Finger Length Ratios of Identical Twins with Discordant Sexual Orientations

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ABSTRACT

A proposed marker of prenatal androgen exposure is the ratio of the index finger to ring finger (2D:4D). Within each sex, this ratio may be lower for those who were exposed to higher levels of androgens and become attracted to women, as compared to those who were exposed to lower levels of androgens and become attracted to men. We examined these patterns in identical twins with discordant sexual orientations. Because these twins are genetically identical, differences in prenatal androgen exposure, as reflected in their different finger length ratios, might contribute to their discordance. For 18 female twin pairs, non-straight (bisexual or lesbian) twins had significantly lower, or more masculinized, 2D:4D ratios than their straight co-twins, but only in the left hand. For 14 male pairs, non-straight twins had, contrary to our prediction, more masculinized finger length ratios than straight co-twins, but this difference was not significant. A reanalysis of present and previous data (Hall & Love, 2003; Hiraishi, Sasaki, Shikishima, & Ando, 2012) suggested that these patterns were robust. Furthermore, males had more masculinized 2D:4D ratios than females. This sex difference did not vary by sexual orientation.

Keywords: sexual orientation, sexual differentiation, finger length ratio, 2D:4D, twins

INTRODUCTION

Because identical twins, who share 100% of their genes, can differ in their sexual orientations, factors other than genetics must account for their differences. For example, different exposure or reactions to prenatal androgens, unique to each twin of a pair, could contribute to their discordant sexual orientations. That is, for one twin, but not for the other, distinct exposure or responses to prenatal androgens may increase the likelihood of a non-straight (bisexual or homosexual) orientation (Bailey et al., 2016; Breedlove, 2017).

Correlational studies in humans, experimental studies in animals, and theoretical reviews all point to the possibility that finger length is sensitive to prenatal androgens. In particular, the ratio of the length of the index to ring finger (2D:4D) is a putative indicator of exposure to prenatal androgens: this ratio is, on average, lower (more masculinized due to androgens) in males than in females (Breedlove, 2010; Grimbos, Dawood, Burriss, Zucker, & Puts, 2010; Hines, 2011; Manning, 2011; Zheng & Cohn, 2011). For example, a meta-analysis suggests that men have lower 2D:4D ratios than women in the left and right hand, $p < .001$, $g = .44$, and $p < .001$, g = .55, respectively (Grimbos et al., 2010). Furthermore, in humans, 2D:4D may be a predictor of sexual orientation. In the following sections we review evidence for sexual orientation differences in 2D:4D and consider that 2D:4D could differ within pairs of identical twins with discordant sexual orientations.

Sexual Orientation Differences in 2D:4D

The measure of 2D:4D has been described as unreliable by some because it produces mixed findings, at least with regards to differences by sexual orientation (McFadden et al., 2005; Putz, Gaulin, Sporter, & McBurney, 2004). The present research therefore relied most heavily on a carefully conducted meta-analysis across 34 independent samples and a total of 5,828 participants in order to draw informed predictions about sexual orientation differences. In this meta-analysis, women of a non-straight sexual orientation showed a lower 2D:4D ratio in the left and right hand, in comparison to straight women, Hedge's *g*'s = .23 and .29, .04 < 95% CIs < .51 (Grimbos et al., 2010). Furthermore, a prominent clinical example with respect to this effect is that of genetic females with congenital adrenal hyperplasia (CAH). Due to a genetic condition, women with CAH are exposed to unusually high levels of testosterone during their intrauterine period (Merke & Bornstein, 2005; Meyer-Bahlburg, Dolezal, Baker, & New, 2008) and display a lower 2D:4D ratio on the right hand than females without CAH (Brown, Hines, Fane, & Breedlove, 2002; Ciumas, Hirschberg, & Savic, 2009; Ökten, Kalyoncu, & Yariş, 2002). In adulthood, women with CAH are also more likely to feel or identify as non-straight than typically developed women (Dittmann, Kappes, & Kappes, 1992; Meyer-Bahlburg et al., 2008; Zucker et al., 1996). Thus, in both clinical and nonclinical samples, a non-straight orientation in women is linked to a lower 2D:4D ratio, possibly due to elevated androgenization during prenatal development (Motta-Mena & Puts, 2017).

Compared to women, the relationship between the 2D:4D ratio and sexual orientation has been less consistent in men. Although hypothesized relationships between higher (more feminized) 2D:4D ratios and homosexuality in men have been obtained in some studies (Lippa, 2003; McFadden & Shubel, 2002), other research found either no link or found more masculinized 2D:4D ratios in non-straight than straight men (Robinson & Manning, 2000). In the meta-analysis by Grimbos et al. (2010), when comparing non-straight and straight men, no significant difference was detected in 2D:4D for either hand, Hedge's *g*'s = -.02, -17 < 95% CIs < .13. It is possible that non-straight men are exposed to the same levels of prenatal testosterone as straight men (reflected in their indistinguishable finger length ratios), but that they respond differently to testosterone, for example, via different gene regulators that may result in a non-straight orientation (Breedlove, 2017). Notably, genetic males with complete androgen insensitivity syndrome (CAIS), which renders them unresponsive to typical levels of androgens during development, are more likely to be attracted to men than women in adulthood (Hines, Ahmed, & Hughes, 2003) and show a more feminized 2D:4D ratio than control males (Berenbaum, Bryk, Nowak, Quigley, & Moffat, 2009). However, as mentioned above, in non-clinical samples, the link between 2D:4D and sexual orientation appears to be inconsistent for genetic males.

