

## *Afenestrata axonopi* n. sp. (Nemata : Heteroderidae) from Brazil

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**Summary** – *Afenestrata axonopi* n. sp. is a parasite of *Axonopus marginatus* (Poaceae), a perennial grass native to Brazil. This new species is close to *A. sacchari* Kaushal & Swarup, 1988 and *A. koreana* Vovlas, Lamberti & Choo, 1992, and is characterized by large vulva-anus distance, vulval lips close together, vulval slit slightly sunken, vulval cone terminus with dorsal-ventral wrinkles, absence of underbridge, and occurrence of variant cysts with thin cuticle at the cone terminus. Second-stage juveniles have two lip annuli and three lateral incisures with dots.

**Résumé** – *Afenestrata axonopi* n. sp. (Nemata : Heteroderidae) provenant du Brésil – *Afenestrata axonopi* n. sp. parasite *Axonopus marginatus* (Poaceae), une graminée pérenne, indigène du Brésil. Cette nouvelle espèce, proche d'*A. sacchari* Kaushal & Swarup, 1988 et d'*A. koreana* Vovlas, Lamberti & Choo, 1992, est caractérisée par la grande distance vulve-anus, la vulve fermée, la fente vulvaire légèrement encaissée, des rides dorso-ventrales sur le cône vulvaire, l'absence de sous-pont et par la présence de certains kystes à cuticule amincie au niveau du cône. Les juvéniles de deuxième stade présentent deux anneaux céphaliques et un champ latéral ponctué comportant trois incisures.

**Key-words** : *Afenestrata*, Heteroderinae, taxonomy.

While surveying native cerrado (savanna) for criconematids, the author found an heteroderid parasitizing *Axonopus marginatus* (Trinius) Chase, 1913 (Poaceae), a native perennial grass. Repeated samplings and further studies led to the description of a new species of *Afenestrata* Baldwin & Bell, 1985.

Females were obtained by root dissection, and cysts by the flotation-sedimentation-sieving techniques (Flegg & Hooper, 1970). Second-stage juveniles (J2) were obtained by hatching eggs from several females. For optical microscopy, the nematodes were fixed with Golden's solution (Hooper, 1970) and mounted in glycerine according to Seinhorst (1959). For scanning electron microscopy, specimens in glycerine were directly coated with gold (Sher & Bell, 1975), and observed with a JEOL JSM 840 A at 5 kV.

### *Afenestrata axonopi*\* n. sp. (Figs 1-3)

#### MEASUREMENTS

See Table 1.

