

## Differences Metric Traits among Three Strains of Apple Snail, *Pomacea canaliculata*

著者	FUJIO Yoshihisa, KURIHARA Hajime, BRAND Elisabeth, von
journal or publication title	Tohoku journal of agricultural research
volume	41
number	3/4
page range	61-68
year	1991-03-30
URL	<a href="http://hdl.handle.net/10097/29916">http://hdl.handle.net/10097/29916</a>

## Differences Metric Traits among Three Strains of Apple Snail, *Pomacea canaliculata*

Yoshihisa FUJIO, Hajime KURIHARA  
and Elisabeth VON BRAND

*Department of Fishery Science, Faculty of Agriculture,  
Tohoku University, Sendai, Japan*

(Received, August 28, 1990)

### Summary

The apple snail strains A, B, and C were examined for six metric traits: egg number per lump, hatchability, body weight at hatching, at 15 days, and at 30 days, and the survival rate at 30 days. As an overall result, significant differences were observed in 1 out of 6 traits between A and B, in all traits between A and C, in 5 out of 6 traits between B and C, indicating that A and B are similar, but C shows divergence from both in metric traits.

A positive correlation was observed between egg number per lump and hatchability, between hatchability and body weight at hatching, and between body weight at 15 days and 30 days in all strains. On the other hand, the correlation between egg number per lump and survival rate, between body weight at hatching and day 15- and day 30-body weight, between hatching body weight and survival rate, between day 15- and day 30-body weight with the survival rate not always found in each of the different strains. This might suggest that the strain differences could be related to the inbreeding level.

### Introduction

Laboratory strains have been developed for used as biological material for genetics and breeding experiments. During the process of maintaining the strains pure, some will keep similar characteristics as the wild population and others have diverged substantially due to selection and inbreeding. Metric trait differences among laboratory strains are also useful for evaluating the existence of genetic factors influencing the traits. In general, the metric traits are measured in term of an end-product such as fertility, hatchability, survival rate or growth rate. The resulting histogram has a continuous distribution and approximates more or less closely to a normal curves.

Several investigators have tried to evaluate the differences in several strains or races in fish (1-11). Most of the reported evaluations were on growth rates and

few investigations have assessed reproductive traits. In oyster (*Crassostrea gigas*), the strain differences have been demonstrated in dimensional characters such as shell size, form and weight and in flatness of shell valve, coloration, adaptability to environmental conditions and also in spawning conditions, in inbreeding of native oysters of Hokkaido, Miyagi, Hiroshima, and Kumamoto (12)

The apple snail, *Pomacea canaliculata*, is useful as experimental organism for shellfish studies in fishery genetics and breeding because of their short life-cycle. The previous paper revealed that three strains of this organism have a chromosome number of  $2n=28$  and male heterogamy (XY) (13). Furthermore, two linked loci with a null allele for leucine aminopeptidase isozyme in apple snail were demonstrated and it suggested a pure mating system and 1 female mates with only 1 male (14).

This paper is an account of metric trait differences in 3 apple snail strains and reports general parameter that can be used as a measurement of the genetic effects in any trait.

### Materials and Methods

Three apple snail strains, A, B, and C, have been maintained in closed colonies in  $180 \times 80 \times 80$  cm aquaria at a density of 100–200 individuals per aquarium. The apple snails are maintained at a temperature of  $23 \pm 2^\circ\text{C}$  and fed with cabbage and carp pellets.

For quantitative analysis, the egg lumps, laid on the aquarium wall, were obtained at random from the different stocks and transferred each into one petridish. The incubation temperature of the eggs until hatching was  $30^\circ\text{C}$ , in an incubator with controlled humidity. After 12 days, the snails hatched. The number of hatched snails were counted, and the unhatched also, to determine the hatchability (%). The recently hatched snails were reared for 30 days at a temperature of  $30^\circ\text{C}$  using plastic vessels of 150 mm diameter with 500 ml water. The culture density was usually 20 snails per vessel coming from one egg lump. The weight before and after the growing period was recorded.

### Results

#### *History and development of the strains*

The history and development of three strains are summarized as follows. The A strain has a light shell color without any color band, and is considered to be the color mutant snail. This strain was originated from three egg lumps obtained from Aomori Prefectural Fresh Water Fish Station in 1984. The B strain is dark colored with a light color band and is considered to be the wild type. It originated from 10 adult snails introduced from The Tokyo University of

Fisheries in 1983. The last, strain C, is slightly different from the B strain because of the lack of the light colored band. This strain originated from 20 adult snails from a population found proliferating in a pond in Okinawa in 1984.

The largest adult sizes were 38.1 g in A strain, 48.8 g in B strain, and 70.3 g in C strain. The maximum size of the adult in strain C was significantly the largest compared to A and B strains. The shell shape was expressed as the ratio between (shell diameter)/(shell length). The ratios were 0.868 in A strain, 0.884 in B strain, and 0.909 in C strain. The differences between all strains are significant.

