FOUR SPECIES OF THE GENUS PARATYLENCHUS MICOLETZKY FROM SOUTHEASTERN SPAIN

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Summary. Four species of the genus Paratylenchus are reported from several natural hibitats in southeastern Spain. Two of them have been recorded previously in Spain (P. microdorus and P. vandenbrande). The other two represent the first record from Spain and the second world record after original descriptions (P. ciccaronei and P. sheri). A complete morphometric table and a brief description of each is given and compared with previous data. Illustrations of SEM face view of P. sheri are also provided.

Specimens of the genus *Paratylenchus* Micoletzky, 1922 occur frequently both in natural and agricultural environments. During a study of the plant parasitic nematode fauna of several natural plant communities four species of this genus were collected: *P. ciccaronei*, *P. microdorus*, *P. sheri* and *P. vandenbrandei*. The genus is considered here as in Raski and Luc (1987).

Materials and methods

Identification was done on specimens killed by gentle heat and fixed in a 4% solution of formaldehyde, then dehydrated and prepared to glycerine according to Seinhorst's method (Seinhorst, 1959, 1962). Specimens for SEM observations were processed using Wergin's methods (Wergin, 1981), coated with gold and observed with a JEOL 50 A scanning electron miscroscope at 10 kV of accelerating voltage.

Body length and curved structures were measured with the aid of a precision curvimeter, straight structures such as maximum body width, stylet, anal body width, etc. were measured using a micrometer-scale in the eyepiece of a high power (1250x) microscope.

PARATYLENCHUS CICCARONEI Raski, 1975 (Fig. 1, Table I)

Female. Body slightly ventrally curved. Lateral field with four smooth incisures, $2.9 \pm 0.3 \, \mu m$ (2.7-3.5) wide. Lip region continuous with body contour, rounded-truncate in shape, 4-5 μm wide. Stylet well developed, occupying 26-31 % of oesophageal length. Basal knobs rounded and lat-

erally directed, 3-3.5 μm across. Dorsal gland orifice at 7.8 \pm 0.8 μm (7-9) from stylet base. Oesophagus typical of the genus. Excretory pore located at the beginning of basal bulb. Hemizonid immediately anterior to excretory pore. Cardia rounded. Vulva enveloped by rounded and prominent vulval flaps, 4.5-6 μm long. Vagina anteriorly directed. Postvulvar uterine sac absent. Ovary always outstretched, with a single row of oocytes. Spermatheca oval, 13-20 μm long, filled with rounded sperms. Tail conoid with acute terminus, similar in length to vulva-anus distance

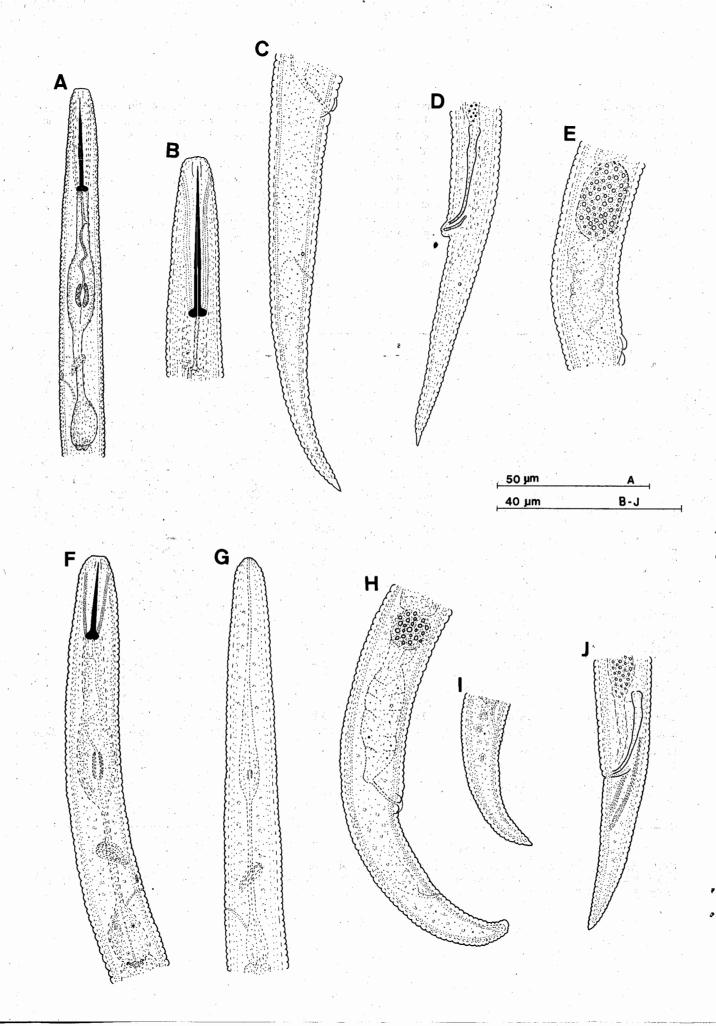
Male. Stylet absent, oesophagus degenerated and non functional. Body narrower than that of female. Spicules and gubernaculum well developed, ventrally curved. Cloacal lips prominent. Tail sharply pointed.

