Received: 17 October 2020 | Revised: 8 April 2021 | Accepted: 18 May 2021

DOI: 10.1002/ajhb.23623

SPECIAL ISSUE ARTICLE

American Journal of Human Biology WILEY

Challenging the binary: Gender/sex and the bio-logics of normalcy

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Abstract

Background: We are witnessing renewed debates regarding definitions and boundaries of human gender/sex, where lines of genetics, gonadal hormones, and secondary sex characteristics are drawn to defend strict binary categorizations, with attendant implications for the acceptability and limits of gender identity and diversity.

Aims: Many argue for the need to recognize the entanglement of gender/sex in humans and the myriad ways that gender experience becomes biology; translating this theory into practice in human biology research is essential. Biological anthropology is well poised to contribute to these societal conversations and debates. To do this effectively, a reconsideration of our own conceptions of gender/sex, gender identity, and sexuality is necessary.

Methods: In this article, we discuss biological variation associated with gender/sex and propose ways forward to ensure we are engaging with gender/sex diversity. We base our analysis in the concept of "biological normalcy," which allows consideration of the relationships between statistical distributions and normative views. We address the problematic reliance on binary categories, the utilization of group means to represent typical biologies, and document ways in which binary norms reinforce stigma and inequality regarding gender/sex, gender identity, and sexuality.

Discussion and Conclusions: We conclude with guidelines and methodological suggestions for how to engage gender/sex and gender identity in research. Our goal is to contribute a framework that all human biologists can use, not just those who work with gender or sexually diverse populations. We hope that in bringing this perspective to bear in human biology, that novel ideas and applications will emerge from within our own discipline.

1 | INTRODUCTION

Biological anthropologists are experts at teasing apart the complexities of biocultural interactions that inform what it is to be human, examining how broad-ranging factors such as market acculturation (Godoy et al., 2005; Liebert et al., 2013), parenting strategies (McKenna et al., 2007; Nelson, 2016), or socially constructed categories of race (Dressler & Bindon, 2000; Gravlee, 2009) relate to physiology including growth and development, immune function, and endocrinology. Yet we have not fully engaged with cutting-edge understandings of variation in gender, sex, and sexuality. This is a critical gap, especially given

[Corrections added after online publication, 30 June, 2021: A word in the abstract section has been revised for clarity.]

renewed debates regarding the boundaries of human sex, where lines of genetics, "sex hormones," and secondary sex characteristics are drawn to defend a strict biologically based sex binary, with attendant implications for the acceptability and limits of gender identity and expression for all people. Whether regulating testosterone levels and bodies of women and girls in sports, legislating the use of gender-specific bathrooms, or enacting broad-sweeping federal definitions of sex, bodily "norms" are being weaponized as a means to discriminate (Karkazis et al., 2012; Nondiscrimination in Health and Health Education or Activities, 2020). Biological anthropology is well poised to contribute to these societal conversations, but first, we need to more deeply consider our own conceptions of sex, gender, and sexuality, and how we implement such understandings in our research.

In this article, we discuss biological variation associated with sex and gender and possible ways forward for conceptualizing and operationalizing these constructs within biological anthropology. We base our analysis in the concept of "biological normalcy," which allows consideration of the relationships between "statistical distributions of biological traits and normative views about what bodies 'should' be like or what constitutes a 'normal' body" (Wiley & Cullin, 2020: p. 1; Wiley, 2021). A classic example of how bionormalcy enables critical interrogation of norms is seen in the case of dietary recommendations normalizing milk consumption culturally as "healthy" and even necessary, despite the statistical norm of lactase nonpersistence (Wiley, 2021). This can be seen as normalizing and even moralizing a biological trait present only in some individuals in some populations (Wiley & Allen, 2017; Wiley & Cullin, 2020). This example aptly demonstrates the fact that many of the statistical distributions that end up being "normalized" are based on samples drawn from predominantly white, "Western" populations (Clancy & Davis, 2019; Henrich et al., 2010), with the psychological, behavioral, and biological traits of these populations referenced as the standard from which other populations deviate (e.g., body size and growth, Thompson et al., 2014).

The model of biological normalcy (Figure 1) is circular. Cultural norms and assumptions inform the development of research questions, methods of data collection, and analyses as well as interpretations of data. Statistical norms are also leveraged (albeit sometimes unconsciously) to create, reinforce, or otherwise inform those very cultural norms and assumptions. However, normalcy has not always been conceptualized in this way. The word "normal" as reflective of something to be desired in reference to an "abnormal" state arose only in the mid to late 19th century (Cryle & Stephens, 2017; Hacking, 1990). Initially, the term "normal" did not represent statistical distributions nor did it carry the morality it is imbued with today. Instead, norms provided a way to reference something "in its own right" and not necessarily through comparison to an ideal. In this way, even anomalies could be understood within a framework of "normal."

With the emergence of statistics in the late 19th century, the concept of the normal became hitched to statistical distributions and to the racist and eugenicist ideas imposed on population traits (Cryle & Stephens, 2017). And with this shift, the concept of the normal intertwines with the history of biological anthropology, as eugenic and white supremacist concepts of human traits and the categorical position of white men as both unmarked and ideal are the very foundation of much of our field (Blakey, 2020; Caspari, 2018; Marks, 2012). Racism and colonialism are equally culpable in the development of value-laden categories of sex and gender and the behavioral norms to which they are often tied. For example, conceptualizations of femininity and masculinity themselves were initially intertwined with racialized categories in an effort to hierarchically demarcate rank, reflecting a colonialist project with the "white ideal" as most differentiated between the sexes (Markowitz, 2001). As a field, biological anthropology continues to suffer from how our history influences *who* practices biological anthropology (e.g., Bolnick et al., 2019).

As biocultural anthropologists, in this article, we aim to broaden the way that human biology engages with categorical thinking about gender and sex and to push for greater recognition of variation in these domains. We are inspired by the decades of strong work into race as a social construct with biological outcomes (Armelagos & Goodman, 1998; Dressler et al., 2005; Graves Jr, 2003; Graves Jr, 2015; Gravlee, 2009; Williams & Mohammed, 2013), and by recent work contextualizing how concepts such as violence are gendered and raced (e.g., Nelson, 2021; Smith, 2021). In our own work, we have grappled with how to better conceptualize and operationalize sex and gender, whether examining energetics and immune

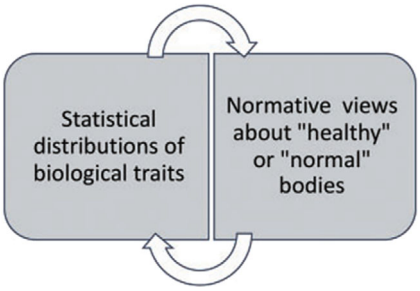

FIGURE 1 Potential relationships between statistical norms and normative views (from Wiley & Cullin, 2020)

function in pubertal girls (Shattuck-Heidorn, Reiches, & Richardson, 2020), sexual decision making among queer adolescent cis men (DuBois et al., 2015), or immune marker and environmental conditions for (cis) men and women (Shattuck-Heidorn, Eick, et al., 2020). In some of our prior work, the category "cis" was unmarked, and at times, in our analytical strategies, we have statistically compared cis men to cis women without a clear justification as to why the sample should be divided by sex as opposed to some other trait(s). Much of our recent scholarship integrates theoretical insights from gender and feminist theory and presents challenges to simple gender/sex binaries through our research questions, study designs, and hypotheses. This is reflected for example, in work expanding understandings of stigma and embodied inequalities among trans and gender diverse people (DuBois, 2012; DuBois et al., 2017), furthering our methodological and theoretical approaches to better encompass gender/sex and sexual diversity (DuBois et al., 2021; Shattuck-Heidorn & Richardson, 2019), and interrogations into the basis for disparities in COVID-19 outcomes (Gibb et al., 2020; Rushovich et al., 2021; Shattuck-Heidorn, Reiches, & Richardson, 2020). Such interdisciplinary merging has enabled us to better conceptualize human gender/sex and enhanced our understanding of variation in embodiment and health.

