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Authors

Mank, JE

Hall, DW

Kirkpatrick, M

et al.

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Sex chromosomes and male ornaments: a comparative evaluation in ray-finned fishes

Judith E. Mank^{1,*}, David W. Hall², Mark Kirkpatrick² and John C. Avise³

¹*Department of Genetics, Life Sciences Building, University of Georgia, Athens, GA 30602, USA*

²*Section of Integrative Biology, 1 University Station C-0930, University of Texas, Austin, TX 78712, USA*

³*Department of Ecology and Evolutionary Biology, University of California, Irvine, CA 92697, USA*

Theory predicts that the mechanism of genetic sex determination can substantially influence the evolution of sexually selected traits. For example, female heterogamety (ZZ/ZW) can favour the evolution of extreme male traits under Fisher's runaway model of sexual selection. We empirically test whether the genetic system of sex determination has played a role in the evolution of exaggerated male ornaments in actinopterygian fishes, a clade in which both female-heterogametic and male-heterogametic systems of sex determination have evolved multiple times. Using comparative methods both uncorrected and corrected for phylogenetic non-independence, we detected no significant correlation between sex-chromosome systems and sexually selected traits in males. Results suggest that sex-determination mechanism is at best a relatively minor factor affecting the outcomes of sexual selection in ray-finned fishes.

Keywords: female preference; genetic sex determination; secondary sexual characters

1. INTRODUCTION

Sexual selection via female preferences has promoted the evolution of elaborate male ornaments in many animal groups. One evolutionary mechanism that can promote the exaggeration of male display traits is indirect selection, in which a female preference evolves because of a genetic correlation that it naturally develops with the male display (reviewed in Kirkpatrick & Ryan 1991). Under a Fisherian runaway process, if the genetic correlation exceeds a critical value, then the female preference and male display can coevolve at an explosive rate (Fisher 1952; Lande 1981). Under a good-genes process, if the male display is genetically correlated with traits that are under positive directional selection then female preference genes that become correlated with male display genes will also become associated with good genes. Both Fisher's runaway and the good-genes processes of sexual selection rely critically on genetic correlations between female preferences and genes either for male displays (in the runaway) or enhanced lifetime fitness (in good-genes). Recent theoretical work has considered how sex linkage might influence these correlations and thereby impact the evolution of sexually selected traits (Hastings 1994; Kirkpatrick & Hall 2004).

Some animal clades, such as mammals, have male heterogamety (that is, males are XY and females XX), while in others, such as birds, females are heterogametic (males are ZZ and females ZW). The models show that Z-linkage of female preference is especially conducive to a Fisherian runaway, whereas X-linkage of female preference coupled with autosomal inheritance of male displays can favour the operation of a good-genes process. These effects can be substantial (see Table 1 of Kirkpatrick &

Hall 2004). Furthermore, the sex chromosomes in some organisms may harbour a disproportionately large fraction of the total genetic variation for male traits and perhaps for female mating preferences (Prowell 1998; Reinhold 1998; Iyengar *et al.* 2002). It thus follows that lineages with different modes of genetic sex determination might show different tendencies to evolve exaggerated male traits. Unfortunately, very few empirical tests of these theoretical predictions are available in the literature. Furthermore, a reporting bias probably exists wherein positive associations between sex chromosome system and sexually selected traits (Iyengar *et al.* 2002; Reeve & Pfennig 2003) may have appeared in print more often than outcomes in which no such empirical relationship was detected. These factors have complicated efforts to assess any general relationship that might exist between male heterogamety and good-genes processes, or between female heterogamety and Fisherian runaway.

Among major vertebrate clades, ray-finned fishes (Actinopterygii) are unrivaled in the evolutionary lability of their sex-determination mechanisms (Solari 1994; Graves & Shetty 2001; Woram *et al.* 2003; Mank *et al.* *in press*; Volf 2005). ZW and XY (as well as other) modes of sex determination have each arisen multiple times and inter-converted recurrently in actinopterygian lineages (Mank *et al.* *in press*), thus making these fish ideal for testing hypothesized associations between exaggerated male ornaments and alternative systems of sex determination. Also, the recent construction of a provisional actinopterygian supertree (Mank *et al.* 2005) now makes it possible to examine the empirical correlation between male ornaments and sex chromosomes in a comparative phylogenetic context. Here, we examine the association between sex determination and the outcome of sexual selection (male ornaments) in ray-finned fishes in order to evaluate the models of indirect selection reviewed above.

* Author for correspondence (jemank@uga.edu).

