





Oncholaimus tripapillatus sp. nov., a New Free-Living Marine Nematode of the Genus *Oncholaimus* Dujardin, 1845 (Nematoda: Enoplida: Oncholaimidae) from the Subtidal Sediment of Dokdo Island, East Sea, Korea, with a New Record of *O. qingdaoensis* Zhang & Platt, 1983

Hyojin Lee D, Heegab Lee and Hyunsoo Rho *D

East Sea Environment Research Center, Korea Institute of Ocean Science & Technology (KIOST), Uljin 36315, Korea; hjlee086@kiost.ac.kr (H.L.); leehg@kiost.ac.kr (H.L.)

* Correspondence: hsrho@kiost.ac.kr; Tel.: +82-54-780-5345; Fax: +82-54-780-5349



Citation: Lee, H.; Lee, H.; Rho, H. Oncholaimus tripapillatus sp. nov., a New Free-Living Marine Nematode of the Genus Oncholaimus Dujardin, 1845 (Nematoda: Enoplida: Oncholaimidae) from the Subtidal Sediment of Dokdo Island, East Sea, Korea, with a New Record of O. qingdaoensis Zhang & Platt, 1983. J. Mar. Sci. Eng. 2021, 9, 1334. https:// doi.org/10.3390/jmse9121334

Academic Editor: Francesco Tiralongo and Anna Di Cosmo

Received: 28 October 2021 Accepted: 25 November 2021 Published: 27 November 2021

Publisher's Note: MDPI stays neutral with regard to jurisdictional claims in published maps and institutional affiliations.



Copyright: © 2021 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (https:// creativecommons.org/licenses/by/ 4.0/). Abstract: During a survey of the free-living marine nematodes of Korea, two species belonging to the genus Oncholaimus Dujardin, 1845 were discovered. A new species, Oncholaimus tripapillatus sp. nov. and a newly recorded species, Oncholaimus qingdaoensis Zhang & Platt, 1983, are reported. Oncholaimus tripapillatus sp. nov. was collected from the washing of subtidal coarse sediments around Dokdo Island of the East Sea, Korea. Oncholaimus tripapillatus sp. nov. is characterized by a relatively long (4071–4435 µm in males and 4514–4661 µm in females) and slender body, a slightly constricted head region, relatively long cephalic setae (10–12 µm), males having a precloacal sexual protuberance bearing two small cone-shaped supplementary spines, five pairs of long cloacal setae (three pairs of precloacal and two pairs of postcloacal setae in subventral position), and three remarkable papillae near the end of the tail, with two pairs of subventral setae. The Korean specimens of Oncholaimus qingdaoensis Zhang & Platt, 1983 are almost identical to the Chinese specimens of the original description from the intertidal sand of Qingdao, China. However, the Korean specimens differ from the Chinese specimens in the longer body length in males (3379–3715 μ m vs. 2380–2640 μ m), the larger spicule length (47–52 µm vs. 34–36 µm), and the presence of ventral tail papillae situated around 14–16 µm from the tail tip. Detailed morphological features and illustrations of two Oncholaimus species from Korea were obtained by differential interference contrast microscopy and scanning electron microscopy. A pictorial key to the species group with distinct tail papillae among the genus Oncholaimus is also provided.

Keywords: taxonomy; marine nematode; Oncholaimus; Dokdo; the East Sea; Korea

1. Introduction

The genus *Oncholaimus* Dujardin, 1845 is distinguished from the other genera of the family by the presence of the largest left ventrosublateral tooth, relatively short spicules, and a monodelphic–prodelphic reproductive system in females [1]. The genus *Oncholaimus* shows worldwide distribution and occurs in various habitats, from littoral sediment to brackish water and deep-sea environments [2,3]. Thus far, about 120 nominated species of the genus *Oncholaimus* have been described across the world [4]. Of these, 24 species of the genus *Oncholaimus* have been described from the northwest Pacific Ocean as follows: 16 species (*O. asiaticus* (Belogurov et. al., 1980), *O. compositus* Belogurov et. al., 1980, *O. eximius* Belogurova, 1978, *O. fucus* (Smolyanko & Belogurov, 1991), *O. furugelmus* (Belogurov, 1977), *O. japonicus* (Belogurov & Belogurov, 1981), *O. olium* (Belogurov et. al., 1975), *O. paracampylocercoides* Smolyanko & Belogurova, 1978, *O. ramosum* (Smolyanko & Belogurova, 1978), *O. natures* (Belogurov et. al., 1978), *O. natures* (Belogurov et. al., 1978), *O. nature*

1972), *O. venustus* (Belogurov et. al., 1972), *O. vesicarius* (Wieser, 1953), *O. zinus* Belogurov & Belogurova, 1978) from Shikotan Island, Kuril Islands, and its adjacent Russian waters of the East Sea (Sea of Japan) [5–8]; two species (*O. dujardinii* de Man, 1876 and *O. secundicollis* Shimada et. al., 2009) from Wakayama and Hokkaido of Japan [9,10]; six species (*O. minor* Chen & Guo, 2014, *O. multisetosus* Huang & Zhang, 2006, *O. qingdaoensis* Zhang & Platt, 1983, *O. sinensis* Zhang & Platt, 1983, *O. xiamenense* Chen & Guo, 2014, and *O. zhangi* Gao & Huang, 2017) from the East China Sea and the Yellow Sea of China [11–13]. Of these, only one *Oncholaimus* species, *O. secundicollis*, has been reported from the rocky intertidal seagrass habitat on the eastern coast of Korea [10,14].

