



Morphology and systematics in mesopsammic nemerteans of the genus *Ototyphlonemertes* (Nemertea, Hoplonemertea, Ototyphlonemertidae)

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Key words: Nemertea, *Ototyphlonemertes*, morphology, phylogenetics, phylogenetic key, phylomorph

Abstract

Interstitial nemerteans of the genus *Ototyphlonemertes* are difficult to organize into traditional morphospecies. They occur in a multitude of slightly different local varieties that form a seemingly continuous morphological cline. In this paper, we summarize most published morphological data on the group, plus 73 new records of geographic varieties from the Mediterranean Sea in the East to the Sea of Japan in the West. We summarize morphological variation, partition traits into character and character states, propose a standardized protocol for examination of live specimens and discuss the phylogenetic structure of the group. The phylogenetic discussion leads to a hypothesis that partitions all varieties (including the established species) into six groups. These are the smallest morphologically homogenous sets of varieties (corresponding to traditional morphospecies) we can diagnose on phylogenetically reliable traits. Variation within the groups appears to be unreliable phylogenetic markers that may distinguish ecological forms rather than relatedness. However, we distinguish four of the groups by combinations of two traits, one apomorphy for a more inclusive group and one plesiomorphy, and the remaining two by one trait each that may be either a unique plesiomorphy or an apomorphy depending on the rooting, and most of them may thus join paraphyletic groups of cryptic monophyletic units. We call this kind of group phylomorph and name them the Duplex-, Pallida-, Cirrula-, Fila-, Lactea- and Macintoshi-morph (referring to the first established species within each group, i.e. *Ototyphlonemertes duplex*, *O. pallida*, *O. cirrula*, *O. fila*, *O. lactea* and *O. macintoshi* respectively). The phylogenetic scheme provides a simple tool to allocate geographical varieties to a group of possible 'species' and a phylogenetic null-hypothesis for further tests with genetic data.

Introduction

Members of the genus *Ototyphlonemertes* (Nemertea, Hoplonemertea, Ototyphlonemertidae) are small and slender nemerteans that inhabit the pore space of coarse-grained, shallow marine sands. Most are less than 10 mm long and 0.3 mm wide, and translucent grayish white. All possess statocysts (diagnostic for the family Ototyphlonemertidae) and lack eyes (although larvae may have eyes Jägersten, 1972; Chernyshev, 2000). They occur on beaches all over the world with the highest diversity in tropical and subtropical regions, and scattered populations up to at least 60° N.

The group consists of a multitude of slightly different local varieties that are difficult to organize into

traditional species. Several distinct morphotypes often occur on single beaches, but over a larger area, continuous variation in traits together with an overlapping distribution of the end states in the variation fuse the morphotypes so to create an impression of a continuous morphological cline. Trying to circumscribe species on morphological characteristics of local varieties makes it difficult to specify their morphological boundaries, and instead trying to join varieties into species on diagnostic traits makes it difficult to find traits that partitions the variation into non-overlapping groups. Presently, the genus contains about 22 recognized 'species,' but most of these are descriptions of local varieties on single beaches.

Recently, Envall (1996) analyzed the phylogenetic

relationships between the established species using morphological data from descriptions and personal observations. The analysis indicated that species possessing 'helically ridged stylet' is a monophyletic group within a paraphyletic group of plesiomorphically similar smooth-styleted species. However, the most interesting discovery was that the apomorphies were distributed along a ladder-shaped tree and many species lacked traits that could join or resolve them on the tree. This means that the apomorphies diagnose nested groups and other traits can neither join nor separate the species in a phylogenetic sense. The analysis furthermore indicated that several traits that diagnose species are homoplastic, i.e. convergent, between species and thus similar for reasons other than common ancestry, for example, similar selection regimes and/or phenotypic plasticity. This suggests that they may diagnose ecomorphs rather than the species (i.e. local varieties) they attempt to distinguish.

In this paper, we summarize morphological data from about 100 geographical *Ototyphlonemertes* varieties from the Mediterranean Sea in the east to the Sea of Japan in the West. We discuss variation, propose a standardized protocol for *in vivo* examination of *Ototyphlonemertes* specimens and discuss the phylogenetic structure of the group. The phylogenetic discussion results in a hypothesized phylogenetic structure that we use as a tool to organize the varieties.

Materials and methods

General

The data-set consists of about 500 morphological records of *Ototyphlonemertes* specimens from 96 local varieties. Twenty-one are the species descriptions of *O. antipai* Müller (1968), *O. americana* Gerner (1969), *O. aurantiaca* (du Plessis, 1891) Gerner (1969), *O. brevis* Corrêa (1948), *O. brunnea* Bürger (1895), *O. cirrula* Mock & Schmidt (1975), *O. correae* Envall (1996), *O. duplex* (Bürger (1895) Envall (1996), *O. erneba* Corrêa (1950), *O. esulcata* Senz (1993), *O. evelinae* Corrêa (1948), *O. fila* Corrêa (1953) (and Mock & Schmidt (1975), *O. lactea* Corrêa (1954), *O. macintoshi* Bürger (1895), *O. martynovi* Chernyshev (1993), *O. nikolaii* Chenyshev (1998), *O. pallida* (Keferstein (1862) Mock (1978), *O. parmula* Corrêa (1950), *O. pellucida* Coc (1943), *O. santacruzensis* Mock & Schmidt (1975) and *O. spiralis* Coe (1940). Four are the varieties Mock & Schmidt (1975)

described from Galapagos archipelago and allocated to *O. americana* and *O. erneba*, respectively called spec. 1 and 2. The remaining 71 are varieties we have recorded from the Mediterranean Sea, the North Sea, the western Atlantic Ocean, the Caribbean Sea, and the eastern and western Pacific Ocean since 1973 (Fig. 1). We summarize variation in the group and means within varieties, partition traits into characters and character states, and propose a standardized protocol for *in vivo* examinations of specimens (Appendix 1).

We use the present data set, the phylogenetic analysis in Envall (1996) and some published and unpublished genetic data to discuss the phylogenetic structure of the group. We do not allocate the new varieties into any of the recognized species, since we have no traits to join them in a phylogenetic sense, and neither do we classify the varieties as species, since we have no traits to support a monophyly for many of them. Both alternatives would confuse phylogenetic consistency of classification rather than improve it. Instead, we treat the recognized species as local varieties (which most of them indeed are), and only make two *a priori* assumptions: (i) that *Ototyphlonemertes* is monophyletic, based on the apomorphy 'lack of adult eyes,' and (ii) that morphologically similar specimens in each locale (i.e. the varieties) are monophyletic, based on morphological similarity and geographical proximity. We then reconstruct the phylogenetic nesting of varieties using the relationships between their traits.

The phylogenetic discussion results in a network that partitions the genus into six distinguishable groups. These are the least inclusive morphologically homogenous groups of varieties we can distinguish with reliable phylogenetic markers. However, we distinguish four of them on unique combinations of two traits, one apomorphy for a more inclusive monophyletic unit and one plesiomorphy, and the remaining two of one trait each that may be either an apomorphy or a plesiomorphy depending on the rooting, and all except one may thus be paraphyletic groups of varieties. We call this kind of group phylomorph and name them the Duplex-, Pallida-, Fila-, Cirrula-, Lactea- and Macintoshi-morph. We then use the hypothesized phylomorphs and geographic location to label each variety with an expression consisting of three parts. The first specifies the phylomorph, the second the sampling locale and the third distinguishable forms of the same phylomorph in single locales. For example, D-TBI specifies a variety of the Duplex-form from Tampa Bay (in Florida) number 1. The phylogenetic

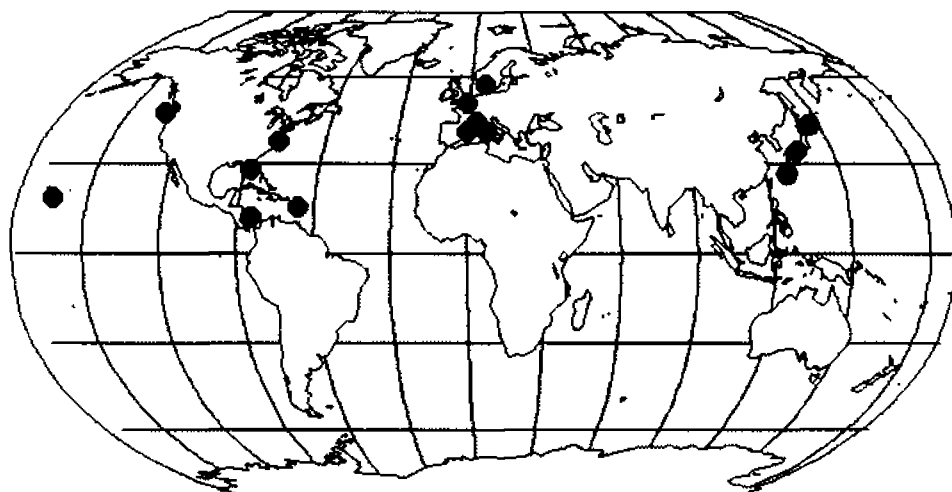


Figure 1. Map showing the distribution of sampling sites of the new *Ototyphlonemertes* varieties.

hypothesis thus functions as a tool to sort the varieties and a null-hypothesis for further tests with genetic data.

We append a list of all varieties (including the recognized species) and their qualitative character states (Appendix 2), but we do not perform a cladistic analysis of the whole character set. The reason is that Envall's (1996) analysis used most of the characters and still could not resolve the recognized species, and many of the characters are furthermore incompletely scored among the varieties. We include the list for comparative purposes for other investigators and on request from one referee. All data are stored in FilemakerPro files and will be available on the nemertean homepage (<http://nemertes.si.edu>) by the time of publication of this manuscript. (However, we continuously include new data and the summary measures may be different by then.)

Population characteristics

Ototyphlonemertes varieties inhabit shallow marine sediments from the upper tidal zone to a depth of at least 15 m. We often find tidal forms in densities of more than 1 specimen dm^{-3} , sometimes more than 100 dm^{-3} , and subtidal forms in densities of less than 1 specimen dm^{-3} . Northern tidal forms appear to increase in density during late spring, summer and autumn, to be more or less absent during winter, while subtropic and tropic tidal forms appear to have a more stable medium density all year around.

External characteristics

Body length

Body length is 1–60 mm, 8.1 ± 6.5 mm (96) (mean, SD and n among varieties). Variation is continuous from 1 to 20 mm and then a few varieties reach lengths up to 60 mm. Variation among varieties (VAV) is $\pm 80\%$ (i.e. $100 \cdot \text{SD}/\text{mean}$) and within varieties (VIV) $\pm 34 \pm 19\%$ ($n = 67$) (i.e. $100 \cdot \text{mean of SD's}/\text{mean of means}$, $\pm \text{SD}$). Body length varies over seasons (Envall, 1996) and also among genetically close and even indistinguishable varieties (Envall & Sundberg, 1998; unpubl. data). For example, the genetically indistinguishable varieties D-Sw1 and D-Vb1 (i.e. *O. correae*) on the West coast of Sweden and the South coast of England are 30 and 15 mm long respectively, and the closely related D-II (*O. duplex*) on the West coast of Italy is only about 4 mm long. (Genetic divergence between the species is about 3% in the mitochondrial CO I gene.)

