

Cryphodera utahensis n. sp., (Heteroderidae), A New Species from Wild Rose in Utah

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Abstract: A new species of *Cryphodera* Colbran, 1966, parasitic on wild rose (*Rosa* sp. L.), is described from Utah, USA. *Cryphodera utahensis* n. sp. most closely resembles *C. podocarpi* (Wouts, 1973) Luc et al., 1978, but is distinct with respect to a number of characters including juveniles with a longer esophagus (206 versus 142 μ m), shorter tail (47 versus 69 μ m), and four rather than three incisures in the lateral field. Females of *C. utahensis* have a terminal protuberance which is more pronounced than in other species. Scanning electron microscope observations of the lip region of males indicate a labial disc surrounded by six lip sectors. However, the remainder of the lip region is comprised of irregularly-shaped plates; similar patterns with longitudinal striae apparently characterize males throughout the genus. The lip pattern of juveniles of *C. utahensis* n. sp. includes an oval labial disc surrounded by six lip sectors; transverse striae extend the height of the lip region without longitudinal striae. The type locality of *C. utahensis* confirms a broader distribution of the genus than was indicated for other species, which are limited to Australia (Queensland) and New Zealand. **Key words:** taxonomy, scanning electron microscopy, morphology, Meloidoderinae.

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The genus *Cryphodera* Colbran, 1966 was erected to accommodate a heteroderid which resembled *Meloidodera* Chitwood et al., 1956 with respect to the absence of a cyst and the striated cuticle, but differed primarily by the posterior position of the vulva (2). *Cryphodera* remained monotypic until Luc et al. (4) synonymized *Zelandodera* (Wouts, 1973) with *Cryphodera*, creating three new combinations. The type species has only been reported from Queensland, Australia, and the remaining species from New Zealand.

Between 1977 and 1981 an undescribed *Cryphodera* sp. was collected from roots and soil associated with wild rose from Clear Creek Canyon, as well as Gates Creek, Utah. Males were only rarely encountered.

MATERIALS AND METHODS

Specimens from the type locality in Utah were fixed in 5% formalin and infiltrated

with glycerin for examination with the light microscope. In addition to whole specimens, some excised heads of females were prepared, as well as terminal protuberances, in glycerin jelly. Measurements and corresponding abbreviations were as previously defined (1). Males and juveniles were infiltrated with glycerin and were mounted for observation with the scanning electron microscope (SEM) as reported by Sher and Bell (5). Females were prepared for SEM by fixing in 3% glutaraldehyde in .025 M phosphate buffer at pH 7.2 for at least 12 h. The specimens were rinsed in distilled water, dehydrated through a graduated series to 100% acetone, and critical-point dried using carbon dioxide. Glycerin-infiltrated and critical-point dried specimens were sputter-coated with 20 nm gold-palladium and examined with a JEOL JSM-35C electron microscope at 5 and 15 KV, respectively.

Specimens of *Cryphodera eucalypti* Colbran, 1966 and paratypes of *C. coxi* (Wouts, 1973) Luc et al., 1978; *C. nothophagi* (Wouts, 1973) Luc et al., 1978, and *C. podocarpi* (Wouts, 1973) Luc et al., 1978 were available from the University of Cali-

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fornia Riverside Nematode Collection (UCRNC) for comparison.

SPECIES DESCRIPTION

Cryphodera utahensis n. sp.

Females (measurements of 20 paratypes): L (including "neck") = 3.38–6.64 mm (mean 5.046 mm, .95% confidence interval \pm .500); width = 1.74–4.66 mm (3.031 mm \pm .384); neck length = 70.0–163.2 μ m (105.63 μ m \pm 12.83) stylet = 41.4–55.8 μ m (48.77 μ m \pm 1.45); stylet shaft and knobs = 18.6–27.0 μ m (19.94 μ m \pm 1.12); DGO = 1.5–3.9 μ m (2.36 μ m \pm .32); median bulb (length) = 24.0–33.6 μ m (26.79 μ m \pm 1.38); median bulb (width) = 22.4–32.0 μ m (25.56 μ m \pm 1.05); valve in median bulb (length) = 4.8–8.0 μ m (6.46 μ m \pm .39); valve in median bulb (width) = 4.3–7.2 μ m (5.62 μ m \pm .45); head end to end of median bulb = 70.4–108.8 μ m (84.85 μ m \pm 5.69); excretory pore from anterior end = 27.2–56.0 μ m (36.82 μ m \pm 3.91); vulva-anus distance (lateral view) = 52.8–88.0 μ m (64.04 μ m \pm 5.76); thickness of cuticle = 9.6–16.0 μ m (12.39 μ m \pm .89); a = 1.13–2.95 (1.73 \pm .21); m = 44.44–62.50 (54.42 \pm 3.32); o = 3.08–8.71 (5.41 \pm .79); length median bulb/width median bulb = .88–1.19 (1.05 \pm .04); excretory pore % of L = 4.07–14.15 (7.59 \pm 1.11); vulva length (four cones) = 48.0–51.2 μ m (49.6 μ m).

