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著者	ANDO Daisei, NAKAJIMA Masamichi, FUJIO Yoshihisa
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Strain Difference of Vertebral Abnormality in the Guppy *Poecilia reticulata*

Daisei ANDO, Masamichi NAKAJIMA and Yoshihisa FUJIO

*Laboratory of Fish Genetics and Breeding Science, Faculty of
Agriculture, Tohoku University, Sendai, 981, Japan.*

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Summary

Vertebral abnormality has been often found in fish stocks. Vertebral abnormality was examined by alizarin staining of the skeleton in 14 different strains of the guppy (*Poecilia reticulata*). The vertebral abnormality was pertinent to the curve of vertebral column and classified into 4 types. Occurrence of each type was different among strains.

The A and F strains showed extremely high incidence (59.5% and 70.8%) but the other strains showed less than 10% and varied from 0.9% to 9.2% among strains. The high incidence indicates the possibility of high inbreeding level. The strain difference suggests the existence of polymorphism of a malformed gene(s).

The genetic differences among strains of the guppy (*Poecilia reticulata*) were demonstrated using electrophoretic markers, and reproductive traits (1-3). It has been also reported that responses to low and high temperature varied among strains, and survival times varied among strains in artificial sea water with a salinity of 35 ppt (4-6).

Vertebral abnormality has been often found in various kinds of fish stock and wild populations. Fused and wavy mutants of the medaka (*Oryzias latipes*) had been often found by Aida (7). The autosomal recessive *fu* (fused) gene manifests itself by fusion or ankylosis of vertebral here and there, while the autosomal recessive *wy* (wavy) gene results in hunchback or lordosis of vertebral column. The present study was focused on the strain differences in occurrence of vertebral abnormality of the guppy.

Materials and Methods

Animal materials

Fourteen strains, S, S3, SC, M1, O, A, F, T, T1, G, B, C, D, and D1 are being

maintained in 60 l aquaria as a closed colony. S, S3, and O strains are the standard-type and S3 is a isolate of the S strain. F, T, T1, G, B, C, D, and D1 strains are the fancy-type, and T1 and D1 are an isolate from the T and D strains, respectively. SC and M1 strains are made from the cross between the S females and a single cobra-type male and between the mosaic-type females and S males, respectively. A, B, and C strains are originated from the hybrids between albino and king-cobra, and A is selected for albino in their segregation. B and C strains are selected for red and green tail, respectively. In addition, size differences are more apparent in male body length although the same tendency is female body length. Two size groups are apparently presented, one group composed by F, T, T1, G, B, C, D, and D1, which are larger than the other group, S, S3, SC, M1, O and A, which are smaller. The fish are kept at a temperature of $23 \pm 2^\circ\text{C}$. The abnormal fish were taken from the stocks continuously throughout the year.

Alizarin staining

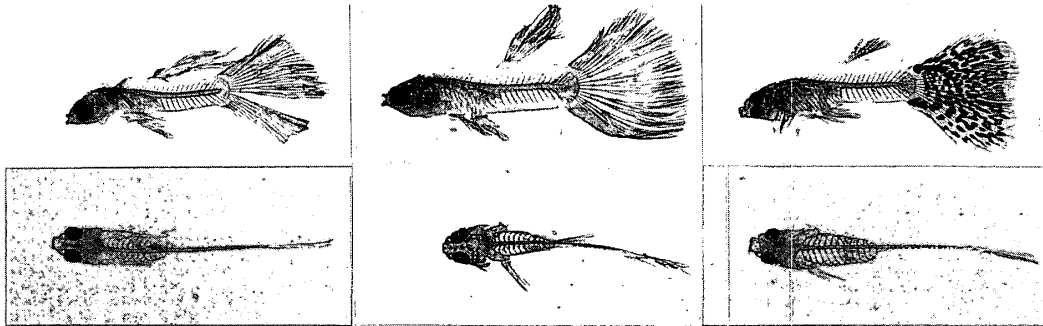
Abnormal fish were cut at the abdomen and fixed with 95% ethanol, and then the internal organ was removed. After fixation, the specimen was put into 1% KOH solution until the skelton could be observed. The specimen was stained with 0.01% alizarin S over night. Decoloration was done with an increased concentration of glycerin. The skeletal specimen was observed by binocular microscope.

