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Article

Garra lorestanensis, a new cave fish from the Tigris River drainage with remarks on the subterranean fishes in Iran (Teleostei: Cyprinidae)

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Abstract

The Iranian subterranean species are reviewed, diagnoses are presented for *Garra typhlops* and *Paracobitis smithi*, and a new species is described. *Garra lorestanensis*, new species, from Loven Cave, the Tigris River drainage in Iran, is distinguished from its congeners by the combination of characters, including lacking pigment and eyes, having a well-developed mental disc, two pairs of barbels, and naked body.

Keywords: Loven Cave, *Iranocypris typhlops*, *Nemacheilus smithi*, Middle East.
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Introduction

The Iranian cave fishes, *Garra typhlops* (Bruun and Kaiser, 1948) and *Paracobitis smithi* (Greenwood 1976), are characterized by lacking pigment and eyes. These subterranean fishes are found in Loven Cave, a well-like pool, the natural outlet of a subterranean limestone system of the Zagros Mountains in the Ab-e Sirum or Ab-e Serum Valley near Tang-e Haft railway station in Lorestan Province, south-west Iran (Bruun and Kaiser 1948; Smith 1953; Movagher 1973; Greenwood 1976; Coad 1996; Proudlove 1997; Sargeran et al. 2008). However, *G. typhlops* is occasionally recorded from the Simarreh River drainage (Mahjoorazad and Coad 2009), with no further confirmed record.

Unlike *P. smithi* (originally described as *Nemacheilus smithi*), the *G. typhlops* (originally described as *Iranocypris typhlops*) is well investigated in some aspects by several authors. Bruun and Kaiser (1948) believe the fish is related to the genus *Barbus* which also have two pairs of barbels. Sargeran et al. (2008) reported two types of the fish based on presence or absence of the mental disc, and examined the hypothesis that are these types showing two different taxa or just two forms. Hashemzadeh-Segherloo et al. (2012) and Farashi et al. (2014) found a mean genetic distance, based on DNA evidence, with an affinity to the genus *Garra*, particularly with *Garra rufa* and suggested that the *Iranocypris* can be a synonym of *Garra*. Coad (2015) also pointed out that bearing three rows of pharyngeal teeth and mouth structures of the fish is more related to the genus *Garra*. Jalili and Eagderi (2014) described osteological structures of *G. typhlops* (with mental disc, which is described here as new species) and compared it with the epigean *G. rufa*. Hamidan et al. (2014) transferred this species from the monotypic genus *Iranocypris* to the genus *Garra*. In addition, Farashi et al. (2015) studied the related caves in the region to find and suggest other suitable alternative habitat to introduce the fish to have a more wide distribution. Recently, Sayyadzadeh et al. (2015) reviewed the members of the genus *Garra* in the Persian Gulf and Oman Sea basins, who recognized six epigean species including *G. barreimiae*, *G. longipinnis*, *G. persica*, *G. rossica*, *G. rufa*, and *G. variabilis*; two subterranean species including *G. typhlops* and *G. widdowsoni*, and described a new species from the region as *G. mondica*.

According to the literatures which discussed about presence of two different types of *G. typhlops* in the Loven Cave, we re-examined these fishes with the aim to test whether they represent two distinct taxa. Moreover, as Bruun and Kaiser (1948) described *G. typhlops* based on specimens without mental disc (see the ventral view

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of head in holotype of *I. typhlops* in Bruun and Kaiser, 1948: plate 1, fig. 2), therefore we considered the discless form as the main *G. typhlops*. Comparing the specimens with mental disc with those without disc (*G. typhlops*), based on morphological characters and osteological features, as well as available reports on genetic differentiation (well discussed by Hashemzadeh-Segherloo et al. 2012; Farashi et al. 2014; Sayyadzadeh et al. 2015), it was revealed that the specimens with mental disc represent an unnamed species which is described here as new taxon.

Material and Methods

After anaesthesia, all specimens were fixed in 5% formaldehyde and stored in 70% ethanol. The morphological measurements were made by a dial caliper and recorded to the nearest 0.1 mm. All measurements were made point to point, never by projections. Methods for counts and measurements follow Kottelat and Freyhof (2007). The terminology of the snout morphology and the oromandibular structures follow Stiassny and Getahun (2007), and Nebeshwar and Vishwanath (2013). Standard length (SL) was measured from the tip of the snout to the end of the hypural notch. The length of the caudal peduncle was measured from behind the base of the last anal-fin ray to the end of the hypural notch, at mid-height of the caudal-fin base. The last two branched rays articulating on a single pterygiophore in the dorsal and anal fins are noted as "1½". In the present study, the subterranean *Garra* species from the Euphrates drainage in Iraq, *G. widdowsoni*, is compared based on original descriptions and available literatures (Hamidan et al. 2014; Sayyadzadeh et al. 2015).

