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NOTES AND COMMENTS

INHERITANCE OF ALLOZYMES OF A MARINE SNAIL (*UROSALPINX CINEREA*)

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SUMMARY

Crosses using virgin oyster drills (*Urosalpinx cinerea*) show that both the ODH and LAP loci of foot muscle tissue have two co-dominant alleles segregating in Mendelian fashion. This constitutes the first demonstration of allozyme inheritance in a marine gastropod.

1. INTRODUCTION

A DISADVANTAGE of many marine species for genetic experiments is that crosses needed to show the Mendelian segregation of presumed alleles are difficult if not impossible to conduct. Obscure and non-genetic, protein variation has been reported for some of these species (Murphy, 1976; Marcus, 1977). Thus, crossing experiments should be done whenever possible.

From 1972 to 1975, genetic experiments using the rocky intertidal gastropod *Urosalpinx cinerea* were implemented to study the inheritance of alleles at two variable enzyme loci. This is the first report of allozyme inheritance in a marine prosobranch.

2. MATERIALS AND METHODS

The oyster drill (*Urosalpinx cinerea*) is easily cultured in the laboratory as there is no pelagic larval stage. Juveniles hatch from attached egg capsules as crawling young.

Details of rearing methods have been published (Cole, 1975). Briefly, juveniles ($\leqq 8$ mm shell length) were isolated from local populations (Nobska Point, Woods Hole, and Wild Harbor, West Falmouth, Massachusetts) and reared to maturity in solitary confinement. Twenty pairs of unknown allozyme genotype were placed in separate cages. Egg capsules were removed from cage walls and hatched. F_1's of each cross were reared together. Eventually parents and offspring were sacrificed for electrophoresis. All cages were immersed in the heated seawater system of Carriker and Van Zandt (1973). Mussels (*Mytilus edulis*) were supplied as food.

A preliminary electrophoretic survey indicated variation at two enzymatic loci, octanol dehydrogenase (ODH) and leucine aminopeptidase (LAP), in *U. cinerea* foot muscle. Only this tissue was used, to avoid possible visceral parasites. Vertical starch-gel electrophoresis was performed using the

discontinuous sodium citrate-histidine (*p*H 7·0) buffer system of Brewer (1970). The best resolution was achieved with a 9 : 1 mixture of Sigma and Electrostarch hydrolysed potato starch in 13 per cent gels. Staining recipes were adapted from Ayala *et al.* (1972).

3. Results

Fourteen pairs produced F_1 snails (table 1). The mortality among the broods varied from 10·9 to 37·0 per cent. Most occurred among recently hatched snails due to anoxia caused by fouling of cage screens. Transferring the young to larger-meshed cages as soon as possible alleviated this problem.

The two loci exhibited different electrophoretic banding patterns. The ODH homozygotes were each single-banded. Alleles producing the more anodal band were termed " fast " and the alternative " slow ". Heterozygotes were three-banded, with both fast and slow bands, plus a middle, " hybrid " band. The LAP patterns showed two bands for each homozygote and three bands for heterozygotes. The slower band of the fast homozygote and the faster band of the slow homozygote overlapped in migration. Increasing the duration of the runs separated these bands in heterozygotes but also made bands more diffuse. The " fast-slow " convention was again used for this locus.

Two co-dominant alleles were observed segregating at the allozymic loci examined (table 1). The observed numbers of genotypes were not significantly different from those expected, assuming Hardy-Weinberg equilibria, except in one case. A significant excess of ODH^F homozygotes was observed for Cross A-4 ($\chi^2 = 5{\cdot}029$; $P < 0{\cdot}05$). No unexpected alleles were observed among progeny from parents homozygous for the same allele. All other ODH and LAP crosses conformed to Hardy-Weinberg expectations (table 1).

4. Discussion

Both loci exhibit simple co-dominant Mendelian inheritance with two alleles segregating at each locus.

