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OCCURRENCE AND TAXONOMICAL SIGNIFICANCE OF ADDITIONAL CEPHALIC SETAE IN SOME LINHOMOEIDAE (NEMATODA)

by

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ABSTRACT

The cephalic sensory structures, papillae and setae, were studied in five species of *Paralinhomoeus* (Linhomoeidae) by light and scanning electron microscopy. A basic cephalic pattern, six labial papillae and ten cephalic setae, was characteristic of all young juveniles. Additional cephalic setae were observed in adults of four species, most frequently in males, giving rise to a variation within species or a sexual dimorphism. The wide-spread occurrence and great variation in number of additional cephalic setae have to be considered before using the number of cephalic setae as a taxonomical character within *Paralinhomoeus* and related genera.

On the basis of present knowledge the delimitation of some genera within Linhomoeinae is doubtful and the genera *Perilinhomoeus* and *Anticyclus* are to be synonymized with *Paralinhomoeus*.

I. INTRODUCTION

Our present classification of the free-living marine nematodes is based almost entirely on morphological characters. As the number of recorded species and genera increases, very minute structural differences are often used to delimit different taxa. However, characters differ in their usefulness for classification and also in their contribution to facilitate proper identification.

An important character in nematode taxonomy is the number and position of cephalic setae. The occurrence of additional setae in the cephalic circle in some linhomoeids and the observed variation in number of these setae, may, as described below, illustrate the difficulties involved in using this particular character within the Linhomoeidae.

Initially only a restricted number of specimens of two closely related species of *Paralinhomoeus*, *P. lepturus* (DE MAN, 1907) and *P. intermedius* (ALLGÉN, 1929), were available for examination. In both species the males were found to differ from juveniles and females regarding the number of cephalic setae. Since the original material had been collected in sediment with pronounced oxygen deficiency, the possibility could not be excluded that the observed variation reflected local populations living under strong environmental stress. Consequently, additional material from other geographical areas was obtained, which included a further three species.

Misinterpretation and failure to observe certain structures cannot be excluded when light microscopy is used to analyse the number and position of the minute cephalic structures. Therefore, in addition to common *en face* preparations, material was examined by scanning electron microscopy which has the advantages, besides increased resolution and depth of field, that the nematodes can be tilted and rotated during examination, allowing observation from the most suitable angles.

The comparative studies presented at the symposium in Ghent on structures other than the cephalic setae will be published separately.

II. MATERIAL AND METHODS

Specimens of five species of *Paralinhomoeus* (Linhomoeidae, Nematoda) were studied. Four of them are referred to species described earlier (Tab. 1). They were collected in the Gullmar Fjord on the west coast of Sweden (species nos. II, III, and V), and in the northern part of the Øresund in the vicinity of Helsingør, Denmark (species nos. I, III, and V). The fifth species was collected on the coast of Bermuda by Professor S. A. Gerlach. It is a new species and will be described elsewhere. The nomenclature used in this paper follows GERLACH & RIEMANN (1973).

TABLE I

Species and number of specimens studied by light microscopy (LM) and scanning electron microscopy (SEM)

	LM			SEM		
	juv	♀	♂	juv	♀	♂
I. *Paralinhomoeus ilenensis* (ALLGÉN, 1933)	2	5	5			
II. *P. lepturus* (DE MAN, 1907)	34	46	40	9	28	42
III. *P. intermedius* (ALLGÉN, 1929)	9	34	28	9	20	22
IV. *Paralinhomoeus n. sp.*	37	8	3	5	2	
V. *P. cf strandibrevicaudatus* (ALLGÉN, 1934)	5	4	4	6	9	12

Preparation for light microscopy (LM). Some of the living nematodes were killed and fixed in cold 4 % neutral formaldehyde in sea water. In order to make preparations for LM and SEM (see below) suitable for comparison a number of nematodes were fixed in ice-cold 4.5 % glutaraldehyde in a solution of 0.2 mol/l cacodylate buffer and filtered sea water (2 : 1). The fixed nematodes were slowly transferred to pure glycerol and mounted in anhydrous glycerol or in Kaiser's glycerol gelatine (*en face* preparations).