Habitat and locality. Soil around the roots of Quercus rotundifolia Lam., pasture and Populus nigra L. all of them at Sierra de Cazorla, Jaén (southeastern Spain).

Discussion. Morphology and measurements of this population fits with the original description, except in a slight difference in stylet length: 29 µm (27-31) vs 31 µm (27-34) in type population. This species is closed to P. neoamblycephalus but differs in tail shape of both female and male. This is the first record of this species after the original description from Dolceacqua, Italy and could be an indication of a mediterranean distribution.

PARATYLENCHUS MICRODORUS Andrássy, 1959 (Fig. 1, Table II)

Female. Body ventrally arcuate. Lateral field with four smooth incisures. Annuli 1 µm wide at mid-body. Lip re-



gion conoid-rounded, bearing 3-4 annuli and continuous with body contour; 4-5.5 μm wide at base. Stylet moderately enveloped; stylet knobs rounded and laterally directed, 3-3.5 μm wide. Orifice of dorsal oesophageal gland at 5 \pm 0.7 μm (3.5-6) from stylet base. Metacorpus with developed valve, 4-4.5 μm long. Isthmus slender, similar to procorpus in length. Excretory pore located at middle of isthmus. Basal bulb pyriform. Vulva with vulval flaps of 4.8 \pm 0.7 μm (4-6) long. Vagina oblique, 7-8 μm long. Ovary always outstretched, with a single row of oogonia, except in the multiplication zone. Spermatheca rounded, 8-9 μm , filled with rounded sperms. Tail ventrally curved with rounded terminus.

Male. Rare, uncommon. Stylet absent. Oesophagus degenerated. Testis outstretched. Spicules and gubernaculum ventrally curved. Cloacal lips absent.

Discussion. Measurements and description of our specimens are close to the numerous data in the literature (Tarjan, 1960; Brzeski and Szczygiel, 1963; Geraert, 1965; Andrassy, 1985). This species has been recorded frequently in Spain (Bello, 1969; Castillo et al., 1985), but this is the first bisexual population and the only one known after those of Wu (1962) and Szczygiel (1974).

TABLE I - Morphometrics of Paratylenchus ciccaronei (measurements in µm).

