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Observations on cucullanid nematodes from freshwater fishes in Mexico, including *Dichelyne mexicanus* sp. n.

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Abstract. A new cucullanid nematode, *Dichelyne mexicanus* sp. n., is described from the intestine of three species of fishes, *Agonostomus monticola* (Bancroft) (Mugilidae, Perciformes) (type host), *Ictalurus balsanus* (Jordan et Snyder) (Ictaluridae, Siluriformes) and *Cichlasoma beani* (Jordan) (Cichlidae, Perciformes), from three rivers (La Maquina River, Veracruz; Chontalcoatlán River, Guerrero and Santiago River, Nayarit) in central Mexico. This species is characterised by the absence of a ventral sucker in the male (subgenus *Dichelyne*) and it differs from its congeners mainly in possessing very unequal and dissimilar spicules (left 0.465-0.768 mm and right 293-548 mm long), an asymmetrical gubernaculum, and two intestinal caeca. Another cucullanid nematode, *Cucullanus caballeroi* Petter, 1977, is reported from *Dormitator maculatus* (Bloch) (Eleotridae, Perciformes) from the La Palma and La Maquina Rivers and Balzapote stream, Veracruz, being briefly described and illustrated; this represents a new host record. Findings of *D. mexicanus* and *C. caballeroi* represent a new record of cucullanid nematodes from fishes in Mexican fresh waters.

The nematode family Cucullanidae Cobbold, 1864, comprising several genera, includes many species parasitising various freshwater, brackish-water and marine fishes worldwide. More rarely they are found in aquatic turtles. Most cucullanids are representatives of the genera *Cucullanus* Müller, 1777 and *Dichelyne* Jägerskiöld, 1902.

In the Americas, most species of *Cucullanus* and *Dichelyne* have been reported from marine fishes, whereas those parasitising freshwater fishes include 16 species of *Cucullanus* (4 in North, 1 in Central and 11 in South America) and 8 of *Dichelyne* (5 in North America and 3 in South America). No cucullanid nematodes have so far been reported from fresh waters in Mexico (Pérez-Ponce de León et al. 1996).

Recent investigations into the helminth parasites of freshwater fishes, carried out by the research teams of the Institute of Biology, National Autonomous University of Mexico and of the Centre for Biological Research, Autonomous University of Morelos State, in several basins of central Mexico revealed the presence of cucullanid nematodes in four species of fish. These proved to belong to an undescribed species of *Dichelyne* and one species of *Cucullanus* known only from the region of Lesser Antilles. The results of this study are presented below.

MATERIALS AND METHODS

The following samples of cucullanid nematodes were examined:

Three males and 6 females of *Cucullanus* from the intestine of *Dormitator maculatus* (Bloch) from the La Palma and La Maquina Rivers and Balzapote stream, Los Tuxtlas, Veracruz, Mexico, collected in November 1996;

Eight males and 2 females of *Dichelyne* from the intestine of *Ictalurus balsanus* (Jordan et Snyder) from the Chontalcoatlán River, Guerrero, Mexico, collected from May 1995 to October 1997;

Two males and 1 female of *Dichelyne* from the intestine of *Agonostomus monticola* (Bancroft) from the La Palma River, Los Tuxtlas, Veracruz, Mexico, collected in November 1996;

One female of *Dichelyne* from the intestine of *Cichlasoma beani* (Jordan) from the Santiago River, Nayarit, Mexico, collected in April 1998.

The nematodes were fixed in hot 4% formaldehyde and cleared with glycerine for examination. Drawings were made with the aid of an Aristoplan microscope drawing attachment. After examination, the specimens were stored in 70% ethanol. All measurements are given in millimetres unless otherwise stated. Type specimens have been deposited in the National Helminthological Collection of the Institute of Biology, National Autonomous University of Mexico (UNAM), in Mexico City and in the Helminthological Collection of the Institute of Parasitology, Academy of Sciences of the Czech Republic (ASCR), in České Budějovice.