An alternative explanation is that the lack of a predictable sexual orientation difference in 2D:4D in males is simply due to statistical reasons. Let us assume, for the sake of argument, that non-straight men do have more feminized finger ratios than straight men, but that this effect is smaller than the corresponding effect for women. Hence, compared with the effect for females, the effect for males might be more prone to measurement error. If so, this effect might be better detected in a more controlled research design than used in most studies, for instance, by comparing identical twins discordant for sexual orientation. Because such comparisons are within pairs of twins, one might have more statistical power to detect predicted effects, even if they are small in magnitude. That is, although there are likely many factors that influence 2D:4D ratios (e.g., nationality or ethnicity; Grimbos et al., 2010), by using matched pairs such as identical twins, who are matched on a host of factors, including genetics, the present study controls for such error variance.

Differences in 2D:4D within Discordant Twin Pairs

To date, two studies have examined finger length ratios in identical twins with discordant sexual orientations. In 7 pairs of female identical twins, the non-straight female twins showed a significantly lower 2D:4D ratio than their straight co-twins in both hands (Hall & Love, 2003). This finding was partially replicated in another study with 8 female pairs: the non-straight twins had a lower 2D:4D ratio on their left hand than their straight co-twins, compared with their straight co-twins; however, a similar effect was not detected in the right hand (Hiraishi et al., 2012).

Within 4 male pairs with discordant sexual orientations, those who identified as non-straight had significantly higher left hand 2D:4D ratios than their straight co-twins (Hiraishi et al., 2012). This finding is consistent with the hypothesis is that the non-straight twins were exposed to lower levels of prenatal androgens than their straight co-twins and with our proposal that such effect in males could be detected if twin pairs were examined.

The Present Study

We investigated whether sexual orientation differences in 2D:4D could be found in identical twins with discordant sexual orientations. If previous findings for twins can be replicated, then, within female pairs, the non-straight twin will display a lower (or more masculinized) 2D:4D ratio than her straight co-twin (Hypothesis 1). In male pairs, the non-straight twin will display a higher, or more feminized, 2D:4D ratio than his straight co-twin (Hypothesis 2).

The main focus of the present research was to examine sexual orientation differences in finger length ratios of identical twins with discordant sexual orientations, however we also tested the effect of sex on 2D:4D. Because we hypothesized that non-straight participants have 2D:4D ratios in the direction of the opposite sex (as compared to straight participants of their sex), a sex difference in 2D:4D should be smaller between non-straight men and women than that between straight men and women (Hypothesis 3).

To test these hypotheses, one set of analyses was performed using newly collected data. A second set of analyses was then performed using a combination of these newly collected data with previously collected data (Hall & Love, 2003; Hiraishi et al., 2012) in order to increase statistical power.

Finally, previous findings have sometimes differed by hand. For example, reported sex differences are somewhat stronger in the right hand than the left hand (Breedlove, 2017; Manning, Kilduff, Cook, Crewther, & Fink, 2014). Yet, in the previous twin studies, sexual orientation differences are stronger in the left hand than the right hand (Hall & Love, 2003; Hiraishi et al., 2012). There are currently no explanations for these differences by hand. We therefore tested whether any effects were stronger in the left or right hand, but made no specific predictions about the direction of this difference.

METHOD

Participants

We advertised for identical twins with discordant sexual orientations via a newsletter at the Department of Twin Research at Kings College London, social media sites, online news sites for gay men and lesbians, and at three gay Pride festivals. Each recruited twin was encouraged to recruit the co-twin.

Participants included 18 female twin pairs and 14 male twin pairs, yielding a total of 32 pairs of identical twins with discordant sexual orientations. Twins self-identified as “straight,” “bisexual,” “gay,” or “lesbian.” They were asked twice about their sexual identities, and all responses were consistent. The number of bisexual women and men (3 and 1 individuals) was low relative to the number of straight women and men (18 and 14) and lesbians and gay men (15 and 13). Furthermore, on 7-point Kinsey Scales (Kinsey, Pomeroy, & Martin, 1948), straight participants reported exclusive or almost exclusive preferences for the other sex, gay men and lesbians reported exclusive or almost exclusive preferences for the same sex, and bisexual participants reported a stronger preference for the same sex than the other sex. For this reason, bisexual participants were grouped with gay men and lesbians into “non-straight”. Excluding bisexual individuals from the analyses conducted below did not affect the magnitude or significance of reported effects. We therefore included data of all twins, including bisexual twins, in our analyses.

For the 18 female pairs, the mean age (SD) was 28.22 years (6.81), and 89% were White, 6% were Black, and 5% were of mixed ethnicities. The mean age of the 14 male pairs was 32.00 years (12.15), and 93% were White and 7% were of mixed ethnicities.

In addition to twins repeatedly reporting that they were identical, five standardized questions about physical and visual similarity were administered to confirm the twins' monozygosity (Bailey, Dunne, & Martin, 2000). A sample question is "During childhood, could you ever have fooled friends by pretending to be your twin?" Items were assessed on scales ranging from 1 to 3, with lower scores reflecting higher similarity within twin pairs. For all twins, their average scores were below 2, suggesting monozygosity. Similar questions about zygosity, given to twins or their parents, are usually 95% accurate or higher, based on comparisons with blood group or DNA analyses (Price et al., 2012). For a subsample of twins who visited our lab, their monozygosity was further confirmed via DNA analyses from saliva samples, conducted by Genetrack Biolabs UK.

Measures and Procedures

Identical twins with discordant for sexual orientation are rare and difficult to recruit. To gather as many data as possible, we aimed for a measure of their digit ratios that did not require them to come into the lab. In fact, 13 pairs (40.62%) visited the lab, whereas 19 pairs (59.38%) participated remotely. In either case, twins were instructed to place their hands, with their palms facing upwards, on a flat surface with small gaps between their fingers. Photographs were then taken with a camera held approximately 30 cm directly above their palm. If twins participated remotely (and photographs were not taken by an experimenter), they were also sent an example photograph to use as guidance. Participants who provided unsatisfactory photographs (e.g., poor focus or resolution, or taken at an incorrect angle) were asked to retake photographs.

