

## BENTHIC NEMATODES FROM THE PACIFIC COAST WITH SPECIAL REFERENCE TO THE CYATHOLAIMIDS

by

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### ABSTRACT

Over thirty genera of free-living nematodes from seventeen families have been found during a survey of the benthic meiofauna of the Pacific Coast near Vancouver. Their distribution appears to be influenced by the quantity of organic matter and associated bacteria in the sediment. The taxonomy of the Cyatholaimidae will be examined, especially those members of the Cyatholaiminae. The use of light and scanning electron microscope observations of pores and punctuations as taxonomic aids within this family will be discussed.

### INTRODUCTION

We report on part of a study of the free-living nematode fauna of coastal British Columbia. Until now, the only comprehensive studies on the free-living marine nematodes of the west coast of North America are those from : Puget Sound, Washington (WIESER, 1959), Oregon (MURPHY, 1961), Northern California (CHITWOOD, 1960), and Southern California and Central America (ALLGÉN, 1947, 1951). Only two reports are available for the Canadian Pacific coast, namely, the redescription of *Oncholaimus vesicarius* (NELSON et al., 1971) and the description of a new species, *Enoplus anisospiculus* (NELSON et al., 1972). Since no comprehensive studies are available for the West Coast of Canada, a primary purpose of our study is to investigate the nematode fauna of this area.

### HABITAT DESCRIPTION

Nematodes were collected from three different sites in Greater Vancouver (Fig. 1), namely :

(i) Iona Island : a tidal flat with a gently sloping beach in the estuary of the Fraser River, south of Vancouver, with jetties on either side of it that minimize the influence of fresh water.

(ii) Stanley Park : a sandy beach at the mouth of Burrard Inlet which contains the dock facilities of a major port. Its slope is somewhat steeper than the beach at Iona Island and it is heavily used for recreational and educational purposes.

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(iii) Belcarra Park : a small steeply sloping beach close to a bed of eel grass (*Zostera* sp.) at the mouth of a deep water fjord.

