Heterodera achilleae n. sp. (Nematoda: Heteroderidae) from Yarrow in Yugoslavia[1]

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Abstract: Heterodera achilleae n. sp., a member of the *H. rostochiensis* group, is described and illustrated from roots of yarrow, *Achillea millefolium* L. in Sarajevo, Yugoslavia. This new, round-cyst species differs from closely related species especially as follows: (1) from *H. leptonepia*, by having stouter larvae (a = 21), with longer stylet (25 μ), and with outlet of dorsal esophageal gland averaging 5.7 μ from base of stylet; (2) from *H. millefolii*, in having excretory pore at base of neck and small, straight vulval slit of 5 μ; (3) from *H. rostochiensis*, in having a B/A ratio (Granek's ratio) of 1.6; (4) from *H. tabacum*, by longer female stylet, two annules on female head, and males with outlet of dorsal esophageal gland further back (5.7 μ). In addition, *H. achilleae* n. sp. differs from the latter three species in having prominent longitudinal striae on the anterior half, or more, of cysts and females. *Key Words:* taxonomy, morphology, new *Heterodera* species, *Achillea millefolium*.

In the fall of 1970, *Heterodera* cysts of the *H. rostochiensis* group from a potato-growing area in Yugoslavia, were sent to the senior author for identification. Examination of this and subsequent material at Beltsville, Maryland, USA, and concurrent morphological studies by the junior author at Sarajevo, Bosnia, Yugoslavia, revealed that this nematode was a new *Heterodera* species. Host tests at Sarajevo showed that yarrow, *Achillea millefolium* L., was heavily attacked, but not potato.

Specimens for the description presented below were collected from yarrow grown in a greenhouse at Sarajevo, Yugoslavia. Procedures used in describing this new species, including measuring, drawing, photomicrographing and preparing specimens, were the same as those used by Golden and Birchfield (1).

Heterodera achilleae n. sp.

FEMALES (25): Length (including neck) 564 μ (434-938); width 321 μ (230-454); L/W ratio = 1.8(1.2 - 2.9); neck length 126 μ (83-188); L/N (length/neck) ratio = 4.6 (3.1 - 6.5); stylet 26 μ (23-28); outlet of dorsal esophageal gland from base of stylet 5.6 μ (3.4 - 6.1).

Holotype (female): Length (including neck) 520 μ; width 326 μ; L/W ratio = 1.6; stylet 26 μ; outlet of dorsal esophageal gland 5.6 μ.

Description: Body pearly white, ovate to subspherical in shape, with elongate, protruding neck, and rounded posteriorly (Fig. 2). Cuticle thick, outer layer rugose in posterior portion, then forming lines extending anteriorly onto neck (Fig. 10). Punctations near or just beneath the surface, appearing often in latitudinal lines as in cysts (Fig. 9). Head slightly set off, variable in shape and bearing two annules (Fig. 1C). Cephalic framework distinct but weakly developed. Stylet strong, slightly curving dorsally and with well-developed knobs sloping posteriorly. Cephalids (anterior and posterior) and esophageal region appearing about as illustrated, with excretory pore at base of neck and 126 μ (112-154) from anterior end (Fig. 1C). Vulva often shaped as illustrated (Fig. 11), measuring 15.8 μ (11.2 - 17.9) on an axis at 90 degrees to vulval slit and 15.9 μ (11.2 - 18.5) on axis with vulval slit, which is only 5 μ (2.8 - 5.6) in length. Vulval bodies (Fig. 13) deep underneath vulva, clustered, and variable in shape, appearing essentially as described by Golden and Ellington (2) in females of *H. rostochiensis* Wollenweber, 1923. Anus small but distinct, located 29 μ (20-39) from nearest edge of vulva and opposite the long axis of vulval slit.

CYSTS (25): Length (including neck) 639 μ (459-852); width 479 μ (316-602); L/W ratio = 1.3 (1.2 - 1.5); neck length 101 μ (72-175); L/N (length/neck length ratio) = 6.6 (4.0 - 9.3); diameter of fenestra (A) 16 μ (12-18); distance from anus to nearest edge of fenestra (B) 27 μ (22-34); B/A ratio (Granek's ratio) = 1.6 (1.3 - 1.9).