In this article, we address the following critical areas: (1) the problematic reliance on binary sex categories used as a priori biological categories across traits; (2) the attendant focus on group means to represent typical "male" and "female" behaviors and biology and accompanying fixation on "difference;" (3) the ways in which binary sex norms reinforce stigma and inequality regarding sex, gender, gender identity, and sexuality; (4) the need for "best practices" to effectively engage sex and gender in research; and (5) methodological suggestions to address the lack of inclusive data collection needed to enhance our understanding of gender and sex and sexual variation.

Our goal is to contribute to a framework that all human biology researchers, not just those who work with gender or sexually diverse populations, can use to inform their thinking as well as decisions about best practices for whether and how to implement sex and gender analyses within their research, both theoretically and methodologically.

2 | PROBLEMS WITH RELYING ON BINARY CATEGORIES FOR GENDER/SEX

For decades, evidence has been amassing that a sex and gender binary is an insufficient way to structure our views of human behavior, psychology, and biology (e.g., Hyde et al., 2019). To engage with this conversation, it is important to establish working definitions of several key terms (Table 1). While many of these are well known, we call attention to the combined term gender/sex in this table. While it is possible to conceptually distinguish "sex" from "gender," in practice, with human biology we are nearly always dealing with both. The combined term sex/gender (Kaiser et al., 2009), or gender/sex (van Anders, 2013), foregrounds how gender and sex are developmentally and

TABLE 1 Key terms and definitions

Term	Definition
Cisgender	• Refers to people whose gender identities align with the sex they were assigned at birth
Transgender	• Umbrella term inclusive of people whose gender identity does not align with their sex assigned at birth.
Transgender Women	• People who identify as women or along the trans feminine spectrum who were assigned male at birth
Transgender Men	• People who identify as men or along the trans masculine spectrum who were assigned female at birth
Gender Diverse/ Nonbinary/ Genderqueer	• These terms refer to a myriad of identities that are somewhere between or outside of the gender binary (regardless of sex assigned at birth).
Sex/Gender	• The biosocial entwinement of sex and gender (Kaiser et al., 2009)
Gender/Sex	• The biosocial entwinement of sex and gender (van Anders, 2015)
Sex	• Broadly refers to biological characteristics generally related to reproductive anatomy or physiology
Gender	• Culturally contextualized social and structural experiences as well as expressions of identity
Female	• Represents female in a conceptual manner consistent with gentics-gonad-genitalia/"3G sex" (Joel 2012) — in this case, XX chromosomes, ovaries, and vagina/vulva complex
Male	• Represents male in a conceptual manner consistent with gentics-gonad-genitalia/"3G sex" (Joel 2012— in this case, XY chromosomes, testes, and the penis

irrevocably entangled throughout the life course (Krieger, 2003; Springer, Hankisky, and Bates, 2012; Springer, Stellman, & Jordan-Young, 2012). We also note that, unless we specify otherwise, in this article, the terms "women" and "men" should be understood to include all people who identify as such, as most published research does not yet specify gender identity. Finally, we sometimes use the phrases "assigned female at birth" and "assigned male at birth" when discussing biological factors, to highlight the fact that the sex-related systems we discuss may be present in a variety of gender identities.

As we enter this complex conversation, we recognize that binary categories based on reproductive biologies or gender identities may make sense to include in analyses in order to address certain questions in human biology. We also recognize that there has been an historical and continuing erasure and ignorance of the bodies and biologies of people who fall outside of a normative model centered on white cis men. As detailed by Springer, Hankisky, and Bates (2012) and Springer, Stellman, and Jordan-Young (2012), a binary categorization of "sex" was and remains important in order to *undo the normalization* of the white, male body and related lack of data and bias against all women's bodies. Prior to the NIH Revitalization Act of 1993, "women and minorities" were not consistently included in clinical research in the United States (Epstein, 2007). In addition to the actual lack of data collected among *all women* prior to this enactment, the problems here are glaring; the very language of the Act reflects the way in which women of color are erased and the term "women" is frequently used as an unmarked reference to *white women* (Bowleg, 2012). Relatedly, gender/sex is too often neutrally raced as white within academic research (Bowleg, 2012; Clancy & Davis, 2019; Hull et al., 1982). As the critical framework of intersectionality and Black feminist theory has amply demonstrated, gender intersects with structural and interpersonal experiences of other social categories, especially race, class, and sexuality (Cho et al., 2013; Collins, 1990; Collins, 2019; Crenshaw, 1991; Hancock, 2016). In sum, we recognize the tension of challenging the a priori use of categories in a world rife with inequity and with insufficient knowledge of the experiences, health, women (cis and trans), especially women of color, and also transgender and gender diverse people more broadly.

Although categories may be useful for addressing major issues of exclusion, feminist scientists have critiqued the concept of binary sex (e.g., Fausto-Sterling, 1993), and, for decades, have deeply considered the analysis of sex and gender within biological and behavioral data (e.g., Bleir, 1984; Fausto-Sterling, 1987; Lowe & Hubbard, 1979). We concur with these critiques, recognizing that the binary sex and gender categories currently in use are problematic. Within human biology, the academic querying of the sex binary has focused mainly on intersexuality. This partly reflects the increased visibility and activism of the intersex community, rightfully focused on ending the practice of surgical "normalization" of intersex infant's genitalia (Karkazis, 2008; Preves, 2003). Intersex variability is now well studied within human biology, with several traits (such as congenital adrenal hyperplasia) frequently turned to for insights into the development of human sex and gender (Hines, Brook, & Conway, 2004; Jordan-Young, 2012). However, although the incorporation of intersex conditions into the human sex model is important and contributes to an emerging understanding of a broader spectrum of sex (Astorino, 2019; Blackless et al., 2000), it nonetheless leaves the sex binary in place for most people.

If one considers a binary to mean that two distributions are largely non-overlapping and internally homogenous, then there is a relative binary human sex system in a limited set of characteristics referred to by some as the genetics-gonad-genitalia triad (i.e., "3G sex") (Joel, 2012, see Ainsworth, 2015 for complexity even here). These 3G characteristics are tightly correlated with one another and have high internal reliability within-individuals. For example, if we know that someone was born with XX chromosomes, we can predict with high accuracy that this person also was born with ovaries and a vagina. However, often what we mean by human sex is not 3G sex and instead is downstream from this relative binary. We do not deny that there are in fact differences that can be identified based on 3G sex and that a binary model of sex can be deployed to carefully consider some questions. However, the blanket assumption of a deep, thoroughgoing binary frequently works to mask variation in physiology, as well as sociocultural contributions to human biology.