Collected photographs were given to three independent raters, who were masked to the sex and sexual orientation of the twins. Each rater used the open-source vector graphics package Inkscape 0.91 to measure finger lengths. Similar computer-assisted measurements have shown higher inter-rater reliability compared to other methods of measuring 2D:4D (Allaway, Bloski, Pierson, & Lujan, 2009). Specifically, each rater drew a line as wide as the finger following the lowest crease at the base of the finger, between the metacarpal and proximal phalanx. They then drew another line beginning at the tip of the finger down towards the base, using a function on the software to have this line automatically snap to the middle of the line at the base, which allowed raters to avoid guessing where the center point of the finger base was. Raters then zoomed in and finely adjusted the top of the line to match the tip of the finger as closely as possible. Lines were measured in pixels.

For each digit, inter-rater reliability (Cronbach's alpha) exceeded .98 for all straight and non-straight males and females. Therefore, the measurements for each digit of each individual twin were averaged across the three raters. These averaged measures were used to calculate the 2D:4D ratio for either hand by dividing the length of the second finger by length of the fourth finger.

A Comparison of Present and Previous Samples

From the previous studies, data from 15 pairs of discordant female twins (7 from Hall & Love, 2003; 8 from Hiraishi et al., 2012) and 4 pairs of discordant male twins (Hiraishi et al., 2012) were available. In Hall and Love, inked prints of the twins' hands were taken. Their fingers were then measured using calipers. In Hiraishi et al., finger lengths were measured via photocopies of the twins' hands also using calipers. Although types of digit measures were different across studies, they all resulted in an equivalent ratio of the second to fourth finger, and this ratio could be compared across studies. Once added to the current data set, the pooled data were from a total of 33 female pairs and of 18 male pairs, yielding a total of 51 pairs of identical twins with discordant sexual orientations.

RESULTS

Twins Recruited for the Present Study

Our first set of analyses concerned findings for twins recruited for this study. Hypothesis 1 stated that for female pairs of identical twins, the non-straight twin would have a lower 2D:4D ratio than her straight co-twin. Hypothesis 2 stated that for male pairs, the non-straight twin would have a higher 2D:4D ratio than his straight co-twin. Using mixed-factorial regression analyses, we predicted, separately by sex, 2D:4D ratios by the twins' sexual orientation as a fixed factor. Twin pairs were included as a random effect to account for repeated measures of finger length ratios within pairs.

For females, non-straight twins had significantly lower (or more masculinized) finger length ratios in the left hand than their straight co-twins, $p = .01$, β [95% CI] = -.31 [-.52, -.09]. This effect was not significant for the right hand, $p = .92$, $\beta = -.02$ [-.40, .36]. For males, no significant differences were found. If anything, non-straight twins had lower (or more masculinized) finger length ratios than their straight co-twins in the left and right hand, $p = .28$, $\beta = -.17$ [-.49, .15], and, $p = .22$, $\beta = -.26$ [-.71, .19], respectively.

To visualize these findings, we computed, within each twin pair and separately for each hand, a difference score by subtracting the finger length ratio of the straight twin from that of the non-straight twin. This resulted in a negative score if the non-straight twin had a lower, or more masculinized, finger length ratio than the straight co-twin, and a positive score if the non-straight twin had a higher, or more feminized, finger length ratio than the straight co-twin. For females, non-straight twins had significantly more masculinized finger length ratios than their straight co-twins in the left hand only (Fig. 1). For males, although not significant, non-straight males had somewhat more masculinized finger length ratios than their straight co-twins (Fig. 2).

We had further hypothesized that females would have significantly higher finger length ratios than males, and that this sex difference would be stronger between straight men and women than non-straight men and women (Hypothesis 3). A mixed-factorial regression analysis was computed, testing whether 2D:4D ratios differed by sex, sexual orientation, and their interaction. This interaction was relevant for Hypothesis 3 as it tested whether the difference between males and females was stronger for straight participants than non-straight participants. The effects of sex and sexual orientation, and their interaction, were further crossed with the effect of hand. Twins pairs were included as a random effect.

Results indicated that independent of sexual orientation and hand, females had higher finger length ratios than males, $p = .02$, $\beta = .27$ [.04, .49]. There was also a significant main effect of sexual orientation, $p = .02$, $\beta = -.17$ [-.32, -.02], which indicated that regardless of sex and hand, non-straight participants had significantly lower or more masculinized finger length ratios than straight participants. The interaction of sex and sexual orientation was not significant, $p = .73$, $\beta = .03$ [-.12, .17]. Thus, there was neither an indication that the sex difference in 2D:4D was more pronounced in straight individuals than non-straight individuals, nor was there an indication that the sexual orientation difference (with non-straight individuals having more masculine ratios) was significantly more pronounced in women than men. These patterns did not significantly differ by hand.

A Comparison of Present and Previous Samples

In our next set of analyses, we combined present data with previously published data (Hall & Love, 2003; Hiraishi et al., 2012), and conducted analyses similar to those described above. In general, effects remained similar.

We first tested, across studies, the effects of sexual orientation on finger length ratios. For each sex, a mixed factorial regression was conducted. The dependent variable was either the left hand finger ratio or the right hand finger length ratio. In each analysis, independent variables were sexual orientation and study sample as fixed effects. In addition, the model included an interaction between sexual orientation and study sample. This interaction was computed to test whether differences in finger length ratios between straight and non-straight twins varied by study. Twin pairs were included as a random effect.