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RESULTS

Dichelyne mexicanus sp. n. Figs. 1, 2

Description: Small nematodes with very thick cuticle. Head end rounded. Mouth formed by two lateral valves, each bearing one dorsolateral and one ventrolateral papilla and lateral amphid; mouth aperture slit-like, dorsoventrally elongated, situated transverselly to longitudinal axis of body. Mouth surrounded by narrow cuticular flange armed at its inner base with row of numerous small denticles. Oesophagus expanded at both ends, opening into intestine through small valve; expanded anterior end of oesophagus forming large pseudobuccal capsule supported internally by marked cuticular lining. Nerve ring encircling oesophagus posterior to pseudobuccal capsule, at 1/3-1/2 of oesophagus length from head end. Deirids located near level of nerve ring or somewhat more posteriorly. Excretory pore situated near posterior end of oesophagus or somewhat anterior to it. Intestine provided with two anterior caeca, one dorsal and one ventral, sometimes reaching to nerve ring. Tail of both sexes conical, sharply pointed. Males as large as females.

Male (10 specimens; measurements of holotype in parentheses): Length of body 2.56-5.65 (3.06), maximum width 0.300-0.669 (0.372). Length of entire oesophagus 0.432-0.772 (0.488) (16% of body length). Distance of nerve ring from anterior extremity 0.204-0.320 (0.231), of excretory pore 0.379-0.679 (0.379), of deirids 0.323-0.584 (0.399). Precloacal sucker absent. Of 11 pairs of cloacal papillae, 5 pairs preanal, 2 pairs adanal and 4 pairs postanal. All preanal papillae subventral, first two pairs (counting from cloacal opening) close to each other. Adanal papillae ventral and lateral. First and third postanal papillae subventral, second and fourth lateral. Distinct unpaired median papilla present on anterior cloacal lip. Spicules unequal, length of larger (left) spicule 0.465-0.768 (0.485), that of smaller (right) spicule 0.293-0.548 (0.363). Distal end of left spicule narrow, bluntly pointed, whereas that of right spicule markedly wide, with conspicuous inner bifurcate support. Gubernaculum Y-shaped, length 0.62-0.115 (0.85); its anterior arms distinctly unequal, left arm being markedly longer than right one. Tail conical, 0.115-0.178 (0.132) long, sharply pointed, ending in small cuticular spike.

Female (4 specimens; measurements of allotype in parentheses): Length of body 2.71-4.23 (length of body fragment of allotype 2.67), maximum width 0.306-0.531 (0.531). Length of entire oesophagus 0.508-0.574 (0.508) (10% of body length). Distance of nerve ring from anterior extremity 0.204-0.280 (0.280), of excretory pore 0.346-0.594 (0.346) mm, of deirids 0.346-0.547 (0.544). Vulva postequatorial, 2.10-2.46 (-); vulvar lips slightly elevated. Short vagina directed anteriorly from vulva. Uteri opposed, containing many eggs; anterior ovary far posterior to oesophagus. Mature eggs almost spherical, thin-walled, their content uncleaved or cleaved into two blastomeres. Tail conical, 0.118-0.145 (-) long, sharply pointed, short region anterior to tip covered by numerous minute spines provided with terminal cuticular spike; pair of lateral papillae (phasmids) located approximately at mid-length of tail.

T y p e h o s t : *Agonostomus monticola* (Bancroft) (Mugilidae, Perciformes).

O t h e r h o s t s : *Ictalurus balsanus* (Jordan et Snyder) (Ictaluridae, Siluriformes) and *Cichlasoma beani* (Jordan) (Cichlidae, Perciformes).

S i t e o f i n f e c t i o n : Intestine.

T y p e l o c a l i t y : La Maquina River, Los Tuxtlas, Veracruz, Mexico.

O t h e r l o c a l i t i e s : Chontalcoatlán River, Guerrero, and Santiago River, Nayarit, Mexico.

P r e v a l e n c e a n d i n t e n s i t y : *A. monticola* in La Maquina River: 11% (9 fishes examined / 1 fish infected), 3 nematodes. *I. balsanus* in Chontalcoatlán: 6% (163/9), 1-2 (mean 1) nematodes per host; and *Cichlasoma beani* in Santiago River: 4.2% (24/1), 1 nematode .

E t y m o l o g y : The specific name is derived from the name of the country of its origin.

D e p o s i t i o n o f t y p e s : Type specimens have been deposited in the National Helminthological Collection of the Institute of Biology, UNAM, in Mexico City (Cat. Nos. CNHE-3327-holotype; CNHE-3328-allotype; CNHE-3329 and CNHE-3330-paratypes) and in the Helminthological Collection of the Institute of Parasitology, ASCR, in České Budějovice (Cat. No. N-728-paratypes).