3 | BIONORMALCY AND THE SEARCH FOR DIFFERENCE

In this section, we draw on bionormalcy to untangle some of the ways in which biological averages and sociocultural norms are mutually reinforcing. Specifically, we demonstrate that there are unnecessary and potentially inaccurate linkages made when binary categories of sex are *exclusively* drawn on to interpret sex-associated biology. The use of binary categories of sex in this way can inadvertently contribute to the normalization of culturally recognized "typical" biologies and undermine capacities to see variation even within these categories defined

as "normal." When used in this way, the *categories themselves* are interpreted as *proxy for pathways* and thus biological differences are concluded to be "sex-based," as opposed to driven by some other mechanism (e.g., the attribution of adverse drug reactions to sex rather than to body size (Greenblatt et al., 2019; Richardson et al., 2015). Intersecting with and mutually reinforcing this issue is a fixation on cisgender heteronormativity and inattention to gender and sexual diversity, despite evidence drawn from our own subdiscipline and closely related fields that gender and sexual diversity is widespread and common (Fuentes, 2019; Meredith, 2015; Monk et al., 2019). Although many contemporary human biologists reject a de-contextualized and simplistic biological determinism (see Smith, 2021 for a recent example), in many ways, when it comes to gender/sex, our field continues to conduct research and provide interpretations of data reinforcing essentialist and reductionist views and concepts.

What human biologists have to offer here is data toward re-orientation to variation and analyses that ground our findings in the biocultural. In the sections below we detail some key examples of how even the traits most associated with maleness and femaleness, including gonadal hormones (here, we challenge the misnomer "sex hormones," while recognizing that these are synthesized throughout the body and not solely in the gonads), secondary sex characteristics, neuroanatomy and the brain are leveraged in interpretations of gender/sex and sex differences. We use language that specifies assigned sex at birth when discussing sex-linked biology, however, we at times fall back on use of the terms "male" and "female" as these are primarily the terms used in this literature by other authors to indicate assigned sex at birth.

3.1 | Traits targeted for difference: Estrogen and testosterone

Moving beyond 3G sex, perhaps nowhere is the binary as firmly entrenched as in our consideration of gonadal hormones. Despite their inaccurate reputations as "male" and "female" hormones, both estrogens, especially estrone and estradiol, and testosterone are important in general human health and development, and hormone levels shift in broadly similar ways over the life course in all humans regardless of sex assigned at birth. However, the study of hormones continues to reflect culturally rooted norms linking estrogens to "femaleness" and testosterone to "maleness. For instance, all infants can have a "mini-puberty" increase in estrogens and testosterone (Frederiksen et al., 2020; Taieb et al., 2003). However, estrogen levels are infrequently measured among infants assigned male and characterization of the androgen surge is lacking among infants assigned female (Becker & Hesse, 2020; Burger et al., 1991; Chellakooty et al., 2003). However, recent evidence suggests not only that all infants have a testosterone surge, but that there are few if any binary group differences in the surge (Corpuz, 2020). Post-infancy, most children have low levels of estrogens and testosterone until puberty (Frederiksen et al., 2020), and during pubertal growth, estrogens act in in similar ways on long bone growth and development for all humans (Bogin, 2020; Dunsworth, 2020; Ellison et al., 2012; Reiches, 2019).

Following puberty, on average females have higher estrogens until menopause when compared to males. But this blunt characterization of averages masks variation within and similarity between these two groups. In fact, significant post-pubertal differences in mean estrogen levels between males and females are largely related to specific menstrual cycle phases and pregnancy. In the early and mid-follicular phase, estrogen levels are modestly different than male levels; it is the rise in estrogens in the late follicular and luteal phase that drives average group difference in estrogen levels (Frederiksen et al., 2020; Handelsman et al., 2014; Verdonk et al., 2019). Pregnancy is also a major source of endogenous variation in estrogens. Given this, comparisons between categories of pregnant/nonpregnant people could arguably be at least as meaningful as binary categories based on sex assigned at birth.

Similar to estrogens, normative interpretations of testosterone as the "male hormone" also belie the biological complexity of this hormone. All humans, regardless of sex assigned at birth, have a rise in testosterone during puberty. On average, males have post-pubertal testosterone levels approximately 10 times higher than females (Jordan-Young & Karkazis, 2019; Kushnir et al., 2010; Salameh et al., 2010). However, this focus on differences in group averages again masks substantial variation within group, and the predominant concept of testosterone as "male" constrains questions related to how testosterone mediates physiology and behavior (van Anders, 2013). For instance, it is well known that absolute levels of testosterone are often not as predictive of outcomes as intra-individual variation (e.g., Archer, 2006). Nonetheless, research often focuses on testosterone levels of cisgender men in relation to normative "male" biology and behavior, while investigation of testosterone and the biology and behavior of cisgender women is comparatively underexplored. Moreover, in our cultural imagination, testosterone is imbued with qualities such as virility, strength, and as being responsible in fact for

"masculinity" itself (e.g., Karkazis & Jordan-Young, 2019). Conceptions of testosterone as driving human male behavioral traits such as male aggression then obscure recognition of the role of learning, early-life experience, and developmental plasticity in such traits (Eliot, 2020). Additionally, such framing fails to account for how, historically, some types of violence (e.g., inter-personal physical violence) are well-recognized and considered within the framework of evolutionary biology, while others are disguised in the service of colonialism, whiteness, and patriarchy (see the February 2021 special issue of *Current Anthropology*, Toward an Anthropological Understanding of Masculinities, Maleness, and Violence, 2021).

Perhaps even more so than with estrogens, when considering transgender, gender diverse, and sexually diverse populations, research inclusive of testosterone measures tends to focus on examining normative masculine/feminine traits and etiologies of what is perceived as "abnormal." The search for a "cause" for sexual and gender diversity itself reflects the impact of norms and their potential to pathologize people and behaviors. For example, studies often aim to "sex" the brain or link fetal androgen exposure to gender identity development or sexuality, though the role of endogenous hormonal levels on either is unclear (Berenbaum, 2018; Jordan-Young, 2011).

In addition to hormonal variation related to stages of development and reproduction, there is significant variation in gonadal hormone levels in relation to diet, activity level, body fat, parenting, and many other factors (Bentley et al., 1998; Bribiescas, 2001; Ellison, 2001; Gettler et al., 2011; Nunez-De La Mora et al., 2007; Valeggia & Ellison, 2001; van Anders, 2013; Vitzthum, 2008). Research in this area makes clear how human neuroendocrinology is embedded and impacted by social factors, not simply driving human behavior. For example, recent longitudinal research has demonstrated how the behavior of male parenting has shaped men's physiology, showing decreased T among fathers who engage in more childcare compared to single, non-fathers or less involved fathers (Gettler et al., 2011; Gray et al., 2002; for review, see Gray et al., 2020). The data in this area points to how the "biology of fatherhood" is culturally embedded, reflecting intergenerational effects as well as developmental plasticity; exposure to fathers early in life influences later life biologies of those who become fathers (Gettler, 2016). Similar patterns have also been observed among mothers of young infants (Kuzawa et al., 2010).