During a continuous ecological investigation on the biodiversity of the free-living marine nematodes around Korean waters, we found two *Oncholaimus* species from the intertidal and subtidal sediments of the eastern, southern, and western coasts of Korea. The present paper deals with morphological descriptions, illustrations, and photomicrographs from differential interference contrast (DIC) microscopy and scanning electron microscopy. Moreover, we also provide a pictorial key and a comparison table of the diagnostic morphological characteristics of the species within the genus *Oncholaimus* with distinct tail papillae in the tail region.

2. Materials and Methods

2.1. Sampling of Taxa

The marine nematodes were obtained from the upper surface of the intertidal sediment from the southern and western coasts of Korea using a hand scoop, as well as by means of a Smith–McIntyre grab from the shallow subtidal benthic environment sediment of Dokdo Islands, located in the East Sea in the northwest Pacific Ocean.

2.2. Sample Processing and Preparation of the Specimens

The samples were filtered through a sieve with 67 µm mesh in the field after rinsing with freshwater for less than 1 min for osmotic shock and then fixed in 5% formalin [15]. In the laboratory, the nematodes were sorted from the mixed meiobenthos under a LEICA 205 C stereomicroscope (Leica, Wetzlar, Germany). The nematodes were transferred to glycerol and mounted between two cover slips on an HS slide for morphological observations [16]. The specimens were measured, examined, and drawn using Nomarski differential interference contrast (DIC) microscopy with an Olympus BX53 microscope equipped with a drawing tube and an Olympus DP26 digital camera with the corresponding Olympus CellSens imaging software (Olympus, Tokyo, Japan). The specimens selected for scanning electron microscopy (SEM) were fixed in 5% buffered formalin and then rinsed twice with distilled water to remove the buffered formalin. After rinsing for 5 min each, the specimens were freeze-dried in a cooling stage, mounted on an aluminum stub, sputter-coated with gold/palladium in a high-vacuum evaporator, and examined with an SEC SNE-3200M Desktop Mini SEM.

2.3. Terminology and Abbreviations

Measurements are in micrometers (μ m). The abbreviations used in the text are as follows: a, body length divided by maximum body diameter; b, body length divided by pharynx length; c, body length divided by tail length; V (%), vulva distance from anterior end as percentage of total body length.

3. Results

3.1. Description of Oncholaimus tripapillatus sp. nov.

Class Enoplea Inglis, 1983 Order Enoplida Filipjev, 1929 Family Oncholaimidae Filipjev, 1916 Genus *Oncholaimus* Dujardin, 1845 *Oncholaimus tripapillatus* sp. nov. (Figures 1–4; Table 1)



Figure 1. *Oncholaimus tripapillatus* sp. nov.—holotype male in lateral view: (**A**) habitus; (**B**) anterior region; (**C**) head region; (**D**) spicule and tail region; (**E**) spicule and tail region in paratype (scale bars: (**A**) = 100 μ m; (**B**) = 20 μ m; (**C**–**E**) = 10 μ m).

4 of 17



Figure 2. *Oncholaimus tripapillatus* sp. nov.—paratype female in lateral view: (**A**) habitus; (**B**) vulva region; (**C**) head region; (**D**) tail region; (**E**) female demanian system (scale bars: (**A**,**E**) = 100 μ m; (**B**) = 50 μ m; (**C**,**D**) = 10 μ m).



Figure 3. *Oncholaimus tripapillatus* sp. nov.—SEM photomicrographs of a male (**A**,**C**,**D**) and a female (**B**): (**A**) head region showing amphideal fovea; (**B**) six low lips and cephalic setae region; (**C**) cloacal region showing precloacal sexual protuberance and precloacal setae (arrow); (**D**) tail region showing three papillae (arrow) and subventral setae (scale bars: $(A-D) = 15 \mu m$).



Figure 4. *Oncholaimus tripapillatus* sp. nov.—DIC photomicrographs, showing a lateral view of a male: (**A**) head region; (**B**) head region showing amphideal fovea (arrow); (**C**) cloacal region showing precloacal spines (arrow); (**D**) tail region showing three papillae; (**E**) spicules and tail region (scale bars: $(A-E) = 20 \mu m$).