Results

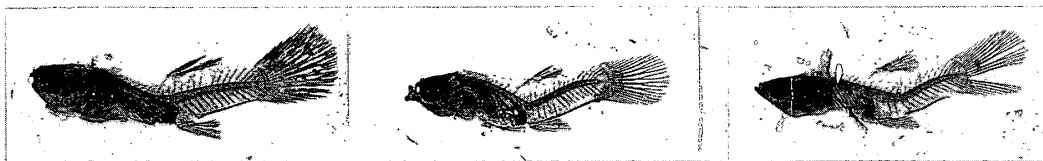
Vertebral abnormality has been often found in each of 14 strains of the guppy. Fig. 1 shows the vertebral abnormality. The vertebral abnormality was pertinent to the curve of the vertebral column and classified roughly into 4 types. In type I, the external body form showed a ventral doglegged outline according to ankylosis of the vertebral column. Type II showed the crooked outline on either side in tail according to the fusion of the vertebral column. In type III, the external body form showed wavy dorso-ventral outline according to the ankylosis and/or fusion of vertebral column, and type IV showed wavy outline on both sides according to the ankylosis and/or fusion of vertebral column. Type I was observed in 12 of the 14 strains but not in A and D strains. Type II was observed in 12 of the 14 strains but not in S3 and T1 strains. Type III was observed in 9 of the 14 strains but not in SC, S3, T1, S, and D strains. Type IV was observed in 8 of the 14 strains but not in B, T, S3, T1, D, and S strains. Occurrence of each type was different among strains.

The number of abnormal individuals varied from 3 to 47 among strains which were maintained in 60 l aquaria at densities of 65 to 576 individuals per aquarium (Fig. 2). There was no significant correlation between the number of abnormal

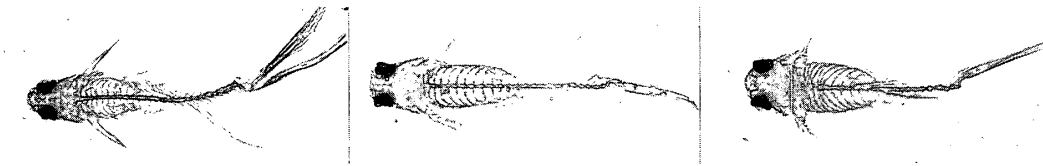
N o r m a l



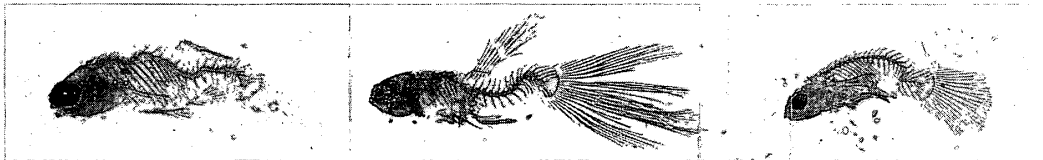
T y p e I



T y p e II



T y p e III



T y p e IV

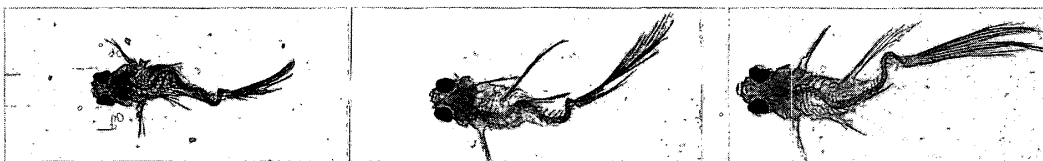


FIG. 1. Vertebral abnormality in the guppy.

individuals and the density of individuals (Fig. 3). Thus, the strain difference was considered as the genetic difference.

Rough estimates of incidence were obtained by the percentage of abnormal individuals against the number of individuals in rearing density (Fig. 2). The A and F strains showed the extremely high incidence. This high incidence indi-

