

Pratylenchus unzenensis n. sp. from *Artemisia* sp.
in Japan (Nematoda: Pratylenchidae)

Takayuki MIZUKUBO

*Laboratory of Nematology and Soil Zoology, Department of Environmental
Biology, National Institute of Agro-Environmental Sciences,
Kannonnai 3-1-1, Tsukuba, Ibaraki 305, Japan*

(Received March 30, 1992; Accepted June 12, 1992)

A lesion nematode collected from the rhizosphere of *Artemisia* sp. at Mt. Unzen (Nagasaki Pref.) was described as *Pratylenchus unzenensis* n. sp. The new species is characterized as follows: $L=370-470\ \mu\text{m}$ (mean 420); $c=12-17$ (14); $V=75-79\%$ (77); spear= $14.5-15.5\ \mu\text{m}$ (14.9); lip annuli=3; tail annuli=21-32 (26); annulated or smooth tail termini. This nematode is distinguished from the similar *P. pratensisobrinus* by a shorter spear ($14.5-15.5\ \mu\text{m}$ (14.9) vs. $15-17\ \mu\text{m}$ (16)), undifferentiated short post-vulval uterine branch ($U=0.9-2.0$ vs. $1.6-2.7$ (2.0) and differentiated) and short spicules in male ($13-14\ \mu\text{m}$ (13) vs. $17-19\ \mu\text{m}$ (18)). *En face* pattern of the new species is characteristic as it has sloping and angular edges of the sub-median segments, considered to be a link between Groups 2 and 3 of the face types proposed by CORBETT and CLARK (1983).

Key words: taxonomy, lesion nematode, new species, *Pratylenchus unzenensis*

INTRODUCTION

During a survey of plant parasitic nematodes in Japan, I found a lesion nematode population closely resembling *P. pratensisoblinus* BERNARD, 1984, which had been synonymized with *P. pratensis* DE MAN, 1880 by FREDERICK and TARJAN (1989). Examination of the specimens of this nematode by optical light microscope and scanning electron microscope (SEM) led to defining it as a new species.

METHODS

Observations were made on the specimens killed by gentle heating, fixed in TAF and mounted in dehydrated glycerin by the quick method of SEINHORST (1959). The specimens were observed and measured for all features at oil immersion at $\times 2,400$ on an Olympus BH-2 light microscope equipped with Normarski interference contrast.

SEM observations were made on a specimen stored in TAF, processed in 30% to 100% ethanol, critical-point dried, and coated with platinum-vanadium (20 nm). The specimen was examined in a Hitachi X-650 SEM at 10 kV.

SYSTEMATICS

Pratylenchus unzenensis n. sp.
(Figs. 1-3; Table 1)*Measurements*

Measurements of the *holotype* female, 12 *paratype* females and 2 *paratype* males are shown in Table 1.

Description

Female ($n=13$): Body slightly curved ventrally when heat-relaxed (Fig. 2 A). Annuli $1.2 \pm 0.10 \mu\text{m}$ (1.0-1.3) wide at midbody. Lip region relatively low, $2.3 \pm 0.20 \mu\text{m}$ (2.0-2.6) high, $7.7 \pm 0.28 \mu\text{m}$ (7.3-8.3) wide, flattened anteriorly, continuous to body contour. Labial framework moderately developed, laterally extending into body at one annule. Lip with 3 annuli on both sides (Fig. 1 B; Figs. 2 D-F). SEM *en face* micrographs (Fig. 1 A) showed division between sub-median and lateral segments; lateral edges of sub-median segments curving slope or angular, assigned between Group 2 and Group 3 of CORBETT and CLARK (1983). Amphid apertures narrow and oblique (Fig. 1 A). Cephalids not seen. Spear 1.9 ± 0.7 times (1.8-2.1) as long as

Table 1. Measurements of *Pratylenchus unzenensis* n. sp.