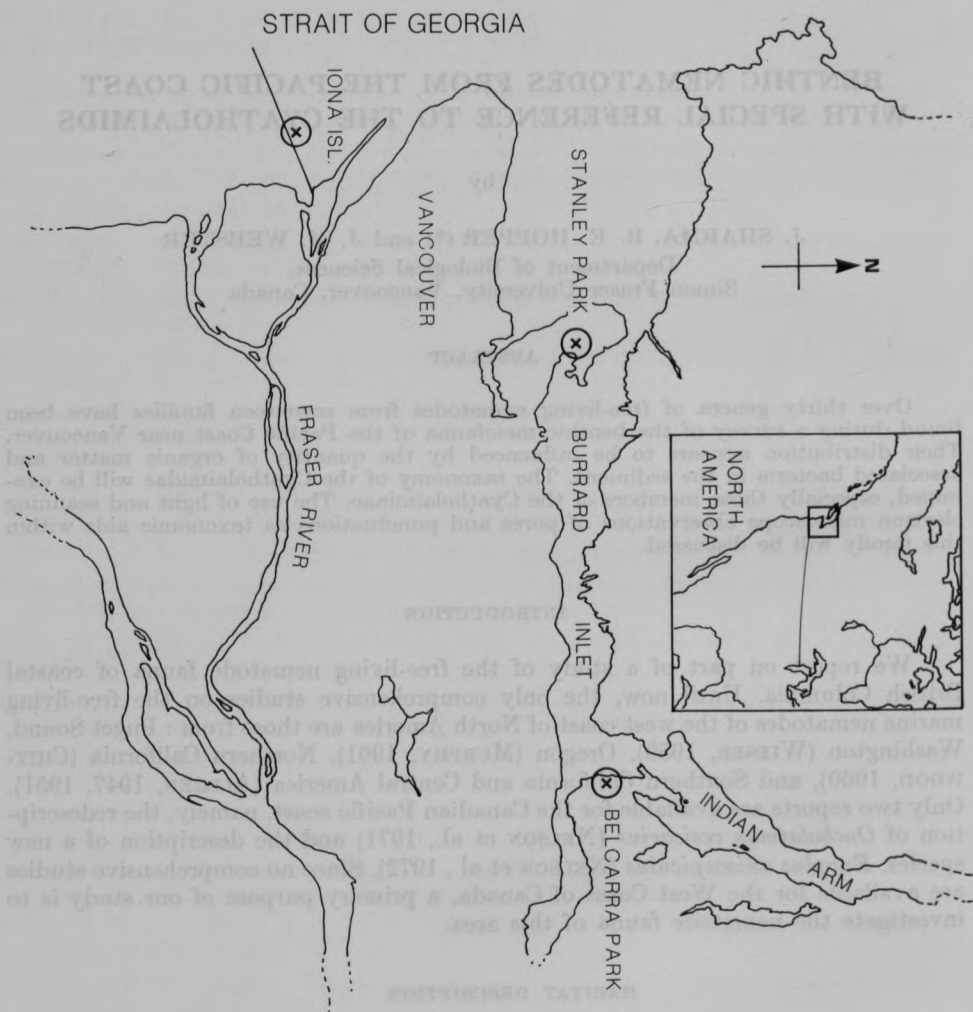


Fig. 1. — Map of Greater Vancouver, British Columbia showing the location of three nematode collection sites.

The sediment at Iona Island is well sorted (as determined by dry sieving) with most of it falling within a narrow range of grain sizes. Both Stanley Park and Belcarra Park sites, however, have poorly sorted sediments with a wide range of grain sizes.

A quantitative estimate of the detrital material in the beach sediments was obtained by measuring with a CHN analyzer the per cent of carbon present. It was lowest in the Iona Island sediments (0.52 %) and highest in the poorly sorted sediments of Belcarra Park (2.35 %).

The degree of bacterial activity in the sediments was assessed by determining the heterotrophic activity of the bacteria. This was done by measuring the amount of radiolabelled glucose the bacteria take up. It was lowest in the Iona Island sediments ( $1.6 \times 10^4$  DPM) and highest in Belcarra Park sediments ( $1.2 \times 10^5$  DPM). Hence, preliminary evidence suggests that there is higher bacterial activity in the more organic sediments at the Belcarra Park site than in the well washed, relatively clean Iona Island site.

#### ECOLOGICAL INVESTIGATIONS

Sediment samples were taken at about mid-tide level at each of the three sites by a hand-held plexiglass corer. The upper 6 cm of the core were examined in 2 cm sections and nematodes were extracted from each section by sieving and centrifugation.

Nematodes representing 20 families and 40 genera (Table I) were recovered from the sediments. Some families that were well represented at Belcarra Park and Stanley Park were absent from all samples from Iona Island. The total number of nematodes in the samples at Iona Island is much lower than that at either of the other two sites. Preliminary observations suggest that the nematodes differ in their distribution at the three sites.

#### TAXONOMIC CONSIDERATIONS

A prominent group of nematodes found in this study is the order Chromadorida which is represented by species from all its families, namely, Comesomatidae, Cyatholaimidae, Choanolaimidae, Chromadoridae, Desmodoridae. The Cyatholaimids are represented by three *Marylynnia* species two of which were collected intertidally and one subtidally. Although on initial observation they all appeared very similar in having a prominent dorsal tooth and multispiral amphid, they differ in some features and especially in the pattern of cuticular pores.

The « pores » referred to here appear, under light microscopy, as circles of varying sizes and shapes amongst the punctations which appear as dots in several Chromadorids. Little attention has been given to the type and number of pores in relation to species identification. One reason for this lack of utilization in taxonomy could be the difficulty of distinguishing the different types at the normal resolution of the light microscope. The lack of complete data on the pores has minimized their inclusion in standard taxonomic descriptions. Hence, their value in taxonomy has not been fully realized. The scanning electron microscope is particularly useful in ascertaining the relative position and distribution of the pores and also their association with the cuticular surface.