Body width

Body width is between 0.08 and 1.3 mm, 0.24 ± 0.16 mm (80). Variation is continuous. VAV is $\pm 67\%$ and VIV $\pm 25 \pm 14\%$.

Stoutness

Body length and width have a relatively low correlation coefficient of 0.54, which suggest that the varieties differ in 'stoutness.' The length/width ratios produce a regression line of approximately

width= $0.014 \times \text{length} + 0.12$ with a standard deviation (i.e. the 'stoutness' range) of $\pm 35\%$. Variation is continuous. We can thus measure the character 'stoutness' (s) as percent deviation in body width from the expected width $E(w)$ as $s = 100 \times (w - E(w)) / E(w)$, where $E(w) = 0.014 \times \text{length} + 0.12$, and then partition the character into the states: slender $< -10\%$ / medium stout $\pm 10\%$ / stout $> +10\%$.

Color

The epidermis is translucent, whitish gray, yellowish, slightly orange or reddish brown. The coloration derives from granules in the epidermal cells. The cerebral region (either the brain or the surrounding tissues or both) may be slightly reddish orange, and the intestinal region may be yellowish, reddish, brownish, green or black depending on the diet. The epidermis and brain coloration is constant within varieties, although larger individuals may have a more conspicuous brain coloration, whereas the intestinal coloration may vary both within varieties and over time.

Body shape

The body is cylindrical to dorso-ventrally flattened, with a more or less pronounced widening of the anterior and/or posterior ends (Figs 2 and 3). The anterior end has been described as pointed, rounded, truncated and slightly or distinctly teardrop-shaped, and the posterior end as pointed, rounded, truncated, widened and slightly or distinctively demarcated from the rest of the body. However, the shapes vary considerably in actively moving worms and we therefore partition the shapes of the anterior end into: narrowing to rounded/distinctly widened, and the shapes of the posterior end into: narrowing to rounded/ widened/demarcated. Larger varieties are often more dorso-ventrally flattened, as could be expected, but also equally long forms may differ in the degree of flattening. In these cases, the flattening appears to be associated with differences in behavior and in the shapes of the anterior and posterior ends. More cylindrical varieties often display a more energetic coiling behavior and have more distinctively widened anterior and posterior ends, while flatter forms often display a relatively tranquil gliding behavior and have rounded to narrowing anterior and posterior ends. This suggests that the characters form functionally linked sets adapted to different environmental conditions.

Epidermis

Epidermis is generally 5–30 μm thick, and often thicker in the anterior and posterior ends than in the mid-body region. The anterior end may be up to two times thicker, and the posterior end up to three times thicker than the mid-body region. The thickness in the posterior end correlates with the development of a caudal adhesion plate (see below).

Adhesion

All *Ototyphlonemertes* varieties possess some means to avoid being flushed out of the sand in strong water currents. Some are overall sticky (similar to nemerteans in general), a state we refer to as continuous adhesion; some have an epidermal specialization in the caudal end called a caudal adhesion plate (see Kirsteuer, 1977), and some coil their posterior end for the same purposes. A caudal plate protudes in live squeezed preparations as a widening of the caudal end and a thickened epidermis (Fig. 3). However, a mere thickening of the epidermis does not necessarily provide evidence for the presence of a caudal plate (Kirsteuer, 1977). It is therefore important to observe the adhesive behavior in free-crawling specimens.

Kirsteuer (1977) studied the distribution of a caudal plate among 'species' and noted that "there was no indication that the presence or absence of this structure is intraspecifically variable" (meaning that the trait may diagnose species). However, he did not specify which species he examined or how he allocated the varieties into species. We have, on the contrary, observed a considerable variation among morphologically similar varieties in different locales, and this is true for several different morphotypes, each presumably joining close relatives. This indicates that the trait may be homoplastic.

Cephalic furrows

Most *Ototyphlonemertes* varieties possess two or four lateral cephalic furrows forming one or two epidermal folds that encircle the body (Fig. 2a). One is situated at or posterior to the statocysts and encircles the body obliquely so to form a dorsal v pointing backward and a ventral v pointing forward, and the other is situated in the midbrain region and encircles the body transversely lacking the dorsal and ventral v's. The furrows may be extremely faint and visible only when the specimens contract. Some varieties have additional epidermal folds at the openings of the cerebral organs,

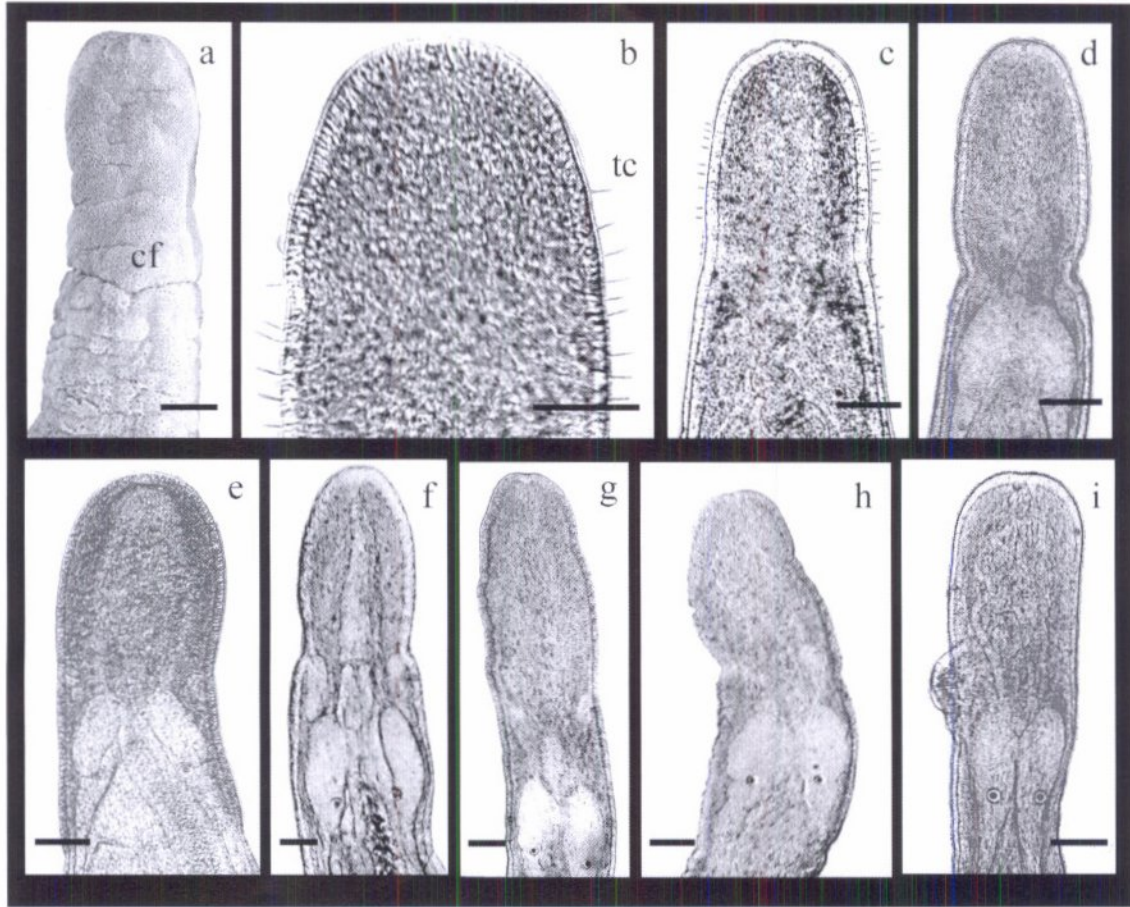


Figure 2. SEM photograph (a), and photomicrographs of live squeezed specimens showing posterior cephalic furrow (cf), tactile cirri (tc) and various shapes of the anterior end. From the varieties a, D-Sw1; b, F-M1; c, P-J2; d, D-SB1; e, D-Vb1; f, P-FP2; g, F-SB1; h, F-M1; i, L-SB1. (Scale bars 100 μ m).

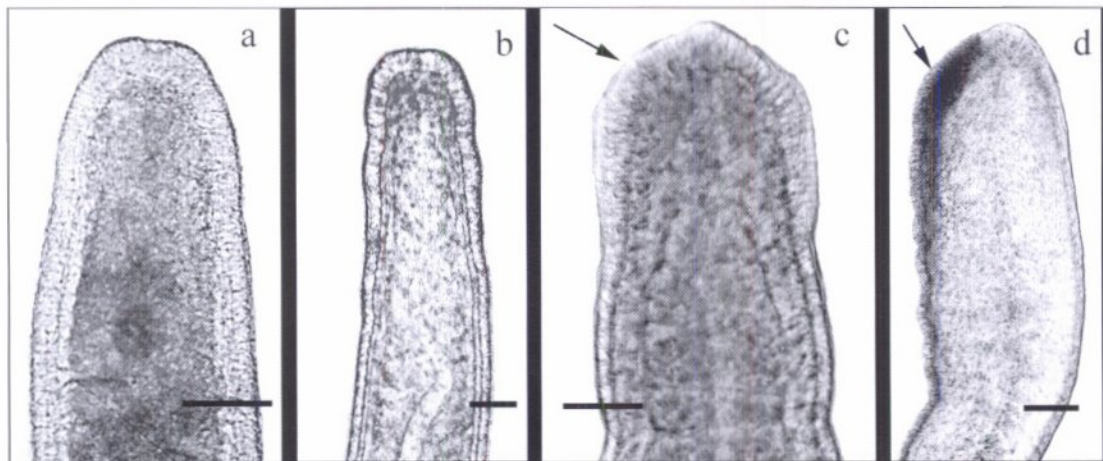


Figure 3. Photomicrographs of live squeezed specimens showing caudal adhesion plate (arrows) and various shapes of the posterior end. From the varieties a, P-FH1; b, D-SB1; c, F-M1; d, F-SB1. (Scale bars 100 μ m).

but these do not appear to compose true cephalic furrows with sensory epithelium. The character may be difficult to assess due to contraction artifacts.

