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Genetic variation in the planktonic chaetognaths *Parasagitta elegans* and *Eukrohnia hamata*

Erik V. Thuesen*, Ken-ichi Numachi, Takahisa Nemoto**

Ocean Research Institute, University of Tokyo, 1-15-1 Minamidai, Nakano-ku, Tokyo 164, Japan

ABSTRACT: Two species of planktonic chaetognaths, *Parasagitta elegans* (Verrill) and *Eukrohnia hamata* (Möbius), from waters off Japan were analyzed electrophoretically and found to display very low levels of genetic variability. Nineteen enzyme loci were examined for 7 population samples of *P. elegans*, and the proportion of polymorphic loci ($P_{0.95}$) and average frequency of heterozygotes per locus (H) were calculated as 0.11 and 0.026, respectively. Fifteen enzyme loci were examined for 2 populations of *E. hamata*, and $P_{0.95}$ and H were calculated as 0.10 and 0.038, respectively. On the basis of allele frequencies at 2 enzyme loci it is possible to suggest that *P. elegans* population samples collected in the Sea of Japan were reproductively isolated from those in the Oyashio. Moreover it would appear that 4 population samples of *P. elegans* in the Sea of Japan are representative of a panmictic population. Differences in allele frequencies between population samples of the Oyashio suggest that these populations are genetically structured. It appears that genetic structuring exists in the 2 population samples investigated for *E. hamata* also. *P. elegans* and *E. hamata* expressed no common alleles over all the loci assayed indicating that the 2 species are phylogenetically very distant within the phylum Chaetognatha.

INTRODUCTION

The small marine phylum Chaetognatha is composed primarily of holoplanktonic predators which feed mainly on copepods. Although they are commonly the largest constituent of carnivorous zooplankton biomass in the earth's oceans, little is known of chaetognath population structure. In recent years advances have been made in understanding the population structure of other marine zooplankters through analyses of electrophoretically detectable differences in enzymes. Since these differences usually result from variations in the amino acid sequence of the enzyme which are controlled by changes in the DNA, they can be considered single locus genetic markers and used to determine genetic differences between conspecific populations (Nei 1972).

Ayala et al. (1975) were the first to perform biochemical genetic studies of zooplankton populations when they analyzed genetic variability in the antarctic krill *Euphausia superba*. As the study of krill populations has attracted the attention of biologists for decades, many other species of krill were also soon analyzed electrophoretically, including *E. mucronata* and *E. distinguenda* (Valentine & Ayala 1976), *E. crystallorophias* (Fevolden & Ayala 1981), *Meganyctiphanes norvegica* and *Thysanoessa raschi* (Fevolden 1982), *E. pacifica* (Fevolden 1986) and *E. krohnii* and *Nematoscelis megalops* (Bucklin & Weibe 1986). In addition to krill, several other planktonic crustacea have been investigated, the copepods *Labidocera aestiva* (Bucklin & Marcus 1985) and *Metridia pacifica* (Bucklin et al. 1989, Bucklin 1991), as well as 4 species of hyperiid amphipods, *Themisto compressa*, *T. abyssorum*, *T. libellula* and *T. gaudichaudii* (Schneppenheim & Weigmann-Haass 1986).

Most of these authors and others (e.g. Schneppenheim & MacDonald 1984, Kühl & Schneppenheim 1986) have generally found these planktonic popula-

* Present address: Oceanic Biology Group, Marine Science Institute, University of California, Santa Barbara, California 93106, USA

** Deceased

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tions to have high levels of genetic homogeneity within the sampling areas. Fevolden & Schneppenheim (1988, 1989) carried out further electrophoretic investigations on *Euphausia superba* and were not able to discriminate among populations in the Pacific, Atlantic and Indian Ocean Sectors of the Southern Ocean.

Notable exceptions to the generalization of genetic homogeneity in the pelagic zooplankton populations in the Southern Ocean are the conclusions of Bucklin and her co-workers. Western Atlantic populations of *Labidocera aestiva* (Bucklin & Marcus 1985), North Atlantic Slope Water euphausiid populations (Bucklin & Weibe 1986) and California Current populations of *Euphausia pacifica* (Bucklin 1986) and *Metridia pacifica* (Bucklin et al. 1989, Bucklin 1991) all showed some evidence of genetic structuring. Although much work has been done on planktonic crustacea, no biochemical analyses have been performed to elucidate the population structure of other zooplanktonic taxa in the marine environment.