Since the earliest record of pores by BASTIAN in 1865, several taxonomic papers have recorded the presence of pores (see Table II). BASTIAN (1865) recorded their presence in various nematode species and the differences he noted suggest that the number of pores varies with species. CHITWOOD and CHITWOOD (1950) described pores, in both the Enoplida and Chromadorida, as the openings of two sublateral rows of unicellular glands situated in the lateral chords. They also noted that these glands occur only in the Aphasmidea (= Adenophorea). Such pores are not confined to the Cyatholaimidae but have been described also from the Eurytominidae (HOPPER, 1970) and the Desmodoridae (HOPPER and CEFALU, 1970). SCHUURMANS,

TABLE I

*Nematode genera present at the three collection sites  
in the Greater Vancouver Area*

		Iona Island	Stanley Park	Belcarra Park
Araeolaimida				
Leptolaimidae	<i>Leptolaimus</i>	—	+	+
Axonolaimidae	<i>Araeolaimus</i>	—	+	+
	<i>Axonolaimus</i>	—	+	+
	<i>Odontophora</i>	—	+	+
	<i>Parascolaimus</i>	+	+	+
Monhysterida				
Siphonolaimidae	<i>Siphonolaimus</i>	—	+	+
Linhomoeidae	<i>Desmolaimus</i>	—	+	+
	<i>Metalinhomoeus</i>	—	—	+
	<i>Tershellinia</i>	+	+	+
	<i>Eleutherolaimus</i>	—	+	+
	<i>Didelta</i>	—	+	+
	<i>Linhomoeus</i>	—	+	+
Monhysteridae	<i>Monhystera</i>	—	—	+
	<i>Steineria</i>	—	+	+
	<i>Theristus</i>	+	+	+
Sphaerolaimidae	<i>Sphaerolaimus</i>	—	—	+
Chromadorida				
Comesomatidae	<i>Sabatieria</i>	+	+	+
Chromadoridae	<i>Ethmolaimus</i>	—	—	+
	<i>Chromadorina</i>	—	+	+
	<i>Euchromadora</i>	—	—	+
	<i>Neochromadora</i>	+	+	+
	<i>Spilophorella</i>	—	—	+
Cyatholaimidae	<i>Paracanthonus</i>	—	—	+
	<i>Cyatholaimus</i>	—	—	+
	<i>Marylynia</i>	+	+	+
Choanolaimidae	<i>Choanolaimus</i>	—	—	+
Enoplida				
Tripyloididae	<i>Bathylaimus</i>	—	+	+
	<i>Paratripyloides</i>	—	—	+
	<i>Tripyloides</i>	+	+	+
Oxystominidae	<i>Oxystomina</i>	—	—	+
Lauratonematidae	<i>Lauratonema</i>	—	—	+
Anticomidae	<i>Anticoma</i>	—	—	+
Enoplidae	<i>Oxyonchus</i>	+	+	+
	<i>Mesacanthion</i>	—	—	+
Oncholaimidae	<i>Oncholaimus</i>	+	+	+
Encheliidae	<i>Eurystomina</i>	—	—	+
	<i>Symplocostoma</i>	—	—	+

STEKHOVEN and TEUNISSEN (1938) recorded the distribution and number of pores in *Plectus granulosus*. However, the Cyatholaimidae is the only family of marine nematodes in which hypodermal pores have been consistently recorded and in which their structure has been the subject of investigation.