Description: Cysts light to dark brown in

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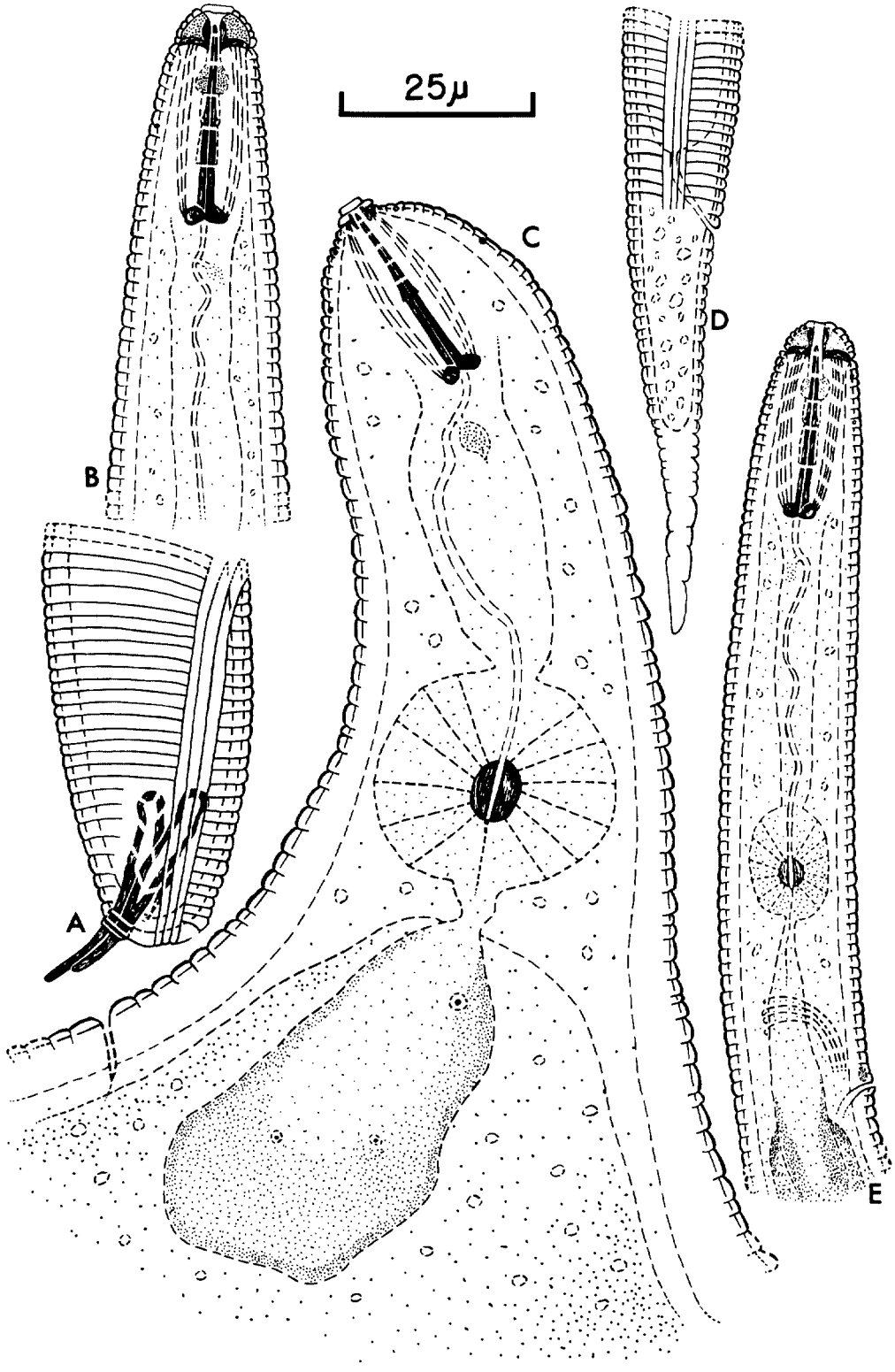

FIG. 1. Drawings of *Heterodera achilleae* n. sp. Male: A = Posterior; B = Anterior. Female: C = Anterior. Second-stage larva: D = Posterior; E = Anterior.

color, ovate to spherical in shape, with protruding neck (Fig. 3); circumfenestrate, abullate, and without distinct vulval bodies as generally seen in females. Excretory pore at or