Criteria	Holotype (♀) ($n=1$)	Paratypes (♀) ($n=12$)		Paratypes (♂) ($n=2$) range
		mean: SD	range	
<i>L</i> (μm)	415	416: 31.1	368-470	366-395
<i>a</i>	25.0	25.8: 2.06	21.9-28.5	29.2-30.0
<i>b</i>	5.3	5.5: 0.47	4.9-6.1	4.9-5.2
<i>b'</i>	3.3	3.4: 0.35	2.9-4.1	3.1-3.4
<i>c</i>	12.1	13.7: 1.33	11.6-16.6	14.2-17.1
<i>c'</i>	3.1	2.9: 0.30	2.4-3.5	2.2-2.8
<i>V</i> (%)	75.2	76.9: 1.04	75.2-78.9	—
spear (μm)	14.9	14.9: 0.32	14.5-15.5	13.9-14.2
<i>m</i> (%)	50.0	51.4: 2.16	47.8-54.5	52.1-54.8
DGO (μm)	2.0	2.4: 0.25	2.0-2.6	2.6-2.8
knob width (μm)	4.0	4.0: 0.36	3.5-4.5	3.3-3.6
excretory pore (μm)	68	72: 2.3	67-75	73
esophagus (μm)	79	76: 5.3	65-82	75-77
vulva-anus	69	66: 8.0	55-87	—
tail (μm)	34	31: 2.6	26-34	23-26
post-uterine branch (μm)	29	23: 4.6	15-30	—
<i>U</i> (%)	1.8	1.5: 0.34	0.9-2.0	—
<i>G</i> ₂ (%)	7.0	5.4: 1.0	3.8-7.0	—
tail annule	29	26: 3.6	21-32	—
<i>T</i> (%)	—	—	—	50.1
spicules (μm)	—	—	—	13.2-13.5
gubernaculum (μm)	—	—	—	4.3-5.0
spicule/spear ratio	—	—	—	0.95

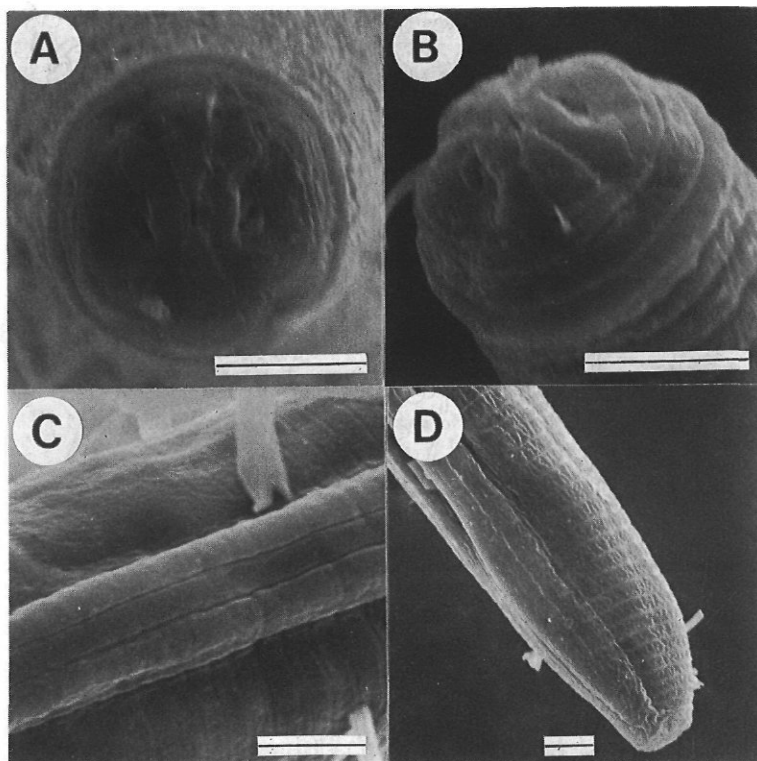


Fig. 1. *Pratylenchus unzenensis* n. sp. Female. A: head (en face view), B: head (lateral view), C: lateral field, D: tail (lateral view). Scale bars=3 μ m.