TABLE II

*A representative chronological list of records  
that describe the presence of pores in free-living marine nematodes*

1. 1865, BASTIAN	fine capillary canals ; lateral integumental pores
2. 1890, DE MAN	round pores
3. 1898, COBB	longitudinal rows of circles in the lateral regions
4. 1917, COBB	longitudinal series of repetitive organs in lateral fields
5. 1918, FILIPJEV	row of circular pores along lateral lines
6. 1933, DE CONINCK and SCHUURMANS STEKHOVEN	one or more rows of finer pointlets in dorsal and ventral fields
7. 1950, CHITWOOD and CHITWOOD	lateral hypodermal glands
8. 1954, WIESER	conspicuous pores
9. 1967, WIESER and HOPPER	longitudinal series of hypodermal pores
10. 1970, VITIELLO	pore complexes described in five genera
11. 1971, WARWICK	two types of lateral pores ; start anterior to excretory pores and extend 2/3 of esophageal length
12. 1972, HOPPER	longitudinal rows pore complexes
13. 1972, HOPPER	pore complexes in <i>Marylynnia</i> sp. and <i>Longicyatholaimus</i> sp. distinguished
14. 1972, HOPPER and CEFALU	longitudinal rows of pore complexes

INGLIS (1963) was the first person to describe cuticular pore structures in detail and he compared them to the «campaniform-type» organs which act as proprioceptors in insects. He described two types of pores : an ovoid type with a transverse sheet of cuticle (type-1) and a circular type with a raised central dome (type-2). Since then the two types of pores have been referred to as hypodermal pores and lateral modified punctations (LMP's) respectively by several authors. Hypodermal pores, which appear as pores with a transverse slit, occur in up to 12 longitudinal rows around the circumference of the body. The most prominent are the two sublateral rows situated in the region of the lateral chords. Normally, the hypodermal pores in these rows are spaced uniformly throughout the body length. There are fewer longitudinal rows of LMP's, each of which appear as pores placed between two punctations, and generally, only a single row of LMP's are present in each lateral aspect. However, their distribution within this row is highly variable between species. WARWICK (1971) refers to these two types of pores as type-a and type-b, respectively.

Although the presence of the two types of pores has been confirmed by several authors, a different interpretation of pore structure resulted from the ultrastructure studies of WRIGHT and HOPE (1968). They describe the pore as a complex consisting of : a shallow depression in the outer cuticle, a slit-like pore extending through

the deeper cuticular layers into an underlying hypodermal cell, and a ring or collar-like development of dense material in the middle cuticle layer. They introduced the term « pore complex » for the type-1 pore described by INGLIS because several components of the cuticle contribute to the image of a circle with transverse bar that is seen on the cuticle surface with a light microscope.

The function of the pore complex is not clear. CHITWOOD and CHITWOOD (1950) used the term « lateral hypodermal glands » to describe the structures beneath these pores in the Enoplida and Chromadorida. DE MAN (1889) drew glands in some of his figures of *Paracanthonus ocellatus* BASTIAN and showed their relationship to pores. SCHUURMANS STEKHOVEN (1935) refers to pores in relation to « skin glands ». Nevertheless, only the term hypodermal has been retained by WRIGHT and HOPE (1968) because the pores are not restricted to a lateral position and their function as glandular openings has not been demonstrated.

Although various authors have noted the presence of hypodermal pore complexes and various names have been assigned to them, the possible taxonomic significance of their kinds and numbers has been ignored. In distinguishing the species of a Canaloid copepod, *Eucalanus*, FLEMINGER (1973) showed that the distribution pattern and number of integumental organs, which include sensilla and glandular pores, are of taxonomic significance. The distribution of hypodermal pore complexes and modified punctations may be equally important in the taxonomy of some nematode groups. The importance of such meristic or countable characters has been emphasized in taxonomy (MAYR, 1969), because in statistical usage, they are discrete variables, and the fact that they can only take on certain values makes them taxonomically more reliable.

The pore patterns for each of the three *Marylynnia* species in our collection are very distinctive (Fig. 2). The arrangement of the hypodermal pore complexes on the cuticle in each of the three species is constant among adults and juveniles and conforms with the general description given above. The LMP's show greater interspecific variation in their cuticular arrangement than do the hypodermal pores. However, the arrangement is consistent within a species.

