



First Record of Two Parasitic Helminths from *Cobia *Rachycentron canadum* (L.) (Carangiformes, Rachycentridae) from Iraqi Marine Waters*

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Abstract: A total of 14 specimens of *Cobia *Rachycentron canadum* (L.)* were caught from Iraqi marine waters during the period from October 2019 till May 2020. Two parasitic helminths (One species each of nematode and acanthocephalan) were isolated from alimentary canal of the cobia. These are the adults of *Iheringascaris iniquies* (Linton, 1901) Deardorff & Overstreet, 1980 and *Serrasentis sagittifer* Linton, 1889. The record of these worms, from this fish, is considered as the first record in Iraq. Adequate description of both *I. iniquies* and adult *S. sagittifer* from cobia for the first time in the Arabian Gulf is given.

Keywords: Marine fish, Nematoda, *Iheringascaris iniquies*, Acanthocephala, *Serrasentis sagittifer*, Iraq.

Introduction

Cobia, a world-wide species, is distributed in most seas and oceans in open waters and rarely enters the estuaries (Froese & Pauly, 2021), with 200 cm in maximum length; it is a predator on crustaceans and fishes (Carpenter *et al.*, 1997). This fish was also recorded in Iraq (Ali *et al.*, 2018). Cobia is one of the important commercial fishes due to its very good food coefficient ratio (FCR) and high growth rates and currently used in mariculture in 23 countries, mostly in south eastern Asia (Liao *et al.*, 2004; Nhu *et al.*, 2011). Al-Mukhtar (2011) recommended using the cobia in mariculture in Iraq.

For long time, *Iheringascaris* Pereira, 1935 is still monotypic species: *I. iniquies* (Linton, 1901), beside other seven genera including

Hysterothylacium which is placed in family Ascarididae, subfamily Raphidascaridinae, which have excretory pore situated close to nerve ring in addition to the unique shape of ventriculus (Gibbons, 2010). However, Gibbons (2010) and Moravec *et al.* (2012a) placed *Iheringascaris* within the family Anisakidae, considering the above taxonomical characters as not enough for creating Raphiascaridae at the family level. Malhotra *et al.* (2012) and WoRMS (2021) put the *Iheringascaris* within Raphidascaridae.

Serrasentis Van Cleave, 1923 belongs to Rhadinorhynchidae beside 22 other genera (Amin, 2013). The genus *Serrasentis* has five nominal species (Barton *et al.*, 2018; WoRMS, 2021), instead of 22 species listed historically

(Gibson & Wayland, 2021). At Pakistani waters, Bilqees (1971) described *S. longiformis* Bilqees, 1971 (now a *taxon inquirendum*) from *Arius serratus* (= *Netuma thalassina*). Bilqees (1972a) described *S. sciaenus* (now uncertain) from *Sciaenus dussumieri* (= *Johnius dussumieri*) and *S. mujibi* (= *S. nadakali* George & Nadakal, 1978) and from *R. canadus* (= *R. canadum*); while Bilqees (1972b) described *Serrasentis giganticus* Bilqees, 1972 (= *S. sagittifer*) from *R. canadum*. Bilqees & Khan (2005) described *S. manazo* Bilqees & Khan, 2005 from the shark *Myrmilo manazo* (= *Mustelus manazo*) from Pakistan. Khatoon & Bilqees (2007) described cystacanth of *S. niger* Khatoon & Bilqees, 2007 (now a *taxon inquirendum*) from *Parastromateus niger*.

At Kuwaiti waters, Amin *et al.* (1984) isolated *S. sagittifer* cystacanths from five marine fishes, viz: *Lutjanus coccineus*, *L. fulviflamma*, *Nemipteris japonicas*, *N. tolu* and *Otolithes ruber*.

In Iraq, three species belonging to Rhadinorhynchidae were recorded. *Micracanthorhynchina kuwaitensis* (misspelled as *Micracanthorhynchus*) Amin and Sey, 1996, *Slendrorhynchus breviclaviproboscis* Amin & Sey, 1996 reported from *Hemiramphus marginatus* off Khor Abdullah by Bannai (2005) and *S. sagittifer* as a larval stage recorded from *Platax teira* and *Acanthopagrus arabicus* in addition to three unidentified larval stages of *Serrasentis* from *Brachirus orientalis*, *Johnius belangeri* and *Otolithes ruber* (Mhaisen *et al.*, 2018). Bannai (2018) added *Upeneus sulphureus* as a new host in Iraq for *Serrasentis* sp., and he examined the food items of the host which included amphipods and

other crustaceans that may role as intermediate hosts for this parasite.

Due to the absence of any parasitological study on *R. canadum* in Iraq, the current study aims to give the information on some of the parasite fauna of this commercial fish.

