Notes brèves

DESCRIPTION OF THE MALE AND OBSERVATIONS ON THE FEMALE OF *MERLINIUS RUGOSUS* (SIDDIQI, 1963) SIDDIQI, 1970 (NEMATODA : TYLENCHIDA) FROM JORDAN

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Merlinius rugosus (Siddiqi, 1963) Siddiqi, 1970 is known from India (Siddiqi, 1963), Iraq (Loof, 1978) and Israel (Cohn *et al.*, 1973). It was also recovered by the author in Jordan, from soil around banana (*Musa* L. (AAA Group) « Dwarf Cavendish ») and wheat (*Triticum* sp.) in the southern Jordan Valley and around grapevine (*Vitis vinifera* L.) in Salt. Males of this species were found in the Jordan Valley populations, and are described here for the first time together with a comparison of the females from Jordan with described material. Specimens used in this study were killed by heat, fixed in TAF, stained in acid-fuchsin lactophenol and processed to glycerol by Baker's (1953) rapid method.

Description of the male (Wheat population)

DIMENSIONS

Males (n = 8) : L = 0.64 - 0.84 (0.73) mm ; a = 31 - 40 (34.9) ; b = 5.3 - 6.5 (5.9) ; c = 11.5 - 17.0 (13.5) ; c' = 2.6 - 3.5 (3.1) ; T = 39 - 53 (45) ; spicules = 18 - 22 (20.5) µm ; gubernaculum = 7.0 - 8.5 (7.3) µm ; stylet = 17.5 - 19.0 (18.5) µm.

DESCRIPTION

Body ventrally arcuate when killed by heat. Cephalic region hemispherical, bearing six or seven annules, and offset from the body by a constriction (Fig. 1, A). « En face » view shows six lobes of equal size, each lobe bearing a lip (Fig. 1, C). Cephalic framework moderately sclerotized. Stylet slender, with rounded knobs directed posteriorly. Excretory pore opposite to the anterior part of the basal oesophageal bulb, at 99 - 119 µm from the anterior end. Hemizonid situated one or two body annules anterior to excretory pore, extending on two annules. Orifice of dorsal oesophageal gland located 2-3 µm behind stylet knobs Median oesophageal bulb oval, large (12.5 - 16.5 × 7 - 10 µm) and with well-developed, refractive valvular apparatus. Basal oesophageal bulb elongate-pyriform. Œsophago-intestinal valve (cardia) prominent and rounded. Nerve ring surrounding middle of isthmus.

Body annules coarse, 1.5 - 2.5 µm in width. Lateral field about 1/3 of mid-body diameter, with six incisures whose number declines anteriorly ; the lateral field is completely areolated in the oesophageal region and irregularly areolated posteriorly. Outer incisures of the field crenate in the tail region. Deirids not observed. Thirty longitudinal citicular striae occurred in mid-body region (Fig. 1, E).

Testis single and outstretched, with spermatocytes arranged in two rows. Vacuolated globules (= ? spermatozoa), measuring 1.5 - 2.0 µm in diameter, were observed in the vas deferens.

Spicules stout, ventrally arcuate and with notched tips. Gubernaculum simple, crescent-shaped in lateral view and non-protrusible. Hypoptygmae present on the posterior cloacal lip. Caudal alae well-developed with crenate edges, extending from about opposite the proximal ends of the spicules to the tail tip. Phasmids open on caudal alae, at 40-47 % of tail length.

Observations on the female

Females of *M. rugosus* from Jordan had a more strongly sclerotized and a more prominently offset cephalic region than was originally illustrated for this species by Siddiqi (1963) (Fig. 1, B). Furthermore, a comparison of measurements between these females and those from India and Iraq shows the Jordanian specimens to have a slightly shorter body and stylet, and the phasmids to occupy a more posterior position (Tab. 1). In addition, females of this population often have slightly narrower and more pointed tail tips than those from India (Siddiqi, 1963) and Iraq (Loof, 1978) (Fig. 1, H).