Description (females) (Figs. 1, 7–9): Body shape varying from reniform with strong dorsal curvature in small individuals to lemon-shaped in large specimens. Neck usually clearly offset from body contour and frequently reflexed ventrally; terminal protuberance well defined (Fig. 1). Freshly collected specimens pearly white, becoming darker and yellowish-brown after fixation in formalin. Subcrystalline layer thin; brown "cement" apparently attaching head and "neck" region to root. Cuticle thinner in neck region becoming uniformly thick throughout body except slightly thicker in terminal protuberance. Transverse striations well defined and similar to juveniles and males in neck region; body markings rough textured and characterized by fine pits between striae (Figs. 1C, 7–9). Head region with two annules, labial disc squarish with four submedial lobes (Fig. 1B);

stylet cone with prominent ventral curvature (Fig. 1A). Gonads with two ovaries; vulva terminal, with anus at base of protuberance (Figs. 1D–9).

Holotype (female): L = 3.50 mm; width = 1.98 mm, neck length = 88.2 μ m; stylet = 45.0 μ m; stylet shaft and knobs = 19.8 μ m; DGO = 2.7 μ m, median bulb (length) = 27.0 μ m; median bulb (width) = 26.1 μ m; valve in median bulb (length) = 7.2 μ m; valve in median bulb (width) = 6.3 μ m; head end to end of median bulb = 81.0 μ m; excretory pore from anterior end = 30.6 μ m; vulva-anus distance = 81.0 μ m, thickness of cuticle = 12.6 μ m; length median bulb/width median bulb = 1.03; excretory pore % = 8.74. Female as in general description; body shape reniform with strong dorsal curvature. Neck reflexed ventrally; neck and stylet twisted relative to rest of body, giving appearance of dorsal curvature of stylet cone.

Males (measurements of 8 paratypes): L = 7.80–10.32 mm (9.210 mm \pm .793); width = 19.9–21.5 μ m (20.37 μ m \pm .97); lip region (height) = 4.4–6.4 μ m (5.41 μ m \pm .59); lip region (width) = 7.6–9.6 μ m (8.68 μ m \pm .60); stylet = 32.0–34.8 μ m (33.32 μ m \pm 1.04); stylet knobs (length) = 2.9–2.8 μ m (2.37 μ m \pm .26); stylet knobs (width) = 4.8–5.2 μ m (4.90 μ m \pm .18); stylet shaft and knobs = 12.8–18.4 μ m (14.71 μ m \pm 1.59); DGO = 2.1–3.6 μ m (2.81 μ m \pm .39); median bulb (length) = 13.6–18.4 μ m (15.60 μ m \pm 1.35); median bulb (width) = 6.0–7.6 μ m (7.20 μ m \pm .54); esophagus length = 108.0–176.0 μ m (140.06 μ m \pm 16.76); excretory pore from anterior end = 122.1–155.6 μ m (135.69 μ m \pm 8.25); hemizonid from anterior end = 109.2–144.8 μ m (130.10 μ m \pm 8.61); annule width = 1.60–1.92 μ m (1.81 μ m \pm .09); tail length = 3.33–6.80 μ m (5.55 μ m \pm .83); testis length = 232.7–574.0 μ m (408.52 μ m \pm 92.0); spicule length = 22.0–30.8 μ m (25.37 μ m \pm 3.25); gubernaculum length = 7.2–10.2 μ m (8.77 μ m \pm 1.00); a = 40.38–53.33 (45.29 \pm 4.10); b = 4.59–5.95 (5.09 \pm .36); b' = 5.69–7.59 (6.64 \pm .63); c = 124.71–245.69 (171.23 \pm 32.74); c' = .21–.32 (.27 \pm .03) m = 46.51–61.90 (55.88 \pm 4.16); O = 6.35–10.71 (8.45 \pm 1.18); T % = 25.47–56.24 (44.16 \pm 8.42); excretory pore % = 13.48–17.07 (14.87 \pm 1.49).

