

On *Achlysiella*, a new genus of obese Pratylenchidae (Nematoda : Tylenchoidea)

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SUMMARY

Achlysiella n. gen. is proposed with *A. williamsi* (Siddiqi, 1964) n. comb. (syn. *Radopholus williamsi*) as type species. The new genus is close to *Radopholus* s.s., particularly in the form of the soil-living vermiform stages, but is distinct in a number of morphological and, particularly, biological, characters. The vermiform female, although usually inseminated, is sexually immature with few cells in the ovaries and has an unusually long oesophageal gland lobe ($b' \times = 3$). On entering a sugar cane root the female swells and becomes sausage-shaped. A gelatinous egg-sac is formed which contains the exceptionally large eggs and later on, juveniles, adult males and vermiform females. The juveniles and adult males do not feed. The obese female is endo- or semi-endoparasitic and usually lies in a cavity in the root cortex. The neck region remains mobile and there is neither a permanent feeding site nor specialized trophic cells. Necrotic areas form where the nematode has fed. The geographic origin and subsequent dispersal of *A. williamsi* are speculated upon and the relationships with other genera containing obese sedentary females are discussed. Several other species of *Radopholus* are thought, on the basis of long oesophageal glands and few ovary cells in the vermiform female, to belong to *Achlysiella*, but until details of their biology are elucidated, no formal proposal of new combinations is considered advisable.

RÉSUMÉ

Achlysiella, un nouveau genre de Pratylenchidae renflés (Nematoda : Tylenchoidea)

Achlysiella n. gen. est proposé, avec *A. williamsi* (Siddiqi, 1964) n. comb. (= *Radopholus williamsi*) comme espèce type. Ce nouveau genre est proche de *Radopholus* s. s., notamment par l'aspect des stades vermiformes, libres dans le sol; il en diffère toutefois par un certain nombre de particularités morphologiques, et surtout biologiques. La femelle vermiforme, bien que généralement fécondée, est sexuellement immature, les ovaires ne présentant que quelques cellules, et son lobe œsophagien est exceptionnellement long ($b' = 3$ en moyenne). Après avoir pénétré dans les racines de canne à sucre la femelle gonfle et devient obèse. Une masse gélatineuse se forme qui contient des œufs de grande taille et, plus tard, des juvéniles, des mâles et des femelles vermiformes. Les juvéniles et les mâles ne se nourrissent pas. La femelle renflée est endo- ou semi-endoparasite et se localise dans une cavité du cortex racinaire. La région antérieure demeure mobile et il n'existe pas de site nutritionnel permanent ni de cellules trophiques spécialisées. Des zones nécrosées signalent l'endroit où le nématode s'est nourri. Des suppositions sont émises concernant l'origine géographique et la dispersion ultérieure de *A. williamsi*; ses relations avec d'autres genres possédant des femelles renflées sont discutées. Prenant en compte la grande longueur des glandes œsophagiennes et le petit nombre de cellules ovariennes chez la femelle vermiforme, il est supposé que d'autres espèces de *Radopholus* pourraient appartenir à *Achlysiella*, mais un transfert formel n'a pas été jugé souhaitable.

In 1960 Williams reported a population of *Radopholus similis* (Cobb, 1893) attacking sugar cane in Mauritius. On the basis of the material described by Williams, Siddiqi (1964) decided that this nematode represented a new species which he described as *Radopholus williamsi*. In 1986 one of us (JB) collected material of diseased sugar cane from Papua New Guinea. Examination of the roots revealed a *Rotylenchulus*-like female with an egg-sac whilst the soil contained adult males and immature females corresponding to *Radopholus williamsi*. Closer examination of the swollen females revealed that they belonged not to *Rotylenchulus* but were close to *Radopholus*. Studies on the bionomics linked the free-living stages with the swollen females parasitic in the roots.

Because of a number of morphological and, particularly, biological differences, a new genus is hereby proposed.