Materials & Methods

A total of 14 specimens of *Rachycentron canadum* were examined for endoparasites during May 2019 till October 2020 from marine waters of Iraq

The isolated nematodes were washed and fixed in hot 4% formalin. Two days later, they were transferred to 70% ethanol for preservation. Three different solutions (Glycerin: water, Amman's lactophenol and glacial acetic acid) were used for clearing the nematode according to what the structure of the parasite could be seen (Berland, 1984; Moravec, 1994). In addition, the enface view of head of nematode was mounted with glycerin jelly (Moravec, 1994; Ali, 2008).

The acanthocephalans, after cleaning from the debris, were put overnight in refrigerator for relaxing and protrusion of the proboscis, then, they were put in cold distilled water, fixed and preserved in cold 70% ethanol (Amin *et al.*, 2018), stained in Mayer-Schuberg's Aceto carmine, cleared in methyl salicylate and mounted in Canada balsam (Palm, 2004). All measurements, unless otherwise indicated, were taken in micrometers. Host taxonomy followed Carpenter *et al.* (1997) and verified by Van der Laan *et al.* (2021).

Results & Discussions

Family: Raphidascarididae

Iheringascaris inquires (Linton, 1901)

Site of infection: Stomach and fore intestine.

Prevalence and mean intensity: 28% and 45, respectively.

Deposition: Iraqi Natural History Research Center and Museum, SN: INHM-NC2-NC21.

Description (Figs. 1 and 2)

Whitish, narrow nematode, maximum wide in the middle of the body, highly striated transversely cuticle form serrated appearance (Fig. 1A). The ala is absent, and the presence of longitudinal groove (Fig. 1A). Three lips longer than the wide. Dorsal lip has double papillae, slightly larger than the subventral lips. Each subventral lip has single papilla. Interlabia triangle and well developed, 32-50 in length. Long esophagus with slightly globular ventriculus. Short caecum, narrow ventral appendage and longer than the caecum. The excretory pore slightly posterior to nerve ring (Fig. 1A). Long, equal spicules. Caudal papillae consist of precloacal and postcloacal papillae. Additional row of postcloacal papillae present. Vulva prequatorial, tail conical in both genders and has phasmid in females.

Immature males and females are whitish slender worm with clear striated cuticle. Well-developed lips, interlabia in the beginning of growth, some of which exist in the last molting (from 4th larval stage to adult). The immature males have long spicules inside the body with spiral shape, may be not well sclerotized, caudal papillae not developed yet. The reproductive system of the immature females

not developed, no eggs. The vulva pre-equatorial.

Males (10 specimens), immature males in square brackets [two specimens]

Total length 17.2-28.5 (24.8) [12.8, 18.7] mm and 0.35-0.57 (0.48) [0.18, 0.22] mm in maximum width, ratio of maximum width to maximum length 1: 44-60 (52) [1: 71, 85], the lips 90-110 [80] in length and 90-163 (140) [110-125] in width. Esophagus 2450-1744 (3284) [2100, 3100] in length and 110-135(123) [110, 125] in width, comprising 12-14 (13.2) [16, 17]% from total length. Nerve ring and excretory pore 320-450 (409) [275, 300] and 470-700 (572) [210, 400] from anterior extremity, respectively. Ventriculus 110-220 (155) [80, 110] in length and 80-160 (125) [75, 90] in width. Caecum 150-420 (275) [167, 200] in length and 70-120 (93) [50, 60] in width. Appendage 700-1950 (1285) [525, 740] in length and 27-70 (580) [25, 35] in maximum width. Ratio of appendage to esophagus 1: 1.5-2.8 (2.6) [1:4], ratio of caecum to each of esophagus and appendage 1:7-19.6 [1:13, 16] and 1: 3-8 (5.4) [1:3, 4], respectively. The spicules similar and unequal (Fig. 1E), right spicule 3450-6200 (4829) in length and 30-45 (36) in width, left spicule 3650-6000 (4612) in length and 30-45 (35) in width. Ratio of spicules length 14-22 (19) % of body length. Caudal papillae 22-31 pairs, 14-19 pairs of precloacal and six postcloacal papillae, six pairs of lateral papillae (Fig. 2A), in addition to presence of medioventral preanal papilla. Tail conical and flexed ventrally, 87-120 (103) [90, 100] in length.

Females (10 specimens), immature females in square brackets [Five specimens]

Length of body 18.5-40 (31) [15.8-20.1 (18.9)] mm and 0.37-0.77 (0.59) [0.22-0.33 (0.27)] mm in maximum width. Ratio of the width to body length 1:46-67 (53) [1:53-88 (71)], lips 100-130 (115) [80-100 (90)] in length and 90-200 (162) [130-140 (137)] in width. Esophagus 3250-4950 (3963) [240-3170 (2620)] in length and 80-150 (122) [75-100 (89)] in width, about 11-19 (13) [12-17 (14)] % from body length. Nerve ring and excretory pore 360-480 [320-360 (342)] and 450-710 (595) [390-500 (450)] from anterior extremity. Ventriculus 140-250 (175) [85-100 (95)] in length and 120-180 (155) [80-100 (88)] in width. Caecum 170-500 (314) [210-230 (221)] in length and 90-350 (180) [45-60 (54)] in width. Appendage 1100-2000 (1531) [520-800 (668)] in length and 80-125 (100) [30-45 (39)] in width. Ratio of appendage to esophagus 1:1.9-3.4 (2.8) [1: 4-5 (4.3)]. Ratio of caecum to each of esophagus and appendage 1: 9.9-19.1 (14.2) [1:11-14 (12)] and 1: 3-8 (5) [1: 2-4 (3)], respectively. Vulva not elevated (Fig. 1F) from body surface and situated at 5650-14200 (11012) [7000-8150 (7488)] and about 30.5-37.9 (35) [35-44 (41)] % from anterior extremity. The subglobular eggs (Fig. 1G), 38-50 (46) × 37-43 (41). The conical tail 330-650 (474) [270-300 (294)] in length, with sharp tip and has pair of lateral phasmids in its middle part (Fig. 1H, 2D).