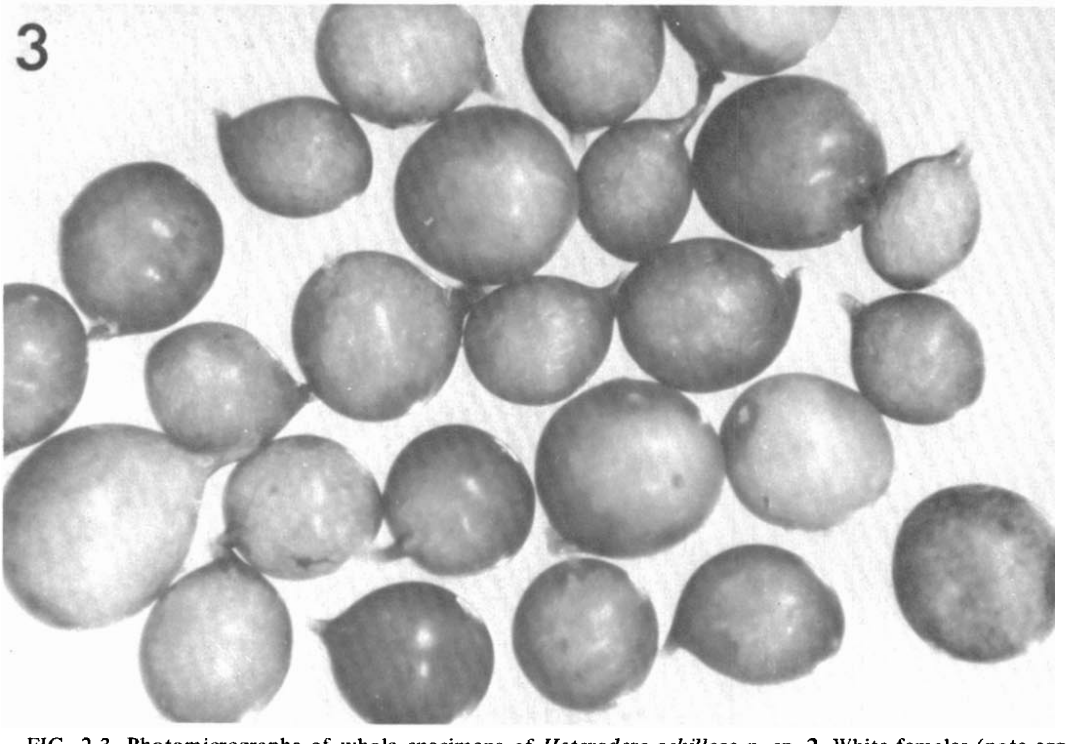

FIG. 2-3. Photomicrographs of whole specimens of *Heterodera achilleae* n. sp. **2.** White females (note egg mass attached to one specimen). **3.** Cysts × 30.

near base of neck, which measures 101 μ (72-175), giving a length/neck ratio of 6.6 (4.0 - 9.3). Fenestra much larger than the small but distinct anus (Fig. 4). In posterior portion, outer layer of cyst wall is essentially rugose and tending to form a consistent type of pattern at anus and fenestra (Figs. 4, 5, 6, 7). At a distance of about 50-100 μ from the fenestra, the cyst pattern changes from a rugose latitudinal type to a longitudinal wavy-line type, the zone of change from one type to another being very distinct (Fig. 8). The prominent wavy lines extend to and onto the neck. Subcuticular punctation (Fig. 9) generally distinct, and arranged in close, parallel rows at right angle to long axis of cyst.

MALES (25): Length 1145 μ (1048-1219); a = 31 (28-34); b = 5.9 (4.8 - 7.0); c = 895 (0-2176); stylet 28 μ (27-29); outlet of dorsal esophageal gland from base of stylet 5.7 μ (5.6 - 6.2); spicules 33 μ (30-37); gubernaculum 11.8 μ (11.2 - 12.9).

Allotype (male): Length 1150 μ; a = 30; b = 6; c = 950; stylet 28.4 μ; outlet of dorsal esophageal gland 5.8 μ; spicules 32 μ; 1 testis.

Description: Body slender, vermiform, tapering slightly at both extremities. Cuticle with prominent annulation; subcuticular annulation less distinct and twice as numerous. Lateral field without areolation, consisting of four lines except fewer at its beginning anteriorly. About midway, body measures 37 μ (34-39) in width. Head slightly offset, bearing five annules and measuring about 10 μ (9-11) at its base and approximately 5 μ (3-5) in height, giving a width/height ratio of 2.2 (2.1 - 2.9). Cephalic framework heavily sclerotized. Stylet, knobs and the very small cephalids (anterior and posterior) commonly appearing as illustrated (Fig. 1B). Median bulb ellipsoidal, with its center 98 μ (80-108) from the anterior end. Excretory pore about two annules posterior to the distinct hemizonid. Testis one. Spicules slightly arcuate, with tips rounded, unnotched. Tail shaped about as illustrated (Fig. 1A), very short and with cloaca often extending beyond terminus.

