

***Stephanofilaria thelazioides* n. sp. (Nematoda: Filariidae) from a hippopotamus and its affinities with the species parasitic in the African black rhinoceros**

J. Boomker¹, O. Bain², A. Chabaud² and N.P.J. Kriek³

¹Department of Pathology, Faculty of Veterinary Science, Medical University of Southern Africa, Medunsa 0204, Republic of South Africa

²Laboratoire de Biologie Parasitaire, Protistologie, Helminthologie, CNRS-URA 114, Muséum National d'Histoire Naturelle, 61 rue Buffon, 75231 Paris Cedex 05, France

³Department of Pathology, Faculty of Veterinary Science, University of Pretoria, Onderstepoort 0110, Republic of South Africa

Accepted for publication 3rd October, 1994

Abstract

Stephanofilaria thelazioides n. sp. (Filarioidea: Filariidae: Stephanofilariinae) is described from a hippopotamus *Hippopotamus amphibius*. This nematode is close to *S. dinniki* Round, 1964, a parasite of the black rhinoceros *Diceros bicornis* in Africa, but differs from it in the number of cuticular spines surrounding the mouth, the arrangement of the cloacal papillae and the measurements of the spicules, gubernaculum and microfilariae. Species of the genus *Stephanofilaria* possess spines on the head which have been derived by modification of the sensory papillae. *S. thelazioides* is the most primitive species of the genus and has the least modified arrangement of these papillae, with six bifid internal labial spines, four bifid external labial spines and four cephalic papillae. The genus appears to have diversified in various mammals which have in common a thick skin, such as rhinoceroses, elephants, buffaloes and now the hippopotamus. It appears to have become adapted secondarily to domestic bovines, initially in Asia and subsequently in North America.

Introduction

During a recent drought in the Kruger National Park, it became necessary to cull a number of hippopotami in several rivers traversing the park. One of these animals had an ulcerated skin lesion of about 5 cm in diameter on the shoulder region. The lesion was excised, preserved in 10% buffered formalin and submitted to one of us (NPJK) for examination and diagnosis.

Histopathological examination revealed numerous nematodes embedded in a hyperplastic epidermis. Preliminary examination of the remainder of the lesion indicated that the worms belonged to the genus *Stephanofilaria* Ihle & Ihle-Landenberg, 1933, but they could not be assigned to any known species.

Materials and methods

Nematodes were dissected out of the lesion mentioned above using a stereoscopic microscope. Fourteen females, including the holotype, and two males, the allotype and a paratype, were recovered. In addition, the anterior and posterior ends of several females, as well as one anterior and two posterior ends of males, were found. All the specimens are housed in the collection of the Museum National d'Histoire Naturelle, Paris, number MNHN 122HS. Measurements were derived from *camera lucida* drawings.

***Stephanofilaria thelazioides* n. sp.**

Type-host and locality: *Hippopotamus amphibius* Linnaeus, 1758, from the Kruger National Park, Republic of South Africa.

Description (Figs 1–3)

Cuticle without frills but transverse striations well-defined and large at level of mid-body (Fig. 2E). Head with projecting cuticular ring, hexagonal in apical view (Fig 1); external border of ring vertical; internal border gently sloping. Six small bifid cuticular internal labial spines on internal aspect of ring; further posterior are 4 cuticular external labial spines and, at their bases, 4 cephalic papillae. Amphids at level of external labial spines. Deirids bifid (Figs 2D, 3B), posterior to nerve-ring. Oral opening hexagonally-rounded; buccal cavity with thin wall. Oesophagus joining intestine immediately posterior to nerve-ring (Figs 2C, 3A).

Female. Lateral alae absent. Vulva near anterior end; vagina slender anteriorly; ovejector initially slightly dilated, then tubular (Figs 2B, C); opisthodelphic; ovaries and oviducts short. Tail almost straight or slightly curved ventrally, with rounded tip and subterminal phasmids (Fig 2H). Few microfilariae present in females examined; head slightly narrower than body; posterior end conical; sheath with same shape as microfilaria.