Table 1 shows that, independent of study sample, non-straight female twins had more masculinized left hand finger length ratios than their straight co-twins, $p = .001$, $\beta = -.28$ [-.44, -.11]. In the right hand of female twins, there was no significant effect of sexual orientation on finger length ratio, $p = .28$, $\beta = -.11$ [-.33, .10]. Thus, Hypothesis 1 was confirmed for the left hands of females, but not their right hands. For males, independent of study, the non-straight twins did not have significantly more feminized left or right finger length ratios than their straight co-twins, $p = .99$, $\beta = .002$ [-.29, .30], and, $p = .59$, $\beta = -.12$ [-.57, .33], respectively. Thus, Hypothesis 2 was not confirmed.

Table 1 also shows there were some significant main effects of study. Whether these average differences by study on finger length ratios were meaningful is unclear. More importantly, there were no significant interactions of sexual orientation and study, suggesting that the main effects of sexual orientation applied to all available data. Figs. 3 and 4 visualize these findings across studies. Non-straight female twins had significantly more masculinized finger length ratios than their straight co-twins in the left hand, but no other sexual orientation difference in ratios was significant in women or men.

We then tested for sex differences in 2D:4D across studies. Independent of sexual orientation and hand, females had more feminized finger length ratios than males, $p = .03$, $\beta = .22$ [.03, .41]. There was also a significant main effect of sexual orientation, $p = .003$, $\beta =$ -

.17 [-.28, -.05], indicating that, regardless of sex and hand, non-straight twins had significantly lower or more masculinized left hand ratios than straight co-twins. Interactions with sex, sexual orientation, hand, or their combination were not significant. For example, the interaction of sex and sexual orientation was not significant, $p = .76$, $\beta = -.02$ [-.13, .09]. Thus, there was no indication that the sex difference in 2D:4D was more pronounced in straight individuals than non-straight individuals nor was there an indication that the sexual orientation difference (with non-straight individuals having more masculine ratios) was more pronounced in women than men. Average sex and sexual orientation differences in finger length ratios are shown in Fig. 5.

DISCUSSION

Present results suggested that non-straight females had more masculinized 2D:4D ratios than their straight co-twins, but only for their left hand. For males, no significant difference between straight twins and their non-straight co-twins was detected in either hand. Further analyses indicated that men had more masculinized finger length ratios than women, regardless of their sexual orientations, and non-straight twins had more masculinized finger ratios than straight twins, regardless of their sex. A reanalysis of present and previous data (Hall & Love, 2003; Hiraishi et al., 2012) suggested that these results were robust.

Sexual Orientation Differences

Non-straight female twins showed lower, or more masculinized, finger length ratios in the left hand (but not in the right hand) than their straight co-twins. Whether the lack of effect in the right hand is meaningful remains unclear. Firstly, it did not appear to be that robust since it was no longer detected in the multiple regression analyses. Secondly, there is no clear pattern of handedness in past work. In previous twin studies, female sexual orientation differences tend to be stronger in the left hand (Hall & Love, 2003; Hiraishi et al., 2012), although in general study populations they are, if anything, stronger for the right hand

(Grimbos et al., 2010). Similarly, sex differences tend to be stronger in the left hand in twin studies (Gobrogge, Breedlove, & Klump, 2008; Hiraishi et al., 2012), but, in general, are stronger in the right hand (Breedlove, 2017). There remains uncertainty over why patterns of laterality might differ among twins versus singletons.

If we assume female sexual orientation differences in their finger length ratios to be valid (and do not consider, for the moment, potential differences by hand), then, perhaps the found difference within female pairs was due to each twin's different prenatal environments. Approximately one-third of identical twins develop in separate placentas (Patterson, 2007), and placentas may differentially regulate the level of testosterone transferred from mother to fetus (Hines, Golombok, Rust, Johnston, & Golding, 2002). Thus, in cases of identical twins developing with separate placentas, each twin could be exposed to different levels of prenatal androgens from the maternal system. For one female twin, but not the other, exposure to elevated levels of prenatal androgens may increase the likelihood of a same-sex sexual orientation.

In men, no predicted sexual orientation differences in 2D:4D were found. This result is consistent with previous evidence suggesting that the relationship between the 2D:4D ratio and sexual orientation is less reliable in males than in females (Grimbos et al., 2010). As mentioned previously, it is possible that exposure levels to prenatal androgens do not differ between non-straight and straight males resulting in similar finger length ratios. Instead, straight and non-straight males may have different responses to the same levels of androgens. For example, their genes may be regulated differently by androgen exposure, leading to different sexual orientations (Breedlove, 2017).

Sex Differences

In general, female twins had higher or more feminized finger length ratios than male twins, indicating that they were exposed to lower levels of prenatal androgens than males.

However, contrary to Hypothesis 3, this sex difference was similar in effect across straight and non-straight men and women. This null finding could be due to a lack of statistical power. Yet, across all available data, the effect size for the interaction between sex and sexual orientation was minimal, $p = .76$, $\beta = -.02$ [-.13, .09], and it appears unlikely that adding further twins would have resulted in a significant interaction. Perhaps because the sexual orientation effect in males is generally difficult to confirm (Grimbos et al., 2010 and the present data), one cannot fully expect that a sex difference in 2D:4D is more prominent in only straight individuals than in a combination of straight and non-straight individuals.

Using Photographs for Digit Measures

The measurement of finger length ratios in pixels, as taken from photographs, provided a unique means of collecting data. Admittedly, this method potentially introduced some variability in the quality of the photographs, especially of those that were taken remotely by the twins. If photographs were particularly poor quality, participants were asked tor retake them, but there remained variation in the sharpness and resolution of the photographs, and this could have introduced measurement error. Yet, the novel method of remote data collection (via photographs) allowed for wider sampling of finger length ratio data than would have otherwise been possible with a rare study population like ours. Further use of this method could, for example, facilitate the gathering of large sample sizes across countries in cross-cultural research involving 2D:4D.