*Achlysiella** n. gen.

DIAGNOSIS

Pratylenchidae. Vermiform nematodes except for the root-parasitic female which becomes swollen. Sexual

* From *Achlys* — goddess of obscurity and *ella* — small.

dimorphism in anterior region marked, males having a higher and more rounded, knob-like head and with the stylet and oesophagus much reduced and non-functional. Female head low, flattened; stylet moderately cuticularized; oesophagus well-developed with a very long dorsal oesophageal lobe ($b' \times = 3$ in vermiform female) with the gland cells discrete. Vulva post-median (60-63 %); genital tracts didelphic outstretched. In the vermiform female the ovaries are immature with only a few cells, although the spermathecae usually contain sperm. Tail conoid with a rounded terminus.

Mature female endo- or semi-endoparasitic in roots; swollen; sausage-shaped with the neck-region attenuated; vulval lips protuberant. Gelatinous egg-sac present containing eggs, juveniles, adult males and vermiform females. Eggs large; juveniles moulting to adult stages without feeding.

Male tail conical with distinct hyaline terminal portion, spicules paired, arcuate. Gubernaculum protrusible, titillae not observed. Bursa reaching almost to tail tip.

TYPE SPECIES

Achlysiella williamsi (Siddiqi, 1964) n. comb.
= *Radopholus williamsi* Siddiqi, 1964
= *Radopholus similis apud* Williams, 1960 nec
Cobb, 1893.

REMARKS

On the basis of the elongated oesophageal gland lobe and the immature ovaries in vermiform females, it is likely that the following species also belong in this genus: *Radopholus capitatus* Colbran, 1971, *R. brevicaudatus* Colbran, 1971, *R. magniglans* Sher, 1968, *R. trilineatus* Sher, 1968 and *R. vacuus* Colbran, 1971.

In fact, Sher (1968) commenting on *R. magniglans* and *R. trilineatus* wrote "the rather short ovaries and absence of well-developed oocytes or eggs might be further evidence that these species undergo further development in the roots of plants".

However, as the biology appears to be the major reliable differentiating character at present, further work is necessary to confirm the position of the above species and hence the new combinations are not formally proposed here.

RELATIONSHIPS

Achlysiella n. gen. is close to *Radopholus* but differs in that: the vermiform female has immature ovaries and a very long gland lobe; the mature female is swollen and sedentary (although with no fixed feeding site) with protuberant vulval lips; a gelatinous egg-sac is formed containing the exceptionally large eggs together with juveniles, adult males and vermiform females; the life-cycle is compressed with the non-feeding juveniles having superimposed moults.

Achlysiella williamsi (Siddiqi, 1964) n. comb.
(Figs 1, 2 A)

MEASUREMENTS

Immature vermiform female (n = 10) : L = 0.41 ± 0.03 (0.35-0.45) mm; a = 25.8 (22.4-30.2); MB = 7.4 (6.5-8.0); b' = 2.9 (2.6-3.4); c = 13.8 (11.6-15.5); c' = 3.0 (2.3-3.4); V = 62.2 (60.3-63.8); body diameter = 16 ± 2.2 (13-20) μm ; tail = 30 ± 10 (21-35) μm ; anal body diameter = 10 ± 0.7 (9-11) μm ; stylet = 14.6 ± 0.7 (13.1-15.4) μm ; h* = 6.1 ± 0.7 (5-7) μm .

Mature swollen female ex root (n = 10) : L = 0.79 ± 0.01 (0.56-0.89) mm; a = 8.8 (8.1-11.2); V = 59 (51-61); maximum diameter = 90 ± 14.8 (64-107) μm .