Sensory cirri

Many *Ototyphlonemertes* varieties possess epidermal tufts of elongated cilia called cirri (Fig. 2) (see Kirsteuer, 1977). Cirri may reach a height of 50 μm (80 μm according to Kirsteuer, 1977) and occur either over the whole body (only one record), anterior to the cephalic furrow and in the extreme hind end, or only in the extreme hind end. Caudal cirri are mostly about 50 μm high, whereas cephalic cirri range from barely higher than body cilia up to the same height as the caudal cirri. Müller (1968) used the distribution of cephalic cirri to diagnose a species (*O. antipai*). Later both Mock & Schmidt (1975) and Kirsteuer (1977) agreed that this distribution is species specific, and Norenburg (1988) proposed a standardized formula to account for it. However, the distribution is difficult to detect. We can conveniently only observe the lateral cirri in squeezed worms, and since these often have a dorsal or ventral orientation, they may be visible only at one side or not at all. It is also difficult to interpret in a phylogenetic sense. The distribution is often unique for different varieties, and thereby difficult to partition into informative states (i.e. possessed by more than one variety), and we have genetic data that indicate that it may vary between genetically identical varieties (i.e. genetic species) in different locales so to be more similar between populations of different genetic species than within the genetic species themselves (Envall & Sundberg, 1998). Envall's (1996) analysis moreover suggested that the mere presence of cephalic cirri is homoplastic (i.e. convergent). The trait is thus difficult to detect, code, and interpret.

Cerebral organs

Ototyphlonemertes varieties possess cerebral organs that are normally developed, lack the gland region, lack the gland region and the chamber, or they lack the organs completely. In live examinations, we can only distinguish whether they possess cerebral organs or not, and whether they appear 'normally' developed or as simple pits.

Internal characteristics

Rhynchocoel

Rhynchocoel length is <8 mm, 2.8 ± 1.6 mm (88), which corresponds to 10–80% of total body length. Variation is continuous. The rhynchocoel may reach the hind end in juveniles, but not in mature specimens of any known variety (possibly excluding *O. esulcata*). Taxonomists traditionally record the relative length based on the implicit assumption that this is less variable than the absolute length, but our data on the contrary indicate that the relative length may vary as much or even more than the absolute length.

Proboscis

The proboscis is either straight or looped in the rhynchocoel. These states seem to be constant within varieties and correlate with particular proboscis types. However, the proboscis often alternates between the two states in actively moving worms and this makes the character difficult to assess. We commonly assess it as looped if it loops in any state of contraction, and to straight if it never loops.

Anterior part of the proboscis

The anterior part of the proboscis is <6 mm long, 1.4 ± 1.1 mm (82). It is covered with an epithelium that is either flat or equipped with glandular protrusions called papillae (Fig. 4) (see Kirsteuer, 1977). Papillae vary in shape from low wart- or flap-like protrusions to high finger-like cones, and may contain spicules called rhabdites or have branched hair-like attachments. Papilla shape is constant within varieties and genetically close relatives.

Diaphragm

Diaphragm length is <500 μm , 126 ± 85 μm (68); width is <120 μm , 70 ± 18 (37) and length/width ratio is <5, 1.5 ± 0.9 (63). The diaphragm occurs in two distinct states (Fig. 5) (see Kirsteuer, 1977). One is approximately as long as it is wide (Fig. 5a–f), and the other is elongated with a long *ductus ejaculatoris* (the duct that connects the poison containing middle chamber to the stylet) (Fig. 5g, h). The length/width ratios have a discontinuous distribution with aggregations above and below 1.5, thus supporting the separation of the two states. However, the ratio does not always accurately identify the types. The short type

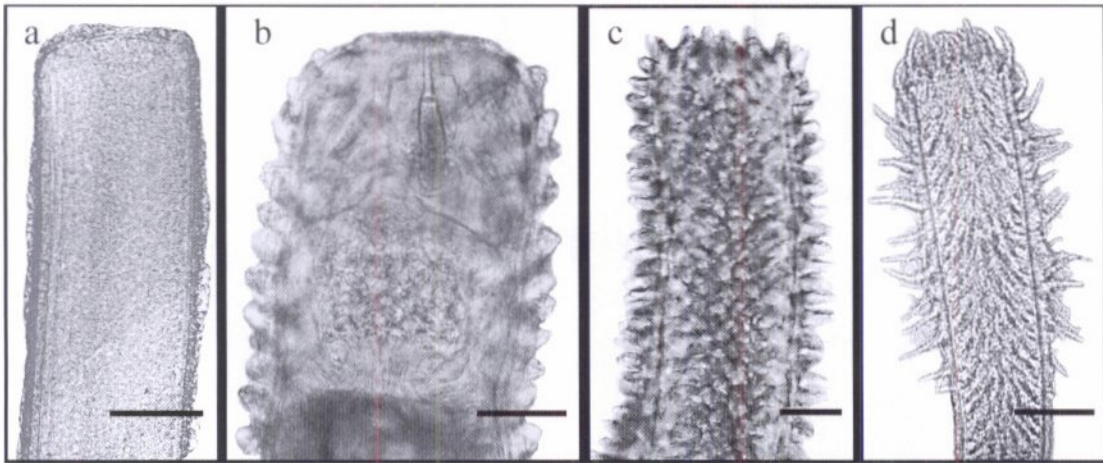


Figure 4. Photomicrographs of live squeezed specimens showing different types of proboscis papillae. From the varieties a, M-Vb1; b, D-Vb1; c, P-FP2; P-CBC6. (Scale bars 50 μ m).

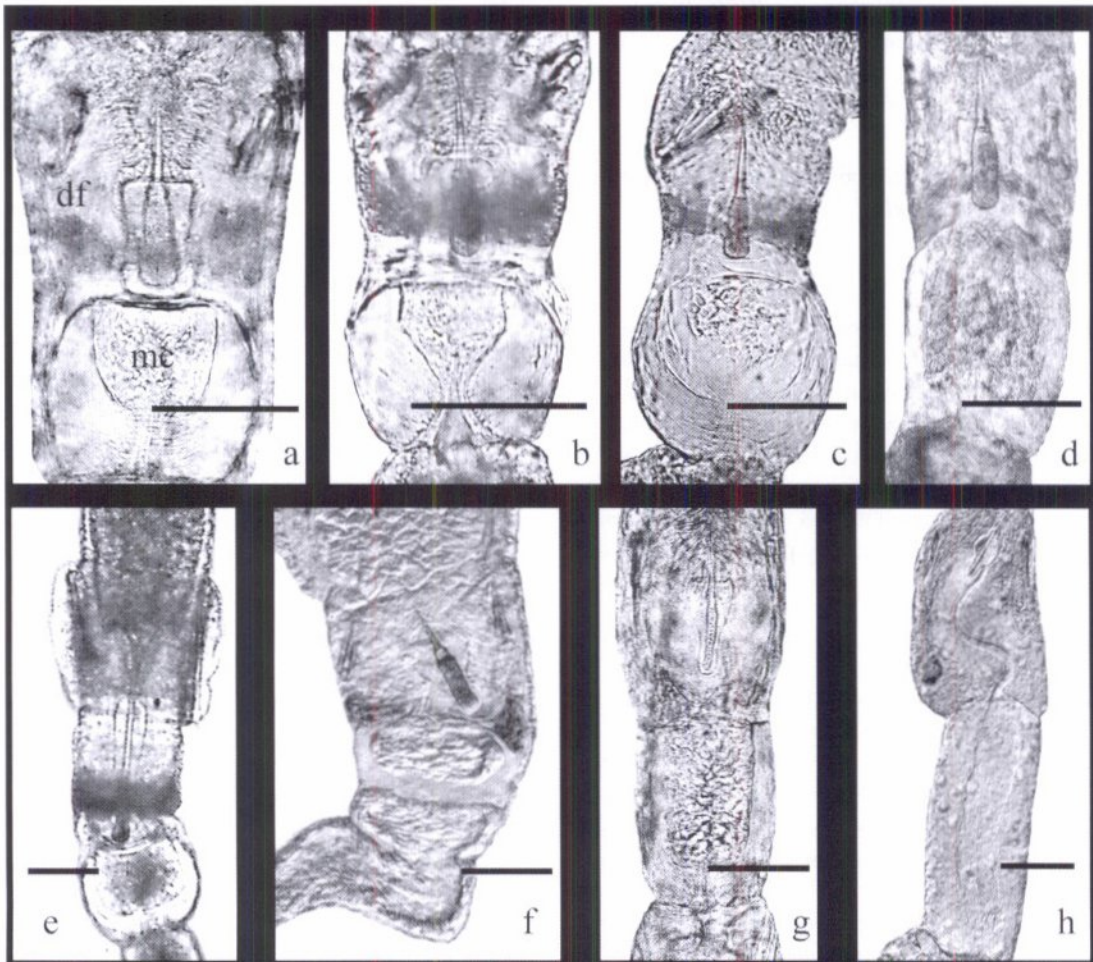


Figure 5. Photomicrographs of live squeezed specimens showing diaphragm (df) of the short type (a–f) and the long type (g and h), and middle chamber (mc) of the short type (a–g) (with the square condition (g)) and the long type (h). From the varieties a, P-FH1; b, P-FP2; c, P-CBC6; d, D-Vb1; e, P-CBC4; f, F-FP1; g, L-SB1; h, M-M1). (Scale bars 50 μ m).

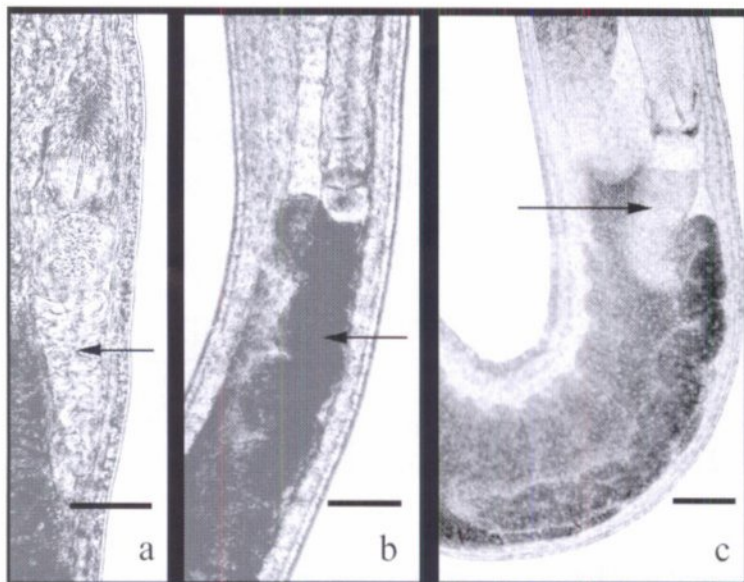


Figure 6. Photomicrographs of live squeezed specimens showing posterior proboscis chamber of the vesicular type (a) (indicated with an arrow), of the opaque type (b) (indicated with an arrow) and of the opaque type with a specialized anterior part (c) (the anterior part indicated with an arrow). From the varieties a, L-SB1; b, P-CBC1; c, F-SB1. (Scale bars 100 μm).

sometimes carries an extremely long stylet and is then considerably elongated (Fig. 5e).

Accessory stylet pouches

All *Ototyphlonemertes* varieties possess accessory stylet pouches that are situated lateral or posterior-lateral to the stylet. Most varieties have two pouches, but two consistently have three. Each pouch contains up to eight accessory stylets, 3.2 ± 1.1 ($n=80$). Variation is fairly small within varieties and among closely related varieties, for example 1–3, or 4–6. The accessory stylets may also consistently point either forward or backward.