#### *Strain differences in metric traits*

The average, standard deviation, and coefficient of variation (CV) for 6 metric traits measured in the 3 strains, A, B, and are presented in Table 1. Significant strain differences for egg number per lump were observed between A and C, B and C, but not between A and B. The hatchability was significantly different between all strains; C displayed the highest and A the lowest value. The CV instead of A was the largest and C the smallest. The distribution of the hatchability obtained is shown in Fig. 1, and the widest distribution obtained was for strain A. Strain B is similar to A. In case of strain C, the values have a narrow distribution, due to an increased frequency of the low hatchability egg lump. The mean body weight at hatching was significantly different between A and C, B and C, but not between A and B, being related probably to the larger egg size of A and B compared to C.

TABLE 1. *Summary of the Averages obtained for Three Strains of Apple Snail*

	A strain	B strain	C strain
Egg number per lump	207.3±140.8 (76)	215.9±106.4 (58)	583.5±273.0 (37)
CV	67.9	49.3	46.8
Hatchability (%)	41.0± 21.7 (76)	53.1± 17.1 (58)	77.4± 13.3 (37)
CV	52.9	32.2	17.2
Body weight at hatching (mg)	3.8± 1.3 (68)	4.3± 1.5 (38)	2.7± 0.6 (31)
CV	34.2	34.9	22.2
Body weight at 15 days (mg)	53.4± 9.9 (19)	63.0± 11.5 (19)	32.3± 10.2 (23)
CV	18.5	18.3	22.2
Body weight at 30 days (mg)	257.0± 48.1 (19)	267.8± 38.0 (19)	167.1± 44.9 (23)
CV	18.7	14.2	26.9
Survival rate at 30 days (%)	87.2± 9.0 (26)	90.3± 6.6 (24)	92.2± 5.5 (33)
CV	10.3	7.3	6.0

Number of egg lumps are enclosed in ( ).

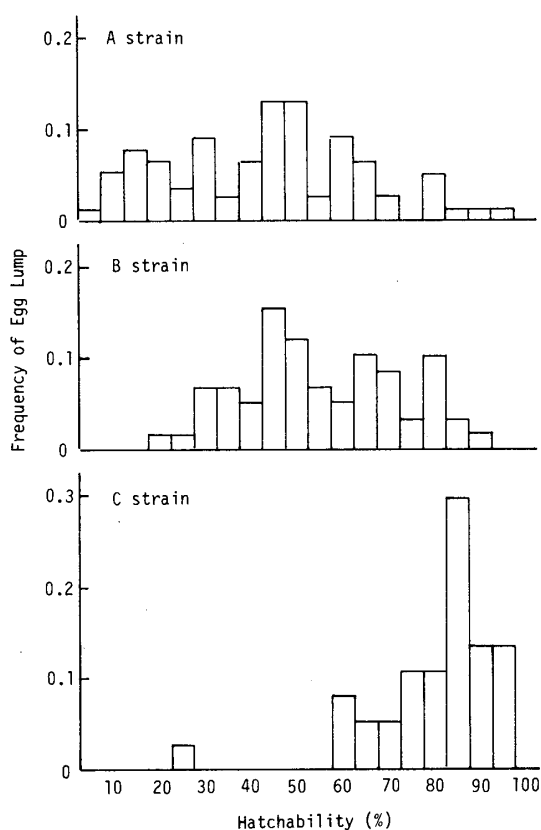


FIG. 1. Distribution of the hatchability in each egg lump of three strains in apple snail.

When the culture density was 20 snails per vessel, coming from one egg lump, the mean body weight at 15 days significantly differed for all strains, while the mean body weight at 30 days was significantly different between A and C, B and C, but not for A and B. Further, as shown in Fig 2, the mean body weight at 15 days decreased in constant proportion with the increase of the culture density  $r = -0.751$ ,  $n = 33$ , in A;  $r = -0.809$ ,  $n = 27$ , in B;  $r = 0.905$ ,  $n = 52$ , in C). Similar results were obtained in mean body weight at 30 days ( $r = -0.915$ ,  $n = 33$ , in A;  $r = -0.948$ ,  $n = 27$ , in B;  $r = -0.919$ ,  $n = 52$ , in C). A significant difference was observed only between A and C for survival rate at 30 days. As an overall result, significant differences were observed in 1 out of 6 traits between A and B, in all traits between A and C, and in 5 out of 6 traits between B and C, indicating divergence of C from A and B strains in metric traits.

The CV for metric traits varied among strains and among traits. The rank order is  $A > B > C$  in egg number per lump, hatchability and survival rate, while the rank is  $A$  and  $B > C$  in body weight at hatching and  $C > A$  and  $B$  in body weight at 15 and 30 days.