Limitations

Although studies on 2D:4D continue to grow in number, the validity of the measure is still much debated. This is partly due to the fact that 2D:4D differences in sexual orientation are in most cases small-to-modest in effect (Grimbos et al., 2010), which has raised concern over its susceptibility to measurement error (Bailey et al., 2016). Especially in small samples, unreliable measures can produce noisy data, resulting in estimated effects that are larger than what they truly are, thereby increasing the chance of Type I error (Loken & Gelman, 2017). In this respect, it is worth focusing on our one hypothesized difference which turned out to be significant: the 2D:4D difference between straight and non-straight females in their left hand. Unlike the recruited 18 pairs, almost 40 pairs would be required to achieve a power of .80 with the small effect reported in a meta-analysis, Hedge's $g = .23$ (Grimbos et al., 2010). However, across previous twin studies, the matched-pair effect size was strong, $dz = .77$ (Hall & Love, 2003; Hiraishi et al., 2012). This effect was similar in magnitude in the newly collected data, $dz = .70$. With this effect size, a power of .80 can be obtained with a minimum of 8 pairs; the newly recruited 18 female pairs exceeded this minimum. Although this calculation makes us more confident that this finding was not spurious, we cannot fully rule out the possibility of Type I error.

Another limitation of the present study is that no statistical corrections were made in the analyses conducted. We note, however, that one set of our calculations were multiple regression analyses which pointed to significant sexual orientation differences while considering multiple comparisons. Because of the statistical adjustments of these multiple regression analyses, we have not further adjusted the alpha level in our simple comparisons between groups. In theory, we could have reduced the results section to include only the complex (and statistically superior) regression analyses, but we consider the simple comparisons to be informative and to aid interpretation of findings.

A final limitation of the present study is a potential selection bias. The majority of non-straight twins openly identified as such. Perhaps non-straight individuals who are "out" are more likely to show gender nonconformity (a correlate of homosexuality; Lippa, 2005; Watts, Holmes, Raines, Orbell, & Rieger, 2018), because their expression makes it more difficult to be closeted about their sexual orientation. If gender nonconformity links to sex-atypical digit ratios, then perhaps individuals who are "out" and participated were also biased toward sex-atypical digit ratios. We did collect data on gender nonconformity and computed correlations with finger length ratios. Gender nonconformity was measured using standardized questionnaires (Rieger, Linsenmeier, Gygax, & Bailey, 2008) in addition to observer ratings of gender nonconformity as seen in photographs of participants (Watts et al., 2018). Correlations between self-reported and observer rated gender nonconformity and 2D:4D were weak and not significant (results not shown). Thus, at least in the present sample, 2D:4D did not relate to gender nonconformity. However, we cannot rule out that participating twins were unusual in either gender-atypical traits or in other, unknown ways, as compared to twins who did not take part in the research.

Conclusion

At least for females, found differences in 2D:4D within pairs of identical twins with discordant sexual orientations emphasize the potential relevance of prenatal androgen exposure in the development of sexual orientation, and this independent of one's genetic makeup. Additional work using other indices of prenatal androgen exposure, including measurement of testosterone levels in amniotic fluid (Auyeung et al., 2009), ano-genital distance (Pasterski et al., 2015), and oto-acoustic emissions (Rahman, 2005) could provide further insight into the development of discordant sexual orientations in genetically identical individuals.

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Compliance with Ethical Standards. The University of Essex's Ethics Committee approved this study ("GR1303").

Conflict of Interest. All authors declare that they have no conflict of interest.

Ethical Approval. All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

Informed Consent. Informed consent was obtained from all individual participants included in the study.

Table 1.

Multiple Regression Analyses for Sexual Orientation and Study predicting Left and Right Hand Ratios for 66 Female and 36 Male Twins.

Measure	Females Left[1]	Females Right[1]	Males Left[1]	Males Right[1]
Sexual Orientation (SO)[2]	-.28 [-.44, .11]*	-.12 [-.33, .10]	.002 [-.29, .30]	-.18 [-.57, .33]
Hall & Love (2003)[3]	.46 [.17, .72]*	.16 [-.15, .46]	N/A	N/A
Hiraishi et al. (2012)[3]	-.78 [-1.09, -.47]***	-.75 [-1.09, -.41]***	-.48 [-.97, -.002] †	-.32 [-.69, .06] †
SO X Hall & Love (2003)[3]	-.02 [-.26, .23]	-.11 [-.43, .21]	N/A	N/A
SO X Hiraishi et al. (2012)[3]	.05 [-.19, .29]	.02 [-.31, .35]	.16 [-.13, .46]	.17 [-.28, .62]

Note. Numbers are standardized regression coefficients, *β's*, with 95% confidence intervals in brackets. [1]Higher scores indicate higher or more feminine ratios. [2]A score of 0 indicates "straight," 1 indicates "non-straight". [3]Statistics reflect contrasts, comparing the main effect or interaction of the previous study to the main effect or interaction in the newly collected data. Twin pairs were a random effect. †$p < .10$. *$p < .05$. ***$p < .0001$