Male (n = 10) : L = 0.39 ± 0.03 (0.35-0.44) mm; a = 27.6 (25.2-30.2); c = 12.5 (11.2-13.6); c' = 2.9 (2.7-3.3); body diameter = 14 ± 1.6 (12-17) μm ; tail = 31 ± 2.4 (26-34) μm ; anal body diameter = 11 ± 0.7 (10-12) μm ; stylet = 10 ± 0.7 (9-11.4) μm ; spicules = 14.9 ± 0.6 (13.7-15.4) μm ; gubernaculum = 9.2 ± 0.6 (8-10) μm ; h* = 9.5 ± 0.8 (8-10.7) μm .

DESCRIPTION

Immature vermiform females : Heat relaxed form slightly arcuate ventrally. Lip region hemispherical, slightly flattened apically and with three to four annules. En face dumb-bell shaped in the dorso-ventral axis with the lateral lips extending to the base of the head. Head slightly offset by a groove. Cuticle annulated, ventral annules at mid-body approximately 1.2 μm apart. Lateral field marked by four lines, the two outer being crenate. Areolation absent except that some specimens have the outer bands areolated anterior to the median bulb. Stylet of moderate strength with rounded basal knobs which can be flattened or slightly concave anteriorly. Dorsal oesophageal gland opening 3-4 μm posterior to knobs. Oesophagus comprising a narrow pro-corpus, ellipsoid median bulb and a long lobe with the individual gland cells well developed and separate. Nerve ring just behind bulb. Hemizonid about 4-5 annules posterior to the nerve ring and immediately anterior to the excretory pore. Hemizonion eight or nine annules further posterior. Vulva post-median and with slightly protuberant lips. Genital system amphi-didelphic, outstretched. Spermathecae well developed with rod-shaped sperm. Ovaries immature with only three to six developing oocytes. Tail conoid with a dorsally convex, smoothly rounded terminus. There are about 18 ventral and 21 dorsal annules. Phasmids about seven (two to nine) annules posterior to anus.

* Length of terminal, non-protoplasmic portion of tail.