	n = 14 females			n = 4 males			
	$\bar{X} \pm SD$	Range	CV %	$\bar{X} \pm SD$	Range	CV %	
L	359 ± 41.6	289 – 444	11.6	385 ± 37.6	341 – 422	9.8	
a	24.9 ± 2.4	20.6 - 29.7	9.7	28.5 ± 1.7	26.8 - 30.7	6.0	
b	3.7 ± 0.3	3.1 - 4.3	8.3	4.3 ± 0.5	3.8 - 4.9	11.6	
b_1	5.8 ± 0.4	5.3 - 6.5	6.1	_	_		
V or T	80 ± 2.0	77 - 82	2.5	32 ± 58	24 - 38	18.4	
G_1	33 ± 4.0	28 – 38	12.2	-	_	, · · · · <u>· · · · · · · · · · · · · · ·</u>	
c	10.7 ± 1.2	8.1 - 12.0	10.8	8.5 ± 0.5	7.8 - 8.8	5.7	
c'	3.7 ± 0.6	3.0 - 4.9	15.8	4.7 ± 0.2	4.4 - 4.8	4.0	
stylet	29 ± 1.4	27 - 31	4.9	- E, 1	- '.		
m	68 ± 2.6	63 - 74	3.8	- • • • • • • • • • • • • • • • • • • •	– , ,		
S	2.6 ± 0.2	2.2 - 3.1	8.5	<u>-</u>		·	
0	27 ± 2.7	23 - 30	10.0	-	2	_ ',	
MB	58 ± 1.4	56 – 60	2.5	<u> </u>	- :		
metacorpus	23 ± 4.1	17 - 30	17.7	<u>-</u>		_: _:	
isthmus	19 ± 2.2	16 - 23	11.7	-			
basal bulb	12 ± 1.5	10 - 15	12.2	_	-	_	
oesophagus	97 ± 8.5	88 - 118	8.8	90 ± 12.2	75 - 103	13.5	
nerve ring	70 ± 7.0	61 – 86	10.0	74 ± 2.8	72 - 76	3.8	
excretory pore	77 ± 8.5	67 – 94	11.0	81 ± 2.8	79 - 83	3.5	
max. body width	14 ± 0.8	13 – 16	5.8	14 ± 1.2	13 – 15	9.1	
anal body width	9 ± 1.2	8 - 12	12.4	10 ± 1.2	8 - 11	12.0	
tail length	34 ± 7.2	27 - 49	21.2	45 ± 4.3	39 – 48	9.5	
Vulva-Anus (VA)	34 ± 2.7	30 - 37	7.9		<u> </u>		
tail/VA	1.0 ± 0.2	0.7 - 1.3	17.6	**	** .	· · · · · · ·	
spicules	- ; . :	i	-	22 ± 1.3	20 - 23	5.8	
gubernaculum		- 1		4.5 ± 0.4	3.9 - 4.7	8.9	

Fig. 1 - (Front page). Paratylenchus ciccaronei A-E: A, female oesophageal region; B, female anterior region; C, female posterior region; D, male tail; E, vulval region. P. microdorus F-J: F, female oesophageal region; G, male oesophageal region. H, female posterior region; I, female tail; J, male tail.

PARATYLENCHUS SHERI (Raski, 1973) Siddiqi, 1986 (Figs. 2-3, Table III)

Female. Lip region conoid-truncate, with an unstriated depression from body contour (4-5.5 μm wide) and strong sclerotization. Small projecting oral lips present. SEM face view (Fig. 3) shows an unstriated, dorso-ventrally flattened head. Stylet robust, occupying 19 (15-23) annuli. Stylet knobs weakly backwardly directed, 4.8 (4-5.5) μm across. Lateral field with four incisures with smooth margins (central two are very faint), 3.2 (2.5-4) μm wide. Orifice of dorsal oesophageal gland 5.9 (4.5-6.5) μm from stylet base. Oesophagus typical of the genus. Metacorpus with well developed valvular apparatus 5.6 (4.5-7.5) μm long, its posterior margin situated at 70 (63-80) μm from anterior end. Excretory pore located near anterior end of basal bulb, immediately posterior to hemizonid. Cardia well developed, 2.5-3.5 μm wide.

Distinct cuticular vulval flap, 6 (5-7) μ m long. Female reproductive system outstretched, 179 (101-268) μ m long; with large round spermatheca 13.5 (11.5-15.5) μ m wide, filled with sperms 1-2 μ m wide. Ovary consisting of a single row of 20 (18-24) oocytes. Tail almost straight to slight ventrally curved, with acute rounded terminus, 0.8 (0.6-1.3) times vulva-anus distance or 3.8 (3.1-4.7) times anal body diameter.

Males. Less common than females. Similar morphology to females except in the absence of stylet and degenerate, non functional oesophagus; metacorpus hardly or not distinguishable. Testis outstretched, 177 (142-196) μm long. Spicules cephalated, ventrally curved.

Female juveniles. Similar morphology to that of adult females, except sexual characters and shorter body length and stylet (even in fourth stage or preadult).

TABLE II - Morphometrics of Paratylenchus microdorus (measurements in µm).