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Table 1. Statistical relationships between chromosomal mode of sex determination and male ornamentation in 154 surveyed species of actinopterygian fishes.

analysis	correction for phylogeny?	test statistic	significance
qualitative (presence versus absence of male ornaments)	no	$r=0.043$	$p=0.504$
	yes (by DISCRETE)	likelihood ratio=3.201	$p=0.525$
quantitative (number of male ornament types)	no	$r=0.041$	$p=0.606$
	yes (by MULTI-STATE)	likelihood ratio=0.329	$p=0.994$

2. MATERIAL AND METHODS

We assembled a bibliographic database on sex-chromosome-based modes of sex determination and male sexually selected characters in 154 species of actinopterygian fishes (the raw data are provided in electronic supplementary material). We focused on male ornament traits that have been shown repeatedly to be the result of female choice in various fish taxa. These include published descriptions of elongate fins (Harrington 1997; Marcus & McCune 1999; Kuwamura *et al.* 2000), breeding tubercles (Kortet *et al.* 2003, 2004), sexual dichromatism defined as nuptial colorations expressed more noticeably in males than in females of a species (Reimchen 1989; Houde & Endler 1990; Stott & Poulin 1996; Amundsen & Forgren 2001), and electric mating calls (Curtis & Stoddard 2003). We omitted from our database sexually dimorphic traits such as gonopodia and body-size differences that are not unambiguously a consequence of female choice (and for which male–male sexual selection and/or natural selection may largely be responsible). Regarding sexual dichromatism, we did not tally counts of a particular ornament type exhibited by males (specific numbers of stripes, spots, patches, etc.), but simply recorded, from published descriptions in field guides and species accounts, whether males displayed any such traits and if so how many such different trait types. This approach is conservative because it avoids overestimating numbers of ‘independent’ traits (Reeve & Pfennig 2003) that in some studies have proved to be pleiotropically related (Fitzpatrick 2004).

Our statistical analyses entailed both qualitative and quantitative appraisals of male ornaments, and were conducted both uncorrected (Ricklefs 1996; Price 1997; Harvey & Rambaut 1998) and corrected (Felsenstein 1985; Harvey & Pagel 1991) for phylogenetic non-independence. In the qualitative assessment, each species was scored for presence versus absence (in published reports) of any sexually selected ornaments; and in the quantitative assessment, each species was scored for total numbers of different male ornament types. For each of these data treatments, phylogeny was either ignored or explicitly accommodated as follows.

First, we treated all 154 surveyed species as independent observations, i.e. without regard to their phylogenetic associations. We calculated the correlation (r) between sex-chromosome type, and presence and number of male ornaments. We also tested for random association using Fisher’s exact test (for presence or absence data) and χ^2 tests (for numbers of male ornament types).

Second, to correct for phylogeny, we used the supertree topology from Mank *et al.* (2005), which we augmented with genus- and species-level phylogenies when a given taxonomic family was polymorphic for sex-chromosome system. These lower-level augmenting phylogenies, each based on robust analyses that yielded well-resolved tree topologies, were included for Cyprinidae (Briolay *et al.* 1998; Cunha *et al.*

2002), Loricariidae (Armbruster 2004), Salmonidae (Phillips *et al.* 2004), Gobiidae (Penzo *et al.* 1998; Thacker 2003), Fundulidae (Grady *et al.* 2001) and Poeciliidae (Lydeard *et al.* 1995; Meyer 1997; Breden *et al.* 1999; Ghedotti 2000).

The cladogram was analysed for possible trait correlations using the maximum likelihood program DISCRETE for presence/absence data, and MULTI-STATE for quantitative data (Pagel 1994, 1997). In the presence/absence analysis, we compared the model of correlated evolution with a null model of independent evolution between male ornaments and chromosomal sex-determination mode (likelihood ratio test, χ^2 distribution with four degrees of freedom). For the quantitative analysis, we compared the correlated model with a null model in which the evolutionary rate of male ornament acquisition was equal for both of the sex-chromosome types (likelihood ratio test, χ^2 distribution with three degrees of freedom). In the absence of sufficient information to date all internal nodes of the supertree, all branch lengths were coded as equal.

3. RESULTS AND DISCUSSION

In none of our data analyses could we reject the null hypothesis of no significant relationship between sex-chromosome type and exaggerated male ornamentation in actinopterygian fishes (tables 1 and 2). Indeed, no test result was even marginally significant. Thus, our analysis suggests that female-heterogametic (ZZ–ZW) lineages are not significantly more or less prone to male ornamentation than male-heterogametic (XY–XX) lineages in these fishes. A previous empirical test (Reeve & Pfennig 2003) found a weak positive association between female heterogamety and the number of male ornaments in fish, but that analysis involved many fewer species (29) and the correlation was not statistically significant.