	Holotype			Paratypes		
Characters	1റ്	1 ♂	2 ♂	1 9	2 ♀	3 ♀
Total body length	4281	4435	4071	4514	4661	4520
a	82	86	83	70	64	72
b	8	8	8	7	8	8
с	46	48	42	41	41	41
Head diameter at the level of cephalic setae	30	30	30	31	32	30
Body diameter at the level of cardia	42	42	43	52	54	46
Maximum body diameter	52	51	49	65	73	63
Cephalic setae length	10	11	12	11	11	11
Amphideal fovea diameter	9	8	11	10	9	10
Amphideal fovea length	5	6	7	6	6	5
Buccal cavity diameter	19	20	20	20	21	22
Buccal cavity length	29	31	31	31	34	33
Anterior end to the largest tooth tip	7	8	7	9	8	9
Anterior end to the other teeth tips	14	16	14	18	15	17
Anterior end to amphids	22	23	25	22	22	23
Anterior end to excretory pores	88	80	85	95	87	91
Anterior end to the nerve ring	251	254	247	274	271	264
Pharynx length	563	548	545	606	602	589
Spicule length	44	42	41	-	-	-
Cloacal setae length	8	7	7	-	-	-
Anterior end to the vulva	-	-	-	2987	3107	2968
Body diameter at level of the vulva	-	-	-	61	70	61
V (%)	-	-	-	66	67	66
Anal body diameter	35	36	37	37	36	35
Tail length	94	92	96	110	114	111
Tail length/anal body diameter	2.7	2.5	2.6	3	3.2	3.1

Table 1. Morphometrics of Oncholaimus tripapillatus sp. nov. (in µm).

3.1.1. Type Material

A holotype male (MABIK NA00156666), in glycerin on an HS slide, was deposited in the nematode collection at the specimen conservation room of the Marine Biodiversity Institute of Korea (MABIK), Seochun, Korea. Two paratype males (KIOST NEM-1-1187 and KIOST NEM-1-1201) and three females (KIOST NEM-1-1211, KIOST NEM-1-1221, and KIOST NEM-1-1225), mounted on HS slides, were deposited in the nematode collection at the specimen conservation room of the Bio-Resources Bank of Marine Nematodes (BRBNM), East Sea Research Institute, Korea Institute of Ocean Science and Technology (KIOST), Korea.

3.1.2. Type Locality and Habitat

The locality was the subtidal zone of Dokdo Islands ($37^{\circ}14'21.00''$ N, $131^{\circ}52'4.00''$ E), Ulleung-gun, Gyeongsangbuk-do, Korea, where the specimens were collected on 22 May 2015 by H. S. Rho. The nematodes were extracted from the subtidal sediments with tiny shell gravels, and detritus was collected at a depth of 5 m.

3.1.3. Etymology

This species' name, *tripapillatus*, comes from the Latin, meaning crested, in reference to the three pairs of papillae on the tail tip.

3.1.4. Measurements

See Table 1 for measurements and morphometrics.

3.1.5. Diagnosis

The body is relatively long (4071–4435 μ m in males and 4514–4661 μ m in females) and slender (a = 82–88 in males and 64–72 in females), with a slightly constricted head, relatively long cephalic setae (10–12 μ m), a large buccal cavity with a thick cuticular wall and three teeth (the left subventral tooth is larger than the right subventral and dorsal teeth), two pre-cloacal supplementary spines in males, five pairs of long cloacal setae (three precloacal and two postcloacal setae in the subventral position), a ventrally arcuated conoid tail with three tail papillae and two pairs of short setae.

3.1.6. Description

Males: Bodies are comparatively long (4071–4435 µm) and slender, slightly tapering toward both extremities (Figure 1A). Maximum body diameter at the mid body level is 49–52 μ m. The cuticle of the body is smooth. The diameter of the head is 30 μ m wide, with a slight constriction posterior to the cephalic seta. The lip region has six conical lips, which are deeply separated, each bearing small rounded inner labial papillae. Six outer labial and four cephalic setae are arranged in a single circle, almost equal in size (10–12 μ m) (Figures 1C, 3A and 4A). The buccal cavity is large (29–31 µm long and 19–20 µm wide), with sclerotized walls and three teeth. The left subventral tooth (22–24 μ m long) is larger than the right subventral and dorsal teeth, and the tip is stretched up to the cephalic setae. The amphideal fovea (8–11 μ m wide) is pocket-like, with an elliptical opening that is located 20–25 μ m from the anterior end (Figures 1C and 4B). The pharynx (545–563 μ m long) is cylindrical and is approximately 12–13% of total body length, with a corresponding body diameter of 42–43 µm. The excretory pore is situated 80–88 µm from the anterior end. The nerve ring is approximately 5.7–6.1% of the total body length from the anterior end. The somatic setae are randomly distributed across the surface of the body (Figure 1B). Two opposed testes are situated to the right side of the intestine. The spicules are short and equal, slightly ventrally curved, with a pointed distal end and a cephalated proximal end. The spicules (41–44 μ m long) are approximately 1.1–1.2 times of the anal body diameter (Figures 1D and 4E). Gubernacula are absent. Five pairs of long and stout subventral setae (7–8 µm long) are located around the cloacal region, of which three pairs are situated on each side of the cloacal opening, while the other two pairs are located on both sides behind the cloacal region (Figure 1D,E and Figure 3C). A precloacal sexual protuberance is present just anterior to the cloacal opening, bearing two small cone-shaped supplementary spines situated on the cloacal protuberance (Figure 4C). The tail is 92–96 μ m in length, conoid, slightly curved ventrally, and approximately 2.5–2.7 times the anal body diameter (Figure 1D,E and Figure 3C). Three remarkable papillae are subventrally situated on the tail tip, of which only the anteriormost papilla consists of a pair. There are two pairs of subventral setae around the tail papillae (Figures 3D and 4D).