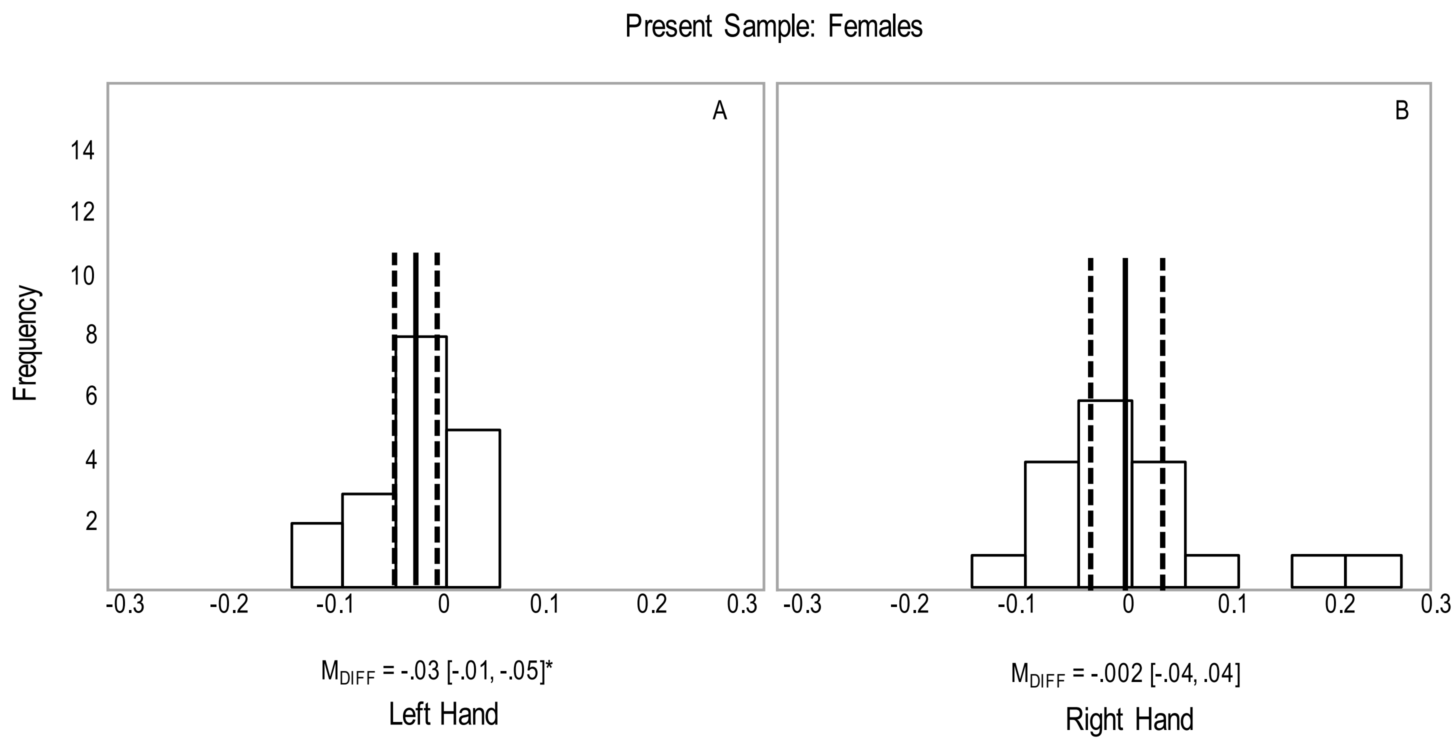

Figure 1. For newly collected data, distributions of differences in 2D:4D ratios between non-straight female twins and their straight co-twins. A shows the left hand and B the right hand. Solid black lines represent the mean of the distribution of difference scores, and the dashed lines represent the 95% confidence intervals of the mean difference. Statistics represent mean differences against zero (M_{DIFF}) with their 95% confidence intervals. * $p < .05$