head diameter. Spear knobs flattened anteriorly (54%: Figs. 2 C, D and G) or broadly rounded (46%: Fig. 2 F), never indented anteriorly, $2.2 \pm 0.16 \mu\text{m}$ (2.0–2.4) high, $4.0 \pm 0.36 \mu\text{m}$ (3.5–4.5) across. Metacarpus oval, valve conspicuous, $51 \pm 4.7 \mu\text{m}$ (41–58) from anterior body end, or $67.2 \pm 6.40\%$ (55.8–77.6) of esophagus length. Esophageal basal lobe extending $123 \pm 9.9 \mu\text{m}$ (107–135) from anterior body end, ventrally overlapping intestine $47 \pm 10.3 \mu\text{m}$ (32–65) or a distance of 2.9 ± 0.62 times (2.0–3.9) the corresponding body diameter. Esophageal nuclei line up. Excretory pore located at 90–106% of esophageal length or at $17.2 \pm 1.12\%$ (15.0–18.8) of body length. Hemizonid flat, 1 to 2 annuli long, immediately anterior to excretory pore.

Gonad outstretched, $148 \pm 20.4 \mu\text{m}$ (117–198) long; ovary with oocytes in a row or partially double row (Fig. 2 M); spermatheca oval to oblong, $53 \pm 11.8 \mu\text{m}$ (42–84) from vulva, $19.6 \pm 3.66 \mu\text{m}$ (13.9–24.4) ($n=8$) long, $10.7 \pm 0.86 \mu\text{m}$ (8.9–11.2) ($n=8$) wide, 1.8 ± 0.28 (1.5–2.3) ($n=8$) in length/width ratios, packed with spermatozoa (Figs. 2 H–J). Post-uterine branch (PUB) less than twice of vulval body diameter, without terminal rudimental ovary (Figs. 2 H, K and L). Vagina narrow-walled, perpendicular to body axis, $7.0 \pm 0.63 \mu\text{m}$ (5.9–7.9) ($n=9$) long, or $44 \pm 3.3\%$ (40–49) of vaginal body diameter. Annuli between vulva and anus 60 ± 5.9 (52–75) ($n=11$) in number.