*Marylynnia* sp. 1 has a cluster of LMP's behind the head extending for about 100  $\mu\text{m}$  with 25-35 units in the cluster. There is no discernible pattern within the cluster, although the placement of the LMP's is similar in all specimens examined. A row of 10-12 LMP's occurs in the preanal-caudal region of both sexes. The male lacks further LMP's but the female has short rows containing 3 to 4 pores located a short distance anterior and posterior to the vulvar region.

*Marylynnia* sp. 2 has a row of 15-20 anteriorly placed LMP's extending about one-third the esophageal length. 8-10 LMP's occur in the caudal region and none were found elsewhere on the body.

In *Marylynnia* sp. 3 the LMP's are not clustered at the anterior end. Instead, two rows of hypodermal pores occur laterally starting 46-73  $\mu\text{m}$  from the anterior end, while the LMP's start one-third of the way down the esophageal region and continue the length of the body.

All three species of *Marylynnia* had pores and modified punctations arranged in varying densities over all the body and therefore data was collected (Table III) so as to differentiate between the species on the basis of their pores. The following observations were made : (i) The distance behind the head of the first hypodermal pore was constant for a given species. (ii) The number of hypodermal pores and LMP's in the esophageal region was constant for a given species. (iii) The number

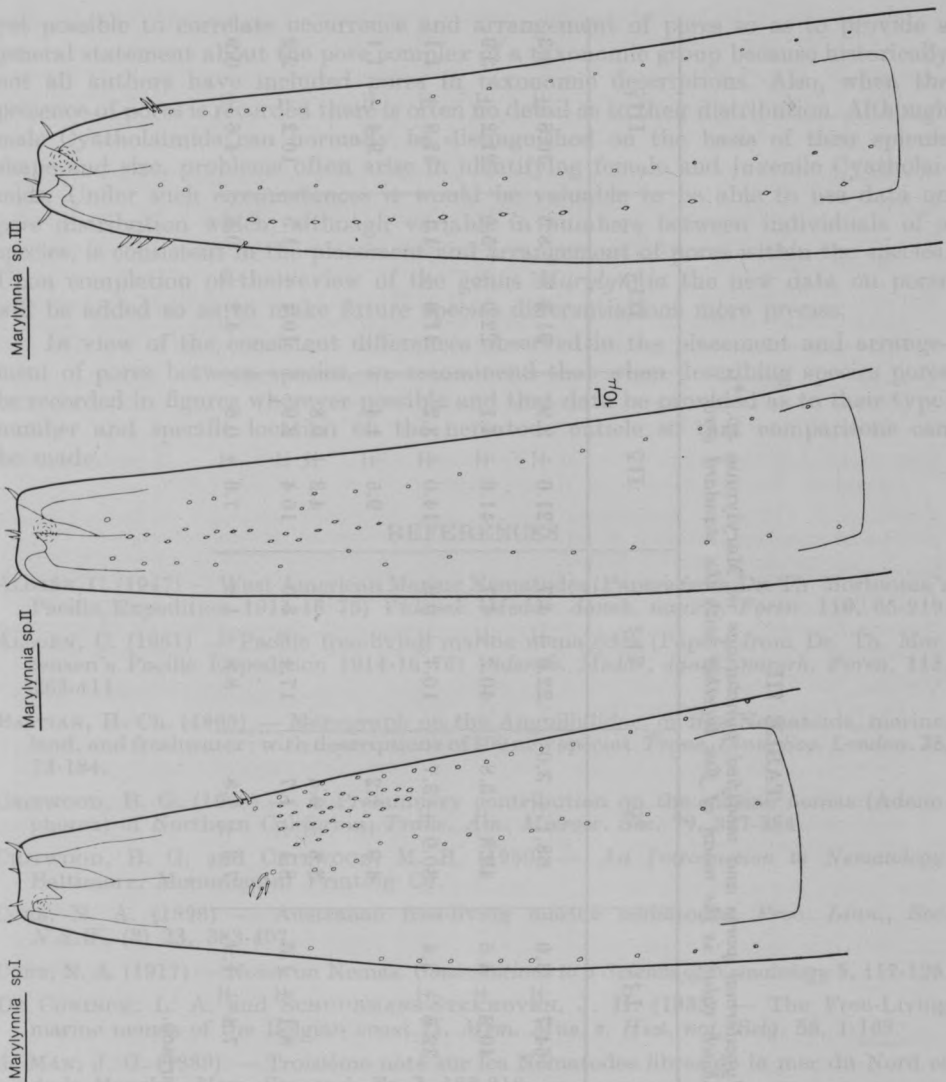


Fig. 2. — The pattern of two types of hypodermal pores in three *Marylynnia* sp. Lateral view.

of pores and LMP's in the tail region were constant for a given species but often difficult to ascertain because the tails of fixed nematodes were frequently curled. (iv) Counting all pores and LMP's over the body is very time consuming, hence the number of pores and LMP's in a row were counted (a) from the base of the esophagus to the anterior end, (b) 15.5  $\mu\text{m}$  on either side of the vulva and (c) on the tail, (v) Varying patterns of distribution may sometimes require other measurements to be made in order to differentiate between some species.

