identities and expressions, including healthcare that reduces the distress of gender dysphoria or incongruence (if this is present);
- Reject approaches that have the goal or effect of conversion, and avoid providing any direct or indirect support for such approaches or services

Principles around delivering competent services

- Become knowledgeable (get training, where possible) about the healthcare needs of transgender and gender diverse people, including the benefits and risks of gender-affirming care;
- Match the treatment approach to the specific needs of clients, particularly their goals for gender identity and expression;
- Focus on promoting health and wellbeing rather than solely the reduction of gender dysphoria or incongruence, which may or may not be present;
- Commit to harm reduction approaches where appropriate;
- Enable the full and ongoing informed participation of transgender and gender diverse people in decisions about their health and wellbeing;
- Improve experiences of health services including administrative systems and via continuity of care.

Principles around working towards improved health through wider community approaches

- Put people in touch with communities and peer support networks;
- Support and advocate for clients within their families and communities (schools, workplaces, and other settings) where appropriate.

Parent(s)/caregiver(s) may provide key information for the clinical team, including report on the young person's gender and overall developmental, medical, and mental health history as well as information about the young person's level of current support and general functioning and wellbeing. Concordance or divergence of report between the adolescent and their parent(s)/caregiver(s) may be important information for the assessment team, including for the designing and shaping of individualized youth and family supports (De Los Reyes et al., 2019; Katz-Wise et al., 2017). Knowledge of the family context, including resilience factors and challenges can help providers know where special supports would be needed during the medical treatment process. Engagement of parent(s)/caregiver(s) is also important for educating families around various treatment approaches, ongoing follow-up and care needs, and potential treatment complications. Through psychoeducation regarding clinical gender care options and participation in the assessment process, which may unfold over time, parent(s)/caregiver(s) may better understand their adolescent child's gender-related experience and needs (Andrzejewski et al., 2020; Katz-Wise et al., 2017).

Parent/caregiver concerns or questions regarding the stability of gender-related needs over time and implications of various gender affirming interventions are common, and should not be dismissed. It is appropriate for parent(s)/caregiver(s) to ask these questions, and there are cases in which the parent(s)/caregiver(s)' questions or concerns are particularly helpful in informing treatment decisions and plans. For example, parent/caregiver report may provide critical context in situations in which a young person experiences very recent and/or sudden self-awareness of gender diversity and a corresponding gender treatment request, or when there is concern for possible excessive peer and/or social media influence on a young person's current self-gender concept. Contextualization of parent/caregiver report is also critical, as the report of a young person's gender history as provided by parent(s)/caregiver(s) may or may not align with the young person's self-report. Gender histories may be unknown to parent(s)/caregiver(s) because gender may be an inward experience for youth, not known by others unless it is discussed.

Some parents may present with unsupportive or antagonistic beliefs about T/GD identities and/or clinical gender care (Clark et al., 2020). Such parent perspectives may in some cases seem rigid, but providers should not assume this is the case. There are many examples of parent(s)/caregiver(s) who, over time with support and psychoeducation, have become increasingly accepting of their T/GD's child's gender diversity and care needs. Helping youth and parent(s)/caregiver(s) to work together on important gender care decisions is a primary goal. However, in some cases, parent(s)/caregiver(s) may be too rejecting of their adolescent child and their child's gender needs to be part of the clinical evaluation process. In these situations, youth may require the engagement of larger systems of advocacy and support to move forward with necessary supports and care (Dubin et al., 2020).

Statement 12:
We recommend that health professionals assessing trans and gender diverse adolescents should only recommend gender affirming medical or surgical treatments requested by the patient when:

Statement 12A:
The adolescent meets the diagnostic criteria of gender incongruence as per the ICD-11 where a diagnosis is necessary to access health care. In countries which have not implemented the latest ICD other taxonomies may be used but efforts should be undertaken to utilize the latest ICD as soon as is practicably possible.

When working with transgender and gender diverse adolescents, health professionals should realize that a classification may give access to care, but pathologizing transgender identities may be experienced as stigmatizing (van Beek et al., 2016). Assessments related to gender health and gender diversity have been criticized, and controversies exist around classification systems (Drescher, 2016). Healthcare professionals should realize they do not diagnose a gender identity per se, as one's gender identity is the subjective experience of being male or female or another gender. Health professionals should assess the overall and gender-related history and transgender care related needs of youth. Through this assessment process, health care providers may provide a classification when needed to get access to transgender-related care. However, a classification involving gender diversity connotes no pathology, in and of itself.

