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# THE MORPHOLOGY OF HETERODERA AVENAE IN SPAIN

by

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A survey of nematodes associated with cereal crops in Spain has been in progress since 1971. From a total of 350 soil and root samples examined to date *Heterodera avenae* Woll. occurred in 35% of samples from wheat (*Triticum* sp.), 30% from barley (*Hordeum vulgare* L.) and oats (*Avena sativa* L.), and 5% from maize (*Zea mays* L.) samples; the nematode has never been found in soil collected from rye (*Secale cereale* L.).

From a detailed study of the nematode specimens, some small morphological differences were identified which suggested there were three different types present, named here as A, B and C (Table I); the terminology does not have any relationship with pathotypes.

Nematodes in the roots were detected by staining in lactophenol and acid fuchsin. Cysts were extracted from the soil by a Fenwick apparatus. Measurements and descriptions are based on specimens mounted in clear lactophenol.

## OBSERVATIONS

Type A seems to be widespread throughout Spain. Three populations from the province of Lérida were selected for morphological examination. Individuals were abundant in both soil and root samples (Table I). The females are lemon-shaped with prominent neck and vulval cone, and pearl white in colour. Head is set off from neck, bearing a prominent labial disc plus a single distinct annule. Spear straight to slightly arcuate. Prorhabdion slightly less than half stylet length. Zig-zag rugose cuticular surface pattern. Metacorpus strongly developed with well developed valves. No egg-sac present.

Cysts are also lemon shaped, but dark brown, almost black, partially covered with a thick, white subcrystalline layer. Wall pattern consisting of irregular zig-zag lines with irregularly arranged punctations. Ambifenestrate with short vulval slit. No underbridge observed. Bullae prominent crowded beneath vulval cone (Fig. 1a).

Eggs cylindrical with rounded ends, with hyaline shell and larval stages folded within four times.

The second stage larvae are vermiform with pointed tail, lip region rounded, set off, 3-4  $\mu$ m high and 8-9  $\mu$ m wide, with labial disc and three indistinct post-labial annules. Cephalic framework heavily sclerotized. Body annules distinct. Lateral fields with 4 incisures forming three bands of which the outers are areolated and disapear near the middle of the tail. Stylet well developed with large basal knobs, whose anterior faces are slightly concave. Prorhabdion, slightly less than half stylet length. The dorsal oesophageal gland opens 6-8  $\mu$ m behind the stylet base. Median oesophageal bulb rounded, very muscular with conspicuous valves. Phasmid pore-like situated 1 annule behind the anus. Genital primordium consists of two cells located at about 55% of body length.

All the biometrical and morphological characters agree with the description of a typical *Heterodera avenae* by William and Siddiqi, 1971 although the vulval slit is shorter in our specimens.

Type B was found in four localities only (Table I), three of them in the province of Navarra and the other in that of Barcelona, all of them in the NW of Spain.

The females, which were found only in the populations from Andosilla and Mendavia were morphologically identical with specimens belonging to type A. Cysts are a little smaller, light brown in colour, with a weak bifurcate underbridge more evident in younger individuals and less prominent bullae (Fig. 1b) than in typical H. *avenae*.

On the basis of these characters, these populations fall in the range of the British Pathotype 3 of *H. avenae*.

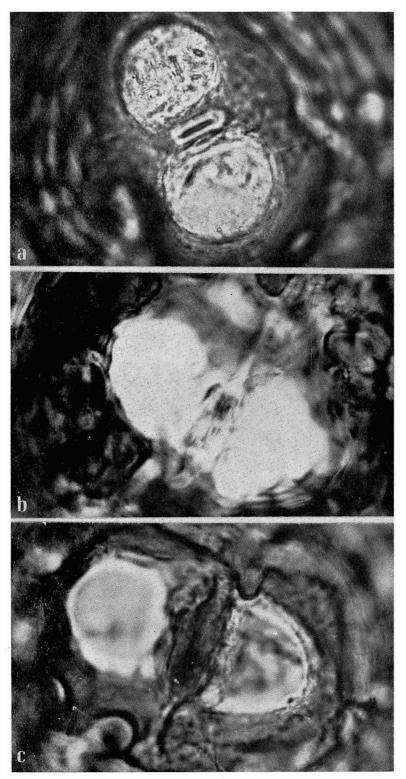


Fig. 1 - Vulval cone of populations of *Heterodera avenae* found in Spain: type A (a); type B (b); type C (c).