#### DESCRIPTION

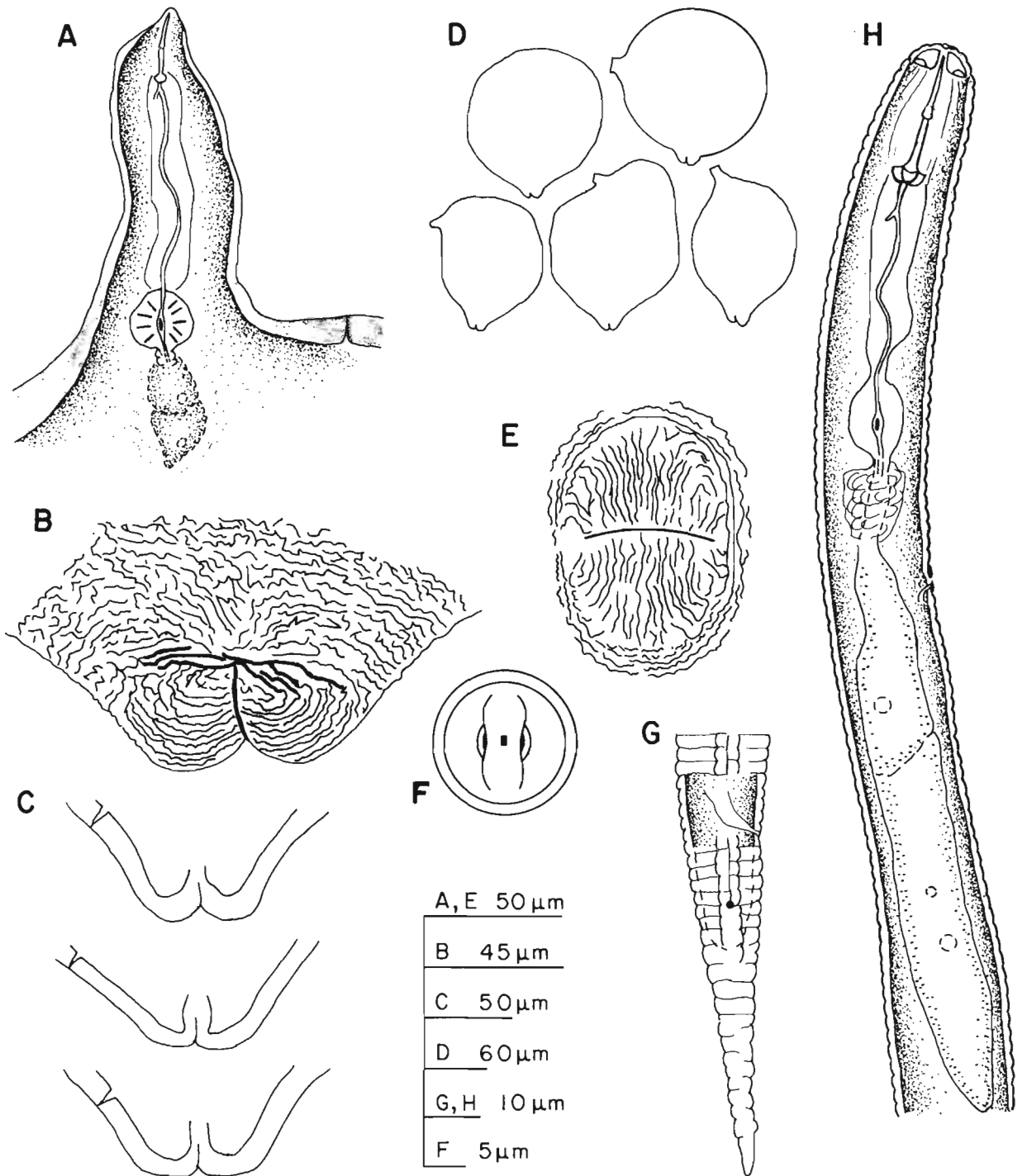
*Females* : Body white to tan, predominantly (90 %) lemon shaped to subspherical, others ovoid or irregular. Neck elongate (n = 23)  $118 \pm 30.9$  (84-216)  $\mu\text{m}$  long, laterally projected and sometimes with asymmetrical expansions. Neck cuticle not tanned, thin (maximum thickness, 6  $\mu\text{m}$ ), often breaking during root dissection. Cuticle thick in the dilated body, 7-13  $\mu\text{m}$  in young females and 12-20  $\mu\text{m}$  in mature females and cysts. Anterior part of the neck smooth or annulated, posterior part with transverse tubercles. Dilated body with lace-like pattern, with deeper grooves in the "shoulder" and vulval regions. At the vulval cone terminus the transverse ridges and grooves are modified into dorsal-ventral wrinkles. Subsurface punctations are not visible in recently molted females, whereas they are refringent in young females, and dark in mature females and cysts. Punctations do not occur at the cone terminus, but fine subsurface transversal lines are present in this region. Labial disc elevated, square, with raised ring in the center. Lips fused forming an irregular cephalic plate. Amphid openings visible. Stylet straight or with slightly bent conus, with round backward sloping knobs. Procorpus bulky, with constriction at junction with metacarpus. Metacarpus ellipsoidal to subspherical, located at shoul-

\* From the host generic name *Axonopus*.