Gender Incongruence and Gender Dysphoria are the two diagnostic terms used in respectively the World Health Organization's International Classification of Diseases (ICD) and the American Psychiatric Association's Diagnostic and Statistical Manual of Mental Disorders (DSM). Of these two widely used classification systems, the DSM is for psychiatric classifications only and the ICD contains all diseases and conditions related to physical as well as mental health. The most recent versions of these two systems, the DSM-5 and the ICD-11 respectively, reflect a long history of reconceptualizing and depsychopathologizing gender related diagnoses (American Psychiatric Association, 2013, World Health Organization, 2019). Compared to the earlier version, the DSM-5 replaced Gender Identity *Disorder* with Gender *Dysphoria* acknowledging the distress experienced by *some* people stemming from the incongruence between experienced gender identity and sex assigned at birth. Compared to the ICD 10th edition, the Gender Incongruence classification was moved from the Mental Health Chapter to a Chapter "Conditions related to Sexual Health" in the ICD-11. One important reconceptualization in comparison to the DSM-5 Gender Dysphoria classification is that distress is not a required indicator of the ICD-11 Gender Incongruence classification (WHO, 2019). After all, when growing up in a supporting and accepting environment, the distress and impairment criterion, an inherent part of every mental health condition, may not be applicable (Drescher, 2012). As such, the ICD-11 Gender Incongruence classification may better capture the fullness of gender diversity experiences and related clinical gender needs.

Criteria of the ICD-11 classification "*Gender Incongruence of Adolescence or Adulthood"* require a marked and persistent incongruence between an individual´s experienced gender and the assigned sex which often leads to a desire to 'transition,' in order to live and be accepted as a person of the experienced gender. For some, this includes hormonal treatment, surgery, or other health care services to make the individual´s body align as much as desired, and to the extent possible, with the person's experienced gender. Relevant for adolescents is the indicator that a classification cannot be assigned '*prior to the onset of puberty'.* Finally, it is noted "*that gender variant behaviour and preferences alone are not a basis for assigning the classification*" (WHO, ICD-11, 2019).

Criteria for the DSM-5 classification "*Gender Dysphoria in Adolescence and Adulthood"* denote 'a marked incongruence between one's experienced/expressed gender and assigned gender, of at least 6 months' duration' (criterion A, fulfilled when 2 of 6 subcriteria are manifest), associated with 'clinically significant distress or impairment in social, occupational, or other important areas of functioning' (Criterion B, APA 2013). As noted before, not all transgender and gender diverse people experience gender dysphoria and this should not preclude them from accessing medical affirming care. For adolescents, the DSM-5 makes two specific remarks, which make it possible to give the classification when secondary sex characteristics have yet to fully develop. First, there should be a marked incongruence between one's experienced/expressed gender and

one's primary and/or secondary sex characteristics *(or in younger adolescents, the anticipated secondary sex characteristics)*. Second, the strong desire to be rid of one's primary and/or secondary sex characteristics because of a marked incongruence with one's experienced/expressed gender (*or in younger adolescents, a desire to prevent the anticipated secondary sex characteristics*).

Of note, a gender related classification is one of the requirements for medical gender affirming care, but such a classification solely does not *indicate* a person *needs* medical affirming care. The range of youth experiences of gender incongruence necessitates professionals provide a range of treatments or interventions based on the individual's needs. Counseling, gender exploration and mental health assessment, and when needed, treatment with mental health providers trained in gender development may all be indicated with or without medical affirming care.

Statement 12B:
There is well-documented (according to local context) evidence of persistent gender incongruence or gender nonconformity / diversity of several years.

Identity exploration and consolidation are experienced by many adolescents (Klimstra et al., 2010; Topolewska-Siedzik & Cieciuch, 2018). Identity exploration during the teen years may include exploration of gender and gender identity (Steensma et al., 2013). Little is known about how processes of adolescent identity consolidation (e.g., the process of commitment to specific identities) may impact a young person's experience(s) of gender. Given potential shifts in gender-related experiences and needs during adolescence, as discussed below, it is important to establish that the young person has experienced several years of persistent gender incongruence or gender diversity prior to initiating gender-affirming hormones or providing gender-affirming surgeries. Establishing evidence of persistent gender incongruence or gender diversity typically requires careful assessment with the young person over time (see Statement 3). Whenever possible and appropriate, the assessment and discernment process should also include the parent(s)/caregiver(s) (see Statement 1). The documentation to demonstrate well documented gender diversity can be provided via history obtained directly from the adolescents and parents/cargivers when this is not documented in the medical records.

