

Nematodes of the order Dorylaimida from Andalucía Oriental, Spain. The genus *Nygolaimus* Cobb, 1913.

IV. Description of *N. seguranus* sp. n. and its relationship with *N. europaeus* (Heyns, 1968) Ahmad & Jairajpuri, 1982

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Summary. *Nygolaimus seguranus* sp. n. is described from natural biotopes in southeastern Spain. The species is characterized by its medium sized (L=1.1-1.7 mm) body, lip region set off by a constriction and 11-12.5 µm wide, deltoid mural tooth 10.5-12.5 µm long at the dorsal side or almost equal to the lip region width, pharyngeal bulb encapsulated by a spiral sheath, V=47.8-57.1%, female tail rounded, conoid (17-20.5 µm, c=66.6-84.4, c'=0.9-1.1), male with no or only one ventromedian supplement, and spicules 27-27.5 µm long. The new species is most similar to *N. europaeus* (Heyns, 1968) Ahmad & Jairajpuri, 1982, the type specimens of which were examined during preparation of the description of the new species. Both species are illustrated, tables of measurements are given, and their relationship is briefly discussed.

Key words: *Nygolaimus seguranus* sp. n., *N. europaeus*, taxonomy, Spain.

In a general nematological survey carried out during the last decade mainly in natural areas in Andalucía Oriental (southeastern Spain) we collected many populations of the genus *Nygolaimus* Cobb, 1913. A few specimens tentatively identified as representing *N. europaeus* (Heyns, 1968) Ahmad & Jairajpuri, 1982 were found in several localities. Subsequently, a more detailed study of these nematodes revealed the presence of at least two species in the Andalusian soils. Comparison of the specimens with type material of *N. europaeus* indicated that most nematodes did not belong to this species, and constituted a new taxon.

This is the fourth in a series of papers on *Nygolaimus* and in it is presented a description of *N. seguranus* sp. n. and its differentiation and relationship with *N. europaeus*.

MATERIAL AND METHODS

Ten female and two male specimens collected in different natural habitats, mostly Mediterranean garigues, in the provinces of Jaén and Málaga have been studied. Nematodes were extracted by a decanting and sieving method, killed by heat, fixed in 4% formaldehyde and mounted in anhydrous glycerin.

Additional details on methodology is available in an earlier paper (Liébanas & Peña Santiago, 1997)

Five paratypes of *N. europaeus* examined during this study were provided by courtesy of Dr. P.A.A. Loof.

DESCRIPTIONS

Nygolaimus seguranus sp. n. (Fig. 1 & Table 1)

Type population:

Female. Very slender nematodes of medium sized body, L=1.13-1.52 mm. Body cylindrical, slightly tapering towards both extremities but somewhat more towards the anterior end. Habitus after fixation ventrally curved, usually "G"- or "J"-shaped, sometimes more irregular, probably due to the processing method. Outer cuticle layer thin along the entire body and smooth. Inner cuticle layer wider than the outer one and somewhat thickened at tail region. Lateral chord occupying one-fourth to two-fifths of the midbody width. Lateral pores obscure. Lip region set-off from the adjacent body by a clear (more or less deep) constriction; some 2.2-2.4 times as wide as high and little more than two-fifths of the body

diameter at neck base; in some specimens it is asymmetrical since the dorsal side appears more elevated than the ventral one. Lips completely amalgamated, with rounded or somewhat angular contour. Labial and cephalic papillae clearly visible, slightly interfering the head contour. Amphid cup-shaped, opening at level of the cephalic constriction and occupying around half of the corresponding body width. Stoma typical of the genus, with sclerotized wall. Mural tooth deltoid, ventral side three-fourths of the lip region diameter, dorsal one practically equal in size. Pharynx consisting of a slender but muscular anterior part which begins at the base of the stoma, then becomes narrower and at the posterior end expands gradually into the basal bulb. Pharyngeal bulb cylindrical, 15-19 times as long as wide, occupying almost half of the body width at the neck base and 54-57% of the total neck length; a conspicuous spiral sheath encapsulates it. Pharyngeal gland nuclei and outlets obscure in the females studied. Nerve ring situated at 27-38% of the total neck length. Cardia conoid, as long as wide. Cardiac cells ovoid, 10 x 5 μm ; in section they are longer than wide. Genital system didelphic-amphidelphic. Ovaries reflexed, medium sized, often reaching and surpassing the oviduct-uterus junction. Oocytes few in number, apparently in a single row. Oviduct consisting of a slender distal part with prismatic cells and a well developed *pars dilatata* with visible lumen. Oviduct-uterus junction distinguished by a clear narrowing but lacking an obvious sphincter. Uterus a wide tube 1-2 times the corresponding body width long and without modifications. Abundant sperm present throughout the genital tract. Vagina typical of the genus, extending inwards to two-fifths of the corresponding body diameter; *pars distalis vaginae* 4 μm long and with straight walls; *pars proximalis* a truncate cone 9 μm long and circled by musculature. Vulva a transverse slit. Prerectum 1.1-1.4 and rectum 0.8-1.1 times anal body width long. Tail convex conoid (more rounded in one paratype), ventrally more straight; the inner cuticle appears only somewhat thickened. Caudal pores obscure in the specimens examined, only visible in the holotype: two pairs, as illustrated.

