

## A study on the transparent-scaled mutant $ne^3$ of the crucian carp, *Carassius auratus langsdorfii*

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**Abstract** There are two reproductive forms in crucian carp, *Carassius auratus langsdorfii*. The bisexual form produces females and males in a ratio of 1:1, whereas the unisexual form is reproduced gynogenetically. A mutant with transparent scales ( $ne^3$ ) was found in the bisexual form, which belonged to the *ne* alleles ( $+>ne^3>ne$ ). In the unisexual form, four transparent-scaled females resembling the *ne* and  $ne^3$  mutants were found in the field, but genetic analysis to confirm their relationship to these genes was not possible because of their gynogenetic nature. Their progenies were transparent-scaled females. In some cases, two variants with a similar scale transparency but differing in the distribution pattern of the transparent scales among the whole scale population were segregated in the progenies.

### Introduction

Natural populations of crucian carp consist of bisexual and unisexual forms (Cherfas, 1972; Yamamoto and Kajishima, 1984). Unisexual females are inseminated by males, but the male characteristics are not inherited. In the bisexual form, the nacreous-like transparent-scaled mutant (*ne*) has been analyzed (Yamamoto, 1977). Only a small amount of guanine is deposited in iridocytes of scales and of the eye reflective layer in this mutant. The *ne* gene exerts no effects on melano-phores and xanthophores.

I purchased in a pet shop a bisexual variant of the crucian carp resembling *ne* but definitely different from it, and genetic analyses proved that this mutant  $ne^3$  belonged to the *ne* alleles. Transparent-scaled fish of the unisexual form resembling *ne* and  $ne^3$  were also purchased or collected from the field, but genetic analysis to define their relationship to *ne* and  $ne^3$  was not possible because of their gynogenetic nature. This paper describes the results of genetic analyses and phenotypic expression of the  $ne^3$  gene and other *ne*- and  $ne^3$ -like genes. These mutant genes have previously been listed by Tomita (1992).

### Materials

In 1979, a transparent-scaled female was collected from the western suburbs of Nagoya. This fish had a considerable number of completely transparent scales amongst normal scales. This transparent-scaled characteristic was different from the *ne* phenotype. Almost all scales of the *ne* mutant have an attenuated amount of guanine, exhibiting nacreous-like transparent appearance, and normal, metallic scales are almost absent.

In 1985, a transparent-scaled male which was very similar in appearance to the 1979 female was purchased from a pet shop in Nagoya. The proprietor of this shop told that this fish had been caught in Lake Biwa.

In 1988, a transparent-scaled female was again collected from the western suburbs of Nagoya.

In 1989, a nacreous-like transparent-scaled female (*ne*-like) was found in the western suburbs of Nagoya.

In 1992, a transparent-scaled female was collected.

In 1992, a transparent-scaled male was purchased from a pet shop in Nagoya. This fish had been caught in Shonaigawa (Shonai river) in Nagoya.

Altogether, one *ne*-like female fish and three female and two male fish resembling *ne* but differing in having some completely transparent scales amongst normal scales were thus obtained, and they were subjected to genetic analysis.

### Results

To assess the reproductive form, a transparent-scaled female in question was mated with a dominant transparent-scaled goldfish male (*T/t*). If the female is bisexual, the progeny should be segregated into fish with normal scales and those with dominant transparent scales in a ratio of 1:1 and the sex ratio should be 1:1. If the female is unisexual, the progeny should be all transparent-scaled females.

A wild-type female was mated with the transparent-scaled male ( $ne^3$ ) purchased in 1985.

The  $F_1$  progenies were 12 wild type (female 5, male 7). Four pairs of the  $F_1$  progenies were used for the test crosses. The  $F_2$  progenies from the first pair were 18 wild type and 4  $ne^3$ . The second  $F_2$  progenies were 22 wild type and 2  $ne$ . The third  $F_2$  progenies were 10 wild type and 3  $ne^3$ . The fourth  $F_2$  progenies were 11 wild type and 3  $ne^3$ . The results indicated that the parent  $ne^3$  male was the heterozygote of  $ne$  and a novel mutant gene allelic to  $ne$  that I designated  $ne^3$ .