The research literature on continuity versus discontinuity of gender affirming medical care needs/requests is complex and somewhat difficult to interpret. A series of studies conducted over the last several decades, including some with methodological challenges (as noted by Temple Newhook et al., 2018; Winters et al., 2018), suggest that gender diversity is not consistent for all children as they progress into adolescence: A subset of youth who experienced gender diversity prior to puberty show reduced (or fully discontinued) gender diversity over time (de Vries et al., 2010; Ristori & Steensma, 2016; Singh et al., 2021, Wagner et al., 2021). However, there has been less research focus on rates of continuity and discontinuity of gender diversity and gender-related needs in pubertal and/or adolescent populations. The data available regarding broad *unselected* gender-referred pubertal/adolescent cohorts (from the Amsterdam transgender clinic) suggest that, following extended assessments over time, a subset of gender diverse adolescents presenting for gender care elect not to pursue gender-affirming medical care (Arnoldussen et al., 2019; de Vries et al., 2011). Importantly, findings from studies of gender diverse pubertal/adolescent cohorts who have undergone comprehensive gender evaluation over time, shown persistent gender diversity and gender-related need, and received resulting referrals for medical gender care, suggest very low levels of regret regarding gender-related medical care decisions (de Vries et al., 2014; Wiepjes

EXHIBIT
7

Updated recommendations for hormone therapy in gender dysphoria in young people

Published: 22/02/2022 at 10:00

Share

The National Board of Health and Welfare today publishes new recommendations regarding hormone therapy of young people under the age of 18 with gender dysphoria. Uncertain science and newly acquired knowledge mean that the National Board of Health and Welfare now recommends restraint when it comes to hormone therapy. At the same time, it is important that children and young people suffering from gender dysphoria are taken seriously, well treated and offered adequate care measures.

Gender dysphoria means that you have a psychological suffering or a impaired ability to function in everyday life, which is caused by gender identity not being consistent with the registered sex. The National Board of Health and Welfare is updating knowledge support for gender dysphoria care for young people and today new recommendations are presented regarding puberty-inhibiting treatment and gender-affirming hormone therapy for young people.

The National Board of Health and Welfare has previously presented statistics showing that the group of young people seeking care for gender dysphoria has increased sharply. Between 2008 and 2018, the number of new cases of diagnosed gender dysphoria multiplied. Particularly large was the increase among those aged 13 to 17 years and with registered sex female at birth.

"The change is greater among young people than older people, and greater within the group with registered sex female than male at birth. Several factors have been put forward as explanations, but it has not been possible to clarify what causes are behind it. As a result, the changes represent an uncertainty that we have had to take into account when it comes to what care should be recommended for minors," says Thomas Lindén, Head of Department at the National Board of Health and Welfare.

Lack of firm conclusions about the efficacy and safety of treatments

At the request of the National Board of Health and Welfare, SBU has produced a literature review that has reviewed all relevant studies on the efficacy and safety of hormone treatments. The report, which is published today, shows that it is not yet possible to draw any firm conclusions about the efficacy and safety of the treatments based on scientific evidence.

"The conclusion is that very little knowledge has been gained about the effects and safety of treatments since 2015," says Thomas Lindén.

- When the knowledge support for the care of children and adolescents with gender dysphoria was developed in 2015, it was stressed the importance of the measures offered in the framework of the clinical work being systematically followed up and evaluated in the best possible way. Now we see that this has not yet been realised, which contributes to the reason for changing the recommendations.

SBU has also compiled studies on changing perceptions of gender identity or interruption of treatment. It is not possible to determine how common it is for people who undergo gender affirming treatment to later change their perception of their gender identity, discontinue treatment or in any aspect regret it. At the same time, it is documented that detransition occurs, and there may also be a dark number.

"For the group that regrets or cancels a initiated treatment, there may be a risk that the treatment has led to poorer health or quality of life," says Thomas Lindén.

The risks outweigh the benefits at present

Based on the results that have emerged, the National Board of Health and Welfare's overall conclusion is that the risks of puberty-inhibiting and gender-affirming hormone therapy for those under the age of 18 currently outweigh the possible benefit for the group as a whole.

"The assessment is that treatment with hormones should continue to be given within the framework of research. Increased knowledge is needed, among other things, about the impact of treatments on gender dysphoria, as well as the mental health and quality of life of minors, in both the short and long term," says Thomas Lindén.

"Pending the completion of a research study, our assessment is that the treatments can be given in exceptional cases. Here we propose a number of criteria that care can be based on in the individual clinical assessments.

At the same time, it is important that young people with gender dysphoria continue to receive care and treatment in the healthcare sector. These include both hormonal treatments where they are deemed justified and, for example, psychosocial interventions, child psychiatric treatment and suicide prevention measures when needed.