Cucullanus caballeroi Petter, 1977 Fig. 3

Description: Medium sized nematodes. Lateral alae absent. Oral opening dorsoventrally elongated, surrounded by narrow membranous ala (collarette) supported by row of numerous minute teeth. Four submedian cephalic papillae and pair of small lateral amphids present. Pseudobuccal capsule (oesophastome) as wide as posterior part of oesophagus. Deirids and excretory pore situated at level of posterior half of oesophagus. Tail conical, pointed.

Male (3 specimens): Length of body 2.26-2.37, width 0.201-0.224. Entire oesophagus 0.478-0.521 (22% of body length), minimum width 0.046-0.056, maximum width in anterior region 0.135-0.158; oesophastome 0.181-0.221 long and 0.082-0.115 wide. Nerve ring, excretory pore and deirids 0.247-0.257, 0.353-0.419 and 0.3.56-0.491, respectively, from anterior extremity. Length of spicules 0.389-0.567. Gubernaculum well sclerotised, 0.052-0.092 long. Precloacal sucker well developed. Caudal papillae including phasmids: 5 pairs preanal and 6 pairs (3 subventral and 2 lateral) postanal. Tail conical, 0.102-0.141 long with pointed tip.

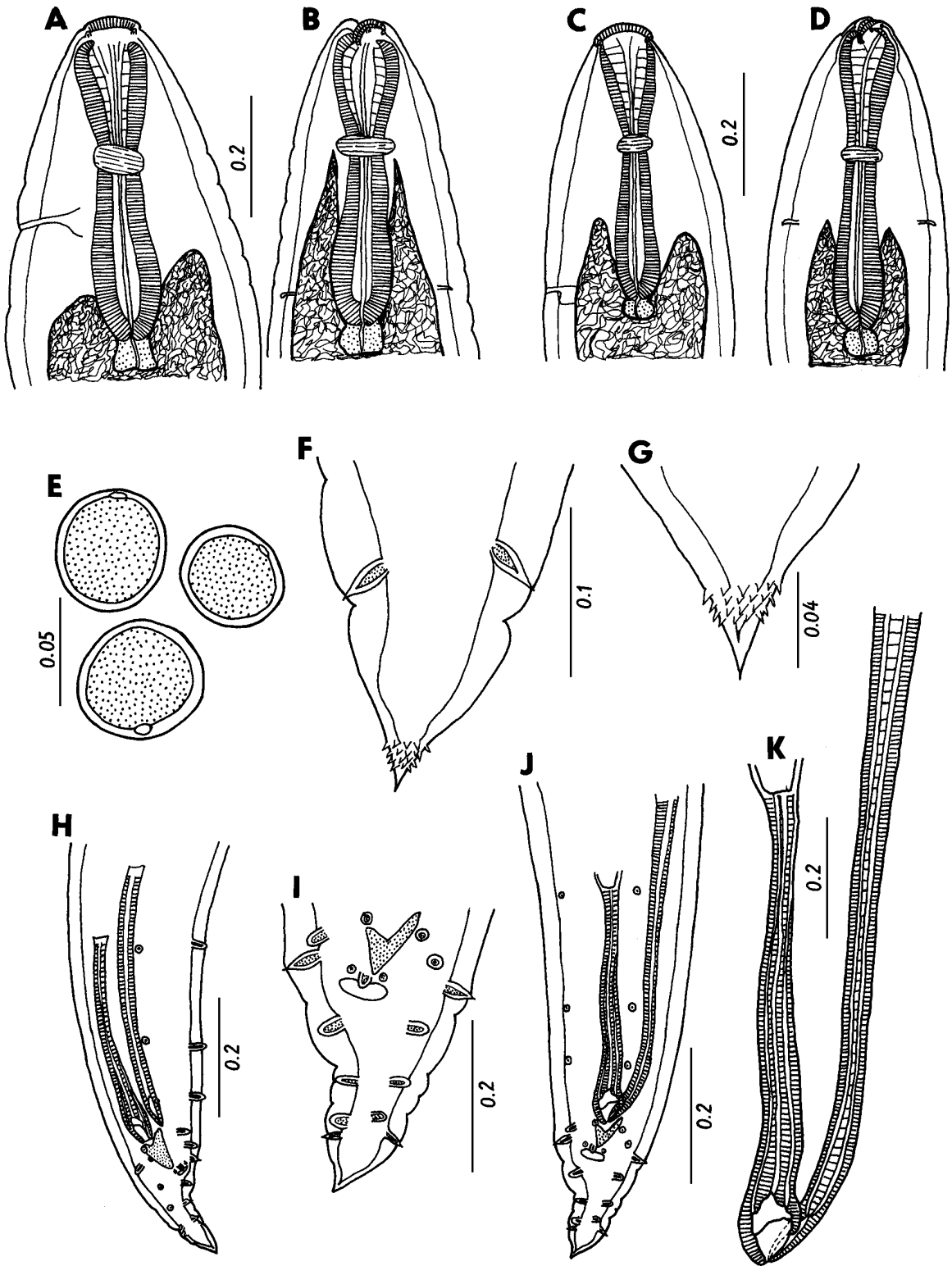

Fig. 1. *Dichelyne mexicanus* sp. n. **A**, **B** – anterior end of female and male from *Agonostomus monticola*; **C**, **D** – anterior end of female and male from *Ictalurus balsanus*; **E** – eggs; **F**, **G** – tail of female, ventral view; **H**, **I**, **J** – posterior end of male, ventral view; **K** – spicules. Scale bars in mm.

