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Systematics of the *Perinereis nuntia* Complex (Polychaeta: Nereididae) in South-eastern Australia

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ABSTRACT. Biochemical and morphological analyses show that two species in the *Perinereis nuntia* group occur in south-eastern Australia. The two species are distinguished by fixed differences at 4 of 8 enzyme loci, by non-overlapping ranges of paragnath counts and by qualitative differences in the types of paragnaths present. Previously, all material from south-eastern Australia had been referred to a single species, *Perinereis nuntia*, with two nominal varieties: *P. nuntia vallata* and *P. nuntia brevicirris*. Analysis of intraspecific variation shows that these varieties have no taxonomic significance. Differences between populations, especially those from markedly different sediments, exaggerate intraspecific variation in paragnath counts. Size related variation of paragnath counts is relatively unimportant.

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Sixteen nereidid genera and about 350 species carry chitinous denticles, "paragnaths", on the eversible proboscis (Fauchald, 1977); this amounts to about 40% of all genera and 80% of all species in the family. In most genera paragnaths are arranged in a regular grouping (Fig.1a,b), and counts of paragnaths in each numbered group are the most important character system used by taxonomists to describe and distinguish taxa. Although this system of meristic characters is ideally suited to statistical analysis of intraspecific and interspecific variation, there has been little work of this nature that could guide taxonomic decision-making. Barnes & Head (1977) and Barnes (1978) provided detailed statistical analysis of the ecological significance of variation in paragnath number in several populations of *Nereis diversicolor* Müller, 1776 (also referred to in the literature as *Hediste diversicolor*) but they studied only that species and did not attempt a taxonomic analysis. No taxonomic studies of nereidids have used statistical methods.

Perinereis nuntia and related species comprise a group characterised by an arc of short bar-shaped paragnaths on group VI of the proboscis (Fig.1a). At least 18 nominal species and varieties have been described, of which *P. nuntia* is by far the most widely reported, being recorded from all continents except Antarctica, North America and northern Europe (Wilson & Glasby, 1993). Published descriptions indicate considerable variation in morphology within and between populations, and trinomens have been widely used to identify supposed morphs and geographical races (Fauvel, 1932; Hartman, 1954; Knox, 1960a; Imajima, 1972). Among recent authors, only Paik (1975) and Hutchings & Turvey (1982) have attempted a more critical appraisal of

morphological variability. Paik examined numerous specimens from two localities in South Korea but provided incomplete data and limited analysis; Hutchings & Turvey compared Australian material with specimens from India, China and New Caledonia. Neither of these papers used statistical methods to compare populations and both papers failed to detect consistent differences within *P. nuntia* and recognised only a single species.

My aim in this study was to use electrophoretic analysis of enzymes to establish species boundaries independently of paragnath morphology. Statistical analysis of paragnath counts would then provide a better understanding of intraspecific and (if more than one species was shown to be present) interspecific variation. These results are applied to a worldwide revision of the *Perinereis nuntia* species group by Wilson & Glasby (1993).

All tables are listed in the Appendix.

Biochemical Systematics

Electrophoresis of enzymes is a powerful means of identifying species and is independent of the morphological characters used in traditional taxonomic analysis (Avise, 1975; Ferguson, 1980). Measures of genetic distance such as Nei's Distance (Nei, 1972) are sometimes used in taxonomic studies, but I have followed Richardson *et al.* (1986) in using the proportion of fixed allelic differences as a more practical and biologically sensible indication of species-level differences. (Richardson *et al.*, 1986 showed that, except in extremely polymorphic organisms, the proportion of fixed differences is the major component in calculation of genetic similarity measures.)

Several studies have applied electrophoretic methods to resolve taxonomic problems in polychaetes. A summary of the more rigorous studies illustrates the application of Richardson *et al.*'s fixed allelic difference criterion. Grassle & Grassle (1974, 1976) revealed a complex of 6 sympatric sibling species where a single species, *Capitella capitata*, (Capitellidae) had previously been recognised. Eight enzyme loci were studied and fixed differences were found for virtually all loci between all possible species pairs. Nicklas & Hoffman (1979) found fixed differences at 4 of 7 loci in two morphologically distinct sympatric species of *Glycera* (Glyceridae). Britton-Davidian & Amoureux (1982) found fixed differences at 4 of 9 loci studied and demonstrated that two forms of *Ophelia* (Opheliidae) represent distinct species; morphologically intermediate worms were shown not to be hybrids (as had previously been suggested), but clearly belonged to one or other of the two species. Ekaratne *et al.* (1982) and Crisp & Ekaratne (1984) confirmed the presence of two sympatric sibling species of *Pomatoceros* (Serpulidae); fixed differences were found at 6 of 19 loci studied. The high level of genetic divergence between species in a wide range of families, most usefully shown by the presence of fixed differences at a large proportion of enzyme loci, is clearly a diagnostic criterion in polychaetes as it is in most other animal groups.