Females: Similar to the males in general appearance, but they differ in tail shape and sexual characteristics (Figure 2A,C and Figure 3B). The conical tail (110–114 μ m long) is slightly arcuate with a blunt posterior end but without a tail papilla. The tail is 3.0–3.2 times the anal body diameter, with two pairs of short terminal setae (Figure 2D). The reproductive system is monodelphic with anterior reproductive branches, lying to the right side of the intestines. They have two intra-uterine eggs, 124–169 μ m in length and 56–67 μ m in width. The vulva is situated at 66%–67% of the total body length from the anterior end (Figure 2B). The demanian system is composed of one osmosium connecting the ductus entericus and the intestine, and one uvette, located at 170 μ m in front of anus, connecting the ductus uterinus with a main tube (Figure 2E).

3.1.7. Differential Diagnosis and Relationships

The genus *Oncholaimus* Dujardin, 1845 is the largest genus of the family Oncholaimidae Filipjev, 1916, containing about 120 species, which have thus far been described from various localities of littoral and deep-sea habitats in oceans all over the world [2,4]. They are distinguished from the other genera of the family by the presence of the largest left ventrosublateral tooth and relatively short spicules, with a monodelphic-prodelphic reproductive system in females [1]. After comparing and compiling the valid species of the genus Oncholaimus on the basis of the taxonomic key characteristics among the congeners, we artificially distinguished a group of 26 species with a conspicuous ventral papilla on the tail, which is the most important diagnostic characteristic of the genus, including the present new species, Oncholaimus tripapillatus sp. nov., and the following: O. aquaedulcis Schneider, 1937; O. bajulus Paramonov, 1937; O. balli Nicholas & Stewart, 1984; O. cobbi (Kreis, 1932); O. deconincki Heyns & Coomans, 1977; O. dimorphicus Tahseen, Siddiqi & Mustaqim, 2016; O. domesticus (Chitwood & Chitwood, 1938); O. hyrcanus Tchesunov, 1979; O. japonicus (Belogurov & Belogurova, 1981); O. longispiculosus Gerlach, 1955; O. longus (Wieser, 1953); O. martini Wieser, 1959; O. menzeli Schneider, 1937; O. oxyuris Ditlevsen, 1911; O. paraoxyuris Salma, Nasira, Saima & Shahina, 2017; O. paroxyuris Schuurmans Stekhoven, 1942; O. qingdaoensis Zhang & Platt, 1983; O. rionegrensis Pastor de Ward et. al., 2013; O. rivalis Gagarin & Gusakov, 2012; O. sahariensis Coomans & Heyns, 1983; O. sidiiqii Tauheed & Ahmad, 2016; Oncholaimus tripapillatus sp. nov.; O. vesicarius (Wieser, 1959); O. vietnamicus Gagarin & Thanh, 2011; O. xiamenense Chen & Guo, 2014; O. zhangi Gao & Huang, 2016. Therefore, we herein provide a pictorial identification key and a comparison table of the diagnostic morphological characteristics for the species group with a conspicuous ventral papilla on the tail, with diagnostic characteristics also including the morphology of the anterior head region and the posterior tail region showing spicule and gubernaculum, which are generally important taxonomic key characters to discriminate closely related nematode species (Figure 5, Table 2).

Table 2. Comparison of diagnostic morphological characteristics of the species group with remarkable tail papillae within the genus *Oncholaimus*. Males only, morphometric values rounded (-: unknown).

	Characters						
Species	Body Length (µm)	a	b	с	Buccal Cavity Height (μm)	Spicule Length (µm)	Number of Tail Papillae
O. aquaedulcis Schneider, 1937	2600-2900	60-71	7–9	28-42	39	100	1
O. bajulus Paramonov, 1937	3144-3676	54-61	7–8	48-50	35	40	1
O. balli Nicholas & Stewart, 1984	1834-2314	52-69	7–8	33-51	26-28	26-34	1
O. cobbi Gerlach, 1954	3452	40	7	47	42	57	1
O. deconincki Heyns & Coomans, 1977	3200-3770	66-80	7–9	36-47	32-41	35-41	1
O. dimorphicus Tahseen et. al., 2016	1925-2076	69–77	6	48-57	-	36–38	1
O. domesticus Chitwood, 1960	2700-3350	54-78	7	39-47	36-38	51-54	1
O. hyrcanus Tchesunov, 1976	2810	57	7	36	29	55	1
O. japonicas Belogurov & Belogurova, 1981	3250-3420	60-89	9–10	52-62	26-31	37-41	1
O. longispiculosus Gerlach, 1955	3090	31	7	46	40	108	1
O. longus Wieser, 1953	5000-6740	130-150	13	104-108	26	42	1
O. martini Wieser, 1959	3890	65	8	43	36	51	1
O. menzeli Schneider, 1937	1885-2084	75-83	6	34-45	22	30	1
O. oxyuris Gerlach, 1957	2970	62	7	42	32	45	1
O. paraoxyuris Salma et. al, 2017	4100	83	11	64	38	50	1
O. paroxyuris Schuurmans Stekhoven, 1942	1512	42	6	47	-	-	1
O. qingdaoensis Zhang & Platt, 1983	2380-2640	92–98	6–7	48-53	25-27	34-36	1
O. rionegrensis Pastor de Ward et. al., 2013	2440-3030	37–51	6–7	30-45	26-33	47-57	1
O. rivalis Gagarin & Gusakov, 2012	3730	57	8	47	35	53	1
O. sahariensis Coomans & Heyns, 1983	3070	63	7	46	27	48	1
O. sidiiqii Tauheed & Ahmad, 2016	2420-2820	59-66	6	39-50	34–38	35-40	1
<i>Ö. tripapillatus</i> sp. nov.	4071-4435	82-86	7–8	42-48	29-31	41-44	3
O. vesicarius Wieser, 1959	3650	52	8	72	32	38	2
O. vietnamicus Gagarin & Thanh, 2011	1758-2226	70–77	5–7	55-59	26-29	52-56	1
O. xiamenense Čhen & Guo, 2014	2480-3020	57-81	6–7	37-42	27-32	44-52	1
O. zhangi Gao & Huang, 2016	3718–3934	55–62	7	34–37	33–35	52–56	1