"Healthcare needs to continue to ensure that children and young people suffering from gender dysphoria are taken seriously, well treated and offered adequate care measures. In the future, this care will become national highly specialized care, and this will increase the opportunities for research and knowledge development in this area of care as well as for further strengthened patient safety and quality," says Thomas Lindén.

Facts

- The National Board of Health and Welfare has an ongoing work to update knowledge support for children and young people with gender dysphoria/gender incongruence.
- The work is carried out in stages and is done on behalf of the Government. Previously, chapters on support and investigation have been published.
- The update is made to weigh in on new knowledge and the changes in the field of care that have taken place since the knowledge support was published in 2015, and to make recommendations for good care based on today's conditions.

Support, investigation and hormone therapy in sex incongruence in children and adolescents

Support, investigation and hormone therapy in gender incongruence in children and adolescents – Partial update of knowledge support, February 2022

Part number: 2022-2-7774

Published: 22/02/2022

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Journal of Adolescent Health ■■ (2017) ■■–■■

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JOURNAL OF ADOLESCENT HEALTH
www.jahonline.org

Original Article

Physiologic Response to Gender-Affirming Hormones Among Transgender Youth

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Article history: Received May 15, 2017; Accepted August 4, 2017
Keywords: Transgender; Transgender youth; Gender transition; Cross-sex hormones; Gender dysphoria

A B S T R A C T

Purpose: The purpose of this study was to examine the physiologic impact of hormones on youth with gender dysphoria. These data represent follow-up data in youth ages 12–23 years over a two-year time period of hormone administration.
Methods: This prospective, longitudinal study initially enrolled 101 youth with gender dysphoria at baseline from those presenting consecutively for care between February 2011 and June 2013. Physiologic data at baseline and follow-up were abstracted from medical charts. Data were analyzed by descriptive statistics.
Results: Of the initial 101 participants, 59 youth had follow-up physiologic data collected between 21 and 31 months after initiation of hormones available for analysis. Metabolic parameters changes were not clinically significant, with the exception of sex steroid levels, intended to be the target of intervention.
Conclusions: Although the impact of hormones on some historically concerning physiologic parameters, including lipids, potassium, hemoglobin, and prolactin, were statistically significant, clinical significance was not observed. Hormone levels physiologically concordant with gender of identity were achieved with feminizing and masculinizing medication regimens. Extensive and frequent laboratory examination in transgender adolescents may be unnecessary. The use of hormones in transgender youth appears to be safe over a treatment course of approximately two years.
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IMPLICATIONS AND CONTRIBUTIONS

This prospective, observational data suggest the safety of cross-sex hormone use among youth with gender dysphoria over a treatment course of at least two years. The findings reported here may help improve provider and caregiver comfort moving forward with hormonal therapy for minors and young adults with gender dysphoria.

Over the past seven years, there has been a significant increase in the number of youth presenting for care related to gender dysphoria in gender-specific clinics, and in primary care settings [1–3]. Gender dysphoria is widely understood as the persistent distress that arises as the result of an incongruence between one's assigned sex at birth (male or female) and one's experienced gender (male, female, both, or neither). Many youth with gender dysphoria seek medical intervention (pharmacological and/or surgical) to bring their phenotypic presentation into closer alignment with their gender of identity. Because there is a paucity of data related to the impact of gender-affirming hormones in youth, providers, caretakers, and community members experience trepidation about the safety and efficacy of their use in transgender adolescents. This article describes the physiologic impact of gender-affirming hormones among adolescents seeking phenotypic gender transition after approximately two years of gender-affirming hormone treatment.

Financial Disclosure: All authors have indicated they have no financial relationships relevant to this article to disclose.
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https://doi.org/10.1016/j.jadohealth.2017.08.005

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Background

Although the presence of primary sex characteristics is often described as a source of distress for transgender youth, the development of "incorrect" secondary sex characteristics during endogenous puberty is the cause of great suffering for many. Some youth describe feeling confused by sexually incongruent development, some feel betrayed by their bodies, and many experience heightened levels of anxiety, depression, and sometimes suicidal thoughts and attempts [3]. The past decade has improved access to care for many youth, but the scarcity of skilled and knowledgeable medical providers has continued to make access to care challenging for transgender youth throughout the country. The dearth of available data regarding medical protocols and outcomes contributes to a lack of continuity about hormone administration, timing, doses, and expected response. Dutch investigation indicates that hormone treatment in adolescence followed by gender confirmation surgery is effective in mitigating gender dysphoria [4]. Despite these findings, there remains on ongoing concern among providers and parents about the safety of hormone use among youth with gender dysphoria.