Parasagitta elegans (Verrill) is a relatively large boreal chaetognath which is most abundant in neritic waters. Its usual southernmost limit of occurrence is the lower boundary of the Pacific Subarctic Water (Bieri 1959), and it is known as an indicator species of the Oyashio Current. *Eukrohnia hamata* (Möbius) is a cosmopolitan pelagic chaetognath which appears close to the surface in arctic and antarctic waters and submerges in lower latitudes (Bieri 1959). In the Sea of Japan, *P. elegans* is generally found north of 37° (Tokioka 1940). The exchange of water in the northern Sea of Japan is fairly limited (Zenkevitch 1963, Nishimura 1969), and meso- and bathypelagic chaetognaths such as *E. hamata*, *E. bathypelagica* and *Caecosagitta macrocephala* that are common in the Pacific Ocean are absent from the Sea of Japan. Although it is thought that in late summer the warm Tsushima Current can reach around the northern end of Honshu, Japan, through the Tsugaru Strait and apparently carry zooplankton populations from the Sea of Japan to the eastern side of Honshu (Furuhashi 1984), it is not known to what extent the exchange of northern zooplankton populations occurs through the shallow northern straits connecting with the Sea of Okhotsk and the Pacific Ocean. The Mamiya Strait, Soya Strait and Tsugaru Strait are 5, 53 and 130 m in depth respectively (Zenkevitch 1963). The present study was undertaken to compare the genetic composition of *P. elegans* in the Sea of Japan with that in the Oyashio and to increase our understanding of the genetic structure of zooplankton populations in general.

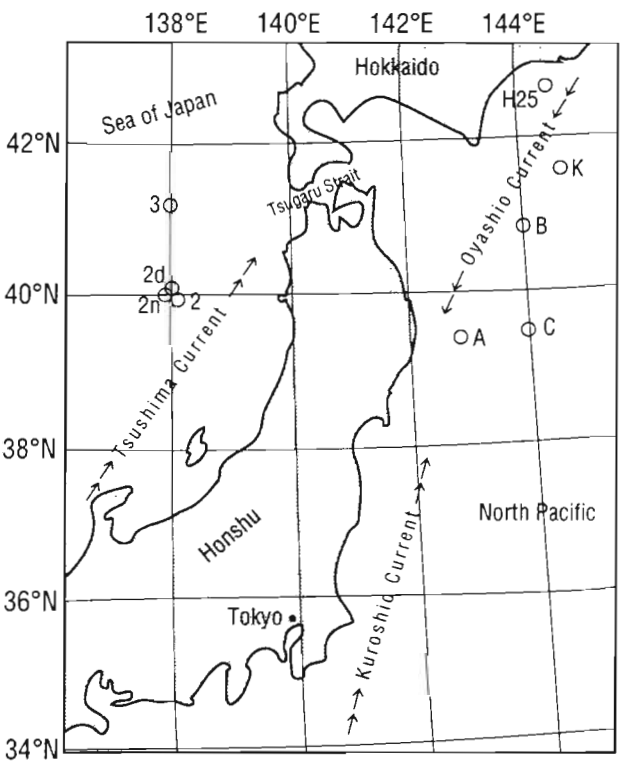

Fig. 1 Locations of sampling stations off the coast of Japan. Station data given in Table 1

METHODS

Chaetognath specimens. Plankton sampling was undertaken on the RV 'Tansei Maru' during cruises KT-87-7 to the Sea of Japan, and KT-87-11 to the Oyashio-Kuroshio mingling waters off the northern coast of Honshu (the main island of Japan), cruise KH-87-4 of the RV 'Hakuho Maru' to the Kuroshio warm core ring area and the Oyashio south of Hokkaido, Japan, and the RV 'Iwate Maru' Salmon I cruise of the Kamaishi Iwate Prefectural Fisheries Station (Fig. 1). Several types of sampling apparatus were used to collect specimens. Oblique tows of ORI nets (Omori 1965), horizontal tows of MTD closing nets (Motoda 1971), and a 113 cm diameter fish larvae net were employed as listed in Table 1.