HOPPER (1972) defined *Marylynnia* sp. as having a greater number of hypodermal pore complexes and LMP's than *Longicyatholaimus* sp. However, it is not

TABLE III

*Hypodermal pores and modified punctations in Marylynnia sp. (\*)*  
 Each value is the mean of five together with the standard error

	I♂	I♀	II♂	II♀	III♂	III♀
Distance behind head of first pore (μm)	64.7 ± 3.6	68.1 ± 3.0	22.6 ± 4.7	21.6 ± 3.9	31.3 ± 8.3	30.8 ± 5.3
No. of hypodermal pores in esophageal region	49.8 ± 4.5	49.8 ± 5.8	40.2 ± 4.7	41.6 ± 4.1	22.0 ± 5.0	21.2 ± 3.9
No. of LMP's in esophageal region	32.6 ± 3.4	30.0 ± 3.7	10.4 ± 0.9	14.0 ± 3.2	11.6 ± 1.1	12.8 ± 1.1
No. of hypodermal pores in vulvar region		4.9 ± 1.2		9.5 ± 1.4		9.4 ± 1.1
No. of LMP's in vulvar region		3.8 ± 0.4		4.2 ± 0.8		
No. of hypodermal pores on tail	8.1 ± 1.5	8.1 ± 1.7	17.4 ± 3.0	16.4 ± 2.9	10.4 ± 1.5	10.2 ± 2.2
No. of LMP's on tail	7.4 ± 1.9	7.2 ± 1.8	8.8 ± 1.8	7.6 ± 0.6	4.2 ± 0.5	4.6 ± 0.9

(\*) As viewed from lateral surface.



yet possible to correlate occurrence and arrangement of pores so as to provide a general statement about the pore complex of a taxonomic group because historically not all authors have included pores in taxonomic descriptions. Also, when the presence of pores is recorded there is often no detail as to their distribution. Although male Cyatholaimids can normally be distinguished on the basis of their spicule shape and size, problems often arise in identifying female and juvenile Cyatholaimids. Under such circumstances it would be valuable to be able to use data on pore distribution which, although variable in numbers between individuals of a species, is consistent in the placement and arrangement of pores within the species. Upon completion of the review of the genus *Marylynnia* the new data on pores will be added so as to make future species differentiations more precise.

In view of the consistent differences observed in the placement and arrangement of pores between species, we recommend that when describing species pores be recorded in figures wherever possible and that data be provided as to their type, number and specific location on the nematode cuticle so that comparisons can be made.

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