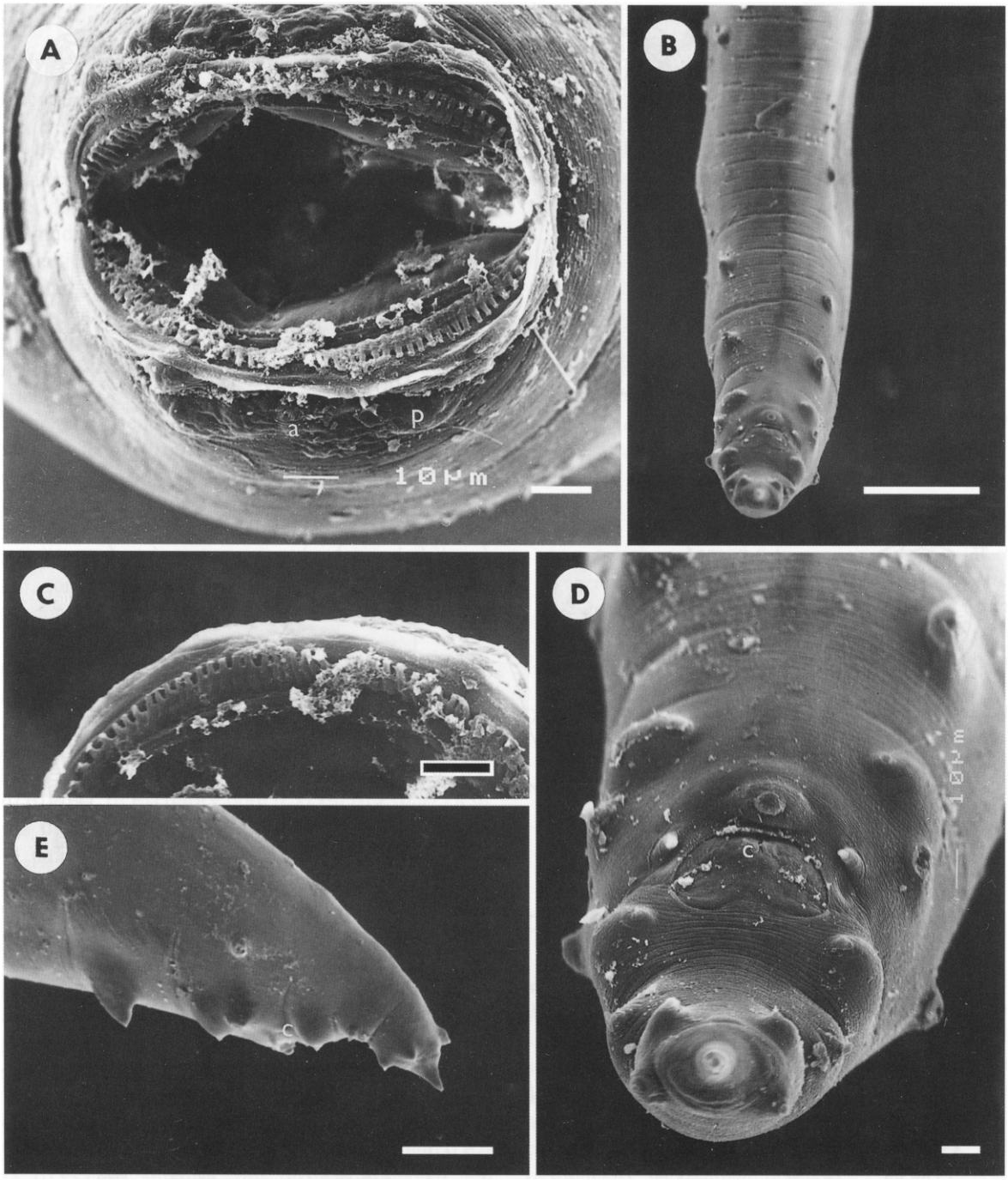

Fig. 2. *Dichelyne mexicanus* sp. n., SEM micrographs. **A** – cephalic end, apical view; **B** – posterior end of male, ventral view; **C** – detail of cephalic teeth; **D** – male tail, ventral view; **E** – posterior end of male, lateral view. Abbreviations: a – amphid; c – cloacal opening; p – cephalic papilla. Scale bars: A, C, D = 10 µm; B = 100 µm; E = 50 µm.

