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## NEW RECORD OF *PUNCTODERA PUNCTATA* FROM SPAIN

by

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**Summary.** During a nematode survey of wet habitats in a mountainous area in the south of Spain an abundant population of a cyst nematode species, identified as *Punctodera punctata* (Thorne) Mulvey *et* Stone was found. The morphology and morphometry of cysts, juveniles and males are briefly described.

During a survey of the Jándula riverbed, in Sierra Morena, Andújar (Jaén) in southern Spain, an abundant population of cysts, second-stage juveniles and males of *Punctodera punctata* (Thorne) Mulvey *et* Stone was found in association with natural aquatic Gramineae (not determined) in the sandy soil of the riverbed. This is the first report of the species from Spain.

Soil samples from the riverbed were collected in January, 1989, when the volume of the river was at its minimum. Cysts, juveniles and males were extracted by means of centrifugation in sugar according to De Grisse (1969) with the modification of Nombela and Bello (1983). The number of cysts per sample was very high (more than 300 per 100 cc of soil), and most of them were full of oval elongate eggs.

### Description

*Cysts* (n = 20): L (excluding neck) =  $542 \pm 66 \mu\text{m}$  (438-714); maximum width =  $334 \pm 64 \mu\text{m}$  (228-480); L/W ratio =  $1.6 \pm 0.2$  (1.3-2.3); medium diameter of fenestrae =  $24.8 \pm 1.7 \mu\text{m}$  (19.4-29.1); distance between fenestrae =  $52 \pm 12 \mu\text{m}$  (38-89).

Light brown in colour, round to ovoid in shape with a long and narrow neck, without prominent vulval cone (Figs. 1 E and 2 A). Circumfenestrated with the two fenestrae (vulval and anal) approximately of the same size (Fig. 2 C), separated by a distance 1.5-3 times longer than diameter of fenestrae, without bullae. Cuticular marks with abundant punctations (Fig. 2 B).

*Second-stage juveniles* (Table I). Head bearing 4 annuli, offset from body contour (Fig. 1 A) and measuring  $3.4 \pm 0.7 \mu\text{m}$  (2.9-4.3) high and  $8.5 \pm 0.8 \mu\text{m}$  (7.5-10) wide. Stylet strong, conus as long as shaft; basal knobs with an-

teriors surfaces slightly concave,  $4.5\text{-}5 \mu\text{m}$  across. Cuticle prominently annulated, annuli about  $2 \mu\text{m}$  wide at mid-body. Lateral fields  $5\text{-}6 \mu\text{m}$  wide, with four lines, areolated in outer bands reducing to two before anus. Orifice of dorsal oesophageal gland  $7\text{-}8 \mu\text{m}$  from base of stylet. Oesophagus strongly developed, occupying about a third of body length. Hemizonid two annuli long, immediately anterior to excretory pore. Median oesophageal bulb oval,  $15\text{-}17 \mu\text{m}$  long  $\times$   $8\text{-}9 \mu\text{m}$  wide. Distance from anterior end to base of median oesophageal bulb  $76.7 \pm 6.8 \mu\text{m}$  (65.5-91.5). Tail long and pointed with rounded terminus and conspicuous hyaline region (Fig. 1 C). Tail length/hyaline region ratio =  $1.5 \pm 0.1$  (1.3-1.7) and hyaline region/stylet length ratio =  $2 \pm 0.2$  (1.7-2.4). Phasmids 2 annuli posterior to anus level.

*Males* (Table I). Body vermiform, C shaped, narrowing in the anterior end. Head bearing 5-6 annuli and offset from body. Stylet robust, basal knobs rounded  $4.5\text{-}5 \mu\text{m}$  across, conus as long as shaft. Orifice of dorsal oesophageal gland  $6\text{-}7 \mu\text{m}$  from stylet base. Median oesophageal bulb narrow, oval (Fig. 1 B), occupying 39-45% of body width at the same level;  $16\text{-}20 \mu\text{m}$  long  $\times$   $9.5\text{-}11 \mu\text{m}$  wide. Hemizonid inconspicuous, 2 annuli before excretory pore. Testis outstretched, with 3-4 rows of spermatogonia. Spicules convergent, ventrally curved with rounded tip (Fig. 1 D).

