Instances of sex inversion in the domesticated swordtail, *Xiphophorus helleri* Heckel (Pisces, Osteichthyes)

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Summary. 3 protogynous hermaphrodites were found in a domesticated strain of *Xiphophorus helleri,* confirming much doubted observations by older authors. It therefore seems that sex determination may vary between strains, certain strains being strictly gonochoristic but others being capable of true hermaphroditism.

It is still an open question whether the swordtail *Xiphophorus helleri,* an ovoviviparous poeciliid fish with polygenic sex determination[1], is occasionally capable of functional sex inversion. Some earlier authors, up to 1940, have described numerous instances of sex inversion in adult females whose female gonads would have changed into typical bilobed testes[2-8]. Some of these authors[6,7] identified the traces of the sex inversion in the so-called 'residual bodies', present in the gonads, which were interpreted as remainders of atretic eggs.

The first doubt about the actual existence of sex inversion in *X. helleri* arose from the fact that the supposedly inverted gonads were bilobate and not oval like the ovaries from which they should have derived[9]. It was shown later that the 'residual bodies' which were found in the supposed sex inverted specimens are actually cysts of the fungus *Ichthyophonus hoferi*[10,11]. Authors who investigated the genetics of poeciliid fishes[12,13] stated, moreover, that they never found sex inversions in their laboratories, with the exception of a few old arrhenoid females that had acquired the male secondary sex characters but were not able to inseminate other females.

It was subsequently demonstrated[13] that 2 kinds of males can be identified in swordtails: some differentiate rapidly while still of small size ($♂♂_F$), and some differentiate slowly ($♂♂_S$) and reach the size of small females before they develop the male secondary sex characters. It was assumed therefore that the immature $♂♂_S$ might have been confused by the previous authors with females and erroneously interpreted as protogynous hermaphrodites after the caudal sword and the copulatory organ of the male were formed.

Although the above-mentioned studies have conclusively demonstrated that many instances of putative sex inversion in the swordtail are actually examples of arrhenoidy, the existence of rare cases of functional proterogyny cannot be ruled out, at least in certain strains of *X. helleri.*

The existence of both early (figure 1) and late differentiating male specimens has been confirmed among hundreds of swordtails which were raised in our laboratory. Some of the slowly differentiating males developed the gonopodium as well as the prolongation of the caudal fin as late as 1 year or more after their birth, thus confirming the existence of an extensive sex variability in the strain. Change from the female to the male livery was also observed in several individuals living together in tanks that contained exclusively females all of which had produced broods at least once. A number of such individuals, transformed aged females, were not able to fertilize virgin females: they must therefore be regarded as arrhenoid females as seen by earlier authors. By contrast, 3 individuals (figure 2) that had assumed male secondary sex characters in earlier stages were able to fertilize several virgin females that had been isolated at very early stages. They engendered progenies which showed markedly high female-male ratios (table). Such data agree with Kosswig's interpretation of polygenic sex determination in *X. helleri*[1]. Thus, we suppose that the individuals which change from the female to the male phase are actually protogynous hermaphrodites, and that their weak male expression is reflected in a significantly low proportion of males among the progeny.

Once it had been ascertained that the 3 individuals were able both to produce some broods and to fertilize virgin females in different phases, their gonads were sectioned and examined histologically. Normal seminiferous tubules and cysts with the various spermatogenetic stages were observed, but it must be pointed out that these gonads are oval in shape, with a central lumen functioning as a testicular duct (figure 4), and are not bilobate like those of male specimens (figure 3).

Thus, the conflicting conclusions reached by old and recent authors can partly be ascribed to an erroneous interpreta-

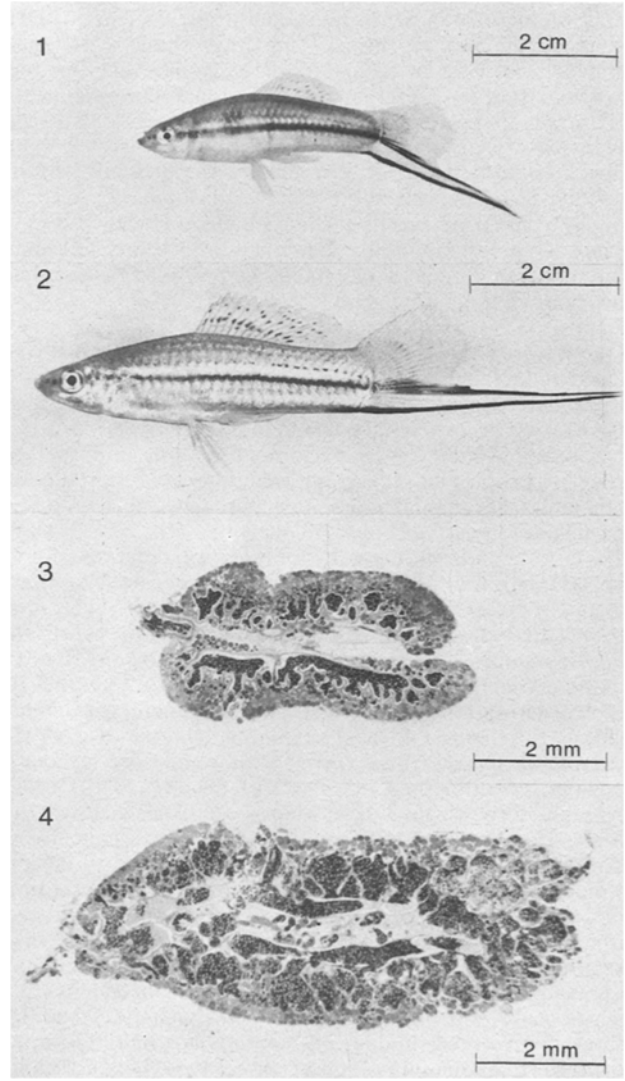

Fig. 1. *Xiphophorus helleri* male. Fig. 2. *Xiphophorus helleri* hermaphrodite in male phase. Fig. 3. Bilobed male gonad of *Xiphophorus helleri* with 2 longitudinal testicular ducts flowing into the vas deferens. Fig. 4. Sex inverted gonad of *Xiphophorus helleri* with a single testicular duct.