SECOND-STAGE LARVAE (25): Length 492 μ (472-515); a = 21(18-22); b = 3.0(2.5 - 3.7); c = 9.0(7.9 - 10.4); stylet 25 μ (24-25); outlet of dorsal esophageal gland from base of stylet 5.7 μ (5.6 - 6.1); tail 55 μ (47-63); hyaline tail terminal 27 μ (20-32); caudal ratio A = 3.9(3.4 - 4.8); caudal ratio B = 17(12-29).

Description: Body vermiform, tapering at both extremities, but much more so posteriorly. Subcuticular annulation less distinct but twice as frequent as on cuticle. Lateral field without areolation, composed of four lines. Widest part of body measures 24 μ (21-28). Head slightly set off, with five annules, and its base measuring 9.0 μ (8.4 - 9.5), its height 3.4 μ (3.3 - 3.9), giving a width/height ratio of 2.6(2.4 - 2.8). Stylet, stylet knobs, cephalids and esophageal region commonly appearing as illustrated (Fig. 1E). Valvated center of prominent median bulb 73 μ (68-77) from anterior end. Excretory pore posterior and adjacent to hemizonid. Tail long, becoming slightly reduced in size on ventral side immediately past anus, and then tapering to a finely rounded terminus (Fig. 1D). Rectum unusually large and prominent. Phasmids very small, located just anterior to midportion of tail.

EGGS (25): Length 110 μ (102-119); width 50 μ (45-56); L/W ratio = 2.2(1.9 - 2.5). Egg shell hyaline, without visible markings.

HOLOTYPE (female): Collected from greenhouse culture by Dr. Olga Klindić at Sarajevo, Bosnia, Yugoslavia, 15 April 1971. Slide T-214t, United States Department of Agriculture Nematode Collection, Beltsville, Maryland, USA.

ALLOTYPE (male): Slide T-215t, same data and collection as holotype.

PARATYPES: Males, females, cysts, larvae and eggs: United States Department of Agriculture Nematode Collection, Beltsville, Maryland; Nematode Collection, Institute for Agricultural Research, Sarajevo, Bosnia, Yugoslavia; and California Nematode Survey Collection, Davis, California, USA.

TYPE HOST AND LOCALITY: Roots of yarrow, *Achillea millefolium*, near Fojnica, Republic of Bosnia, Yugoslavia.

DIAGNOSIS: *Heterodera achilleae* n. sp., as described above, differs from closely related species especially as follows: (1) Larvae of *H. leptonepia* Cobb and Taylor, 1953 are extremely slender (a = 39), have shorter stylet (18 μ) and outlet of dorsal esophageal gland about 12 μ from base of stylet; (2) female of *H. millefolii* Kirjanova and Krall, 1965 with excretory pore at midneck and vulval slit bow-shaped, about 35 μ in length; (3) cysts of *H. rostochiensis* Wollenweber, 1923 with a B/A ratio (Granek's ratio) averaging 4.5 and without prominent longitudinal striae; (4) in *H.*