Methods

Collection and treatment of samples. Seven sampling localities were chosen in south-eastern Australia to encompass a wide range of habitats. Worms were collected by turning rocks and shallow digging. Such opportunistic collecting probably results in a bias towards larger size ranges, however more objective methods such as sieving sediment fragment many worms. Sample sites are given in the following list (numbers refer to localities shown in Fig.2). Collection dates are followed by a number in parentheses indicating the number of worms collected; samples used for electrophoresis were collected on dates marked with an asterisk.

1. Apollo Bay breakwater, Vic., 38°45'S 143°41'E. Lee shore of artificial breakwater; sediment highly heterogeneous with much quarried rock and some sand

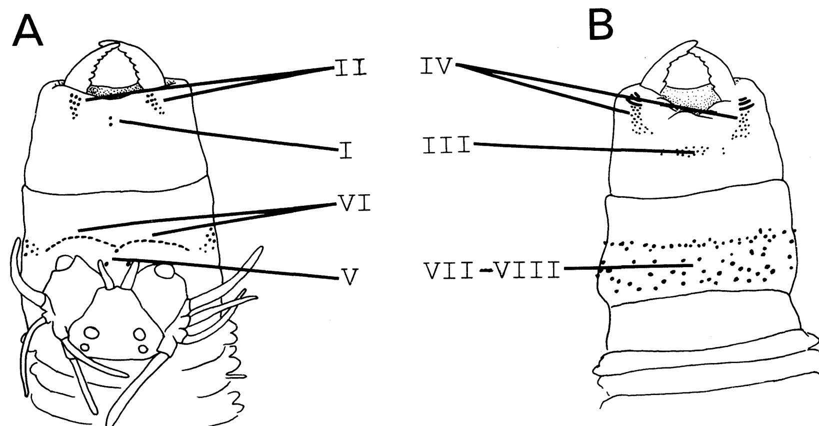

Fig.1. Arrangement of paragnaths in the *Perinereis nuntia* complex (A, dorsal view of extended proboscis; B, ventral view).

and mud fractions present; probably little fluctuation in salinity throughout the year. Dates: 2 Nov. 1984 (20); 30 Oct. 1985 (20); 23 Aug. 1986* (20); 30 Aug. 1987 (20).

2. Mouth of Separation Creek, Vic., 38°38'S 143°54'E. Boulder bed of exposed ocean beach; sediment well-sorted sand; high autumn rainfall and flow from the creek result in seasonal fluctuations in salinity, especially at low tides. Dates: 15 Nov. 1984 (11); 28 Oct. 1985 (20); 13 Apr. 1986* (20); 24 Aug. 1986 (20).

3. Werribee, Melbourne Water main drain, Vic., 37°58'S 144°54'E. Exposed locality in Port Phillip Bay; poorly sorted calcareous sediment with some rocks surrounding mouth of sewage effluent drain; considerable fluctuation in flow results in seasonal variations in salinity and organic input (the locality was described in more detail by Poore & Kudenov, 1978). Dates: May 1985* (21); 18 Sept. 1985 (19); 20 Mar. 1986 (14); 2 June 1986* (20).

4. Flinders, south end of Kennon Cove, Vic., 38°29'S 145°01'E. Sheltered locality in Western Port; poorly sorted muddy sand with seagrass *Heterozostera tasmanica* and some rocks; some fluctuation in salinity due to a small freshwater spring. Dates: 3 Sept. 1984 (21); 7 Oct. 1985 (20); 23 Mar. 1986* (20); 11 June 1986* (20).

5. Quarantine Bay, NSW, 37°05'S 149°54'E. Exposed rocky ocean shoreline with some heterogeneous sediment under rocks; probably little seasonal fluctuation in salinity. Date: 17 July 1987* (2).

6. Batemans Bay, north shore of Clyde River under Princess Highway bridge, NSW, 35°43'S 150°11'E, 17 July 1987. Poorly sorted muddy sand with little rock; salinity probably nearly fully marine with some seasonal fluctuation. Date: 17 July 1987* (7).

7. Coal and Candle Creek, Hawkesbury River estuary, near Akuna Bay, NSW, 33°37'S 151°14'E. Sheltered locality; poorly sorted muddy sand with much rock; salinity probably nearly fully marine but fluctuates due to high rainfall at irregular intervals. Date: 19 Oct. 1986* (5).

