Asian Journal of Andrology (2013) 15, 152–154

www.nature.com/aja

SHORT COMMUNICATION

Sperm speed is associated with sex bias of siblings in a human population

Jim A Mossman[1,2], Jon Slate[2], Tim R Birkhead[2], Harry D Moore[3] and Allan A Pacey[4]

Recent studies investigating possible causes of male subfertility have largely focused on how lifestyle or environmental factors impact on the process of spermatogenesis. Markedly, fewer studies have investigated those risk factors that result in reduced sperm quality, such as poor sperm motility. The speed at which sperm swim is a major predictor of fertility and is extremely variable in human populations. It has been hypothesized that offspring sex may be adaptively manipulated to maximize the offspring's reproductive fitness (e.g., parents with genes for good male fertility traits, such as high sperm speed, would produce primarily sons and fewer daughters because the offspring will inherit advantageous male fertility genes). Conversely, parents with poor male fertility genes would produce primarily daughters. We tested whether there was an association between how fast a man's sperm swam and the sex bias of his siblings in a sample of men attending clinic for fertility investigations with their partner and with a wide range of semen characteristics, including sperm speed. We found that the sex bias of a man's siblings is associated with his sperm speed; men with female-biased siblings had significantly slower sperm (judged using computer-assisted sperm analysis (CASA)) than men from male-biased sibships. This observation suggests family composition is an important factor that needs to be considered in future epidemiological and clinical studies of human fertility.
Asian Journal of Andrology (2013) **15,** 152–154; doi:10.1038/aja.2012.109; published online 3 December 2012

Keywords: computer-assisted sperm analysis (CASA); fertility; intralocus sexual conflict; male; offspring; sex ratio; sexual antagonism

INTRODUCTION

In most animal species, sperm motility (their ability to swim) is vital for fertilisation success and is highly variable within populations.[1] Despite its critical role, the identification of genetic and environmental factors responsible for fast or slow sperm motility remains poorly understood.

In some species, it is known that the sex ratio of offspring (number of males/sibling size) can be facultatively adjusted to maximize offspring fitness.[2] In the context of this study, the term 'fitness' is used to describe fertility potential, although more generally the term captures multiple evolutionarily important traits such as number of offspring, health, longevity, etc. In certain environmental conditions, it may be beneficial to produce mainly males and in alternative environments it may be beneficial to produce mainly females,[3] causing sexual antagonism for offspring gender. The ability of mammalian mothers to adjust the sex ratio of their offspring is poorly understood although empirical and theoretical studies in other species have shown that: (i) it is possible;[4] and (ii) it is adaptive.[2] Little is known, however, whether humans are able to manipulate the sex ratio of offspring to maximize their reproductive success and that of their offspring. Furthermore, to our knowledge, no study to date has investigated possible associations of an index of reproductive fitness and sibling composition.

One important component of lifetime fitness in humans is fertility. Variation in a reproductive trait, such as sperm motility, has been shown to correlate with fertility outcomes and therefore reproductive success.[5] As part of ongoing investigations into maternal effects on sperm motility, the aim of the present study was to determine if components of family composition—the number of brothers, sisters, and the sibling sex bias[6]—were associated with the swimming speed of a man's sperm.

MATERIALS AND METHODS

Fresh ejaculates and questionnaire data were obtained from 500 men attending an andrology laboratory in South Yorkshire, UK.[7] From all the samples tested, only men with clinically normal sperm parameters, judged using World Health Organization (WHO) reference values,[8] were included in the main analysis (n=129, **Table 1**). However, we also conducted an identical analysis on the larger cohort of men (n=463; **Supplementary Tables 1 and 2** and **Supplementary Figure 1**) to test for the robustness in a larger sample including men who were likely to be subfertile.

During the questionnaire interview, men were asked how many biological (maternal) brothers and sisters they had. Since 95% of the samples' mothers in the smaller cohort were at least 50 years old and unlikely to have further children, we considered that the number of siblings recorded at the time of investigation would not change appreciably. Sibling sex bias (from 1 : 1) was estimated as number of brothers minus number of sisters ('brothers–sisters'). Calculating the sex bias in this way allowed us to include men with no siblings (n=30).

