

NEW SPECIES OF *GYRODACTYLUS* (MONOGENOIDEA,
GYRODACTYLIDAE) FROM THE WHITE SANDS PUPFISH,
CYPRINODON TULAROSA, IN NEW MEXICO

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ABSTRACT—A new species of *Gyrodactylus* (Monogenoidea: Gyrodactylidae) is described from the skin of the White Sands pupfish, *Cyprinodon tularosa* Miller and Echelle, 1975 (Cyprinodontiformes: Cyprinodontidae) from Salt Creek, White Sands Missile Range, Tularosa Basin, New Mexico. The new species is compared with the 6 species of Gyrodactylidae known to parasitize pupfish in the southern United States.

RESUMEN—Una nueva especie de *Gyrodactylus* (Monogenoidea: Gyrodactylidae) se describe de la piel del pez cachorrillo *Cyprinodon tularosa* Miller and Echelle, 1975 (Cyprinodontiformes, Cyprinodontidae) de Salt Creek, White Sands Missile Range, Tularosa Basin, New Mexico. La nueva especie se compara con seis especies de Gyrodactylidae que parasitan al pez cachorrillo al sur de los Estados Unidos.

Native populations of the White Sands pupfish, *Cyprinodon tularosa* Miller and Echelle, 1975 (Cyprinodontiformes, Cyprinodontidae), occur only in Salt Creek and Malpais Spring (type locality) on the White Sands Missile Range, Tularosa Basin, New Mexico. Non-native pupfish populations at Mound Spring, White Sands Missile Range, and in the Lost River, Holloman Air Force Base, New Mexico (Pittenger and Springer, 1999), are descended from the native population in Salt Creek (Stockwell et al., 1998). The fish is listed as threatened by the state of New Mexico because of its limited distribution (Miller and Echelle, 1975; Pittenger and Springer, 1999). During research on the evolutionary ecology of White Sands pupfish by CAS, gyrodactylids were discovered parasitizing the skin of the fish. These helminths belonged to a new species of *Gyrodactylus* von Nordmann, 1832 (Monogenoidea, Gyrodactylidae) that is described herein.

White Sands pupfish were collected by beach seine from Salt Creek at Range Road 316, White Sands Missile Range, Tularosa Basin, Sierra County, New Mexico (Township 12S, Range 6E, NE¼ section 3) during 2003. Fish were sacrificed in MS-222 (500 mg/L) and im-

mediately placed in hot (60°C) 5% formalin solution. Gyrodactylids were subsequently removed from the sediment using a small probe and stereomicroscope. Some specimens were mounted unstained in Gray and Wess medium for study of sclerotized structures; other specimens were stained with Gomori's trichrome and mounted as whole mounts in Canada balsam for study of soft anatomy (Kritsky et al., 1978; Humason, 1979). Illustrations were prepared with the aid of a camera lucida or microprojector. Anatomical terminology was that presented by Mizelle and Kritsky (1967a) and Kritsky and Mizelle (1968). Type specimens were deposited in the United States National Parasite Collection (USNPC), Beltsville, Maryland and the British Museum (Natural History) (BM[NH]), London, United Kingdom. For comparative purposes, the following museum specimens were examined: holotype, paratype, *Gyrodactylus cyprinodontis* Mizelle and Kritsky, 1967 (USNPC 62951); holotype, 2 paratypes, *Gyrodactylus hargisi* Williams and Rogers, 1971 (USNPC 71759, 71760).

Gyrodactylus tularosae sp. n.

Description—Body fusiform; greatest width usually at level of uterus containing embryo;