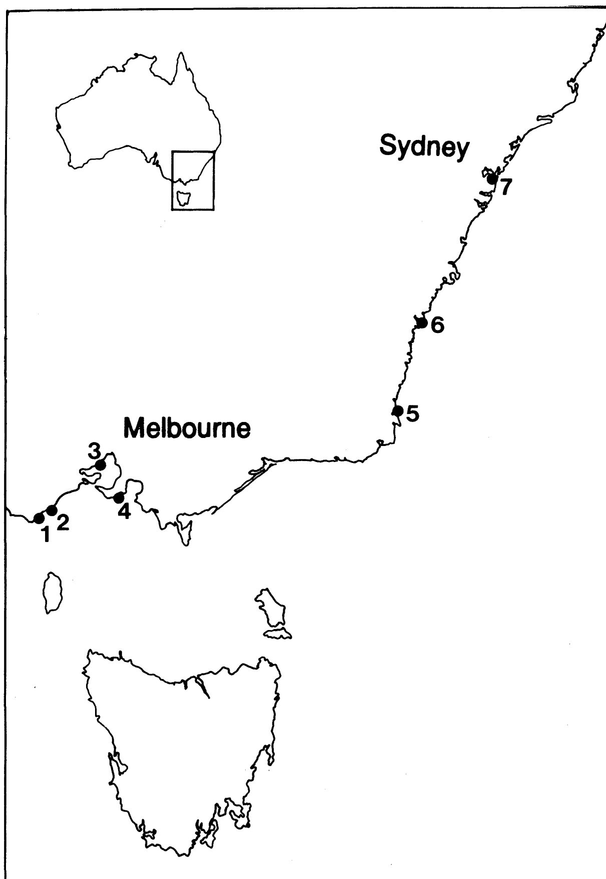

Fig.2. Sample sites referred to in the text.

Sampling strategy and numbers of specimens. Richardson *et al.* (1986) demonstrated that sample sizes of three to five specimens per population are sufficient to allow detection of different species in biochemical systematics. In this study, ten specimens were collected from each of populations 1 to 4, two specimens from population 5, seven from population 6, and four from population 7. Morphological analysis was based on four replicate collections over two years of about 20 specimens from each of sites 1 to 4 (see above). Approximately 80 worms per site were thus analysed; this is of the same order as the sample sizes used by Barnes & Head (1977).

Electrophoretic methods. Worms were kept alive in clean sediment or algae and seawater until returned to the laboratory. In the laboratory individual worms were kept in separate jars of seawater at 16°C for one to four weeks before being used for electrophoresis. Within this period few worms died. A tissue sample comprising two to ten posterior segments was severed from a live worm. The worm was returned to seawater in an individually labelled jar and kept alive to provide further fresh tissue samples. At the conclusion of electrophoretic procedures the remainder of the worm (including the taxonomically crucial proboscis) was preserved in formalin for morphological analysis. The tissue sample was homogenised and run immediately with two to three times its own volume of 0.2 mM Cleland's reagent in distilled water and centrifuged. Electrophoresis was carried out at 4°C using supernatant solutions applied to cellulose acetate supporting gels (Cellogel, Chematron, Milan). Details of buffers, running conditions and stain formulations are provided in Table 1.

Morphological methods. Worms used only for morphological analysis were carried back alive to the laboratory where they were briefly dipped in 95% ethanol. This causes worms to die instantaneously with the proboscis everted (the same method was used by Barnes & Head, 1977), after which they were fixed in 4% formalin and later transferred to 70% ethanol.

A size measure of each worm was obtained by dissecting the proboscis at the base of the right jaw (or the left jaw if the right was damaged) to reveal the full length of the jaw which was measured to the nearest

0.5 mm with an eyepiece micrometer on a stereo microscope. Body size and jaw length in nereidids are linearly related (Olive, 1980; Glasby, 1986) and jaw length measurements have the advantages of being unaffected by preservation technique and can be recorded from broken specimens. Paragnaths in each of the standard "groups" of the proboscis were counted, with left and right side counts distinguished for symmetrical groups. Several distinctive bar-shaped paragnaths were present on group IV in addition to conical paragnaths (Fig.1b); these were recorded separately.

Results

1. Allelic variation. The electrophoretic data (Table 2) were collected to test the null hypothesis "that all 7 populations represent a single species". Two loci, AK and LDH, were monomorphic in all populations. Four loci (AAT, GPI, IDH and MDH) showed fixed differences for alternative alleles at each locus, thus distinguishing populations 6 and 7 from populations 1 to 5. Two further enzyme systems, HK and PGM, stained for multiple bands and showed uninterpretable variation, probably the result of genetic polymorphisms. For both HK and PGM there was evidence of unique alleles in populations 6 and 7, but in the absence of genetic interpretations these data were not utilised further. In terms of genetic distance measures these data are equivalent to Nei's D of 0.69 between populations 1 to 5 and 6 and 7. By the standards of Richardson *et al.* (1986), Nei's D of 0.69 and the existence of fixed differences at 50% of loci is overwhelming evidence for the rejection of the null hypothesis. Two species are present, one represented by populations 6 and 7, the other by populations 1 to 5.

Identification of the two species depends on examination of type material of *Perinereis nuntia* and related taxa. Meaningful reassessment of the type material in turn depends on a better understanding of the nature and significance of inter- and intraspecific variation, which is the goal of this paper. Thus, for the remainder of this paper the northern species (from localities 6 and 7) will be referred to as *Perinereis* sp. A and the southern species (from localities 1-5) as *Perinereis* sp. B. Formal description of these species, with a revision of the *Perinereis nuntia* species-group, is reported separately (Wilson & Glasby, 1993).

2. Intraspecific morphological variability in *Perinereis* sp. B. Three factors which potentially contribute to intraspecific morphological variation in *Perinereis* sp. B were investigated, using paragnath counts and jaw length measurements: size-dependent variation, differences between populations and differences between subspecific morphs. Paragnath data from *Perinereis* sp. B were also used to investigate two additional questions of taxonomic interest: i) are paragnath counts from different groups on the proboscis correlated; ii) are some paragnath counts more variable than others. Sample sizes for *Perinereis* sp. A were too small to analyse intraspecific variation in this species.