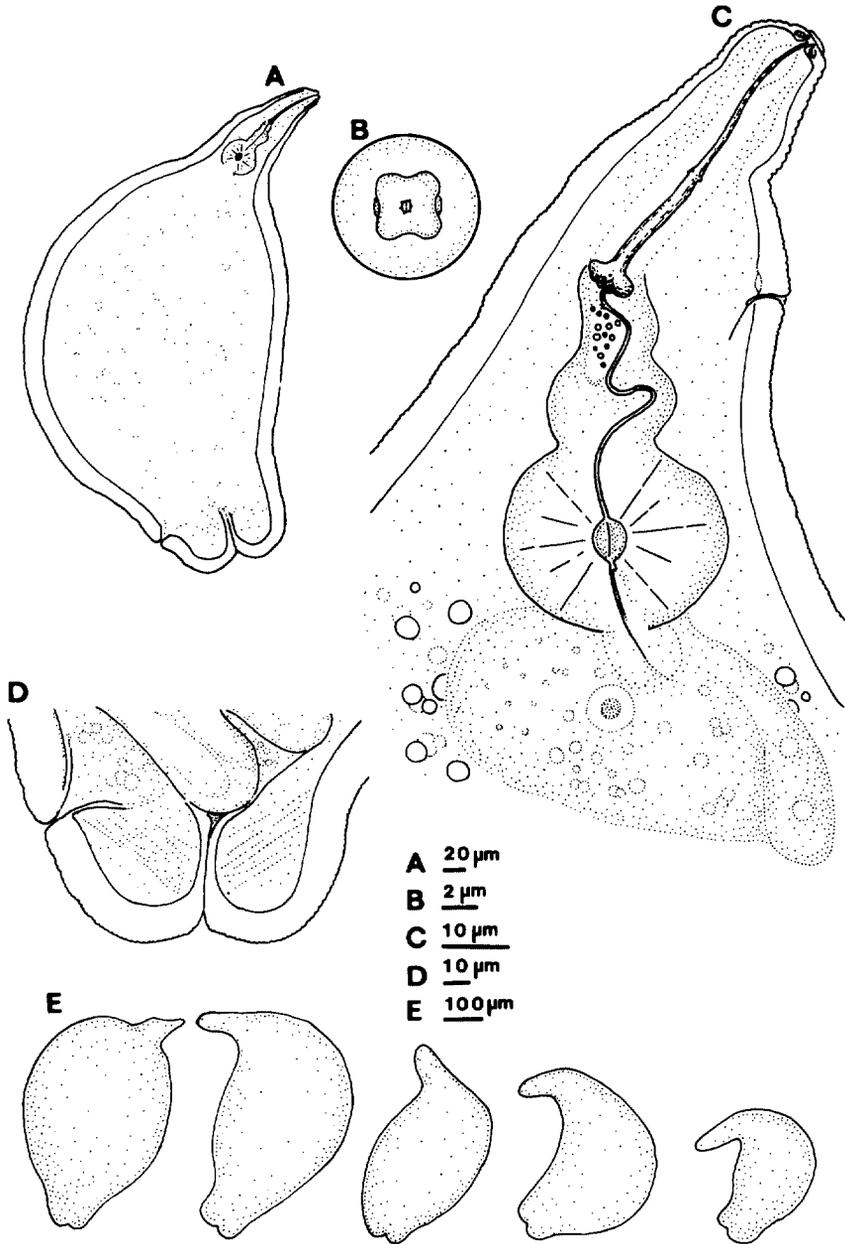


Fig. 1. Drawings of females of *Cryphodera utahensis* n. sp. A) Entire specimen (lateral). B) Face view as observed with SEM. C) Anterior portion (lateral). D) Terminal protuberance showing position of anus and vulva. E) Outline of specimens of varying size and shape.

Description (males) (Figs. 2, 4, 5): Body cylindrical, tapering near anterior terminus; posterior half of heat-killed specimens twisting about 90 degrees. Lip region distinctly offset, with five annules and irregular longitudinal striae (Figs. 2A & D, 4, 5). Amphid opening slit-like. Lateral field with four incisures generally areolated (Fig. 2E), cen-

ter ridge not areolated in tail region. Cephalids sometimes visible at about third and ninth annule; hemizonid about 1½ annules long. Stylet knobs often with posterior slope, sometimes with minute anterior indentation (Fig. 2A-C). Median bulb narrow and elongate; gland lobe very broad, highly variable in length and with indis-