Male. Testes at mid-body; vas deferens runs anteriorly and reflexes near posterior extremity of oesophagus. *Area rugosa* with pattern of small beads packed close together, situated latero-ventrally (Figs 3D,E,G); at border of *area rugosa* bead-like pattern replaced by small longitudinal ridges (batonnets). Cuticle expanded in pre- and postcloacal regions, ventrally and laterally, forming caudal vesicle with lateral alae. Fifteen pairs of caudal papillae, arranged in 3 groups: one group of ventral ventro-lateral papillae comprising single precloacal papilla, one adanal pair, 2 strongly pedunculate postcloacal pairs and 2 subterminal pairs (Figs 3D, E); one group of 2 latero-ventral rows of pedunculate papillae, hind-most being at level of cloaca, the anterior-most 300 μm anterior to cloaca; and one group of 2 lateral rows of 3–4 pedunculate pairs in pre- and postcloacal regions (Fig 3F). Left spicule long and thin (Fig 3H) with shaft slightly longer than blade and spirally twisted distal tip (Fig 3I). Right spicule with shaft and blade only slightly differentiated; membranous alae present along blade; tip obtusely conical (Fig 3J). Gubernaculum lightly sclerotised.

Measurements (in micrometres unless otherwise stated)

Females. (Measurements are for holotype, with range in parentheses). Body 6.8 (6.3–6.8) mm long and 135 (135–180) wide; nerve-ring and deirids 110 (90–150) and 210 (140–275) respectively from anterior end; buccal capsule length 10 (10–11); oesophagus 150 (144–171) long; vulva 35 (22–35) from anterior end; vagina 220 (130–220) long; ovejector 280 long; tail 60 (50–60) long. Microfilariae expressed from ovejector 180 (160–195) long and 8 (8–10) wide.

Males. (Measurements are for allotype, with paratype fragments in parentheses). Body 3.2 (3.2) mm long, 80 (90) wide; length of buccal cavity 7 (7); nerve-ring and deirids 100 (80) and 160 (127) respectively, from anterior extremity; tail 30 (25, 25) long; left spicule 945 (880, 925) long with blade 500 (500, 475) long; right spicule 130 (120, 148) and gubernaculum 35 (25, 35) long.

Discussion

Sonin (1977) and Johnson (1987) revised the species of the genus *Stephanofilaria* and no new species have been added subsequently. Like Johnson (1987), we are of the opinion that *Stephanofilaria andamani* Sinha & Das, 1958 from the water buffalo *Bubalus bubali* in the Andaman islands and *Stephanofilaria srivastavi* Bhattacharjee, 1967 from the elephant *Elephas maximus* in India are *species inquirendae*, because “no taxonomic descriptions have been published in support of either name” (Johnson, 1987). Four species are currently recognised: *Stephanofilaria dedoesi* Ihle & Ihle–Landenberg, 1933 is a parasite of cattle, goats and water buffalo in Indonesia. Johnson (1987) considered *S. assamensis* Pandit, 1936 from India, *S. kaeli* Buckley, 1937 from Malaysia and *S. okinawaensis* Ueno & Chibana, 1977 from Japan (erroneously assigned to *S. rono* Kono, 1965 by Ivashkin, Shmytova & Koishibaev, 1971) as probable synonyms of *S. dedoesi*. All these species (or subspecies?, or synonyms?) have several features in common: cuticle with frills; head with a circle of internal labial spines, a circle of external labial spines and four cephalic papillae; female with the vulva at the level of the nerve-ring and a short conical tail; male with the left spicule 150–230 μm long, right spicule 40–50 μm long, gubernaculum present and lateral alae absent; and microfilariae 85–140 μm long with an elongate sheath.