Table I - Comparative measurements of different populations of Heterodera avenae.

Type A							
	Pop. nº 1 Sample 540 Wheat «Triple Dwarf»	Pop. nº 2 Sample 430 Wheat «Triple Dwarf»	Pop. nº 3 Sample 630 Barley «Ayer»	Average of the three: populations	Pop. nº 1. Wheat from Andosilla (Navarra)		
Females							
n	4				5		
$L \mu m (1)$	550-760 (680*)				454-735 (627)		
Bμm	375-490 (435)				300-525 (412)		
Spear µm	28.5 (n=1)				20-22 (21.2)		
L/B			-		1.4-1.6 (1.5)		
Neck length µm					109-150 (128)		
Excretory pore (2)					116 (n=1)		
Vulva-anus distance					42-57 (48.5)		
Cysts							
n	20	20	20	60	20		
L μm (1)	648-918 (765)	567-905 (723)	638-958 (793)	567-958 (760)	552-904 (719)		
B μm	486-716 (586)	380-702 (530)	527-750 (613)	380-750 (576)	416-698 (498)		
L/B	1.1-1.6 (1.4)	1.1-1.5 (1.3)	1.1-1.5 (1.3)	1.1-1.6 (1.3)	1.2-1.7 (1.4)		
Cone top $(3)$							
n	6	6	6	18	8		
L μm	46-51 (48.5)	48-56 (52)	44.5-53 (48.7)	44.5-56 (49.7)	39-58 (47.5)		
b μm	22-24 (23.2)	22-27 (24)	20.5 - 26.5(22.6)	20 5-27 (23.2)	18-29 (24.6)		
s µm	5.10 (7)	6.5-9 (7.5)	6.5-8 (7.3)	5-10 (7.3)	5-9.5 (7.3)		
Distance between semi- fenestrae μm	6-9 (7)	7-8.5 (7.6)	5-7.5 (6)	5-9 (6.9)	6-9 (7.7)		
	(-/		0 (0)		,		
Eggs n	20	20	20	60	20		
L µm	118-136 (125)	117-133 (125)	122-141 (132)	117-141 (127)	116-136 (126)		
B μm	45-59 (51)	38-52 (44)	36-52 (48)	36-59 (48)	41-54 (49)		
L/B	2.1-2.9 (2.5)	2.2-3.3 (2.5)	2.5-3.1 (2.8)	2.1-3.3 (2.6)	2.3-3.3 (2.6)		
Second stage larvae							
n	10	20	4	34			
Lμm	583-639 (609)	552-592 (576)	554-609 (588)	552-639 (591)			
Spear µm	25-30 (27)	25.5-28 (26.4)	25-26.5(26)	25-30 (26.4)			
a	27-31 (28.5)	26-27 (26.3)	28-29.5(29)	26-31 (27.9)			
с	8-9 (8.4)	8-9 (8.3)	7.8-8.2 (8)	7.8-9 (8.2)			
tail μm	69-76 (72)	63-74 5(68.5)	71-76 (73.2)	63-76 (71.2)			
hyaline tail µm	42-56 (49)	44-50 (49)	50-52 (50.5)	42-56 (49.5)			
hyaline tail/spear length	1.5-2 (1.8)	1.7-1.9 (1.8)	1.8-2 (1.9)	1.5-2 (1.8)			
tail length/anal body width	3.8-4.5 (4.3)	3.9-4.5 (4.3)	4.3-4.6 (4.4)	3.8-4.6 (4.3)			
	,	•	•	•	• •		

Type A

(1) Excluding neck.
(2) Distance of excretory pore from the anterior end of body.
(3) Oostenbrink and Den Ouden, 1954.

(\*) Mean values in brackets.