[1]Department of Ecology and Evolutionary Biology, Box G, Brown University, Providence, Rhode Island 02912, USA; [2]Department of Animal and Plant Sciences, University of Sheffield, Western Bank, Sheffield S10 2TN, UK; [3]Centre for Stem Cell Biology, Department of Biomedical Sciences, University of Sheffield, Western Bank, Sheffield S10 2TN, UK and [4]Academic Unit of Reproductive and Developmental Medicine, Department of Human Metabolism, School of Medicine and Biomedical Sciences, Sheffield S10 2SF, UK
Correspondence: Dr JA Mossman (Jim_Mossman@brown.edu)
Received: 18 May 2012; Revised: 30 July 2012; Accepted: 24 August 2012; Published online: 3 December 2012

Table 1 A description of the sample (n=129)

Descriptor	*Mean*	*Standard deviation (s.d.)*	*Range*
Participant age (year)	34.0	5.6	20.0–51.0
Sperm concentration ($\times 10^6$ ml^{-1})	71.7	43.2	23.0–304.0
Morphology (strict criteria) (%)	9.82	3.65	2.00–17.50
Number of brothers	0.87	0.96	0–6
Number of sisters	0.88	0.96	0–4
Number of brothers minus sisters	−0.02	1.44	−4–6
Number of brothers plus sisters	1.75	1.26	0–6
Sperm speed (PC1)	0.00	11.16	−26.37–26.96

To estimate how fast sperm swam, we used computer-assisted sperm analysis (CASA) (see the Supplementary Information for a detailed description of the protocol). A principal component analysis was conducted to reduce the complexity of colinear sperm motility variables. The principal axis method was used to extract the components of the principal component analysis, and this was followed by a varimax (orthogonal) rotation. Sperm motility variables obtained during CASA and used in the principal component analysis were as follows: (i) curvilinear velocity; (ii) average path velocity; (iii) straight line velocity; (iv) amplitude of lateral head displacement; and (v) beat cross frequency. The first principal component (PC1) explained 72.3% of the variance between these variables and was used as an index of sperm speed. Furthermore, PC1 was most highly correlated with average path velocity (r=0.94, t=30.44, df=127, P<0.0001), a significant predictor of *in vitro* fertility in humans.[9] A full description of the study protocols can be found in the Supplementary Information.

RESULTS

For the men with clinically normal sperm parameters (n=129), there was a positive relationship between sperm speed and the number of brothers minus the number of sisters ($F_{1,124}$=7.62; P=0.007; **Figure 1** and **Table 2**). There was, however, no relationship between sperm speed and the number of brothers plus sisters, the sibship size ($F_{1,124}$=1.09; P=0.30; **Table 2**). Therefore, men from larger sibships did not have faster sperm than men from smaller sibships, but men with more male-biased siblings tended to have faster sperm. This relationship was not driven solely by the number of male or the number of female siblings, as these terms explained less variation (R^2) of PC1 than the model fitting 'brothers–sisters'. Number of brothers was not associated with PC1 ($F_{1,124}$=1.72, P=0.19), but the number of sisters was associated with PC1 ($F_{1,124}$=7.14, P=0.009).

For the larger cohort of men (n=463), the results of the analyses were qualitatively similar, although less of the variance in PC1 was explained by the components of family composition (**Supplementary Table 2**). For both cohorts of men, we found no association between the proportions of the ejaculate that swam with WHO 1999 (A+B) criteria and any measure of sibling composition (data not shown).

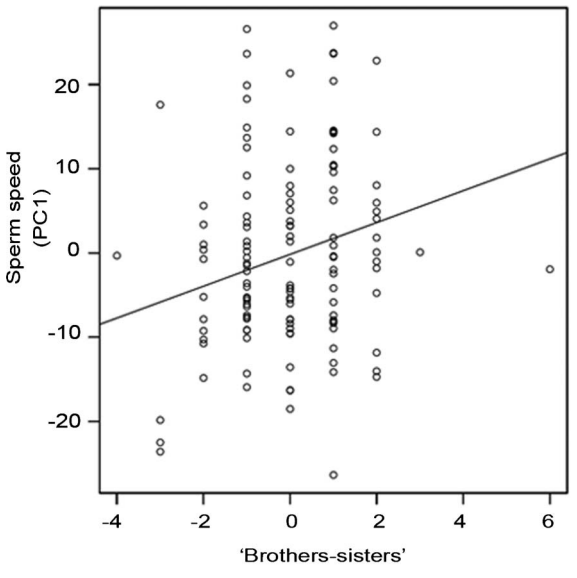

Figure 1 Sperm speed (PC1) and sibling composition ('brothers–sisters') in 129 men (smaller cohort). There was a significant positive correlation between PC1 and 'brothers–sisters' (**Table 2**).