Male. General morphology similar to female, but with smaller size and shorter odontostyle. Genital system diorchic with opposite testes. Ventromedian supplement absent. Spicules relatively stout and ventrally curved, 1.5 anal body widths long. Lateral guiding pieces relatively short and robust, with rounded end. Tail slightly ventrally curved. Spermatozoa spindle shaped.

Specimens (one female and one male) from Sierra

de la Pandera, province of Jaén:

Female. This specimen has a longer ($L=1.7$ mm) body and is visibly flattened. The lip region is more angular (see Fig. 1E), presenting very clear labial and cephalic papillae. In other morphological details it is very similar to specimens in the type population.

Male. This specimen is longer than males in the type population, becoming flattened posteriorly. It has one ventromedian supplement separated from the adanal pair by a distance little more than the anal body width. The spicules appear less ventrally curved, probably as a consequence of the flattening process.

Female from Sierra de la Cruz, province of Jaén:

The identity of this specimen is doubtful because of the morphology of the odontostyle (shorter and stouter than the type population) and mainly by the atypical morphology of the tail, whose internal core presents a very prominent terminal projection (see Fig. 1M) reaching the outer layer cuticle. It is probably not conspecific with the females of the type population.

Type habitat and locality. Soil around roots of *Lonicera arborea*, *Berberis hispanica*, *Crataegus monogyna* (a high altitude Mediterranean garigue) in two adjacent localities (Nava Noguera and Los Campos) in Sierra de Segura, province of Jaén, Spain.

Other habitats and localities. One female and one male were collected in association with *Echinoparatum boissieri*, *Salvia lavandulifolia* and *Lavandula latifolia* (other Mediterranean garigue), and *Asphodelus* sp. in Sierra de la Pandera, province of Jaén, Spain.

Type material. Four females (holotype and paratypes) and one male deposited in Departamento de Biología Animal, Vegetal y Ecología, Universidad de Jaén, Spain on slides n° 0228-0232. One female paratype deposited at Instituut voor Dierkunde, Rijksuniversiteit, Gent, Belgium; and at Muséum National d'Histoire Naturelle, Paris, France.

Etymology. The specific epithet "securanus" refers to Sierra de Segura, the geographical area where the species was collected.

Differential diagnosis and relationships. The new species is characterized by its medium sized ($L=1.1-1.7$ mm) body, lip region set off by a constriction and 11-12.5 μm wide, deltoid mural tooth 10.5-12.5 μm long at the dorsal side or almost equal to the lip region width, pharyngeal bulb involved by a spiral sheath, $V=47.8-57.1\%$, female tail rounded conoid