Increasingly, the exogenous administration of gonadal hormones and environmental exposures also constitute important sources of endocrine variation. Cisgender and transgender people alike access exogenous hormones to address a variety of concerns related to health and aging. Arguably, many cisgender people (the group perceived as "normal") in fact access what could also be called "gender affirming care" (e.g. breast augmentation, treatment for gynomastia, hormonal replacement therapies, and Viagra), although this term is generally only applied to transgender people (the group perceived as "abnormal") when accessing care. Regarding transgender health specifically, expression of one's gender identity including through gender affirming hormonal therapies, is associated with positive mental health outcomes including lower levels of anxiety and depression (Keo-Meier et al., 2015; White Hughto & Reisner, 2016), and reduced suicidality (Hughto et al., 2020). Moving forward, much remains to be learned that will be of benefit to these communities. As these critical areas are researched, it will be important to prevent normative, binary conceptions of hormones as sex-specific and linked to stereotypical behaviors from driving research agendas and hypotheses.

In sum, estrogens and testosterone play an important role in the biologies of all people, regardless of sex assigned at birth or gender identity. Sex-related differences and similarities depend on a multitude of factors including age/life stage, menstrual status, reproductive state, ecology and the environment, other aspects of health, and interactions with exogenous hormones. Given this, it is perhaps unsurprising that even the distribution of secondary sex characteristics associated with these hormones exists beyond a clear binary. For example, the flexibility of secondary sex characteristics is evidenced among transgender people who affirm their gender through exogenous administration of estrogen or testosterone. Phenotypic changes occur that are not unlike adolescent puberty, including shifts in body and facial hair growth, fat distribution, musculature, and vocal changes. Variation in secondary sex characteristics is similarly evident in "female hirsutism," a condition in which "excess," "male-typical" body hair is present in women, which affects at least 5–15% of the population in the US, depending on ethnicity (Yildiz et al., 2010). The very measurement of "hirsutism" was invented by anthropologists to assist in classifying race (Carlin & Kramer, 2020; Danforth & Trotter, 1922). Finally, many adolescent males develop transitory breast tissue during puberty, and about a third of adult men will develop breast tissue at some point in their lives (Braunstein, 1993; Johnson & Murad, 2009; Lemaine et al., 2013). White, Eurocentric, cisgender, and heterosexual norms clearly inform how variation in breast tissue and body hair is characterized and socially responded to within the overarching binary of gender/sex.

3.2 | Traits targeted for difference: Neuroanatomy and the brain

When considering how biological and behavioral characteristics (including psychological traits) are interpreted as "male" or "female," it is important to recognize how small most differences in such traits are. The brain, and purported downstream behavioral differences, provides an ideal site to examine how we conceptualize sex differences and similarities. Recent research demonstrates that, consistently, apparent human sex differences are developmentally dependent on the environment, may shift over the life course, and overall, demonstrate significant plasticity and canalization (Fausto-Sterling et al., 2012a, 2012b; Hyde, 2005; Maney, 2016). For classically recognized "sex differences," such as female verbal advantage, the actual effect size is often quite small (e.g., for verbal advantage is Cohen's d is <.20) and the distributions are highly overlapping (Figure 2(A)); for most other traits like vocabulary or reading comprehension, there are not consistent sex differences or they are even smaller (Hyde, 2014; Hyde & Linn, 1988). Brain regions purported to explain broad male and female differences also show minimal effect sizes and high degrees of overlap yet are often seen as providing evidence of deep and throughgoing difference. For example, small effect sizes ($d = 0.31$) in intrahemispheric connectivity (Figure 2 (B)) are nonetheless leveraged as explanation for differences in learning "capacities" and as the basis for related policy recommendations for sex-segregated education (Ingalhalikar et al., 2014; Maney, 2016).

In contrast to hypotheses focused on difference, the gender similarities hypothesis recognizes that most biobehavioral "sex differences" actually reflect largely overlapping distributions with relatively small differences in average scores (Hyde, 2005). This hypothesis has held up

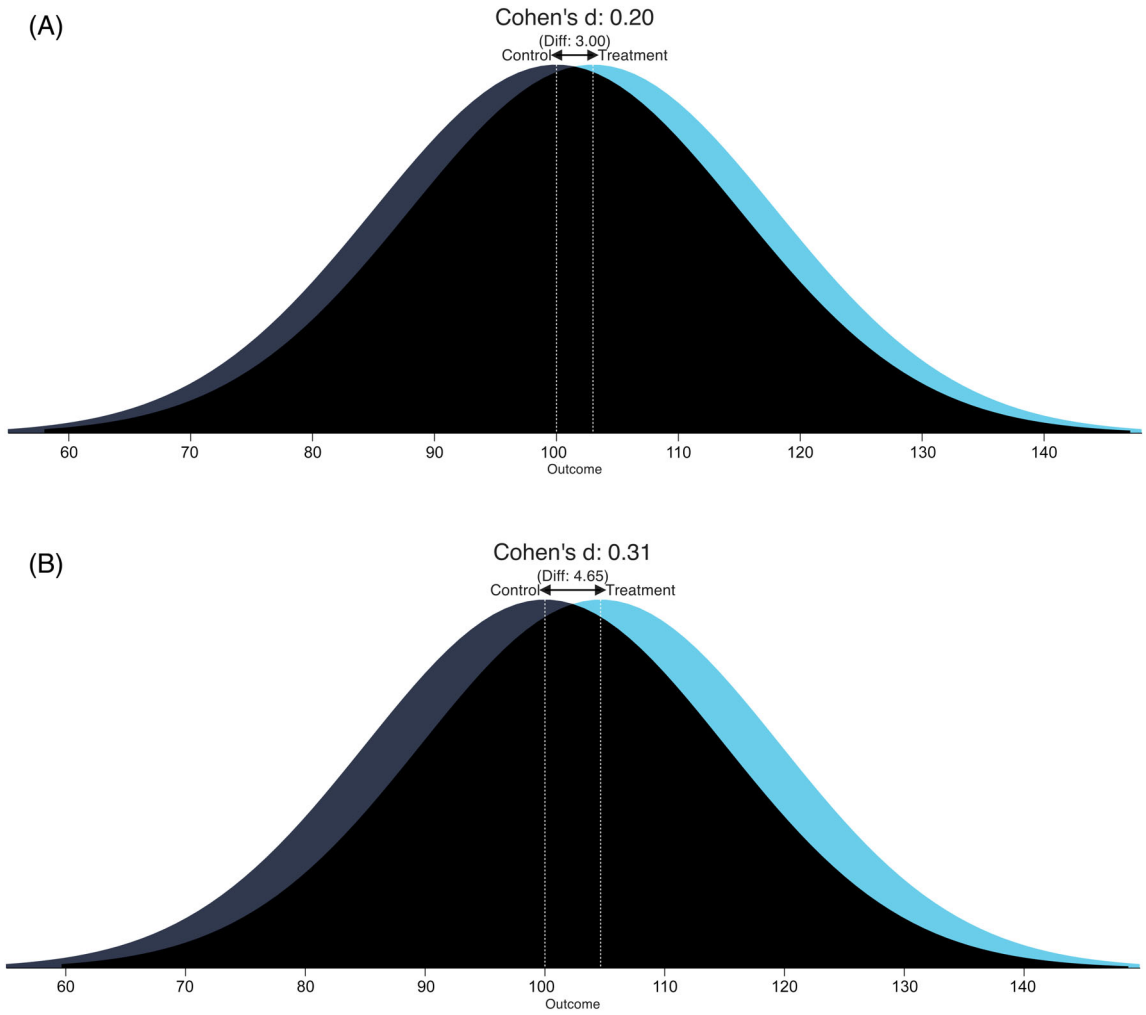

FIGURE 2 Representation of distributions of common "sex differences". (A) The female advantage in verbal ability, usually <$d = 0.20$ (Hyde, 2014). There is a 92% overlap in the distributions. Figure created using Magnusson, 2020. (B) Intrahemispheric connectivity in the brain, female advantage, $d = 0.31$. There is an 87.7% overlap in the distributions. Figure created using Magnusson, 2020