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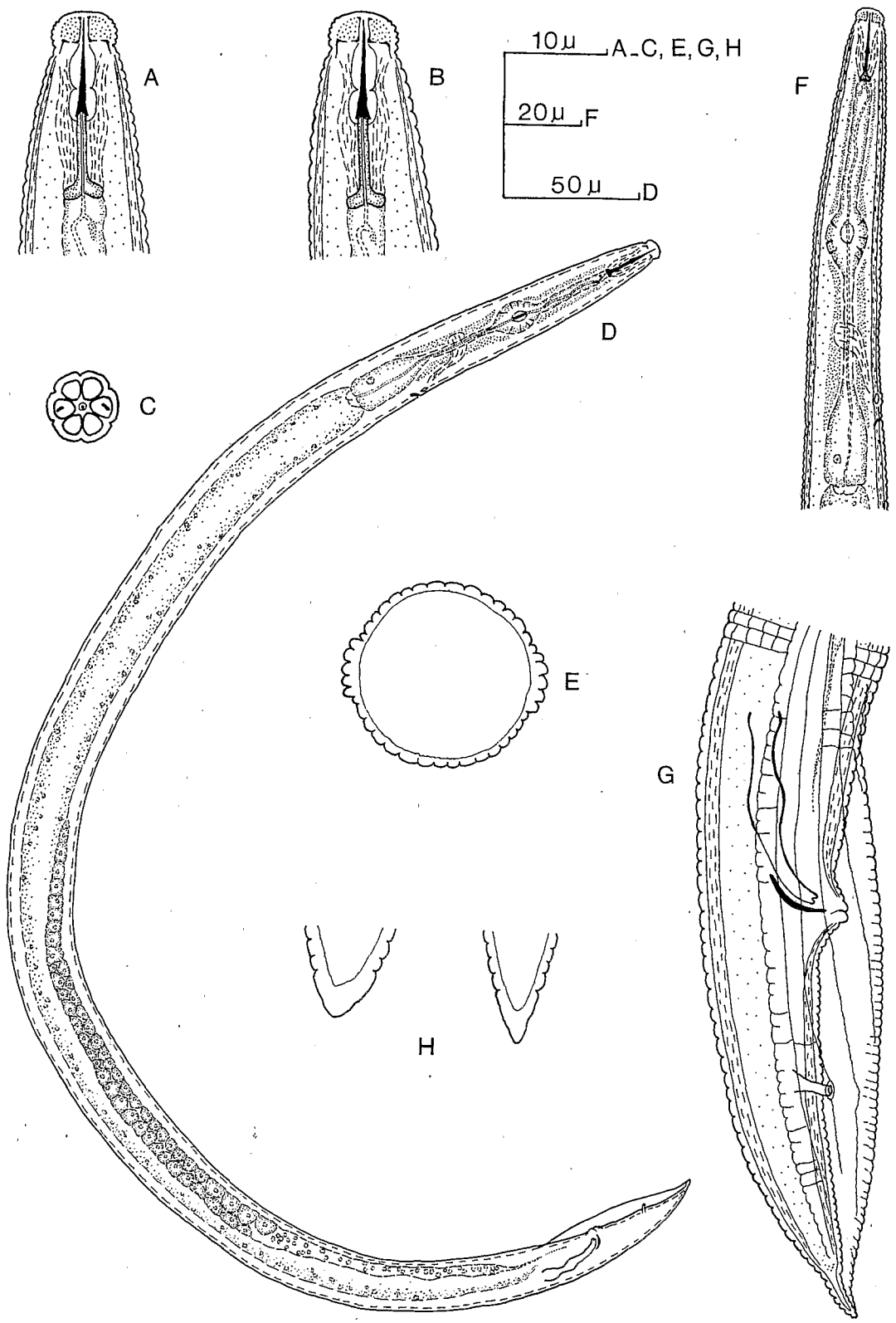

Fig. 1. *Merlinius rugosus* (Siddiqi, 1963) Siddiqi, 1970. Male. A : anterior region; C : *en face* view; D : whole specimen; E : transverse section at mid-body; F : oesophageal region; G : caudal region. Female. B : anterior region; H : variations in tail terminus shape.

Table 1

Measurements of females of *Merlinius rugosus* (Siddiqi, 1963) Siddiqi, 1970 from different locations

	Jordan		*India*	*Iraq*
	Jordan Valley (orig.)	*Salt (orig.)*	*(Siddiqi, 1963)*	*(Loof, 1978)*
n	8	1	4	20
L (mm)	0.63-0.83 (0.75)	0.83	0.8-0.9	0.73-0.94
a	26-32 (29)	34	30-32	—
b	4.6-6.1 (5.6)	5.3	4.4-5.1	—
c	15-19 (16.5)	16	15.5-17	—
c'	2.2-3.0 (2.5)	2.7	3.0	2.6-3.1
V	53-57 (55)	55	55-56	52-58
Stylet (µm)	18.-019.5 (18.6)	19.5	23	20-23
phasmids (% tail length)	31-45 (38.7)	42 (?)	33	20-23
Tail annules	20-27 (23.7)	26	24	19-28
Long striae	28-36	?	32-36	—

Discussion

Kheiri (1972) synonymized *M. rugosus* with *M. quadrifer* (Andrássy, 1954) Siddiqi, 1970, but this was rejected by Loof (1978) who detailed the differences between the two species. The apparent absence of males in the former species was a main character used by Loof (1978) to differentiate it from the latter species, which is bisexual, but, in the author's opinion, this character can no longer be used with certainty. However, it may be relevant that, despite the relatively high frequency of males in the Jordan Valley populations of *M. rugosus*, all the females examined appeared to have empty spermathecae.