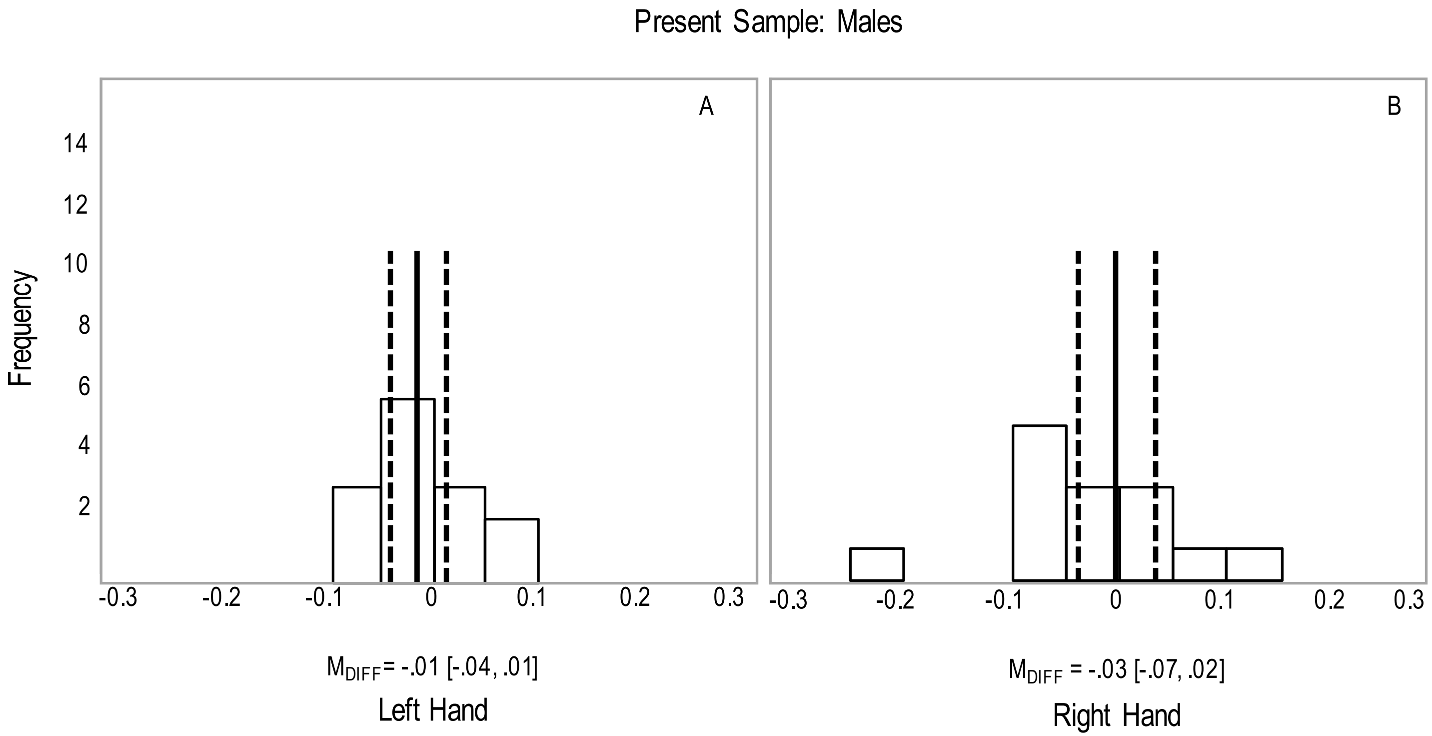

Figure 2. For newly collected data, distributions of differences in 2D:4D ratios between non-straight male twins and their straight co-twins. A shows the left hand and B the right hand. Solid black lines represent the mean of the distribution of difference scores, and the dashed lines represent the 95% confidence intervals of the mean difference. Statistics represent mean differences against zero (M_{DIFF}) with their 95% confidence intervals.

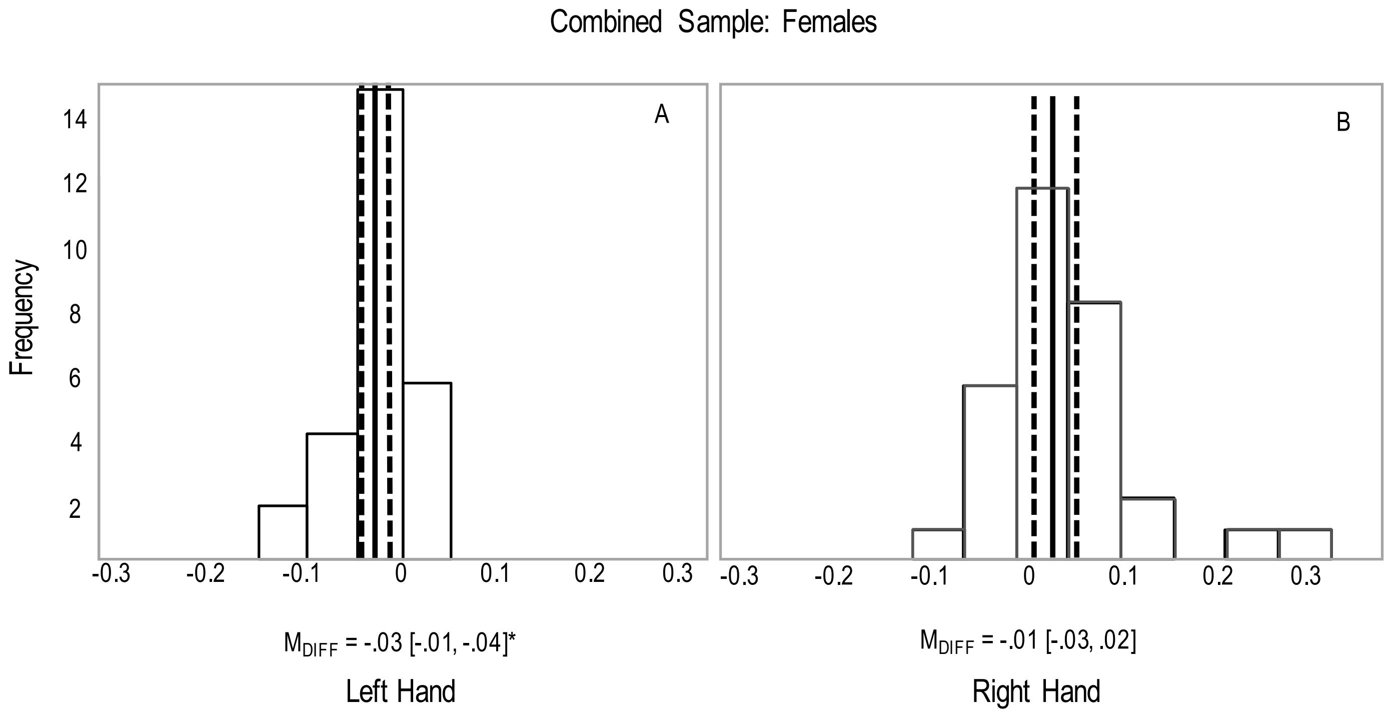

Figure 3. For a combination of newly collected and previous data, distributions of differences in 2D:4D ratios between non-straight female twins and their straight co-twins. A shows the left hand and B the right hand. Solid black lines represent the mean of the distribution of difference scores, and the dashed lines represent the 95% confidence intervals of the mean difference. Statistics represent mean differences against zero (M_{DIFF}) with their 95% confidence intervals. * $p < .05$.