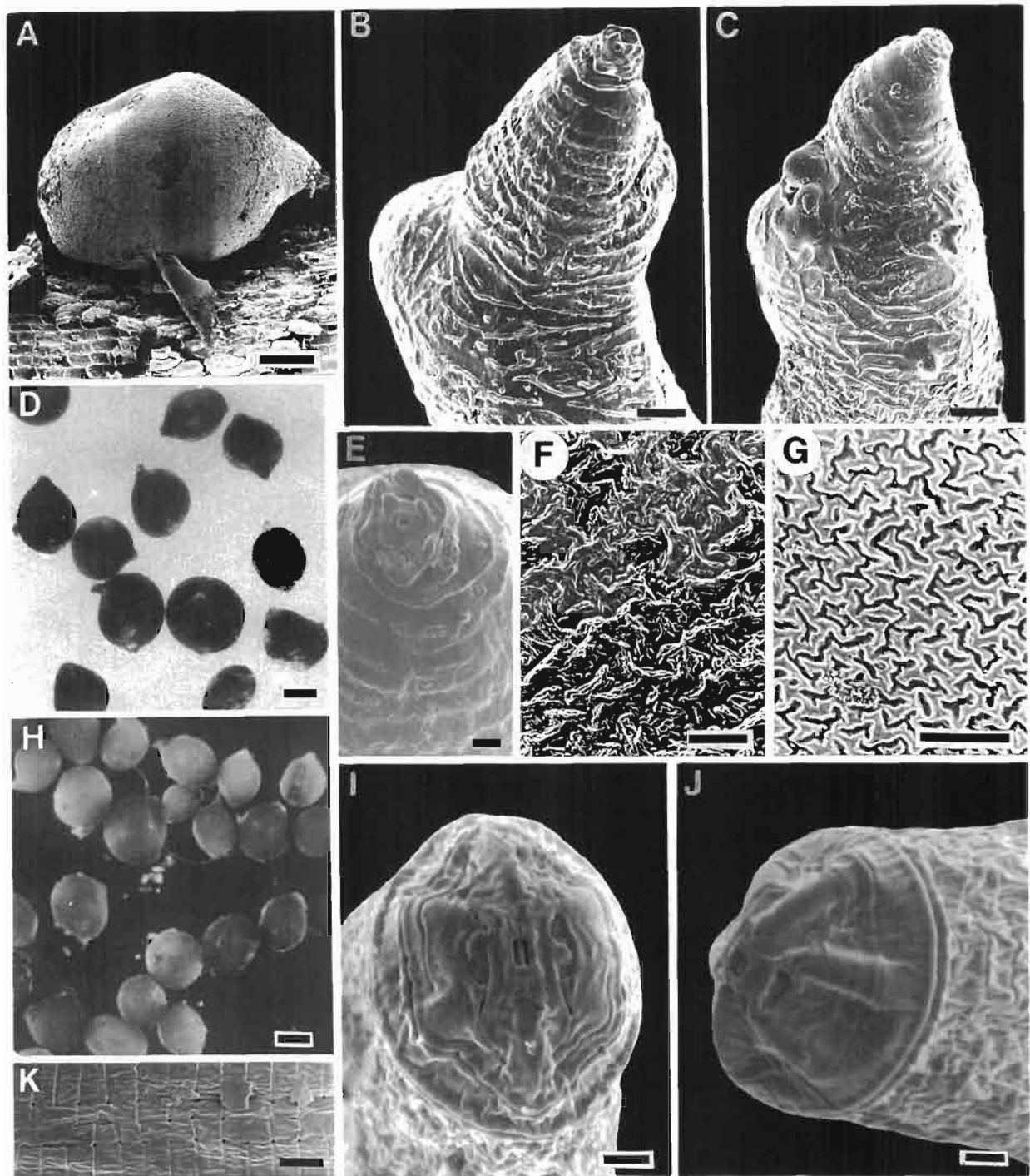
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**Table 1.** Morphometrical characters of *Afenestrata axonopi* n. sp. (all measurements in  $\mu\text{m}$ ).