Middle chamber

Middle chamber length is $<1000 \mu\text{m}$, $152 \pm 184 \mu\text{m}$ (68). It occurs in two distinct states (Fig. 5) (see Kirsteuer, 1977). One is onion-shaped (bulbous) (Fig. 5a–f) and the other is tubular (Fig. 5h) (see also Kirsteuer, 1977). Some varieties have an intermediate condition that is short but distinctly square (Fig. 5g). The length/width ratio is <12 , 1.7 ± 2.0 (61). Variation is discontinuous with aggregations above and below 1.5, thus supporting the separation of the two states.

Posterior chamber

Posterior chamber length is $<5 \text{ mm}$, $0.8 \pm 0.7 \text{ mm}$ (79). It occurs in three distinct states (Fig. 6). The first has a vesicular content making it appear bright and reflective (Fig. 6a), the second has a granular content making it appear black when trans-illuminated and opaque white with reflected light (Fig. 6b), and the third is similar to the second type, but has a specialized anterior part (Fig. 6c).

Ratio anterior part/posterior chamber

The length ratio between the anterior part and posterior chamber is 0.2–5, 2.1 ± 1.9 (79). VAV is $\pm 90\%$ and VIV $\pm 34 \pm 22\%$. The character appears phylogenetically informative, but an extreme flexibility of the proboscis makes exact measurements difficult and the two parts also seem to develop asynchronously. We have data that indicate that the anterior part grows to full length more rapidly than the posterior does, resulting in an initial steep ratio increase followed by a slow decrease. It is important to consider these developmental properties, since some varieties easily dispatch their proboscis and single specimens therefore may have a regenerating proboscis with an unrepresentative ratio for its variety.

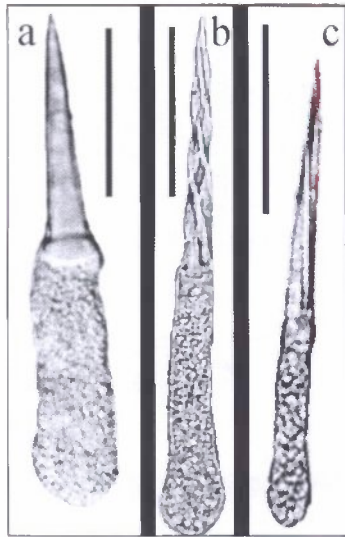


Figure 7. Photomicrographs of live squeezed specimens showing stylets of the smooth type (a), a type with crossing ridges and (b) a type that appears as two intertwined strings (c). From the varieties a, D-Vb1; b, M-Vb1; c, M-SB1. (Scale bars 25 μm).

Stylet

The stylet is either smooth (Fig. 7a) or ridged (Fig. 7b, c) (see also Kirsteuer, 1977). The ridges may appear as crossing strands (Fig. 7b), two or three twined strands (Fig. 7c), or two parallel strands (Chernyshev, 1998). Stylet length is $<180 \mu\text{m}$, $39 \pm 24 \mu\text{m}$ (74). VAV is $\pm 62\%$ and VIV $\pm 16 \pm 14\%$. The relatively small VIV suggests that the length can distinguish single varieties, but the length varies considerably among morphologically similar varieties in different locales. Stylet maximum width (closest to the basal knob) is $<15 \mu\text{m}$, $4.6 \pm 3 \mu\text{m}$ (29), and length/width ratio is 9.7 ± 5.6 (32).

Basis

Basis length is $<80 \mu\text{m}$, $31 \pm 10 \mu\text{m}$ (73); width is $<20 \mu\text{m}$, $10.1 \pm 4.9 \mu\text{m}$ (52), and length/width ratio is 3.7 ± 1.7 (55). VAV is ± 32 , ± 49 and $\pm 46\%$, respectively, and VIV ± 14 , ± 18 and $\pm 18\%$, respectively. The length/width ratios have a discontinuous distribution with aggregations below and above 3.5, thus supporting the character states thick and thin (see Kirsteuer, 1977; Envall, 1996). The basis is regularly or irregularly cylindrical, barrel-shaped, conical, and with or without a median constriction, but the shape varies considerably within varieties.

Stylet/basis length ratio

The stylet/basis length ratio is 1.3 ± 0.5 (73). VAV is $\pm 49\%$ and VIV $\pm 12\%$. The small VIV suggests that the ratio may distinguish varieties and genetic data support that it has a small variation among closely related varieties. Kirsteuer (1977) wrote that "the proportions of the armature, i.e. ratios of stylet length to base length, stylet length to stylet width and base length to base width, showed only a slight intraspecific variation and differences between species turn out to be significant (at the 95% level) when the relevant data are compared with a *t*-test." However, it is unknown which species he examined and he did not provide any data.

Statocyst and statolith

All *Ototyphlonemertes* varieties possess a pair of statocysts situated in the posterior parts of the ventral lobes of the paired ganglia (Fig. 8) (see Kirsteuer, 1977). Each statocyst contains a statolith consisting of two to more than 20 granules. Statocyst maximum diameter is $<50 \mu\text{m}$, $24 \pm 10 \mu\text{m}$ (46), statolith maximum diameter is $9.9 \pm 2.8 \mu\text{m}$ (36), and granule maximum diameter is $4.0 \pm 2.1 \mu\text{m}$ (30). The statocyst occurs in two distinct shapes, spherical or ovoid. A spherical shape is mostly associated to a median position in the ganglia and poly-statoliths (>12 granules), whereas an ovoid shape is associated to a lateral position on the inner margin of the ganglia and oligo-statoliths (<12 granules). However, the statocysts are embedded in soft tissue and the shape varies considerably in squeezed preparations. The granule number varies little within varieties and among closely related varieties. It can be 2, 3, 4, irregular clusters of 4–10, or morula-shaped balls consisting of about 12, 16 or more than 20 granules. Those that consist of 3 to 10 granules vary more both in numbers and sizes of granules, and they mostly occur in subtidal forms. Taxonomists traditionally discern oligostatoliths, consisting of 2–10 granules, and polystatoliths, consisting of 10 to more than 20 granules. We further partition the oligostatoliths into 2 and 3–10 granules, and the polystatoliths into type 1, appearing as a rosette consisting of 1 central and 6 surrounding granules, and type 2, appearing as a rosette consisting of 2–3 central and 8–10 surrounding granules. However, there are intermediate conditions between both 2 and 3–10, and type 1 and 2.

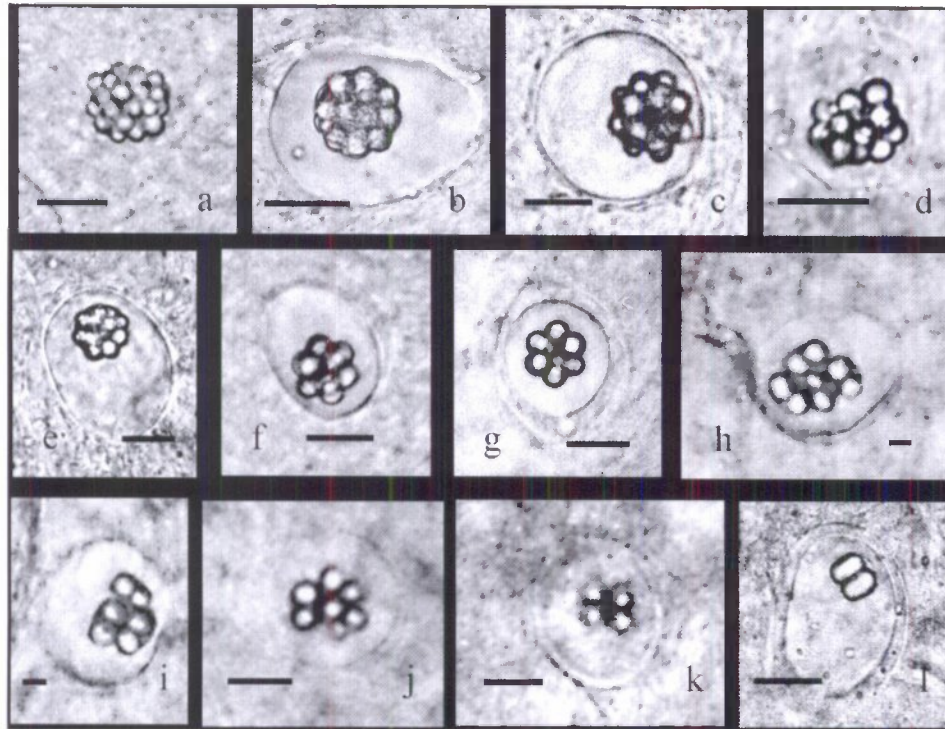


Figure 8. Photomicrographs of live squeezed specimens showing different statoliths. From the varieties a, M-J1; b, M-Vb1; c, L-SB1; d, L-LP1; e, M-CBC1; f, F-M1; g, F-FP1; h, i P-FP5; j, k, P-J1; l, D-Vb1. (Scale bars 10 μm).

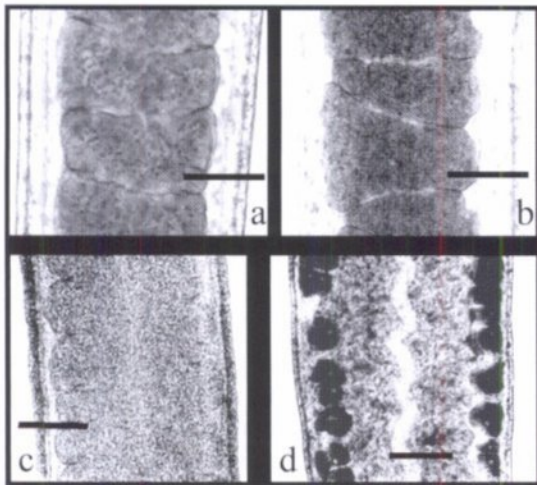


Figure 9. Photomicrographs of live squeezed specimens showing intestine with deep (a, b) and shallow diverticula (c, d). From the varieties a, F-M1; b, F-SB1; c, D-Vb1; d, M-CBC1. (Scale bars 100 μm).

Intestine and surrounding tissue

The intestine varies mainly in the depth of the lateral folds that form the intestinal pouches called divertic-

ula (Fig. 9). Diverticula range from a total absence to deep pouches making the intestine appear as a stack of platelets. The intestine is sometimes surrounded by a layer of translucent tissue that can reach a thickness of about 1/2 intestine width. This tissue has sometimes been called 'postcerebral extension of the cephalic gland' and sometimes 'parenchyma', but these terms may denote homologous conditions (see Envall, 1996). However, in live examinations we can only deduce whether translucent tissue is present or not. Large amounts often occur together with deep diverticula and uniform body width, together giving the specimens an overall tubular appearance, and lesser amounts often occur together with shallow or no diverticula and widening towards the hind end, together giving the specimens a more sack-shaped appearance.