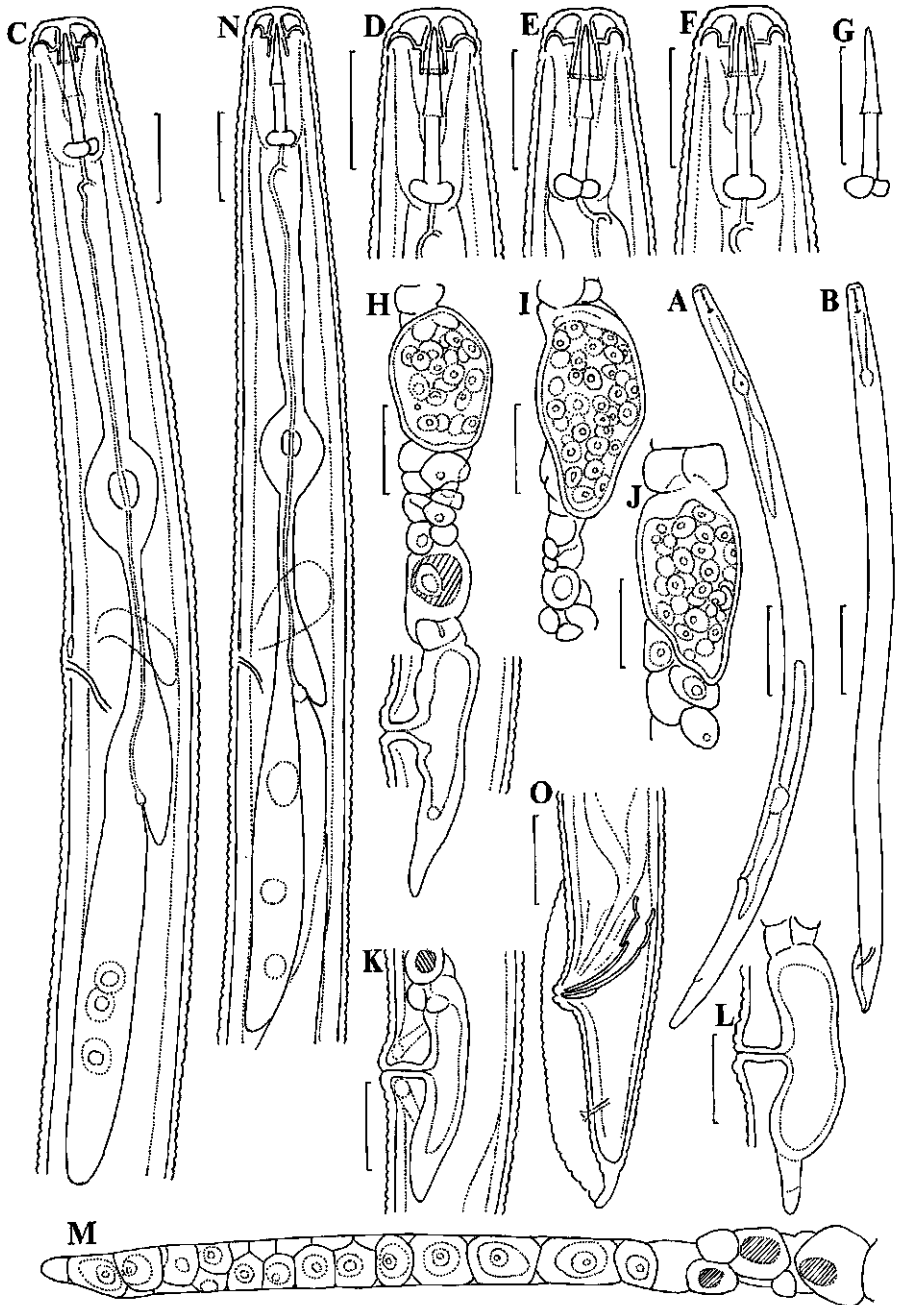


Fig. 2. *Pratylenchus unzenensis* n. sp.—Female. A: entire, C: esophageal region, D–F: heads, G: spear, H–J: spermathecae, K, L: vulval region, M: anterior gonad,—Male. B: entire, N: esophageal region, O: posterior end. Scale bars: A & B=50 μ m; C–O=10 μ m.

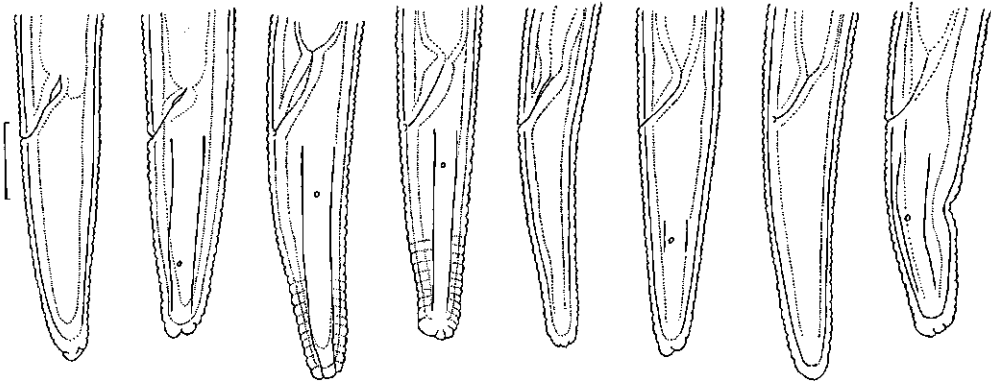


Fig. 3. Morphological variation of female tails of *Pratylenchus unzenensis* n. sp. Scale bar = 10 μ m.