across multiple meta-analyses of studies on diverse traits ranging from cross-national mathematics test scores (Else-Quest et al., 2010) to most measures of sexuality (Petersen & Hyde, 2010). A recent meta-synthesis, a statistical examination of the now large body of meta-analyses, also supports the general concept of gender similarities (Zell et al., 2015). Even authors committed to interpreting minimal sex differences as reflective of evolved male and female tendencies concede that the majority of the differences are extremely slight (Archer, 2019). Finally, a recent comprehensive synthesis of 30-years of brain research found that *size not binary sex*, explains observed differences within and between groups of men and women (Eliot et al., 2020). Despite this, the concept of pervasive and immutable male and female difference continues to drive policies and practices, being used to justify interventions in sex and sports, same-sex educational programs, and even medicine (Eliot, 2009; Karkazis, Jordan-Young, Davis, & Camporesi, 2012; Richardson et al., 2015). This pattern of reifying biology and normalizing group averages to represent typical biologies and behavior is consistently evident in disputes over how to characterize the human brain.

Scientists have spent decades looking for sex differences in the human brain to help explain observed sociocultural and behavioral differences in women and men. Often what occurs is the identification of a statistically significant difference, for instance, that females have a larger splenium (DeLacoste-Utamsing & Holloway, 1982). This is then disseminated as evidence to explain all manner of sex stereotypes, such as women's ability to multitask, even as these same differences are found inconsistently or not supported by later work (Bishop & Wahlsten, 1997; Bryant et al., 2019; Eliot, 2009). This pattern of identification and refutation has occurred with a number of areas of the brain (Fausto-Sterling, 2008). Nonetheless, as with other biological fixations in the past (e.g., overall brain size), sex differences in specific aspects of brain anatomy remain a target in the search for difference to explain observed behavioral traits (see Journal of Neuroscience, Special Issue Prager, 2017).

Key here for critically interpreting brain sex difference research is to note that most brain differences attributed to innate sex biology are small, highly overlapping, and lack internal consistency. Brains, unlike 3G sex, are "mosaic"; a man with an area of the brain characterized as typically "male" also has areas characterized as typically "female," and a majority categorized as neither "male" nor "female" (Joel et al., 2015; Maney, 2016). Of course, even with very slight differences, if a large number of traits is compiled, one can achieve relatively high (~75%) accuracy when retroactively "sexing" an individual brain; some researchers claim this as evidence of innate sex-based difference (Del Giudice et al., 2016). Others critique this interpretation, as it is a basic statistical fact that any large set of traits that differ slightly between two groups can, for an individual, predict group membership with some accuracy. Yet this does not mean there are innate biological differences between the groups, or that it is possible to predict which traits an individual has based on group membership (Joel, 2011). Overall, we draw on this example to suggest that the largely overlapping distributions of brain regions, plasticity of brain development, and the lack of correlation between "sexed" brain regions in individuals challenge the definition of just what "sex" is (Eliot, 2009; Eliot et al., 2020; Maney, 2016; Rippon, 2020).

As it stands, the brain continues to be a focus for research aiming to locate a biological basis for perceived gender/sex differences. The implications for thinking about relationships between biology and behavior make this a particularly controversial area of inquiry and one in need of an anthropological lens. Too frequently, differences are emphasized in order to move forward gender-based policies or practices, such as the biological arguments made to justify the institution of sex-specific education programs, though there are only slight mean differences in boy's and girl's learning and no evidence of these differences being driven by brain structure or function (Eliot, 2009). Biology is also clearly leveraged to uncover the "cause" of identities and behaviors perceived as "abnormal." For example, the brains of transgender people (much like sexually diverse people in the recent past) are often examined in order to unearth explanations for what are considered "non-normative" gender identities and sexualities, or to enable comparison between transgender and cisgender people, further reinforcing binary and stereotypical gendered expectations (LeVay, 1991; Nguyen et al., 2019). Moreover, when seeking biological bases for perceived sex differences, researchers have targeted transgender and gender diverse people specifically to test hypotheses without sufficient consideration of the needs of the community (Llaveria Caselles, 2021).

3.3 | The stigmatizing effects of rigid sex difference frames

Our collective failure to recognize the overlapping nature of much of human biology, even that directly linked to 3G sex, such as gonadal hormones, has ramifications not only for our research paradigms, but also in our greater (and connected) sociopolitical worlds. One key area where the trouble with the binary becomes increasingly apparent is forensics. Estimation of sex from skeletal remains are based on methods of scoring traits using an

ordinal scale. Much like the brain, instead of scoring results indicating discrete binary categories, traits are recognized as overlapping with individuals displaying mixtures of traits and without capacity to account for individual gender identity (Garofalo & Garvin, 2020). This is a pressing issue in contemporary forensics cases, where it is vitally necessary for instance, that trans and gender diverse people are properly identified and not disrespected through misgendering and deadnaming.

Another current example is the controversy surrounding testosterone levels among elite athletes, where regulations around "doping" have become conflated with natural human variation and production of endogenous hormones. These controversies, along with the even more recent public debate and discourse about trans youth in sports, raise the issue of how to achieve equality for all women, without excluding or discriminating against intersex, transgender, and gender diverse athletes. Most recently, the case of Caster Semenya, an intersex cisgender woman, exposes the failure of these categories to affirm or sufficiently protect any*body* given the at once arbitrary and also discriminatory definitions of what it means to be female (Karkazis & Jordan-Young, 2015; Jordan-Young & Karkazis, 2019; Pielke et al., 2019). The recent International Association of Athletics Federation regulation requires naturally elevated testosterone levels be medically lowered to the "normal" female range for a woman to compete as a woman. In terms of testosterone, this ruling clearly demonstrates the way in which testosterone is marked as the "male hormone" with insufficient recognition of the degree of similarity and overlap of hormonal levels across binary categories (e.g., among elite athletes, 16.5% of men had low testosterone and 13.7% of women had high testosterone with overlap, see Healy et al., 2014). In terms of biological normalcy, this also points to the degree to which certain gendered biological traits (i.e., hormones) are targeted for "normalization" while others (e.g., limb length) are not scrutinized or policed through the same gendered lens.