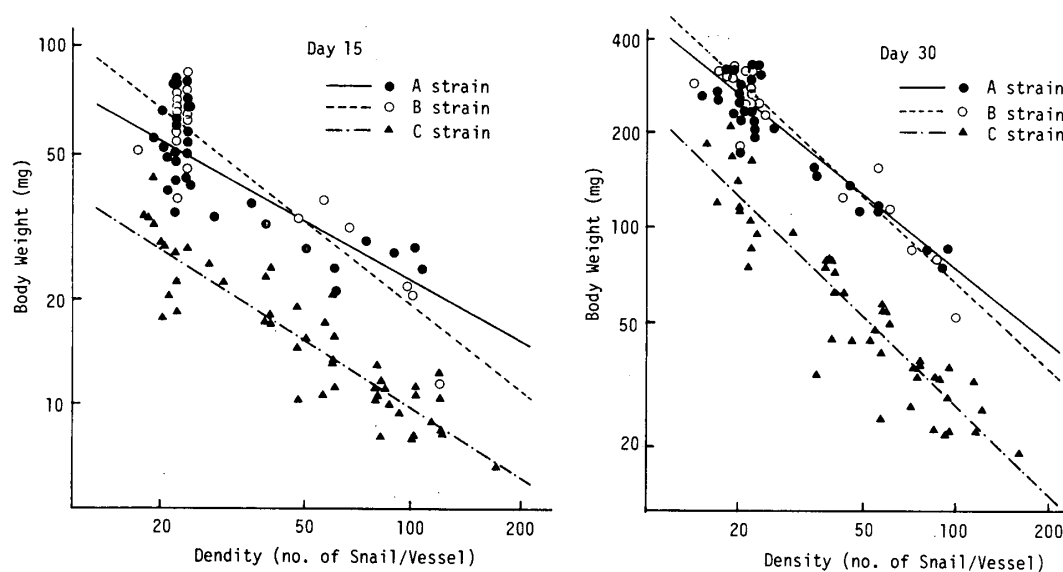


FIG. 2. Mean body weight at 15 and 30 days in relation to the culture density in three strains of apple snail.

Data points represent the mean body weight in each egg lump.

Regression lines describe how the mean body weight (Y) varied with the culture density (X);  $Y=209X^{-0.569}$  in A strain,  $Y=678X^{-0.779}$  in B strain, and  $Y=183X^{-0.638}$  in C strain at 15 days;  $Y=2614X^{-0.778}$  in A strain,  $Y=3751X^{-0.878}$  in B strain, and  $Y=1988X^{-0.934}$  in C strain at 30 days.

#### Correlation among 6 metric traits

Table 2 presents the correlation coefficient in 15 comparisons among 6 metric traits in three strains of the apple snail. Three out of 15 comparisons showed a positive correlation, but five showed no correlation in all strains. The observed positive correlations in all strains were between egg number per lump and hatchability, between hatchability and body weight at hatching, and also between day 15-body weight and day 30-body weight. On the other side, in strain A we found a positive correlation between hatching body weight and day 15, day 30-body weight and survival rate after 30 days. The other positive correlation observed in this strain was between day 15-body weight and the survival rate after 30 days. In strain B, the only additional positive correlation was between day 30-body weight and survival rate after 30 days. In strain C, the only additional positive correlation found was between the egg number per lump and the survival rate after 30 days. These observed differences between strains, but also the amount of positive or non existing correlations between the different metric traits within one particular strain, might suggest that each strain presents a different inbreeding level.

TABLE 2. *Correlation Coefficient among 6 Metric Traits in Three Strains of Apple Snail*

A strain						
	(1)	(2)	(3)	(4)	(5)	(6)
(1) Egg number per lump						
(2) Hatchability	0.385*					
	(n=76)					
(3) Body weight at hatching	-0.212	0.480*				
	(n=68)	(n=68)				
(4) Body weight at 15 days	-0.313	0.235	0.657*			
	(n=28)	(n=28)	(n=28)			
(5) Body weight at 30 days	-0.182	0.180	0.514*	0.906*		
	(n=28)	(n=28)	(n=24)	(n=29)		
(6) Survival rate at 30 days	0.028	0.323	0.511*	0.406*	0.255	
	(n=28)	(n=28)	(n=24)	(n=28)	(n=28)	
B strain						
	(1)	(2)	(3)	(4)	(5)	(6)
(1) Egg number per lump						
(2) Hatchability	0.460*					
	(n=58)					
(3) Body weight at hatching	0.005	0.625*				
	(n=38)	(n=38)				
(4) Body weight at 15 days	0.167	-0.125	0.260			
	(n=25)	(n=25)	(n=7)			
(5) Body weight at 30 days	0.139	-0.094	0.532	0.895*		
	(n=24)	(n=24)	(n=7)	(n=25)		
(6) Survival rate at 30 days	0.200	-0.175	-0.456	0.397	0.450*	
	(n=23)	(n=23)	(n=7)	(n=24)	(n=24)	
C strain						
	(1)	(2)	(3)	(4)	(5)	(6)
(1) Egg number per lump						
(2) Hatchability	0.438*					
	(n=37)					
(3) Body weight at hatching	0.261	0.378*				
	(n=37)	(n=31)				
(4) Body weight at 15 days	-0.211	-0.178	0.181			
	(n=50)	(n=50)	(n=30)			
(5) Body weight at 30 days	-0.237	-0.169	0.162	0.931*		
	(n=50)	(n=50)	(n=30)	(n=64)		
(6) Survival rate at 30 days	0.473*	0.204	0.069	0.037	0.062	
	(n=50)	(n=50)	(n=30)	(n=63)	(n=63)	