Figure 5. Pictorial key to the species group with remarkable tail papillae of the genus *Oncholaimus*. Source of the figures: (**a**) Schneider (1937); (**b**) Paramonov (1937); (**c**) Nicholas and Stewart (1984); (**d**) Gerlach (1954); (**e**) Heyns and Coomans (1977);

(f) Tahseen, Siddiqi and Mustaqim (2016); (g) Chitwood (1960); (h) Tchesunov (1976); (i) Belogurov and Belogurova (1981); (j) Gerlach (1955); (k) Wieser (1953); (l) Wieser (1959); (m) Schneider (1937); (n) Gerlach (1957); (o) Salma, Nasira, Saima and Shahina (2017); (p) Schuurmans Stekhoven (1942); (q) Zhang and Platt (1983); (r) Pastor de Ward et. al., (2013); (s) Gagarin and Gusakov (2012); (t) Coomans and Heyns (1983); (u) Tauheed and Ahmad (2016); (v) *O. tripapillatus* sp. nov.; (w) Wieser (1959); (x) Gagarin and Thanh (2011); (y) Ghen and Guo (2014); (z) Gao and Huang (2016).

Oncholaimus tripapillatus sp. nov. was discovered in the shallow subtidal coarse detritus and shell gravels collected around Dokdo Islands of the East Sea, northwest Pacific Ocean. Oncholaimus tripapillatus sp. nov. is characterized by a combination of the following features: (1) the presence of a slightly constricted head; (2) relatively long cephalic setae ($10-12 \mu m$); (3) the presence of a precloacal sexual protuberance and two small, stout cone-shaped spines situated just anterior to the cloaca in males; (4) five pairs of long cloacal setae (three pairs of precloacal and two pairs of postcloacal setae in the subventral position); (5) a conoid tail, with three conspicuous papillae at the end of the tail in males; (6) the presence of two pairs of short and stout setae around the tail papillae. Among the genus Oncholainus, there are 25 species with conspicuous ventral papillae on the tail region. Of these, only four species-i.e., O. domesticus (Chitwood & Chitwood, 1938), O. qingdaoensis Zhang & Platt, 1983, O. rionegrensis Pastor de Ward et al., 2013, and O. sahariensis Coomans & Heyns, 1983—are reported to have the preanal supplementary spines in males. Oncholaimus tripapillatus sp. nov. closely resembles these four species based on the presence of a conoid tail with remarkable ventral papillae and a preanal supplementary spine in males. However, Oncholaimus tripapillatus sp. nov. differs from O. domesticus by the cloacal setae pattern (arranged in a longitudinal row of five pairs of cloacal setae rather than arranged in a transverse semicircle of 13–15 cloacal setae) and the position of the tail papillae (at the end of the tail rather than in the distal third of the tail). Oncholaimus tripapillatus sp. nov. is distinguished from O. sahariensis by a relatively longer body length in males ($4071-4435 \ \mu m \ vs. \ 2720 \ \mu m$), the number and arrangement of the cloacal setae (arranged in a longitudinal row of five pairs of cloacal setae rather than in a transverse semicircle of 16 cloacal setae), and the position of the tail papillae (at the end of tail rather than in the middle of tail). Oncholaimus tripapillatus sp. nov. is distinguished from O. rionegrensis by a longer body length in males (4071–4435 µm vs. 2440–3030 µm), a relatively shorter spicule length (41–44 μ m vs. 47–57 μ m), the number of cloacal setae (five pairs vs. nine pairs), the number of tail papillae (three vs. one), and the height of the tail papillae (2.4–2.7 μm vs. 7 μm). Oncholaimus tripapillatus sp. nov. also resembles O. *qingdaoensis* in possessing a constricted head, relatively long cephalic setae, longitudinally arranged cloacal setae in a row, and the presence of preanal supplementary spines in males. However, Oncholaimus tripapillatus sp. nov. differs from O. qingdaoensis in body length in males (4071–4435 μ m vs. 2350–2640 μ m) and the number of tail papillae (three vs. one). Moreover, Oncholaimus tripapillatus sp. nov. has a precloacal sexual protuberance situated just anterior to the cloaca opening; this feature is not shared by O. gingdaoensis.