In 2006, the use of gonadotropin-releasing hormone (GnRH) analogs for suppression of endogenous puberty was introduced by a team of professionals in the Netherlands, and significantly altered the landscape of transgender youth care [5]. For those youth who present early enough for care, having endogenous puberty placed on hold allows them an opportunity to explore gender, learn more about exogenous hormones, and get a deeper understanding of the challenges of navigating the world as an individual of transgender experience [6]. However, most youth are presenting for care well into, or even beyond their endogenous pubertal development. Most youth presenting well into endogenous puberty desire hormones to bring their bodies into closer alignment with their gender.

Feminizing hormones

The use of gender-affirming hormones in transgender adults for the purpose of phenotypic gender transition is well documented, and demonstrates both efficacy and safety [7,8]. Weinand and Safer reviewed published longitudinal data examining the impact of hormones in transgender adults. Among transfeminine adults (those assigned male at birth who identify somewhere along the feminine spectrum) who were administered feminizing hormones for phenotypic changes, a small risk of venous thrombolytic events ranging between 1% [8] and 8% [9] was reported depending on the type of estrogen used in the treatment protocol and other existing risk factors (smoking, proximity to surgery, hypercoagulable states). Other cardiovascular events reported were also rare. Among these studies, findings consistently describe no increased cancer risk for transfeminine individuals undergoing hormone therapy. Changes in physiologic measures for transfeminine individuals include mixed results about changes in lipid profile [10]. Liver function tests and hematocrit did not change with feminizing hormone use [10,11]. An increase in prolactin levels, enlarged pituitary glands, and prolactinomas (six cases) has been reported with feminizing hormone therapy [7,12]. Increased mortality among the transfeminine population is a result of AIDS-related complications, suicide, substance abuse, and cardiovascular disease [13].

Masculinizing hormones

Among adult transmasculine individuals (those assigned female at birth who identify somewhere along the masculine spectrum of gender) who underwent hormone therapy with testosterone, there was no reported increase in cardiovascular disease, cancer, or reproductive tract sequelae. Hemoglobin and hematocrit increased in those taking testosterone, but remained within normal male range. Increased insulin resistance and fasting glucose were both noted as sequelae for transmasculine individuals taking hormones. A higher rate of polycystic ovary syndrome diagnoses among pre-hormone transmasculine individuals may contribute to some of these findings in that cohort [14]. Finally, there has been no increase in mortality noted for transmasculine individuals taking testosterone. Overall, the authors concluded that gender-affirming hormone therapy is safe, with careful monitoring for potential complicating factors [7].

Present study

Little has been reported about transgender adolescents and their response to hormone therapy. A recent retrospective article by Jarin et al. reported the minimal impact of hormone treatment on 116 adolescents aged 14–25 years with gender dysphoria who were treated over time. Jarin et al. demonstrated that among adolescents treated for a period of 1–6+ months, the only findings were an increase in hemoglobin, hematocrit, and body mass index, and a lowering of high-density lipoprotein levels in those using testosterone for masculinization. Among those using estrogen for feminization, lower testosterone and alanine aminotransferase (ALT) were reported [15]. These findings are consistent with data from adults undergoing phenotypic gender transition with exogenous hormones, and indicate short-term safety of hormone use. The results from this retrospective study are useful for helping to allay some of the concerns that providers and parents have about safety. This study is limited by its retrospective design, and the challenges faced by many trying to collect data from adolescents—sporadic follow-up, frequent relocation, and inconsistent medication adherence.

The Center for Transyouth Health and Development at Children's Hospital in Los Angeles is the largest clinic dedicated to the care of transgender and gender non-conforming children, adolescents, and young adults in the United States. The center currently provides services for approximately 925 youth between the ages of 3 and 25 years, and provides multidisciplinary care that includes mental health, medical intervention, advocacy, and referral resources for transgender and gender non-conforming youth and their families. The center has been providing gender-affirming hormones for adolescents and young adults since 1993. Growth of the clinic numbers has been unprecedented, and demand has far outpaced capacity.

This article offers preliminary results from a prospective study examining the physiologic impact of gender-affirming hormones in a cohort of adolescents aged 12–24 years with gender dysphoria over approximately two years of hormone use.

Methods

Self-identified transgender youth between the ages of 12 and 24 years presenting consecutively for care at the center

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between February 2011 and June 2013 were screened for participation in this prospective study. Eligibility criteria for the study included age between 12 and 24 years old, self-identification of an internal gender identity different from the sex assigned at birth, presence of gender dysphoria, a desire to undergo phenotypic gender transition, naivety to cross-sex hormones or less than three months of previous hormone use, and ability to read and comprehend English. Participants under the age of 18 required consent from their legal guardians to participate in the study.