The chaetognath specimens removed from 1 net tow were considered to comprise 1 population sample. Population samples of *Parasagitta elegans* were collected at stations in the Sea of Japan and Oyashio (Table 1). Populations samples of *Eukrohnia hamata* were collected in the Oyashio and Kuroshio warm core ring area (Table 1). Chaetognaths were immediately removed from the plankton samples before the addition of any preservatives, and identified with a binocular microscope. Only mature individuals in good condition were taken for analyses. Chaetognaths are commonly cannibalistic predators and occasionally contain visible parasites. Only specimens without visi-

Table 1. Chaetognath population samples

No.	Date	Stn	Net	Depth (m)
Parasagitta elegans				
51	20 Apr 1987	A	Ring net	0–68
31	10 Jun 1987	2	MTD	250
28	10 Jun 1987	2d	ORI	0–669
13	10 Jun 1987	2n	MTD	50
41	11 Jun 1987	3	ORI	0–334
15	12 Jun 1987	B	ORI	0–322
30	12 Sep 1987	H25	MTD	200
Eukrohnia hamata				
40	12 Jun 1987	C	ORI	0–347
35	13 Aug 1987	K	MTD	250

ble gut contents or parasites were used to avoid the possibility of sample contamination. While still living, chaetognaths were rinsed very briefly in distilled water to remove excess salt, immediately blotted dry on filter paper to remove excess water, and frozen in liquid nitrogen. Specimens were transported to the laboratory and remained in liquid nitrogen until use with one exception, when animals which had been originally frozen in liquid nitrogen were transported while stored in dry ice.

Chaetognaths were homogenized with a glass rod in welled glass plates either on ice or in a 5 °C cold room. Rectangular pieces of filter paper (5.0 × 1.5 mm) were applied directly to the sample homogenate without the addition of any extraction buffer, adapting the methods of Numachi (1971) to smaller sample size. These filter papers containing chaetognath cell lysates were then applied to the starch gels. This method of preparation allowed each chaetognath specimen to be examined at every enzyme locus.

Starch gel electrophoresis and interpretation of electrophoretic data. Horizontal starch gel electrophoresis was performed following the methods outlined by Numachi et al. (1979). Four buffer systems were selected for the preparation and running of gels. Three CAEA buffer systems [citric acid and N-(3-amino-propyl)-diethanolamine with (1) NADP, (2) NADP and EDTA and (3) NADP, ATP and Mg^+, all at pH 7.0], were adapted from Clayton & Tretiak (1972), and the preparation of TBE buffer (Tris, boric acid and EDTA with NAD and ATP; pH 8.7) followed Kraus & Neely (1964). Starch gels of 12.5% and 6 mm thick were run at a constant current (CAEA buffers 3.6 mA cm^{-2}; TBE buffer 2.0 mA cm^{-2}) until a control marker of amide black 10B migrated a distance of 5 cm.

Each gel was sliced into 6 sheets 1 mm thick and stained for enzymes according to Shaw & Prasad (1970) and Numachi et al. (1979). A survey of 34 enzyme systems was carried out to identify enzyme loci capable of serving as genetic markers. Sixteen enzyme systems were found to be routinely assayable and were chosen for investigation. Stained gels were preserved between cellophane sheets for future analysis following the methods given by Numachi (1981). The naming of loci and alleles follows that described by Shaklee et al. (1990). The most common allele at each enzyme locus was scored as 100 and all other alleles were referred to as 100 plus or minus the distance (in mm) of electrophoretic mobility greater or less than this standard allele.