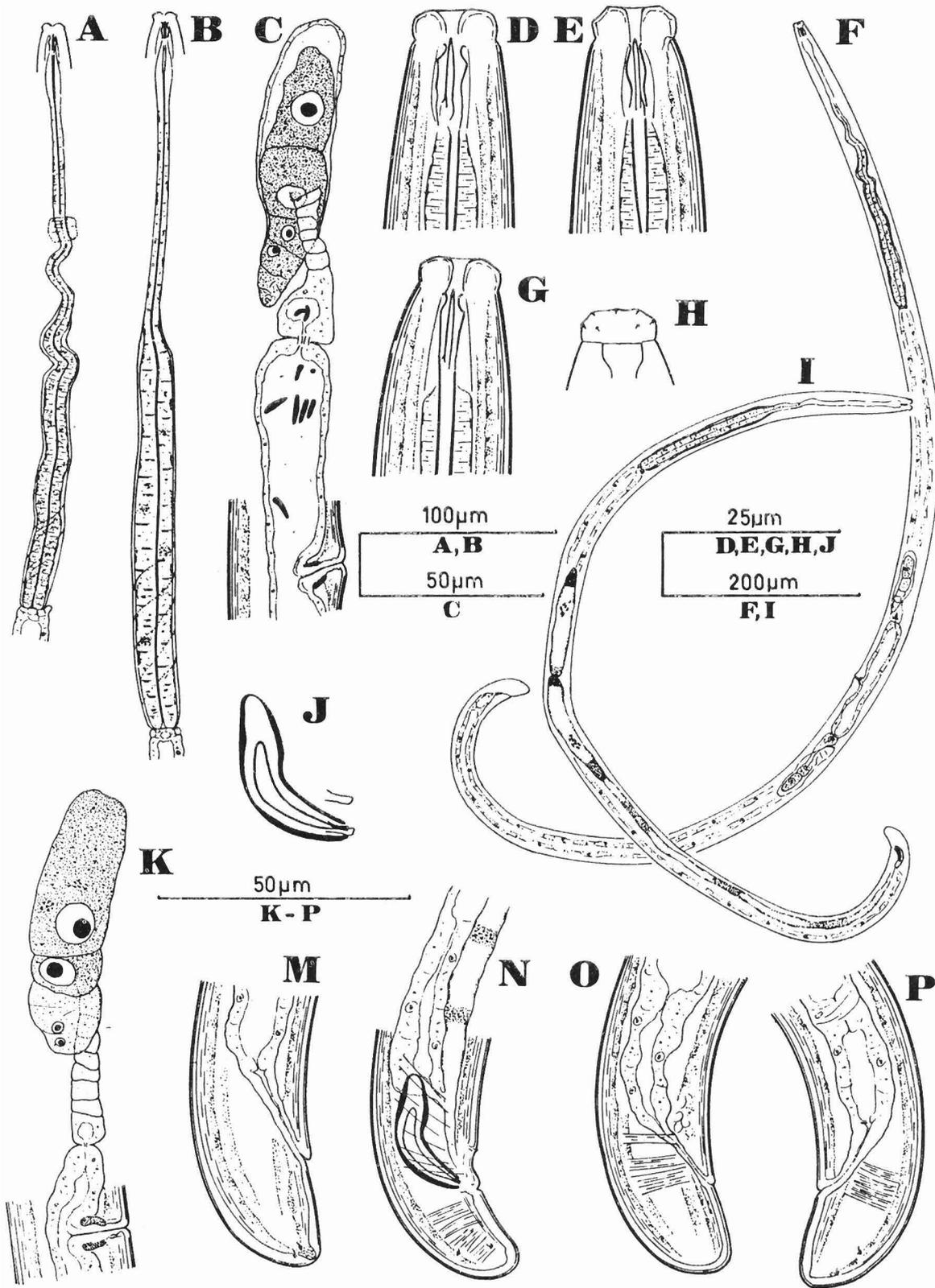


Fig. 1. A-J, N-P: *Nygolaimus seguranus* sp. n. A, B: Neck region; C: Anterior genital branch; D, E, G: Lip region in median view; F: Female entire; H: Lip region in surface view; I: Male entire; J: Spicule and lateral guiding piece; N: Male tail; O, P: Female tail; K: Anterior genital branch of *N. europaeus* (Heyns, 1968) Ahmad & Jairajpuri, 1982 from Yunquera, province of Málaga; M: Female tail of *Nygolaimus* sp. from Sierra de la Cruz, province of Jaén.