The present worms (*I. inquires*) have highly striated and serrated cuticle, possessing additional pair of lateral row of caudal papillae in the male, so hence they are in agreement with the genus *Iheringascaris* Pereira, 1935 according to Gibbons (2010). The description of most of the current specimens is agreeable

with the description of *I. inquires* from the same type host (*R. canadum*) by Deardorff & Overstreet (1981) who resurrected the genus *Iheringascaris*, which was originally placed as a synonym of *Thynascaris* (= *Hysterothylacium*) by Hartwich (1974).

The maximum length of females of the present study reached 40 mm in comparison with 30 mm and 29.7 mm by Deardorff & Overstreet (1981) and Moravec *et al.* (2012a), respectively. Bruce & Cannon (1989) recorded *I. inquires* from cobia at Australian waters, with some variations in the length of females that considered as interspecific differences caused by the variation in the geographical distribution. Based on two specimens of this parasite, Petter & Sey (1997) reported *I. inquires* from cobia in the Arabian Gulf at Kuwaiti coasts. Moravec *et al.* (2012a) added new data on the parasite with the aid of SEM technique from *R. canadum* from Atlantic Ocean, off Carolina (United States), by absence of the alae and presence of longitudinal groove and the number of caudal papillae (33 pairs). Interestingly, the presence of the groove instead of alae was confirmed here by light microscope study too as well as number of caudal papillae of the present study (22-31) is somewhat close to 33. Malhotra *et al.* (2012) described the second species in the genus (*I. goai*) from intestine of *Sillago sihama* and spotted sea catfish *Arius maculatus* from eastern coast of India; Moravec *et al.* (2012a) considered the latter species as to need more investigation on its validity. Based on the nature of cuticle as highly striated, Moravec *et al.* (2012b) transferred two species of *Hysterothylacium* (*H. neocornuta* Rao & Shyamasundari, 1992 and *H. japonica* Rao & Shyamasundari, 1992)