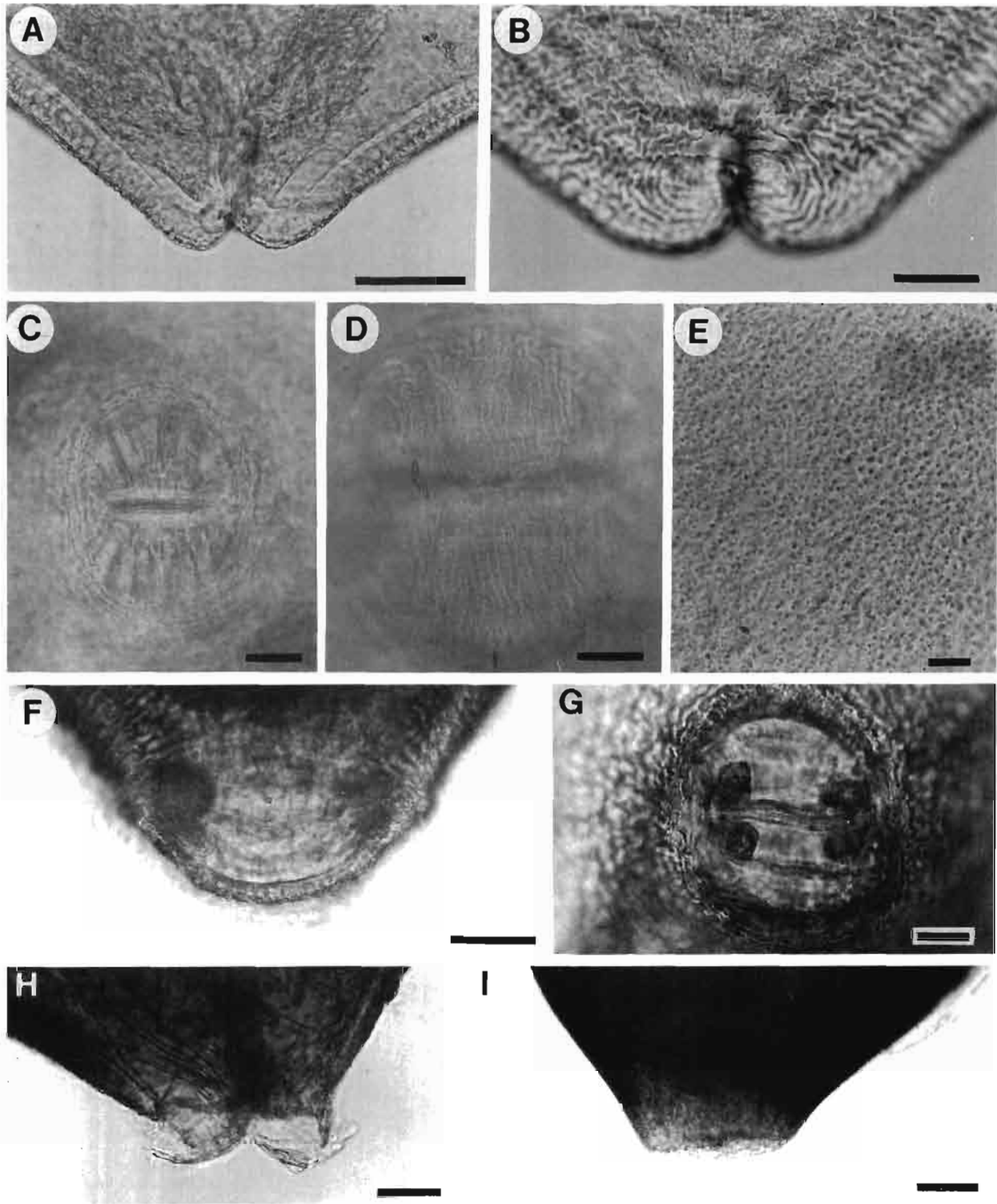
Character	Female		Cyst		Juvenile		Holotype
	n	X $\pm$ SE (min-max)	n	X $\pm$ SE (min-max)	n	X $\pm$ SE (min-max)	
L					42	384 $\pm$ 18.2 (340-415)	
L. dilat. portion	23	500 $\pm$ 70.7 (371-656)	31	558 $\pm$ 73.7 (408-703)			545
Width	23	394 $\pm$ 51.1 (304-523)	31	459 $\pm$ 71.7 (352-608)	41	18.3 $\pm$ 1 (16.2-20.4)	419
a	23	1.3 $\pm$ 0.2 (1.1-1.6)	31	1.2 $\pm$ 0.1 (1-1.5)	41	21.1 $\pm$ 1.1 (19.5-24.7)	1.3
Stylet	20	23.4 $\pm$ 1.3 (21-26.7)			37	21.6 $\pm$ 0.4 (20.6-22.5)	24.8
Stylet conus	15	11.7 $\pm$ 1.2 (9.7-14.8)			33	9.9 $\pm$ 0.6 (8.8-11.2)	12.9
Stylet shaft	14	8.9 $\pm$ 1.1 (7.2-10.8)			37	9.3 $\pm$ 0.5 (8.1-10.3)	8.9
M %	15	50.5 $\pm$ 2.9 (45.8-55.4)			33	45.5 $\pm$ 2.2 (42.1-51.4)	52.1
Knobs (height)	20	2.8 $\pm$ 0.5 (2-3.5)			41	2.6 $\pm$ 0.2 (2-2.9)	3
Knobs (width)	20	4.8 $\pm$ 0.6 (4-6)			41	5 $\pm$ 0.2 (4.7-5.5)	4.9
DGO	20	5.7 $\pm$ 1.1 (4.2-7.5)			37	5.2 $\pm$ 0.6 (3.7-6.6)	5.6
O %	20	24.4 $\pm$ 4.5 (17.9-32.6)			35	24 $\pm$ 2.5 (20.9-30.3)	22.6
Metacarpus (length)	11	27.4 $\pm$ 3.8 (23-35)			41	13 $\pm$ 1 (10.8-14.9)	25
Metacarpus (width)	11	22 $\pm$ 2.3 (18.3-27)			41	9.2 $\pm$ 0.7 (7.8-10.8)	23.3
Hemizonid-ant. end					40	82.6 $\pm$ 3.8 (72.5-90.2)	
Exc. pore-ant. end	11	135 $\pm$ 22 (105-170)			39	85.4 $\pm$ 3.8 (75.9-93.1)	
Exc. pore %					39	22.3 $\pm$ 1.1 (19.5-25.4)	
Vulva-anus distance (lateral view)	42	88.1 $\pm$ 11 (70-115)					81
Vulval slit length	33	47.3 $\pm$ 3.8 (40.2-55.2)					
Tail					38	54 $\pm$ 3.8 (43.4-60.8)	
c					38	7.1 $\pm$ 0.5 (6.3-9.3)	
c'					38	4.6 $\pm$ 0.3 (3.8-5.6)	
Hyaline region					39	34.1 $\pm$ 3 (29.2-42.4)	
Hyaline region %					38	63.4 $\pm$ 4.3 (55.3-73.5)	