DISCUSSION

What explains the relationship between male-biased siblings and sperm speed? It has been suggested that a fraternal birth order effect, whereby a maternal immune response against 'masculinizing' H–Y genes of focal males with older male siblings, can influence some male traits, such as the probability of homosexuality.[10] However, similar arguments cannot explain our data because: (i) men with more brothers have faster, not slower sperm; and (ii) we also found a negative relationship between sperm speed and the number of sisters a man has ($F_{1,124}$=7.14, P=0.009, **Table 2**), whereas the fraternal birth order hypothesis predicts no relationship. Instead, we favor an evolutionary explanation, which can be explained as follows.

There is considerable evidence, from experiments,[11,12] meta-analyses[13] and studies of wild populations[14] of different species that there is a negative genetic correlation between male and female fitness-related traits. In other words, genes that are beneficial for fitness in males can be deleterious for fitness in females (and *vice versa*), a concept termed 'intralocus sexual conflict'. As a result, parents could ultimately benefit from producing the sex of offspring that would have the maximum fitness given the compliment of genes that they will inherit. Furthermore, it is known that in many species, the offspring sex ratio can be adaptively manipulated to maximize the fitness of offspring.[2,15] If we assume that in humans, sperm speed is an indicator of male fitness (fertility), which has been shown,[9] and that there is a negative genetic correlation between male and female fitness, then females that carry genes for high sperm speed (and/or females who mate with males with genes for high sperm speed) should produce male-biased sibships. Similarly, females with genes for low sperm speed should produce female-biased sibships. Therefore, there should be a positive relationship between a man's sperm speed and the proportion of his siblings that are males. This is exactly the pattern we observe. Note that this explanation does not require any causality between sibling sex bias and sperm speed and rests only on assumptions with good empirical support. There is no requirement for genes that influence sperm speed to be sex-linked.

Table 2 Models of sibling sex associations with sperm motility (PC1) in humans. All 129 men were included in each model. Four models are presented corresponding to: (i) the number of brothers—number of sisters of a focal man; (ii) the number of brothers+sisters; (iii) the number of sisters; and (iv) the number of brothers. Model degrees of freedom (DF) are shown

Model	R^2	F *value*	P *value* (>F)	*Coefficient* (±*s.e.*)	T	P *value* (>T)
Number of brothers–sisters		$DF_{(3,124)}$				
Sperm concentration ($\times 10^6$ ml^{-1})	0.040	5.90	0.017*	−1.34 (0.44)	−3.03	0.003*
Sperm morphology	0.065	9.61	0.002*	0.72 (0.24)	3.03	0.003*
Brothers–sisters	0.052	7.62	0.007*	1.76 (0.64)	2.76	0.007*
Model explained	0.157					
Number of brothers+sisters		$DF_{(3,124)}$				
Sperm concentration ($\times 10^6$ ml^{-1})	0.040	5.61	0.019*	−1.35 (0.45)	−2.99	0.003*
Sperm morphology	0.065	9.13	0.003*	0.72 (0.24)	2.95	0.004*
Brothers+sisters	<0.01	1.09	0.30	−0.74 (0.71)	−1.05	0.30
Model explained	0.113					
Number of sisters		$DF_{(3,124)}$				
Sperm concentration ($\times 10^6$ ml^{-1})	0.040	5.89	0.017*	−1.35 (0.44)	−3.06	0.003*
Sperm morphology	0.065	9.58	0.002*	0.70 (0.24)	2.93	0.004*
Number of sisters	0.049	7.14	0.009*	−2.43 (0.91)	−2.67	0.009*
Model explained	0.154					
Number of brothers		$DF_{(3,124)}$				
Sperm concentration ($\times 10^6$ ml^{-1})	0.040	5.64	0.019*	−1.34 (0.45)	−2.96	0.004*
Sperm morphology	0.065	9.18	0.003*	0.75 (0.24)	3.06	0.003*
Number of brothers	0.012	1.72	0.19	1.29 (0.99)	1.31	0.19
Model explained	0.117					

*$P<0.05$.

A previous study in humans provided some evidence for a positive association between sperm speed, judged as curvilinear velocity and average path velocity, and the offspring sex.[16] Balli *et al.*[16] found that faster sperm were associated with female offspring, contrary to our results in the present study, where we found that increased numbers of sisters were associated with slower sperm. However, we did not directly test for associations between offspring sex and sperm speed. Instead, we observed positive associations between the sex bias in the focal man's siblings and his sperm speed, suggesting a possible negative genetic correlation between the genes that make sperm fast or slow and the offspring gender. Neither the present study nor that of Balli *et al.*[16] found any association between the proportion of sperm that swam with WHO (A+B) characteristics, and either sibling sex composition or offspring gender, respectively.