Phylogenetic discussion

Then, how shall we organize these varieties? One option is to describe all as separate species, but then the descriptions may not diagnose some of them. Envall's (1996) analysis and genetic data (Envall & Sundberg,

Table 1. Character matrix showing different combinations of a key set of phylogenetically reliable characters that occur among *Ototyphlonemertes* varieties, and the names of the phylomorphs they typify (see text for explanation)

Statolith	Stylet	Basis	Diaphragm	Middle chamber	Morphotype
2 granules	Smooth	Thick	Short	Short	Duplex-type
3-10	Smooth	Thick	Short	Short	Pallida-type
Poly	Smooth	Thick	Short	Short	Cirrula-type
Poly	Spiral	Thick	Short	Short	Fila-type
Poly	Spiral	Thin	Long	Short	Lactea-type
Poly	Spiral	Thin	Long	Long	Macintoshi-type

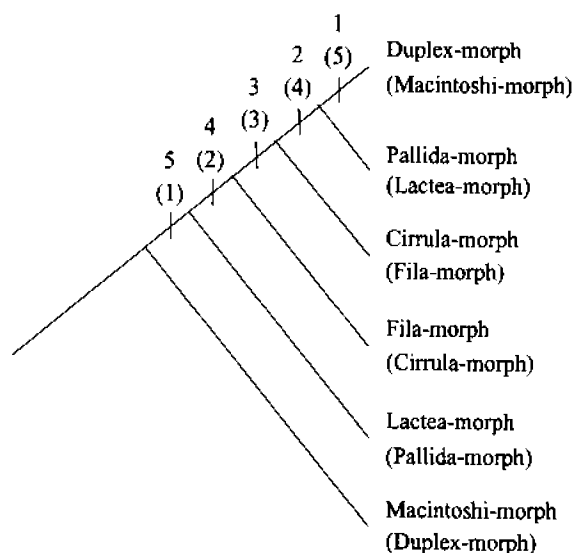


Figure 10. Phylogenetic tree showing the relative position of the 6 phylomorphs (see text). Text within brackets show an alternative rooting in the Duplex-morph.

1998; unpubl. obs.) indicate that several traits that distinguish the varieties are homoplastic (i.e. convergent), e.g. anterior cirri distribution and all quantitative traits, and may thus diagnose ecological forms rather monophyletic units. Such species would make allocation of new varieties to any of the described species even more difficult, and ultimately lead to the alternatives of joining all varieties into one species or describing all as geographic species. Neither of these alternatives would produce a phylogenetic organization or increase the knowledge of the phylogenetic nesting within the group. Another option is to group varieties into species on diagnostic characters. However, the reliable apomorphies seem to be distributed along the stem of a ladder-shaped tree (Envall, 1996) and thus diagnosing groups that are nested in each other. Such species are

non-Linnean and will become less morphologically homogenous the more inclusive they get. However, we have another option to organize the varieties into groups that are both morphologically homogenous and have a certain kind of phylogenetic significance. We then first extract the most reliable phylogenetic markers from the character set. Envall's analysis (1996) suggested that statolith shape, stylet structure, basis shape and middle chamber shape are single apomorphies, and present data set and genetic data suggest that the diaphragm structure belong to this category. These traits sum up to a data set consisting of five characters and 11 states, and thus 48 possible combinations of character states. Of these, only six occur among known *Ototyphlonemertes* varieties (Table 1). Assuming that the combinations have originated in an evolutionary sequence, we can order them parsimoniously separated by as few steps as possible. This gives us a sequence of combinations that are separated by only one hypothesized transformation between each. Allocating all described species into these combinations produces a network (Fig. 10) that is congruent with the tree Envall (1996) proposed. This character set merely reduces resolution and partitions the whole group into six morpho-types. These groups are similar in all characters of the reduced character set and also in several other characteristics. We call this kind of group phylomorph and name them the Duplex-, Pallida-, Cirrula-, Fila-, Lactea-, and Macintoshi-morph.

The Duplex-morph joins intertidal forms with a short diaphragm, bulbous middle chamber, smooth stylet, thick basis, cirri at both ends, one cephalic furrow (sometimes indistinct) that encircles the body posterior to the statocysts, normally developed cerebral organs, low proboscis papillae (with some exception), two accessory stylet pouches containing one to four accessory stylets each (pointing both forward and backward), ovoid statocysts with statoliths consisting of on average two granules (rarely up to eight).

Body length <40 mm, width <0.7 mm. Color translucent whitish (with the exception of the reddish-brown *O. brunnea*), with or without a reddish hue in the brain region. Intestinal region yellow, orange, pink, red, brown or green. Anterior end narrowing, rounded or teardrop; posterior end slightly pointed, rounded, truncated or distinctively widened, with or without a caudal adhesion plate (no record explicitly mentions an adhesive behavior with the caudal end). Cephalic cirri often sparse and short, length <35 μm . Rhynchocoel length <4.5 mm, corresponding to <50% of total body length. Anterior proboscis part length

<2.8 mm, proboscis papillae height <20 μm . Diaphragm length and width <120 μm . Gland secretions lateral to the basis absent or present. Middle chamber length and width <120 μm . Posterior chamber length <3 mm. Anterior/posterior chamber length ratio 0.5–2.5. Stylet length 20–40 μm , length/width ratio 4–10. Basis pear-shaped, conical, barrel-shaped or regularly cylindrical; length 20–42 μm ; length/width ratio 2–3.5. Stylet/basis length ratio mostly 1, but may reach 1.7. Statocyst maximum diameter 10–50 μm . Statolith maximum diameter 7–15 μm . Granule maximum diameter 3–10 μm . Tissue between intestine and body wall muscle layers absent, sparse or extensive. The phylomorph has a worldwide distribution and includes the recognized species *O. antipai*, *O. aurantiaca*, *O. brunnea*, *O. correae*, *O. duplex*, *O. evelinae* and *O. martymovi*.

The Pallida-morph joins the great majority of the subtidal and a few intertidal forms. They have short diaphragm, bulbous middle chamber, smooth stylet, thick basis, cirri at both ends (mostly many and high cirri in the anterior end), high proboscis papillae, two accessory stylet pouches (often containing four to six stylets each), ovoid statocysts with statoliths consisting of 3–12 granules (larger numbers often form irregular clusters of differently sized granules).

Body length <30 mm, width <0.7 mm. Color translucent whitish or reddish-brown, with/without a reddish hue in the brain region. Intestinal region yellow, orange, pink, red, brown or green. Cephalic furrow present, posterior to statocysts and/or in the midbrain region. Anterior end narrowing, rounded, or slightly teardrop-shaped; posterior end slightly pointed, rounded, truncated or distinctively widened with or without a caudal adhesion plate. Cirri length <35 μm . Cerebral organs normally developed or forming simple pits. Rhynchocoel length <10 mm, often extending more than 50% of total body length (sometimes reaching hind end in juveniles). Proboscis anterior part length <5.5 mm. Proboscis papillae 15–60 μm , sometimes carrying arborescent hair-like attachments or containing rhabdites. Diaphragm length <355 μm , width <300 μm , length/width ratio <4.5 (extremely elongated forms always occur together with an extremely elongated stylet and never have elongated *ductus ejaculatoris*). Two accessory stylet pouches, each containing 2–7 stylets that point both forward and backward. Gland secretions lateral to the basis absent or present. Middle chamber length and width <260 μm . Posterior chamber length <2.5 mm. Anterior/posterior chamber length ratio 1–4 (often 2–

3). Stylet length 9–170 μm , length/width ratio 2–14. Basis pear-shaped, conical, barrel-shaped or regularly cylindrical with or without median constriction; length 15–70 μm , length/width ratio 1.5–3.5. Stylet/basis length ratio 0.8–4. Statocyst maximum diameter 8–74 μm . Statolith maximum diameter 5–15 μm . Granule maximum diameter 1–6 μm (often variable sizes). Tissue between intestine and body wall muscle layers absent, sparse or extensive. The phylomorph has a worldwide distribution and includes the recognized species *O. erneba*, *O. pallida* and *O. santacruzensis*.

The Pallida-forms are about 13% stouter than *Ototyphlonemertes* forms in general ($p=0.01$), and also significantly stouter than Duplex- and Fila-forms ($p=0.01$ and 0.002, respectively), but not than Macintoshi-forms ($p=0.21$) (these have a larger internal variation). The values confirm our intuitive impression that subtidal forms (mostly of the Pallida-morph) generally are stouter than tidal forms, but they also allocate these differences specifically to the varieties of the Pallida-morph and their hypothesized closest relatives within the Duplex- and Fila-morphs. This suggests that the Pallida-morph is specifically adapted to a subtidal existence.

The Cirrula-morph joins intertidal forms with short diaphragm, bulbous middle chamber, smooth stylet, thick basis, short diaphragm and morula-shaped polystatoliths consisting of 10–20 granules.

Body length <40 mm, width <0.7 mm. Color translucent whitish, with/without a reddish tinge in the brain region. Intestinal region yellow, orange, pink, red, brown, or green. Anterior end narrowing, rounded, or slightly, or distinctively teardrop-shaped; posterior end slightly pointed, rounded, truncated, or distinctively widened with a well developed caudal adhesion plate. Cirri absent, present in both ends, or present over the whole body; length <35 μm . Cephalic furrow present, posterior to the statocysts (more or less distinct). Cerebral organs absent or forming simple pits. Rhynchocoel length <4 mm, corresponding to <50% of total body length. Anterior proboscis part length <0.9 mm. High papillae, >15 μm . Diaphragm length and width <200 μm (it may be elongated due to a long stylet). Accessory stylet pouches 2, each containing about 4–6 accessory stylets pointing both forward and backward, gland secretions lateral to the basis absent or present. Middle chamber length and width <100 μm (except in one form where it reaches a length of about 1000 μm). Posterior chamber length <0.7 mm. Anterior/posterior chamber length ratio 0.4–2.3. Stylet length 15–22 μm (N513 has a long

but not measured stylet), length/width ratio 3–4. Basis pear-shaped, conical, or irregular cylindrical; length 15–24 μm ; length/width ratio about 3. Stylet/basis length ratio mostly 1, but may reach 4.5. Statocyst maximum diameter 12–20 μm . Statolith maximum diameter 10 μm . Granule maximum diameter 3–4 μm . Tissue between intestine and body wall muscle layers absent, sparse, or extensive. The phylomorph has a documented distribution in the West Atlantic and the Pacific Ocean and includes the recognized species *O. cirrula* and *O. brevis*.

The Fila-morph joins intertidal forms with short diaphragm, bulbous middle chamber (except for the original *O. fila*, which possesses an elongated middle chamber), spiral stylet, thick basis lacking gland secretions lateral to the basis, cirri in both ends; medium-high flap-like proboscis papillae, two accessory stylet pouches containing about one to four accessory stylets that point both forward and backward, and morula-shaped statoliths composed of about 12 granules (type 1).