Stephanofilaria zaheeri Singh, 1958 is also a parasite of cattle in Asia. Nevertheless, it may be distinct from *S. dedoesi* in that it has more internal and external

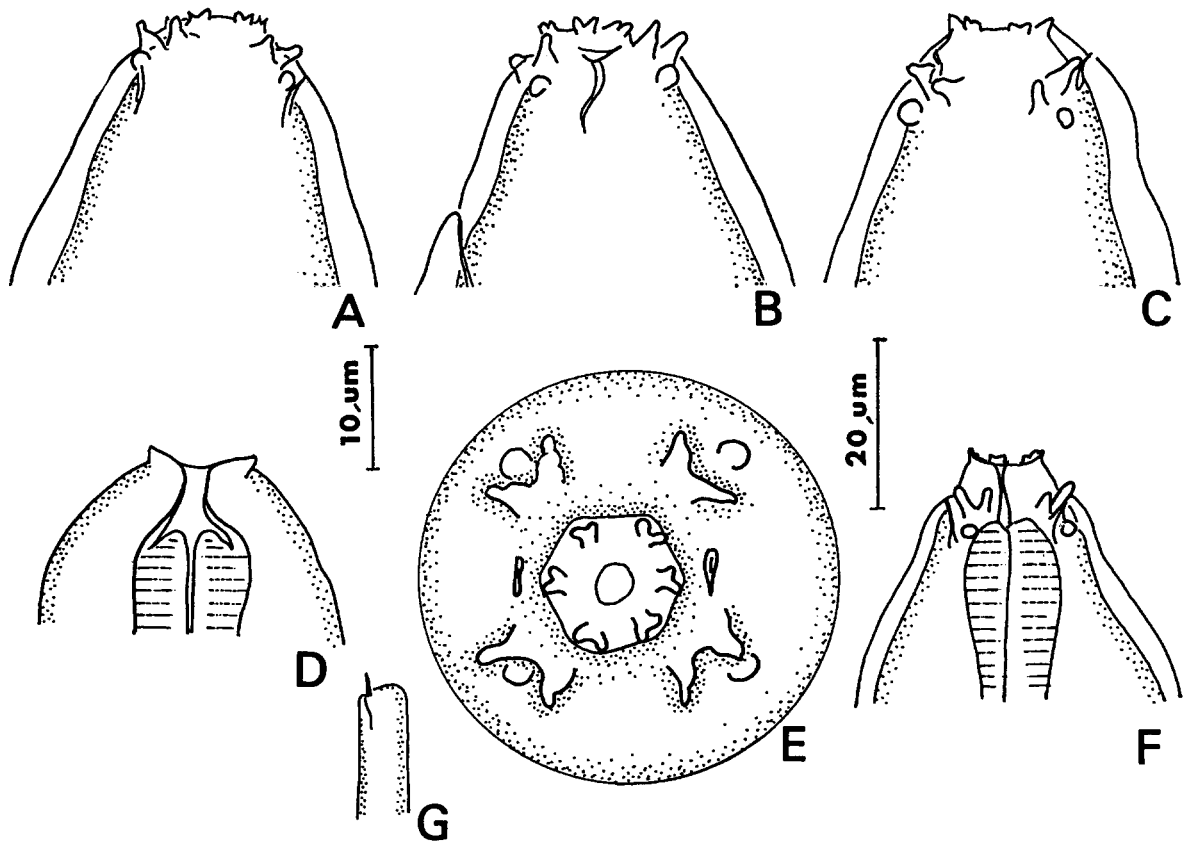


Fig. 1. *Stephanofilaria thelazioides* n. sp. Cephalic extremities of the female (A–E), the male (F) and the microfilaria (G). A, dorsal view; B, left lateral view; C, ventral view (since the head is slightly curved dorsally the internal labial papillae are more visible in A and the external labial papillae in C); D, longitudinal optical section, lateral view; E, apical view; F, lateral view; G, ventral view. Scale-bars: A,B,C,D,F, 20 μm ; E,G, 10 μm .

labial spines (23–24 vs 15–18, and 28–32 vs 16–23). Singh (1958) stated that four external labial and four cephalic papillae are present, but these are difficult to see in such a small worm.