3.2. Description of Oncholaimus Qingdaoensis Zhang & Platt, 1983

Class Enoplea Inglis, 1983 Order Enoplida Filipjev, 1929 Family Oncholaimidae Filipjev, 1916 Genus *Oncholaimus* Dujardin, 1845 *Oncholaimus qingdaoensis* Zhang & Platt, 1983 (Figures 6–8, Table 3)



Figure 6. *Oncholaimus qingdaoensis* Zhang & Platt, 1983—lateral view of a male: (**A**) habitus; (**B**) anterior region; (**C**) head region; (**D**) spicule and tail region (MABIK NA00156663); (**E**) spicule and tail region (scale bars: (**A**) = 100 μ m; (**B**) = 50 μ m; (**C**,**D**) = 20 μ m; (**E**) = 10 μ m).



Figure 7. *Oncholaimus qingdaoensis* Zhang & Platt, 1983—lateral view of a female: (**A**) habitus; (**B**) vulva region and demanian system; (**C**) head region; (**D**) tail region (scale bars: (**A**,**B**) = 100 μ m; (**C**,**D**) = 20 μ m).



Figure 8. *Oncholaimus qingdaoensis* Zhang & Platt, 1983—DIC photomicrographs, showing the lateral view of a male: (**A**) head region; (**B**) head region showing amphideal fovea (arrow); (**C**) ventral cloacal seta (arrow); (**D**) cloacal region showing precloacal spins (arrow); (**E**) spicules and tail region (scale bars: (**A**–**E**) = 20μ m).

Table 3. Morphometrics of Oncholaimu	us qingdaoensis (in µm)	
--------------------------------------	-------------------------	--

Characters	1 ♂ੋ	2 ♂	3♂	1 ♀	2 ♀
Body length	3379	3505	3715	4061	3550
a	82	86	85	75	67
b	7	7	7	7	7
с	46	45	44	56	43
Head diameter at the level of cephalic setae	28	29	29	32	31
Body diameter at the level of the cardia	36	40	38	44	47
Maximum body diameter	41	41	44	54	53
Cephalic setae length	10	12	11	11	11
Amphideal fovea diameter	10	12	11	10	12
Amphideal fovea length	5	6	7	5	7
Buccal cavity diameter	17	19	18	17	21
Buccal cavity length	32	31	33	35	34
Anterior end to the largest tooth tip	5	5	7	6	8
Anterior end to the other teeth tips	13	12	13	12	14
Anterior end to amphids	22	20	23	22	22
Anterior end to excretory pores	84	79	77	68	81
Anterior end to the nerve ring	229	241	243	265	247
Pharynx length	492	533	555	560	514
Spicule length	47	50	52	-	-
Cloacal setae length	8	8	8	-	-
Anterior end to the vulva	-	-	-	2812	2990
Body diameter at level of the vulva	-	-	-	54	56
V (%)	-	-	-	69	84
Anal body diameter	30	29	32	38	40
Tail length	74	79	85	72	83
Tail length/anal body diameter	2.5	2.7	2.7	1.9	2.1

3.2.1. Material Examined

The examined material included three males (MABIK NA00156663, KIOST NEM-1-2565, and KIOST NEM-1-2604) and two females (KIOST NEM-1-1719 and KIOST NEM-1-2605), mounted in anhydrous glycerin between two coverslips on an HS slide, sealed with nail polish. One male specimen (MABIK NA00156663) was deposited in the nematode collection at the specimen conservation room of the Marine Biodiversity Institute of Korea (MABIK), and the other specimens were deposited in the Bio-Resources Bank of Marine Nematodes (BRBMN), Korea Institute of Ocean Science and Technology.

3.2.2. Locality and Habitats

One male (MABIK NA00156663) and one female (KIOST NEM-1-1719) were collected from the intertidal sandy sediments of Namildae beach (34°55′34.34″ N, 128°05′43.11″ E), Sacheon-si, Gyeongsangnam-do on 19 January 2017 by H. Lee and H. J. Lee. Two males (KIOST NEM-1-2565 and KIOST NEM-1-2604) and one female (KIOST NEM-1-2605) were collected from the intertidal sandy sediments of Ujeon beach (34°58′18.00″ N, 126°08′11.00″ E), Sinan-gun, Jeollanam-do on 26 May 2020 by H. Lee and H. S. Rho.

3.2.3. Measurements

See Table 3 for measurements and morphometrics.