Body length <20 mm, width 0.5 mm. Color translucent whitish, with/without a reddish tinge in the brain region. Intestinal region colorless, yellow, orange, pink, red, brown, or green. Anterior end pointed, rounded or teardrop-shaped; posterior end slightly pointed, truncated, or distinctively widened with a well-developed caudal adhesion plate. Cephalic furrow present, posterior to the statocysts. Cerebral organs normally developed or simple pits. Cirri length <25 μm . Rhynchocoel <5 mm, corresponding to <50% of total body length. Anterior proboscis part length <2.7 mm. High papillae, <15 μm (except *O. parmula*). Diaphragm length and width <120 μm . Middle chamber length and width <130 μm . Posterior chamber length <1.8 mm. Anterior/posterior chamber length ratio 0.5–4. Stylet length 15–48 μm ; length/width ratio 7–11.5. Basis pear-shaped, conical, barrel-shaped, or regularly cylindrical; length 15–51 μm ; length/width ratio 2–6. Stylet/basis length ratio mostly 1. Statocyst maximum diameter 15–25 μm . Statolith maximum diameter 7–12 μm . Granule maximum diameter 1–4 μm . Tissue between intestine and body wall muscle layers absent, sparse, or extensive. The phylomorph has a documented distribution in the West Atlantic and Pacific Ocean (not yet found in Europe), and includes the recognized species *O. fila* and *O. parmula*.

The Lactea-morph joins intertidal forms with long diaphragm (which may be extremely shortened), short (often squarish) middle chamber, spiral stylet, thin

basis, spherical statocysts containing morula-shaped statoliths composed of 12–20 granules. They lack cirri, cerebral organs, proboscis papillae and gland secretions lateral to the basis.

Body length <60 mm, width <1.5 mm. Color translucent whitish to orange, with/without a reddish hue in the brain region. Intestinal region colorless, yellow, orange, pink, red, brown, or green. Anterior end pointed, rounded or teardrop-shaped; posterior end slightly pointed, rounded, truncated, or distinctively widened with a well developed caudal adhesion plate. Cephalic furrow either absent, present in the midbrain region and/or present posterior to the statocysts. Rhynchocoel length <8 mm, corresponding to about 50% of total body length. Proboscis anterior part length <3.5 mm. Diaphragm length <400 μm , width <120 μm . Accessory stylet pouches 2, each containing 2–6 accessory stylets pointing forward, backward or both. Middle chamber length <800 μm , width <120 μm . Posterior chamber length <3.3 mm. Anterior/posterior chamber length ratio 0.4–4. Stylet length 14–150 μm ; length/width ratio 6–30. Basis pear-shaped, conical, barrel-shaped, or regularly, or irregularly cylindrical; length 15–80 μm ; length/width ratio 3–8. Stylet/basis length ratio mostly 0.4–2. Maximum diameter of the statocyst 10–34 μm ; statolith 5–14 μm ; granule 1–4.5 μm . Tissue between intestine and body wall muscle layers mostly absent. The phylomorph has a documented distribution in the West Atlantic and Pacific Ocean (not yet found in Europe), and includes the established species *O. americana* and *O. lactea*.

The Macintoshi-morph joins intertidal and a few subtidal forms with long diaphragm, long tubular middle chamber, spiral stylet, thin basis, spherical statocysts and morula-shaped statoliths composed of 20 or more granules. They mostly lack cirri, cerebral organs, proboscis papillae and gland secretions in the diaphragm.

Body length <60 mm, width <1.5 mm. Color translucent whitish to orange, with/without a reddish hue in the brain region. Intestinal region colorless, yellow, orange, pink, red, brown or green. Anterior end pointed, rounded, or teardrop-shaped. Posterior end slightly pointed, rounded, truncated or distinctively widened with a well developed caudal adhesion plate. Cephalic furrow absent, present in the midbrain region and/or present posterior to the statocysts. Rhynchocoel length <8 mm long, corresponding to about 50% of total body length. Anterior proboscis part length <3.5 mm. Diaphragm length <400 μm . Accessory stylet pouches 2, each with 2–6 ac-

cessory stylets pointing forward, backward or both. Middle chamber length $< 800 \mu\text{m}$. Posterior chamber length $< 3.3 \text{ mm}$. Anterior/posterior chamber length ratio 0.4–4. Stylet length 14–150 μm , length/width ratio 6–30. Basis pear-shaped, conical, barrel-shaped or regularly or irregularly cylindrical; length 15–80 μm ; length/width ratio 3–8. Stylet/basis length ratio mostly 0.4–2. Maximum diameter of the statocyst, 10–34 μm ; statolith, 5–14 μm ; granule, 1–4.5 μm . Tissue between intestine and body wall muscle layers mostly absent. The phylomorph has a worldwide distribution and includes the recognized species *O. macintoshi*, *O. pellucida* and *O. spiralis*.

Phylogenetic key

These six proposed phylomorphs are similar in all phylogenetically reliable characters and in other characteristics, but despite this, most of them lack unique apomorphies (except for the most derived type). Assuming that the root of the network is situated within the Macintoshi-morph (which appears most probable from genetic evidence), the Macintoshi-morph is diagnosed by the plesiomorphic trait 'long middle chamber,' because all other forms possess the apomorphic state 'short middle chamber.' The Lactea-morph is diagnosed by a combination of 'long diaphragm' and 'short middle chamber,' whereof 'long diaphragm' is plesiomorphic and 'short middle chamber' diagnoses a monophyletic unit consisting of all forms except Macintoshi. The Fila-morph is diagnosed by a combination of 'spiral stylet' and 'short diaphragm,' whereof 'spiral stylet' is plesiomorphic and 'short diaphragm' diagnoses a monophyletic unit consisting of all forms except Lactea and Macintoshi. In a similar vein, combinations of two character states diagnose most of the phylomorphs, except the most plesiomorphic and the most apomorphic. This means that they may be arbitrarily delimited paraphyletic groups of monophyletic units in a morphological continuum. Such groups will lack distinct boundaries and, instead, graduate smoothly into each other via forms that are intermediate in precisely the characters states we use to distinguish them. Despite this, the organization helps us to organize the variation in the genus and it also provides a preliminary phylogenetic hypothesis for analyses on genetic data.

We allocate varieties into these phylomorphs using apomorphies in a consecutive sequence from either end of the network according to the scheme in Figure

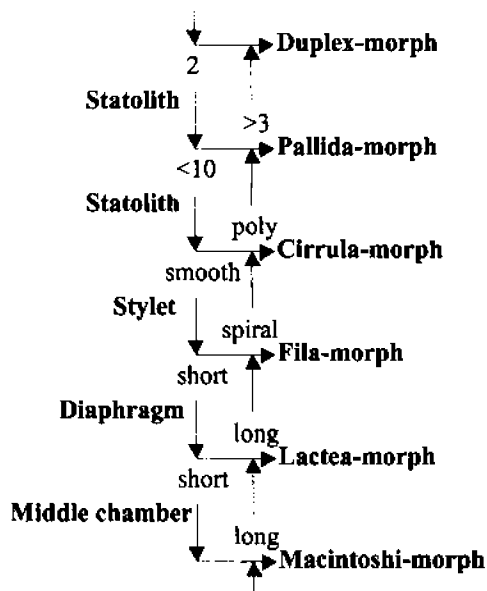


Figure 11. Phylogenetic key for allocation of *Ototyphlonemertes* varieties into six phylomorphs using four characters and five hypothesized character state transitions (see text for explanation). Start either from the top or the bottom. Starting from the top, check if they have statoliths consisting of two granules (on average within geographical varieties). If yes, go to the right and place it in the Duplex-morph. If no, go to the next step. Check if they have statoliths consisting of < 10 granules. If yes, go to the right and place it in the Pallida-morph, etc. The procedure is the same if we start from the bottom, except that we have to use the other state in each hypothesized transition. Check if they have long middle chamber. If yes, go to the right and place it in the Macintoshi-morph. If no, check if they have long diaphragm. If yes, place it in the Lactea-morph. If no, go to the next step, etc.

11. The root of the network is ambiguous. Envall's analysis suggested that it should be situated within the Duplex-morph, but later genetic data instead suggest that it should be situated within the Macintoshi-morph. However, as we distinguish both terminal morpho-types on character states that are apomorphic in respective phylogenetic hypotheses, and the rest on combinations of two plesiomorphic states, the position of the root is irrelevant for the function of the key. We get identical groups in both directions.

Summary

Ototyphlonemertes varieties are difficult to organize into typified assemblages like traditional species. The reason seems to be that the smallest monophyletic units are not diagnosable with morphological characteristics and diagnosable units are nested in each

other. The first makes it impossible to distinguish the smallest monophyletic units with morphological characteristics and the second makes it difficult to group varieties into separate sets of morphologically similar organisms like our traditional species. Assuming the root is located within the Macintoshi-morph, the monophyletic units are (1) the Duplex-morph (diagnosed by a statolith consisting of two granules); (2) the Duplex- and Pallida-morphs (diagnosed by a statolith with less than ten granules); (3) the Duplex-, Pallida- and Cirrula-morphs (diagnosed by a smooth stylet); (4) the Duplex-, Pallida- Cirrula-, and Fila-morphs (diagnosed by a short diaphragm); (5) the Duplex-, Pallida-, Cirrula-, Fila- and Lactea-morphs (diagnosed by a short middle chamber) and (6) the genus (diagnosed by an adult lack of eyes). These monophyletic units become less and less morphologically homogenous the more inclusive they get. The phylomorphs, on the other hand, are homogenous in most characters, but despite this, lack phylogenetically diagnostic traits (except for the most derived, probably the Duplex-morph). However, their morphological homogeneity is appealing for our intuitive wish to discern groups of similar organisms, and their assumed phylogenetic connections also represent the most resolved phylogenetic hypothesis we can obtain with morphological data. The arrangement thus produces both a phylogenetic kind of traditional species and null-hypothesis for future studies of the phylogenetic structure within *Otocyphlonemertes*.

Acknowledgements

This work was partially supported by NSF PEET award DEB 9712463, and constitutes contribution 516 from the Smithsonian Marine Station at Fort Pierce (SMSFP), 611 from the Smithsonian Caribbean Coral Reef and Ecosystems program (CCRE) at Carrie Bow Cay, Belize, and 237 from Northeastern University Marine Science Center (MSC). JLN extends his heartfelt thanks to N.W. "Pete" Riser for introducing him to this marvelous group of worms and for providing ad-

vice, insight and many specimens. JLN is grateful for a Sigma Xi Grant-in-Aid of Research, a Lerner-Gray Grant for Marine Research, a Theodore Roosevelt Memorial Grant, and a Friday Harbor Laboratories grant, all in support of work at Friday Harbor. JLN also is grateful for assistance from the staffs at MSC, University of Washington Friday Harbor Laboratories, SMSFP, CCRE, Smithsonian Tropical Research Institute in Panama, and especially to Barbara Littman of the Smithsonian Institution's National Museum of Natural History. ME especially would like to acknowledge the financial contributions from Rådman Ernst och fru Collianders Stiftelse. Both authors are grateful to the many colleagues who have supported their collecting efforts with food, drink, lodging and stimulating company.