*Eggs*. Oval elongate, embryonated with folded juveniles, measuring (n = 25): L =  $112.8 \pm 4.1 \mu\text{m}$  (108-121); W =  $51.4 \pm 3.9 \mu\text{m}$  (47-63); L/W =  $2.2 \pm 0.1$  (1.8-2.4).

### Discussion

Measurements and descriptions of the three stages of our population are similar to those given by Webley and

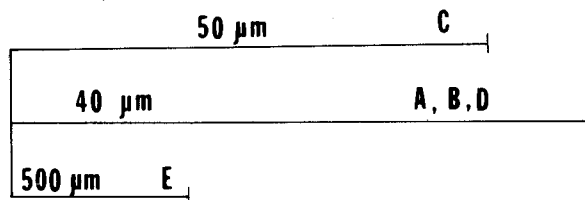
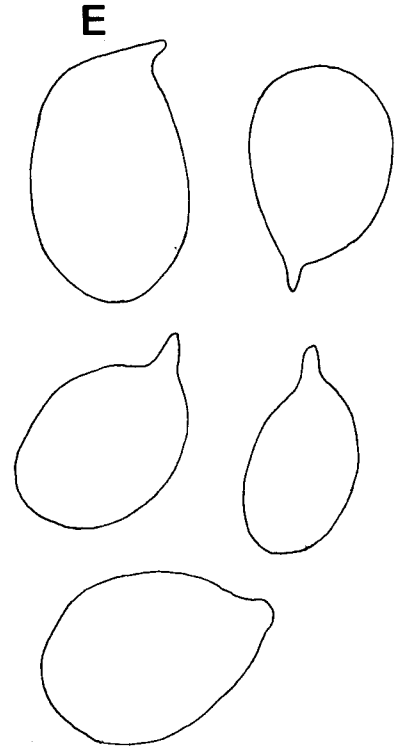
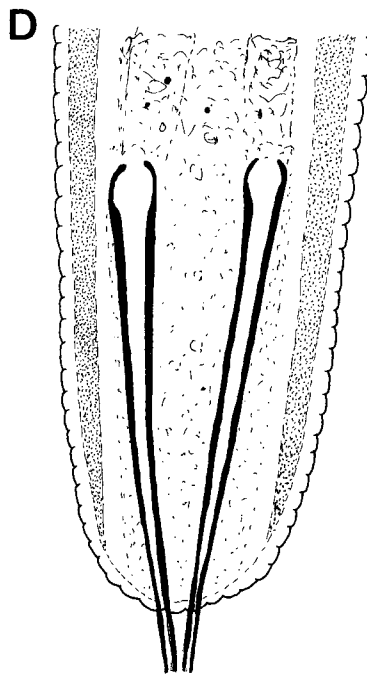
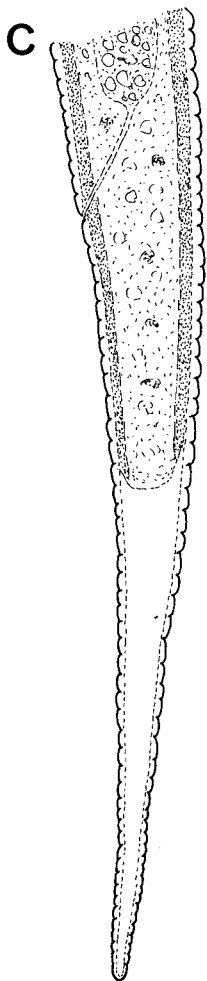
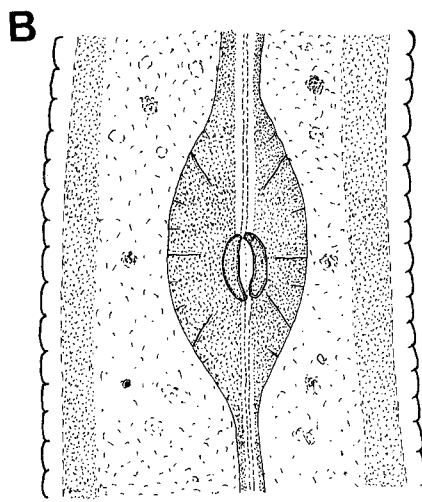
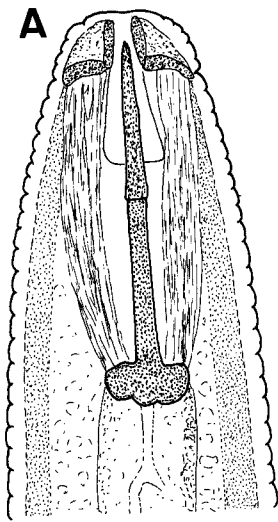