Demographic data were collected via computer-assisted survey at baseline after participants were screened and consented. Baseline and follow-up physiologic data were abstracted from the medical charts of the participants. One hundred one participants were evaluated for physiologic parameters at baseline. Follow-up physiologic data were available for 59 youth (34 transmasculine and 25 transfeminine participants). The Committee on Clinical Investigations at Children's Hospital Los Angeles approved this study. This study was supported by the Saban Research Institute at Children's Hospital Los Angeles, The National Center for Research Resources, and the National Center for Advancing Translational Sciences, National Institutes of Health, and by the Eunice Kennedy Shriver National Institute of Child Health and Human Development.

Hormone regimens

Transfeminine youth were started on hormone protocols that usually included a testosterone blocking agent and feminizing medications. Spironolactone (100–200 mg orally per day) or a GnRH analog was used for testosterone blocking and induction of feminizing features with 17 β estradiol, and in some cases, the addition of progesterone. At the time of this study, spironolactone and GnRH analogs were not covered by most insurance plans; therefore, seven (28%) of these youth did not have their endogenous testosterone blocked specifically in the first two years of treatment. One transfeminine young person was on GnRH analogs since early puberty. Eighteen participants used an escalating dose of oral estradiol ranging from 1 to 6 mg each day; four switched to injectable estradiol over the course of treatment, and one was off of hormones at the follow-up visit. Six participants initially started, and continued using injectable estradiol at doses ranging from 20 to 30 mg delivered intramuscularly every 14 days.

Transmasculine youth were all treated with testosterone cypionate via subcutaneous delivery at escalating doses ranging from 12.5 mg to 75 mg weekly. At follow-up, most youth were at a dose of 50–75 mg weekly. Two transmasculine youth were on simultaneous GnRH analogs that were started earlier in adolescence. Doses for both cohorts were adjusted based on clinical response and circulating levels of sex steroids.

Statistical analysis

Descriptive statistics were used to report baseline characteristics. Categorical variables were summarized using frequencies and percentages. Paired samples *t* tests were used to determine whether there were statistically significant differences in physiologic parameters measured before initiation of hormone therapy and at two-year follow-up.

Results

Demographic information

Physiologic data for 59 youth (25 transfeminine participants and 34 transmasculine participants) were available for follow-up comparison at 21–31 months following the initiation of exogenous hormones for phenotypic transition. Twenty-five (42%) of the participants were assigned male at birth and identified somewhere along the female gender spectrum (transfeminine), and 34 (58%) were assigned female at birth and identified somewhere along the masculine gender spectrum (transmasculine). Youth ranged in age from 12 to 23 years at initiation of therapy, with a mean age of 18 years. Thirteen (22%) youth started hormones younger than age 16 years. More than half (55%) of the participants identified as Caucasian, 29% Latino/a, 10% African-American, 2% Asian Pacific Islander, and 3% identified as other ethnicity. Almost all (91%) of the transmasculine participants identified their gender as male, 6% as gender queer, 3% bi-gender, and none as female. Of the transfeminine youth, almost all identified their gender as female (91%), one youth identified as a gender bender, and one as "other" (Table 1).

Physiologic parameters (transfeminine youth)

Transfeminine youth showed statistically significant changes in high-density lipoprotein (HDL), aspartate aminotransferase, potassium, prolactin, and hemoglobin at follow-up. Although these metabolic parameters were statistically significant, they were not clinically significant, and did not present a safety concern. A decrease in hemoglobin is an expected physiologic response to the lowering of testosterone, which stimulates the production of erythropoietin and subsequently hemoglobin. Only one participant dropped into borderline anemia, all others were within the normal range for cisgender females. An increase in the mean HDL, although statistically significant, lacks clinical significance because it is within the normal range for cisgender females. An overall increase in prolactin levels is an expected change related to increased estrogen levels [16]. Physiologic parameters including blood pressure and glucose were within normal clinical range for most of the participants. Additionally, prolactin levels statistically, but not clinically, increased with hormone use. Blood pressure and lean body mass index at follow-up did not change.