Several reservations about our findings deserve mention. Perhaps our analyses simply lacked the statistical power to detect weak correlations that nonetheless exist. Or, perhaps the (inevitably) provisional and incomplete structure of the supertree employed, or its lack of information on branch lengths, somehow obscured a positive evolutionary association between sex-chromosome systems and sexually selected traits. However, because the evolution of both sex-determination mode and male ornaments under sexual selection are rapid in fishes (see below), most of the still-detectable evolutionary effects of sex-chromosome changes should be concentrated near branch tips of the supertree, rather than in deeper portions where phylogenetic uncertainties might often be greatest.

Apart from such ‘technical’ concerns, complicating biological and evolutionary factors might also have come into play. First, some male ornaments may be more the result of male–male competition than of female choice (Andersson

Table 2. Fisher's exact test (presence-absence data) and chi-squared test (quantitative data) for possible associations between male ornaments and sex-chromosome system in 154 species of actinopterygian fishes. The body of each table shows numbers of species observed (and expected under the null hypothesis of random association) to display various combinations of these traits.

male ornaments	sex chromosome system	
	ZZ-ZW	XX-XY
absent	42 (39.2)	62 (64.8)
present	16 (18.9)	34 (31.2)
	d.f. = 1, $p > 0.2$	
no ornaments	42 (39.2)	62 (64.8)
one ornament type	7 (10.6)	21 (17.4)
two ornament types	9 (8.3)	13 (13.7)
	$\chi^2 = 2.38, p > 0.2$	

1994; Gould & Gould 1997), and this would lower any expected correlation between sex-determination mode and exaggerated male traits. Second, although male display genes are sex-linked in some clades (Prowell 1998; Reinhold 1998; Lindholm & Breden 2002), they are primarily autosomal in others (Ritchie & Phillips 1998; Fitzpatrick 2004), and unfortunately their genetic bases remain completely unknown in most fish taxa (a conspicuous exception being the Poeciliidae; Lindholm & Breden 2002). If preference and display trait genes are often autosomal in the Actinopterygii, any evolutionary effects of sex linkage for the remaining genes might be difficult to detect.

Third, a general pattern may have failed to emerge because the mode of indirect selection (Fisherian, good-genes or otherwise) has varied across actinopterygian taxa. To test this possibility, analyses that examined subsets of the full phylogeny could be employed (at least in principle), but the trade-off would be a serious loss of statistical power with the fewer comparisons possible. Fourth, the prediction that sex linkage can have a substantial effect on the evolution of male characteristics assumes that quantities such as genetic variances in male displays and female preferences do not vary in a systematic way with sex linkage (Kirkpatrick & Hall 2004). If this assumption is incorrect, then all predictions would have to be altered accordingly.

A fifth potential concern is that sex-determination systems in fishes might have changed states more rapidly than the male ornaments they theoretically influenced. However, this seems unlikely. Although mechanisms of sex determination are indeed highly labile during fish evolution (Mank *et al.* in press), evidence for particular taxa (e.g. *Poecilia* and *Xiphophorus*) suggests that rates of male ornament evolution are probably even higher (Endler 1980; Meyer 1997). Finally, the fast pace of evolution for sex-determining mechanisms and male ornaments might have constrained the extent of influences from sex-chromosome systems on male ornament evolution, and thereby made any association between these variables more difficult to detect (especially in the phylogenetically uncorrected analyses). However, the other side of that coin is that rapid evolution in male ornaments and sex-determination mode should generally have limited unwanted complications otherwise arising from phylogenetic inertia (Blomberg & Garland 2002).

In any event, for all of these biological and technical reasons, our current findings should be viewed as provisional. Further ethological and phylogenetic evaluations will be important, but critical tests of the possible associations between sex chromosomes, male ornaments and female preferences would profit especially from close genetic dissections of sexually selected traits in many more fish taxa. Only when the exact genetic underpinnings of these phenotypic traits are directly understood in many independent fish lineages will it become more evident as to whether the theoretical association between male ornaments and female heterogamety has been empirically realized. Especially if this association does not exist regularly in nature (as our current results suggest), then it will also be important to revisit and perhaps modify the theory itself to take into account additional biological considerations.

In conclusion, our phylogenetic analyses suggest that the particular mode of sex determination has had no consistent and discernible impact on the evolution of sexually selected traits in ray-finned fishes. According to recent theory, an association between male heterogamety and male ornaments should probably have been observed if good-genes processes of sexual selection predominated in fishes, whereas an association between female heterogamety and showy males might have been observed if Fisherian sexual selection was the predominant force. Although various technical and biological complications in our current assessments must be acknowledged (see above), the lack of a clear empirical association between sex-chromosome type and male ornamentation in ray-finned fishes suggests that sex-determination mode has been at best only one of many evolutionary and ecological factors affecting the outcome of sexual selection in this large vertebrate clade.

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