Туре в			Type C			
Pop. nº 2 Barley from Mendavia (Navarra)	Pop. nº 3 Cereals from San Adrian (Navarra)	Pop. n 4° Wheat Caldas de Montbuy (Barc.)	Average of the four populations	Pop. nº 1 Oats Azuqueca de Henares (Guadalajara)	Pop. nº 2 Wheat «Mexipac» Lebrija (Sevilla)	Average of the two populations
5 561-798 (648) 352-503 (423) 22.5 (n=1) 1.4-1.6 (1.5) 110-135 (125) 115 (n=1)			$\begin{array}{c} 10\\ 454\text{-}798\ (637)\\ 300\text{-}525\ (418)\\ 20\text{-}22.5(21.8)\\ 1.4\text{-}1.6\ (1.5)\\ 109\text{-}150\ (126.5)\\ 115\text{-}116\\ 42\text{-}57\ (48.5) \end{array}$			
20 561-821 (691) 356-602 (476) 1.2-1.7 (1.5)	20 589-794 (687) 328-657 (504) 1.1-1.7 (1.4)		60 552-904 (699) 328-698 (492) 1.1-1.7 (1.4)	20 540-890 (690) 430-700 (580) 1.1-1.6 (1.2)	20 611-927 (807) 444-720 (620) 1.1-1.5 (1.3)	40 540-970 (748) 430-720 (600) 1.1-1.6 (1.2)
4 39-52 (44) 19-27 (24.7) 6-10 (7)	$\begin{array}{c} 3\\ 47\text{-}51 & (49)\\ 24\text{-}28.5(26)\\ 5.5\text{-}6.5 & (5.6) \end{array}$	$\begin{array}{c} 2\\ 47\text{-}49.5(48.5)\\ \textbf{23}\text{-}24.5(24)\\ 9\text{-}10.5(9.7)\end{array}$	$\begin{array}{r} 17\\ 39{\text{-}58} & (47.2)\\ 18{\text{-}29} & (24.8)\\ 5{\text{-}10.5}(6.6)\end{array}$	$\begin{array}{c} 10\\ 41\text{-}51.5(45.2)\\ 19\text{-}24 (21.4)\\ 6\text{-}8.5 (6.8)\end{array}$	13 40-55 (45.5) 19-25.5(21.1) 5-9 (6.5)	$\begin{array}{c} 23\\ 40{\cdot}55 & (45.3)\\ 19{\cdot}25{\cdot}5(21.3)\\ 5{\cdot}9 & (6.7) \end{array}$
6-9 (7.4)	6-6.6 (6.4)	5-8 (6.5)	5-9 (7)	5.5-10.5(8)	5-10 (6.1)	5-10.5(7.2)
20 118-136 (125) 41-57 (47) 2,2-3.3 (2.6)	20 118-141 (133) 41-63 (52) 2.1-3.4 (2.6)		$\begin{array}{r} 60\\ 116\text{-}141\ (128)\\ 41\text{-}63\ (49)\\ 2.1\text{-}3.4\ (2.6) \end{array}$	20 126-142 (131) 34-50 (40.5) 2.6-3.9 (3.2)	20 108-139 (119) 47-60 (52,5) 2-2.6 (2.3)	40 108-142 (125) 34-60 (46.5) 2-2.9 (2.7)
$\begin{array}{c} 7\\ 540{-}608\ (573)\\ 25.5{-}28\ (26\ 6)\\ 25{-}26.5(25.5)\\ 8{-}10\ (9.1)\\ 61{-}65\ (62.8)\\ 40{-}46\ (42.1)\\ 1.5{-}1.7\ (1.6)\\ 3{-}4\ (3.7)\\ \end{array}$	$\begin{array}{c} 10\\ 560{-}626\ (586)\\ 26{-}29\ (26.8)\\ 24{,}5{-}28\ (26.7)\\ 7{,}5{-}9\ (8.2)\\ 66{-}79\ (71.5)\\ 45{-}51{,}5(47.6)\\ 1{,}5{-}2\ (1.7)\\ 4{-}4{,}4\ (4.1)\\ \end{array}$	· ·	27 540-650 (593) 25.5-27 (26.6) 24.5-28 (26.3) 7.5-10 (8.8) 61-79 (67.1) 40-51 (43.7) 1.5-2 (1.6) 3-5 (4) Phasmid 2 - 4 annules behind anus level	$\begin{array}{c} 20\\ 553.642\ (598)\\ 24-28.5(25.8)\\ 26-30\ (28.1)\\ 8-11\ (9)\\ 53-72\ (65.5)\\ 41-49\ (46.2)\\ 1.5-2\ (1.8)\\ 3.9-4.5\ (4.3)\end{array}$	$\begin{array}{c} 20\\ 549{\textbf{-}}621\ (588)\\ 24{\textbf{-}}29\ (25.7)\\ 26.5{\textbf{-}}31\ (28.1)\\ 7.7{\textbf{-}}9.9\ (8.3)\\ 56{\textbf{-}}71\ (68.4)\\ 39{\textbf{-}}51\ (46)\\ 1.5{\textbf{-}}2\ (1.7)\\ 3.9{\textbf{-}}4.6\ (4.2)\\ \end{array}$	$\begin{array}{c} 40\\ 549{-}642\ (593)\\ 24{-}29\ \ (25.7)\\ 26{-}31\ \ (28.1)\\ 8{-}11\ \ (8.6)\\ 53{-}72\ \ (66.9)\\ 39{-}51{-}5(46.1)\\ 1{-}5{-}2\ \ (1.7)\end{array}$