Preparation for scanning electron microscopy (SEM). The same fixatives used for LM were used for the primary fixation of the living nematodes. Each specimen, still in fixative, was then temporarily transferred to a slide and covered by a cover glass supported by glass rods. The identification of species and the examination

of sex and degree of maturity were performed by LM. After 1-3 days in the fixative, the specimens were washed in 0.2 mol/l cacodylate buffer and postfixed in 1 % osmium tetroxide in the same buffer for 1-2 h. at room temperature. After being washed in buffer, the specimens were dehydrated by passing them through a graded series of ethanol of increasing concentration to pure freon or by graded ethanol-acetone series, critical point-dried with CO_2 (COHEN, 1974), mounted on metal stubs, coated with gold in argon atmosphere, and examined in a Jeol JSM-U3 or a Jeol JSM-35 scanning electron microscope. The temporary study by LM before postfixation sometimes impaired the quality of the SEM preparations.

The number of specimens of each species and category (juveniles, females, males) studied by LM and SEM is given in Tab. 1. Only fully mature specimens are included in the categories « females » and « males ».

III. RESULTS

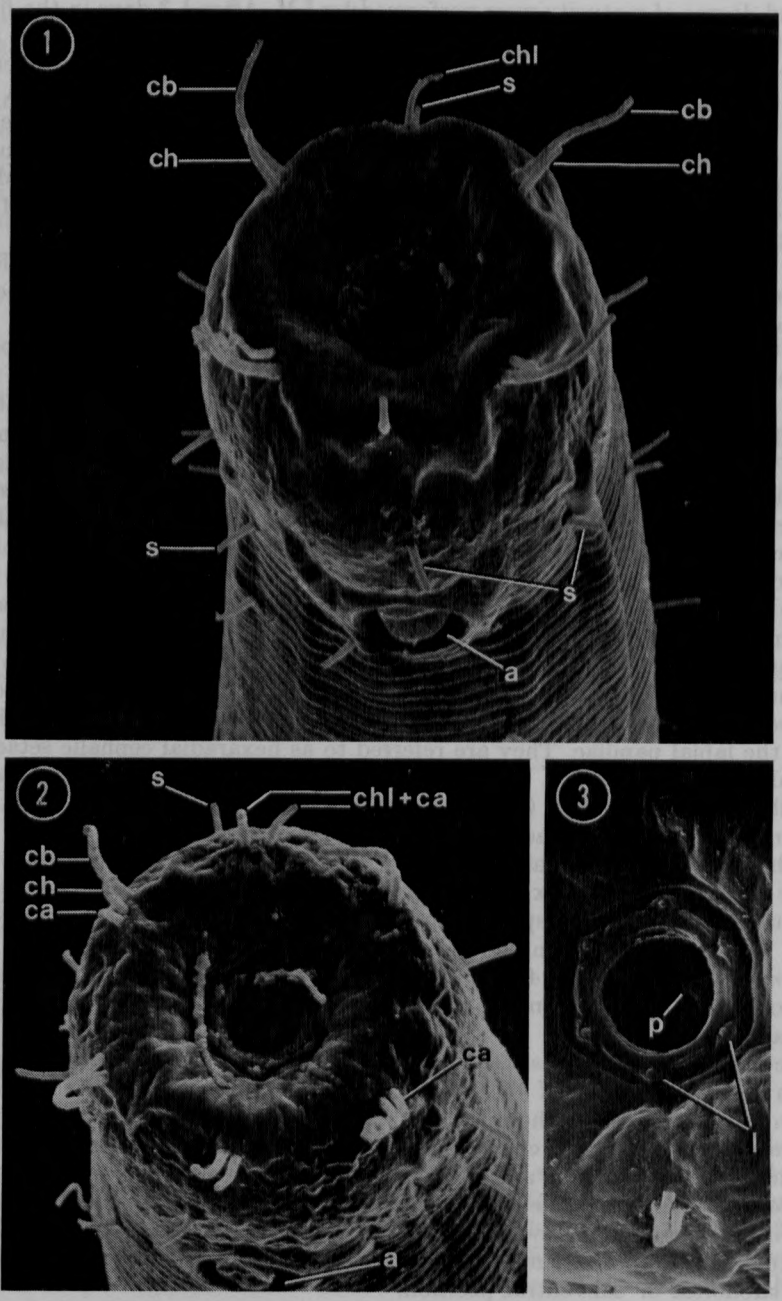
Within Linhomoeidae the arrangement of the cephalic sense organs (papillae, setae) is used to distinguish different genera (or groups of genera). A circle of six labial papillae and a circle of ten cephalic setae are reported to be characteristic of the genus *Paralinhomoeus* and some related genera. In the following this arrangement is called the basic pattern. Regular deviations from this pattern were observed in four of the five species studied.

