days. Within 10-20 days after the injection, a state of maximal bloating has been reached, whereby not only is the abdomen distended to capacity, intersegmental membranes being stretched to the limit, but also the labium, which cannot be retracted anymore, and even the neck are bloated (figure 1). The bloated flies never copulate, nor do they lay eggs. However, this pathological state does not affect viability: the mean longevity of ascitic flies is the same as that of controls. Although the tumor grows incessantly, it is clearly not the volume of tumorous tissue that causes the bloating, but rather a genuine ascitic state, generated either by an overproduction of hemolymph or by water retention. In ascitic flies the abdomen becomes transparent so that most of the internal organs are visible. The latter retain their normal aspect, except the fat body which is strongly reduced.

It is interesting to note that flies injected with a *l(3)gl* disc, taken directly from a larva, in general do not develop the ascitic reaction. By contrast, when disc tissue has been previously cultured in 2 successive hosts over a period of about 40 days, a single small fragment of the tumor will induce the reaction in 80–90% of the cases.

It is also noteworthy that the lethal *l(3)gl* larvae become ascitic towards the end of their life. At the age of 7 days when normal larvae are ready to pupate, the lethal larvae are distinguished by a reduced size of some internal organs, especially the salivary glands and the fat body. Imaginal rudiments, imaginal discs, gonads, and parts of the brain are also underdeveloped. Lethal larvae at this stage are more transparent than their normal sibs. They can survive up to 15 more days under favorable conditions, without a supplementary molt, whereby they become very large and transparent (figure 2). During this period, while organs and anlagen remain stationary or degenerate, with the exception of the brain and the one remaining disc, the volume of the lethal larva increases essentially due to an excess amount of hemolymph. This suggests the supposition that there may be a connection between the bloated state of *l(3)gl* larvae and the ascites induced in normal flies by *l(3)gl* tissue. It should however be recalled that certain other mutations also produce bloated lethal larvae, in particular *l(3)tr* of *D. melanogaster*[10] and *l(3)gl* of *D. hydei*[11].

Results of the electrophoretic analysis of hemolymph proteins in the wild-type and *l(3)gl* larvae as well as in the host flies following injection of the lethal disc tissue are illustrated in figures 3 and 4. As can be seen, protein fractions 2, 3, 7 and 8b are detectable only in the *l(3)gl* larvae, among which fraction 7 increases rapidly in concentration during their survival period (figure 4, B and C). It is of interest to note that the same protein fraction accumulates in the hemolymph of host flies carrying the *l(3)gl* tissue. In flies with disc tissue which had been previously cultured in vivo for 530 days, fraction 7 became the most prominent protein band (figure 3, F). Thus, it appears that both the development of the ascitic state in *l(3)gl* larvae and the induction of the ascitic reaction in host flies after injection of the *l(3)gl* disc are correlated with the accumulation of a specific protein in the hemolymph.

In *Drosophila*, as in other insects, the hemolymph proteins are known to be synthesized in the fat body[12,13]. As mentioned above, the fat body in the *l(3)gl* larvae becomes greatly reduced at the end of the survival period. Similarly, a large part of the fat body in the abdomen of the bloated host fly is destroyed. Consequently, the protein accumulated in the hemolymph probably has its origin in some tissue other than the fat body. To what extent this protein is directly involved in the induction of the ascitic state is not yet clear. Further experiments are needed to explain the causal relationships of the phenomena reported here.

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Genetic variability in 3 co-occurring forms of the starfish genus *Othilia (= Echinaster)*