Indices of genetic variation were calculated for electrophoretic data according to Ayala & Valentine (1978). Heterozygosity (H) was measured as the observed frequency of heterozygotes per locus averaged over all loci in the sample. The proportions of enzyme loci in a sample which were polymorphic at greater than 5% and 1% of the loci observed were expressed as $P_{0.95}$ and $P_{0.99}$, respectively. χ^2 analyses were performed to determine if the population samples were representative of panmictic populations or genetically structured. When necessary, χ^2 calculations adjusted for small sample size were performed according to Sokal & Rohlf (1981). Nei's (1972) measure of genetic distance (D) was computed in order to quantify the genetic differentiation which was observed between chaetognath population samples, and a dendrogram was constructed using the unweighted pair-group method using arithmetic averages (UPGMA; Sneath & Sokal 1973) to illustrate the genetic divergence between population samples.

RESULTS

Nineteen loci encoding 16 enzymes were resolved for *Parasagitta elegans* and were the primary focus of this study (Table 2). In addition, 15 loci encoding 12 enzymes of *Eukrohnia hamata* were stained with sufficient clarity for analysis (Table 2). The glycolytic enzymes lactate dehydrogenase, octopine dehydrogenase, strombine dehydrogenase and alanopine dehydrogenase could not be detected in either of these chaetognaths, although many buffer systems at several different pHs were used in the preliminary survey. *Eukrohnia hamata* shared no common alleles with *P. elegans* at any of the enzyme loci examined.

Alleles and allele frequencies of the 8 polymorphic enzyme loci examined for 7 population samples of *Parasagitta elegans* are presented in Table 3. Alleles and allele frequencies of the 3 polymorphic enzyme loci resolved for the 2 population samples of *Eukrohnia hamata* are presented separately (Table 4). Both species of chaetognaths were found to display low levels

Table 2. *Parasagitta elegans* and *Eukrohnia hamata*. Chaetognath enzyme loci. na: not assayed

Enzyme (E.C. no.)	Buffer	*P. elegans* loci	*E. hamata* loci
Acid phosphatase (3.1.3.2)	CAEA 2	*ACP*	*ACP*
Adenylate kinase (2.7.4.3)	CAEA 2	*AK*	*AK*
Asparate aminotransferase (2.6.1.1)	CAEA 2	*AAT*	*AAT*
Esterase (3.1.1.1)	CAEA 1	*EST*	*EST-1, EST-2*
Glutathione reductase (1.6.4.2)	CAEA 1	*GLR*	*GLR*
Glycerol-3-PO_4 dehydrogenase (1.2.1.12)	TBE	*G3PDH*	*G3PDH-1, G3PDH-2*
Isocitrate dehydrogenase (1.1.1.42)	CAEA 3	*ICD-1, ICD-2*	*ICD-1, ICD-2*
Leucine aminopeptidase (3.4.11.?)	TBE	*LAP*	*LAP*
Malate dehydrogenase (1.1.1.37)	CAEA 3	*MDH*	*MDH*
Malic enzyme (1.1.1.40)	CAEA2	*ME-1, ME-2*	*ME*
Mannose phosphate isomerase (5.3.1.8)	TBE	*MPI*	na
Peptidase [LeucylGlycine] (3.4.13.?)	CAEA 3	*PEP-1, PEP-2*	na
Phosphoglucomutase (2.7.5.1)	TBE	*PGM*	*PGM*
6-Phosphogluconate dehydrogenase (1.1.1.44)	CAEA 1	*6-PGD*	*6-PGD*
Phosphohexose isomerase (5.3.1.9)	CAEA 3	*PHI*	na
Pyruvate kinase (2.7.1.40)	CAEA 2	*PK*	na

Table 3. *Parasagitta elegans*. Allele frequencies of polymorphic loci. nd: not determined