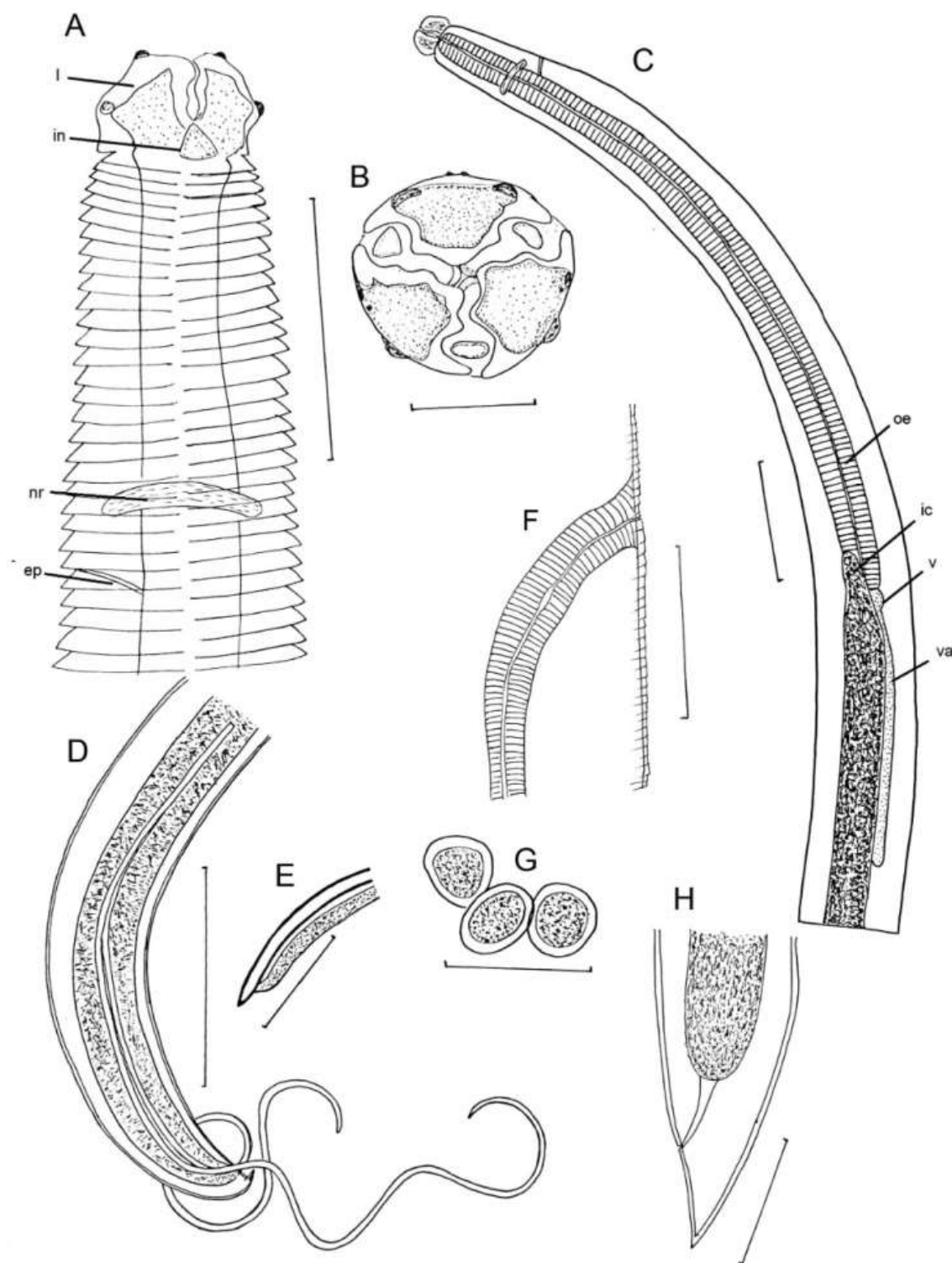


Fig. (1): *Iheringascaris inquires* adult. A: anterior end, B: lips, enface view, C: anterior part, D: posterior end of male, E: tip of spicule, F: vagina, G: eggs, H: posterior end of female. Abbreviations: ep: excretory pore, ic: intestinal caecum, in: interlabia, l: lip, nr: nerve ring, oe: oesophagus, v, ventriculus, va, ventral appendage. Scale bar A= 200 µm, B, E, G= 100 µm, H, F= 500 µm, D= 1000 µm.