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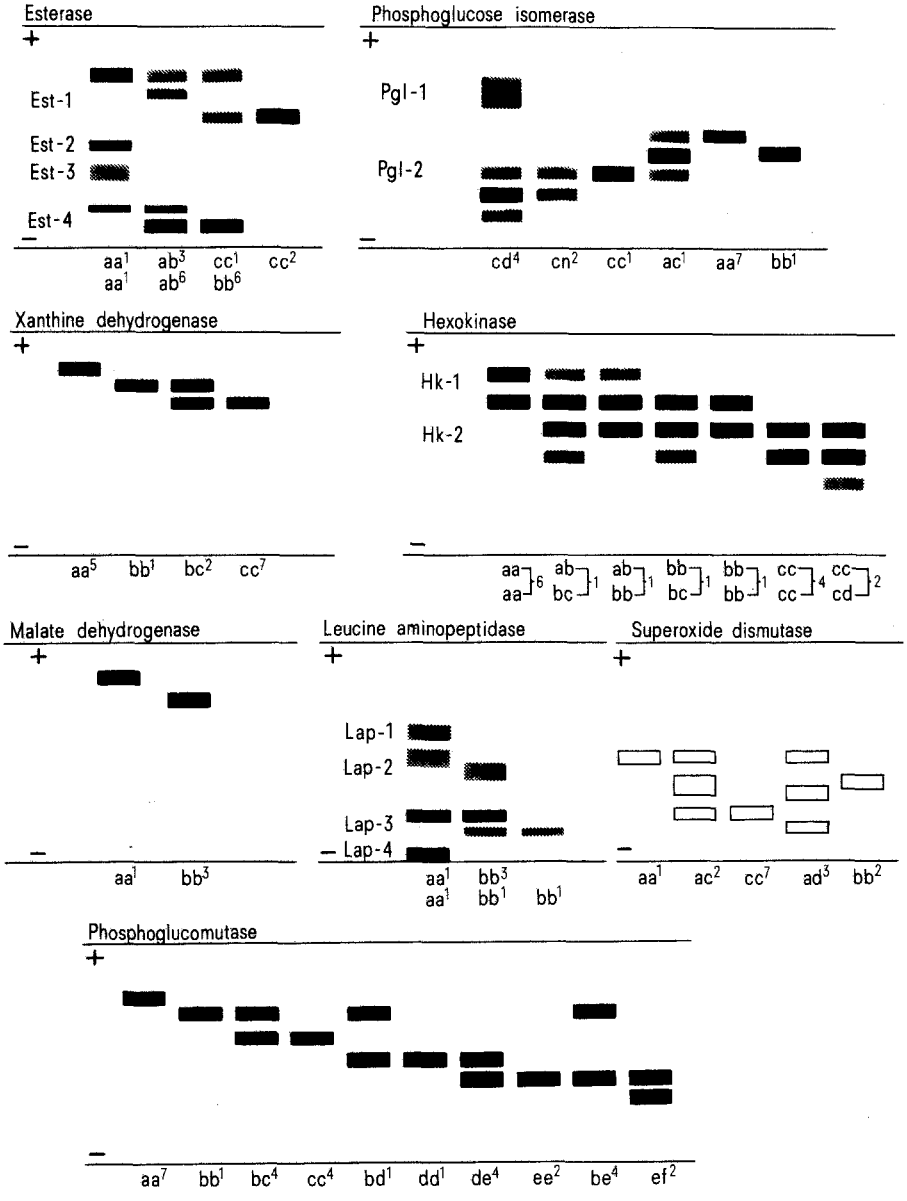
Summary. High levels of genetic variability have been found in 3 (grey, brown, orange) co-occurring forms of the starfish *Othilia*. These high levels appear to correspond to the generalistic habit of this starfish. The biochemical data, coupled with morphological observations, indicate that the grey and orange forms are morphs of the same species and that the brown form is a species separate from the grey and orange forms.

To date, information concerning genetic variation in starfish has been limited[2-4]. Manchenko and co-workers have only recently initiated an extensive survey of allozymic variation among several starfish species from the Sea of Japan[5-10]. In the present study, we compare allozymic variation among 3 co-occurring forms (grey, brown, orange) of the starfish genus *Othilia (= Echinaster)* from the Gulf of Mexico. In addition, allozymic and morphological data have been coupled in order to determine whether these morphs are morphs within a species or diverging species.

Materials and methods: 26 grey, 19 brown and 24 orange specimens of *Othilia* were collected from off the coast of Panama City, Florida and maintained without food for 4–



Allozyme patterns for 8 enzymes in 3 forms of *Othilia*. Dark bands indicate relatively stronger activities seen compared to the stipled bands. The open bands represent achromatic SOD activity. The superscripts indicate in which form(s) a particular pattern was seen: 1 - grey, brown, and orange; 2 - grey only; 3 - brown only; 4 - orange only; 5 - grey and brown; 6 - brown and orange; 7 - grey and orange.

21 days prior to sacrifice. Pyloric caeca were removed and homogenized 1:2 w/v in 9 mM Tris-3 mM citrate-1.2 mM EDTA pH 7 buffer at 4 °C. Homogenates were centrifuged at 27,800 × g for 30 min. The resultant supernatants were subjected to horizontal starch gel electrophoresis. The enzymes examined and buffer systems employed are included in the table. Enzyme staining was modified after Shaw and Prasad[13]. Observations of 17 external skeletal features were made from the dried specimens[14].

Results and discussion: The isozyme patterns of the 8 enzyme systems examined are illustrated in the figure. The various patterns are generated by a minimum of 16 loci in each of the 3 forms. Allelic variation is detectable at 8 of the 16 loci examined in the grey form, 9 of 16 in the orange form, and 11 of 16 in the brown form (table). The grey form is polymorphic (criterion of $p < 0.95$) at 50% (8 of 16) of the loci examined; the brown and orange forms are both polymorphic at 56% (9 of 16) loci examined. 5 loci *(Pgi-1, Est-2, Est-3, Lap-1* and *Lap-4)* are monomorphic in all 3 forms.