The basic pattern is illustrated by the scanning electron micrograph of a *P. lepturus* female (Fig. 1). The circular mouth opening is surrounded by six minute lips, each bearing a small, labial papilla (cf. Fig. 3). The lips and their papillae are located in distinct positions, lateral, subdorsal, and subventral. The second circle of sense organs consists of ten cephalic setae. Six of them are short and located in the same radii as the labial papillae. They are referred to as hexaradial cephalic setae (*ch*, or in lateral position *chl*, in Fig. 1). The four longer setae (*cb*) of the cephalic setae are bilaterally arranged (cf. DE CONINCK, 1950). They are situated close to the subdorsal and subventral hexaradial setae. Four of the five species (exception : *P. ilenensis*) have subcephalic setae (*s* in Fig. 1) in distinct positions.

The basic pattern of six papillae and ten cephalic setae was observed in all young juveniles of the five species studied, and in the adults of *P. ilenensis* (*I* in Fig. 4). It was also found in the majority of the females of *P. lepturus* (*II*) and *P. intermedius* (*III* in Fig. 4). Of the five species studied, only *P. ilenensis* was found to have the same number of cephalic setae in juveniles, in adult females, and in adult males.

The males of *P. lepturus* were found to have 12-16 cephalic setae (Fig. 4). In addition to the ten setae of the basic pattern, two additional setae were always found in lateral positions. These two additional setae were observed already in old juveniles with distinct male characters. In adult males additional setae were found also near the short subdorsal and subventral hexaradial setae. However, the number of the latter additional setae varied from one to four. In Fig. 2 a male is shown with four additional setae ; two almost lateral, one almost subdorsal and one almost subventral. A few of the females studied were also found to have one additional seta in subdorsal or subventral position.

The males of *P. intermedius* were found to have six additional setae, one near each of the six short hexaradial cephalic setae. The four additional setae in almost subventral and subdorsal positions are easy to identify since they are somewhat shorter than the hexaradial ones (indicated in Fig. 4) and situated just outside the circle formed by the original cephalic setae.



Figs. 1-3. — Scanning electron micrographs of *Paralinhomoeus lepturus*. 1. Anterior end of a female (× 1800) ; 2. Anterior end of a male (× 1800) ; 3. Mouth opening and labial papillae (× 3000).

a = amphid, ca = additional cephalic seta, cb = bilateral cephalic seta, ch = hexaradial cephalic seta, chl = hexaradial cephalic seta in lateral position, l = labial papilla, p = tooth-like cuticular projection, s = subcephalic seta.