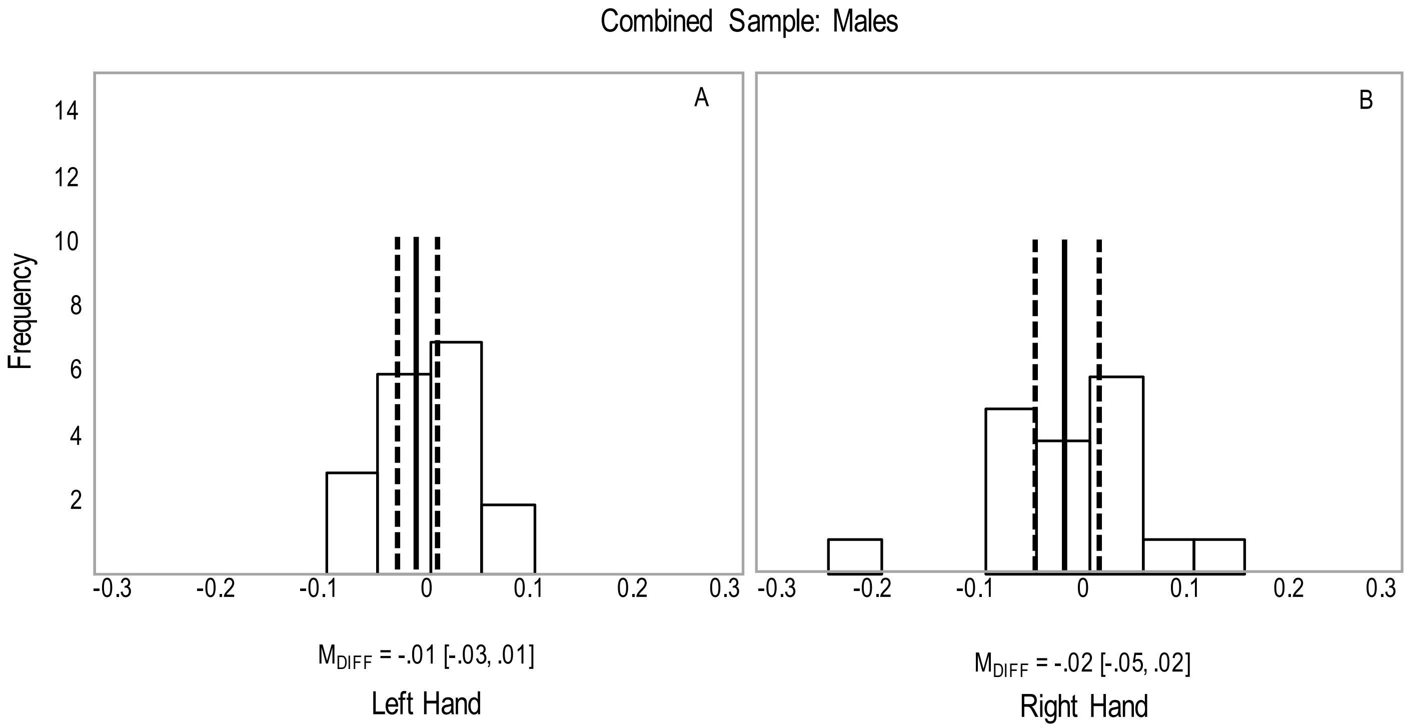

Figure 4. For a combination of newly collected and previous data, distributions of differences in 2D:4D ratios between non-straight male twins and their straight co-twins for A, the left hand and B, the right hand. Solid black lines represent the mean of the distribution of difference scores, and the dashed lines represent the 95% confidence intervals of the mean difference. Statistics represent mean differences against zero (M_{DIFF}) with their 95% confidence intervals.

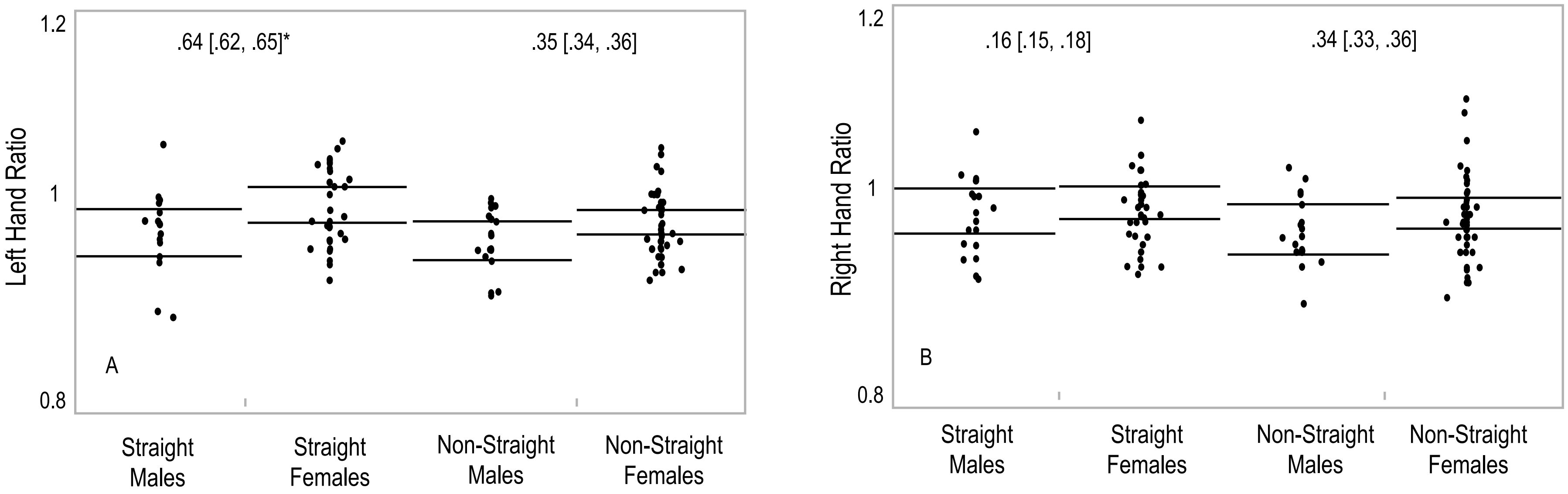

Figure 5. Sex differences in 2D:4D. Panels A and B show sex differences in the left and right hands, respectively, between 18 straight males and 33 straight females, and between 18 non-straight males and 33 non-straight females. Dots represent finger length ratios of individual twins, averaged across all ratings. Lines are the means' 95% confidence intervals. On the y-axis, higher scores indicated a higher second to fourth finger ratio. Numbers represent Hedge's *g's* with their 95% confidence intervals. * $p < .05$.

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