į.	n = 16 females				n = 3 males		
	$\tilde{X} \pm SD$	Range	CV %	$\hat{X} \pm SD$	Range	CV %	
L	359 ± 44.5	289 – 436	12.4	348 ± 42.7	300 – 381	12.3	
a e e e e e e e	24.5 ± 2.6	20.6 - 29.0	10.9	26.1 ± 2.6	23.1 - 28.0	10.1	
b	4.2 ± 0.4	3.5 - 5.2	10.2	4.0 ± 0.4	3.6 - 4.3	9.4	
b_1	6.8 ± 0.6	5.7 - 7.9	8.6	,	-		
V or T	82 ± 1.6	80 - 85	1.9	26 ± 8.0	17 - 33	31.5	
G_1	41±8.9	30 – 60	21.9		_		
c	11.7 ± 1.4	8.9 - 14.5	11.9	11.7 ± 1.8	10.0 - 13.5	15.0	
c'	3.3 ± 0.5	2.7 - 4.3	15.8	3.3 ± 0.4	3.0 - 3.8	12.5	
stylet	15 ± 0.9	14 - 16	6.0	_	_		
m	60 ± 2.3	55 – 62	3.8		· · · · · · ·	<u>-</u>	
S	1.6 ± 0.1	1.6 - 1.7	3.2	_	_		
D.G.O.	5.0 ± 0.7	3.5 - 6.0	14.7	<u> </u>		· —	
0	33 ± 5.0	23 - 40	15.0		_	10 <u>1</u> . 4	
MB	53 ± 2.4	49 – 56	4.5	_	_	_	
procorpus	16 ± 2.3	14 - 20	14.3	_	_	_ 1	
metacorpus	17 ± 2.3	14 - 20	13.2	_	_		
isthmus	19 ± 1.6	17 - 21	8.9	<u> </u>	_ '	· <u>-</u>	
basal bulb	12 ± 1.1	10 - 14	9.6		- 1 - 1 - 1 - 1 - 1 - 1 - 1 - 1 - 1 - 1	÷	
oesophagus	85 ± 5.5	75 – 95	6.5	87 ± 2.5	84 - 89	2.9	
nerve ring	61 ± 3.6	53 – 68	6.0	54 ± 4.5	50 - 59	8.3	
excretory pore	73 ± 5.6	60 - 83	7.7	67 ± 5.5	61 - 72	8.3	
max. body width	15 ± 1.3	12 - 17	9.2	13.3 ± 0.6	13 - 14	4.3	
anal body width	9 ± 0.7	8.5 - 11	7.4	9 ± 1.0	8 - 10	11.1	
tail	31 ± 5.2	24 – 39	16.9	30 ± 6.6	26 - 38	21.9	
Vulva-anus (VA)	34 ± 9.3	20 - 49	27.8	, t	_		
tail/VA	1.0 ± 0.2	0.7 - 1.3	19.6	- · · · · -	_	 .	
spicules	<u> </u>	<u> </u>	_	20.6 ± 0.6	20-21	2.8	
gubernaculum	_	_	_	3.8 ± 0.3	3.5 - 4.0	7.5	

Habitat and locality. Soil around the roots of the following hosts: Quercus rotundifolia Lam., Q. faginea Lam., Rosmarinus officinalis L., Populus nigra L., Pinus pinaster Aiton, P. nigra L. and pasture, all of them at Sierra de Cazorla, Jaén, Spain.

Discussion. The species was described by Raski (1973) from grass and weed soil, two miles South of Digne (France). Since then it has not been recorded; this second world record in Southeastern Spain is confirmation of its presence in the Mediterranean area as was established by Raski (1973). Morphology and morphometry of our specimens are in close conformity with those of the original description, the strong cephalic sclerotization being very evident. Identification of our specimens has been kindly confirmed by Prof. D. J. Raski (Davis). Two females have been deposited in the nematology collection of the Department of Nematology, University of California, Davis, USA.

P. sheri can be differentiated from P. perlatus Raski, 1975 in the shape of the lip region (conical with protruding

lips surrounding the oral aperture vs truncate), head sclerotization light and stylet length (21-25 μm vs 19-22). From P. arculatus Luc et De Guiran, 1962 can be differentiated in head shape (truncate) and tail length (c = 7.8-13 vs = 15-18).

PARATYLENCHUS VANDENBRANDEI De Grisse, 1962 (Fig. 2, Table IV)

Female. Body ventrally curved in C shaped. Lateral field with three lines, central being very faint. Lip region round-truncate, 2.5-3 µm wide. Stylet comparatively long, occupying the procorpus completely or 29-38% of oesophageal length. Metacorpus muscular with long valve, 5-5.5 µm long. Basal bulb pyriform, moderately developed. Excretory pore at end of isthmus. Hemizonid immediately anterior to excretory pore.