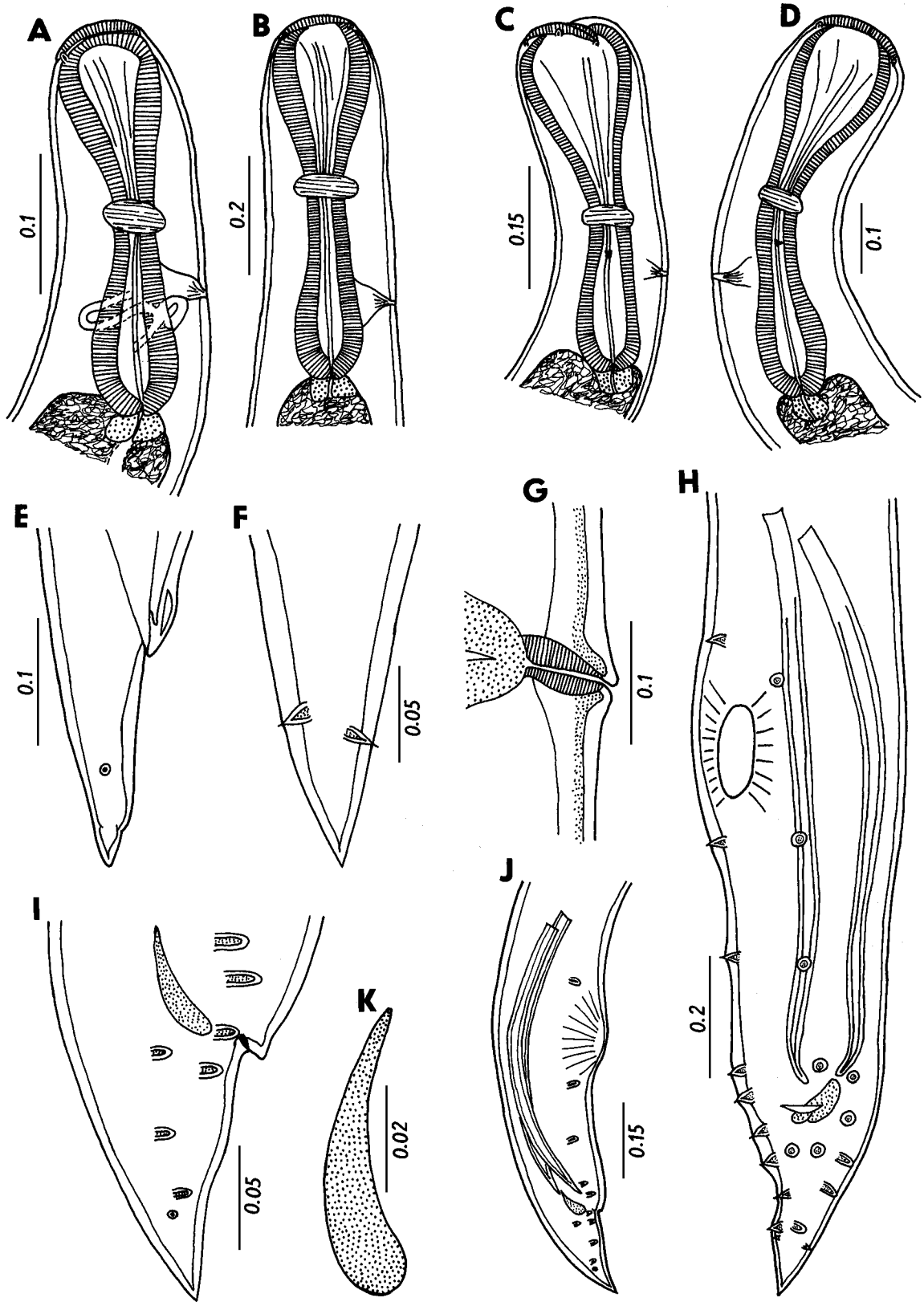

Fig. 3. *Cucullanus caballeroi* Petter, 1977. **A**, **B** – anterior end of female; **C**, **D** – anterior end of male; **E**, **F** – tail of female, lateral and ventral views; **G** – vulva; **H** – posterior end of male with spicules and 10 pairs of papillae, ventral view; **I**, **J** – posterior end of male, lateral view; **K** – gubernaculum, lateral view. Scale bars in mm.

Female (4 specimens): Length of body 2.62-3.40, maximum width 0.234-0.326. Entire oesophagus 0.580-0.686 long (22% of body length); length of oesophastome 0.194-0.204, maximum width 0.099-0.115. Distance of nerve ring, excretory pore and deirids from anterior extremity 0.280-0.287, 0.419-0.495, 0.445-0.603, respectively. Tail conical, 0.131-0.191 long; pair of small lateral papillae (phasmids) present near tail tip. Vulva postequatorial, 1.57-2.10 from anterior extremity. Vulvar lips elevated. Uteri opposed, containing immature eggs.

Type host: *Dormitator maculatus* Bloch (Eleotridae, Perciformes).

Site of infection: Intestine.

Localities: La Palma and La Maquina Rivers and Balzapote stream, Los Tuxtlas, Veracruz, Mexico.

Prevalence and intensity: In La Palma River: 3.2% (31 fishes examined / 1 fish infected), 5 nematodes; in La Maquina River: 22% (9/2), 2 nematodes, and in Balzapote stream: 25% (4/1), 2 nematodes.

DISCUSSION