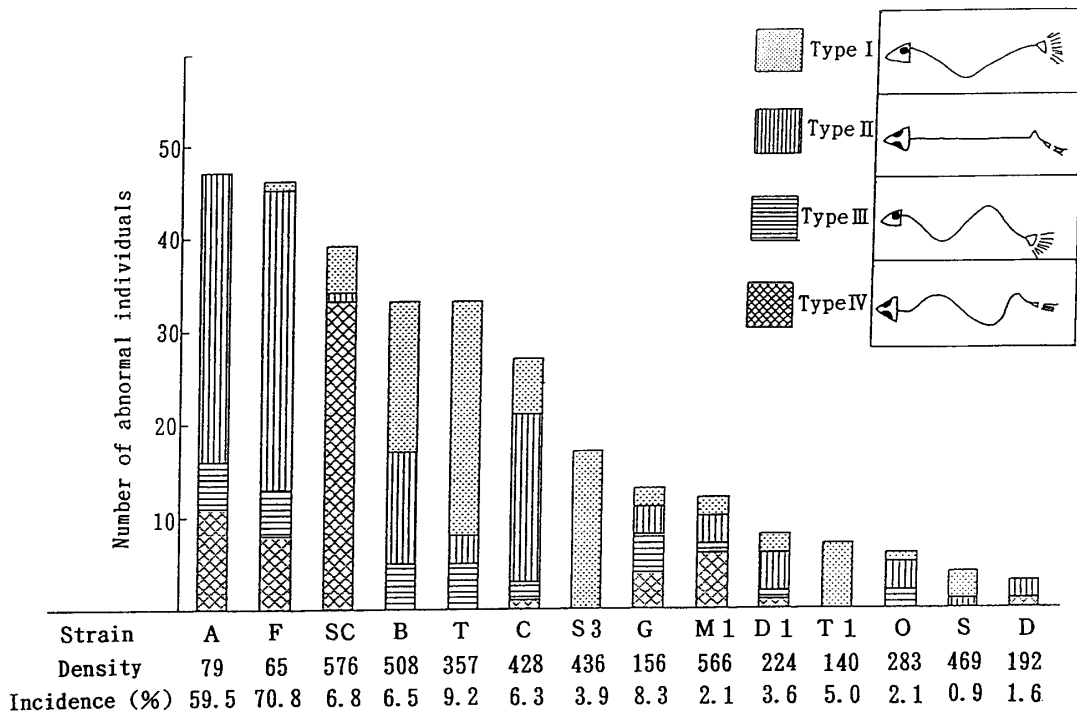


FIG. 2. Occurrence of vertebral abnormality in 14 strains of the guppy.

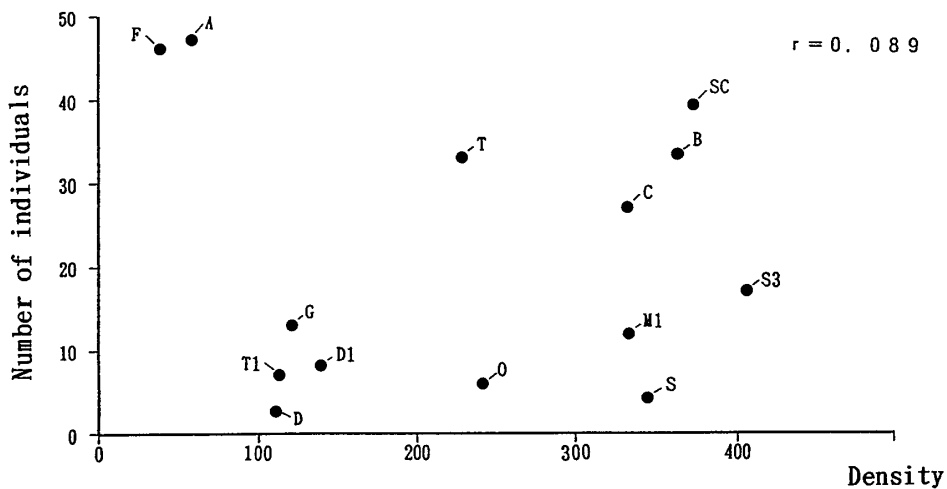


FIG. 3. Correlation between vertebral abnormality and rearing density.

cates the possibility of high inbreeding level, because A and F strains had the notably low density. On the other hand the remaining 12 strains showed the low incidence, less than 10% and the incidence varied from 0.9% to 9.2% among strains. The D1 strain, which was an isolate of the D strain, had significantly higher incidence than the D strain, and the S3 strain, which was an isolate of the S strain, had significantly higher incidence than the S strain. The T1 strain,

which was an isolate of the T strain, had significantly lower incidence than the T strain. The B and C strain, which originated from the same stock had the same level of the incidence. Regarding the fluctuation of incidence, it indicates the possibility that the gene(s) for vertebral abnormality are fluctuating due to genetic random drift.

Discussion

The genetic differences of the guppy strains are demonstrated by electrophoretic markers⁽¹⁾. The present study is a demonstration of the difference in occurrence of vertebral abnormality among the guppy strains which are genetically different. These vertebral abnormalities are thought to be inherited. The strain of the guppy for the study of the gene expression would be made by selective breeding. The existence of polymorphism of a malformed gene(s) was suggested by the fluctuation of incidence among strains and among related strains. Such a selection experiment is performed in the wavy (*wy*) mutant of the medaka (*Oryzias latipes*) and the wavy characters are selective to normal and wavy, meaning the presence of modifiers of the major gene (8). Phenogenetics of the vertebral fused (*fu*) of the medaka (*Oryzias latipes*) was carefully studied by Ogawa (9), who found that the expressivity varies significantly even among the same sib groups reared at a constant temperature and also increases with increase of temperature.

High incidence in the A and F strains indicates the possibility of high inbreeding level, because they are the lowest density of individuals. The most striking observed consequence of inbreeding in the guppy is the reduction of the mean phenotype value shown by characteristics connected with reproductive capacity, such as fertility ratio, offspring survival at birth, and offspring size (10). Since the general effect of inbreeding is to increase of homozygosity, the occurrence of the vertebral abnormality is thought to increase with the increase of homozygosity.

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