3.2.4. Description

Males: Bodies are comparatively long (3379–3715 µm) and slender, typically cylindrical in appearance (Figure 6A). The maximum body diameter at the mid body level is $41-44 \mu m$. The head is constricted immediately posterior to the cephalic setae (Figures 6C and 8A). The head region (28–29 µm in width) has typically six rounded lips, slightly attenuated toward the end, with a minute rounded inner labial papilla. Six outer labial and four cephalic setae are arranged in one circle, almost equal in size (10–12 μ m), inserted at the level of the amphideal fovea. The buccal cavity (31–33 µm deep, 17–19 µm wide) has a thick cuticular wall and three well-developed teeth. The left subventral tooth is the largest (26–28 μ m), with its tip stretched up to the cephalic setae, while the right subventral and dorsal teeth are almost equal in size (19–21 μ m). The amphideal fovea is pocket-like, 10–12 μ m in width (corresponding head diameter of 37%–41%), and 5–7 µm in height, located 20–23 µm from the anterior end (Figures 6C and 8B). The pharynx (492–555 µm long) is cylindrical, approximately 15% of the total body length, with a corresponding body diameter of 36–40 μ m. The excretory pore is situated 77–84 μ m from the anterior end. The nerve ring is situated 229–243 µm from the anterior end, with a corresponding body diameter of $35-38 \mu$ m, and 44%-47% of the length of the pharynx (Figure 6B). The somatic setae are sparsely distributed on the surface across the body. Two opposed testes are located on the right side of the intestine. The spicules are short and straight (47–52 µm long), proximally cephalated and pointed distally (Figure 6D,E and Figure 8E). The gubernacula are absent. Four pairs of long and stout setae are located in the cloacal region in a row (Figures 6D,E and 8C). Two unapparent pairs of precloacal spines are situated at the cloacal opening (Figures 6E and 8D). The tail is 74–85 μm long, approximately 2.5–2.7 times the anal body diameter. A conspicuous ventral tail papilla is situated approximately 14-16 µm from the tail tip (Figure 6D,E and Figure 8E). Two pairs of ventral setae (3–4 μ m) are situated in the tail papilla.

Females: Very similar to the males but differ in several dimensions, tail shape, and sexual characteristics. The body is 3550–4061 μ m long, with a maximum body diameter of 53–54 μ m (Figure 7A,C). The conical tail (72–83 μ m long) is slightly curved ventrally, without a ventral papilla (Figure 7D), and approximately 1.9–2.1 times the anal body diameter. The reproductive system is monodelphic with anterior reproductive branches, lying to the right side of the intestines. The vulva is situated at 69–84% of the total body length from the anterior end (Figure 7B). The demanian system is composed of one

osmosium connecting the ductus entericus and the intestine, and one uvette, located at $870 \mu m$ in front of the anus, connecting the ductus uterinus with a main tube (Figure 7B).

3.2.5. Distribution

China (Qingdao), Korea (Sacheon, Sinan).

3.2.6. Differential Diagnosis and Relationships

Oncholaimus qingdaoensis Zhang & Platt, 1983 was first described from intertidal sand in Qingdao, China. Oncholaimus qingdaoensis has been previously reported only in that type of locality. The present Korean specimens of Oncholaimus qingdaoensis were discovered from the intertidal sandy sediments of the southern and western coasts of Korea. Oncholaimus *qingdaoensis* is easily distinguished from its congeners as having the following characteristic combinations: (1) the presence of a constricted head, (2) relatively long cephalic setae, (3) the presence of four pairs of long and stout cloacal setae longitudinally arranged in a row in males, (4) the presence of two pairs of precloacal spines in males, and (5) a conoid tail with conspicuous ventral papillae in front of the tail tip in males. The present Korean specimens of O. qingdaoensis resemble O. domesticus (Chitwood & Chitwood, 1938), O. hyrcanus Tchesunov, 1979, O. longus (Wieser, 1953), and O. xiamenense Chen & Guo, 2014, mainly in possessing a conoid tail with conspicuous ventral papillae in front of the tail tip in males. The present Korean specimens of *O. qingdaoensis* are clearly distinguished from O. domesticus and O. hyrcanus by the absence of a precloacal sexual protuberance in males and by a cloacal setae pattern (longitudinally arranged in a row of cloacal setae rather than arranged in a transverse semicircle of cloacal setae). The Korean specimens differ from O. longus in terms of body length (3.4–3.7 mm vs. 5–6.7 mm in males), number of cloacal setae (four pairs vs. seven pairs), and by possessing two pairs of cloacal spinae (absent in O. longus). Moreover, the Korean specimens are also distinguished from O. xiamenense based on the body length (3.4-3.7 mm vs. 2.5-3 mm in males), number of cloacal setae (four pairs vs. six pairs), and the presence of a constricted head and precloacal spins (absent in the original description of O. xiamenense). The present Korean specimens agree well with Zhang and Platt's (1983) original description in general features, especially for a constricted head, the detailed shape of the tail, and the number and shape of cloacal setae. However, the Korean specimens differ from the Chinese specimens in the longer body length in males $(3379-3715 \ \mu m \ vs. \ 2380-2640 \ \mu m)$, the larger spicule length $(47-52 \ \mu m \ vs. \ 34-36 \ \mu m)$, and the presence of ventral tail papillae situated approximately 14–16 μm from the tail tip.

Author Contributions: Conceptualization, data curation, writing—original draft preparation, and writing—review and editing, H.L. (Hyojin Lee) and H.R.; investigation, H.L. (Heegab Lee) and H.R.; funding acquisition, H.R. All authors have read and agreed to the published version of the manuscript.