According to Petter (1974a), the genus *Dichelyne* includes three subgenera: *Dichelyne* Jägerskiöld, 1902 (precloacal sucker absent; 11 pairs of caudal papillae), *Cucullanellus* Törnquist, 1931 (precloacal sucker present; 11 pairs of caudal papillae), and *Neocucullanellus* Yamaguti, 1914 (more than 11 pairs of caudal papillae). It is apparent that *D. mexicanus* sp. n. belongs to the subgenus *Dichelyne.*

At present, eight species of the genus *Dichelyne* have been described from America in freshwater fishes, five in North America: *D. cotylophora* (Ward et Magath, 1917); *D. robustus* (Van Cleave et Mueller, 1932); *D. diplocaecum* Chandler, 1935; *D. lepisosteus* Casto et McDaniel, 1967; *D. bullocki* Stromberg et Crites, 1972), and three in South America (*D. leporini* Petter, 1989; *D. moraveci* Petter, 1995; *D. pimelodi* Moravec et al., 1997). Of these, in contrast to *D. mexicanus*, three species (*D. cotylophora, D. lepisosteus, D. bullocki)* have a ventral sucker in the male and belong, in agreement with Petter´s (1974a) conception, to the subgenus *Cucullanellus.* The remaining species, except for *D. diplocaecum*, are characterised by the presence of only one intestinal caecum and similar, almost equally long spicules. *D. diplocaecum* has two intestinal caeca, resembling thus *D. mexicanus*, and it was also described from fishes of the genus *Ictalurus* Rafinesque (*I. furcatus* [LeSueur]); unfortunately, this species was described by Chandler (1935) from the USA only from two young females and, therefore, should now be considered a *species inquirenda*.