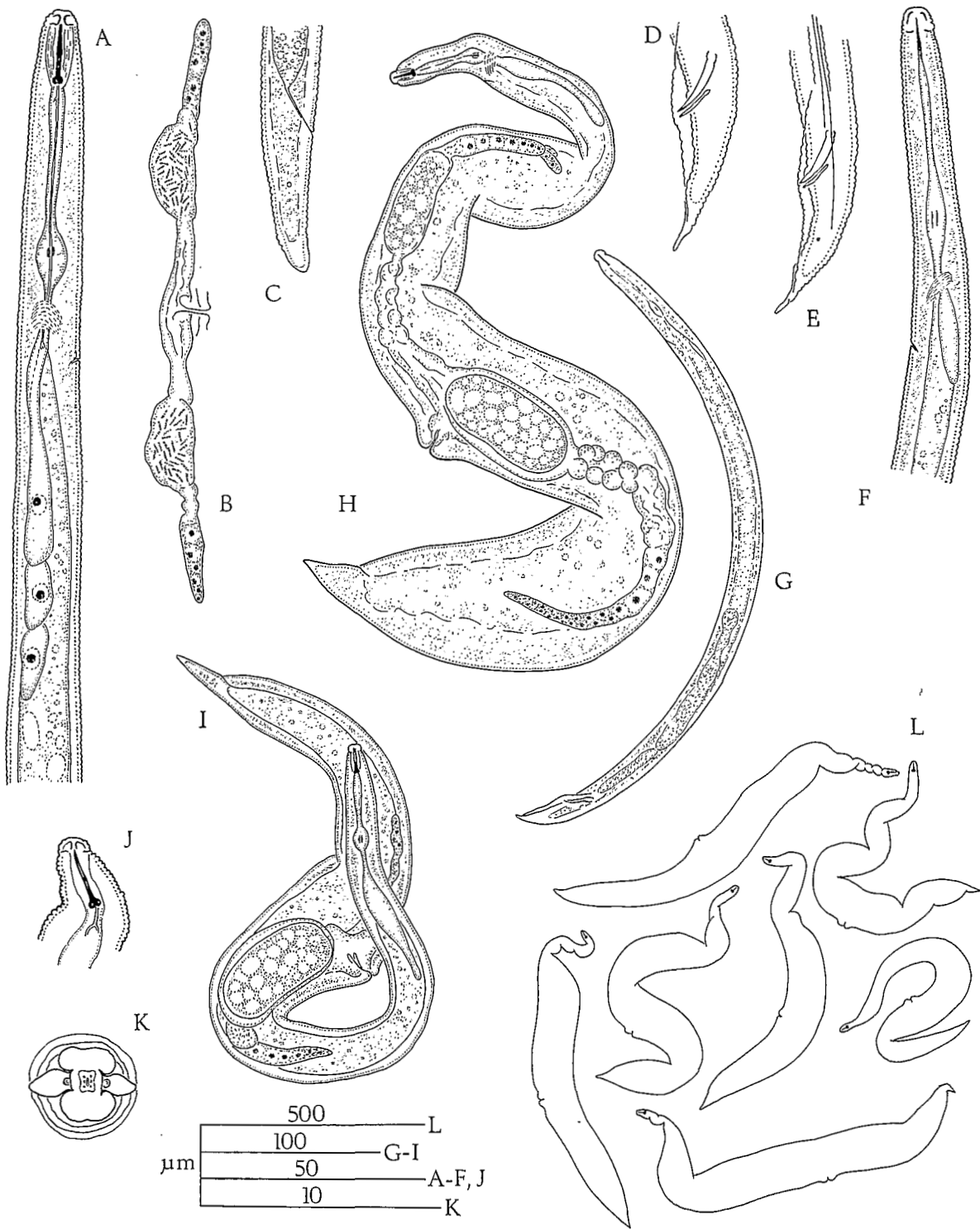


Fig. 1. *Achlysiella williamsi* (Siddiqi, 1964), n. comb. - Vermiform immature female - A : Oesophagus; B : Genital tracts; C : Tail. Male - D, E : Tails; F : Oesophagus; G : Entire male. Mature swollen female - H, I, L : Entire females; J : Head region; K : Head, en face view.