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Appendix 1. Standardized protocol for *in vivo* examinations of *Ototyphlonemertes* specimens. The proposed character states are only suggestions

1	Sampling site	
2	Sampling depth	
3	Body length and width	
4	Epidermis color	Whitish/reddish brown
5	Reddish in brain region	Absent/present
6	Shape of anterior end	Rounded to pointed/teardrop shaped
7	Shape of posterior end	Rounded to pointed/widened/demarcated
8	Adhesive behavior	Continuous/caudal end
9	Cephalic furrow(s)	Absent/ in the statocyst region/in the midbrain region
10	Cirri	Absent/ in the hind end/in the cephalic region and in the hind end/over whole body
11	Anterior cirri: pattern and lengths	
12	Cerebral organs	Absent/pits/normally developed
13	Rhynchocoel length	
14	Proboscis anterior part length	
15	Proboscis papillae	Absent/flap-or knob-like/ finger-like cones/ with arborescent hairlike attachments/with rhabdites
16	Diaphragm length and width	Short/long
17	Gland secretions lateral to basis	Absent/present
18	No. Accessory stylet pouches	
19	No. Accessory stylets in each sack	
20	Direction of accessory stylets	Forward/backward/both
21	Middle chamber	Bulbous/squarish/tubular
22	Middle chamber length and width	
23	Posterior chamber	Opaque/vesicular/vesicular with specialized anterior region
24	Posterior chamber length	
25	Stylet	Smooth/ridged, structure of ridges
26	Stylet length and width	
27	Basis	Thin/thick
28	Basis length and width	
29	Statocyst	Spherical/ovoid
30	Statocyst maximum diameter	
31	Statolith, no. granules	2/3-10/type 1/type 2
32	Statolith maximum diameter	
33	Granule maximum diameter	
34	Intestinal diverticula	Shallow/deep
35	Parenchyma	Absent/medium/well-developed

Appendix 2. All recorded *Ototyphlonemertes* varieties and their qualitative character states. **1. Habitat:** 1. Tidal, 2. Subtidal; **2. Epidermis color:** 1. Whitish, 2. Reddish; **3. Reddish in the cerebral region:** 1. Absent, 2. Present; **4. Cephalic furrow at midbrain:** 1. Absent, 2. Present; **5. Cephalic furrow at or posterior to statocysts:** 1. Absent, 2. Present; **6. Cirri:** 1. Absent, 2. Hind end, 3. Hind end and cephalic region, 4. Whole body; **7. Cerebral organs:** 1. Absent, 2. Pits, 3. Normal; **8. Proboscis papillae:** 1. Absent or wart-like, 2. Finger-like, 3. Finger-like with hair, 4. Finger-like with rhabdites; **9. Diaphragm type:** 1. Short, 2. Long; **10. Gland secretions:** 1. Absent, 2. Present; **11. Accessory stylet pouches number:** 1. Two, 2. Three; **12. Accessory stylet pouches position:** 1. Lateral to stylet, 2. Posterior to stylet; **13. Accessory stylets direction:** 1. Backward, 2. Forward, 3. Both; **14. Middle chamber structure:** 1. Bulbous, 2. Elongate, 3. Square, 4. Tubular; **15. Posterior chamber structure:** 1. Granular, 2. Vesicular, 3. Granular with specialized anterior region; **16. Stylet structure:** 1. Smooth, 2. Ridged; **17. Ridged stylet structure:** 1. Two straight strands, 2. Two or three twined strands, 3. Crossing ridges; **18. Statolith type:** 1. Two-partite, 2. Three-partite, 3. Four-partite, 4. Variably 3-10, 5. Type 1 morula, 6. Type 1.5, 7. Type 2, 8. Compounded; **19. Intestine type:** 1. Sack-like with shallow diverticula, 2. Stacked platelets with deep diverticula. The label of the varieties refers to phylomorph (C, D, F, etc.) and sampling locale. Geographic abbreviations are: B = Spain, Barcelona; Bb = USA, Florida, South Hutchinson Island, Bathub Beach; BM = France, Banyuls sur Mer; Bu = Bulgaria; CBC = Belize, Carrie Bow Cay; Cr = Croatia, Rovinj; Dai = Japan, Hokkaido, Daikoku Island; Dan = Belize, Dangriga; FH = USA, Washington, Friday Harbor; FK = USA, Florida, Florida Keys (Pidgeon Key and Tennessee Reef); FP = USA, Florida, Fort Pierce (Fort Pierce Inlet, Pierce Shoal and Capron Shoals); Gal = Galapagos; Hw = USA, Hawaii archipelago; Ges = Japan, Okinawa, Gesashe; LP = USA, Maine, Liberty Pt.; m = USA, Florida, Miami (Biscayne Bay and Miami South Beach); Man = USA, Massachusetts, Ellenville, Manomet; N = France, Nice (Antibes, Menton); Na = Italy, Naples (Bay of Naples and Island of Ischia); PB = USA, Florida, Palm Beach; PGB = Russia, Vladivostok, Peter the Great Bay; R = France, Roscoff; San = USA, Florida, Sanibel Island; Sar = USA, Florida, Sarasota (Ana-Maria Island); SB = West Indies, Saint Barthélemy; SD = USA, California, San Diego; Ses = Japan, Okinawa, Sesoko; Set = Japan, Honshu, Seto; SM = France, Saint-Malo; SS = Brazil, Santos, Island of São Sebastião; Sw = Sweden, Tjörn; Vb = UK, Gorran Haven, Vault beach; WH = USA, Massachusetts, Woods Hole (1 marks descriptions in Mock & Schmidt, 1975). To compare a sampled variety with the recorded varieties you should: 1. Allocate the variety to a phylomorph using the phylogenetic key. 2. Compare character states with recorded varieties of the phylomorph in the region. 3. Go to the nemertean homepage (<http://nemertes.si.edu>) and check remaining characters

Variety	Species	Distribution	Coordinates	1																		
				1	2	3	4	5	6	7	8	9	0	1	2	3	4	5	6	7	8	9
C-SS1	<i>O. brevis</i>	Atlantic, SW	23° 51' S, 45° 20' W	1			1	2	3	1	2	1	1	1	1	1	1	1	1	1	5	
C-Gal1	<i>O. cirrula</i>	Pacific E	0° 31' S, 90° 46' W	1	1	2			4	2	2	1	2	1		3	1			1	7	
C-Set1		Pacific NW	33° 42' N, 135° 20' E	1	1	1	1	2	1	1		1		1	1	4	1	1	6	1		
D-Sw1	<i>O. corruca</i>	Atlantic NE	58° 52' N, 11° 10' E	1	1	2	1	2	3	3	1	1	1	1	1	3	1	1	1	1	1	
D-Vb1		Atlantic NE	50° 14' N, 4° 47' W	1	1	2	1	2	3	3	1	1	1	1	1	3	1	1	1	1	1	
D-SS1	<i>O. evelinae</i>	Atlantic SW	23° 51' S, 45° 20' W	1	1/2	1	1	2	3	3	1	1	1	1	1	3	1	1	1	1	1	
D-Bb1		Atlantic W	27° 20' N, 80° 16' W	1	1	2	1	2	3	3	1	1	2	1	1	3	1	1	1	1	1	
D-M1		Atlantic W	25° 47' N, 80° 07' W	1	1	1/2	1	2	3	3	1	1	1	1	1	3	1	1	1	1	1	
D-FK1		Atlantic W	24° 42' N, 81° 09' W	1	1		1	2	3	3	1	1	1	1	1	3	1	1	1	1	1	
D-Bu1	<i>O. antipai</i>	Black Sea W	43° 23' N, 28° 29' E	1	1	2	1	2	3	3	1	1	1	1	1	1	1	1	1	1	1	
D-SB1		Caribbean E	17° 37' N, 61° 46' W	1	1	2	1	2	3	3	1	1	2	1	1	3	1	1	1	1	1	
D-CBC1		Caribbean W	16° 48' N, 88° 04' W	1	1	1	1	2	3	3		1	1		1	1	1	1	1	1	1	
D-Dan1		Caribbean W	17° 00' N, 88° 14' W	1	1		1	3	3	1	1	1	1		1	1	1	1	1	1	1	
D-Sar1		Gulf of Mexico E	27° 21' N, 82° 34' W	1	1	1/2	1	2	3	3	1	1	2	1	1	3	1	1	1	1	1	
D-San1		Gulf of Mexico E	26° 26' N, 82° 06' W	1	1	1	1	2	2	3	1	1	1	1	1	3	1	1	1	1	1	
D-BM1	<i>O. aurantiaca</i>	Mediterranean N	42° 30' N, 3° 10' E	1	1/2	1	1	1	3	3	1	1	2	1	1	3	1	1	1	1	1	
D-N1		Mediterranean N	43° 40' N, 7° 20' E	1	2	2	1	2	3	3	1	1	1	1	3	1	1	1	1	1	1	
D-Na1	<i>O. duplex</i>	Mediterranean N	40° 42' N, 13° 56' E	1	1	1/2	1	2	3	3	1	1	1	1	1	3	1	1	1	1	1	
D-Na2	<i>O. brunnea</i>	Mediterranean N	40° 49' N, 14° 14' E	1	2						3	1	1	1		1	1	1	1	1	1	
D-B1		Mediterranean W	41° 30' N, 2° 09' E	1	1	2	1	2	3	3	1	1	2	1	1	3	1	1	1	1	1	
D-Hw1		Pacific Central	21° 30' N, 158° 00' W	1	1	2	1	2	3	3	1	1	1	1	1	1	1	1	1	1	1	
D-Ses1		Pacific W	26° 28' N, 127° 57' E	1	1	2	1	2	3	3	1	1	1	1	1	3	1	1	1	1	1	
D-PGB1	<i>O. martynovi</i>	Sea of Japan	43° 07' N, 131° 56' E	1	1	2	1	2	3	3	1	1	1	1	1	1	1	1	1	1	1	
F-FP1		Atlantic W	27° 28' N, 80° 17' W	1	1	1	1	2	3	3	1	1	1	1	1	2	1	3	2	3	5	
F-Bb1		Atlantic W	27° 20' N, 80° 16' W	1	1	1	1	2	3	3	1	1	1	1	1	2	1	3	2	3	5	
F-PB1		Atlantic W	26° 42' N, 80° 02' W	1	1	1	1	2	3	3	1	1	1	1	1	3	1	3	2	3	5	
F-M1		Atlantic W	25° 47' N, 80° 07' W	1	1	1	1	2	3	3	1	1	1	1	1	2	1	3	2	3	5	
F-SS1	<i>O. flta</i>	Atlantic SW	23° 51' S, 45° 20' W	1	1						3	3	1	1	1	1	2		2	3	5	