Table 1
Descriptive characteristics (n = 58 participants' survey results available for analyses)

	Transfeminine youth (n = 23) n (%)	Transmasculine youth (n = 35) n (%)	Total (n = 58) n (%)
Race/Ethnicity			
African-American/black	2 (9)	4 (11.4)	6 (10)
Asian/Pacific Islander	0 (0)	1 (3)	1 (2)
Caucasian	10 (43)	22 (63)	32 (55)
Latino(a)	10 (43)	7 (20)	17 (29)
Other	1 (4)	1 (3)	2 (3)
Gender identity			
Male	0 (0)	32 (91)	32 (55)
Female	21 (91)	0 (0)	21 (36)
Gender queer	0 (0)	2 (6)	2 (3)
Gender bender	1 (4)	0 (0)	1 (2)
Bi-gendered	0 (0)	1 (3)	1 (2)
Other	1 (4)	0 (0)	1 (2)

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Table 2
Physiologic parameters of transfeminine youth

		Baseline		24-Month follow-up		*p* value
	n	Mean (SD)	Range	Mean (SD)	Range	
Systolic BP (mm Hg)	25	122.68 (14.4)	98–151	124.84 (12.10)	96–148	.465
Diastolic BP (mm Hg)	25	71.08 (10.66)	45–86	70.80 (7.68)	57–84	.898
BMI (kg/m^2)	24	24.60 (4.73)	17.7–34.0	24.86 (5.34)	17.1–37.8	.554
Cholesterol (mg/dL)	23	168.96 (41.68)	93–256	166.13 (23.97)	125–218	.718
HDL (mg/dL)	23	43.83 (10.42)	24–62	50.91 (14.34)	27–75	<.001[b]
Triglycerides (mg/dL)	23	135.52 (83.85)	44–383	115.96 (66.22)	45–317	.259
AST (U/L)	23	72.52 (42.19)	18–139	30.83 (17.39)	15–87	<.001[b]
ALT (U/L)	23	30.09 (12.74)	15–57	25.37 (11.59)	5–55	.236
Potassium (mEq/L)	23	4.25 (.31)	3.5–4.9	4.47 (.36)	3.8–5.1	.040[a]
Glucose (mg/dL)	23	90.26 (11.14)	66–110	91.96 (14.00)	74–129	.625
Hemoglobin (g/dL)	22	15.31 (1.13)	12.9–18.1	14.05 (1.24)	11.8–16.0	<.001[b]
Testosterone free (pg/mL)	24	80.90 (49.83)	.7–200.2	28.54 (43.17)	.3–167.4	<.001[b]
Testosterone total (ng/dl)	24	425.88 (233.82)	4–927	169.00 (217.61)	6–782	<.001[b]
Estradiol (pg/mL)	23	26.16 (14.55)	2–61	286.04 (492.04)	5–1,993	.018[a]
Prolactin (ng/mL)	13	8.27 (5.98)	1.0–22.0	11.99 (5.44)	3.2–19.7	.047[a]

ALT = alanine aminotransferase; AST = aspartate aminotransferase; BMI = body mass index; BP = blood pressure; SD = standard deviation.
p values were calculated using paired-samples *t* tests.
[a] Significant at $p < .05$.
[b] Significant at $p < .001$.

Induction of desired feminine physical features as well as achieving a female range of sex steroids is the goal of hormone use in transfeminine patients; therefore, the decrease in mean testosterone and increase in mean estradiol levels is expected, and contributes to desired clinical changes. The mean estradiol level at 24 months is higher than expected with standard dosing (286.04 pg/mL) with a very large range (5–1,993 pg/mL). For those youth taking estradiol via oral route, the levels at follow-up have a smaller range than those getting intramuscular injections (19–331 pg/mL and 5–1,933 pg/mL respectively), and lower mean at follow-up (135.3 vs. 577.6 pg/mL). These differences are likely because of the fluctuating levels of medication delivered via injections versus oral delivery. Mean total testosterone level decreased from 425.9 ng/dL to 169 ng/dL (Table 2).

Physiologic parameters (transmasculine youth)

Transmasculine youth experienced statistically, and mildly clinically, significant changes in blood pressure and HDL. Statistical, but not clinically significant increases in triglycerides, aspartate aminotransferase, ALT (ALT), potassium, and hemoglobin were noted. The change in mean hemoglobin reflects and expected shift that occurs related to testosterone induced amenorrhea and increased production of erythropoietin. Induction of desired masculine physical features and achieving a male range of circulating sex steroids is the goal of testosterone therapy, and is reflected in the changes in total testosterone serum levels of this cohort. Mean total testosterone level increased from 41.2 ng/dL to 533.3 ng/dL. Mean estradiol serum level at follow-up was