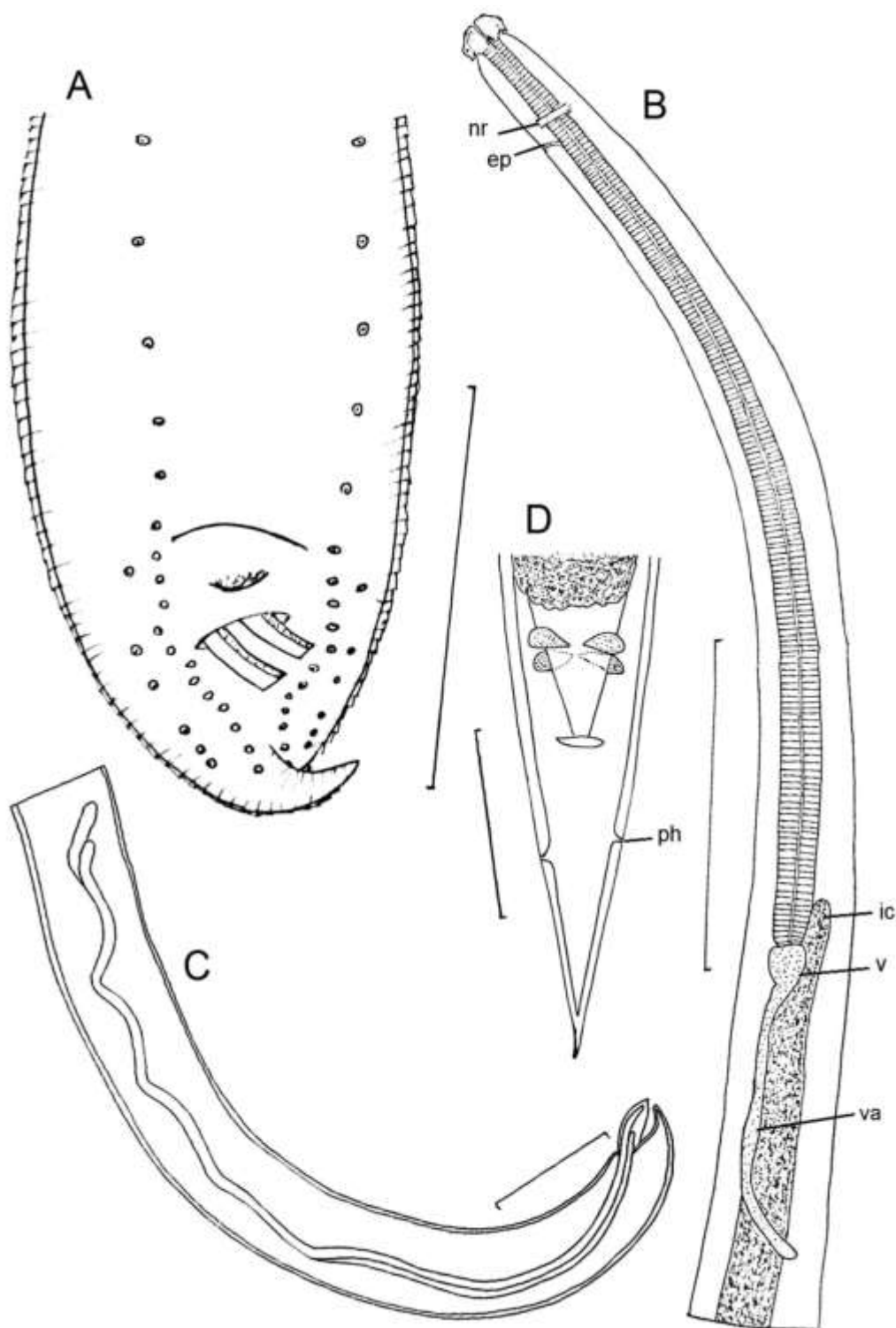


Fig. (2): *Iheringascaris iniquies*. A- posterior part of male demonstrating the distribution of caudal papillae, B- anterior part of juvenile male, C- posterior part of juvenile male, D- tail of juvenile female. Abbreviations: ep: excretory pore, ic: intestinal caecum, nr: nerve ring, ph: phasmid, v: ventriculus, va: ventral appendage. Scale bars, A, C-D= 200 µm, B= 1000µm.

from Indian waters to *Iheringascaris* and became *I. neocornuta* (Rao & Shyamasundari, 1992) and *I. japonica* (Rao & Shyamasundari, 1992), respectively.

The morphology of adult and immature of *I. inquires* of the present study, indicated that the position of vulva along body length is being similar between them. The spicules are shorter in the immature males than in the adult, the ratio of esophagus to length of body is somewhat longer in the immature than in the adult. The record of adult *I. inquires* is considered as the first in Iraq.

Family: Isthmosacanthidae

Serrasentis sagittifer (Linton, 1889)

Site of infection: Fore intestine, close to caeca.

Prevalence and mean intensity: 71% and 14.8, respectively.

Deposition: Iraqi Natural History Research Center and Museum, SN: INHM-AC1-AC25.

Description (Figs. 3 and 4)

Whitish large cylindrical worms, females larger than males. The posterior end of body wider than anterior end. The big worms possess transverse pseudo segmentation anteriorly. Proboscis cylindrical or claviform, flexed ventrally, armed with hooks longitudinally, the ventral rows more robust than dorsal rows (Figs. 3A, B), hooks 3 and 4 the larger ones. Neck short and conical in shape. Proboscis receptacle has double membrane, ganglion in the middle of proboscis receptacle. Lemnisci long and consist three times than proboscis receptacle. The neck aspinous, the anterior trunk surrounding with number of hooks (Figs. 3A-C), that conforms irregular rings, 7-9 in number with 16-20 hooks. The ventral combs

(number of small spines formed hemi circle) extending to middle of the body, the number of them larger in the females than in the males. The first group of combs close to each other compared with the posterior ones.

Males (12 specimens, fig. 3A)

Trunk length 20-90 (50.160) mm, anterior end 0.5-0.82 (0.7) mm in width, posterior end 0.65-1.25 (0.96) mm in width. Proboscis 700-1050 (898) in length and 250-400 (342) in width, armed with longitudinal hooks, 22-24 (22.8) in number, each row possessed 16-19 (17.4) hooks equal in length, the mean length of four specimens from tip to base of proboscis 30-75 (50), 30-75 (53), 33-80 (57), 38-57 (62), 45-73 (65), 50-70 (65), 50-68 (63), 50-67 (62), 48-63 (56), 42-63 (55), 38-60 (52), 33-58 (48), 33-55 (43), 28-55 (37) and 25-50 (34). The neck 300-400 (365) in length and 260-430 (350) in width in the base. Proboscis receptacle 1280-2400 (1765) in length and 170-340 (295) in width. Lemnisci 2300-7700 (5423) in length and 90-150 (120) in width. The spines in the anterior part of body 7-9 (8) irregular rings, the length of larger hook 50-85 (71). Number of ventral combs 19-24 (22), length of larger spine in the comb 70-110 (92). The testes tandem and separated with level of 1-2 combs. Anterior and posterior testes 650-1500 (1095) and 730-1800 (1183) in length and 180-480 (311) and 170-380 (311) in width, respectively. Cement glands four, very long, 9150-28060 (20190) in length and 90-170 (109) in width. Saefftigen's pouch 1400-2250 (1871) in length, copulatory organ (Fig. 3A) 550-1200 (890) in length and 450-1000 (810) in width.