Vulva enveloped by a vulval cuticular flap, 3.5 ± 0.4 μm (3-4) long. Ovary outstretched, with a single row of oogonia even in the multiplication zone. Spermatheca almost spherical, 7.5-9 μm wide, filled with rounded sperms

Table III - Morphometrics of Paratylenchus sheri (measurements in μm).

	n = 26 females				n = 7 males	
	$\bar{X} \pm SD$	Range	CV %	$\bar{X} \pm SD$	Range	CV %
L	463 ± 44	352 – 531	9.5	484 ± 37	434 – 537	7.7
a	23.9 ± 2.4	18.2 - 28.8	10.1	30.2 ± 1.6	28.4 - 32.9	5.3
b	4.2 ± 0.4	3.4 - 5.1	9.4	4.7 ± 0.7	3.8 - 6.0	15.0
b_1	6.7 ± 0.6	5.5 - 8.2	9.5	_	_	
V or T	79 ± 1.3	78 - 82	1.6	36 ± 3.1	31 - 40	8.5
G_1	38 ± 9.1	21 – 54	32.9		<u> </u>	· · · · · · · · · · · · · · · · · · ·
c	10.9 ± 1.2	7.8 - 13.0	11.1	10.1 ± 1.6	9.6 - 13.8	14.4
c'	3.8 ± 0.5	3.1 - 4.7	12.9	4.1 ± 0.4	3.5 - 4.9	10.4
stylet	23 ± 1.0	21 - 25	4.5	- :	-	· –
conus	14 ± 1.0	12 – 15	7.0	· –	_	- ,
S	1.7 ± 0.1	1.5 – 1.9	8.2		<u> </u>	-
D.G.O.	5.9 ± 0.8	4.7 - 6.7	13.1	–	-	_
0	26 ± 4.1	20 – 39	15.8		. <u> </u>	-
metacorpus	22 ± 2.5	17 - 28	11.6	-	· ·	
isthmus	23 ± 2.2	18 - 28	9.8	-	-	_
basal bulb	14 ± 1.3	13 – 17	9.4	-	<u> </u>	
oesophagus	109 ± 7.5	94 - 121	6.8	103 ± 12	88 – 116	11.6
nerve ring	77 ± 5.7	63 – 86	7.4	,		-
excretory pore	93 ± 9.7	73 - 114	10.4	88 ± 10.6	76 – 100	12.1
annuli	1.5 ± 0.3	1-2	16.5	1.6 ± 0.4	1.3 - 2	11.4
tail	43 ± 5.5	31 – 57	12.8	44 ± 5.1	35 – 49	11.6
tail annuli	27 ± 4.8	20 - 37	17.8	– ,		'. <u>-</u>
spicules	· - · - ·		-	26 ± 1.2	24 – 28	4.7
gubernaculum	. ¹ ,	– ' - '	-	5.5 ± 0.3	5-6	5.4

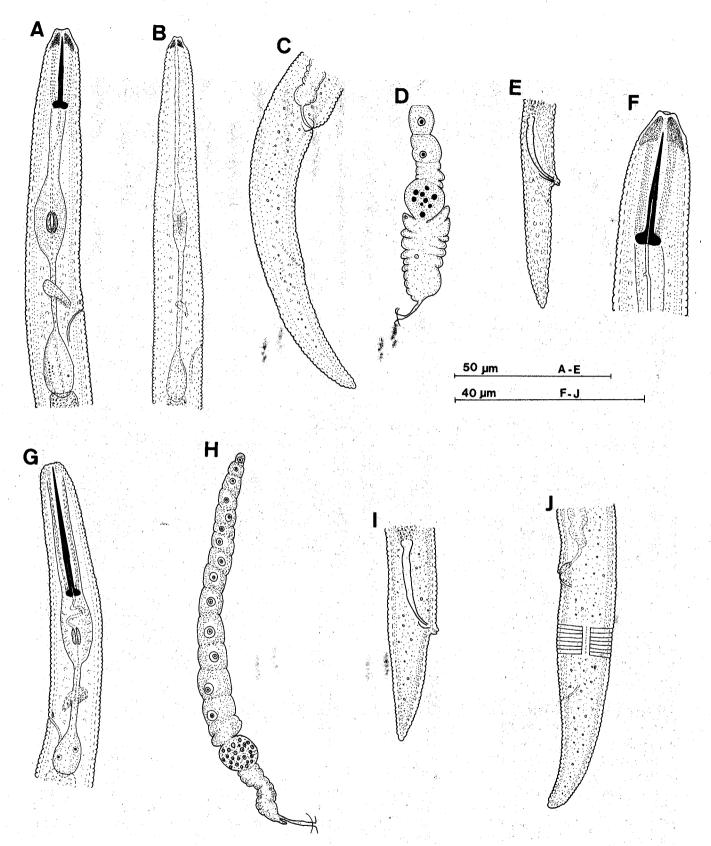


Fig. 2 - Paratylenchus sheri A-F: A, female oesophageal region; B, male oesophageal region; C, female posterior region; D, detail of female gonad; E, male tail; F, female anterior region. P. vandenbrandei G-J: G, female oesophageal region; H, female gonad; I, male tail; J, female posterior region.