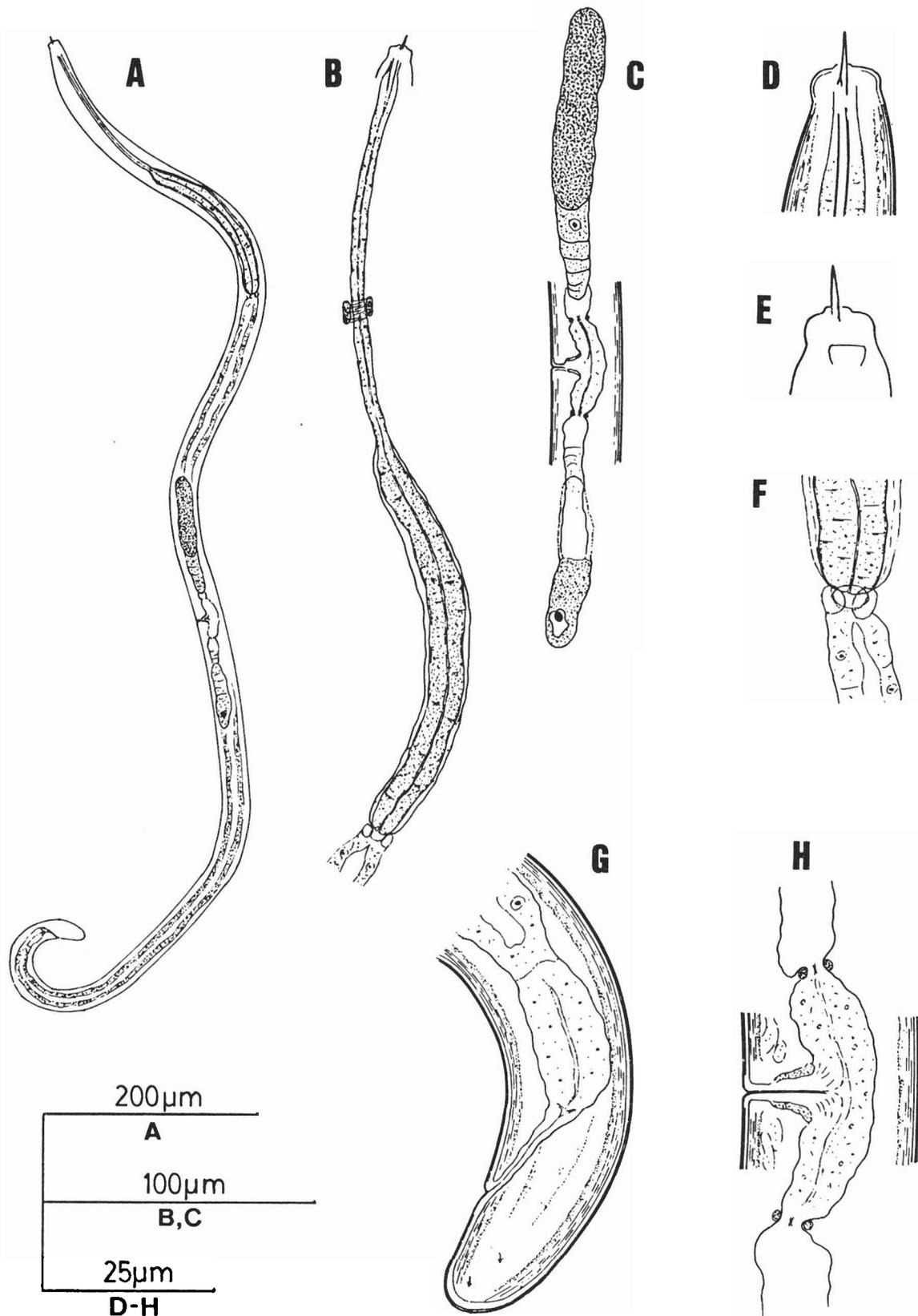


Fig. 2. *Nygolaimus europaeus* (Heyns, 1968) Ahmad & Jairajpuri, 1982 (female, type population). A: Entire; B: Neck region; C: Genital system; D: Lip region in median view; E: Same in surface view; F: Cardia; G: Tail; H: Vagina and uterus.