Stephanofilaria stilesi Chitwood, 1934 is a parasite of *Bos taurus* in the Nearctic region, but it has also been found in Russia. It differs from the previous species in the configuration of the cephalic structures, i.e. the external labial papillae are not transformed into cuticular spines (Anderson, 1968), there are five submedian cuticular spines (lateroventral according to Hibler, 1966), and frills on the cuticle of the body are absent. The microfilariae are also much smaller (40–60 μm long, 2–4 μm wide) and possess an ovoid sheath (Hibler, 1966).

Stephanofilaria dinniki Round, 1964 is a parasite of the black rhinoceros *Diceros bicornis* (Linnaeus, 1758) in East and South Africa. This species has no frills on

the cuticle of the body. Round (1964) stated that the head bears a crown of 11–12 peribuccal spines (but illustrates 16), eight spines grouped in pairs slightly posteriorly and four cephalic papillae. This species also differs from the others in that the vulva is situated more anteriorly, the vagina is longer, the nerve-ring is further posterior, and the tail of the female is longer than wide and its tip is rounded; the spicules are also longer, the caudal alae are present and the cloacal papillae are pedunculate.

The last named species is the only one that bears close resemblance to our specimens. It was not possible to do comparative studies because the material from the rhinoceros had been lost. Nevertheless, the description of Round (1964) is precise and clearly shows the distinctive characteristics of the head, the caudal papillae and the dimensions of the spicules, gubernaculum and microfilariae. *S. dinniki* differs from our speci-