Only 16 of 65 possible correlations between paragnath number and jaw length are significant at the 0.01 level and even the highest values generated coefficients of determination (r) values of about 0.2 (Table 3). That is, only about 20% (at most) of the variation in paragnath number can be accounted for by variation in size. A scatter plot of total paragnath number versus jaw length indicates the weak relationship between these quantities and the large amount of variation that is independent of size (Fig.3). Since paragnath numbers in all but group VI are free to increase in two dimensions with growth of the worm, a significant correlation might be expected between paragnath number and the square of jaw length. However, there was no such correlation. There seems to be no simple relationship between size and paragnath number in *Perinereis* sp B.

Numbers of paragnaths in each group are significantly different in about half of all 84 possible pairwise comparisons between population samples (Table 4). The most similar pairs of population samples (in terms of mean paragnath counts) are Apollo Bay and Separation Creek (column A, Table 4), and Werribee and Flinders (column F, Table 4). Canonical variates analysis of the same data (Fig.5) shows some separation of the sampled populations of *Perinereis* sp. B on the first two discriminant functions but does not produce discrete clusters as seen in the interspecific comparison in Figure 4.

Two methods of multivariate analysis were used to look for possible morphs or varieties in *Perinereis* sp. B. Canonical variates analysis was used to determine if differences existed between *a priori* designated groups, and principal components analysis was used as an exploratory technique for investigating intra-specific variation where there is no *a priori* basis for designating groups. Paragnath counts for group V on the proboscis have previously been used as the basis for recognising subspecific varieties of *P. nuntia*. The validity of the named varieties was investigated by assigning specimens from populations 1 to 4 to groups on the basis of 1, 2 or 3 paragnaths present in group V, and canonical variates analysis was performed using the remaining paragnath group counts. This procedure was followed both for the pooled data from populations 1 to 4 and for each population separately. These analyses failed to produce any separation of the designated groups. In all analyses, all specimens plotted in a single cluster and no separation can be seen between the notional sub-specific varieties. Principal components analysis of paragnath data also failed to produce any clusters or partial separation of specimens.

The idea that paragnath counts for the different groups on the proboscis might be strongly correlated is relevant because, if so, exhaustive paragnath counts would include some redundant data. Pearson correlation coefficients were calculated between all paragnath groups for all four populations of *Perinereis* sp. B. Coefficients of determination (r) values calculated from these are

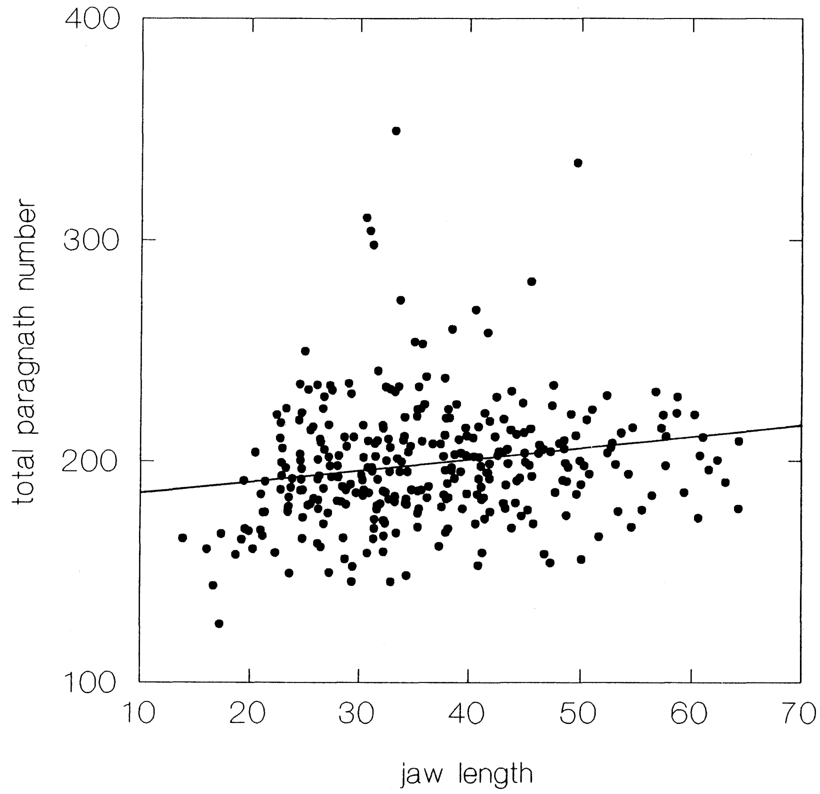

Fig.3. Scatter plot of total paragnath number versus jaw length, *Perinereis* sp. B.