peduncle tapering toward haptor (Fig. 1a). Cephalic lobes well developed, each containing head organ, spike sensilla. Cephalic glands comprising 3 bilateral groups of unicellular glands: 2 pairs prepharyngeal, single pair postpharyngeal. Pharynx having 2 tandem bulbs, posterior bulb larger, anterior bulb with 8 papillae frequently emerging from mouth. Esophagus moderately long; intestinal ceca nonconfluent posterior to gonads, with moderately thick wall. Testis subovate, lying posterodorsal to ovary. Proximal portion of vas deferens not observed. Two tandem seminal vesicles simple dilations of vas deferens; larger proximal vesicle lying to right or left of body midline at level of uterine pore; distal vesicle small, emptying into copulatory organ through small dorsal pore. Two bilateral prostatic glands, 1 lying on each side of distal seminal vesicle; each prostatic gland with 1 or 2 nuclei, emptying into copulatory organ by short duct. Copulatory organ armed with large spine, 3 to 5 (usually 5) spinelets; medial spinelets smaller than lateral spinelets (Fig. 1b). Ovary containing large oocyte surrounded by thin peripheral layer of germinal cells; oviduct short; uterus with 1 or 2 generations of embryos (or empty); uterine pore midventral at level of proximal seminal vesicle. Vitellarium absent or replaced by up to 7 apparently syncytial masses located in posterior trunk, peduncle; 2 anterior syncytial masses with reticulate cytoplasm. Haptor subcircular. Hooks similar, intrahamular; shank uniform, with imperceptible proximal enlargement; hooklet with short open point, sloping shaft, globose heel, tapered toe with diagonal termination; filamentous hooklet loop about one-half shank length (Fig. 1c). Superficial anchor root folded medially, flattened, with thickened edges; anchor fold present, securing ends of superficial bar; anchor shaft slightly arched; point recurved, elongate (Fig. 1d). Superficial bar with moderately long anterolateral terminal rami; superficial-bar shield tapered posteriorly, extending to mid-length of anchor shaft; deep bar variable, with attenuated ends inserted into anchor base (Fig. 1d).

Measurements—Table 1.

Type Host—White Sands pupfish, *Cyprinodon tularosa* Miller and Echelle, 1975

Site—Skin

Type Locality—Salt Creek (Tularosa Basin),

White Sands Missile Range, Sierra County, New Mexico.

Collection Date—12 September 2003.

Specimens Deposited—Holotype, USNPC 94779; 20 paratypes, USNPC 94780, BM(NH) 2004.4.20.1–8

Etymology—This species is named for its host, *Cyprinodon tularosa*.

Remarks—North American species of *Cyprinodon* are known to be parasitized by 6 species of Gyrodactylidae: *Gyrodactylus nevadensis* Mizelle and Kritsky, 1967, *Gyrodactylus saratogensis* Mizelle and Kritsky, 1967, and *Gyrodactylus cyprinodontis* from the Amargosa pupfish, *Cyprinodon nevadensis nevadensis* Eigenmann and Eigenmann, 1889 in Saratoga Springs, Death Valley, California (Mizelle and Kritsky, 1967a); and *Gyrodactylus hargisi*, *Gyrodactylus mobilensis* Williams and Rogers, 1971, and *Fundulotrema prolongis* (Hargis, 1955) Kritsky and Thatcher, 1977 from the sheepshead minnow, *Cyprinodon variegatus* Lacépède, 1803 on Dauphin Island, Alabama (Williams and Rogers, 1971) Williams and Rogers (1971) considered *Gyrodactylus* sp. of Hargis (1955) on *C. variegatus* from Alligator Harbor, Florida, to be a synonym of *G. hargisi*.

Gyrodactylus tularosae is easily differentiated from *G. mobilensis* by having folded superficial anchor roots (roots straight in *G. mobilensis*) and hooklet points shorter than the hooklet shafts (hooklet point longer than shaft in *G. mobilensis*). However, morphologies of the haptor sclerites of *G. tularosae*, *G. nevadensis*, *G. cyprinodontis*, *G. hargisi*, and *G. saratogensis* are essentially identical. Differentiation of the latter 5 species rests primarily in the comparative lengths of their anchors, with *G. cyprinodontis* possessing the largest anchors followed by those of *G. tularosae*, *G. nevadensis*, *G. hargisi*, and *G. saratogensis*, respectively (Table 1). Based on comparative anchor length, *G. tularosae* is most similar to *G. cyprinodontis*. Anchor lengths for *G. tularosae* and *G. cyprinodontis* are significantly different ($t_{23} = -14.66$; $P < 0.001$). Although Mizelle and Kritsky (1967b) indicated that the description of *G. cyprinodontis* was based on 8 specimens, these authors did not provide a standard deviation nor specifically indicate the number of measurements for anchor length that were taken. Thus, our determination of significance was based on the assumption that 8 measurements for anchor