Females (12 specimens, fig. 3B)

Trunk length 19.25-100 mm in length and 0.43-0.98 (0.77) mm in width. Width of posterior

end 0.9-2.44 (1.5) mm, proboscis length 800-1050 (931) and 280-450 (397) in width, armed with 22-24 (23) longitudinal hooks, each row contains 16-20 (19) hooks, measurement of hooks, 30-55 (46), 33-70 (54), 43-80 (64), 55-80 (69), 50-90 (73), 54-90 (71), 40-80 (63), 40-80 (63), 38-75 (57), 38-75 (57), 40-73 (55), 38-70 (52), 35-70 (50), 35-65 (49), 30-65 (46) and 28-60 (43). The neck 220-310 (277) in length and 260-330 (287) in width. Proboscis receptacle 1600-3172 (2002) and maximum width 190-537 (331). Lemnisci 2450-7000 (5398) and maximum width 130-150 (145). Spine rings in the anterior of body 7-8 in number, the biggest spine 48-115 (82) in length. Spine combs 21-32 (26) in number, biggest spine 70-120 (95) in length. Female reproductive system short, 2400-3200 (2900). Eggs spindle shaped, 50-108 (75) × 12-25 (19).

***Serrasentis sagittifer* cystacanths**

Site of infection: Mesenteries, intestine.

Description (Five specimens, fig. 4F)

Small whitish worms, 3.7-11.45 (6.69) mm in trunk length, 0.3-0.6 (0.44) mm in maximum width, flexed ventrally from both ends (n=4). Posterior extremity 250-450 (345) wider than anterior end. Proboscis 800-1000 (930) in length and 250-360 (320) in width. Proboscis receptacle 900-1600 (1375) in length and 200-300 (253) in width. Lemnisci long and reach to middle of body, 2350-4850 (3025) in length and 70-120 (88) in width. Proboscis hooks armature, anterior trunk spine 8-9 rows, longest spine 40-80 (55), ventral spine combs like in the adult, 19-26 (23) in number, longest spine 50-100 (64). The early males have two oval testes in level of combs 10-14. Cement glands and female reproductive system not distinguishable.

The current worms (*S. sagittifer*) possess cylindrical proboscis and double membrane in proboscis receptacle. Anterior trunk armed with hooks and surrounded with tegument, four long cement glands taxonomically agree with Rhadinorhynchidae, and the specimens have ventral combs of hooks fit with *Serrasentis* Van Cleave, 1923 (Amin, 2013; Smales, 2015). The taxonomic position of *Serrasentis* within higher taxa passed controversially changes in recent years. Verweyen *et al.* (2011), Amin (2013), Smales (2015) and Barton *et al.* (2018) placed it within Rhadinorhynchidae, order Echinorhynchida. Huston *et al.* (2020), based on molecular studies on number of marine acanthocephalans transferred *Serrasentis* with two other genera to Isthomosacanthidae, order Polymorphida.

The characters of this acanthocephalan from cobia could be identified as *S. sagittifer* (Linton, 1889), which is a wide distribution species. It was reported from fishes, mostly as cystacanths, from various regions e.g. Brazil (Luque *et al.*, 1995), India (Bhattachary, 2007), Australia (Smales, 2014), Egypt (Mohamadain & Adel, 2015) and Arabian Gulf and Gulf of Oman (Amin *et al.*, 1984; Kardousha, 2005; Hosseini *et al.*, 2013; Tavakol *et al.*, 2015). Amin (2013) listed just 14 valid species of *Serrasentis*, while Barton *et al.* (2018) discussed the validity of 14 species of *Serrasentis* and reduced species to five valid species. *S. sagittifer* shares with *S. lamelliger* and *S. nadakali* by possessing more than 15 ventral combs. However, *S. sagittifer* is distinguished by 22-24 rows in proboscis armature with 14-18 hooks per row and long cement glands (13.3-45.0 mm) in comparison with 24-28 rows of 20-26 hooks per row; short cement glands (3.3 mm) in *S. nadakali*;