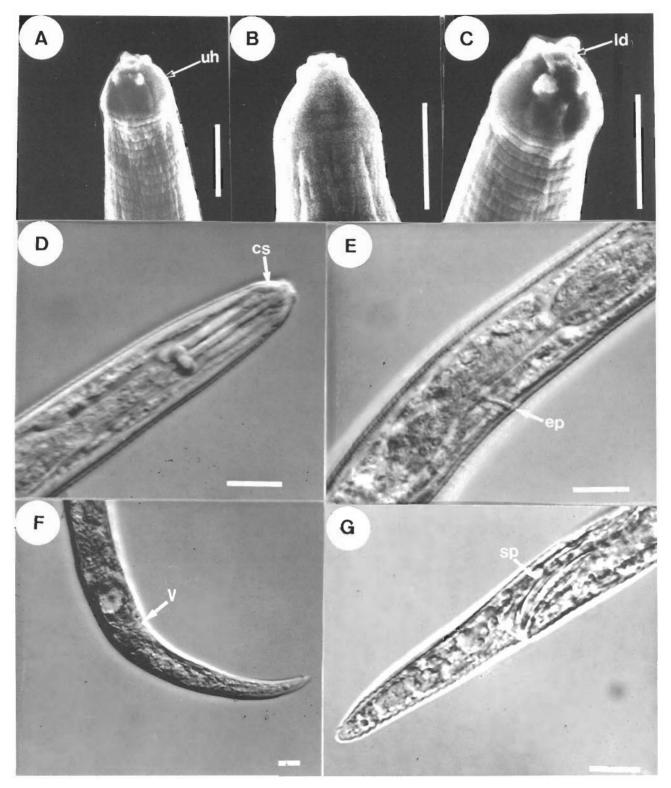


Fig.3 - Paratylenchus sheri. A-C: SEM micrographs. A, female anterior region; B and C, face views. D-G: ligth micrographs, D, female anterior region; E, female oesophageal region; F, female tail; G, male tail. (Scale bars: A-C = $5 \mu m$, D-G = $10 \mu m$). (uh = unstriated head; ld = labial disc; cs = cephalic sclerotization; ep = excretory pore; V = vulva; sp = spicules).

of about 1 µm in diameter. Postvulval uterine sac reduced. Tail conoid, with rounded terminus.

Male. Similar to female except in the absence of stylet and degenerated and non-functional oesophagus. Cloacal lips developed. Spicules and gubernaculum ventrally curved, well developed.

Discussion. This is a quite distinctive species of the genus by the presence of three lateral incisures of the lateral

field, a well posterior position of the vulva (80-86%) and the comparatively long stylet (25-29 μ m). Morphology of our specimens fits well with original description of De Grisse (1962) and measurements are similar to that of Raski (1975). It has been previously recorded in Spain by Artero (1976) on ornamental plants at Malaga (South Spain).

We thank Dr. N. Vovlas (Bari) for this help in taking the SEM photos and Prof. D.J. Raski (Davis) for confirming the identity of *P. sheri*.

TABLE IV - Morphometrics of Paratylenchus vandenbrandei (measurements in µm).