4 | STRESS BIOLOGY AND THE BINARY

Research related to psychosocial stress provides an ideal opportunity to examine the dynamic interplay of gender/sex and to ask how sex-related biology in tandem with gendered experiences contributes to biological outcomes. Here, as above, we suggest that integration of gender experience, gender identity, and biological measures of hypothesized pathways and moderators are necessary to elucidate understandings of stress and gender/sex embodiment.

Studies of stress are necessarily complex, requiring assessment of individual perception of threat in coincidence with short-term (acute) and/or long-term (chronic) exposure and measures of psychobiological response. During a stress response, the body activates the sympathetic-adrenal-medullary system and hypothalamic–pituitary–adrenal (HPA) axis, enabling short-term adaptive responses through increased cortisol levels, heart rate, and blood pressure, facilitating the mobilization of energy stores and redirecting blood flow (James & Brown, 1997). However, this activation comes at a cost, with chronic stress leading to systemic dysregulation and pathological long-term outcomes including increased risks of Type-II diabetes and cardiovascular disease (McEwen & Seeman, 1999; Padgett & Glaser, 2003). Field-based studies have primarily focused on effects of chronic stress, while lab-based research primarily measures stress reactivity (e.g. to social evaluative threat) in order to identify physiological pathways linking stress experience to health (Dickerson & Kemeny, 2004). Research also examines specific aspects of diurnal cortisol profiles, such as the cortisol awakening response (CAR), which measures the surge in cortisol associated with waking (Pruessner et al., 1997). Finally, collection of multiple biomarkers of stress and health enable the calculation of allostatic load (AL), a characterization of stress-induced "wear and tear" on multiple systems of the body, and an important predictor of morbidity and mortality (McEwen & Seeman, 1999).

4.1 | Gonadal hormones and the stress response

When considering sex and biology linked to psychosocial stress, many studies aim to detect sex-based differences in stress reactivity. Overall, there are few differences in average baseline cortisol levels between (presumably cisgender) men and women, but experimental studies do identify differences in responses to psychosocial stress. For example, men are found to show greater HPA axis responsivity (i.e., cortisol secretion) to lab-based social stressors such as public speaking compared to women (Kirschbaum et al., 1992). On the other hand, women are sometimes (but not always) found to have a higher CAR than men (Almeida et al., 2009; Fries et al., 2009; Hill et al., 2013).

The normative concept of an innate sex binary heavily influences how differences in stress biology between (presumably cisgender) men and women are interpreted. When differences are found, an innate biological lens is nearly exclusively used to consider causal mechanisms (e.g., sex-differences in evolved stress responses), with minimal consideration of gendered

experiences and infrequent systematic measures of gonadal hormones (Juster et al., 2019). To be sure, when measured, relationships have been found between gonadal hormones and some aspects of stress-related biology. For example, early research found lower concentrations of cortisol secretion during the follicular phase and higher levels of cortisol secretion during the luteal phase, which has been interpreted as evidence of the mediating role of estrogen (Schöneshöfer & Wagner, 1977). Further work has supported the role of estrogen in attenuating cortisol reactivity; women with low estrogen show similar reactivity patterns to men while women with high estrogen align with women taking oral contraceptives (Kirschbaum et al., 1999).This suggests that estrogen exposure attenuates cortisol responsiveness, possibly through effects on cortisol binding globulin, thereby modifying levels of circulating free cortisol (Kajantie & Phillips, 2006). However, such hormonal differences do not explain all differences in cortisol secretion or reactivity. For instance, researchers find that CAR does not differ across the follicular, luteal, or menstrual phase (Kudlielka & Kirschbaum, 2003; Wolfram et al., 2011). Further, recent research that examined the role of progesterone, estradiol, and testosterone in stress induced HPA-activation found that sex differences disappear when statistically controlling for gonadal hormones, suggesting that while gonadal hormones are involved, responses are not in fact sex-specific (Juster et al., 2016). A recent review by the same author emphasized that when considering overall differences in stress responses, research should not only consider related hormones, but also needs to incorporate a nuanced understanding of the role of experiences related to gender, gender identity, and sexuality in anticipation, exposure, and response to stressors (Juster, 2019).

4.2 | Gender/sex experience, stigma and stress response

Gender/sex-based inequalities, norms, enacted stigma and discrimination clearly impact the biologies of people of all genders/sexes, albeit in different ways, and there is a significant body of experimental research on gender-based stress in response to threats. Both (presumably cisgender) men and women show heightened cortisol following experimentally induced gender-based exclusion in the workplace; however, women are much more likely to report having actually experienced this type of stressor as well as sexist threats in their lives (Taylor, 2016). Relatedly, recent experiences of sexism and gender-based discrimination, including experiences in the workplace, have negative effects on the health and well-being of women (Berg, 2006; Pascoe & Richman, 2009; Sojo et al., 2016). Experimental stress tests also demonstrate that women who report experiencing more chronic sexism in their lives have higher cortisol responses in experimental employment-related situations that cued the experience of sexism (Townsend et al., 2011).

Of course, social experiences of gender are also raced and contingent on other social categories. Consider the starkly revealing study of the impact of employment discrimination on Black men, which found that white men were twice as likely to receive a job callback as Black men with identical applications, and that Black men with a clean record are as likely to receive a job callback as white men with a recent felony (Pager et al., 2009). The racist discrimination revealed in this study can be understood as a result of racist stereotypes not only of Blackness, but of Black men in particular in the United States. There is a large body of literature demonstrating the effects of racism on stress and health in the United States (Bailey et al., 2017; Clark et al., 1999), yet much remains to be done to further incorporate gender/sex, social identity, and localized contexts. Recent work such as the investigation of how heterosexism and racism impact stress as measured through salivary cortisol among sexually diverse Latinx (Parra & Hastings, 2020) exemplifies the utility of considering intersectional experiences of gender, race, and sexuality.

As these examples demonstrate, the intersectional role of gender in peoples' lives must be interpreted from a multilevel framework. Such a framework will consider multiple ways in which gender impacts individuals, from institutional and structural levels (such as abortion and healthcare access, or community-level beliefs about gender roles), to interpersonal relationships, to individually held beliefs (Homan, 2019; Shattuck-Heidorn & Richardson, 2019; Springer, Hankisky, & Bates, 2012). The effects on individuals are an outcome of multiple levels interacting. For example, consider how not just relational aspects of gender (i.e., a sexist threat), but also an individual's own beliefs and internalized gender norms influence how individuals respond to situations. For instance, (presumably cis) women who are more aware of sexism and feel less personal control report more distress from sexist events (Landry & Merucrio, 2009), and in experimental work, women who indicate that "being a woman" is important to their identity have stronger cardiovascular stress responses to a sexist social threat test (Eliezer et al., 2010). Masculinity beliefs may also influence stress response in women (Juster & Lupien, 2012) and cortisol reactivity in men (Himmelstein et al., 2019).