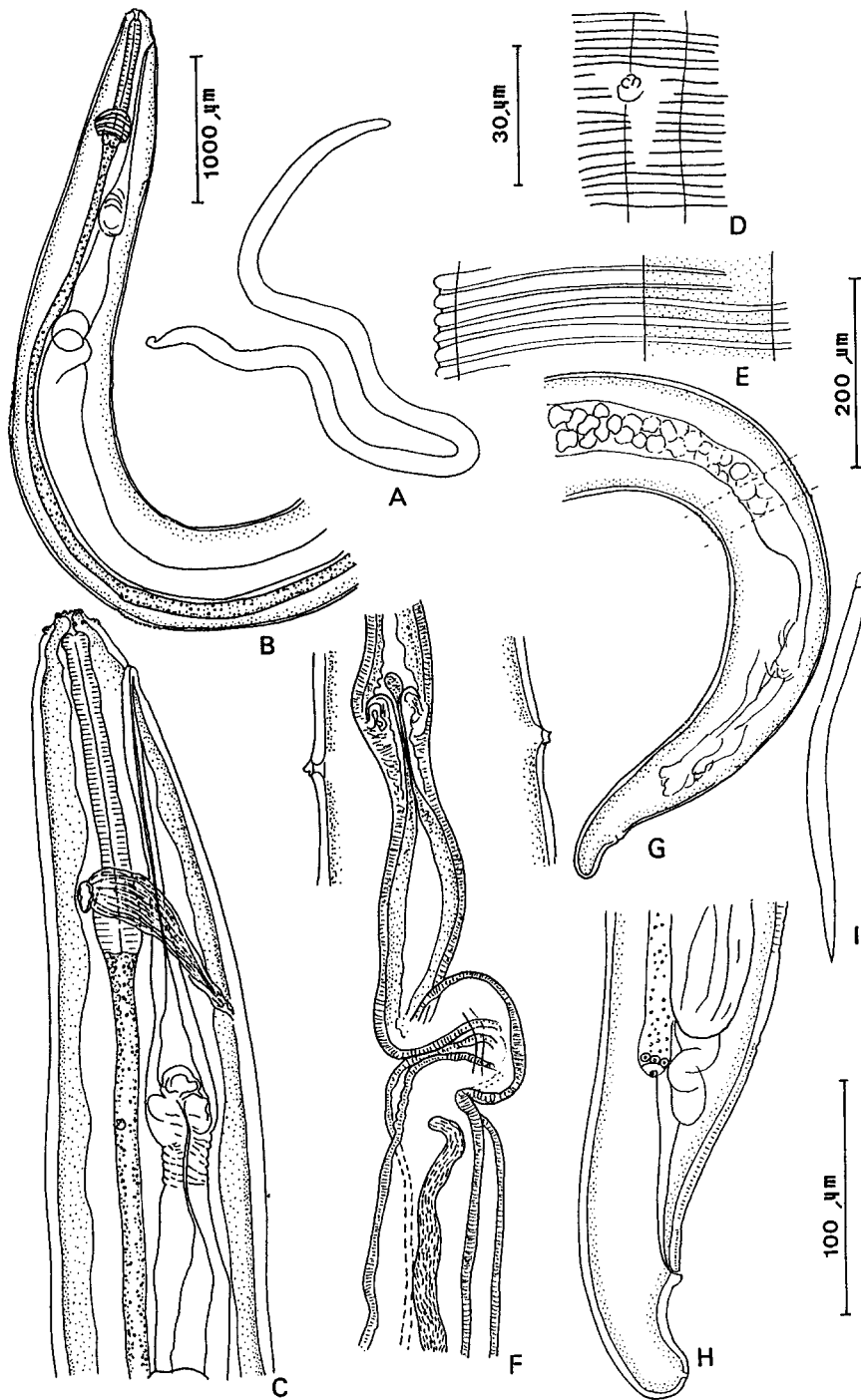


Fig. 2. Stephanofilaria thelazioides n. sp. Female. A, entire body; B, anterior region in right lateral view; C, anterior extremity in right lateral view showing the oesophagus and vagina; D, deirid; E, cuticular ornamentation; F, deirids and the division of the ovejector; G, posterior region; H, posterior extremity; I, microfilaria. Scale-bars: A, 1,000 μm ; B, G, 200 μm ; C, F, H, 100 μm ; D, E, I, 30 μm .

