



Reexamining *Pseudoglaridacris confusus* (Hunter, 1929) nom. nov. (Cestoda: Caryophyllidae) from buffalo fish, *Ictiobus* spp. in Arizona, USA

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Abstract

Specimens of *Pseudoglaridacris confusus* (Hunter, 1929) nom. nov. (Caryophyllidae) first described as *Glaridacris confusus* Hunter, 1929 and subsequently designated as *Pseudoglaridacris confusa* Oros, Uhrovič, Scholz, 2018 were collected and examined from 3 species of buffalofish, *Ictiobus cyprinellus* (Valenciennes), *I. bubalus* (Raf.), and *I. niger* (Raf.) in 2 Arizona lakes, Roosevelt and Apache lakes, on the Salt River, Arizona. We update our first and only 1969 redescription of this Arizona population, with new observations and measurements, compare with those in the original description, and assess the status of *P. confusus* in light of the recent taxonomic treatments of this species. The morphology of *P. confusus* is revisited using light microscopy revealing new features. Emphasis is placed on the morphological variability of its scolex. The re-examination of over 50 specimens from the Apache and Roosevelt lakes provided an opportunity to revise the taxonomy of the species and correct and update some old statements and concepts by other observers.

Keywords: *Pseudoglaridacris confusus*; Zoogeography; Caryophyllideans

Introduction

Mackiewicz [1] reviewed the morphology, anatomy, development, zoogeography, and systematics of the Caryophyllidea van Beneden (Cestoidea) and indicated the existence of 89 species and 37 genera in these monozoic cestodes. Mackiewicz [2] provided perspectives on the evolution, biology and zoogeography of 128 species in 45 genera of caryophyllids from the Nearctic, Neotropical, Palearctic, Ethiopian, Oriental, and Australian regions. Mackiewicz [3] later recognized about 150 species in 41 genera worldwide (except for the Neotropical Region). Of these, 14 genera and 90 species belonging to 3 families were described from the Indo-Malayan region from catfishes

(Siluriformes: Bagridae, Clariidae, Heteropneustidae, Schilbeidae and Siluridae), cyprinid, and cobitid fishes [3]. Ash, et al. [4,5] invalidated a large number of inadequately described species from clariid and hetero-pneustid catfishes in India and neighboring countries. Most recently, however, Scholz, et al. [6] and Scholz, et al. [7] recognized only 117 species and 46 genera in order Caryophyllidea Carus, 1863 parasitizing teleost fish (Cypriniformes, Siluriformes, and some catfish). Chubb [8] reviewed the seasonal ecology of the Caryophyllidea in the various climate zones of the world.

The order consists of four families (Balanotaeniidae, Capingentidae, Caryophyllaeidae, Lytocestidae) [3]. Mackiewicz [1,3] stated that "The caryophyllideans are

exclusively intestinal parasites of freshwater teleosts with 85 species reported from cypriniform and 29 from siluriform fishes. Only few species of the *Lytocestus* with 2 species and monotypic *Lytocestoides* are known from mormyrids (Osteoglossiformes), characiform and cichlid (Perciforms) fishes, respectively." Within families, 40 % of caryophyllideans parasitize North American suckers (Catostomidae), 26 % cyprinids, 10 % clariid catfishes and just a few caryophyllideans have been reported from 12 additional fish families" [7]. Freshwater annelids of the family Naididae, especially species previously placed in the Tubificidae (*Tubifex tubifex*, *Limnodrilus* spp.), serve as the only intermediate hosts [1].

Amin [9-12] recognized 15 species in 8 families of caryophyllid cestodes mostly from suckers (Catostomidae) in lake and river systems in Arizona and Wisconsin (Table 1). These included *Pseudoglaridacris confusus* (Hunter, 1929) Oros, Uhrovič, Scholz, 2018 nom. nov. for *Pseudoglaridacris confusa* Oros, Uhrovič, Scholz, 2018. These latter authors redescribed the genus *Glaridacris* and created a new genus, *Pseudoglaridacris* with 2 additional species: *Pseudoglaridacris laruei* (Lamont, 1921) Oros, Uhrovič, Scholz, 2018, and *Pseudoglaridacris oligorchis* (Haderlie, 1953) Oros, Uhrovič, Scholz, 2018. See collections, following, for details.