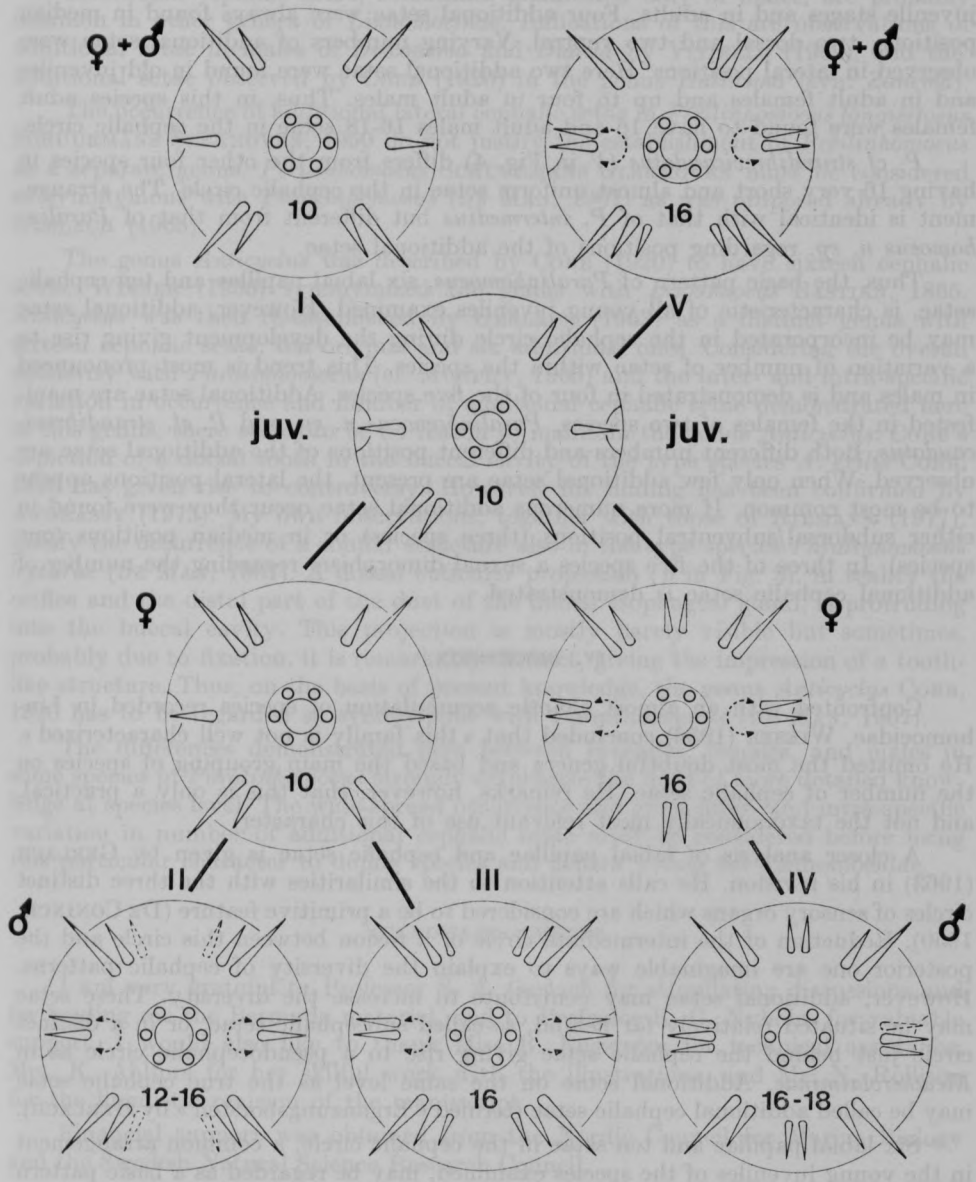


Fig. 4. — Schematic diagram illustrating the numbers and groups of cephalic setae in the five species :

I. *Paralinhomoeus ilenensis*
II. *P. lepturus*
III. *P. intermedius*
IV. *Paralinhomoeus n. sp.*
V. *P. cf strandibrevicaudatus.*

In *Paralinhomoeus n. sp.* (*IV* in Fig. 4) additional setae were observed in late juvenile stages and in adults. Four additional setae were always found in median positions, two dorsal and two ventral. Varying numbers of additional setae were observed in lateral positions. Here two additional setae were found in old juveniles and in adult females and up to four in adult males. Thus, in this species adult females were found to have 16 and adult males 16-18 setae in the cephalic circle.

P. cf strandibrevicaudatus (*V* in Fig. 4) differs from the other four species in having 16 very short and almost uniform setae in the cephalic circle. The arrangement is identical with that of *P. intermedius* but different from that of *Paralinhomoeus n. sp.* regarding positions of the additional setae.

Thus, the basic pattern of *Paralinhomoeus*, six labial papillae and ten cephalic setae, is characteristic of all young juveniles examined. However, additional setae may be incorporated in the cephalic circle during the development giving rise to a variation of number of setae within the species. This trend is most pronounced in males and is demonstrated in four of the five species. Additional setae are manifested in the females of two species, *Paralinhomoeus n. sp.* and *P. cf. strandibrevicaudatus*. Both different numbers and different positions of the additional setae are observed. When only few additional setae are present, the lateral positions appear to be most common. If more numerous additional setae occur they were found in either subdorsal/subventral positions (three species) or in median positions (one species). In three of the five species a sexual dimorphism regarding the number of additional cephalic setae is demonstrated.