Funding: This research was supported by a grant from the National Marine Biodiversity Institute of Korea (2021M01100) and by the research projects of the Korea Institute of Ocean Science and Technology (PG52510, PE99913).

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: Not applicable.

Conflicts of Interest: The authors declare no conflict of interest.

References

- 1. Smol, N.; Muthumbi, A.; Sharma, J. *Handbook of Zoology. Gastrotricha, Cycloneuralia and Gnathifera*; Nematoda. 7.3 Order Enoplida; Walter De Gruyter: Berlin, Germany, 2013; Volume 2.
- Gerlach, S.; Riemann, F. The Bremerhaven Checklist of Aquatic Nematodes. A Catalogue of Nematoda Adenophorea excluding the Dorylaimida. Veröffentlichungen Inst. Meeresforsch. Bremerhav. Suppl. 1974, 4, 405–736.

- 3. Zeppilli, D.; Bellec, L.; Cambon-Bonavita, M.-A.; Decraemer, W.; Fontaneto, D.; Fuchs, S.; Gayet, N.; Mandon, P.; Michel, L.N.; Portail, M.; et al. Ecology and trophic role of *Oncholaimus dyvae* sp. nov.(Nematoda: Oncholaimidae) from the lucky strike hydrothermal vent field (Mid-Atlantic Ridge). *BMC Zool.* **2019**, *4*, 1–15. [CrossRef]
- Shimada, D.; Suzuki, A.C.; Tsujimoto, M.; Imura, S.; Kakui, K. Oncholaimus langhovdensis sp. nov.(Nematoda: Enoplea: Oncholaimida), a new species of free-living marine nematode from Langhovde, Dronning Maud Land, East Antarctica. Species Divers. 2017, 22, 151–159. [CrossRef]
- Belogurov, O.; Belogurova, L.; Pavlyuk, O. Morphology and systematic position of two new species of marine nematodes (Nematoda: Oncholaimidae) from Far-Eastern seas: *Oncholaimium olium* sp. n. and *Pseudoncholaimus vesicarius* (Wieser, 1959) comb. nov. *Biol. Morya* 1975, 2, 25–30.
- 6. Belogurov, O.; Belogurova, L. Morphology of *Oncholaimus paropisthonchus* sp. n., *Oncholaimus zinus* sp. n., *Oncholimium unicum* sp. n. and key for genera of the subfamily Oncholaiminae(Nematoda). Zool. Zhurnal **1978**, *57*, 977–986.
- Belogurov, O.; Belogurova, L.; Pavlyuk, O. Three new species of free-living nematodes from the family Oncholaimidae from the Far-Eastern seas of the USSR, and a differential key for *Pseudoncholaimus* species. In *Coastal Plankton and Benthos of the Northern Parts of the Sea of Japan*; Akademiya Nauk SSSR, Dal'nevostochnyĭ Nauchnyĭ Tsentr; Institut Biologii Morya: Vladivostok, Russia, 1980; pp. 57–64.
- 8. Smolyanko, O.; Belogyrov, O. On the study of the morphology and taxonomy on two nematode species of the genus *Oncholaimium* (Nematoda, Oncholaimidae) and comments on the independence of this genus and on synonymy of *O. olium* and *O. sinensis*. *Zool. Zhurnal* **1991**, *70*, 17–24.
- 9. Wieser, W. A collection of marine nematodes from Japan. Publ. Seto Mar. Biol. Lab. 1955, 4, 159–181. [CrossRef]
- 10. Shimada, D.; Kajihara, H.; Mawatari, S.F. Three new species of free-living marine nematodes (Nematoda: Enoplida) from northern Japan. *Species Divers.* **2009**, *14*, 137–150. [CrossRef]
- 11. Zhang, Z.; Platt, H.M. New species of marine nematodes from Qingdao, China. *Bull. Br. Mus. Nat. Hist. Zool.* **1983**, 45, 253–561. [CrossRef]
- 12. Chen, Y.; Guo, Y. Three new species of free-living marine nematodes from East China Sea. *Zootaxa* **2014**, *3841*, 117–126. [CrossRef] [PubMed]
- Gao, Q.; Huang, Y. Oncholaimus zhangi sp. nov.(Oncholaimidae, Nematoda) from the intertidal zone of the East China Sea. Chin. J. Oceanol. Limnol. 2017, 35, 1212–1217. [CrossRef]
- 14. Lee, H.J.; Rho, H.S.; Jung, J. New Record of the Genus Oncholaimus Nematode Species (Nematoda: Oncholaimidae) from the East Sea of Korea. *Korean J. Environ. Biol.* **2015**, *33*, 170–176. [CrossRef]
- 15. Kristensen, R.M.; Higgins, R.P. Marine Tardigrada from the southeastern United States coastal waters I. *Paradoxipus orzeliscoides* n. gen., n. sp.(Arthrotardigrada: Halechiniscidae). *Trans. Am. Microsc. Soc.* **1989**, 262–282. [CrossRef]
- Shirayama, Y.; Kaku, T.; Higgins, R.P. Double-sided microscopic observation of meiofauna using an HS-slide. *Benthos Res.* 1993, 44, 41–44. [CrossRef]