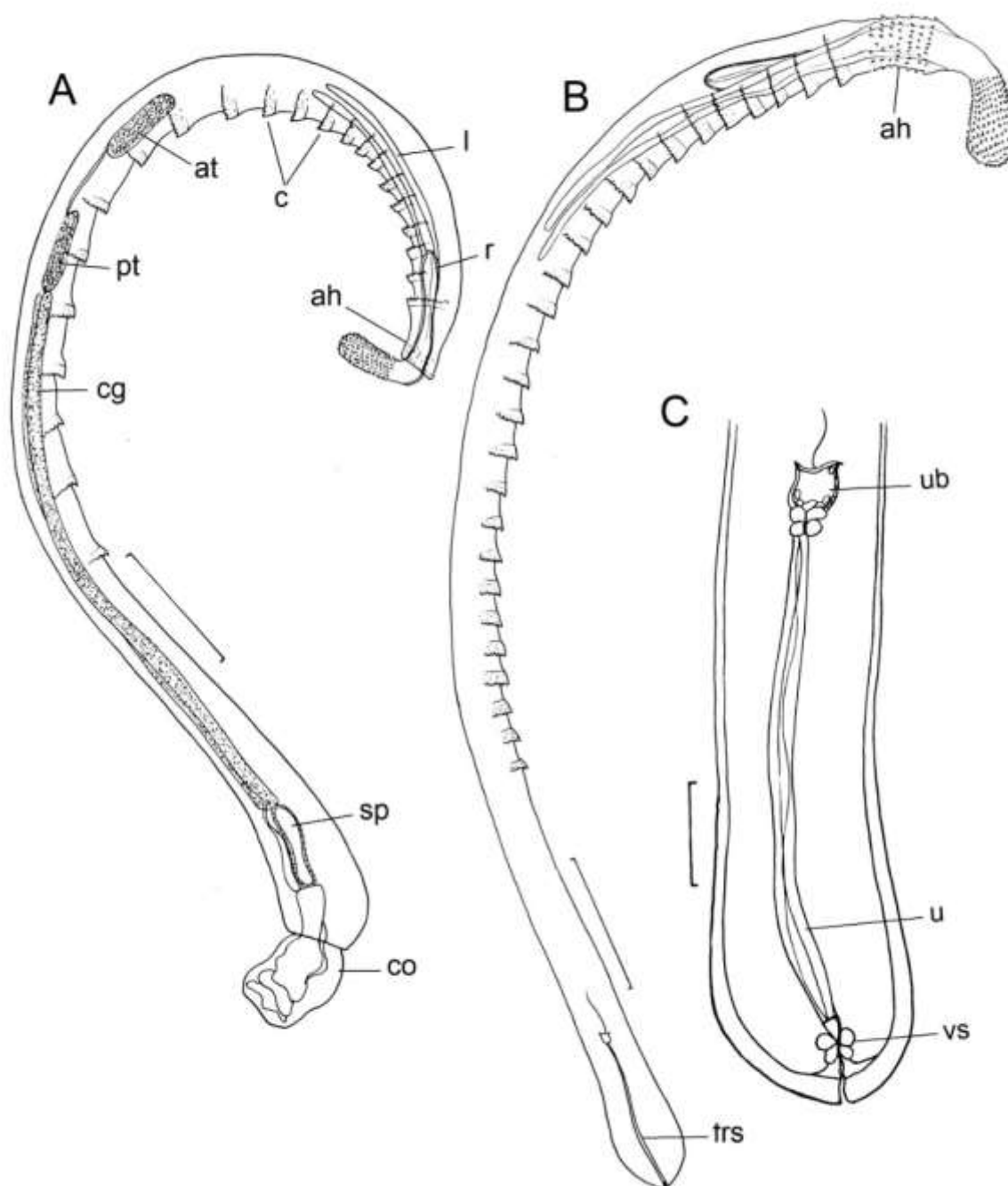


Fig. (3): *Serrasentis sagittifer*, A- adult male, 2- mature female (no eggs), C- posterior end of female with reproductive system. Abbreviations: ah: anterior hooks, at: anterior testis, c: combs, cg, cement gland, co: copulatory organ, frs: female reproductive system, l: lemnisci, pt: posterior testis, r: receptacle, sp: Saefftingen's pouch, u: uterus, ub: uterus bell, vs: vaginal sphincter. Scale bars: A, B= 200 µm, C= 300 µm.