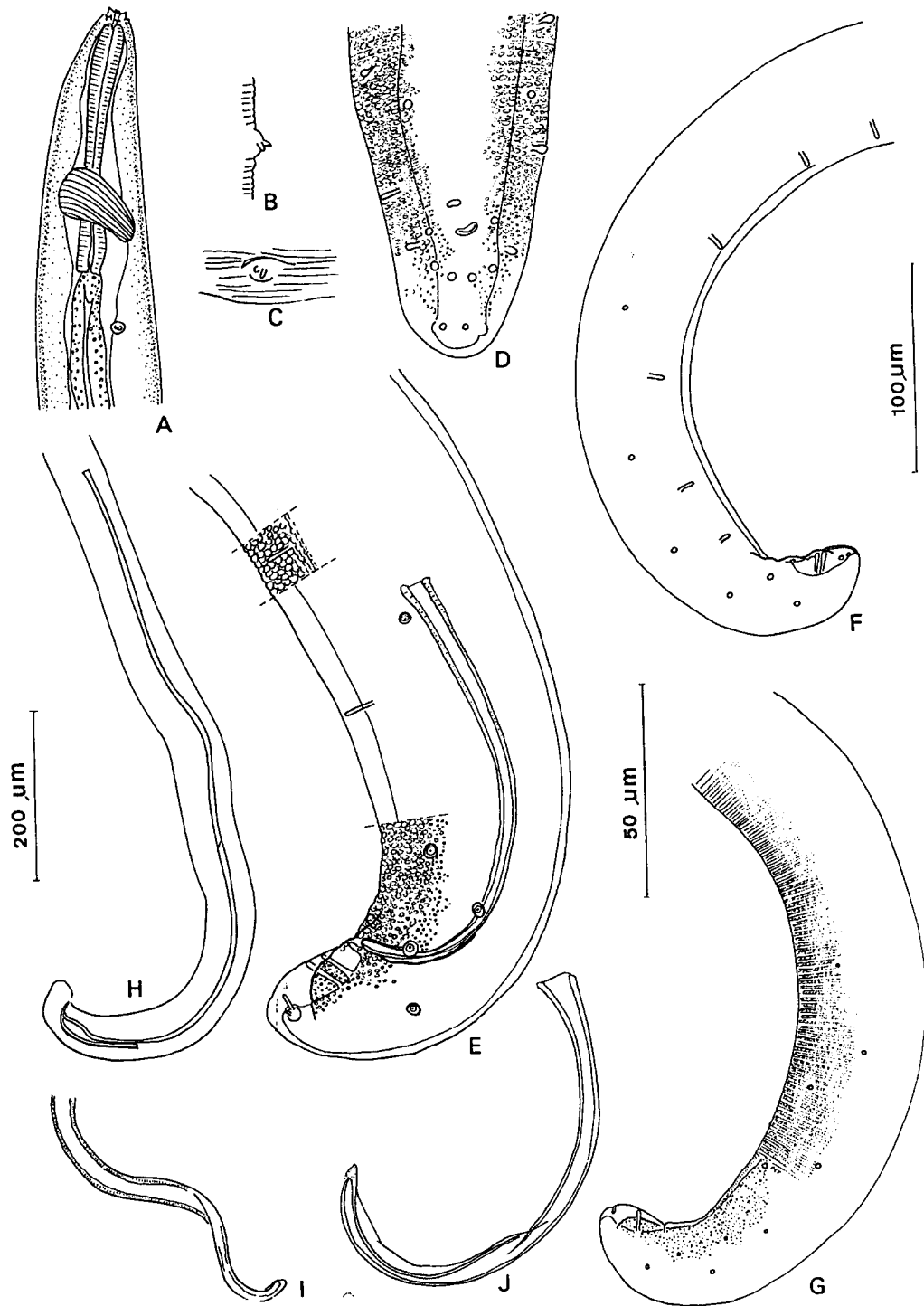


Fig. 3. Stephanofilaria thelazioides n. sp. Male. A, anterior extremity, right lateral view; B, deirid, median view; C, deirid, lateral view; D, posterior extremity, ventral view; E, posterior extremity, left lateral view, showing the right spicule and gubernaculum (the cuticular ornamentation is not drawn at mid-body); F, posterior extremity, right lateral view, showing the arrangement of the cloacal papillae; G, posterior extremity, left lateral view, showing the cuticular ornamentation; H, spicules; I, tip of the left spicule; J, right spicule. *Scale-bars:* H, 200 μm ; A, D, F, G, 100 μm ; B, C, E, I, J, 50 μm .

mens in the following respects: the head bears 12 (or 16?) internal labial spines arranged regularly, while our specimens have six distinct bifid spines; the three pairs of ventral postcloacal papillae are close together, while in our specimens one pair is adanal and two pairs are clearly more posterior; the left and right spicules and the gubernaculum are, respectively, 62–115, 530–750 and 11–14.5 μm long, while in our specimens they are 880–945, 120–148 and 25–35 μm in length; and the microfilariae are 120–150 μm long and have a cuticular cap, while in our specimens they are 180 μm long and without a cap.

In view of these differences, we consider the specimens from the hippopotamus to be a new species for which we propose the name *Stephanofilaria thelazioides* n. sp. The specific name refers to the affinities of *Stephanofilaria* with the genus *Thelazia* Bosc, 1819, previously established by Anderson (1957), Anderson & Bain (1976) and Bain (1981), which are especially apparent in the new species.