**Fig. 1.** *Afenestrata axonopi* n. sp. Female. A : Anterior region; B, E : Cuticle pattern at the vulval cone; C : Vulval cone shapes – Cyst : D ; Shapes – J2 : F : En face pattern; G : Posterior region; H : Anterior region.



**Fig. 2.** *Afenestrata axonopi* n. sp. Female : A. Female on root surface of host; B, C : Anterior region; E : En face pattern; F, G : Cuticle patterns; H : Whole body – Cyst ; D : Whole body – J2 : I : En face pattern; J : Lip region; K : Lateral fields. (Bar equivalent : A = 100  $\mu$ m; B = 2  $\mu$ m; C, F = 5  $\mu$ m; D, H = 220  $\mu$ m; E, I, J = 1  $\mu$ m; G = 10  $\mu$ m; K = 2.5  $\mu$ m.).



**Fig. 3.** *Afenestrata axonopi* n. sp. Vulval region, Mature female : A, B : Lateral view; C : Protractor muscles; D : Cuticle pattern at cone terminus; E : Subsurface punctations – Cyst; F : Variant cone with thin cuticle at cone terminus; G : End-on view beneath cone terminus; H, I : Variant ruptured vulval cones. (Bar equivalent : A, I = 10  $\mu$ m; B, C, F-H = 5  $\mu$ m; D = 3  $\mu$ m; E = 2  $\mu$ m.)

der level. Oesophageal glands ( $n = 6$ )  $36 \pm 6.5$  (27-42)  $\mu\text{m}$  long and  $18.4 \pm 5.3$  (12-30)  $\mu\text{m}$  wide. Vulval cone present ( $n = 34$ )  $55.8 \pm 10.9$  (36-78)  $\mu\text{m}$  high and  $143 \pm 18.3$  (110-192)  $\mu\text{m}$  wide at the base, predominantly (75%) with round, narrow terminus, the rest with flat or wide terminus. Vulva terminal. Vulval lips close together, not widely separated. Vulval slit slightly sunken. Ten to twelve vulval protractor muscles distinct. Phasmids not observed. Extruded gelatinous matrix containing a few eggs in some females. Subcrystalline layer present.