Tail termini with some variations in shape (Fig. 3): using tail tip shape codes of FREDERICK and TARJAN (1989), mostly fall into SHM (subhemispherical), rarely in TRN (truncate) (Fig. 1 D), but never in distinct BLP (bluntly pointed) condition; termini smooth (38%) or irregularly annulated (62%) ($n=13$). Tail terminal cuticle 2.2 ± 0.21 μ m (2.0–2.6) thick; phasmids pore-like, centered in lateral fields (Fig. 3), 16–19 μ m behind anus.

Lateral fields 6.0 ± 0.68 μ m (5.3–6.6) ($n=5$) wide, i.e. $37.1 \pm 3.17\%$ (32.0–40.0) of the widest body diameter, consist of 3 bands or 4 incisures; outer bands partially areolated at mid-body region (Fig. 1 C) and completely areolated at tail region (Fig. 1 D). Lateral fields extend at tail tip and continue around the tail terminus (Fig. 1 D).

Males ($n=2$): Body straight to arched when killed by gentle heating (Fig. 2 B). Annuli finer than in female, 1.0–1.1 μ m apart at mid-body. Lip region relatively low, 2.4–2.6 μ m high, 6.6–6.9 μ m wide, flattened anteriorly, continuous to body contour. Labial framework moderately developed, laterally extending into body at 1 annule. Spear shorter than in female, 2.0–2.1 times as long as head diameter. Spear knobs smaller than female, 3.3–3.6 μ m across, broadly rounded (Fig. 2 N). DGO close to spear base. Metacarpus oval; valve conspicuous, 48–50 μ m from anterior body end, or 63–66% of esophagus length. Esophageal basal lobe extending 116 μ m from anterior body end, ventrally overlapping the intestine 40–42 μ m or a distance of 3.0–3.3 times the corresponding body diameter. Esophageal nuclei line up. Hemizonid flat, immediately anterior to excretory pore. Testis outstretched, with spermatogonia in double rows, 198 μ m long. Spicules arch-shaped, slightly shorter than spear; gubernaculum crescent-shaped. Bursal alae 36 μ m long. Tails' terminal cuticle 4.6–5.9 μ m thick, ending in finely rounded terminus (Fig. 2 O); phasmids 12.5–15.8 μ m behind anus or 9.9–10.6 μ m from tail tip. Lateral fields 4.6 μ m wide, i.e., 35% of widest body diameter.

Type specimens

Holotype female (slide no. T36-5) and 11 *paratype* females and 2 *paratype* males (slides nos. T36-1–T36-4, T36-7–T36-15) are deposited in the Herbarium and Insect

Museum of the National Institute of Agro-Environmental Sciences. One *paratype* female (slide no. T36-6) is deposited in the University of California Davis Nematode Collection (UCDNC), Davis, CA, U.S.A.

Type host and locality

Specimens collected from the rhizosphere of *Artemisia* sp. at Nita-toge, Mt. Unzen, Nagasaki Prefecture, Japan by T. MIZUKUBO in August, 1988.