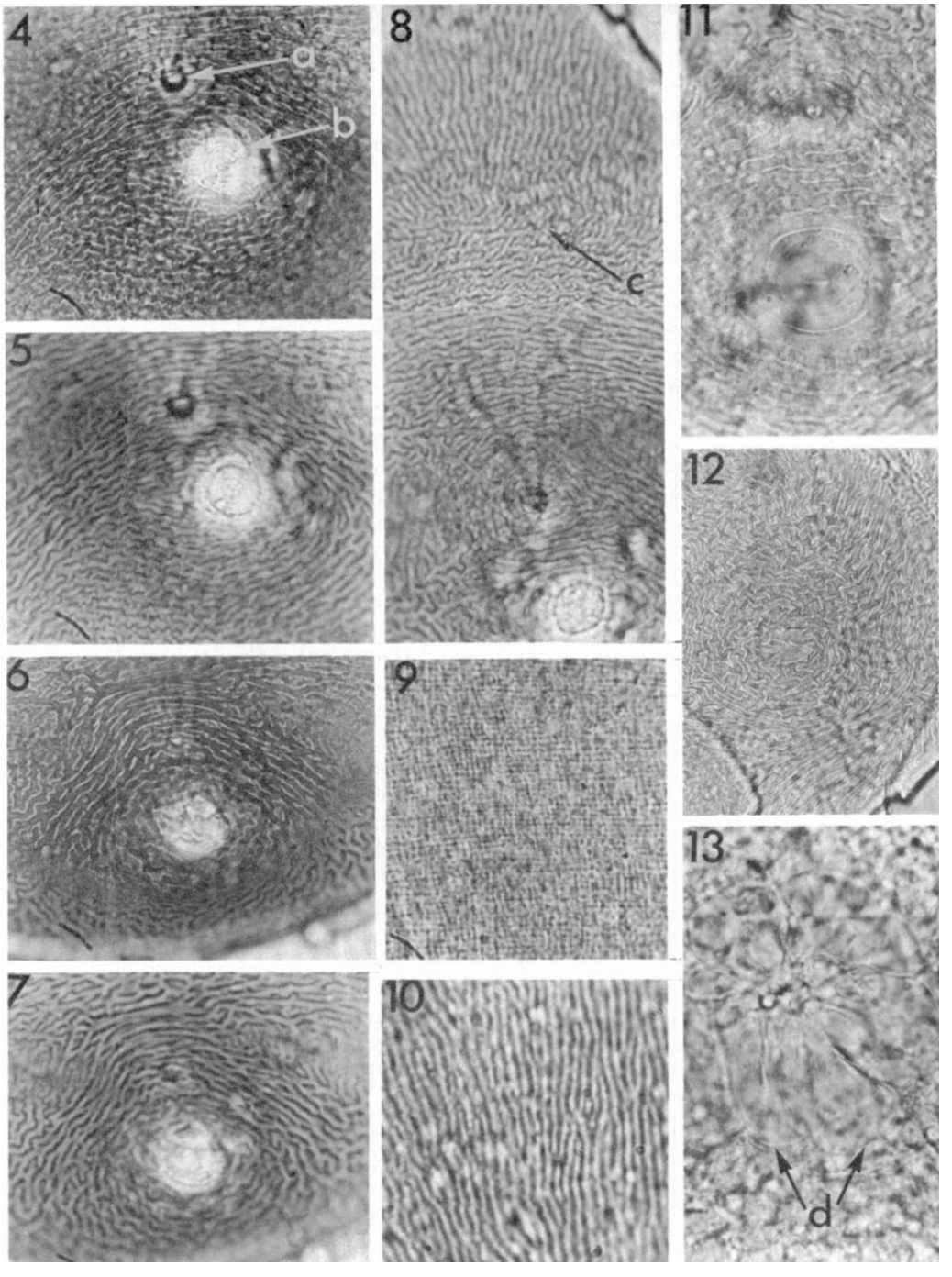

FIG. 4-13. Photomicrographs of portions of cysts (4-9) and females (10-13) of *Heterodera achilleae* n. sp.: **4** and **6** - surface views of two different cysts at posterior portion showing anus (a), fenestra (b) and fenestral pattern; **5** and **7** - same as 4 and 6, respectively, except at a deeper focus; **8** - cyst surface, showing fenestra at bottom, then the anus, followed by latitudinal rugose striae and then a transitional zone (c) at which the striae become longitudinal and wavy (composite of two photomicrographs); **9** - subcuticular, latitudinal punctation at midbody of cyst; **10** - wavy, longitudinal striae on cuticular surface of female at midbody; **11** - vulval and anal area of female; **12** - surface view of the posterior portion of another female, showing variation in pattern and some of the transitional zone in upper and lower right hand corners; **13** - vulval bodies (d) at deep focus beneath the vulva. Figure **4, 5, 6, 7, 8** and **12** at × 350; **9, 10, 11** and **13** at × 760.

tabacum Lownsbery and Lownsbery, 1954 females with three head annules and stylet averaging 22.4 μ, cysts without prominent longitudinal striae, and males with outlet of dorsal esophageal gland averaging 3.6 μ from base of stylet.

The presence in this species of prominent longitudinal striae on portions of the cysts and an irregular pattern in the fenestral area suggest for the first time close similarities in cyst wall pattern of a *Heterodera* species to *H. leptonepia*. The nature and arrangement of the punctation also indicate such a relationship.

This new species is already known from areas in Bosnia, Croatia and Slovenia, indicating rather widespread distribution in Yugoslavia. Its detailed distribution and other results of extensive research performed in Yugoslavia on hosts and biology will be given in forthcoming publications by the second author.

The common name, yarrow cyst nematode, is proposed for this parasite of *A. millefolium*. This is the second *Heterodera* species known to attack yarrow, the other species being *H. millefolii*, the milfoil cyst nematode, in Russia.

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