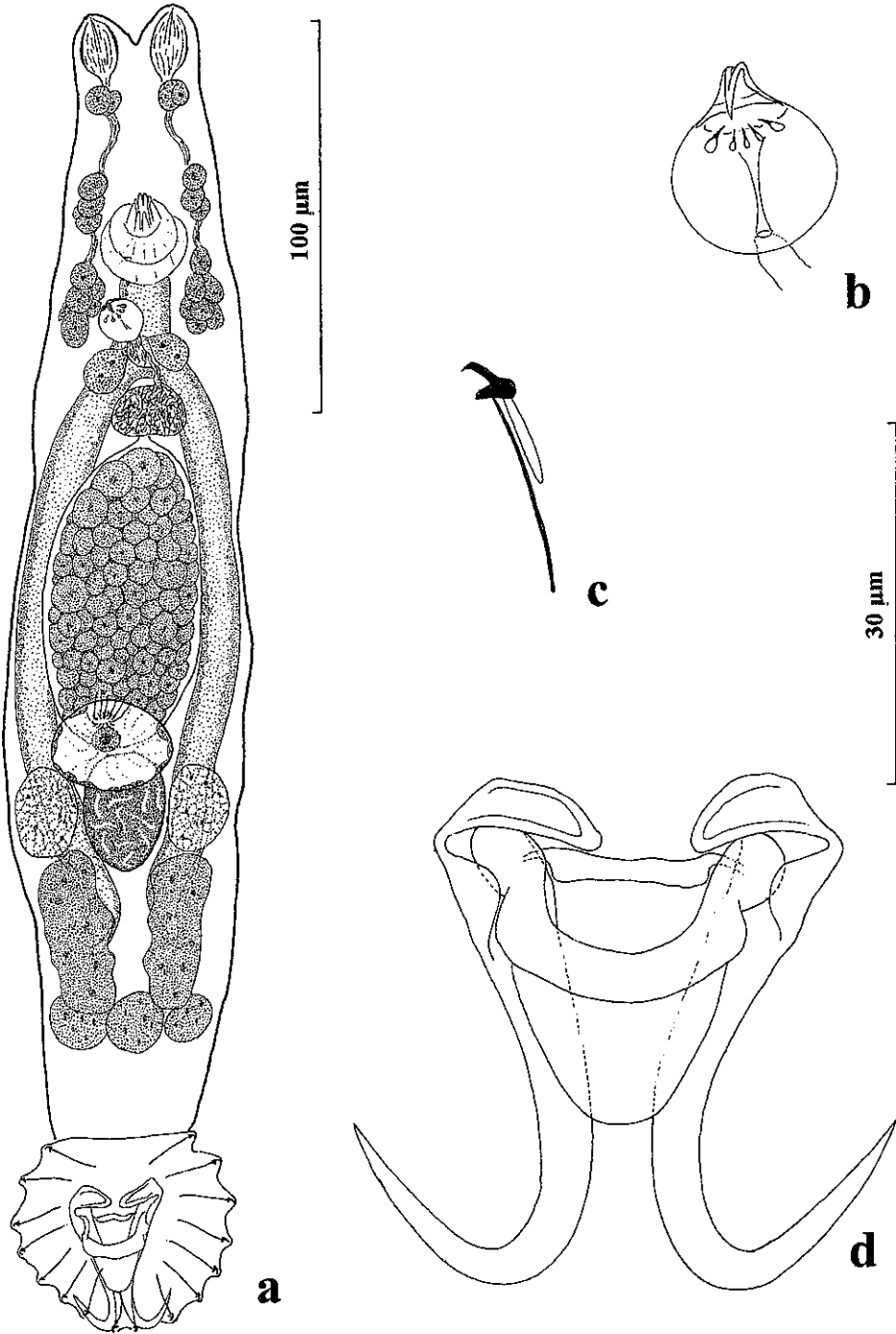


FIG. 1—*Gyrodactylus tularosae* sp. n. 1a) Whole mount (ventral view). 1b) Copulatory organ. 1c) Hook. 1d) Anchor-bar complex. All drawings are to the 30- μ m scale except Fig. 1a (100- μ m scale).