Locus	Alleles	Sea of Japan				Oyashio		
		2	2d	2n	3	A	B	H25
AAT	n	31	28	13	41	47	15	30
	*92	0	0	0	0	0.011	0	0
	*96	0	0	0	0.012	0.245	0.067	0.450
	*100	1.000	1.000	1.000	0.988	0.745	0.933	0.550
GLR	n	31	nd	9	41	51	15	30
	*100	0.968	–	0.944	0.963	0.863	1.000	0.833
	*103	0.032	–	0.056	0.037	0.137	0	0.167
G3PDH	n	31	28	13	41	47	15	30
	*100	1.000	0.982	1.000	1.000	1.000	1.000	1.000
	*103	0	0.018	0	0	0	0	0
ICD-1	n	31	nd	13	41	nd	15	30
	*98	0.016	–	0	0	–	0	0
	*100	0.984	–	1.000	1.000	–	1.000	1.000
ICD-2	n	31	28	13	41	51	15	30
	*95	0.016	0.054	0.038	0.073	0.039	0	0.017
	*100	0.984	0.946	0.962	0.927	0.961	1.000	0.983
MDH	n	31	28	13	41	51	15	30
	*100	0.935	0.982	0.962	0.988	0.980	1.000	1.000
	*106	0.065	0.018	0.038	0.012	0.020	0	0
ME-1	n	31	28	13	41	51	15	30
	*96	0	0	0	0.012	0	0	0
	*100	1.000	1.000	1.000	0.976	1.000	1.000	1.000
	*102	0	0	0	0.012	0	0	0
PHI	n	31	28	13	41	51	15	30
	*90	0.016	0	0.038	0	0	0	0
	*94	0	0	0	0	0	0.067	0
	*96	0.048	0	0	0.037	0.010	0	0
	*98	0.097	0	0	0.012	0.029	0	0.017
	*100	0.806	0.964	0.962	0.939	0.951	0.933	0.967
	*102	0	0	0	0	0.010	0	0
	*104	0.032	0.036	0	0.012	0	0	0

Table 4. *Eukrohnia hamata*. Allele frequencies of polymorphic loci

Locus	Allele	Station C	Station K
6PGD	n	40	32
	*96	0.250	0.016
	*100	0.750	0.984
MDH	n	40	33
	*95	0.138	0.152
	*100	0.812	0.666
	*105	0.050	0.182
ME	n	40	35
	*95	0.013	0
	*100	0.987	1.000

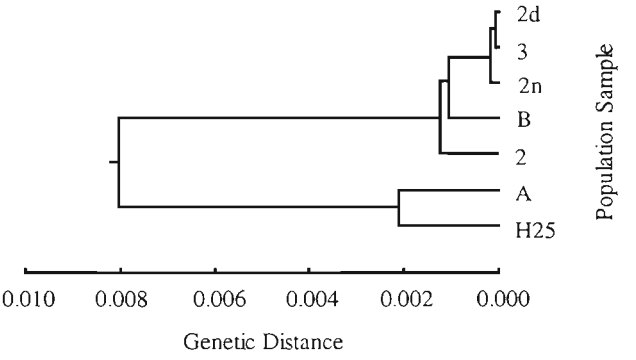

Fig. 2. *Parasagitta elegans*. Dendrogram generated by UPGMA of genetic distance between population samples. Stns A, B & H25 are in the Oyashio and the others are in the Sea of Japan

Table 5. *Parasagitta elegans* and *Eukrohnia hamata*. Genetic heterozygosity (H) and proportion of polymorphic loci at 5% ($P_{0.95}$) and 1% ($P_{0.99}$) in chaetognath population samples

Sample	No.	$P_{0.95}$	$P_{0.99}$	H
Parasagitta elegans				
2	31	0.111	0.278	0.025
2d	28	0.062	0.250	0.016
2n	13	0.055	0.222	0.021
3	41	0.111	0.278	0.023
A	51	0.118	0.294	0.036
B	15	0.111	0.111	0.025
H25	30	0.167	0.222	0.035
Average		0.105	0.236	0.026
Eukrohnia hamata				
C	40	0.133	0.200	0.047
K	35	0.067	0.133	0.029
Average		0.100	0.167	0.038

of genetic heterozygosity (Table 5) and low proportions of polymorphic loci (Table 5) in all population samples.

Expected phenotype frequencies matched with observed phenotype frequencies in most of the population samples according to χ^2 analyses. However, deviations from Hardy-Weinberg equilibrium due to homozygous excesses were observed at the *AAT* and *GLR* loci ($\chi^2_{adj[1]} = 3.99$, $p < 0.05$ and $\chi^2_{adj[1]} = 4.26$, $p < 0.05$, respectively) in the Stn A population sample and at the *AAT* locus ($\chi^2_{adj[1]} = 4.33$, $p < 0.05$) in the Stn H25 population sample. The occurrence of rare alleles at the *PHI* locus in population samples from the Sea of Japan also resulted in statistical deviations from Hardy-Weinberg equilibrium, however expected χ^2 distributions were observed when rare heterozygotes were excluded from the data set.