Diagnosis and relationships

Pratylenchus unzenensis n. sp. is characterized in the following character-states: female—3-lip annuli, short body ($L=368-470\ \mu\text{m}$ (mean 416)), smaller c -value (11.6–16.6 (13.7)), relatively small V -value (75–79% (77)), moderately long spear (14.5–15.5 μm (14.9)), moderately distanced DGO (2.0–2.6 μm (2.4) from spear base), post-uterine branch (PUB) of moderate length ($U=0.7-2.0$ (1.5)), undifferentiated PUB, tail with 21–32 (26) annuli, oval or oblong spermatheca with spermatozoa, irregularly annulated or smooth tail terminus; male—much short spicule (13–14 μm) and short gubernaculum (4.3–5.0 μm). By having 3-lip annuli, V -value at most 80%, c -value at most 20, the new species is distinguished from all the congeneric species except *P. cerealis* HAQUE, 1966, *P. elamini* ZEIDAN et GERAERT, 1991, *P. emarginatus* EROSHENKO, 1978, *P. exilis* DAS et SULTANA, 1979, *P. kasari* RYSS, 1982, *P. manohari* QURAIISHI, 1982 (syn. of *P. cerealis* according to FREDERICK and TARJAN (1989)), *P. morettoii* LUC et al., 1986, *P. pratensis* (DE MAN, 1880), *P. pratensisobrinus* BERNARD, 1984 (syn. of *P. pratensis* according to FREDERICK and TARJAN (1989)), *P. teres* KHAN et SINGH, 1975, *P. yassini* ZEIDAN et GERAERT, 1991, and *P. zaeae* GRAHAM, 1951.

These are separated from *P. unzenensis* n. sp. by the character-states shown in parentheses: *P. cerealis* ($V=79-80\%$, $c=19$); *P. elamini* ($V=72-77\%$, $c=16-21$, *en face*=Group 1); *P. emarginatus* ($V=68-73\%$, tail annuli=19–20, $c=14-21$); *P. exilis* ($L=490-560\ \mu\text{m}$, spear=17–18 μm , $V=73-76\%$, tail annuli=17–20); *P. kasari* ($L=560-770\ \mu\text{m}$, $c=16-20$, tail annuli=32–44); *P. manohari* ($V=78-80\%$, $c=18-20$, tail annuli=13–15); *P. morettoii* ($L=560-930\ \mu\text{m}$, spear=14–19 μm (16), tail tip=finely pointed, *en face*=Group 1); *P. pratensis* ($L=400-630\ \mu\text{m}$, $c=15-21$ (19), rather regularly annulated tail tip, male spicule=17–19 μm , *en face*=typical Group 3); *P. pratensisobrinus* (spear=15–17 μm (16), DGO=1.0–2.5 μm (1.7), $U=1.6-2.7$ (2.0), differentiated PUB, male spicules=17–19 μm); *P. teres* ($V=70-77\%$ (72), spear=17–18 μm); *P. yassini* ($V=71-76\%$, $c=13-19$, *en face*=Group 1); *P. zaeae* ($V=69-75\%$, spear=15–17 μm , *en face*=Group 1).

Remark

This new species most closely resembles *P. pratensisobrinus* and *P. pratensis*. Although FREDERICK and TARJAN (1989) regarded the former as a junior synonym of the latter, they did not articulate their reasoning. BERNARD (1984) distinguished *P. pratensisobrinus* from *P. pratensis* in such characters as spear length (generally longer), tail length (smaller c and larger c') and number of tail annuli (more numerous). Despite some degree of overlap among all of them, he considered these differences sufficient to separate the two species. I consider that they should remain as two distinct species, since they show an obvious difference in c -value, which was adopted by FREDERICK's and TARJAN's (1989) tabular key to the species as being diagnostic for differentiation

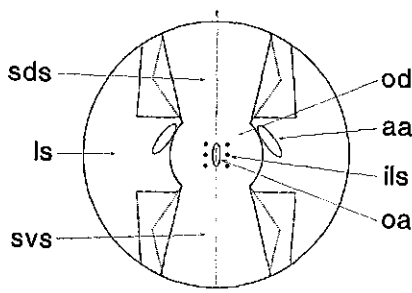


Fig. 4. Diagram of 'face' showing oral disc and variations of the first lip annule (modified from CORBETT and CLARK, 1983). Abbreviations: aa: amphid aperture, ils: inner labial sensilla, ls: lateral segment, oa: oral aperture, od: oral disc, sds: sub-dorsal segment, svs: sub-ventral segment. —: Group 3, - - -: typical Group 2, ·····: a variation of Group 2 (assigned between Groups 2 and 3).