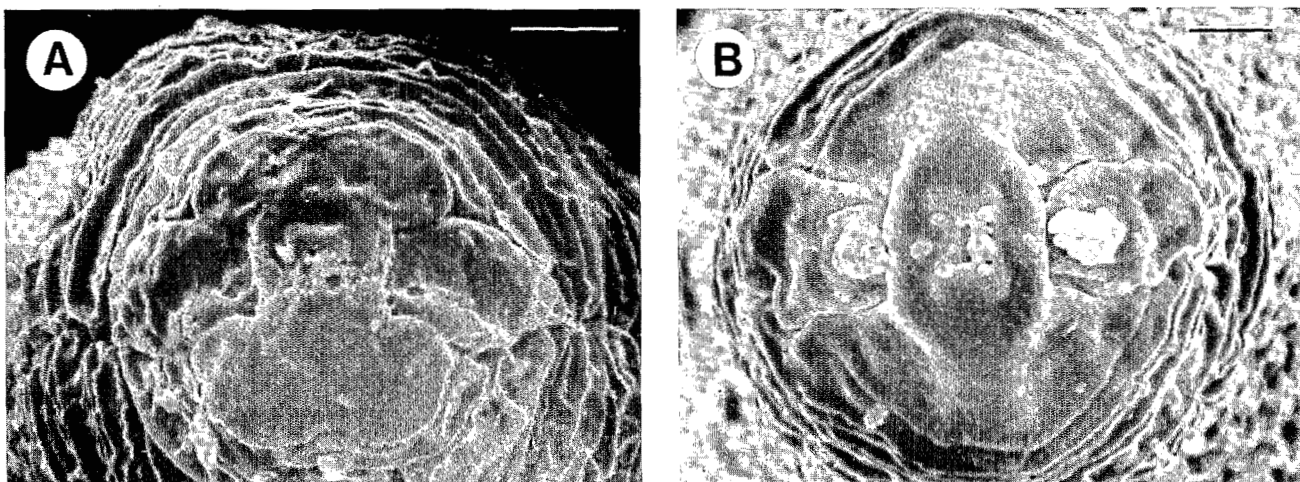


Fig. 2. *Achlysiella williamsi* (Siddiqi, 1964), n. comb. A : Female head (SEM en face view). *Radopholus similis*. B : Female head (SEM en face view). (scale bar = 1 μ m).

Mature swollen female : Endo- or semi-endoparasitic in the root. Body swollen, sausage-shaped, with the neck region narrower and mobile. Vulval lips strongly protuberant, genital system fully developed with numerous oocytes and usually one egg in the uterus. Eggs large (92 \times 58 μ m). Posterior tract may reach almost to the anus and be reflexed one or more times. Anterior tract reaching up to the oesophageal gland lobe in some specimens.

Male : Heat-relaxed form open C-shaped. Head offset, as high as broad and with five or six annules. Stylet and oesophagus degenerate. Bursa crenate, reaching almost to tail tip. Spicules paired, arcuate; gubernaculum simple, protrusible. Tail with a more or less digitiform terminus.

Juveniles : Non-feeding but with fully developed stylet. Often enclosed in one or two cast cuticles.

Note : The vermiform female and male of *Achlysiella williamsi* n. comb. were compared with the type material of *Radopholus williamsi*. No differences were noted apart from the following : (1) in the *R. williamsi* type material the phasmid in the vermiform female is more anterior [4-5 annules posterior to the anus as opposed to 7 (2-9)]; (2) the male spicules in *R. williamsi* are slightly longer (18 : 15 μ m). These differences are not considered significant.

DIAGNOSIS

Having the characters of the genus.

TYPE HOST AND LOCALITY

Sugar cane, L'Étoile, Mauritius.

TYPE SPECIMENS

Holotype female and allotype male at Rothamsted Experimental Station, Harpenden, Herts., U.K.

VOUCHER SPECIMENS

Ex rhizosphere and roots of *Saccharum officinarum*, *S. robustum* and *S. spontaneum*, Ramu Valley, Papua New Guinea and held at CABI Institute of Parasitology, St. Albans, Herts., U.K. Additional specimens (three males, two vermiform females and three mature swollen females) at Laboratoire des Vers, Muséum national d'Histoire naturelle, Paris, France.

BIONOMICS

The partly or fully swollen mature female is found within a cavity in the root cortex, although in smaller roots the posterior section of the nematode body can be exposed (Fig. 3). No swelling or galling of the root is produced but necrotic lesions, purplish in colour and similar to those produced by *Pratylenchus*, are evident on the root surface around established females. The neck region is mobile and the body can be twisted around within the cavity. Histological studies show that there is no fixed feeding site, the cavity being surrounded by areas of necrotic cells which presumably mark previous