Two crosses were made between the  $ne^3$  females and  $ne^3$  males that were obtained in the above experiment. In one cross, the progenies were 23  $ne^3$ . In the other cross, the progenies were 14  $ne^3$  and 4  $ne$ . In this case, the parent fish were therefore the heterozygotes of  $ne^3$  and  $ne$ . The  $ne^3$  and  $ne$  genes are the alleles, and the  $ne^3$  gene is dominant over the  $ne$  gene ( $+ne^3 > ne$ ).

An  $ne$  female derived from the  $ne$  mutant that was established by Yamamoto (1977) was mated with an  $ne^3$  male produced in the first experiment. The progenies were 9  $ne^3$  and 8  $ne$  in a ratio of 1:1. The parent  $ne^3$  male was therefore heterozygous ( $ne/ne^3$ ).

The  $ne^3$ -like female collected in 1979 was mated with a  $T/t$  goldfish male. The progenies were 16 transparent-scaled females. The  $ne^3$ -like female was therefore gynogenetic.

The  $ne^3$ -like female collected in 1988 was mated with a  $T/t$  goldfish male. The progenies were 14  $ne^3$ -like females and 2 ( $ne^3$ -like)' females. The ( $ne^3$ -like)' females were distinct from the  $ne^3$ -like females in that the majority of scales were transparent and only a few normal, metallic scales were present, contrasting with the  $ne^3$ -like females which had many transparent scales but the majority of scales were normal, amongst which the transparent scales were locally distributed. One of the two  $ne^3$ -like females obtained above was mated with a  $T/t$  goldfish male. The progenies were all  $ne^3$ -like females (36 fish).

The parent  $ne^3$ -like female used in the preceding experiment was again mated with another  $T/t$  goldfish male. The progenies were 65  $ne^3$ -like females and one ( $ne^3$ -like)' female. The  $ne^3$ -like females produced mostly the  $ne^3$ -like females and a few ( $ne^3$ -like)' females. Thus, the 1988  $ne^3$ -like female reproduced gynogenetically and the progenies were segregated into two slightly different types.

The nacreous-like transparent-scaled female ( $ne$ -like) collected in 1989 was mated with a  $T/t$  goldfish male. The progenies were 42  $ne$ -like females, indicating that the  $ne$ -like mother was a gynogenetic form. These  $ne$ -like females had often a few metallic scales amongst the nacreous-like transparent scales.

The  $ne^3$ -like female collected from the field in 1992 was mated with a  $T/t$  goldfish male. The progenies were 60  $ne^3$ -like fish (The sexes were not discerned because the fish were still juvenile when this paper is being prepared). The  $ne^3$ -like mother was therefore most probably gynogenetic.

### Discussion

Two types of transparent-scaled mutants have previously been described, i.e., the incomplete dominant  $T$  in the goldfish (Chen, 1928) and the recessive nacreous-like transparent-scaled in the goldfish ( $n$ : Matsui, 1993;  $g$ : Kajishima, 1977) and in the crucian carp ( $ne$ : Yamamoto, 1977). The  $T$  gene affects the expression of melanophores, xanthophores and iridocytes. The  $T$  homozygote ( $T/T$ ) often lacks iridocytes, melanophores and xanthophores, and shows transparent, colorless body. The  $T$  gene exerts a pleiotropic action on the pigment cells.

The  $n$  mutant of Matsui (1933) was lost. The mutant  $g$  established by Kajishima (1977) was similar to  $n$  of Matsui. The  $g$  and  $ne$  genes have been inferred to be the same gene (Kajishima, personal communication).

In the present study, I have described a novel mutant gene  $ne^3$  of the crucian carp, which is allelic to  $ne$ . Four more mutants which are apparently  $ne$ -like or  $ne^3$ -like were found, but whether they are allelic to  $ne$  or  $ne^3$  was not confirmed because of their gynogenetic nature.

### References

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