Table 1. Measurements and diagnostic features of *Nygalaimus seguranus* sp. n. and *N. europaeus* (Heyns, 1968) Ahmad & Jairajpuri, 1982. (All measurements in μm except L in mm).

| Species | <i>N. seguranus</i> sp. n. | | | | | | <i>N. europaeus</i> | | |
|--------------------------------|----------------------------|-----------------------|------------------|-----------------|--------|----------|-----------------------|----------|--------------|
| | Garigue Segura | | | Garigue Pandera | | Meadow* | Type population | Garigue* | <i>Abies</i> |
| | Jaén | | | Jaén | | Jaén | | Málaga | Málaga |
| | Holo-type (female) | Paratypes (5 females) | Allo-type (male) | (female) | (male) | (female) | Paratypes (5 females) | (female) | (female) |
| L | 1.37 | 1.3±0.11 (1.13-1.52) | 1.10 | 1.72 | 1.44 | 1.72 | 1.2±0.04 (1.15-1.28) | 1.30 | 1.15 |
| a | 45.6 | 46.0±7.06 (40.3-59.6) | 40.7 | ? | ? | 48.6 | 47.8±2.41 (45.5-52.4) | 48.4 | 47.9 |
| b | 4.2 | 3.8±0.37 (3.3-4.2) | 3.7 | 4.6 | 3.8 | 4.8 | 4.2±0.20 (4.0-4.5) | 4.3 | 3.8 |
| c | 66.8 | 73.6±6.86 (65.6-84.2) | 50.0 | 84.2 | 70.6 | 78.5 | 67.6±4.26 (61.3-72.6) | 90.1 | 76.6 |
| V(%) | 53.0 | 53.1±2.79 (47.8-57.1) | — | 50.2 | — | 46.7 | 48.8±2.20 (46.2-52.4) | 53.0 | 52.1 |
| G ₁ /T ₁ | 10.1 | 10.4±2.75 (7.6-15.7) | 60.3 | 9.3 | 54.8 | 7.0 | 6.2±2.14 (4.5-10.4) | 5.3 | 7.7 |
| G ₂ /T ₂ | 11.2 | 8.9±2.49 (5.5-12.3) | 39.1 | 7.9 | 33.6 | 9.8 | 5.8±1.67 (3.8-8.6) | 9.0 | 7.9 |
| c' | 1.1 | 0.96±0.08 (0.9-1.1) | 1.3 | 0.9 | ? | 1.0 | 1.1±0.10 (0.9-1.2) | 0.9 | 1.0 |
| Lip region width | 12 | 11.6±0.47 (11-12) | 10.5 | 12.0 | 12.5 | 11 | 10.0 | 11.5 | 11.0 |
| height | 5.5 | 5.1±0.34 (4.5-5.5) | 4.5 | 4.5 | 5.0 | 5.0 | 4.2±0.24 (4.0-4.5) | 5 | 4.5 |
| Amphid aperture | ? | (n=1) 5.5 | 5 | ? | ? | 5 | 4.7±0.23 (4.5-5.0) | ? | ? |
| Mural tooth | | | | | | | | | |
| ventral side | 9 | 9.4±0.02 (8.0-11) | 8.0 | 10.0 | 9.5 | 8.0 | 8.8±0.4 (8-9) | 10.0 | 9.5 |
| dorsal side | 12 | 11.8±0.68 (10.5-12.5) | 9.5 | 11.5 | 11.0 | 9.5 | 9.5±0.63 (8.5-10.0) | 12 | 9.5 |
| Nerve ring-ant. end | 125 | 114.2±13.4 (97-134) | ? | ? | ? | 109 | 91.6±2.24 (88-94) | 100 | 100 |
| Neck length | 328 | 351±15.7 (328-375) | 295 | 376 | 381 | 362 | 289±15 (269-307) | 309 | 300 |
| Phar. bulb length | 182 | 195.2±8.32 (182-205) | 156 | 206 | 206 | 220 | 144±13 (137-170) | 164 | 159 |
| Body width-neck base | 27.5 | 27.5±1.75 (25.3-30) | 27 | 36 | 36 | 30.5 | 25.4±0.73 (24.5-26) | 24.5 | 24 |
| midbody | 30 | 31.8±1.77 (30-35) | 27 | 40 | 35 | 35.5 | 25.6±1.85 (22-27) | 27 | 24 |
| anus | 19 | 19.1±0.58 (18-19.5) | 17.5 | 22 | 26 | 21 | 16.7±0.87 (15.5-18) | 15.5 | 15 |
| Anterior ovary/testis | 63 | 88.5±29.4 (59-138) | 106 | 161 | 166 | 116 | 65.2±21.4 (41-105) | 40.5 | 54.5 |
| Anterior gen. branch | 138 | 136.3±26.1 (109-178) | 663 | 161 | 793 | 121 | 95.1±17.9 (72-125) | 69 | 89 |
| Posterior ovary/testis | 80 | 73.1±45.8 (37.5-178) | 122 | 119 | 145 | 116 | 60.6±11.4 (47.5-80.0) | 94 | 57 |
| Posterior gen. branch | 153 | 121.8±33.2 (83-169) | 430 | 136 | 487 | 169 | 92.1±14.9 (72-114) | 118 | 90.5 |
| Vagina length | 13 | 12.4±0.73 (11-13) | ? | 15.5 | — | 14.5 | 14.7±0.43 (14.5-15.5) | 10.5 | 14.0 |
| Vulva-ant. end | 726 | 707±31.7 (647-747) | ? | 867 | — | 807 | 595±38 (533-638) | 693 | 599 |
| Prerectum | 25 | 22.7±3.34 (19-28) | 30 | 37.5 | ? | 20.5 | 23.2±1.6 (21-25) | 15.5 | 14.5 |
| Rectum/Cloaca | 19 | 18.4±1.52 (16-20.5) | 24 | 21 | 28 | 22 | 19.5±0.89 (18.0-20.5) | 15.5 | 15.5 |
| Tail | 20.5 | 18.2±1.20 (17-20.5) | 22 | 20.5 | 20.5 | 25 | 17.6±1.46 (15.5-20.0) | 14.5 | 15.0 |
| Spicules | — | — | 27 | — | 27.5 | — | — | — | — |
| Lateral guiding pieces | — | — | 5 | — | 5 | — | — | — | — |