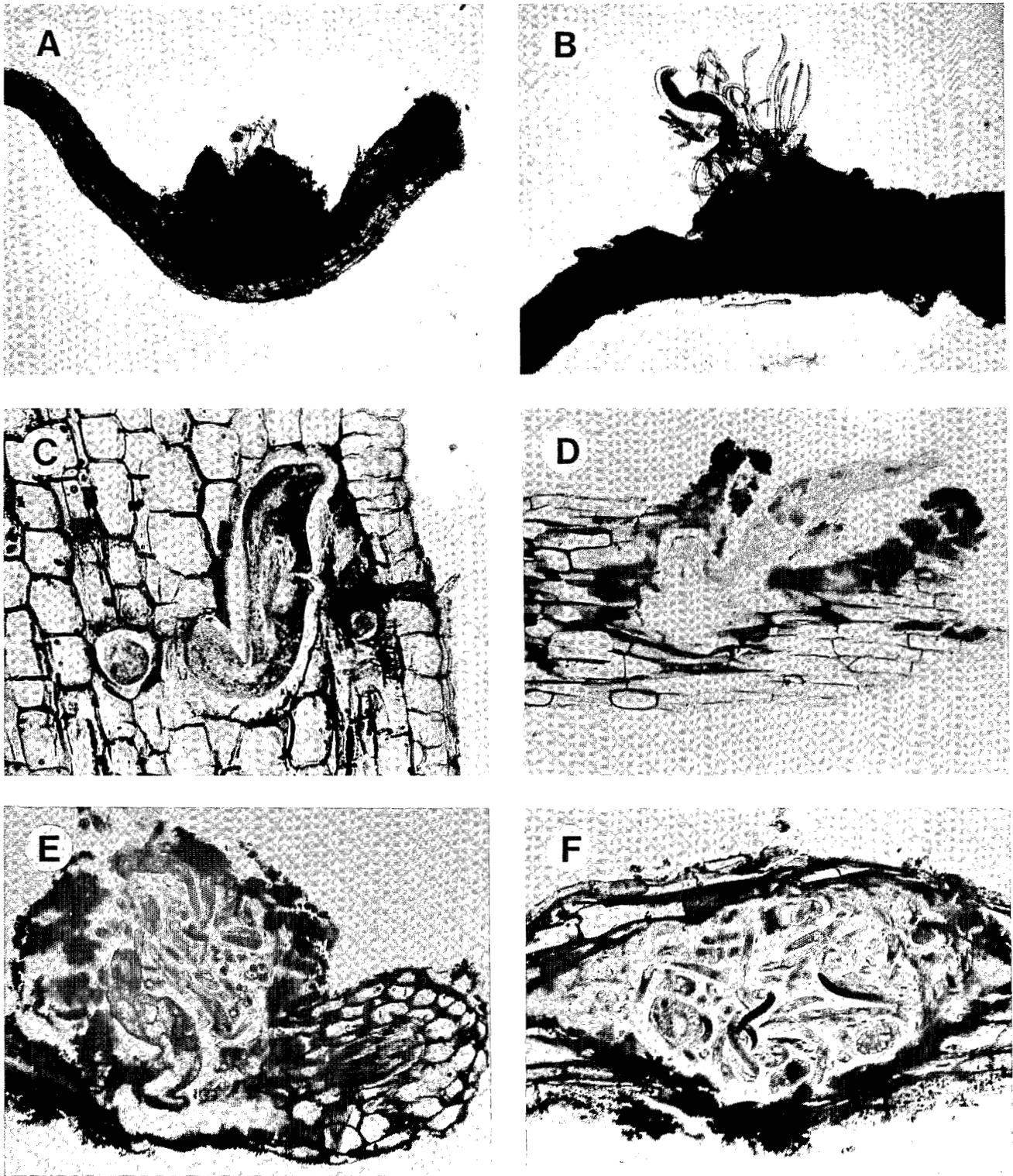


Fig. 3. *Achlysiella williamsi* (Siddiqi, 1964), n. comb. A, B : Ruptured egg sacs attached to sugar cane root; C, D : Swollen females in root cavity. E : Swollen female with egg-sac formed mostly outside the root and containing juveniles and eggs; F : Mass of juveniles and eggs within an egg-sac still contained within the root.

feeding areas. A gelatinous egg-sac [530 (500-600) × 410 (333-480) µm; n = 7] is produced around the vulval region. When exposed outside the root the periphery of the sac toughens.