**Cysts:** Light to dark brown, predominantly (90%) lemon shaped to spherical, the rest ovoid or irregular. Bullae and underbridge absent. Vulval cone without fenestration, its cuticle typically thickened and relatively little tanned. In 13 of the 134 females and cysts observed the cuticle was thin at the cone terminus, irregularly torn, and sometimes the vulval bridge or the entire vulval cone was lost. Two hundred twenty-six (120-370) eggs per cyst,  $99.6 \mu\text{m}$  long,  $38.4 \mu\text{m}$  wide.

**Juveniles:** Body ventrally curved after fixation. Cephalic region offset ( $n = 38$ )  $4 \pm 0.4$  (3.2-4.9)  $\mu\text{m}$  high and  $9.8 \pm 0.5$  (8.8-10.5)  $\mu\text{m}$  wide, with two annuli lacking anastomoses. *En face* pattern with labial disc fused to submedian lips; adjacent submedian lips fused with each other and, with few exceptions, partially fused with second head annulus. Anterior cephalids located at the 7th to 9th body annulus, and posterior cephalids located three to four annuli more posterior. Stylet robust, straight, knobs large with flat anterior surface. Metacarpus ellipsoidal, at ( $n = 39$ )  $55 \pm 2.2$  (51-60)  $\mu\text{m}$  from anterior end. Oesophageal glands filling body cavity, terminating at ( $n = 37$ )  $181 \pm 8.5$  (157-195)  $\mu\text{m}$  from anterior end. Hemizonid lens-like, one and a half annuli long, just anterior to the excretory pore and twelve to fourteen annuli anterior to the hemizonion. Genital primordium at ( $n = 30$ )  $209 \pm 11.7$  (181-241)  $\mu\text{m}$  from anterior end. Tail tapering. Phasmids pore-like, five annuli posterior to anus. Lateral field with three areolated incisures with dots.

**Males:** Unknown.

#### DIAGNOSIS AND RELATIONSHIPS

The distinguishing characters of *A. axonopi* n. sp. are the large vulva-anus distance, the closed together and dorsal-ventrally wrinkled vulval lips, and the slightly sunken vulval slit.

*A. axonopi* n. sp. is close to *A. sacchari* Kaushal & Swarup, 1988 and *A. koreana* Vovlas, Lamberti & Choo, 1992. *A. axonopi* n. sp. can be distinguished from *A. sacchari* by the underbridge (absent *vs* present), the position of vulval lips (close together *vs* widely separated), the degree of sunken of the vulval slit (slight *vs* deep), by the shorter DGO ( $5.2 \pm 0.6$  *vs* about 8  $\mu\text{m}$ ) in females, and the number of lip annuli (2 *vs* 3-4) and lateral incisures (3 *vs* 4) in J2. *A. axonopi* n. sp. can be dis-

tinguished from *A. koreana* by the larger vulva-anus distance ( $88.1 \pm 11$  *vs* 63  $\mu\text{m}$ ), the cuticle pattern at the vulval cone terminus (dorsal-ventrally wrinkled *vs* tuberculated), the vulval cone dimensions ( $55.8 \pm 10.9$  high  $\times$   $143 \pm 18.3$  wide *vs*  $113 \pm 12.2 \times 73 \pm 10.4 \mu\text{m}$ ), and the larger stylet in females ( $23.4 \pm 1.3$  *vs* 16.8  $\mu\text{m}$ , from authors' drawing) and in J2 ( $21.6 \pm 0.4$  *vs*  $18 \pm 1.5 \mu\text{m}$ ).

#### TYPE HOST AND LOCALITY

Roots and rhizosphere of *Axonopus marginatus* (Trinius) Chase, 1913 (Poaceae), alongside Estrutural highway, Km 1, Brasilia, Brazil.

#### TYPE SPECIMENS

Holotype female and paratypes were deposited in the Nematode Collection of Departamento de Fitopatologia, Universidade de Brasilia, Brasilia (DF), Brazil. Two groups of five female paratypes and ten J2 paratypes were each deposited at Muséum National d'Histoire Naturelle, Paris, France, and Department of Nematology, University of California, Riverside, CA, USA.