The different species of the genus *Stephanofilaria* can be classified phyletically. They have a cephalic ring which is already present in the larval stages (Hibler, 1966). The spines on the head, equally characteristic of the genus, are derived by modification of the sensory papillae, and these spines have become more modified as the species evolve. Several other characteristics give an indication of the degree of specialisation in each species: female and male tails that are shortened to a lesser or greater degree; caudal papillae which may or may not be numerous and have a lesser or greater tendency to accumulate in the cloacal region; and the considerable variation in the length of the spicules.

Based on the above, *S. thelazioides* from the hippopotamus appears to be the most primitive. *S. dinniki* from the rhinoceros is closely related, but the disposition of the spines on the head and the cloacal papillae are slightly more specialised. The species parasitising the domesticated Asiatic mammals (water buffalo, cattle, goats and elephants) are clearly more evolved, as is indicated by the multiplication of the internal labial and external labial spines and the shorter tail, as well as by the development of cuticular frills covering the body. *S. stilesi*, a parasite of cattle in North America, has the most advanced morphology in that the cuticular spines on the head are replaced by a small newly formed group of latero-ventral spines and in that the tail of the male is very short with the papillae arranged around the cloaca (Ivashkin, Timofeeva & Khromova, 1961).

Hippopotami, rhinoceroses, elephants and water buffaloes are mammals without obvious zoological affinities, but they have a thick skin in common. The evolution of the genus *Stephanofilaria* seems to be associated with this favourable biotope rather than with the phylogeny of the hosts. We assume that this genus is of African origin, was introduced into Asia and that its occurrence in cattle is recent.

Acknowledgements

We thank Dr Roy Bengis, State Veterinarian, Skukuza, Kruger National Park, for submitting the lesion in the skin of the hippopotamus, and Mme Roselyne Tchep-rakoff, Muséum National d'Histoire Naturelle, for her technical assistance. Part of this study was made possible with a grant to the first author from the Foundation for Research Development.

References

- Anderson, R.C. (1957) The life cycles of dipetalonematid nematodes (Filarioidea: Dipetalonematidae); the problem of their evolution. *Journal of Helminthology*, **31**, 203–224.
- Anderson, R.C. (1968) The comparative morphology of cephalic structures in the superfamily Filarioidea (Nematoda). *Canadian Journal of Zoology*, **46**, 181–199.
- Anderson, R.C. & Bain, O. (1976) Keys to the genera of the order Spirurida. Part 3. Diplotriaenoidea, Aproctoidea and Filarioidea. *In: Anderson, R.C., Chabaud, A.G. & Willmott, S. (Eds) CIH keys to the nematode parasites of vertebrates*. No. 3, Wallingford, UK: CAB International, pp. 59–116.
- Bain, O. (1981) Filariids and their evolution. *In: Willmott, S. (Ed) Evolution of helminths*. Workshop Proceedings EMOP 3. *Parasitology*, **82**, 167–168.
- Hibler, C.P. (1966) Development of *Stephanofilaria stilesi* in the horn fly. *Journal of Parasitology*, **52**, 890–898.
- Ivashkin, V.M., Timofeeva, T.N. & Khromova, L.A. (1961) Stephanofilariasis in cattle. *Trudi Gelmintologicheskoi Laboratorii*, **11**, 109–114. (In Russian).
- Ivashkin, V.M., Shmytova, L.A. & Koishibaev, G.K. (1971) Stephanofilariasis in herbivores. *Veterinariya Moscow*, **3**, 66–68. (In Russian).
- Johnson, S.J. (1987) Stephanofilariasis – a review. *Helminthological Abstracts, Series A. Animal and Human Helminthology*, **56**, 287–299.
- Round, M.C. (1964) A new species of *Stephanofilaria* in skin lesions from the black rhinoceros (*Diceros bicornis* L.) in Kenya. *Journal of Helminthology*, **38**, 87–96.
- Sonin, M.D. (1977) [Filariata of animals and man and diseases caused by them. Part 4. Onchocercidae.] *Osnovy Nematologii*, **28**, 220 pp. (In Russian).