IV. DISCUSSION

Confronted with an almost chaotic accumulation of species recorded in Linhomoeidae, WIESER (1956) concluded that « this family is not well characterized ». He omitted the most doubtful genera and based the main grouping of species on the number of cephalic setae. He remarks, however, that this is only a practical, and not the taxonomically most relevant use of this character.

A closer analysis of labial papillae and cephalic setae is given by GERLACH (1963) in his revision. He calls attention to the similarities with the three distinct circles of sensory organs which are considered to be a primitive feature (DE CONINCK, 1950). Reduction of the intermediate circle or a fusion between this circle and the posterior one are imaginable ways to explain the diversity of cephalic patterns. However, additional setae may contribute to increase the diversity. These setae may be situated relatively far behind, so-called subcephalic setae, or in a distinct circle just behind the cephalic setae giving rise to a pseudocephalic circle as in *Eleutherolaiminae*. Additional setae on the same level as the true cephalic setae may be called additional cephalic setae (termed « Ergänzungsborsten » by GERLACH).

Six labial papillae and ten setae in the cephalic circle, a common arrangement in the young juveniles of the species examined, may be regarded as a basic pattern in Linhomoeinae and very few exceptions are reported. However, occasional observations of additional cephalic setae have been reported earlier within *Paralinhomoeus*, by DE MAN (1907), SCHULZ (1932), and VITIELLO (1969). In some descriptions such setae have been depicted but not mentioned. Recently RIEMANN (1977) reported additional setae in lateral positions in a male of *P. lepturus*, and he also mentions the possibility of sexual dimorphism. These reports and the results obtained in the present study indicate that additional setae is a regularly occurring feature in many species of *Paralinhomoeus*.

Additional cephalic setae, sometimes manifested only in males, are probably common in other genera of *Linhomoeinae*. Indications of this are observations of additional setae in males of *Disconema* and *Didelta* by VITIELLO (1969), and the additional setae observed by COBB (1920) in the genus *Halinema* (syn. *Zanema*).

The occurrence of additional lateral cephalic setae in *Perilinhomoeus longisetosus* SCHUURMANS STEKHOVEN, 1950 do not justify the establishment of *Perilinhomoeus* as a separate genus. *Perilinhomoeus* SCHUURMANS STEKHOVEN must be considered as synonymous with *Paralinhomoeus* (DE MAN, 1907) as was proposed already by GERLACH (1963).

The genus *Anticyclus* was described by COBB (1920) to have sixteen cephalic setae. WIESER (1956) synonymized the genus with *Linhomoeus* BASTIAN, 1865. *Anticyclus* was then re-established by GERLACH (1963) as a distinct genus with sixteen cephalic setae, ten original and six additional ones. Considering the overall similarity with *Paralinhomoeus* (cf. MURPHY, 1965) and the inter- and intra-specific variation in occurrence and number of additional cephalic setae demonstrated here in this genus, there seems to be no reason to maintain the genus *Anticyclus*. COBB's depiction of a dorsal tooth in the buccal cavity of the type species *A. exilis* COBB, 1920 has given rise to controversy. However, his finding has been confirmed by ANDRÁSSY (1973). My own observations, together with those of RIEMANN (1977), justify the occurrence of a similar structure also in the type species *Paralinhomoeus lepturus* (DE MAN, 1907). A dorsal cuticular projection (*p* in Fig. 3), in reality the orifice and the distal part of the duct of the dorsal esophageal gland, is protruding into the buccal cavity. This projection is mostly barely visible but sometimes, probably due to fixation, it is remarkably distinct, giving the impression of a tooth-like structure. Thus, on the basis of present knowledge, the genus *Anticyclus* COBB, 1920 has to be regarded as synonymous with *Paralinhomoeus* (DE MAN, 1907).

The differences demonstrated here between juveniles, females, and males in some species of *Paralinhomoeus* strongly emphasize the need of more detailed knowledge at species level. The wide-spread occurrence and great inter- and intra-specific variation in number of additional cephalic setae must be considered before using this particular character to define species and genera within the Linhomoeidae.

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