* - Specimens whose identification as *N. seguranus* sp. n. or *N. europaeus* is not sure (see the text).

(17-20.5 μm , c=66.6-84.4, c'=0.9-1.1), male without or only a single ventromedian supplement, and spicules 27-27.5 μm long.

N. seguranus sp. n. is very similar to *N. europaeus* (Heyns, 1968) Ahmad & Jairajpuri, 1982 but can be separated from this species by its wider lip region (11-12 vs 10 μm in the type population), longer mural tooth (10.5-12.5 vs 8.5-10 μm), longer neck (328-381 vs 269-304 μm) and pharyngeal bulb (182-206 vs 137-159 μm), wider body (30-35 vs 22-27 μm at

midbody, 18-22 vs 15-18 μm at anus), different morphology of the genital tract, longer uterus (more than the corresponding body width vs 0.5-0.8 body widths), shorter vagina (less than half of the corresponding body width vs more than half), and presence of males with females containing sperm (vs male unknown and females without sperm).

Remarks. The material recorded by Castillo *et al.* (1985) as *Nygalaimus* cf. *europaeus* belongs to the new species described here.

***Nygolaimus europaeus* (Heyns, 1968)
Ahmad & Jairajpuri, 1982
(Fig. 2 & Table 1)**

Type population (five female paratypes):

Female. Very slender nematodes of medium sized, 1.15-1.28 mm long. Body cylindrical, tapering slightly towards both extremities but more so towards the anterior end. Habitus after fixation irregularly ventrally curved: sigmoid, "C"- or "G"-shaped. Outer cuticle layer thin and apparently smooth. Inner layer wider than the outer one along the entire body and thickened at tail region. Lateral chord occupying 25-29% of the midbody diameter. Lateral pores obscure. Lip region probably offset by a weak constriction but its precise morphology was difficult to determine in the specimens examined; it is 2.2-2.4 times as wide as high and about two-fifths of the body diameter at the neck base. Lips amalgamated, somewhat angular. Labial and cephalic papillae not well observed but certainly not interfering with the head contour. Amphid aperture situated behind the cephalic constriction and occupying about half of the corresponding body width; fovea funnel-like. Stoma typical of the genus, with somewhat sclerotized walls. Mural tooth deltoid, always extruded in the specimens examined; its ventral and dorsal sides almost equal in length and little shorter than the lip region width. The pharynx consists of a slender but muscular anterior part expanding gradually into the basal bulb. Pharyngeal bulb cylindrical, 11-14 times as long as wide and occupying about half (47-55%) of the total neck length; it is encapsulated in a conspicuous spiral sheath. Pharyngeal gland nuclei and outlets not clearly visible in the five females studied. Nerve ring located at about one-third (29-34%) of the total neck length. Cardia conoid, practically as long as wide. Cardiac cells ovoid, longer than wide in section. Genital system didelphic-amphidelphic. Ovaries reflexed, relatively short, not reaching the sphincter level. Oviduct joining the ovary subterminally and consisting of a slender distal part of prismatic cells and a poorly developed *pars dilatata*. Junction between oviduct and uterus distinguished by a clear narrowing encircled by the muscular ring. Uterus a very short tube, 0.5-0.8 times the corresponding body diameter; long. Vagina extending inwards more than half of the corresponding body diameter; *pars distalis vaginae* a slender tube 4 μm long with straight walls, shorter (about one half) than *pars proximalis* which is a truncate cone 12 μm long with weakly sigmoid, convergent contour, being surrounded by muscles. Vulva a transverse slit. Prerectum 1.2-1.5 and rectum 1.1-1.2 anal body widths long. Spermatozoa not seen into the genital tract. Tail rounded conoid. Caudal