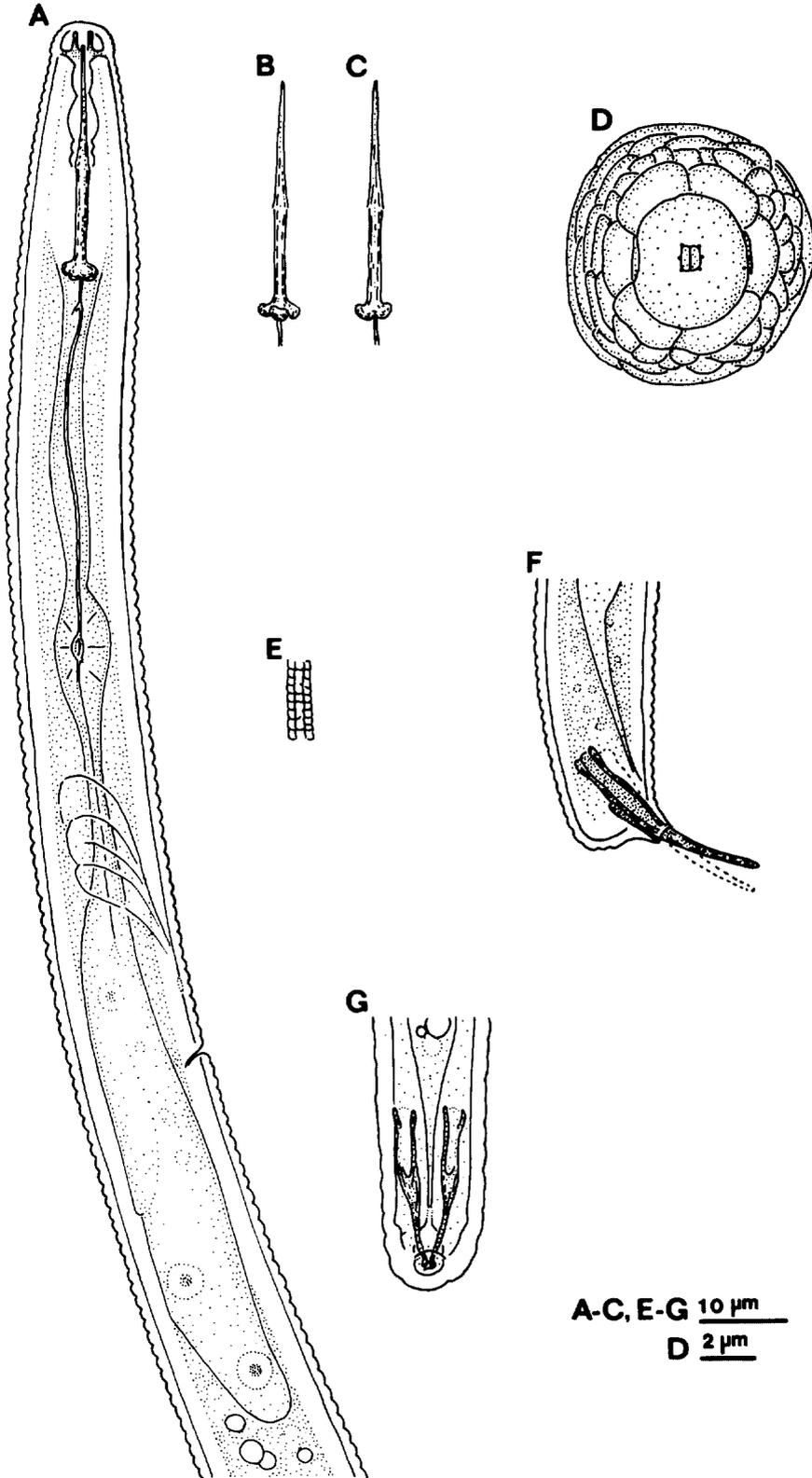


Fig. 2. Drawings of males of *Cryphodera utahensis* n. sp. A) Anterior portion (lateral). B) Stylet (dorsal). C) Stylet (ventral). D) Face view as observed with SEM. E) Lateral field. F) Tail (lateral). G) Tail (ventral).

tinct esophago-intestinal junction. Testis highly variable in length, filled with large irregularly shaped sperm. Tail very short, terminus angular in lateral view with spicule sheath protruding about 6 μm from main body contour; phasmids indistinct (Fig. 2F-G). One very small male (L = .58 mm) encountered.

Juveniles (measurements of 20 paratypes shown in Table 1).

Description (juveniles) (Figs. 3, 6): Body cylindrical, slightly tapered anteriorly, sharply tapered posteriorly (Fig. 3A). Lip region slightly offset with five annules; no longitudinal striae. Amphid opening pore-like (Figs. 3H, 6). Lateral field with four incisures, little or no areolation (Fig. 3F). Cephalids sometimes visible at third and

ninth annule; hemizonid about two annules long; caudalid about 1½ annules long and about four annules anterior to anus. Stylet knobs with slight anterior indentation (Fig. 3B-D). Median bulb proportionately broader and less elongate than in male. Gland lobe with three glands relatively distinct; large conspicuous subventral gland nuclei, less conspicuous dorsal gland nucleus, distinct esophago-intestinal junction. Phasmid about one annule width in diameter, with broader spherical ("lens-like") structure beneath surface of cuticle; tail tapering (Fig. 3E-G).

Diagnosis: Cryphodera utahensis n. sp. most closely resembles *C. podocarpi* but is distinct on the basis of several characters. The esophagus of juveniles of *C. utahensis*

Table 1. Measurements of second-stage juveniles of *Cryphodera utahensis* n. sp. (n = 20).