TABLE 1.—Measurements (all in μm) of 5 species of *Cyrodactylus* infesting individuals of *Cyprinodon* species in the United States.¹

	<i>G. tatarosae</i>	<i>G. cyprinodontis</i> ²	<i>G. nevadensis</i> ²	<i>G. hargisi</i> ²	<i>G. saratogensis</i> ²
Body length	366 (322–400; $n = 13$)	353 (322–407)	400 (326–447)	201 (147–266)	202 (158–236)
Body width	72 (62–91; $n = 14$)	72 (53–85)	74 (71–146)	45 (40–49)	59 (46–82)
Haptor length	60 (51–68; $n = 14$)	67 (60–73)	52 (49–55)	41 (33–54)	43 (35–52)
Haptor width	64 (57–72; $n = 14$)	70 (58–84)	57 (55–59)	46 (40–56)	57 (44–74)
Pharynx	24 (21–27; $n = 15$)	23 (20–25)	21 (20–23)	20 (15–26)	17 (12–20)
Copulatory organ	15 (13–17; $n = 12$)	12 (11–13)	11	10 (8–13)	10–11
Anchor	42 (39–45; $n = 17$) ($SD = 1.16$)	49 (45–50)	33 (32–34)	30 (29–31)	26 (24–27)
Deep bar	14 (12–17; $n = 6$)	15 (12–17)	12–13	14–16	10
Superficial bar	20 (18–22; $n = 9$)	23 (21–25)	25–26	18–20	21
Hook	21–22 ($n = 19$)	24 (22–25)	17 (16–18)	20 (17–24) ³	20–21
Hooklet	5 (4–6; $n = 17$)	6–7	5	6–7	4–5
Ovary length	23 (16–27; $n = 9$)	—	—	—	—
Ovary width	27 (22–32; $n = 9$)	—	—	—	—
Testis length	35 (24–47; $n = 14$)	—	—	—	—
Testis width	29 (20–36; $n = 14$)	—	—	—	—

¹ Measurements were obtained using a calibrated flar micrometer and compound microscope, represent straight-line distances between extreme points, and are expressed as the mean followed by the range and number (n) of structures measured in parentheses; body length includes that of the haptor.

² Measurements of these species are from the respective original descriptions (Mizelle and Kritsky, 1967*b*; Williams and Rogers, 1971).

³ Hook length represents addition of measurements of the hooklet and hook shank, which were presented separately by Williams and Rogers (1971).

length were originally obtained, and the standard deviation was conservatively estimated at one-fourth of the reported range for this dimension in *G. cyprinodontis*.

Although anchor length might distinguish species of *Cyrodactylus* infesting *Cyprinodon*, it might not be useful for phylogenetic reconstruction. In fact, the host phylogeny based on mitochondrial DNA (Echelle and Echelle, 1992; Echelle et al., 2005) has *Cyprinodon tularosa* as sister to a clade that includes *C. variegatus* and more distantly related to the Death Valley pupfishes. Thus, one would expect *G. tularosae* to clump with the parasites from sheepshead minnows. Phylogenetic evaluation of the *Cyrodactylus* species associated with *Cyprinodon* using molecular data might be more useful than morphometrics in evaluating historic host-parasite relationships, as well as providing insights to phylogeographic links.

That *Cyrodactylus tularosae* is distinct from *G. cyprinodontis*, *G. hargisi*, *G. nevadensis*, and *G. saratogenis* is further supported by the historical biogeography of its host. The White Sands pupfish has been geographically isolated from *Cyprinodon variegatus* for about 2 million years (Echelle et al., 2005), while the 2 native habitats of *C. tularosa* (Salt Creek and Malpais Spring) within the Tularosa Basin were subsequently isolated from each other 5,000 to 7,000 years ago, a result of a lava flow that disrupted the proto-Salt Creek drainage (see Pittenger and Springer, 1999). Measurable genetic divergence of the respective subpopulations of White Sands pupfish has subsequently occurred (Stockwell et al., 1998). Although gyrodactylids have been observed on *C. tularosa* from Malpais Spring, the prevalence and intensity seems to be relatively low (Stockwell, pers. obser.) Further, experimental work suggests that *G. tularosae* from the Salt Creek population of pupfish easily transfers and reproduces on those from Malpais Spring (Vinje and Stockwell, unpublished). Because gyrodactylids lack means for transmission from one host to another outside of an aquatic environment, the ancestral stock of *G. tularosae* was probably exposed to the same isolation that resulted in the divergent evolution of its host, and concomitant speciation of gyrodactylids would have occurred. Although morphological divergence in these gyrodactylids has been minimal, experimental infestations of *G. tularosae* on *C.*

variegatus suggests that this gyrodactylid is adapted to the White Sands pupfish because the helminth does develop comparatively large infrapopulations on sheepshead minnows (Moen and Stockwell, pers. obser.)

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