Type B

Type C

Type C was found very widespread throughout Spain. Two populations were selected as representative (Table I). Differences appear in the vulval cone, where the underbridge is sometimes rather thick in the center but very weak and bifurcated on the extremities (Fig. 1c); it is, therefore, very easily lost in slide preparation. Bullae are also less prominent than in type A.

The morphometric characters of the second stage larvae are similar to those of the typical *H. avenae*, except that the anterior faces of the spear knobs are deeply concave. However, this is not a constant feature and varies among specimens within the same population.

From these features type C can be identified as *H. mani* Mathews. This differs from *H. avenae* as follow (Mathews, 1971):

« In *Heterodera avenae* the larvae have stylet knobs whose anterior faces are deeply concave and the lateral fields has four lines; cysts are more spherical with a distinct bifurcating underbridge and well defined fenestration. The host range of *H. mani* does not include wheat, oats or barley ».

Meagher (1974) does not consider the morphological differences sufficient to establish two distinct species. On the basis of the observations on the Spanish populations, and taking into account that type C was found on the roots of wheat, it is considered that *H. avenae* and *H. mani* are the same species. All the populations found in Spain are *H. avenae* and minor differences in morphology may perhaps be attributed to the effect of environment.

#### SUMMARY

Three types of *Heterodera avenae* Woll., distinguished by morphological characters, have been found in Spain. The possibility that *H. mani* Mathews and *H. avenae* might be conspecific is discussed.

#### RIASSUNTO

### Morfologia di Heterodera avenae in Spagna

Tre tipi di *Heterodera avenae* Woll., distinti da caratteri morfologici sono stati trovati in Spagna. È discussa la possibilità che *H. mani* Mathews sia sinonimo di *H. avenae*. \_\_\_\_\_

Mathews H. J. P., 1971 - Two new species of cyst nematodes, *Heterodera mani* n. sp. and *H. iri* n. sp. from Northern Ireland. *Nematologica*, 17: 553-566.

MEAGHER J. W., 1974 - The morphology of the cereal cyst nematode (Heterodera avenae) in Australia. Nematologica, 20: 1-8.

- Oostenbrink M. and Den Ouden H., 1954 De structur van de kegeltop als taxonomisch kenmerk bij Heterodera-soorten met citroenvormige cysten. *Tijdschr. Pl. Ziekt.*, 60: 146-151.
- WILLIAMS T. T. and SIDDIQI M. R., 1972 Heterodera avenae C.I.H. Descriptions of Plant Parasitic Nematodes Set 1 n. 2.

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