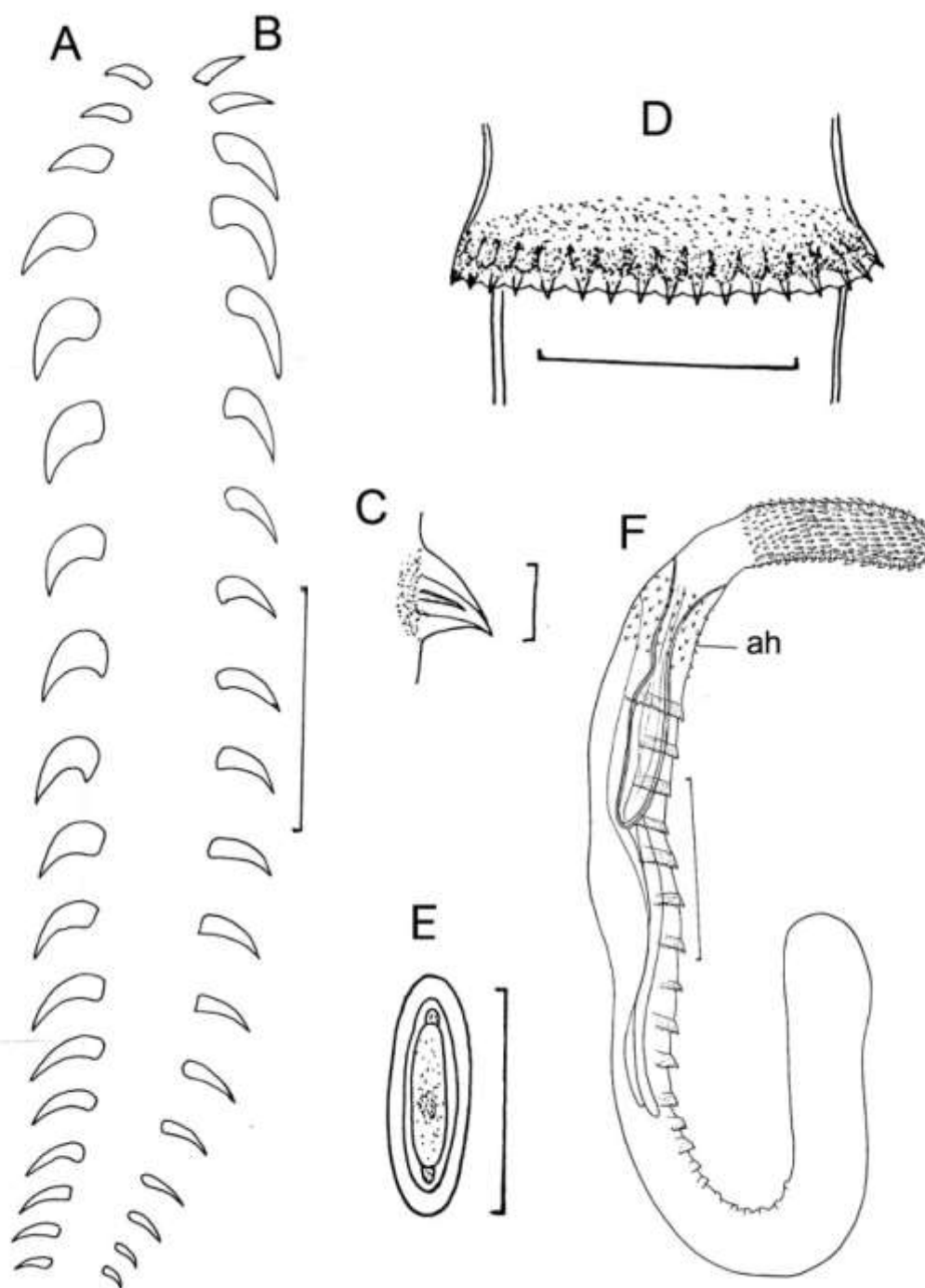


Fig. (4): *Serrasentis sagittifer*, A: ventral comb row, B- proboscis dorsal hook row, C- anterior hook, D- hook comb from female worm, E- egg, F- cystacanth. Abbreviations: ah: anterior hooks. Scale bars: A, B= 200 µm, C= 60 µm, D= 500 µm, E= 100 µm, F= 300 µm.

Proboscis armature has 24-31 longitudinal rows of three hooks or six transverse rows (12-16 hooks); definitive host is *Naucrates doctor* (Carangidae) in *S. lamelligeter* (Barton *et al.*, 2018). *S. manazo* has three small spines located dorsally on the posterior trunk, while *S. indicus* possessing proboscis armature of 18-20 rows, 10-12 hooks per row and 5-15 ventral combs.

The present report is considered as the first for adult worm of *S. sagittifer* from *R. canadum* in Iraq and Arabian Gulf and hence, this fish is considered as a new third host in Iraq for the parasite. This acanthocephalan was previously reported from Iraq as cystacanths from *Acanthopagrus arabicus* (reported as *Acanthopagrus latus*) and then from *Platax teira* (Mhaisen *et al.*, 2018).

Conclusions

The present investigation is the first parasitological study on cobia, *R. canadum* in Iraq. It demonstrated the record of adults of two parasitic worms which have high specificity to cobia, the type and the only host for the nematode *I. iniquus* and for the adult *S. sagittifer* in the World.

Conflict of interest

The authors declared that they have no conflict of interest.

Ethical approval

All applicable national and international guidelines for the care and use of animals were followed.

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التسجيل الأول لإثنين من الديدان الطفيلية في أسماك السكن (*Rachycentron canadum* (L.) من المياه البحرية الإقليمية للعراق (Carangiformes: Rachycentridae)

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المستخلص: جمعت 14 عينة من أسماك السكن (*Rachycentron canadum* (L.) من المياه البحرية العراقية خلال الفترة من شهر تشرين الأول 2019 ولغاية أيار 2020. عزل من القناة الهضمية لهذه الأسماك نوعين من الديدان الطفيلية: نوع من الديدان الخيطية ونوع من الديدان شوكية الرأس، الطور البالغ للدودة للخيطية (*Iheringascaris inquires* (Linton, 1901) Deardorff بالإضافة إلى شوكية الرأس *Serrasentis sagittifer* Linton, 1889. تسجيل هذين الدودتين، من هذه السمكة، يعدّ الأول في العراق. الوصف التفصيلي للدودة الخيطية *I. inquires*، كما ان وصف بالغات شوكية الرأس *S. sagittifer* من سمكة السكن يعدّ الأول من الخليج العربي.

كلمات مفتاحية: أسماك بحرية، ديدان خيطية، *Iheringascaris inquires*، شوكية الرأس، *Serrasentis sagittifer*، العراق.