Host species	Parasite species	State
<i>Carpoides Cyprinus</i> (LeSueur); quillback	<i>Rowardleus pennensis</i> Mackiewicz and Deutsch, 1976	W
<i>Catostomus clarkii</i> Baird and Girard; desert sucker	<i>Isoglaridacris hexacotyle</i> (Linton, 1897) Mackiewicz, 1968	A
<i>Catostomus commersoni</i> (Lacépède); white sucker	<i>Biacetabulum biloculoides</i> Mackiewicz and McCrae, 1965	W
<i>Catostomus commersoni</i> (Lacépède); white sucker	<i>Biacetabulum macrocephalum</i> MacCrae, 1962	W
<i>Catostomus commersoni</i> (Lacépède); white sucker	<i>Biacetabulum</i> sp. Hunter, 1927	W
<i>Catostomus commersoni</i> (Lacépède); white sucker	<i>Glaridacris catostomi</i> cooper, 1920	W
<i>Catostomus commersoni</i> (Lacépède); white sucker	<i>Hunterrella nodulosa</i> Mackiewicz and McCrae, 1962	W
<i>Catostomus commersoni</i> (Lacépède); white sucker	<i>Isoglaridacris folius</i> Fredrickson and Ulmer, 1967	W
<i>Catostomus commersoni</i> (Lacépède); white sucker	<i>Monobothium hunteri</i> Mackiewicz, 1963	W
<i>Catostomus insignis</i> Baird and Girard; Sonora sucker	<i>Isoglaridacris hexacotyle</i> (Linton, 1897) Mackiewicz, 1968	A
<i>Cyprinus carpio</i> Linn.; common carp	<i>Atractolytocestus huronensis</i> Anthony, 1958	W
<i>Cyprinus carpio</i> Linn.; common carp	<i>Khawia iowensis</i> Calentine and Ulmer, 1961 (W)	W
<i>Erimyzon sucetta</i> (Lacépède); lake chubsucker	<i>Pseudoglaridacris laruei</i> (Lamont, 1921) Oros, Uhrovič, Scholz, 2018	W
<i>Erimyzon sucetta</i> (Lacépède); lake chubsucker	<i>Isoglaridacris agminis</i> Williams and Rogers, 1972	W
<i>Erimyzon sucetta</i> (Lacépède); lake chubsucker	<i>Isoglaridacris multivitellaria</i> Amin, 1986	W
<i>Ictiobus</i> spp.; buffalofish	<i>Pseudoglaridacris confusus</i> (Hunter, 1929) Oros, Uhrovič, Scholz, 2018	A

Table 1: Caryophyllid cestodes collected from fish in Arizona (A) and Wisconsin (W) as reported in Amin.

In Arizona, Amin [9,10] gave a new description of *P. confusus* as *Glaridacris confusus* from 3 species of buffalo fish, *Ictiobus* spp., in 2 lakes, Apache and Roosevelt lakes on the Salt River. Since its original description in Hunter (1930), it has been reported from hosts along the Mississippi River and New York (Amin [9]). Buffalo fish appear to be the common usual hosts. Later reports list 8 species of fish hosts in wider geographical areas of the US and Canada [13,14]. The southernmost distribution of *P. confusus* appears to be Oaxaca and Veracruz, Mexico from *Ictiobus meridionalis* (Günther) [15]. The number of true hosts of *P. confusus* appears to be considerably less than has been reported (Amin [9]). In his

Ph.D. dissertation, Mackiewicz [16] reviewed reports that were based on its erroneous identification from various hosts confusing it with *Pseudoglaridacris laruei* (Lamont, 1921) Oros, Uhrovič, Scholz, 2018, *Glaridacris catostomi* Cooper, 1920, and *Spartoides wardi* Hunter, 1929. In addition to buffalo fish, *P. confusus* was also verified from *Dorosoma cepedianum* Lesueur (Gizzard shad) in Tallahatchie River, Mississippi by Hunter (1930) and from *Carpoides carpio* Rafinesque (common carp sucker) and from *I. cyprinellus* in Lake Texoma, Oklahoma by Mackiewicz JS [17] and McAllister CT [18].