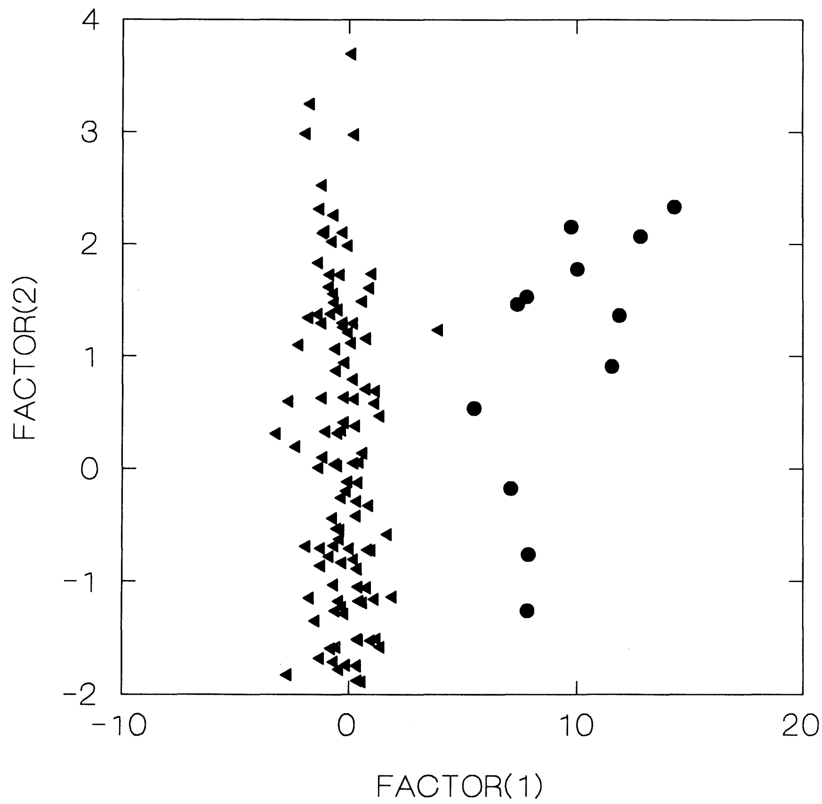

Fig.4. Canonical variates analysis of *Perinereis* spp. A and B based on paragnath counts for groups I, IIL, IIR, III, IVLC, IVLB, IVRC, IVRB, V, VIL, VIR, VII-VIII (circles = sp. A, triangles = sp. B).

low; even for paired symmetrical groups (II, IV, and VI), for which correlation coefficients are significant at p less than 0.01, coefficients of determination are in the range 0.11 to 0.58. That is, only 11% to 58% of the variability in one paragnath group can be accounted for by the opposite pair.

The possibility that there are differences in variability between different paragnath groups is of interest both in terms of taxonomic utility (characters showing little variability are likely to be more useful in taxonomy) and for more fundamental biological reasons (differences in variability between characters might reflect different selective pressures operating or differences in genetic control of expression of the characters). Relative variability of characters is measured by the coefficient of variation (Sokal & Rohlf, 1981); these data are provided for *Perinereis* sp. B in Table 5. Sokal & Brauman (1980) provide a test for comparing coefficients of variation of two dependent (correlated) variables based on the t-distribution. By this method there are no significant differences in the variability of paragnath groups II, III, IV (cones) and VII-VIII within any of the study populations. (Paragnath groups I, IV (bars), V and VI differ in their probabilities of a zero measurement or have different dimensions. Lande (1977) showed that it is not valid to compare coefficients of variation of such characters and these were therefore excluded from inter-character comparisons.) Another possibility is that differences in the variability of the same character might be evident between different populations, but by Sokal & Brauman's (1980) test there are no significant differences in variability of paragnath counts between any of the 4 Victorian populations of *Perinereis* sp. B.

3. Interspecific differences between *Perinereis* spp. A and B. The paragnath data show both quantitative and qualitative differences between the two species. Canonical variates analysis of the same data (Fig.4) differentiate the two species clearly: *Perinereis* sp. A is represented by a cluster distinct from *Perinereis* sp. B. In contrast, although there is some separation of populations 1 to 4 of *Perinereis* sp. B on the first two discriminant functions, there are no discrete clusters. (The two specimens representing population 5 of *Perinereis* sp. B, are morphologically and geographically intermediate between the other populations of *Perinereis* spp. A and B, however the electrophoretic data unequivocally identify these specimens as *Perinereis* sp. B.) Canonical loadings from the canonical variates analysis highlight those dependent canonical factors (paragnath groups) which best separate the populations (Table 6). Oral ring paragnath counts (groups V, VI and VII-VIII) contribute most to factor 1, which separates the two species. In contrast, maxillary ring paragnath counts (groups I, II, III and IV) load more highly on factor 2. Some separation of populations within species occurred along factor 2, although this information could not legibly be presented in Figure 4, which shows the interspecific comparison. Frequency histograms of those