Fig. 1 - *Punctodera punctata*: A, head end of second-stage juvenile; B, median oesophageal bulb of male; C, tail of second-stage juvenile; D, tail end of male (frontal view); E, cysts.

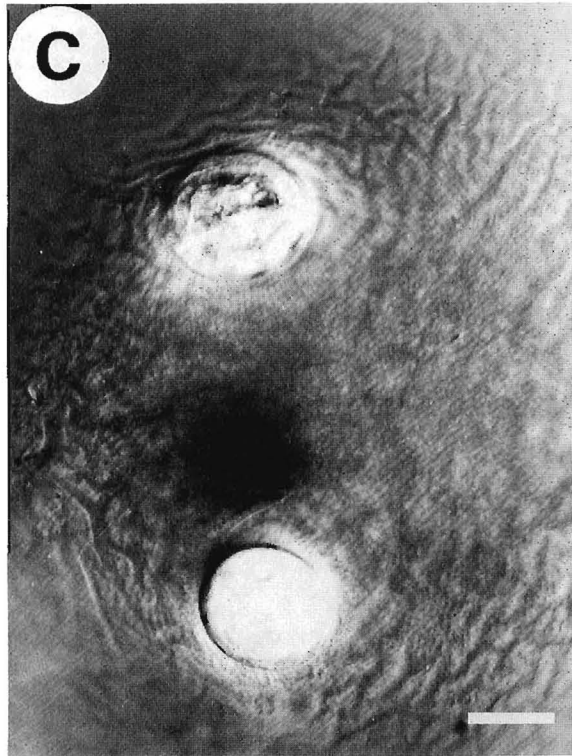
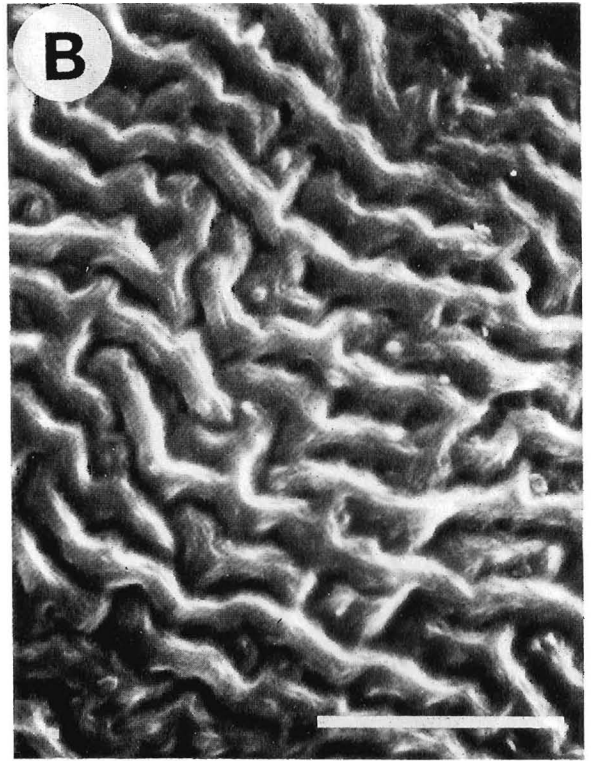
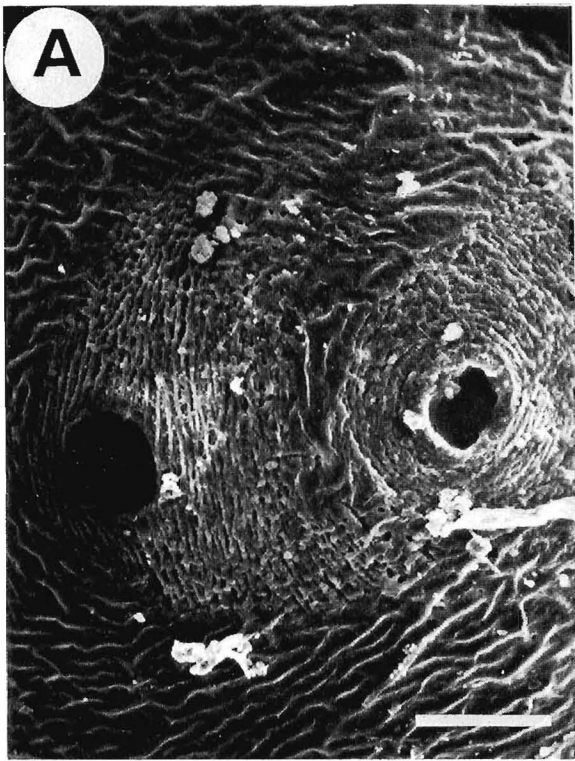