Allele frequencies at polymorphic loci were mostly similar across all population samples for *Parasagitta elegans*. Noticeable exceptions were found at the *AAT* and *GLR* loci (Table 3). The 2 most polymorphic loci of *Eukrohnia hamata* displayed different allele frequencies between the 2 population samples (Table 4). Comparisons of genetic distance (Table 6) showed that *P. elegans* population samples were very similar. Two of the 3 Oyashio population samples (Stns A & H25) were more closely related to each other and to the third Oyashio population sample than to the population samples from the Sea of Japan. The Sea of Japan population samples showed higher similarity to each other. These relationships are represented diagrammatically in a dendrogram generated by UPGMA (Fig. 2). Although the genetic distances between population samples were small, analysis by UPGMA resulted in the formation of 2 groups: 2 Oyashio population samples, A and H25, forming one group and the Sea of Japan samples along with the small homogeneous Oyashio population sample from Stn B forming the other group. Nei's D was calculated for the 2 *E. hamata* population samples at 0.0052.

Table 6. *Parasagitta elegans*. Nei's genetic distance between population samples

Sample	H25	B	3	2n	2d	2
A	0.0023	0.0030	0.0037	0.0038	0.0044	0.0050
2	0.0134	0.0014	0.0011	0.0011	0.0012	
2d	0.0126	0.0006	0.0002	0.0003		
2n	0.0118	0.0007	0.0002			
3	0.0116	0.0007				
B	0.0095					

DISCUSSION

Population structure of chaetognaths

Population structuring has been defined as genetic differences arising among populations due to barriers which prevent interbreeding (cf. Avise & Felley 1979). The low percentages of polymorphic loci and low level of heterozygosity displayed at these loci rendered both *Parasagitta elegans* and *Eukrohnia hamata* difficult subjects for the investigation of such population structuring. However the differences in allele frequencies which were observed between population samples suggest that some genetic structuring of these populations over the study area does exist.

Although only a few differences were seen in allele frequencies between *Parasagitta elegans* population samples from the Sea of Japan and Oyashio, analysis by UPGMA showed a tendency to group 2 of the Oyashio population samples together. This result was due to the higher frequencies of *AAT*96* and *GLR*103* alleles found in these population samples. Effects of small sample size may partially explain the lower frequencies found in the third Oyashio population sample. The differences in allele frequencies at these 2 loci suggest that barriers exist between the *P. elegans* populations of the Sea of Japan and the Oyashio. If these results are not obscured by small sample size, the observed differences in allele frequencies at this station may present further evidence of genetic structuring and possibly leakage of Sea of Japan populations through the Tsugaru Strait. The similarities in allele frequencies at 3 polymorphic loci between the Stn A and Stn H25 population samples would appear to suggest that these 2 samples come from a single panmictic population, however the marked difference in the allele frequencies at the *AAT* loci indicates the opposite. The Stn A population sample was taken over 500 km downstream in the Oyashio and almost 5 mo previous to the Stn H25 population sample.

The differences in allele frequencies at the 2 $P_{0.95}$ loci, *6PGD* and *MDH*, of *Eukrohnia hamata* suggest that the 2 population samples came from at least partially isolated populations, despite smaller differences in spatial and temporal distributions. In contrast to the evidence found supporting the idea of genetic structuring of chaetognath populations in the Pacific study area, the Sea of Japan population samples appear to represent a panmictic population similar to those found in antarctic euphausiids.