Character	Range	Confidence interval			Standard deviation
		Mean	95%	99%	
Linear (μm)					
Body length	508.0-596.8	5.33.89 \pm	10.27 \pm	14.04	21.947
Body width	17.8- 24.0	20.78 \pm	.62 \pm	.84	1.325
Lip region (height)	4.4- 6.0	5.19 \pm	.18 \pm	.24	.391
Lip region (width)	10.0- 11.5	10.64 \pm	.26 \pm	.36	.559
Stylet length	35.2- 41.0	37.63 \pm	.69 \pm	.94	1.469
Stylet knobs (height)	2.8- 4.0	3.35 \pm	.17 \pm	.23	.335
Stylet knobs (width)	6.4- 7.6	7.16 \pm	.15 \pm	.21	.325
Stylet shaft and knobs	17.5- 20.5	18.86 \pm	.41 \pm	.57	.883
DGO	4.0- 7.0	5.45 \pm	.33 \pm	.46	.712
Medium bulb (length)	14.0- 19.0	15.94 \pm	.82 \pm	1.12	1.758
Medium bulb (width)	8.9- 13.0	11.90 \pm	.43 \pm	.58	.909
Esophagus length	136.9-241.0	206.08 \pm	12.48 \pm	17.06	26.669
Excretory pore to					
anterior end	108.0-134.4	120.62 \pm	3.42 \pm	4.67	7.305
Hemizonid to anterior end	103.0-126.5	119.23 \pm	2.99 \pm	4.09	6.392
Annule width	1.5- 1.9	1.78 \pm	.07 \pm	.09	.141
Tail length	40.0- 51.5	46.86 \pm	1.46 \pm	1.99	3.120
Tail terminus length (hyaline region)	22.2- 30.5	27.54 \pm	1.16 \pm	1.59	2.485
Phasmid to tail tip	38.5- 47.5	41.99 \pm	1.32 \pm	1.81	2.740
Genital primordium to tail tip	143.5-226.0	189.10 \pm	9.02 \pm	12.39	18.144
Ratios					
a	21.3- 33.5	25.83 \pm	1.12 \pm	1.53	2.391
b	2.1- 3.9	2.64 \pm	.19 \pm	.26	.404
b'	1.9- 3.0	2.19 \pm	.13 \pm	.17	.268
c	10.1- 13.0	11.35 \pm	.36 \pm	.49	.764
c'	2.7- 3.6	3.14 \pm	.10 \pm	.14	.222
Percentages					
m	44-53	50.2 \pm	1.18 \pm	1.62	2.530
o	14-18	14.5 \pm	.95 \pm	1.29	2.030
Excretory pore	20-25	22.6 \pm	.74 \pm	1.01	1.580
Genital primordium	28-41	35.1 \pm	1.52 \pm	2.08	3.250