Materials, Collections and Methods

Although *P. confusus* has been reported in all the 4 impoundment lakes on the Arizona Salt River (Apache, Roosevelt, Canyon, and Sahuaro lakes), only the first two lakes appear to be significantly populated by this cestode. Apache Lake (33°34'43"N 111°15'48"W), the second largest of the 4 Salt River Project Reservoirs, is about 104 km northeast of Phoenix in Maricopa and Gila counties. Apache Lake, was formed by Horse Mesa Dam impounding the Salt River, which was completed in 1927. Apache Lake is located about 8 km downstream from Roosevelt Lake, the largest in Gila County, created in 1911 about 170 km from Phoenix by car; (47°57'22"N 118°58'50"W, at Grand Coulee Dam), and upstream from the smaller Canyon Lake and Saguaro Lake closest to Phoenix.

The 3 species of buffalo fish, *Ictiobus cyprinellus* (Valenciennes) large-mouth buffalo fish, *I. bubalus* (Raf.) small mouth buffalo fish, and *I. niger* (Raf.) black buffalo fish, inhabit Apache Lake (collected at Burnt Corral) whereas Roosevelt Lake is inhabited only by *I. cyprinellus* (collected at Tonto, Grape, Vine, and 3 Bar). Fish were captured by netting and boat shocking using current from 123-v, 2650-3000-w generator.

A total of 154 specimens were collected from 96 individuals of 3 species of buffalo fish examined (18.75%) from both lakes, Apache and Roosevelt lakes in 1966 and 1967. The zoo-plankton feeder with large terminal mouth, *I. cyprinellus*, from both lakes showed the lowest infection prevalence (12.12% and 7.14%) and intensity (0.24 and 0.09) in Roosevelt and Apache lakes, respectively. The small benthic invertebrate feeders with small fleshy subterminal mouths, *I. bubalus* and *I. niger* from Apache Lake, had considerably higher prevalence (38.46% and 75.00%) and intensity (4.00 and 11.25), respectively.

After capture and transfer to the laboratory in coolers on ice, the fish were dissected within a few hours after capture. Upon recovery, worms were placed at once in warm 70% ethanol then processed and stained with Semichon's carmine. Worms were then processed in ascending concentrations of ethanol for dehydration reaching 100%. Worms were ultimately cleared in oil of winter green before whole mounting in Canada balsam. Measurements are in mm as noted in Table 2; the range is followed by the mean values between parentheses. Width measurements represent maximum width. Microscope images were created using 10X

and 40X objective lenses of a BH2 light Olympus microscope (Olympus Optical Co., Osachi-shibamiya, Okaya, Nagano, Japan) attached to an AmScope 1000 video camera (United Scope LLC, dba AmScope, Irvine, California), linked to an ASUS lab top equipped with HDMI high-definition multimedia interface system (Taiwan-USA, Fremont, California). Images from the microscope were transferred from the labtop to a USB and stored for subsequent processing on a computer. We found microscope images to be considerably more informative than schematic line drawings as they depict the natural appearance of anatomical structures.

Results

Buffalo fish eggs (unknown species) were introduced in Roosevelt Lake in 1918 by the US Bureau of Fisheries [19] and buffalo fish have been reported in all 4 lakes on the Salt River. Buffalo fish do not limit their feeding to any particular depth. Diet does not appear to be affected by fish size (age) but mouth position, feeding habits and diet are definitely related to their parasitic fauna (Amin [10] and others included therein).

Pseudoglaridacris confusus was the only species of caryophyllid cestode found in all 3 species of buffalo fish introduced to Arizona over 100 years ago. Mackiewicz [17], however, listed 8 species of Caryophyllidae in *I. bubalus* and *I. cyprinellus* from Lake Texoma. It is suggested that the parasite fauna of fish is reduced following introduction into new habitats because of the differential distribution of supporting intermediate hosts.

Amin [9,10] redescribed *Pseudoglaridacris confusus*, as *Glaridacris confusus*, from the Arizona population collected from the 3 species of buffalo fish captured in Apache and Roosevelt lakes. He included 8 line drawings; one of a whole worm, 5 of scolex variations and 2 of immatures. We have restudied specimens in our personal collection. Measurements from about 100 specimens were made in 1967-1968 [9,10] and new measurements and observations from an additional 50 specimens from the original collection were recently made and added to the earlier measurements creating composite measurements included in Table 2. We provide below a brief revised description of our qualitative morphological findings and a comparative morphometrical data (Table 2) high-lightening variations from the only other description of Hunter [20] which included line drawings of a scolex, reproductive system, and a few sections.