Another North American species described from *Ictalurus* is *D. robustus,* which was inadequately described by Van Cleave and Mueller (1932) from *I. nebulosus* (LeSueur) from the USA. It differs from *D. mexicanus* in having only one caecum and a different arrangement of caudal papillae. Although it was mentioned by Van Cleave and Mueller (1932) that cotypes were deposited in the US National Museum, we were informed by Dr. Hoberg that the type specimens of *D. robustus* are not present in the US National Helminth Collection in Beltsville, Maryland.

Dichelyne species from South American freshwater fishes differ from *D. mexicanus* as follows: all of them have only one intestinal caecum and equal spicules; moreover, *D. moraveci* and *D. pimelodi* have markedly longer spicules (more than 1 mm long), whereas in *D. leporini* the distal ends of spicules are narrow, almost pointed (Petter 1989, Moravec et al. 1993, 1997). None of these species has numerous minute spines near the female tail tip.

Unique features of *D. mexicanus* seem to be the very unequal and dissimilar spicules and an asymmetrical gubernaculum. The right, shorter anterior arm of the gubernaculum in *D. mexicanus* corresponds to the shorter spicule, whose distal end is conspicuously expanded; it may well be that the distal end of this spicule partly substitutes the function of the gubernaculum.

Most *Dichelyne* species possess only one (dorsal) intestinal caecum; the presence of two caeca (one dorsal and one ventral) as in *D. mexicanus,* has been described only for *D. diplocaecum* Chandler, 1935, but two caeca were also illustrated for some others species, e.g. *D. tripapillatus* (Gendre, 1927), *D. exiguus* (Yamaguti, 1954) or *D. indentatus* (Rasheed, 1968) (see Ivashkin and Khromova 1976). The presence of numerous fine caudal spines on the tip of the female tail, characteristic of *D. mexicanus*, has also been described in other species, e.g., *D. rasheedae* Petter, 1974, *D. spinicaudatus* Petter, 1974 and *D. alatae* De et Maity, 1995 (see Petter 1974b, De and Maity 1995, Gibbons and Saayman 1996).

Dichelyne mexicanus has been recorded from three fish host species belonging to different families and two different orders. At present it is difficult to determine its true definitive host, but the finding of the female with eggs in *A. monticola* suggests that probably this fish acts as the definitive host, whereas *I. balsanus* or *C. beani* may serve as paradefinitive or postcyclic hosts (Moravec 1994).

At present 16 species of *Cucullanus* have been reported in America from freshwater fishes: 4 species in North America (*C. truttae* Fabricius, 1794; *C. sphaerocephalus* (Rudolphi, 1809); *C. clitellarius* Ward et Magath, 1917; *C. pybusae* [Ardenson, 1992]), 11 species in South America (*C. pinnai* Travassos, Artigas et Pereira, 1928; *C. zungaro* Vaz et Pereira, 1934; *C. pauliceae* Vaz et Pereira, 1934; *C. mogi* Travassos, 1948; *C. colossomi* Díaz-Ungría, 1968; *C. oswaldocruzi* Santos, Vicente et Jardim, 1979; *C. grandistomis* (Ferraz et Thatcher, 1988); *C. brevispiculus* Moravec,

Kohn et Fernandes, 1993; *C. pimelodellae* Moravec, Kohn et Fernandes, 1993; *C. pseudoplatystomae* Moravec, Kohn et Fernandes, 1993; *C. rhamphichthydis* Moravec, Kohn et Fernandes, 1997), and 1 species from Central America (*C. caballeroi* Petter, 1977).

In all biometrical features the present specimens are very similar to *C. caballeroi*, a species described by Petter (1977) from eleotrid fishes, *Gobiomorus maculatus* and *Eleotris pisonis*, from the rivers of Guadeloupe, Lesser Antilles. There is some difference in the length of spicules (about 0.330 mm according to the illustration vs. 530 mm), but we consider it to be within the intraspecific variability of the species. Petter (1977) reported 11 pairs of caudal papillae, including phasmids and she distinguished *C. caballeroi* mainly by the size of papillae of the last but one preanal pair. Petter (1995) has considered the distribution of male caudal papillae in *Cucullanus* to be stable within species and she used this for the separation of the South American species of *Cucullanus*; however, Moravec et al. (1997) have shown on the example of *C. pinnai* that there exists a considerable intraspecific variability in both the distribution and the size of papillae.

The present finding of *C. caballeroi* in *D. maculatus* represents a new host record and the first record of this nematode species in Mexico.

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