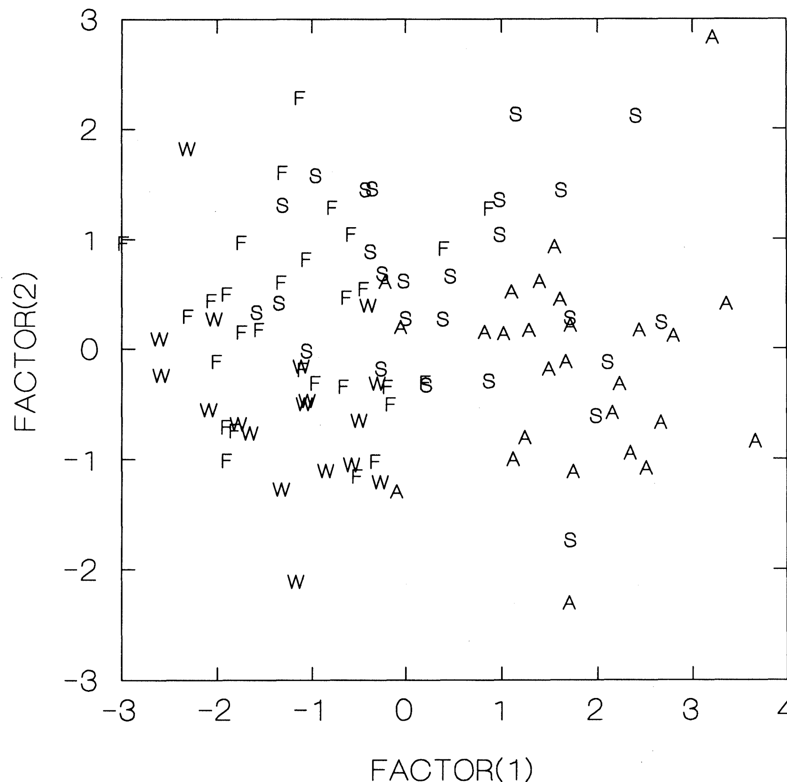

Fig.5. Canonical variates analysis of samples from four localities for *Perinereis* sp. B based on paragnath counts for groups I, IIL, IIR, III, IVLC, IVLB, IVRC, IVRB, V, VIL, VIR, VII-VIII (A = Apollo Bay, S = Separation Creek, W = Werribee, F = Flinders).

paragnath groups identified by the canonical variates analysis, Areas II, VI and VII-VIII, show little overlap between the two species (Fig.6). No intraspecific comparisons of populations show differences of this order.

Finally, re-examination of the specimens revealed a qualitative difference that distinguishes the two species. *Perinereis* sp. A specimens carry an additional band of minute conical paragnaths ventrally on Area VII-VIII. This band is posterior to, and in addition to,