Sources	Amin (1968, 1969 & present paper)		Hunter (1930)
Hosts	<i>Ictiobus Cyprinellus, I. bubalus, I. niger</i>		<i>Ictiobus sp., I. bubalus, Dorosoma cepedianum</i>
Geography	Roosevelt & Apache lakes on the Salt River, Arizona	N	Talahatchie River, Mississippi
Total length	0.90 – 4.69 (2.150)*	105	3.00 – 7.00; up to 20.00
Pre-vitelline	0.25 – 0.94 (0.560)	83	---
Post-vitelline	0.65 – 3.75 (1.590)	83	---
Scolex length	0.20 – 0.77 (0.473)	70	---
Scolex width	0.22 – 0.94 (0.598)	70	---
Width below scolex	0.18 – 0.65 (0.446)	70	---
Testes number	27 – 42 (35)		25 – 35
Testes length	0.10 – 0.21 (0.155)	40	0.100 – 0.300
Testes width	0.08 – 0.14 (0.103)	40	0.100 – 0.13
Vitellaria diameter	0.07 – 0.11 (0.098)	35	0.067 – 0.135
Width at male gonopore	0.20 – 0.77 (0.461)	81	0.20 – 0.80
Width at uterine coils	0.16 – 0.69 (0.474)	81	0.20 – 0.80
Cirrus sac diameter	0.04 – 0.14 (0.10)	53	0.10 – 0.16
Wings of ovary length	0.23 – 0.364 (0.297)	35	0.40 – 0.75
Wings of ovary width	0.10 – 0.21 (0.170)	35	0.10 – 0.12
Commissure diameter	0.08 – 0.19 (0.115)	30	0.030 – 0.12
Ovoid egg Length	0.035 – 0.045 (0.040)	40	0.037 – 0.048 (0.042) (N=50)
Ovoid egg width	0.025 – 0.032 (0.029)	40	0.020 – 0.031 (0.025) (N=50)

*Range (mean); all measurements in mm.

Table 2: Morphometric description of *Pseudoglaridacris confusus* collected from buffalofish, *Ictiobus* spp., in Arizona and Mississippi.

Brief Morphological Description of Adults

Body small and broad, widest at scolex and dorso-ventrally flattened, covered with thin cuticle enclosing cortical parenchyma and inner longitudinal muscles. Measurements and counts in Table 2. Scolex oval at base, fanning anteriorly to widest point at middle then tapering to a dome-shaped apical end cut by 6 well-defined loculi separated by strong ribs. Testes round, relatively large, in 2 central parallel rows. Vitelline glands much smaller than testes, ovoid-elongate, in 2 lateral fields, extending posteriorly to anterior end of ovaries with few post-ovarian vitellaria usually in an open U-shape form. Vas deference detectable about middle of testicular field and ending dorsal and anterior to cirrus sac having strong muscular wall filling about half medullary parenchyma. Granular ovary dumb-bell-shaped, thick and short, and vagina opening in a receptaculum siminis anterior and dorsal to thick ovarian commissure. Uterine coils do not extend anterior to cirrus sac. Male and female genital openings separate. Posterior end bluntly obtuse with slight middle invagination where excretory pore drains. Egg round-ovoid, relatively

thick-shelled, containing ovum and vitelline globules.

Discussion

The taxonomic status of *Glaridacris confusus* Hunter, 1929 has not been challenged until Oros, et al. [21] erected the genus *Pseudoglaridacris* Oros, Uhrovič, Scholz, 2018 and included 3 former members of *Glaridacris* in it: *Pseudoglaridacris confusa* Oros, Uhrovič, Scholz, 2018, *Pseudoglaridacris laruei* (Lamont, 1921) Oros, Uhrovič, Scholz, 2018, and *Pseudoglaridacris oligorchis* (Haderlie, 1953) Oros, Uhrovič, Scholz, 2018. Oros, et al. [21] diagnosed the new genus, differentiated it from *Glaridacris*, provided systematic summaries and remarks but did not redescribe or measure these 3 species. *Pseudoglaridacris* was distinguished from *Glaridacris* in having compact rather than follicular ovary, bothrioloculodiscate rather than cuneiform scolex, genital atrium rather than well separated gonopores, lateral position of vitelline follicles rather than also having median follicles, and a smaller size body. We accept the designation of *Pseudoglaridacris* with qualifications based on our

specimens from Arizona. In our specimens:

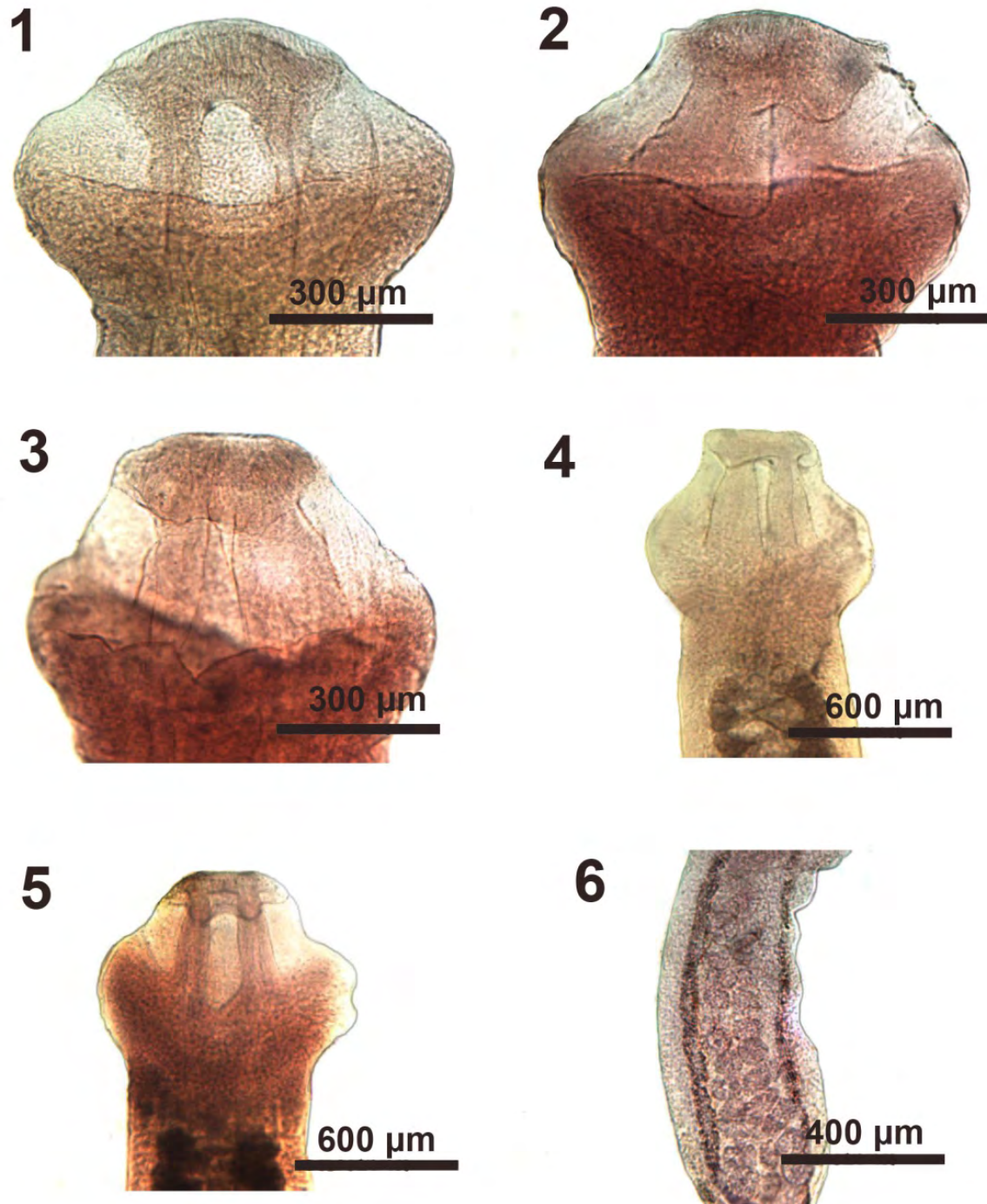
1. The male and female gonopores ARE separate even though they may appear to be in a genital atrium depending on the state of contraction of the specimen.
2. The scolex appeared dome-shaped apically in all our specimens (Figures 1-5), a shape not mentioned in Oros, et al. [21] who depicted a different bluntly-apically-pointed scolex (Figure 3A) with what appears to be additional smaller anterior loculi. The scolex, however, appeared cuneiform (wedge-shaped) in Hunter's [20], (Figure 2) material.
3. We have never seen a V-shaped post-ovarian vitelline bands in our many specimens (Figures 7-10), neither did Hunter [20]. Figure 3B in Oros et al. [21] from Mississippi and Figure 2G in Scholz and Pérez-Ponce de León [15] from Mexico depict V-shaped post-ovarian vitelline bands. Figure. 2G of the latter authors, unlike our specimens, depicted an elongate slender specimen with unusually narrow scolex, poorly defined loculi, slender ovary arms, and a reduced number of post-ovarian vitelline glands forming V-shaped band touching lateral lobes of ovary.
4. While we accept the assignment of *G. confusus* in *Pseudoglaridacris*, we do not see a cause for changing the second name from *confusus* to *confusa*. Oros, et al. [21] explained the *Pseudo-* part of *Pseudoglaridacris* but did not give a reason for changing *confusus* to *confusa*. Oros, et al. [21] did not redescribe their *P. confusa* but in their taxonomic summary, indicated that "*Glaridacris confusa* Hunter, 1929" is a synonym. That name, "*confusa*" actually, does not exist. Hunter [20] stated "It was named *G. confusus* because of its superficial resemblance to the *Lytocestinae*." We thought that this should be respected and have decided to accept *Pseudoglaridacris confusa* as *Pseudoglaridacris confusus* nom. nov. Other authors have also used the name *Pseudoglaridacris confusa* including McAllister, et al. [18] and Scholz and Perez-Ponce de Leon [15].