Beyond the binary, heterosexist and cisgender norms also have a negative impact on transgender, gender diverse, and sexually diverse people, particularly through "minority-stressors" which are experienced above and

beyond general day-to-day stress (Brooks, 1981; Hendricks & Testa, 2012; Meyer, 2003). Marginalization and stigma for people whose gender expressions, identities, and/or sexual orientation fall outside of cisgender heterosexual norms are known to contribute to excess stress and increase health risks (Hatzenbuehler & McLaughlin, 2014; Hughto et al., 2015). Binary categories as well as bodily norms together arguably create and facilitate the maintenance of these conditions of inequality that trans and gender diverse people endure, exacerbating both enacted and internalized stigma. These experiences become embodied, for instance, stigma-induced stress related to "coming out" as transgender and the use of gender-specific public restrooms is associated with elevated waking cortisol levels among transgender men (DuBois et al., 2017). Stress about "coming out" is also associated with reduced decline in blood pressure while sleeping which, if experienced chronically, can greatly increase risk for cardiovascular disease (DuBois, 2012).

These experiences of minority stress endured in the context of binary-induced marginalization and stigma have devastating effects on sexual and gender minority people. Forty-one percent of transgender people in the United States have attempted suicide in their lifetime (James et al., 2015). Transgender and gender diverse people, particularly trans women of color, continue to face significant social and structural inequalities, economic instability, high levels of mistreatment, discrimination, and violence (James et al., 2015; Puckett, Maroney, et al., 2020; Hughto et al., 2015). Enacting cultural, policy, and legislative shifts toward inclusion helps shift the fixation on binary norms to instead recognize the breadth of human variation, particularly where gender/sex and identity is concerned.

As this brief tour of the complexities of psychosocial stress in relation to gender/sex, sexuality, and gender identity shows, an intersectional, multilevel understanding of gender/sex is needed to enable sophisticated analyses of stress-linked biology and health outcomes; the contextualization and elucidation of complex physiological pathways from this framework are much needed. This includes the need for further research investigating the role of historical trauma (see Gone et al., 2019 for review on impacts for Indigenous populations in United States and Canada) and intergenerational transmission of stress (e.g., via epigenetic pathways) linking experience to health and health disparities (e.g., Thayer & Kuzawa, 2011). Given that the stress response reflects psychobiological experience within cultural context, when considering gender/sex, feminist theory ideally informs the integration of socially relevant factors in research. Unless we consistently consider the dynamic and embodied interaction of gender/sex, observed differences between those in gendered categories will likely continue to be assumed to be rooted in innate biology, and the experiences of sexual and gender minority people will continue to be erased. Psychosocial stress offers a clear framework for complex consideration of the gendered/sexed biologies of everyday life, and one that our field is well poised to apply across varied sociocultural environments.

5 | GENDER/SEX "ENTANGLEMENT" AND NUANCED METHODOLOGIES

In this final section, we unpack some of existing tensions between (1) the need to reduce exclusion by collecting group-level data on the one hand (e.g., of cis women and trans and gender diverse people) and the coincident recognition that (2) binary group-level categorization in human biology can reify essentialist explanations, contribute to erasure, and exacerbate aspects of marginalization and stigma. Here, we suggest how biological anthropology can look to how other fields are theorizing and operationalizing gender/sex, which can assist us as we further develop and implement best practices in our work.

Our call to use caution in deploying binary sex categories overlaps with an increased focus on sex-differences, heightened by the 2014 National Institute of Health (NIH) mandate for inclusion of sex as a biological variable in clinical and preclinical research (Clayton & Collins, 2014; Eliot & Richardson, 2016; Richardson et al., 2015; Ritz, 2017; Tannenbaum et al., 2016). This NIH policy shift mandates inclusion in preclinical research of both female and male cellular materials and animal models as well as analyses targeting the identification of sex differences (Clayton & Collins, 2014), and has inspired debate regarding how human sex should be considered in clinical and population-based research (e.g., Ritz, 2017). Within this debate, we can find reasonable suggestions for how researchers interested in operationalizing gender/sex might best do so.

5.1 | Recommendations: The use of gender/sex categories in research

Here, we elaborate recommendations made in allied fields of health sciences, including a development plan

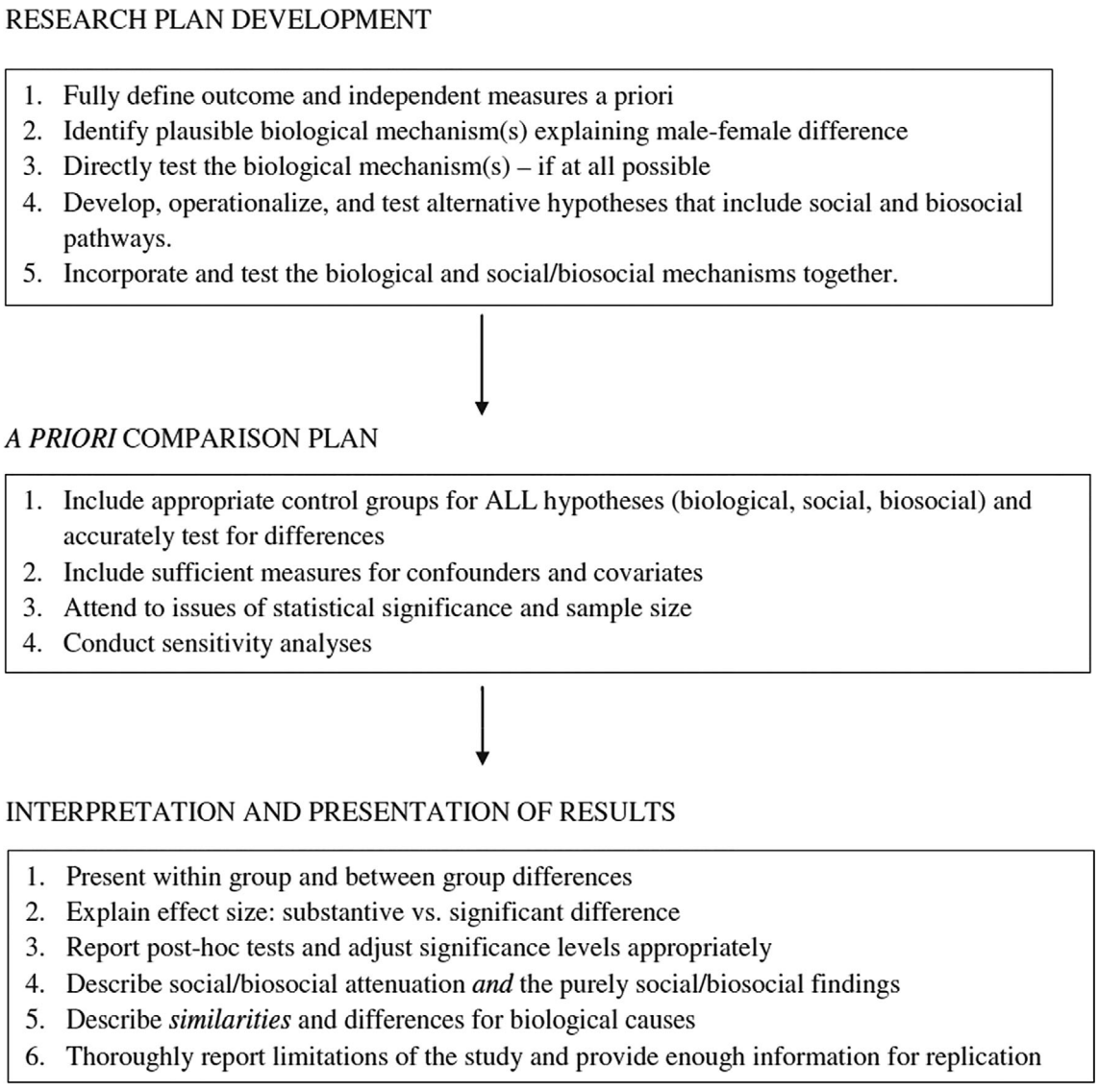

FIGURE 3 Good practice guidelines for researching gender/sex in humans. From Springer, K. W., Stellman, J. M., & Jordan-Young, R. M. (2012). Beyond a catalogue of differences: a theoretical frame and good practice guidelines for researching gender/sex in human health. Social science & medicine, 74(11), 1817–1824

for researching gender/sex in human health as proposed by Springer, Stellman, and Jordan-Young (2012) (see Figure 3).