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Variety	Species	Distribution	Coordinates	1																		
				1	2	3	4	5	6	7	8	9	0	1	2	3	4	5	6	7	8	9
F-SS2	<i>O. parvula</i>	Atlantic SW	23° 51' S, 45° 20' W	1	1	2	1	3	2	4	1	1	1	1	1	1	2	3	5			
F-SB1		Caribbean E	17° 37' N, 61° 46' W	1	1	1	1	2	3	3	1	1	1	1	1	1	3	2	3	5	2	
F-Sar1		Gulf of Mexico E	27° 21' N, 82° 34' W	1	1	2	1	2	3	3	1	1	1	1	2	1	3	2	3	5	2	
F-San1		Gulf of Mexico E	26° 26' N, 8° 06' W	1	1	1	1	2	3	3	1	1	1	1	3	1	3	2	3	5	2	
F-Gal1 ¹		Pacific E	0° 31' S, 90° 46' W	1	1	1	1	2	3	3	1	1	1	1	3	1	2	3	5	2		
F-Hw1		Pacific Central	21° 35' N, 158° 07' W	1	1	2	1	2	3	3	1	1	1	1	3	1	3	2	3	5		
F-Hw2		Pacific Central	19° 57' N, 155° 52' W	1	1	1	1	2	3	3	1	1	1	1	1	1	2	3	5			
F-Ges1		Pacific W	26° 36' N, 128° 09' E	1	1	2	1	2	3	3	1	1	1	1	3	1	3	2	3	5	2	
F-Set1		Pacific NW	33° 42' N, 135° 20' E	1	1	2	1	2	3	3	1	1	1	1	3	1	3	2	2	5	2	
L-LP1		Atlantic NW	42° 25' N, 70° 54' W	1/2	1	1	1	2	3	1	2	1	1	2	1	2	2		7			
L-FP1		Atlantic W	27° 28' N, 80° 17' W	1	1	2	1	2	1	1	2	1	1	1	3	2	2	2	7			
L-PB1		Atlantic W	26° 42' N, 80° 02' W	1	1	1	1	2	2	1	1	2	1	1	1	3	2	2	2	6	1	
L-SS1	<i>O. luacea</i>	Atlantic SW	23° 51' S, 45° 20' W	1	1	2	1	3	1	1	2	1	1	1	3	2	2	3	5	1		
L-SB1		Caribbean E	17° 37' N, 61° 46' W	1	1	2	1	2	2	1	1	2	1	1	1	3	2	2	2	6		
L-CBC1		Caribbean W	16° 48' N, 88° 04' W	1	1	2		2	2	1	1	2	1	1	1	3	2	2	2	6	1	
L-CBC2		Caribbean W	16° 48' N, 88° 04' W	1	1	1		2	3	1	2		1	1	3		2	3	6			
L-FH1	<i>O. americana</i>	Pacific NE	48° 32' N, 123° 05' W	1	1	1		2	1	1	1	2	1	1	1	1	2	2	2	7	1	
L-Ses1		Pacific W	26° 28' N, 127° 57' E	1	1	2	1	2	1	1	1	2	1	1	1	3	2	2	2	7	1	
L-Dai1		Pacific NW	42° 56' N, 144° 53' E	1	1	1	1	2	1	1	1	2	1	1	1	2	2	1	2	3	5	1
M-R1		Atlantic NE	48° 41' N, 4° 09' W	1	1	1	1	2	1	1	1	2	1	1	1	3	4	1	2	3	7	1
M-Vb1		Atlantic NE	50° 14' N, 4° 47' W	1	1	1		2	1	1	1	2	1	1	1	3	4	1	2	3	7	1
M-WH1	<i>O. pellucida</i>	Atlantic NW	41° 31' N, 70° 40' W		1	2				2	1		1		4			2	3	5		
M-Man1		Atlantic NW	41° 53' N, 70° 32' W	1	1	1		2	3	1	1	2	1	2	1	4	2	2	2	6	2	
M-Bb1		Atlantic W	27° 20' N, 80° 16' W	1	1	2	1/2	2	2	1	1	2	1	1	1	4	2	2	2/3	6/7	1	
M-M1		Atlantic W	25° 47' N, 80° 07' W	1	1	2	1	2	2	1	1	2	1	1	1	4	2	2	2	7	1	
M-M2		Atlantic W	25° 47' N, 80° 07' W	1	1	1	1	1	1	1	1	2	1	1	2	2	4		3	8		
M-FK1		Atlantic W	24° 42' N, 81° 09' W	1	1	2	1	1	2	1	1	2	1	1	1	4		2	2	7/8	1	
M-SB1		Caribbean E	17° 37' N, 61° 46' W	1	1	2	1	2	2	1	1	2	1	1	1	4	2	2	2	6	1	
M-CBC1		Caribbean W	16° 48' N, 88° 04' W	1	1	2	1	2	2	1	1	2	1	1	1	4	2	2	2	7	1	
M-CBC2		Caribbean W	16° 48' N, 88° 04' W	2	1	2		1		1	1	1	1	1	1	4		2	3	7		
M-CBC3		Caribbean W	16° 48' N, 88° 04' W	2	1	2	2	2	1	1	1	2	1	1	2	3	4	1	2	2	7	
M-Na1	<i>O. macintoshi</i>	Mediterranean N	40° 49' N, 14° 14' E		1					1	1	2		1	4		2	3	7			
M-N1		Mediterranean N	43° 40' N, 7° 20' E	1	1	2	1	2	1	1	1	2	1	1	1	3	4	2	2	3	7	1
M-FH1		Pacific NE	48° 32' N, 123° 05' W	1	2	1	1	2	1	1	1	2	1	1	2	2	4	1/2	2	3	6	1
M-SD1	<i>O. spiralis</i>	Pacific NE			2					3	1		1	3	4		2	3	5			
M-Gal1 ¹	spec.1	Pacific E	0° 31' S, 90° 46' W	1	1	1	1	1	1/2	1	1	2	1	1	1	4		2	2	5		
M-Gal2 ¹	spec.2	Pacific E	0° 31' S, 90° 46' W	1	1	1	1	1	1/2	1	1	2	1	1	1	4		2	2	5		
M-Hw1		Pacific Central	21° 4' N, 158° 1' W	1	1	1	1	2	1	1	1	2	1	1	2	4		2	7	1		
M-Set1		Pacific NW	33° 42' N, 135° 20' E	1	1	1	1	2	2	1	1	2	1	1	1	1	4	2	2	3	7	1
M-Set2		Pacific NW	33° 42' N, 135° 20' E	1	1	1	1	2	1	1	1	2	1	1	2	1	4	2	2	2	7	1
M-PGB1	<i>O. nikolaii</i>	Pacific NW	43° 07' N, 131° 56' E	1	1	1		2	1	1	1	2	1	1	2	4		2	1	7		
P-SM1	<i>O. pallida</i>	Atlantic NE	48° 38' N, 2° 04' W	1	1	1	1	2	3	3	2	1	2	1	1	3	1	1	1	2	1	
P-WH1		Atlantic NW	41° 24' N, 70° 31' W	1	1	1	1	2	3	1	3	1	2	1	1	3	1	1	1	2	1	
P-Man1		Atlantic NW	41° 54' N, 70° 32' W	1	1	1	1	2	3	2	3	1	2	1	1	3	1	1	1	2		
P-Bb1		Atlantic W	27° 20' N, 80° 16' W	1	2	2	1	2	3	3	4	1	2	1	1	3	1	1	1	2	1/2	
P-FP1		Atlantic W	27° 28' N, 80° 17' W	2	1	1	1	2	3	3	2	1	2	1	1	3	1	1	1	3	1	
P-FP4		Atlantic W	27° 28' N, 80° 17' W	2	1	2	1	2	3	3	2	1	2	1	1	3	1	1	1	2	1	
P-FP5		Atlantic W	27° 28' N, 80° 17' W	2	1	2	1	2	3	3	2	1	1	1	1	3	1	1	1	4		

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Appendix 2. contd.

Variety	Species	Distribution	Coordinates	I																	
				1	2	3	4	5	6	7	8	9	0	1	2	3	4	5	6	7	8
P-M1		Atlantic W	25° 47' N, 80° 07' W	1	2	2	1	2	3	3	2	1	1	1	1	3	1	1	1	2	1
P-FK1		Atlantic W	24° 42' N, 81° 09' W	2	2	2		2	3	3	2	1	1	1	1	3	1	1	1	2	1
P-FK2		Atlantic W	24° 42' N, 81° 09' W	1	2	2	1	2	3	3	4	1		1	1	3	1	1	1	2	1
P-FK3		Atlantic W	24° 42' N, 81° 09' W	2	1	1	1	2	3	3	2	1	1	1	1	3	1	1	1	3	
P-SS1	<i>O. erneba</i>	Atlantic SW	23° 51' S, 45° 20' W	1		1	2	3	3	4	1	2	1	1	3	1		1	2		
P-CBC1		Caribbean W	16° 48' N, 88° 04' W	1	1	1	1	2	3	3	2	1	2	1	1	3	1	1	1	1/2	1
P-CBC2		Caribbean W	16° 48' N, 88° 04' W	1/2	1	2	1	2	3	3	2	1	1/2	1	1	3	1	1	1	4	1
P-CBC3		Caribbean W	16° 48' N, 88° 04' W	1/2	1	1	1	2	3	3	4	1	1	1	1	3	1	1	1	2	1
P-CBC4		Caribbean W	16° 48' N, 88° 04' W	2	2	2	2	2	3	3	2	1	1	1	1	3	1	1	1	2	
P-CBC6		Caribbean W	16° 48' N, 88° 04' W	2	1	2	1	2	3	3	2	1	2	1	1	3	1	1	1	3	1
P-CBC8		Caribbean W	16° 48' N, 88° 04' W		1	2			3	1	2	1		1	1		1		1	2	
P-Cr1	<i>O. esulcata</i>	Mediterranean N	45° 03' N, 13° 38' E	1	1				3		1				1		1		3		
P-FH1		Pacific NE	48° 32' N, 123° 05' W	1	1	1	2	2	3	3	2	1	1	1	1	3	1	1	1	4	1
P-FH2		Pacific NE	48° 32' N, 123° 05' W	2	1		2	2	3	3	2	1		1	1	3	1		1	3	1
P-Gal1 ¹		Pacific E	0° 31' S, 90° 46' W	1	2	2	1	2	3	3	2	1	2	1	1	3	1	1	1	2	1
P-Gal2	<i>O. santacruzensis</i>	Pacific E	0° 31' S, 90° 46' W	1	1		1	2	3	3	2	1	2	1	1	3	1	1	1	3	1
P-Hw1		Pacific Central	21° 28' N, 157° 46' W	1	1	2	1	2	3	2	2	1	1	1	1	3	1	1	1	2	1
P-Hw2		Pacific Central	21° 35' N, 158° 07' W	1	1	2	1	2	3	3	2	1	1	1	1	3	1	1	1	2	
P-Ses1		Pacific W	26° 28' N, 127° 57' E	1	1	2	1	2	3	2	2	1	1	1	1	3	1	1	1	2/3	1
P-Set1		Pacific W	33° 42' N, 135° 20' E	1	1	1	1	2	3	3	2	1		1	1	3	1	1	1	3	1