As a triple-banded electrophoretic pattern was observed for ODH heterozygotes, this enzyme in *Urosalpinx cinerea* foot muscle is apparently a dimer with randomly associating subunits (Brewer, 1970). The interpretation of LAP banding patterns is more difficult. Murphy (1976) has reported similar LAP patterns from Californian limpets (genus *Acmaea*). He proposed that acmaeid LAP is composed of " . . . two polymeric enzymes which share a common polymorphic subunit as well as having different monomeric subunits. Thus homozygotes show one band pair and heterozygotes two ". Results obtained with *Urosalpinx cinerea* support this interpretation, though migration distances of the closest bands of homozygotes overlap giving three bands in heterozygotes under the electropherotic conditions used.

In *U. cinerea*, two polymorphic allozymic loci and a polymorphic shell-colour locus manifested by hatchlings have now been shown to be genetic (Cole, 1975; this report). These markers may aid in elucidating adaptational and evolutionary processes of natural populations (T. Cole, in prep.).

TABLE 1

Segregation at ODH and LAP loci in Urosalpinx cinerea *foot muscle. Male genotypes are given first in the parental allozyme phenotype column. Two co-dominant alleles were observed for each locus. Numbers in parentheses are those expected under Hardy-Weinberg equilibrium conditions. Only Cross A-4 for ODH exhibited significant deviations ($\chi^2 = 5{\cdot}029$; $P < 0{\cdot}05$). Percentage mortality for each cross was calculated as the number surviving to be electrophoresed divided by the number hatched.*

Cross	Parental allozyme phenotypes	% Mortality	Progeny phenotypes: ODH SS	ODH FS	ODH FF	LAP SS	LAP FS	LAP FF
A-3	ODH SS × SS	11·8	112 (112·0)	—	—	—	—	—
	LAP FS × SS		—	—	—	53 (56·0)	59 (56·0)	—
A-4	ODH FS × FS	37·0	14 (17·0)	29 (34·0)	25 (17·0)	—	—	—
	LAP SS × FS		—	—	—	30 (33·5)	38 (33·5)	—
A-6	ODH SS × SS	15·2	39 (39·0)	—	—	—	—	—
	LAP SS × SS		—	—	—	39 (39·0)	—	—
A-8	ODH SS × SS	13·6	70 (70·0)	—	—	—	—	—
	LAP SS × FF		—	—	—	—	70 (70·0)	—
A-9	ODH SS × FS	27·7	51 (47·0)	43 (47·0)	—	—	—	—
	LAP FS × FF		—	—	—	—	52 (47·0)	42 (47·0)
A-10	ODH SS × SS	14·9	80 (80·0)	—	—	—	—	—
	LAP SS × SS		—	—	—	80 (80·0)	—	—
A-11	ODH SS × SS	16·1	78 (78·0)	—	—	—	—	—
	LAP FS × SS		—	—	—	47 (39·0)	31 (39·0)	—
A-12	ODH SS × SS	10·9	106 (106·0)	—	—	—	—	—
	LAP FF × FF		—	—	—	—	—	106 (106·0)
A-13	ODH SS × SS	30·9	154 (154·0)	—	—	—	—	—
	LAP FS × FS		—	—	—	37 (38·5)	84 (77·0)	33 (38·5)
A-14	ODH SS × SS	28·1	123 (123·0)	—	—	—	—	—
	LAP SS × SS		—	—	—	123 (123·0)	—	—
A-15	ODH FS × SS	25·5	12 (17·5)	23 (17·5)	—	—	—	—
	LAP SS × SS		—	—	—	35 (35·0)	—	—
A-16	ODH SS × SS	16·3	87 (87·0)	—	—	—	—	—
	LAP FF × SS		—	—	—	—	87 (87·0)	—
A-17	ODH SS × SS	11.1	88 (88·0)	—	—	—	—	—
	LAP FF × FS		—	—	—	—	39 (44·0)	49 (44·0)
A-18	ODH SS × SS	16·8	79 (79·0)	—	—	—	—	—
	LAP SS × FF		—	—	—	—	79 (79·0)	—

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