Kühl & Schneppenheim (1986) computed genetic distance values between population samples of 2 species of antarctic krill to be on the order of 0.0002. They suggested that such low values were indicative of large, panmictic zooplankton populations in the antarctic pelagic ecosystem. Similar allele frequencies between population samples of a hyperiid amphipod in the antarctic environment have been cited as further support of this statement (Schneppenheim & Weigmann-Haass 1986). The *D*-values for population samples of *Parasagitta elegans* (Table 6, Fig. 2) may be interpreted as supporting the idea of panmixis for those population samples in the Sea of Japan with barriers existing with the Oyashio population samples. The one population sample from the Oyashio which refutes this supposition was much smaller than the others possibly accounting for this apparent deviation. The value of D for the 2 population samples of *Eukrohnia hamata* was similar to that displayed by the *P. elegans* population samples from the Oyashio. The *D*-values reported by Bucklin and co-workers (Bucklin & Marcus 1985, Bucklin 1986, Bucklin & Weibe 1986) for planktonic copepods and krill are all of an order of magnitude larger than that reported here for chaetognaths. Albeit their studies were undertaken with relatively low numbers of samples and/or very low numbers of enzyme loci.

Homozygous excesses have been repeatedly observed among marine molluscs (Singh & Green 1985). Greater frequencies of homozygotes have been found in juveniles than in adults, possibly as a result of selection against homozygosity (Koehn et al. 1973). Moreover, the extent of homozygous excess has been observed to vary greatly among loci (Singh & Green 1985). Bucklin & Marcus (1985) observed many heterozygote deficiencies in the planktonic copepod *Labidocera aestiva*, which they ascribed to restricted gene flow between populations or differences in factors of selection at work on the populations. In contrast, departures from expected χ^2 frequencies have been observed as heterozygote excesses in krill (Fevolden 1984). The homozygous excesses found in the 2 Oyashio population samples could be interpreted as an example of the Wahlund effect, i.e. the mixing of 2 separate populations in the sample.

One possible mechanism for such population mixing could be a Bering Sea population combining with one from the Subarctic North Pacific in the Oyashio and therefore accounting for the observed homozygous excesses. Kotori et al. (1986) found that *Parasagitta elegans* apparently reproduced during winter under the ice in a large lagoon in northern Hokkaido. Transport out of such bays in the Bering Sea, Sea of Okhotsk and coastal waters of the Kamchatka Peninsula may also be able to influence the genetic composition of Oyashio *P. elegans* populations, if neritic breeding populations have different genetic compositions. The effect of founder events or localized patchiness on zooplankton population structure is unknown. It is also possible that current mingling in the Oyashio-Kuroshio area affects

chaetognath population structure as chaetognaths reach reproductive maturity. Two studies that attempted to elucidate the relationships between physical processes in the ocean and the genetic structure of a pelagic copepod, *Metridia pacifica*, found no statistically significant correlation between hydrographic features and allozyme allele frequencies (Bucklin et al. 1989, Bucklin 1991).

To date there have been no studies on the life histories and ecology of chaetognaths in the Oyashio Current. Reproduction spans from 6 to 10 mo in pelagic populations of *Parasagitta elegans* in the eastern North Pacific (Terazaki & Miller 1986) to 2 yr in neritic populations in the Arctic (Dunbar 1962). Terazaki & Miller (1986) suggested that there may be genetic differences between stocks of *P. elegans* on the basis of discrepancies in average body lengths. However, Fevolden (1986) found it impossible to correlate electrophoretically detectable genetic differences to such size discrepancies in stocks of *Euphausia superba* near the Antarctic Peninsula.

A different explanation for the observed homozygous excesses relates to the reproductive biology of *Parasagitta elegans*. Chaetognaths are hermaphroditic, and some species are evidently capable of self-fertilization (Jägersten 1940, Nagasawa 1987). In the course of sorting live *P. elegans* for electrophoretic analysis, mature individuals with sperm streaming out of the seminal vesicles and progressing anteriorly along the body towards the seminal recepticles were observed on several occasions, observations similar to those made by Jägersten (1940). The possibility of self-fertilization in *P. elegans* cannot be excluded, and a section of the population utilizing self-fertilization could be responsible for the observed homozygous excesses. The other possible explanation, selective advantage for homozygotes, is not analyzable with the present data, since all of the specimens used in the study were mature adults.