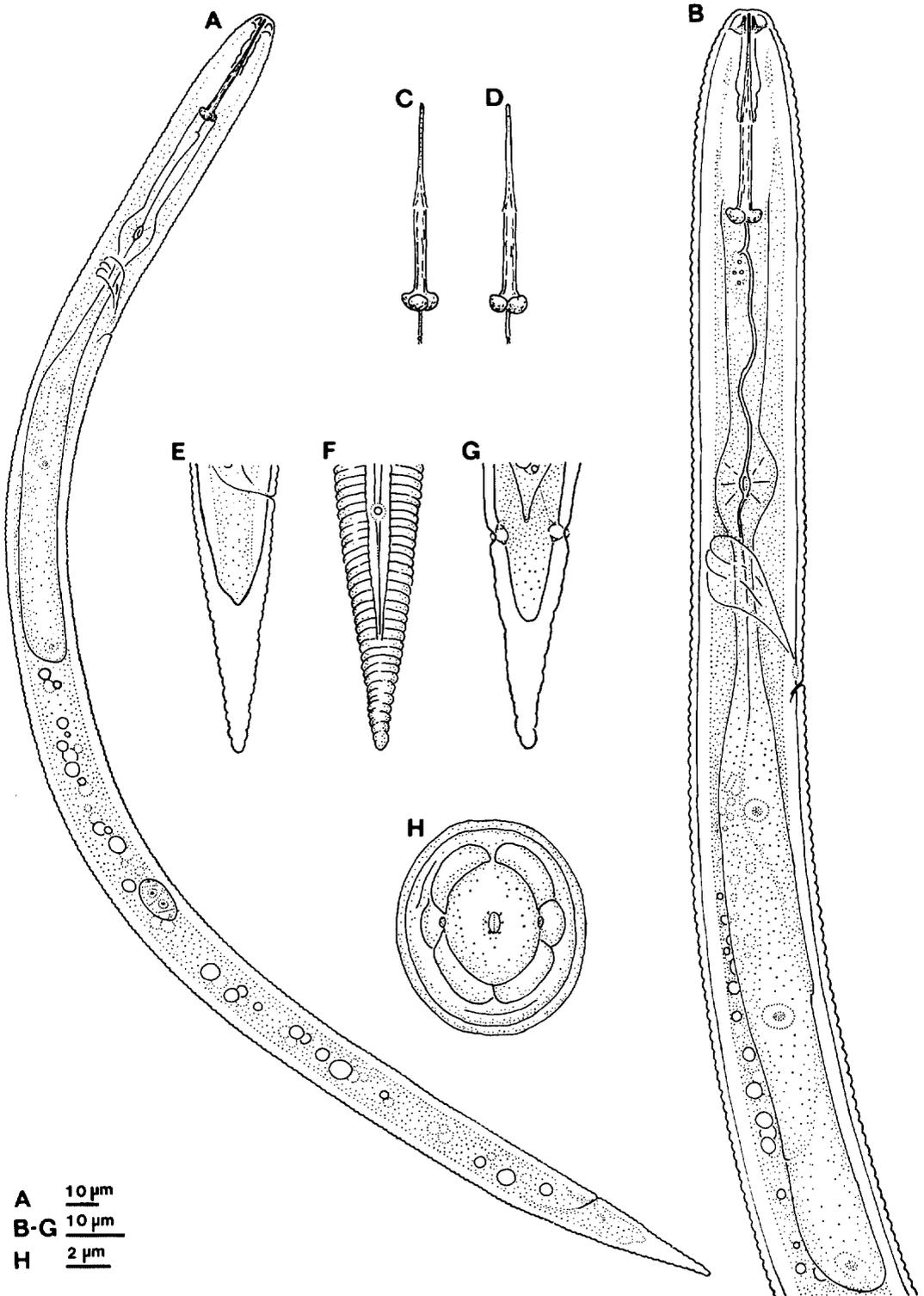


Fig. 3. Drawings of juveniles of *Cryphodera utahensis* n. sp. A) Entire specimen (lateral). B) Anterior portion (lateral). C) Stylet (dorsal). D) Stylet (ventral). E) Tail (lateral). F) Tail surface with lateral field (lateral). G) Tail (ventral). H) Face view as observed with SEM.

is much longer (206.1 versus 142.0 μm) consequently *b* is less (2.6 versus 3.8). Juveniles of *C. utahensis* have a shorter tail (46.9 versus 68.9 μm). In addition, the lateral field of juveniles of *C. utahensis* has four incisures whereas *C. podocarp*i has three. Males of *C. utahensis* tend to be more slender than those of *C. podocarp*i so that the *a* ratio is greater (45.3 versus 35.7); the stylet tends to be shorter in *C. utahensis* (33.3 versus 37.0 μm). The overall shape of females of *C. utahensis* is distinct from *C. podocarp*i and other *Cryphodera* spp. as a result of the more pronounced terminal protuberance.

Holotype (female): Collected by A. H. Bell and M. Mundo-O. on 4 May 1981. Catalog number 52, UCRNC, Department of Nematology, University of California, Riverside.

Paratypes (57 females, 9 males, 88 juveniles): Same data as holotype. Specimens distributed in type collections as follows: 6 females, 1 male, 6 juveniles, USDNC, Division of Nematology, University of California, Davis; 6 females, 1 male, 6 juveniles, USDA Nematology Investigations, Beltsville, Maryland; 6 females, 6 juveniles, Rothamsted Experimental Station, Harpenden, England; 39 females, 7 males, 70 juveniles, UCRNC, Department of Nematology, University of California, Riverside.

Type habitat and locality: Roots and surrounding soil of wild rose (*Rosa* sp. L.; blooms were not available for specific identification), Clear Creek Canyon, Sevier County, Utah. Additional specimens have been collected from wild rose at Gates Creek, Utah.

Detailed surface morphology: The face view of *C. utahensis* n. sp., as observed with SEM, is strikingly different between males and juveniles. The lip region of the male includes a broadly oval labial disc, with a large prestoma region surrounded by orifices of six labial sensillae (Figs. 2D, 4, 5).

Slit-like openings of amphids occur at each lateral margin. The labial disc is surrounded by six sectors, four submedial and two lateral, of nearly equal size. The remainder of lip region consists of a pattern of highly irregularly shaped plates (i.e., regions defined by superficial cuticular incisures) which include longitudinal and transverse striae (Figs. 2D, 4, 5).

Specimens of *C. eucalypti*, the type species, were available for comparison with *C. utahensis* n. sp. but had been prepared for light microscopy in 1966 and were of marginal quality for SEM. However, SEM examination of several views of a few specimens made it possible to prepare a composite illustration of the head regions (Fig. 10). The lip region of males is characterized by a labial disc surrounded by six separate lip sectors, similar to *C. utahensis* n. sp. However, the remainder of the lip region, unlike *C. utahensis* n. sp., consists of a single transverse row of about 12 rectangular plates (Fig. 10A–B). The plates are not necessarily symmetrically arranged, and the numbers of plates on the dorsal side may vary relative to the ventral side.

Light microscope observations of paratypes of *C. podocarp*i, *C. nothophagi*, and *C. coxi* confirm the presence of longitudinal striae on the lip region of males of each and suggest a similar face pattern among members of the genus. The pattern for *C. coxi* and *C. nothophagi* appears to include a single row of rectangular plates as in *C. eucalypti*, whereas *C. podocarp*i has a pattern of irregularly shaped plates similar to *C. utahensis* n. sp.

The lip region of juveniles of *C. utahensis* n. sp. is characterized by an oval to broadly spindle-shaped labial disc, with the longer axis being dorso-ventral (Figs. 3H, 6). The prestoma is also oval with six orifices of labial sensillae. Two pore-like openings of amphids occur at the margin of each lateral side of the disc. The labial



Fig. 4. SEM face view of male of *Cryphodera utahensis* n. sp. $\times 7,400$.