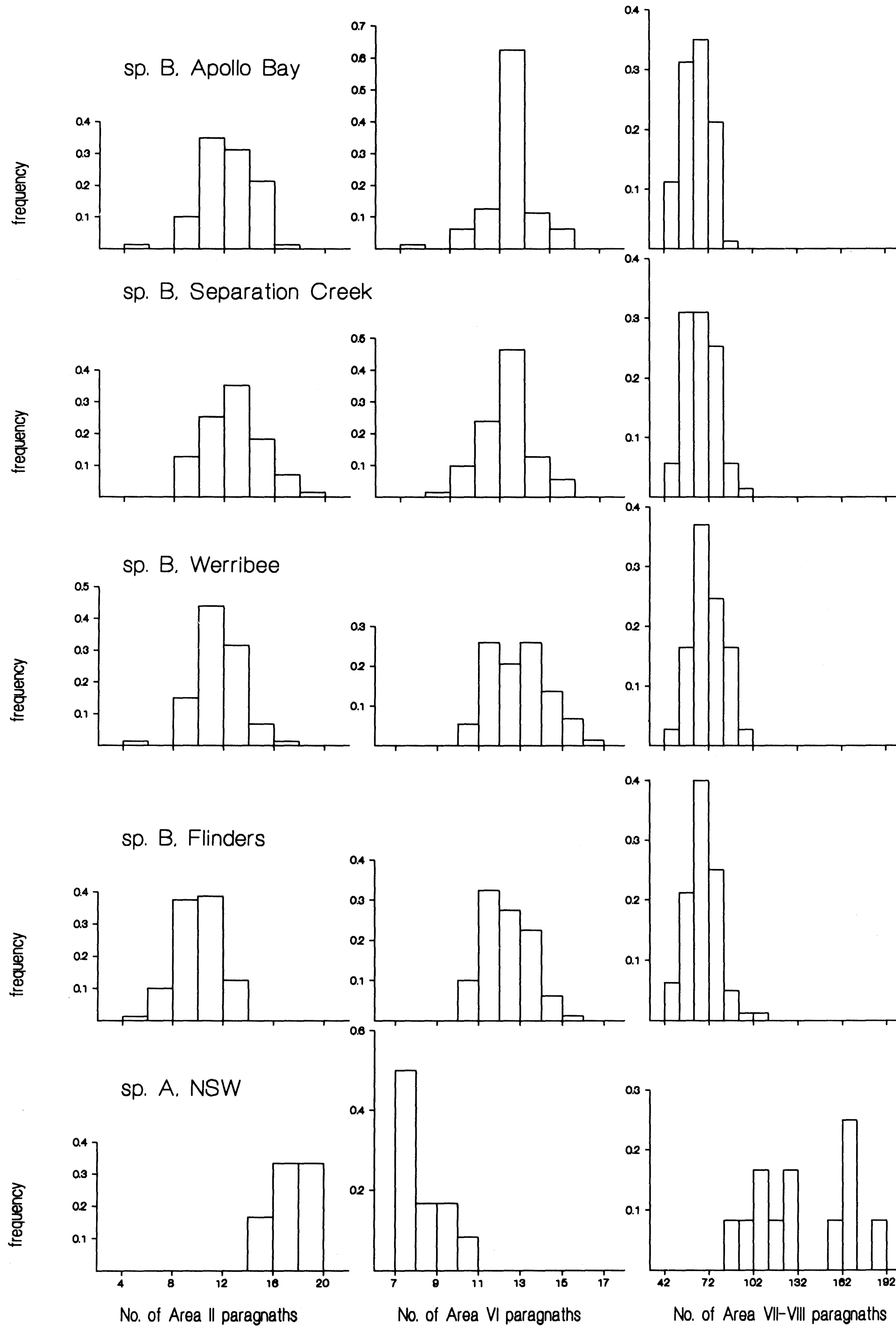

Fig.6. Frequency histograms of paragnath counts for selected groups, *Perinereis* spp. A and B.

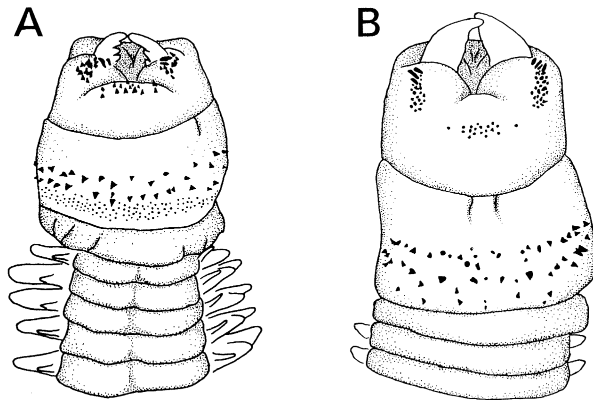

Fig.7. Ventral view of extended proboscis, *Perinereis* spp. A and B.

the irregular double row of larger conical paragnaths that is present in both species (Fig.7).

Discussion

Previous treatments of *Perinereis nuntia* and related taxa suffer from the basic handicap that the level of intra-specific morphological variability was unknown, as was the true taxonomic significance of supposedly diagnostic characters. In this study I have relied on genetic differences between species (as determined by electrophoresis of enzymes) to establish species boundaries independently. Analysis of variation in paragnaths from the same populations shows that differences between populations contribute more to intraspecific variation than differences due to size. Canonical variates analysis of paragnath data separates species convincingly and efficiently, and allows the characters that account for most of the separation of species to be quickly identified. The two species studied can be distinguished on the basis of having non-overlapping ranges of paragnath counts and from qualitative differences in the type of paragnaths present. There is no evidence for recognition of any subspecific morphs or varieties in *Perinereis* sp. B and lesser differences (though they may still be statistically significant) can be attributed to intraspecific variation between populations.

Detailed analysis of variation and correlation in paragnath number has been carried out in a study of another species of nereidid, *Nereis diversicolor*, by Barnes & Head (1977). The statistical technique used by Barnes & Head to represent the pattern of correlations between different paragnath groups was to draw a dendrogram based on unweighted pair group averaging of the correlation coefficient matrix. This method could conceivably be taxonomically useful, however the low r values (in the range 0.11 to 0.58 for paired symmetrical groups) for *Perinereis* sp. B preclude the use of this technique. Comparable values for r from the data of Barnes & Head (1977) on *Nereis diversicolor* are in the range 0.64 to 0.74. Dendrograms of paragnath count correlation coefficients are unlikely to be taxonomically useful in nereidid taxa with levels of intraspecific variability similar to *Perinereis* sp. B.

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APPENDIX

Table 1. Enzymes examined, buffers and running conditions.

Enzyme (abbreviation; EC no.)	Running buffer	Running conditions
Adelnylate kinase (AK; 2.7.4.3)	TEB	30 m @ 300V
Aspartate amino transferase (AAT; 2.6.1.1)	TM	40 m @ 1.1mA/cm
Glucosephosphate isomerase (GPI; 5.3.1.9)	TEB	30 m @ 300V
Hexokinase (HK; 2.7.1.1)	TEB	30 m @ 300V
Isocitrate dehydrogenase (IDH; 1.1.1.42)	TM	60 m @ 1.1mA/cm
Lactate dehydrogenase (LDH; 1.1.1.27)	TM	60 m @ 1.1mA/cm
Malate dehydrogenase (MDH; 1.1.1.37)	TM	60 m @ 1.1mA/cm
Phosphoglucomutase (PGM; 2.7.5.1)	TEB	30 m @ 300V

TEB = Tris-EDTA-borate buffer pH 8.7 after Shaklee *et al.*, 1973; TM = Tris-maleate buffer pH 7.8 after Richardson *et al.*, 1980; all stain formulations after Richardson *et al.*, 1980, except HK which is after Richardson, 1983.