Our aim is to challenge a number of practices that generally go unquestioned. These include (1) the automatic inclusion of binary sex categories without an analytic plan to engage with gender/sex entanglement, (2) conducting comparative analyses based on binary sex categories that are unwarranted by the hypotheses, (3) relying on categories of binary sex as proxies to explain observed difference rather than examining physiological mechanisms, and (4) relying on a priori binary categories of sex or gender rather than collecting data from our study participants about their gender experience and identities (detailed below).

When considering biosocial hypotheses, we follow others in recommending that researchers take an intersectional approach to understanding gender (Agénor, 2020; Springer, Hankisky, & Bates, 2012). As mentioned prior, intersectionality is a critical analytical framework, developed within Black feminist theory and critical race theory, which understands gender as operating in intersection with structural and interpersonal experiences of other social categories, especially race, class, and sexuality (Collins, 1990; Crenshaw, 1991). In other words, there is no universal "gender" but rather, gender is meaningful in light of other identity categories. For biological anthropologists, we caution against interpreting this "intersection" as indicating that the analytical consideration of multiple "identity" categories fulfills

the goals of intersectionality; intersectionality is an analytical framework for the consideration of societal inequities and social justice (Bilge, 2013; Hancock, 2016). For instance, an intersectional perspective on gender could motivate investigation of the high levels of discrimination and violence enacted against both cis and trans women of color *if* it situated such an analysis within larger societal and structural drivers of these experiences (Crenshaw, 1991; James et al., 2015; Parra & Hastings, 2020). Increasingly, scientists are working to find robust methodological approaches that can be deployed in primarily quantitative fields and also stay true to the epistemic goals of intersectionality (Bauer, 2014; Bauer & Scheim, 2019; Bowleg, 2008; Bowleg & Bauer, 2016; Else-Quest & Hyde, 2016; Evans et al., 2018).

5.2 | Recommendations: Capturing gender/sex diversity through data collection

Overall, we recommend collecting data inclusive of gender identity and lived gender experience, instead of relying exclusively on phenotypic and secondary sex characteristics to assign individuals to binary sex categories. Integrating gender identity allows our work to grapple with the complexity of what it means to be human, including when viewed through evolutionary frameworks. For instance, if the gender of a sample of study participants is assumed homogenous because recruitment criteria specified gender as part of eligibility criteria (e.g., calls for women 18–45), it would be important to specify whether identity data were also collected. Conscientious use of terminology is also important; the term "woman" pertains to identity and is thus inclusive of transgender women. However, many studies are inclusive of cis women only, with the term "woman" actually referring to assigned sex at birth. Similar issues are at play with sexuality; studies may assume heterosexuality among participants, rather than measure it specifically.

A call to meaningfully engage with gender identity does not suggest that gender identity-related terms translate in all cultural contexts. Meaningful engagement at the local level is clearly required to ensure appropriate language and concepts. To facilitate this, we offer an example of a multi-part question to assess gender identity (see example Table 2) that can be adapted for use along with data collection regarding sexual orientation when appropriate. When sexual activity or health is of interest, language can be used that is respectful of the varied ways people experience their bodies (e.g., Reisner et al., 2014 and see the Trans PULSE Canada survey (Bauer et al., 2012). Ideally, all questionnaires also have write-in options and/or multiple categorical options, are grounded in ethnographically obtained research, and are locally informed and linguistically appropriate (e.g., Puckett, Brown, et al., 2020).

TABLE 2 Example TGD-inclusive measure of gender/sex

Q1. What sex were you assigned at birth (meaning on your original birth certificate)?
1. Female
2. Male
3. Undetermined

Q2. What is your current gender identity?
1. Woman/Female
2. Man/Male
3. Trans woman /Trans female
4. Trans man /Trans male
5. Indigenous or other cultural gender minority identity (e.g. Two-Spirit)
6. Genderqueer/Gender Non-Binary/Gender Fluid
7. Different identity (please specify)_____

Q3. What gender do you currently live in your day to day life?
1. Woman/Female
2. Man/Male
3. Sometimes a man/male sometimes a woman/female
4. Something other than a man/male or woman/female

Note: *Adapted based on recommendations from: Bauer, G. R., Braimoh, J., Scheim, A. I., & Dharma, C. (2017). *Transgender-inclusive measures of gender/sex for population surveys: Mixed methods evaluation and recommendations. PLoS ONE*, *12*(5), 1–28.

6 | CONCLUSIONS

As human biologists, gender/sex is central to how we understand and organize our thinking about human evolution as well as health in contemporary and historic contexts. The entwinement of gender and sex is complex, as is much of the science exploring this variation and how it develops. It is increasingly necessary for human biologists to engage novel methodologies to ensure we are capturing and engaging with gender/sex diversity. As detailed above, research in human biology and other disciplines challenges the understanding and the use of binary sex as a meaningful category explaining human biological variation across contexts.

The work reviewed here is a small part of a large field of research that pushes us to continue to consider the ways in which human bodies and identities resist static categorization. Hormones vary and function in complex ecological and social environments, brains and bodies develop over time in response to varied experiences and inputs, and societal structures of gender norms, race and racism, and sexuality influence and mediate human

biology. As the common-sense notion of binary categories for human gender/sex are destabilized, our discipline is well-positioned to meaningfully explore the complexity of gender/sex in terms of human variation and to understand that variation within a sociocultural context, including race, sexuality, and gender diversity. Our field has contributed substantially to an understanding of human biology in a socioecological context. We look forward to a generation of work from biological anthropologists who are incorporating intersectional analyses of gender/sex and gender identity into our understandings of human diversity.

ACKNOWLEDGMENTS

The authors would like to thank Dr. Andrea Wiley and Jennifer Cullin for organizing the SAR workshop focused on bionormalcy and for providing the opportunity to engage this useful model with colleagues at the workshop. We would also like to thank editors Virginia Vitzhum and Jennifer Cullin for this Special Issue.

CONFLICT OF INTERESTS

The authors have no conflict of interests.

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How to cite this article: DuBois, L. Z., & Shattuck-Heidorn, H. (2021). Challenging the binary: Gender/sex and the bio-logics of normalcy. *American Journal of Human Biology*, *33*(5), e23623. https://doi.org/10.1002/ajhb.23623