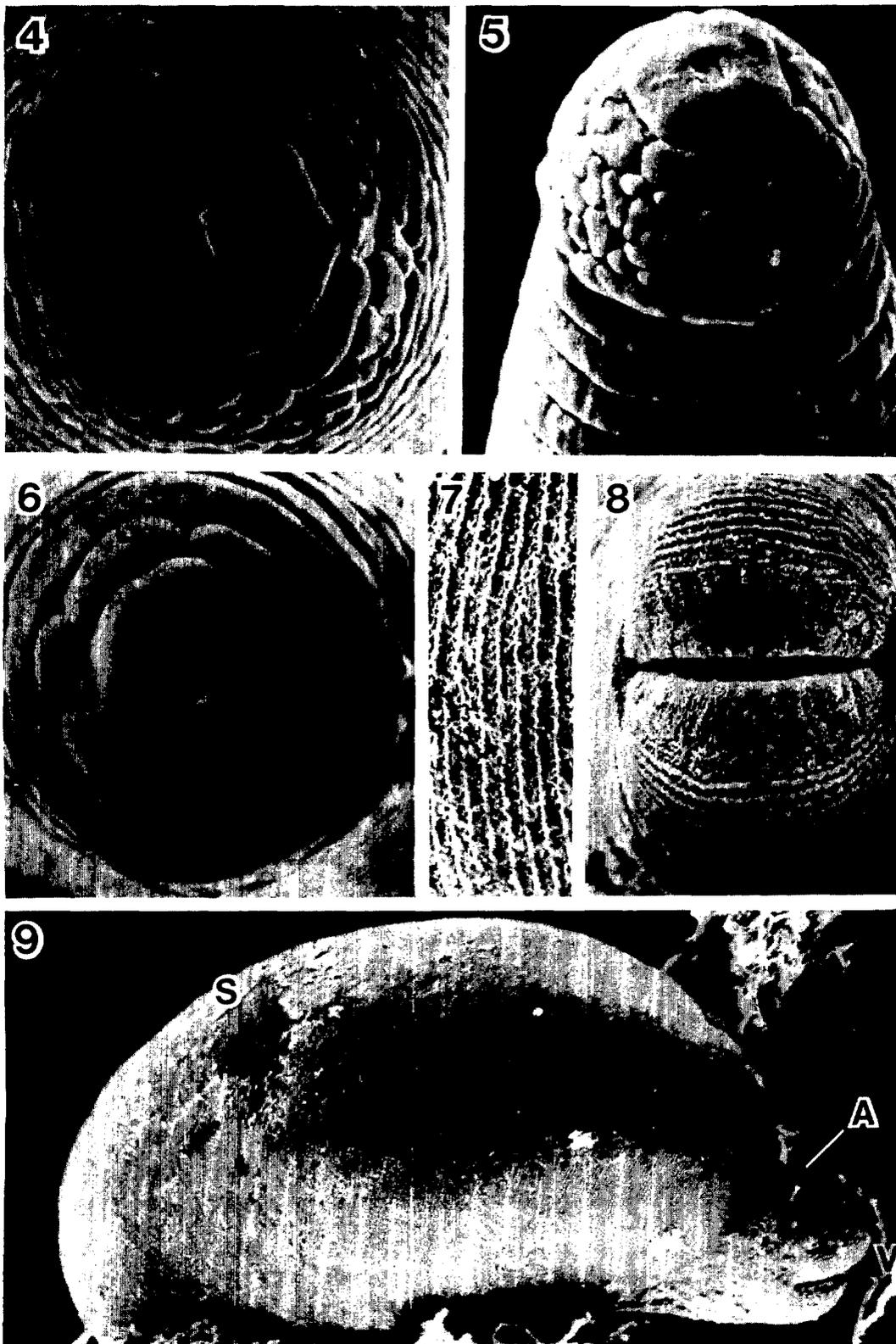
Fig. 5. SEM submedial view of lip region of *Cryphodera utahensis* n. sp. showing pattern of irregularly shaped plates. $\times 6,000$.

Fig. 6. SEM face view of juvenile of *Cryphodera utahensis* n. sp. $\times 7,400$.

Fig. 7. SEM of body striae of female of *Cryphodera utahensis* n. sp. $\times 1,650$.

Fig. 8. SEM of terminal protuberance of female of *Cryphodera utahensis* n. sp. $\times 750$.

Fig. 9. SEM of entire female of *Cryphodera utahensis* n. sp. with head embedded in root of rose. $\times 226$, A = anus; S = portion of subcrystalline layer; V = vulva.