Rare alleles causing deviations from expected χ^2 frequencies were found at the *PHI* locus in the population samples from the Sea of Japan. Some workers have ignored rare alleles in χ^2 calculations (Fevolden 1986) and others have interpreted them as being significant (Bucklin & Marcus 1985). MacDonald et al. (1986) found discrepancies between populations of *Euphausia superba* due to rare alleles but considered them insignificant in the face of other data overwhelmingly supporting the idea of a large panmictic population of krill. Differences in electrophoretic techniques have prohibited the comparison of results concerning the structure of the relatively well-investigated antarctic krill populations, and it remains unknown what changes occur in the frequencies of rare alleles over time. Furthermore, comparisons of different biochemical genetic techniques have indicated that selection occurring at allozyme loci may obscure population structuring over large geographic areas (Karl & Avise 1992).

Genetic homogeneity of chaetognaths

Parasagitta elegans and *Eukrohnia hamata* both were found to have low levels of $P_{0.99}$, $P_{0.95}$ and H when compared with those of other marine zooplankton (Table 7). Much discussion and debate has taken place concerning advantages and mechanisms behind enzyme polymorphisms in species and populations (Kimura & Ohta 1971, Selander & Kaufman 1973, reviewed in Nei & Koehn 1983). The 'environmental heterogeneity-trophic diversity' model of Nelson &

Table 7. Comparison of genetic variation in zooplankton. Parameters as in Table 5

Taxa	Location	$P_{0.99}$	$P_{0.95}$	H	Loci observed	Data source
Euphausiacea						
Euphausia superba	Southern Ocean	0.53	0.40	0.118	15	Kuhl & Schneppenheim (1986)
E. crystallorophias	Southern Ocean	0.35	0.29	0.094	17	
E. krohnii	North Atlantic	1.00	0.75	0.230	8	Bucklin & Weibe (1986)
Copepoda						
Labidocera aestiva	Western Atlantic	1.00	0.78	0.25	6	Bucklin & Marcus (1985)
Metridia pacifica	California Current	0.64	0.60	0.18	9	Bucklin et al.(1989)
Amphipoda						
Themisto libellula	Southern Ocean	0.88	0.33	0.133	9	Schneppenheim & Weigmann-Haass (1986)
T. gaudichaudii	Southern Ocean	0.70	0.50	0.230	10	
Chaetognatha						
Parasagitta elegans	Sea of Japan and Oyashio	0.24	0.11	0.026	19	Present study
Eukrohnia hamata	Western North Pacific	0.17	0.10	0.038	15	Present study

Hedgecock (1980) and its predecessors (Selander & Kaufman 1973, Ayala 1976) have most often been applied to interpretations of zooplankton electrophoretic data (Valentine & Ayala 1976, Ayala & Valentine 1979, Fevolden 1984). The low levels of enzyme polymorphism and genetic heterogeneity observed in *P. elegans* and *E. hamata* match well with that predicted for fine-grained adaptive strategists by the 'environmental heterogeneity-trophic diversity' model proposed by Nelson & Hedgecock (1980). Comparisons of the genetic diversity data for *P. elegans* and *E. hamata* are difficult to perform, since both display such low levels of variation. Although some difficulties are inherent in these methods of investigation and analysis, it should be considered profitable to analyze biochemical genetic differences in planktonic taxa to gain insights into their biology, ecology and distribution patterns. Newer techniques of molecular biology should prove to be very fruitful towards achieving this goal (Powers et al. 1990, Falkowski & Laroche 1991).

Phylogenic implications

Much speculation concerning the phylogenic relationships between the 100+ species that belong to the phylum Chaetognatha has occurred, but few analytical analyses have been undertaken (Bieri 1991). Tokioka (1965a, b) erected a taxonomic system which described the higher taxonomic categories of the Chaetognatha and split the most common genus, *Sagitta*, into 9 different genera. In contrast to the low levels of conspecific variation which were observed in this study, electrophoretic results show that a very large phylogenetic gap exists between *Eukrohnia hamata*, a member of the order Phragmophora Tokioka, and *Parasagitta elegans*, a member of the order Aphragmophora Tokioka. Our data are therefore in agreement with Tokioka's division of these 2 chaetognath suprataxa. A large-scale electrophoretic study of chaetognath enzyme loci would no doubt be very valuable to elucidate phylogenetic relationships within the phylum.

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