tion of histological data and to a lack of experimental control by some of the older authors. But they can also be ascribed to the existence of a certain degree of sex variability in certain strains: some strains may produce a percentage of true hermaphrodites, as shown in this paper, and some are totally gonochoristic[12,13].

A possible hybridization between *X. helleri* and *X. maculatus* in aquarium tanks has been accounted as responsible for a labile sex differentiation[15]. It has been ascertained however in the present experiments that the gonads of the protogynous hermaphrodites are oval and not different from the gonads of the pure female individuals, and that the protogynous hermaphrodites do occur in domesticated strains that any aquarium breeder would identify as true swordtails *X. helleri.*

Further research should ascertain whether proterogyny is present in natural populations of *X. helleri.*

Xiphophorus helleri: progenies of different virgin females paired with 3 exceptional protogynous hermaphrodites in male phase (♂ 1, 2, 3)

Parents	Number of broods	Offspring ♂♂	♀♀	♂♂ (%)
$♂_1 \times ♀_1$	4	33	78	29.7
$♂_1 \times ♀_2$	4	18	71	20.2
$♂_2 \times ♀_3$	4	17	61	21.8
$♂_2 \times ♀_4$	4	16	70	18.6
$♂_3 \times ♀_5$	4	22	92	19.3
$♂_3 \times ♀_6$	4	15	75	16.7

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Developmental G6PD polymorphism in *Drosophila melanogaster:* Evidence for non-structural variants

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Summary. G6PD isozyme variation in *Drosophila melanogaster* is investigated in the larval stage through electrophoretic and genetic analyses. As current structural models for this gene-enzyme system fail to apply in these laboratory populations, the authors suggest a regulatory hypothesis to explain their observations.

The electrophoretic phenotype of G6PD in *Drosophila melanogaster* was shown to have a complex relationship to its structural genes, because of regulatory factors affecting isozyme patterns[1]. An association between G6PD isozyme patterns and vestigial marker was also observed and could not be explained in terms of structural genes only[2]; these studies were performed on adult single fly homogenates.

As far as we know, the only available information about G6PD developmental expression is from Steele et al.[3], who measured total enzyme activity of larval stage, and Wright and Shaw[4], who monitored electrophoretically *D. melanogaster* embryos up to the 48th h from deposition.

In this communication we report some observations concerning G6PD isozyme pattern in *D. melanogaster* larvae. Our results provide evidence that G6PD electrophoretic variation is inherited in a peculiar non-Mendelian fashion that deserves attention.

We sampled 3rd instar larvae from Canton strain, from an unrelated strain carrying the vg marker and from 6 populations having a Canton × vg cross as their common origin, plateaued for a quantitative trait after about 70 generations of selection[5]. We raised these populations in mass culture at 25 °C for several generations, and then took random samples of about 100 individuals in the proper developmental phase. Cellogel electrophoresis was carried out on pooled homogenates of the above 100 flies samples, following the procedure detailed in Pieragostini et al.[2].

As represented in figure 1, samples from vg strain exhibited a fast band, selection lines a slow one; Canton samples gave single bands even slower than selection lines.

Canton and vg strain, being genetically unrelated, could reasonably be homozygous for the slow and fast allele respectively and exhibit single bands; larval stage variation would then be similar to adult stage variation[6]. However, if this were our case, we would expect multiband patterns in the progeny from Canton and vg strain (selection lines); our multiple sampling procedure would certainly reveal the presence of segregational homozygotes with the fast form of the enzyme. Heterozygotes too would give their contri-

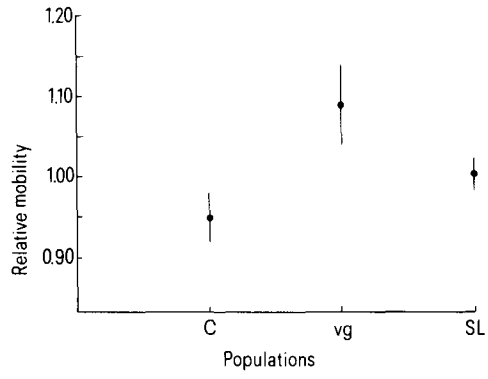

Fig. 1. G6PD phenotypes in larval populations of *Drosophila melanogaster.* From left to right, Canton strain (C, number of samples = 16), vg strain (vg, number of samples = 11), selection lines (SL, including $PMvg^+$, $PFvg^+$, $PKvg^+$, PMvg, PFvg, PKvg, number of samples = 10 each) average mobilities. They are expressed in standard units (mm from origin of samples/mm from origin of standard) and are represented with their 't_{95}' confidence intervals. 2-banded zymograms, in mixed control runs, confirmed non-homology of bands.