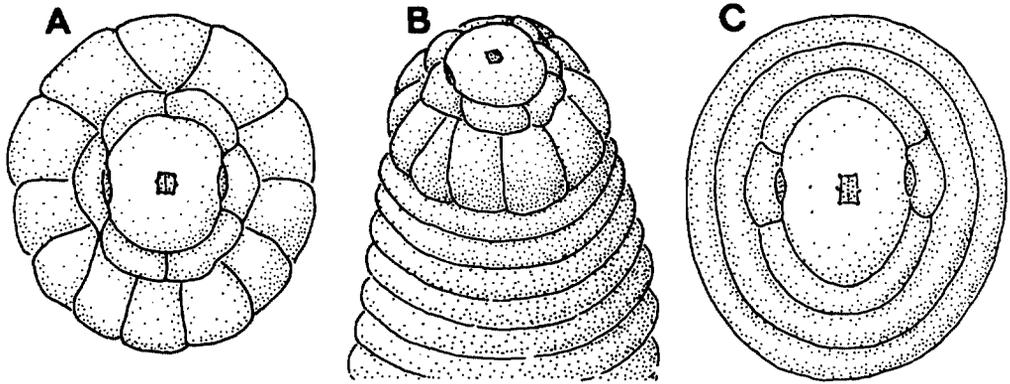


Fig. 10. Drawings of lip region of *Cryphodera eucalypti* Colbran, 1966 as observed with SEM. $\times 4,600$. A) Face view of male. B) Submedial view of male showing rectangular plates. C) Face view of juvenile.

disc is surrounded by six lip sectors, including four elongate submedial sectors and two shorter but slightly broader lateral sectors. Adjacent submedial sectors may be in direct contact with one another or may be slightly separated. Some transverse striae may be incomplete, and no longitudinal striae occur on the lip region (Figs. 3H, 6). The lip region of juveniles of *C. eucalypti* resembles that of *C. utahensis* n. sp., except the four submedial lip sectors are fused to form a pair of medial lips and the openings are slit-like (Fig. 10C).

The lip region of females of *C. utahensis* is smooth with a squarish labial disc characterized by four submedial lobes. Openings of six labial sensillae occur at the margin of the prestoma, and a small slit-like amphid opening occurs at each lateral margin of the labial disc (Fig. 1B). Striations basically extend over the entire body, although they become more pronounced on the posterior protuberance and are modified to a rugose pattern in the perivulval region (Figs. 7, 8, 9). Processing specimens for SEM generally removes all but minute portions of the subcrystalline material (Fig. 9).

DISCUSSION

Discovery of *C. utahensis* n. sp. confirms a broader distribution of *Cryphodera* than indicated by descriptions of other species of the genus, which are limited to Australia and New Zealand. In addition, Wouts (6) has noted evidence that species occur in Japan and the Soviet Union. Worldwide distribution of *Cryphodera* supports the

suggestion that the ancestor which gave rise to *Cryphodera* was in Pangaea (3).

Within the United States we have found *Cryphodera* only in Utah, although Wouts and Sher (7) report that a sample with an undescribed species of *Cryphodera* was intercepted from Oregon. Within Utah, *C. utahensis* n.sp. was collected at two similar localities. Gates Creek and Clear Creek Canyon are about 50 miles apart; both appear to be virgin areas with a cool climate at an elevation of about 1500 M. In each case, specimens were collected from wild rose on the banks of streams.

Cryphodera is frequently classified with *Meloidodera* Chitwood et al., 1959 in a common subfamily *Meloidoderinae* Golden, 1971 on the basis of a number of similarities including striated cuticle in females, short spicules ($< 30 \mu\text{m}$), wide separation between vulva and anus, absence of a cyst, and the lens-like plasmid in juveniles (excluding *M. charis* and *M. belli*, see Wouts [6]) (3). However, a cladistic analysis of Heteroderidae (3) indicates that since no shared derived characteristics have been proposed between *Meloidodera* and *Cryphodera*, there is no basis for suggesting a unique shared common ancestor. This observation is probably best reflected in a taxonomic system which does not place the two genera in Meloidoderinae. Luc et al. (3) rejected Meloidoderinae and other subfamilies of Heteroderidae, considering current data inadequate to elucidate relationships among genera. Examination of new characters will be useful for clarification

of the phylogenetic relationship between *Meloidodera* and *Cryphodera*.

The longitudinal striae on the lip region of males of *C. utahensis* apparently occur throughout the genus as well as on *M. floridensis*, *M. charis*, and *M. belli* (Baldwin and Bell, unpublished observations). Wouts and Sher (7) noted that the large phasmids and longitudinal lip striations of certain "primitive" Heteroderidae resemble similar characteristics in Hoplolaimidae. Comparative detailed studies of the phasmid and lip region may contribute to knowledge on the distribution of specific character states and establishment of homologies. Such information will better indicate if polarity (primitive versus derived) of large (versus pore-like) phasmids and longitudinal lip striations (versus smooth lips or transverse striae, alone) can be established on the basis of out-group comparisons with Hoplolaimidae for phylogenetic analysis.

Cryphodera sp. and *Meloidodera* spp. also resemble one another in that both induce a single uninucleate giant cell in their host, in contrast to the cluster of syncytia associated with *Heterodera sensu lato* and certain other Heteroderidae (Mundo and Baldwin, unpublished observations). This

similarity between the two genera may reflect more fundamental shared characteristics in digestive enzymes of the nematodes, which induce such specific host responses.

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