\*  $P < 0.05$ .

### Discussion

The obvious and significant differences between each strain of the apple snail, analyzing 6 metric traits, reaffirm genetic differentiation between strains. These strains are also different in their salinity tolerance leading to the conclusion that the strains have different genotypes (15). We could even think that differentiation can occur by parentage or by residual segregation if the strains have been separated before the fixation was completed. The genetic properties of a population can be expressed in terms of the gene frequencies and genotype frequencies based on the qualitative traits such as isozymes. On the other hand, the quantitative differences exhibited in metric traits are expressible in terms of the metric units (mean and variance) by which the trait is measured on an individual. When the strain difference is done by comparison of the mean is each of the strains, the environmental factor is necessary to be uniform. For example, the mean is the day 15- and day 30-body weight decreased in constant proportion with the increase of culture density.

The CV for metric traits varied among strains, for example, the widespread distribution for egg number per lump, hatchability, and survival rate after 30 days shown by strain A and B, and low in strain C. The increase of CV in hatchability was related with the increased egg lump frequency with low hatchability. The strain differences observed suggest the existence of a differential inbreeding depression as a result of the homozygosity of different deleterious recessive alleles. Associated with this, the original effective population size was different among strains, being the rank order  $C > B > A$ . Also, it is necessary to point out the different provenance of the original population. On the other side, the increase of frequency with low hatchability in apple snail might depend on this genetic load. However, any difference of phenotypic variance highly inbred lines and  $F_1$  (hybrids), must be attributed to differences of the environmental component, because the amount of genetic variance is negligible in the hybrids as well as in the inbred lines. The inbred lines can show a greater susceptibility to environmental variations than the hybrids, but it is not considered an universal phenomenon (16). In the relation of the CV with the genetic load in a population, the deeper analysis remains for the future.

### References

- 1) Wohlfarth, G., Moav, R., and Hulata, G., *Heredity*, **34**, 341 (1975)
- 2) Refstie, T., Steine, T.A., and Gjedrem, T., *Aquaculture*, **10**, 231 (1977)
- 3) Refstie, T., and Steine, T.A., *Aquaculture*, **14**, 221 (1978)
- 4) Gunnes, K., and Gjedrem, T., *Aquaculture*, **15**, 19 (1978)
- 5) Webster, D.A., and Flick, W., *Can. J. Fish. Aquat. Sci.*, **38**, 1701 (1981)
- 6) Smitherman, R.O., Dunham, R.A., and Tave, D., *Aquaculture*, **33**, 197



- (1983)
- 7) Ayles, G.B., and Baker, R.F., *Aquaculture*, **33**, 269 (1983)
  - 8) Ferguson, M.M., Danamann, R.G., and Allendor, F.W., *Can J. Genet. Cytol.*, **27**, 289 (1985)
  - 9) Koljonen, M.L., *Aquaculture*, **57**, 253 (1986)
  - 10) Macaranas, J.M., and Fujio, Y., *Tohoku J. Agri. Res.* **39**, 19 (1988)
  - 11) Macaranas, J.M., and Fujio, Y., *Aquaculture*, **85**, 69 (1990)
  - 12) Imai, T., and Sakai, S., *Tohoku J. Agri. Res.*, **12**, 125 (1961)
  - 13) von Brand, E., Yokosawa, T., and Fujio, Y., *Tohoku J. Agri. Res.*, **40**, 81 (1990)
  - 14) Fujio, Y., and von Brand, E., *Nippon Suisan Gakkaishi*, **56**, 1039 (1990)
  - 15) Kijima, A., Umeda, H., and Fujio, Y., *Fish Genetics and Breeding Sci.*, **13**, 35 (1988) (in Japanese)
  - 16) Falconer, D.S., "Introduction to Quantitative Genetics" 2nd. E. Longman, London, U. K. p. 340 (1981)