· •	$\tilde{X} \pm SD$	Range	CV %	n = 1 male
L	251 ± 21.5	219 – 293	8.6	333
a	19.2 ± 1.8	16.8 - 23.4	9.3	262
b	3.4 ± 0.3	2.8 - 3.9	8.1	3.8
b_1	5.2 ± 0.4	4.2-5.6	7.8	·
V or T	82 ± 1.5	80 – 86	1.9	22
G_1	33 ± 9.1	21-56	27.7	
c	13.3 ± 2.6	10.3 - 19.9	20.0	13.9
c'	2.3 ± 0.3	1.7 - 2.7	14.2	2.7
stylet	27 ± 1.2	25 – 29	4.3	· –
m	68 ± 2.7	62 – 72	4.0	_
S	2.7 ± 0.3	2.2 - 3	10.1	
D.G.O.	4.7		<u>-</u>	<u> </u>
0	17		<u> </u>	_
metacorpus	16 ± 4.0	13 – 26	24.3	· <u> </u>
isthmus	14 ± 2.0	11-18	14.2	. –
basal bulb	9 ± 2.1	7 – 13	22.3	
oesophagus	74 ± 8.6	66 – 98	11.5	
nerve ring	53 ± 5.5	46 – 67	10.2	, * * ₁ —
excretory pore	61 ± 6.7	51 – 75	11.0	· · · —
max. body width	13 ± 1.0	11 – 15	7.5	12.7
anal body width	8.5 ± 1.1	6-10	12.7	8.7
tail	20 ± 4.1	13 - 27	20.8	24.
lateral field	2.1 ± 0.4	1.3 - 2.7	20.7	·
annuli	0.9 ± 0.1	0.8 - 1.0	13.3	· -
spicules	<u> </u>	$(\mathcal{F}_{\omega_{0}})$, $\mathcal{F}_{\omega_{0}}$	- · · · · · · · · · · · · · · · · · · ·	23
gubernaculum	<u> </u>		_ , .	4.7

Literature cited

Artero J., 1976 - Estudio de la nematofauna de las plantas ornamentales introducidas en la provincia de Malaga. Tesis Doctoral, Fac. Cienc., Univ. Granada, 331 pp.

Andrassy I., 1985 - Paratylenchus microdorus. C.I.H. Description of Plant-parasitic Nematodes. Set 8 No. 107, 2 pp. Farnham Royal IIK Commonwealth Agricultural Burgaux.

Royal, UK, Commonwealth Agricultural Bureaux.

Bello A., 1969 - Estudio de la nematocenosis de las Islas Canarias e influencia del factor antropogeno sobre las mismas. Bol. R. Soc. Esp. Hist. Nat., 67: 35-52.

CASTILLO P., PENA R. and JIMENEZ MILLAN F., 1985 - Modelos de distribucion vertical de las especies de nematodos en un biotopo natural. I. Bol. Serv. Plagas, 11: 155-162.

Brzeski M. W. and Szczyciel A., 1963 - Studies on the nematodes of the genus Paratylenchus Micoletzky (Nematoda: Para-

tylenchinae) in Poland. Nematologica, 9: 613-625.

DE GRISSE A., 1962 - Paratylenchus vandenbrandei n. sp. (Nematoda: Criconematidae) nouvelle espèce de Paratylenchus associée aux racines d'agave au Kenya. Nematologica, 8: 229-232.

GERAERT E., 1965 - The genus Paratylenchus. Nematologica, 11:

301-334.

- RASKI D.J., 1973 Paratylenchoides gen. n. and two new species (Nematoda: Paratylenchidae). Proc. helminth. Soc. Wash, 40:
- RASKI D.J., 1975 Revision of the genus Paratylenchus Micoletzky, 1922 and descriptions of new species. Part II of three parts. J. Nematol., 7: 274-295.

RASKI D.J. and Luc M., 1987 - A reappraisal of Tylenchina (Ne-

the in

mata) 10. The superfamily Criconematoidea Taylor, 1936. Revue Nématol., 10: 409-444.

Seinhorst J.W., 1959 - A rapid method for the transfer of nematodes from fixative to anhydrous glicerine. Nematologica, 4: 67-69.

Seinhorst J.W., 1962 - On the killing, fixation and transferring to glycerine of the nematodes. Nematologica, 8: 29-32.

Szczygiel A., 1974 - Plant parasitic nematodes associated with strawberry plantations in Poland. Zesz. Problem. Post. Nauk. Roln., 154: 9-132.

TARJAN A.C., 1960 - A review of the genus Paratylenchus Micoletzky, 1922 (Paratylenchinae: Nematoda) with a description of two new species. Ann. New York Acad. Sci, 84: 329-390.

Wergin W.P., 1981 - Scanning electron microscopic techniques and applications for use in nematology, pp. 175-204, Vol. 3. In: Plant Parasitic Nematodes (B.M. Zuckerman and R.A. Rhode, eds), Academic Press, New York and London. Wu L.Y., 1962 - Paratylenchus brevihastatus n. sp. (Criconema-

tidae: Nematoda). Čan. J. Zool., 40: 391-393.