Morphometrically, our specimens from Arizona were considerably smaller than those of Hunter [20] (Table 2) from Mississippi but the ovary was shorter and wider (Figures 7-10) making the total mass of ovarian tissue about the same. The state of contraction of this and may be of other structures, could be related. All other structures were of comparable sizes. The scolex in our Arizona specimens characteristically was dome-shaped, apically (Figure 1-5), never wedge-shaped or pointed anteriorly. The arrangement of testes vs. pre-ovarian vitelline band is shown in a young specimen (Figure 6).

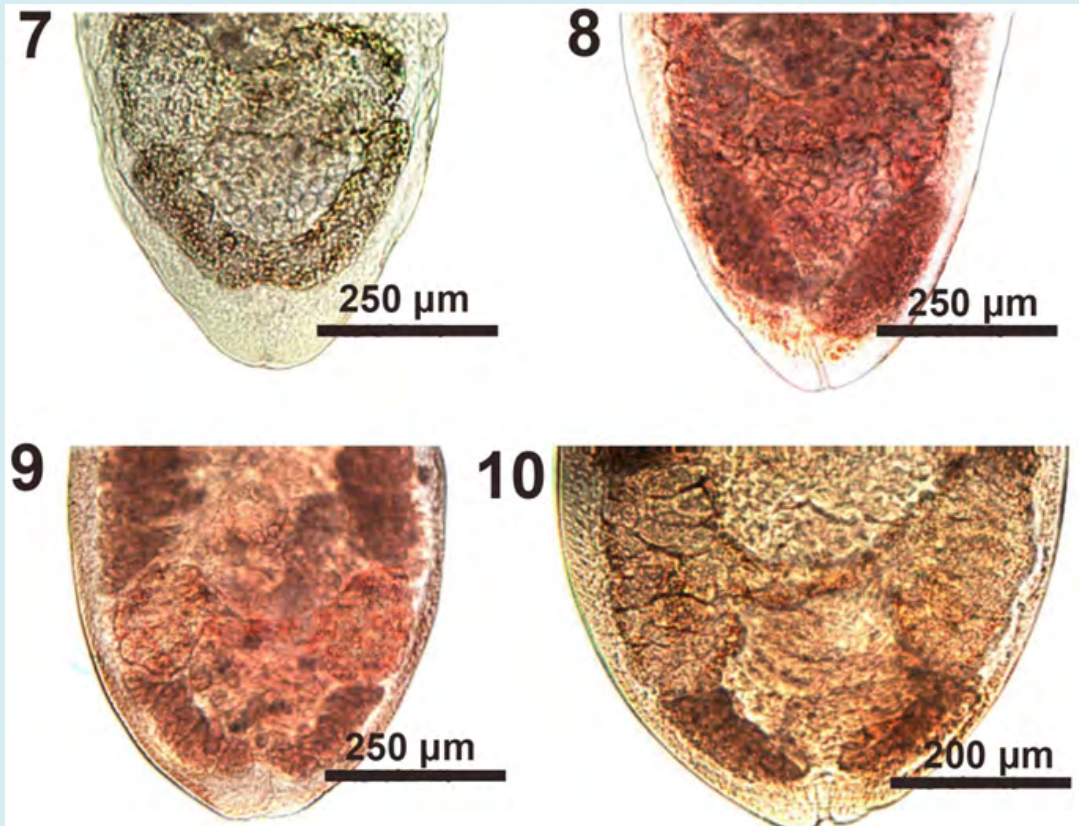
Quantitatively, infection parameters appear to be related to the anatomy and position of the buffalo fish mouth parts since all 3 species appear to have the potential of serving as