M. rugosus is most easily distinguished from *M. quadrifer* by the shape of the female tail, which is conoid with a narrowly-rounded tip in the former species and subcylindrical with a broadly-rounded tip in the latter (Loof, 1978).

The differences noted between the Jordanian and previously described populations of *M. rugosus*, such as the more prominently offset cephalic region and the differently-shaped tail tip of the Jordanian specimens, are not considered of significant magnitude for species differentiation. Similar variations have been noted in other, closely-related species. For instance, *Tylenchorhynchus clarus* Allen, 1955 has a cephalic region varying from continuous to slightly offset (Elmiligy, 1969), whilst *M. nanus* (Allen, 1955) Siddiqi, 1970 exhibits some variation in tail tip shape (Saltukoglu, Geraert & Coomans, 1976). Similarly, morphometric differences between nematode populations, such as occurred between this and other populations of *M. rugosus*, are not always of major taxonomic significance since these features can be influenced by environmental factors. It is concluded, therefore, that the Jordanian populations are representative of *M. rugosus*, and that the variations of characters observed are only of intraspecific level.

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Accepté pour publication le 19 juin 1979.

MORE ABOUT THE DROP BY DROP DISTRIBUTION OF A NEMATODE SUSPENSION

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Samples containing exactly equivalent numbers of small nematodes such as infective juveniles of zoo- and phyto-parasitic species, are often prepared by distributing a homogeneous suspension of nematodes into samples of equivalent volumes. When the required volumes of suspension are pipetted at one time, the error variance observed on their nematode content should be as a rule, more or less closely equivalent to the mean count, displaying a Poisson distribution (Peters, 1941). In practice however, the observed variance may be higher (Moriarty, 1963) and in some recent papers authors have proposed a modified procedure to improve reproducibility. Wilson (1976) made a complete sample with two partial pipettings, arguing, «... thus any tendency for the pipetting procedure to select water rather than larvae was randomized. » Castro and Fairbairn (1969) made ten partial pipetings and obtained 1.4 to 2.6 % coefficient of variation. Reversat (1976) using a drop by drop distribution made 300 partial samplings to achieve 3 % accuracy.

New facts are presented about the reliability of the last procedure. In the apparatus used (Fig. 1), the suspension of nematodes, maintained homogeneous by gentle air bubbling or by stirring, flows out by gravity through a narrow plastic tubing and is delivered drop by drop (100 drops = 3.25 ml) at its end.

In a first experiment, five 250 ml suspensions of freshly hatched juveniles of *Heterodera oryzae*, ranging from 2,000 to 10,000 individuals per ml, mixed by air bubbling were treated successively. After 50 ml were withdrawn, 24 consecutive drops were collected separately and their nematode content was determined (Fig. 2). The relationship between variance and the mean agreed with the Poisson law, with diluted suspensions, but diverged with increased nematode density. Lellouch (1964), discussing a similar relationship between variance and the mean obtained for red cells in blood counts, attributed this effect to the volume of particles, which is not negligible compared with the sample volume. Adapting his argument to the present case, we can consider the maximum number of nematodes in a drop (n), equal to the ratio between the volume of the drop (32.5 µl) and the volume occupied by one nematode (a).

If volume a has a probability p to contain a nematode, the average number of nematodes in a drop is $\mu = np$ and the variance equals $\sigma^2 = npq$ with $p + q = 1$, wich leads to the formula $\sigma^2 = \mu(1 - \frac{\mu}{n})$. By using the estimations m and s^2 of μ and σ^2, n can be calculated from the formula $n = m^2/(m - s^2)$. The mean numerical value for n, calculated with this formula from the four highest values (given on Fig. 2) of m is 491. From this, a curve giving the relationship between m and s^2 can be traced according ot a second formula, wich is the reciprocal of the first : $s^2 = m\text{-}m^2/n$, which fits the experimental results (dotted line in Fig. 2). Further, if we consider a drop (32.5 µl), filled with 491 spherical particles arranged in the densest state, the void ratio, i.e. ratio of volume of voids to volume of particles, equals 0.35 (Means & Parcher, 1964). From this, the volume of the particle can be calculated by the relation : (32.5 µl - 49a)/491a = 0.35. The result is a = 0.049 µl, wich corresponds to a spherical particle with a diameter of 0.45 mm, while the length of the *Heterodera oryzae* juvenile has a

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