#### *Afenestrata* Baldwin & Bell, 1985 = *Afrodera* Wouts, 1985

#### DIAGNOSIS AMENDED

Heteroderinae. **Female:** Cyst stage present. Body lemon shaped, sub spherical or spherical, with projecting neck. Cuticle thick with superficial lace-like pattern in dilated body. D-layer absent. Vulval cone present. Vulva terminal, vulval lips close together or widely separated, vulva slit slightly or deeply sunken. Fenestration and bullae absent. Underbridge present or absent. **Male:** Body twisted posteriorly. Three or four lateral incisures. Spicules straight. Cloacal tubus present. No tail. No phasmids. **Juvenile:** Stylet less than 30  $\mu\text{m}$ . Three or four lateral incisures. Oesophageal glands filling body cavity. Phasmids punctiform.

#### TYPE SPECIES

*A. africana* (Luc, Germani & Netscher, 1973) Baldwin & Bell, 1985  
= *Sarisodera africana* Luc, Germani & Netscher, 1973  
= *Afrodera africana* (Luc, Germani & Netscher, 1973) Wouts, 1983

#### OTHER SPECIES

*A. sacchari* Kaushal & Swarup, 1988  
*A. koreana* Vovlas, Lamberti & Choo, 1992  
*A. axonopi* n. sp.

#### Discussion

The characters listed in Table 2 distinguish *Afenestrata* species from the closed related *Brevicephalodera bamboosi* Kaushal & Swarup, 1988, type and only species of

**Table 2.** Main characters of *Afenestrata* species and *Brevicephalodera bamboosi* Kaushal & Swarup, 1988.

Character	<i>A. africana</i>	<i>A. sacchari</i>	<i>A. koreana</i>	<i>A. axonopi</i>	<i>B. bamboosi</i>
<b>FEMALE</b>					
Stylet	25-27	26 (23-30)	16.8 (A)	23.4 ± 1.3	23.8 (21-28)
Vulval cone	present	present	present	present	absent
Vulval lips	widely separated	widely separated	close together	close together	widely separated
Vulval sunken	deep	deep	slight	slight	slight
Vulval slit (µm)	64 (62-68)	NO DATA	49 (42-56)	47.3 ± 3.8	55
Vulva-anus dist. (µm)	68 (49-83)	77	63	88.1 ± 11	NO DATA
Underbridge	absent	present	absent	absent	absent
Cuticle pattern at vulval cone terminus	T (B)	I (B)	TU (B)	DV (B)	NO DATA
<b>JUVENILE</b>					
Stylet (µm)	21 (19-22.5)	24.8 (22-29)	18 ± 1.5	21.6 ± 0.4	20 (19-23)
Lip annuli	2	3-4	2	2	2-3
Incisures	4	4	3	3	3
En face pattern	labial disc fused to subm. lips	NO DATA	labial disc fused to subm. lips	labial disc fused to subm. lips	NO DATA
<b>MALE</b>					
Stylet	26.5 (24.5-28)	26 (23-28)	UNKNOWN	UNKNOWN	25 (24-28)
Lip annuli	4-5	3-4	UNKNOWN	UNKNOWN	2-3
Spicules (µm)	40 (38-44)	36 (30-44)	UNKNOWN	UNKNOWN	35 (31-39)
Cloacal tubus	present	present	UNKNOWN	UNKNOWN	absent

(A) from drawing of Vovlas *et al.* (1992).

(B) transversal (T), irregular (I), dorsal-ventral (DV) wrinkles or tubercles (TU).

the genus. Although a detailed redescription of *B. bamboosi* is needed, its distinction from *Afenestrata* is doubtful. The profile of the vulval region of *Brevicephalodera* was tentatively defined without vulval cone, whereas it is present in *Afenestrata*, a variation similar to that reported among species in *Heterodera* A. Schmidt, 1871 (Golden, 1986) and *Atalodera* Wouts & Sher, 1971 *sensu* Souza & Huang (1994). Furthermore the variation in the cyst shape – from globose to kidney-shaped – do not really distinguish *Brevicephalodera* from *Afenestrata*, since such a level of variation is reported in *Verutus* Esser, 1981, *Punctodera* Mulvey & Stone, 1976 and *Globodera* Skarbilovich, 1959 (Golden, 1986; Luc *et al.*, 1988).

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