pores difficult to observe, but apparently two pairs, lateral: one at the middle of the tail, other subterminal.

Male. Unknown.

Specimens (two females) from Sierra de las Nieves, province of Málaga:

One female, collected in association with Spanish fir (*Abies pinsapo* Boissier, 1837) is, in our opinion, conspecific with the type population. The other, collected from a Mediterranean garrigue, has a longer (12 μm) mural tooth and shorter vagina and probably belongs to another species.

Remarks. Our description and morphometrics are in accordance with those of Heyns. Only a few differences in the range of several measurements were evident.

TAXONOMIC NOTES

The new species described here and *N. europaeus* (Heyns, 1968) Ahmad & Jairajpuri, 1982 and *N. captivitatis* (Andrássy, 1962) Ahmad & Jairajpuri, 1982 form a species group which are distinguished from other species in the genus by having present a conspicuous spiral sheath encapsulating the pharyngeal bulb. This feature constitutes an apomorphic state of the character and probably indicates a close evolutionary relationship among the three species. *Nygolaimus europaeus* and *N. securanus* sp. n. are very similar in general morphology and morphometrics (see above), whereas *N. captivitatis* (described on the basis of a single female) differs from these two species by having present a narrowing at the middle of the pharyngeal bulb which may be regarded as being an autapomorphic state.

Conversely, the geographical distribution of the species is at present restricted to Central and Southern Europe, as *N. captivitatis* was found in Hungary (Andrássy, 1962) and Poland (Wasilewska, 1967), *N. europaeus* is known from Germany and Poland (Heyns, 1968) and Spain (present paper) and *N. securanus* sp. n. is known only from Spain. This biogeographical pattern may result from the effect of the quaternary ice age and is indicative of a recent common origin of the species.

Erection of a new genus to accommodate these three species was considered and rejected for two reasons. Firstly, the existence of only one synapomorphy (presence of a spiral sheath encapsulating the pharyngeal bulb) is not sufficient to support the erection of a new taxon of generic level. Secondly, there is a paucity of information available on the geographical distribution of *Nygolaimus* species.

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Peña Santiago R., Liebanas G. Нематоды отряда Dorylaimida из Восточной Андалусии, Испания. Род *Nygolaimus* Cobb, 1913. IV. Описание *N. seguranus* sp. n. и его отношение к *N. europaeus* (Heyns, 1968) Ahmad & Jairajpuri, 1982.

Резюме. Из природных биотопов юго-восточной Испании описывается *Nygolaimus seguranus* sp. n., отличающийся средними размерами тела, (L=1,1-1,7 мм), губной областью диаметром 11-12,5 мкм, отделенной перетяжкой, дельтовидных онхиумом с длиной его дорсальной стороны 10,5-12,5 мкм, почти равным диаметру губной области, пищеводным бульбусом, заключенным в спиральное покрытие, V=47,8-57,1%, закругленным хвостовым концом самки (17-20,5 мкм, c=66,6-84,4, c'=0,9-1,1), хвостом самца с единственным суплементом или без него и спикулами длиной 27-27,5 мкм. Новый вид наиболее близок к *N. europeus* (Heyns, 1968) Ahmad & Jairajpuri, 1982, типовые экземпляры которого также были исследованы при подготовке первоописания. Приводятся иллюстрации и измерения для двух этих видов, кратко обсуждаются их филогенетические связи.