(12–15 (14) vs. 15–21 (19)). LOOF (1991) also treated them as different species.

The face morphology of the present new species deserves careful attention. Its face does not adequately fulfill any of the face types (Groups 1, 2 or 3) proposed by CORBETT and CLARK (1983). As was mentioned in the description section, the face of this species comes between Groups 2 and 3, i.e., present species has a face with lateral edges of the sub-median segment widening gradually, then, at the middle of the edges, tapering towards the outer edge of the face and resulting in more or less angular edges of the segments (Fig. 4; broken line). In this respect, the structure of the present species differs from that of Group 2, which was described as having "sub-median segments that are fused to the oral disc and are narrower at their inner extremity widening towards the outer edge of the face" (CORBETT and CLARK, 1983), which did not include a statement on the converging edges of the sub-median segments near the outer edge of the face. Group 3 was described as having "a more distinctive dumb-bell shaped pattern of the sub-median segments" (CORBETT and CLARK, 1983). Unfortunately, the above description was fairly subjective and was not necessarily successful in defining Group 3 nor in distinguishing between Groups 2 and 3. I consider that Group 3 may be defined, from SEM micrographs of the members or a diagram of the face proposed by CORBETT and CLARK (1983), as having lateral edges of the sub-median segments extending at right angle to a median line of the face, then shifting their turn parallel with a median line to the edges of the face, forming rectangular sub-median segments. Provided that the above understanding of Group 3 is acceptable, the face of the present species differs from this category 3; its sub-median segments are by no means rectangular, even though their edges are angular.

This morphological property of the sub-median segments in the present species is not a preparation artifact of the specimen, since ANDERSON and TOWNSEND (1980), who apparently dealt with artifact specimens, indicated an obvious Group 3 pattern (rectangular segments) for *Pratylenchus penetrans*, which did not largely differ from the one observed by CORBETT and CLARK (1983) on the same species.

CORBETT and CLARK (1983) predicted that "examination of more species may show there to be only two groups, with no discontinuity in face pattern between the

types presented in Groups 2 and 3". Although present species has a face pattern that can be a link between Groups 2 and 3, I consider that these face types are still available. A pattern of the face in the present species would be assigned into Group 2, since the sub-median segments show lateral edges oblique to a median line of the face, never perpendicular to the latter, which presently seems to be the only criterion distinguishing Groups 2 and 3 (Fig. 4).

According to CORBETT and CLARK (1983), *P. pratensis* has a face pattern belonging to Group 3. Hence, the present new species would be distinguished from *P. pratensis* on the face morphology basis.

REFERENCES

- ANDERSON, R. V. and J. L. TOWNSEND (1980) Variation of the first head annule in Canadian populations of *Pratylenchus penetrans* (Nematoda: Pratylenchidae) from three host plants. *Can. J. Zool.* **58**: 1336-1340.
- BERNARD, E. C. (1984) Hoplolaimidae (Nematoda: Tylenchida) from the Aleutian Island with descriptions of four new species. *J. Nematol.* **16**: 194-203.
- CORBETT, D. C. M. and S. A. CLARK (1983) Surface features in the taxonomy of *Pratylenchus* species. *Revue Nématol.* **6**: 85-98.
- FREDERICK, J. J. and A. C. TARJAN (1989) A compendium of the genus *Pratylenchus* FILIPJEV, 1936 (Nemata: Pratylenchidae). *Revue Nématol.* **12**: 243-256.
- LOOF, P. A. A. (1991) The family Pratylenchidae THORNE, 1949. In *Manual of Agricultural Nematology* (W. R. NICKLE, ed.). Marcel Dekker, Inc., New York, p. 363-421.
- SEINHORST, J. W. (1959) A rapid method for the transfer of nematodes from fixative to anhydrous glycerin. *Nematologica* **4**: 67-69.