Interestingly, we observed the same results regardless of whether the men were from the cohort with good sperm parameters, judged using WHO (1999) thresholds,[8] or were possibly subfertile. It is therefore likely that the results are robust even when possibly subfertile men are included in the analysis.

Future studies are required to identify if this is a universal relationship between human populations and across species, and to unravel the mechanisms underpinning the link between sibling sex bias and sperm speed. However, our data provide good support for intralocus sexual conflict, influencing both male fertility and offspring sex bias in humans.

AUTHOR CONTRIBUTIONS

JAM conceived the study, collected the data, obtained participant consent and conducted the analyses. JS provided analytical assistance and conceptual advice. HDM and AAP provided help for participant recruitment. AAP, HDM and TRB provided technical assistance for CASA. All authors contributed to the manuscript preparation.

COMPETING FINANCIAL INTERESTS

The authors declare no competing financial interests.

ACKNOWLEDGMENTS

We are grateful to the participants in this study, the staff at the Andrology Laboratory in the Jessop Wing of Sheffield Teaching Hospitals, and to A-M Hernandez, E Villa-Cuesta and B Woodward for comments on the manuscript. Geoff Hobson provided technical assistance for CASA setup. Jim A. Mossman was funded by a University of Sheffield PhD studentship.

Supplementary Information accompanies the paper on *Asian Journal of Andrology*'s website (http://www.nature.com/aja).

1 Birkhead TR, Hosken DJ, Pitnick S. Sperm Biology: An Evolutionary Perspective. Oxford: Academic Press; 2009. p567.
2 Trivers RL, Willard DE. Natural selection of parental ability to vary the sex ratio of offspring. *Science* 1973; **179**: 90–2.
3 Pryke SR, Rollins LA, Griffith SC. Context-dependent sex allocation: constraints on the expression and evolution of maternal effects. *Evolution* 2011; **65**: 2792–9.
4 Katsuki M, Harano T, Miyatake T, Okada K, Hosken DJ. Intralocus sexual conflict and offspring sex ratio. *Ecol Lett* 2012; **15**: 193–7.
5 Hirano Y, Shibahara H, Obara H, Suzuki T, Takamizawa S *et al*. Relationships between sperm motility characteristics assessed by the computer-aided sperm analysis (CASA) and fertilization rates *in vitro*. *J Assist Reprod Genet* 2001; **18**: 213–8.
6 Charnov EL. The Theory of Sex Allocation. Princeton, NJ: Princeton University Press; 1982. pp7–20.
7 Mossman JA, Slate J, Birkhead TR, Moore HD, Pacey AA. Mitochondrial haplotype does not influence sperm motility in a UK population of men. *Hum Reprod* 2012; **27**: 641–51.
8 WHO. WHO Laboratory Manual for the Examination of Human Semen and Sperm–cervical Mucus Interaction. Cambridge: Cambridge University Press; 1999. pp60–1.
9 Liu DY, Clarke GN, Baker HW. Relationship between sperm motility assessed with the Hamilton–Thorn motility analyzer and fertilization rates *in vitro*. *J Androl* 1991; **12**: 231–9.
10 Blanchard R, Klassen P. H–Y antigen and homosexuality in men. *J Theor Biol* 1997; **185**: 373–8.
11 Rice WR. Sexually antagonistic male adaptation triggered by experimental arrest of female evolution. *Nature* 1996; **381**: 232–4.
12 Chippindale AK, Gibson JR, Rice WR. Negative genetic correlation for adult fitness between sexes reveals ontogenetic conflict in Drosophila. *Proc Natl Acad Sci USA* 2001; **98**: 1671–5.
13 Poissant J, Wilson AJ, Coltman DW. Sex-specific genetic variance and the evolution of sexual dimorphism: a systematic review of cross-sex genetic correlations. *Evolution* 2010; **64**: 97–107.
14 Foerster K, Coulson T, Sheldon BC, Pemberton JM, Clutton-Brock TH *et al*. Sexually antagonistic genetic variation for fitness in red deer. *Nature* 2007; **447**: 1107–10.
15 Kruuk LE, Clutton-Brock TH, Albon SD, Pemberton JM, Guinness FE. Population density affects sex ratio variation in red deer. *Nature* 1999; **399**: 459–61.
16 Balli KS, Patton WC, Jacobson JD, Corselli J, King A *et al*. Sperm velocity in seminal plasma and the association with gender of offspring. *Arch Androl* 2004; **50**: 37–40.