In this study the genetic distance (D) is 0.030 between the grey and brown forms, 0.035 between the brown and orange, and 0.007 between the grey and orange[15]. For the grey form the average heterozygosity (H) is 0.255 ± 0.069; for the brown, 0.255 ± 0.058; and the orange, 0.260 ± 0.065[14]. The grey and orange forms are morphologically distinct in only 3 of 17 external morphological characters examined. On the other hand, the brown form is morphologically distinct from both the grey and orange forms in 12 of 17 morphological characters[14].

The biochemical and morphological data support a conclusion that the grey and orange forms are morphs of the same species. Morphological data indicate that the brown form is distinct from the grey-orange complex. Additional data will be required to determine the status of the grey-orange polymorphism and to specify the relationship of the brown form to the grey-orange complex.

The percentage of polymorphic loci, as well as the average heterozygosity seen here in *Othilia* is high in comparison with other invertebrates studied[16]. However, these values are within those which have been recorded for other starfish[2,3]. The enzymes surveyed in the present study

Allele frequencies of 16 enzyme loci in grey, brown and orange forms of *Othilia*

Enzyme (EC number)*	Buffer system**	Locus	Allele	Grey	Brown	Orange
Phosphoglucose isomerase	A	*Pgi-1*	a	1.000	1.000	1.000
(5.3.1.9)		*Pgi-2*	a	0.134	0.026	0.229
			b	0.077	0.158	0.042
			c	0.731	0.816	0.687
			d	0.000	0.000	0.042
			null	0.058	0.000	0.000
Hexokinase	A	*Hk-1*	a	0.077	0.132	0.167
(2.7.1.1)			b	0.846	0.868	0.750
			c	0.077	0.000	0.083
		Hk-2	a	0.000	0.105	0.042
			b	0.750	0.579	0.708
			c	0.212	0.316	0.250
			d	0.038	0.000	0.000
Phosphoglucomutase	A	*Pgm*	a	0.095	0.000	0.059
(2.7.5.1)			b	0.548	0.719	0.470
			c	0.000	0.000	0.118
			d	0.214	0.281	0.294
			e	0.119	0.000	0.059
			f	0.024	0.000	0.000
Malate dehydrogenase	B	*Mdh*	a	1.000	0.947	1.000
(1.1.1.37)			b	0.000	0.053	0.000
Xanthine dehydrogenase	A	*Xdh*	a	0.154	0.579	0.000
(1.2.1.37)			b	0.404	0.421	0.458
			c	0.442	0.000	0.542
Esterase	C	*Est-1*	a	0.596	0.605	0.458
(3.1.1.1)			b	0.000	0.079	0.000
			c	0.404	0.316	0.542
		Est-2	a	1.000	1.000	1.000
		Est-3	a	1.000	1.000	1.000
		Est-4	a	0.000	0.158	0.271
			b	1.000	0.842	0.729
Leucine aminopeptidase	C	*Lap-1*	a	1.000	1.000	1.000
(3.4.11.1)		*Lap-2*	a	1.000	0.684	1.000
			b	0.000	0.316	0.000
		Lap-3	a	0.558	0.447	0.646
			b	0.442	0.553	0.354
		Lap-4	a	1.000	1.000	1.000
Superoxide dismutase	B	*Sod*	a	0.000	0.000	0.020
(1.15.1.1)			b	0.558	0.974	0.917
			c	0.192	0.000	0.000
			d	0.250	0.000	0.063
			e	0.000	0.026	0.000

* Enzyme Commission number. ** Buffer systems used are: A, Tris-citrate-EDTA pH 7.0; B, Tris-borate-EDTA pH 8.6; C, borate-NaOH gel pH 8.5, electrode pH 8.0.

included monomeric and dimeric forms, non-specific and major metabolic enzymes. As both monomorphic and polymorphic loci were found for each category, the high proportion of polymorphic loci cannot be attributed to the experimental bias which may relate to enzyme structure or function[17,18]. A preliminary examination of a distinct *Othilia* form from off the west-central coast of Florida suggests that high levels of genetic polymorphism also exist in this *Othilia* form. Thus, the high degree of genetic variability among *Othilia* in this study cannot be designated as a geographic phenomenon.

There has been some interest in comparisons of genetic variability between habitat generalists and specialists. In his survey, Nevo has found a significant correlation between genetic variability and ecological heterogeneity[16]. He concludes that generalists are relatively more polymorphic and heterozygous than specialists.

Othilia is a cosmopolitan genus, occurring in tropical and subtropical waters around the world. Studies on feeding behavior indicate that *Othilia* is an opportunist, feeding at the water-sediment interface[17]. These observations indicate that *Othilia* (relative to other starfish) is a generalist. Therefore, consistent with Nevo, the high levels of polymorphism seen in the 3 forms of *Othilia* would seem to correspond to the generalistic habitat of this starfish.

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