definitive hosts. *Ictiobus cyprinellus* has a thin-lipped terminal mouth and feed by scooping the water for zoo-planktons. *Ictiobus bubalus* and *I. niger*, in another category, have small, thick-lipped sub-ventral mouth and they bottom feed mostly on benthic invertebrates including a larger concentration of intermediate hosts rendering them considerably more frequently and heavily infected than *I. cyprinellus*. Mixed infections of more than one species of caryophyllid cestodes in the same host do not appear to be uncommon. Up to 4 species of cestodes may be found in the same host individual but double infections are usually more common [16]. The argument that earlier infections may elicit immunological reactions preventing subsequent infections does not appear to be true for caryophyllid cestodes [16]. The absence of competition in the fish intestinal tract is an important factor in cases of multiple infections. Niche segregation might not only be of spatial nature but the mode of attachment may also be involved. *Pseudoglaridacris confusus* is weakly attached to mucosal folds and its lack of intimate mechanical and possibly physiological association with host tissue may be partially responsible for its wide range of host specificity in up to 8 host species. On the other hand, other caryophyllid cestodes like *Monobothrium hunteri* Mackiewicz, 1963 is firmly attached [16] and more host specific.

In *P. confusus*, allometric growth was observed to proceed at a considerably larger rate in regions containing reproductive structures posterior to first vitellaria compared to pre-vitelline region of scolex and neck. Hunter ([20], Tables 1&2, graphic chart 2) demonstrated this phenomenon from a study of 106 specimens in 7 length classes reaching 20 mm long worms. Hunter's [20] pioneering concept applied also to other caryophyllid as well as pseudophyllid examples. Amin [10] also studied growth patterns in *Isoglaridacris hexacotyle* (Linton, 1897) Mackiewicz, 1968 from *Catostomus insignis* Baird & Girard in the lower Salt River, Tempe, Arizona, and demonstrated the acceleration of growth of the post-vitelline region, the region of reproductive activity, compared to the pre-vitelline region including the scolex and neck, as well as the growth in the scolex, cirrus sac, and in width at the common gonopore region. Fischthal, et al. [22] showed comparable growth of the post-vitelline region of *Pseudoglaridacris laruei* from *C. commersoni* in New York. We have additionally observed the same phenomenon in non-tapeworm helminths also enhancing the increased reproductive potential of worms. Amin [10] also studied a total of 136 metacercariae of *Clinostomum marginatum* Rudolphi, 1819 collected from 17 *C. insignis* and 4 *C. clarkii* Baird & Girard in the Upper Salt River, Arizona, placed in 6 size classes ranging from 2.49 to 6.04 mm in length. The hind body posterior to the constriction at the level of the ventral sucker including the developing reproductive structures grew at a considerably faster rate than the forebody anterior to the ventral sucker.



Figures 1-6: Microscope images of adult *Pseudoglaridacris confusus* from 3 species of buffalo fish in Apache and Roosevelt lakes, Arizona. 1-5. Variations in shape of scolex. Note the dome-shaped apical end, the wide middle and the well ribbed loculi. 6. Part of a young specimen showing the medullary testicular field and the vitelline bands confined to two lateral fields.



Figures 7-10: Microscope images of adult *Pseudoglaridacris confusus* from 3 species of buffalo fish in Apache and Roosevelt lakes, Arizona. The posterior portion of 4 specimens showing the thick and short ovarian arms and the open U-shaped post-ovarian vitelline characteristic of the Arizona population. Also note the unusual arrangement of the asymmetrical and unequal sides of the post-ovarian vitelline bands (Figure 8) and the cirrus sac (Figure 9).

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Declarations

- **Compliance with Ethical Standards**
- **Conflict of Interest:** The authors declare no conflicts of interest or competing interests.
- **Ethical Approval:** The authors declare that they have observed all applicable ethical standards.
- **Availability of Data:** All presented and related data are available by contacting the senior author.

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