Fig. 2 - *Punctodera punctata*: A, SEM micrograph of vulval cone, 845  $\times$  (scale bar = 20  $\mu$ m); B, cuticular pattern of cyst, 3380  $\times$  (scale bar = 10  $\mu$ m); C, photomicrograph of vulval and anal fenestrae, 500  $\times$  (scale bar = 20  $\mu$ m).

TABLE I - *Morphometric data (in  $\mu\text{m}$ ) of Punctodera punctata.*

	Second-stage juveniles n = 20			Males n = 18		
	$\bar{X} \pm \text{SD}$	Extr. Val.	CV %	$\bar{X} \pm \text{SD}$	Extr. Val.	CV %
L	533 $\pm$ 16	501 - 559	3.0	1036 $\pm$ 129	803 - 1240	12.5
a	26.6 $\pm$ 1.5	24.0 - 29.6	5.6	33.4 $\pm$ 3.4	28.7 - 39.8	10.2
b	2.5 $\pm$ 0.1	2.3 - 2.7	6.3	5.8 $\pm$ 0.5	5.1 - 6.8	9.3
T	-	-	-	51 $\pm$ 7.2	39 - 58	14.2
c	7.0 $\pm$ 0.3	6.6 - 7.8	4.6	subterminal	-	-
c'	5.4 $\pm$ 0.5	4.2 - 6.3	9.8	»	-	-
Stylet	24.7 $\pm$ 1.1	23 - 27	4.6	25.5 $\pm$ 2.4	21 - 28	9.5
DGO	7.1 $\pm$ 1.1	7 - 8	5.3	6.5 $\pm$ 0.7	6 - 7	10.7
MB (*)	33 $\pm$ 1.6	31 - 36	4.9	48 $\pm$ 2.8	45 - 52	5.7
Nerve ring	93 $\pm$ 6.7	84 - 102	7.2	103 $\pm$ 13.0	83 - 119	12.6
Excretory pore	107 $\pm$ 4.9	97 - 114	4.5	133 $\pm$ 16.1	105 - 156	12.0
Oesophagus	209 $\pm$ 13.1	193 - 240	6.3	175 $\pm$ 20.7	145 - 212	11.8
Maximum body width	20 $\pm$ 0.9	18.5 - 21.5	4.4	31 $\pm$ 2.5	27 - 34	8.0
Anal body width	14 $\pm$ 1.4	12 - 18	9.9	-	-	-
Tail length	76 $\pm$ 3.6	67 - 83	4.8	-	-	-
Tail hyaline region	50 $\pm$ 4.2	41 - 58	8.4	-	-	-
Spicules	-	-	-	34 $\pm$ 1.8	31.5 - 37	5.3
Gubernaculum	-	-	-	8.9 $\pm$ 0.2	8.5 - 9	2.8

(\*) MB = distance of median bulb anterior end  $\times$  100/total oesophageal length.

Lewis (1977) except for the slightly shorter stylet of males and second-stage juveniles and the longer distance between dorsal oesophageal gland and stylet knobs. According to the study of several populations of *P. punctata* by Wouts *et al.* (1986) our measurements of juveniles are similar to those of the Rothamsted population except in the shorter stylet. Therefore our Jándula population can be included in the second form proposed by these authors.

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