Table 3
Physiologic parameters of transmasculine youth

		Baseline		24-month follow up		*p* value
	n	Mean (SD)	Range	Mean (SD)	Range	
Systolic BP (mm Hg)	34	115.62 (15.15)	76–153	128.03 (11.40)	102–157	<.001[b]
Diastolic BP (mm Hg)	34	67.15 (12.57)	40–95	72.32 (12.15)	48–96	.024[a]
BMI (kg/m^2)	35	27.27 (6.17)	16.9–44.2	27.99 (5.53)	19.8–38.2	.188
Cholesterol (mg/dL)	35	163.57 (33.42)	122–256	164.49 (41.40)	94–339	.840
HDL (mg/dL)	35	51.74 (11.37)	30–72	44.49 (12.73)	26–78	.001[a]
Triglycerides (mg/dL)	35	109.86 (92.43)	36–486	144.40 (87.91)	47–403	.044[a]
AST (U/L)	34	55.85 (33.55)	20–129	40.00 (27.42)	16–155	.034[a]
ALT (U/L)	35	22.23 (10.64)	7–53	32.86 (16.67)	14–80	<.001[b]
Potassium (mEq/L)	35	4.23 (.36)	3.5–5.2	4.55 (.41)	3.4–5.5	.002[a]
Glucose (mg/dL)	35	89.20 (17.48)	47–148	84.66 (11.39)	61–119	.172
Hemoglobin (g/dL)	34	13.02 (.97)	10.8–14.8	15.50 (1.12)	11.4–16.9	<.001[b]
Testosterone free (pg/mL)	35	5.79 (7.71)	.3–47.0	117.43 (80.51)	5.7–310.5	<.001[b]
Testosterone total (ng/dl)	35	41.17 (45.31)	7–288	533.26 (331.31)	63–1,272	<.001[b]
Estradiol (pg/mL)	34	81.93 (81.95)	7–372	50.79 (43.21)	9–185	.061

ALT = alanine aminotransferase; AST = aspartate aminotransferase; BMI = body mass index; BP = blood pressure; SD = standard deviation.
p values were calculated using paired-samples *t* tests.
[a] Significant at $p < .05$.
[b] Significant at $p < .001$.

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50.8 pg/mL, higher than most cisgender men, but lower than many cisgender women. Among this sample, mean body mass index, total cholesterol, and random glucose levels did not change over the course of treatment (Table 3).

Discussion

To our knowledge, this is the first prospective study examining the physiologic changes that occur among minors and young adults undergoing treatment with gender-affirming hormones for the purpose of phenotypic gender transition. As the demand for care continues to exponentially increase across the United States, much more data are needed about the impact of hormonal therapy on both physical and mental health in transgender adolescents. This study was limited by the variability in the adherence rates to medication, a problem common to both adolescents and those undergoing care for gender dysphoria. Barriers to access to medication, medical provider follow-up visits, and the natural tendency for youth to geographically relocate pose challenges to collecting data among this cohort. Additionally, the youth in this study were predominantly Caucasian and Latino/a and therefore these results may not be generalizable to youth of other ethnicities.

Current guidelines recommend monitoring physiologic parameters during induction of puberty at baseline and as often as every three months during the first year of hormone use [17,18]. Given the findings of this study, Weinand [7], and Jarin et al. [15], extensive and frequent laboratory examination in transgender adolescents may be unnecessary simply in response to the induction of puberty with gender-affirming hormones. Specifically, monitoring complete blood counts and chemistries including LFTs are unnecessary except perhaps in transwomen who plan to go on spironolactone in which case baseline potassium and creatinine levels would be reasonable. In transwomen, monitoring prolactin is unnecessary unless galactorrhea, or other clinically concerning symptoms occur. Increases in blood pressure seen in transmasculine individuals warrant ongoing monitoring and standard treatment for those who develop hypertension. In other cases where further medical investigation is warranted, it should be undertaken according to clinical necessity.

Frequent concerns about the safety of hormone use in individuals younger than 18 years can create barriers for youth to access medically necessary interventions that have been demonstrated to improve the lives of transgender adolescents. Concerns about the impact of cross-sex hormones on metabolic parameters are starting to be assuaged with clinical experience, retrospective analysis, and more recently, the undertaking of prospective, longitudinal investigations. Among this cohort of youth reported here, there were several statistically significant changes in mean values of physiologic parameters over time, but these did not translate to clinical safety concerns. Hormone levels were impacted as anticipated, and reflect the therapeutic goals of the care. These data indicate that gender-affirming hormone therapy is safe over a time period of approximately two years. Future studies and follow-up information that includes longitudinal results are necessary.

Acknowledgment

The authors would like to acknowledge the youth who participated in this study.

Funding Sources

This work was supported in part by The Saban Research Institute Clinical Research Academic Career Development Award, The National Center for Research Resources and the National Center for Advancing Translational Sciences, National Institutes of Health (NIH), through Grant Award Number KL2TR000131 and by the Eunice Kennedy Shriver National Institute of Child Health and Human Development, NIH, through Grant Award Number 5R01HD082554-03.

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