Table 2 Frequency of 11 alleles in samples from seven populations of the *Perinereis nuntia* complex.

Locus:	allele	Locality sample numbers						
		1	2	3	4	5	6	7
AAT:	133	–	–	–	0.03	–	–	–
	123	–	–	–	–	–	1.0	1.0
	100	1.0	1.0	1.0	0.97	1.0	–	–
AK:	100	1.0	1.0	1.0	1.0	1.0	1.0	1.0
GPI:	100	1.0	1.0	1.0	1.0	1.0	–	–
	75	–	–	–	–	–	1.0	1.0
IDH:	111	–	–	–	–	–	1.0	1.0
	100	1.0	1.0	1.0	1.0	1.0	–	–
LDH:	100	1.0	1.0	1.0	1.0	1.0	1.0	1.0
MDH:	123	–	–	–	–	–	1.0	1.0
	100	1.0	1.0	1.0	1.0	1.0	–	–

Sample sizes for each locality: 1-3, n = 10; 4, n = 30; 5, n = 2; 6, n = 4; 7, n = 3.

Table 3. Pearson product-moment correlation coefficients for numbers of paragnaths in each group on jaw length for samples from four Victorian populations of *Perinereis* sp. b.

	Apollo Bay	Separation Creek	Werribee	Flinders	all populations
I	-0.08	0.28	-0.07	-0.04	-0.01
IIL	-0.15	0.04	0.05	-0.09	-0.06
IIR	0.05	0.09	0.03	-0.17	-0.03
III	0.02	0.44**	0.27	0.20	0.12
IVLC	0.08	0.44**	0.17	0.46**	0.15
IVLB	0.12	0.31*	-0.03	0.04	0.06
IVRC	-0.13	0.29	0.24	0.42**	0.05
IVRB	0.00	0.34**	0.07	0.06	0.07
V	0.25	0.10	0.06	0.06	0.14
VIL	0.03	0.24	0.43**	0.06	0.21**
VIR	-0.03	0.33*	0.29	0.27	0.24**
VII-VIII	0.05	0.35*	0.16	0.34*	0.26**
Total Paragnaths	0.00	0.43**	0.28	0.43**	0.22**

2-tailed probabilities: * = $p < 0.01$; ** = $p < 0.001$

Table 4. Significance tests for means and variances of paragnath groups for pairwise comparisons of samples of four populations of *Perinereis* sp b.

	Means						Variances					
	A	B	C	D	E	F	A	B	C	D	E	F
I	+	+	+		+	+						+
IIL			+	+	+	+						+
IIR			+	+	+	+						+
III	+	+	+	+	+			+	+			
IVLC	+	+	+	+	+					+		
IVLB					+	+				+		
IVLT	+	+	+	+	+					+		
IVRC		+	+	+	+			+	+	+	+	+
IVRB	+			+	+		+					
IVRT		+	+	+	+			+	+	+	+	
V	+			+						+		
VIL												
VIR												
VII-VIII		+		+								

Means tested by Mann-Whitney U test (t test returned almost identical results at a higher level of significance); variances by F test. Significance level: + = $p < 0.01$. Codes for column headings: A = Apollo Bay versus Separation Creek; B = Apollo Bay versus Werribee; C = Apollo Bay versus Flinders; D = Separation Creek versus Werribee; E = Separation Creek versus Flinders; F = Werribee versus Flinders.

Table 5. Coefficients of variation (V) for numbers of paragnaths in each group for samples from four populations of *Perinereis* sp. B (V = s/Y where s is the sample standard deviation and Y is the sample mean; Sokal & Brauman, 1980).

	Apollo Bay	Separation Creek	Werribee	Flinders	all populations
I	29.63	34.15	43.04	36.29	39.07
IIL	20.54	19.84	16.93	20.36	21.55
IIR	18.09	18.09	19.76	20.05	20.43
III	18.56	17.18	15.38	16.88	20.05
IVLC	12.33	17.38	13.03	17.07	19.14
IVLB	22.63	16.88	28.32	22.24	22.95
IVLT	12.04	16.13	12.03	15.70	17.94
IVRC	19.69	19.29	11.88	16.56	21.73
IVRB	28.82	17.58	23.80	24.53	24.28
IVRT	19.11	18.17	11.29	15.14	20.42
V	34.19	48.57	26.50	37.85	37.38
VIL	11.60	11.97	11.92	11.35	11.79
VIR	10.47	11.74	12.66	10.96	11.59
VII-VIII	15.30	16.24	14.30	15.73	15.75

Table 6. Canonical loadings (correlations between conditional dependent variables and dependent canonical factors), *Perinereis* spp. A & B, samples 1 to 7.

Paragnath groups	Canonical factors	
	1	2
I	-0.150	**0.349**
IIL	-0.244	**0.399**
IIR	-0.261	**0.398**
III	-0.003	**0.534**
IVLC	0.035	**0.727**
IVRC	0.024	**0.633**
V	-0.064	0.022
VIL	**0.300**	-0.108
VIR	**0.283**	-0.128
VIIVIII	**-0.640**	0.076

(figures in bold are discussed in the text)