Contained within the egg-sac are eggs, juveniles of all stages, adult males and immature adult females which may already be inseminated. The number of eggs and nematodes within each sac (n = 10) varies from 15 to 91 (x̄ = 38). Eggs develop to vermiform adults in distilled water in 14-16 days at 25°. Moults are often superimposed so that a J4 enclosed in both the J3 and J2 cuticle is not uncommon. Juveniles, males and immature females (usually inseminated) can also be found in the soil but the females do not swell until root penetration and feeding has occurred. Although the swollen females are almost invariably solitary, groups of up to five have been seen in the cavity. However, whether this is a result of a multiple invasion or re-infection from an existing egg-sac is not clear.

DISCUSSION

Papua New Guinea is the major centre of diversity of *Saccharum* spp. and is thought to be the origin of the thick-stemmed "noble" canes (= *S. officinarum*) of commerce (Purseglove, 1972). In the Ramu Valley *Achlysiella williamsi* was found quite commonly on *S. officinarum* and the wild species *S. robustum* and *S. spontaneum*. As the centre of origin of the closely related genus *Radopholus* is thought to be Australasia, and as the major centre of diversity of *Saccharum* is Papua New Guinea, it seems probable that *A. williamsi* evolved from *Radopholus* stock either within Papua New Guinea itself or nearby.

At present *A. williamsi* (syn. *R. williamsi*) is recorded from Mauritius, the type locality (Williams, 1960), over a wide area of the Pacific region (Kirby & Kirby, 1977; Orton Williams, 1980) and Queensland, Australia (Khair, 1986). The distribution is probably wider as a cursory examination under the stereomicroscope of sugar cane roots infested with *A. williamsi* females would almost certainly result in them being misidentified as *Rotylenchulus*.

If *A. williamsi* did evolve in the Papua New Guinea region it was probably spread via rooted sugar cane cuttings carried by man. Noble canes have long been grown for chewing purposes in the Pacific region and have been transported by the Pacific people during their long migrations by ocean-going canoe (Purseglove, 1972). For nematodes to be transported, rooted cuttings would have to be carried and such is likely to have been the case. During the period of European influence in the region, sugar cane was certainly transported as rooted cuttings in order to better survive the long sea journeys (Purseglove, 1972) and this probably accounts for the introduction of *A. williamsi* to Mauritius and possibly other, yet to be confirmed, localities far removed from the Australasian area.

The discovery of a nematode with vermiform stages more or less corresponding to *Radopholus*, yet a mature swollen female with a gelatinous egg-sac and compressed life cycle more akin to *Rotylenchulus*, at first sight raises doubts as to the correct familial placement of *Achlysiella* n. gen. *Rotylenchulus*, *Senegalonema* and *Acontylus* are placed under *Rotylenchulidae* by Siddiqi (1986) but *Hoplolaimidae* by Fortuner (1987) whilst *Radopholus* is under *Pratylenchidae* in both recent systems (Luc, 1987; Siddiqi, 1986). Of the *rotylenchulid* genera, *Achlysiella* shares more characters with *Acontylus* (obese sedentary female, protection of eggs in an egg mass, degeneration of stylet and oesophagus in male). In addition *Acontylus* is only known from Australia (Meagher, 1968) and *Achlysiella* almost certainly originated in the same region. However, these morphological similarities are not of phylogenetic significance but are superficial. Fortuner (1987) makes the valid point that "enlargement of the female body is not a reliable character for the ... differentiation of families" whilst Luc *et al.* (1987) illustrate the effect of convergent evolution in the development of obese sedentary females in diverse families. *Achlysiella* n. gen. shares more fundamental morphological characters — the anterior placement of the dorsal oesophageal gland opening, shape of female head, en face view, relatively weak development of stylet and labial framework, long conoid tail — with the *pratylenchids* than with the *rotylenchulids* and clearly belongs to the *Pratylenchidae*. *Achlysiella* should be regarded as an intermediate stage between the tendency for endo-parasitic females of *Pratylenchus* and *Radopholus* extracted from roots to be more swollen than those from soil (a feature not found in *Hoplolaimidae*, incidentally) and the more advanced genera where obese females develop a sophisticated trophic nurse-cell system.

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