For osteological examinations, fish specimens were cleared and stained with alizarin red S and alcian blue according to the protocol of Taylor and van Dyke (1985). Terminology of skeletal elements follows Rojo (1991) and other special publications on Cyprinid osteology (Harrington, 1955; Howes, 1981).

Abbreviations used: SL, standard length. HL, lateral head length. GUIC, Collection of the Ichthyology Museum, Department of Fisheries Sciences, Faculty of Natural Resources, the University of Guilan, Guilan Province, Iran. VMFC, Vatandoust and Mousavi-Sabet Fish Collection, Tehran.

Results

Garra lorestanensis, new species

(Figs. 1, 2, 3)

Holotype: VMFC GL-H, 55 mm SL. Iran, Lorestan prov.: Loven Cave, the Tigris River drainage, the Persian Gulf basin, 33°04'39"N 48°35'33"E, 23 April 2014, H. Mousavi-Sabet, A. Jouladeh-Roudbar & S. Vatandoust.

Paratypes: VMFC GL-P1 to VMFC GL-P3, 3 specimens, 27.2-58.0 mm SL, the same locality as holotype, 17 June 2012, S. Eagderi. GUIC GL-P1 and GUIC GL-P2, 2 specimens, 31.6 and 45.1 mm SL, the same locality as holotype, 17 June 2012, S. Eagderi.

Material for osteological examination: VMFC GL-NT1 to VMFC GL-NT3, none type, 3 specimens, 55.8-74.0 mm SL, the same locality as holotype, 17 June 2012, S. Eagderi.

Diagnosis: *Garra lorestanensis* sp. nov. can be distinguished from its congeners by lacking pigment and eyes (vs. presence of eyes and pigments in all epigean species), having mental disc (vs. absence of mental disc in *G. typhlops*), two pairs of barbels (vs. three pairs of barbels in the sympatric subterranean loach, *Paracobitis smithi*; and one pair of barbels in *G. variabilis*) and naked body (body fully covered by scales or scales restricted to lateral midline in *G. widdowsoni*). *Garra lorestanensis* sp. nov. is also distinguished from the congeners by a K2P nearest-neighbor distance of 3.8% to *G. typhlops* and 4.2% to *G. rufa* (suggested by Hashemzadeh-Segherloo et al. 2012; Farashi et al. 2014; Sayyadzadeh et al. 2015).

Description: See Figures 1-3 for general appearance and Table 1 for morphometric data of holotype and 5 paratypes. Relatively stout species with wide head, moderately compressed laterally, more compressed

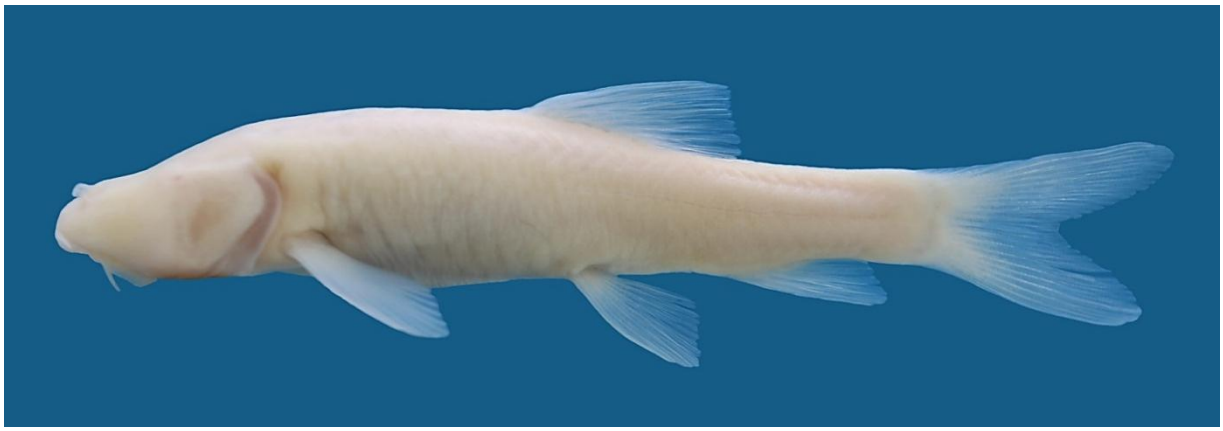

Figure 1. *Garra lorestanensis*, VMFC GL-H, holotype, 55 mm SL; Iran: Loven Cave.

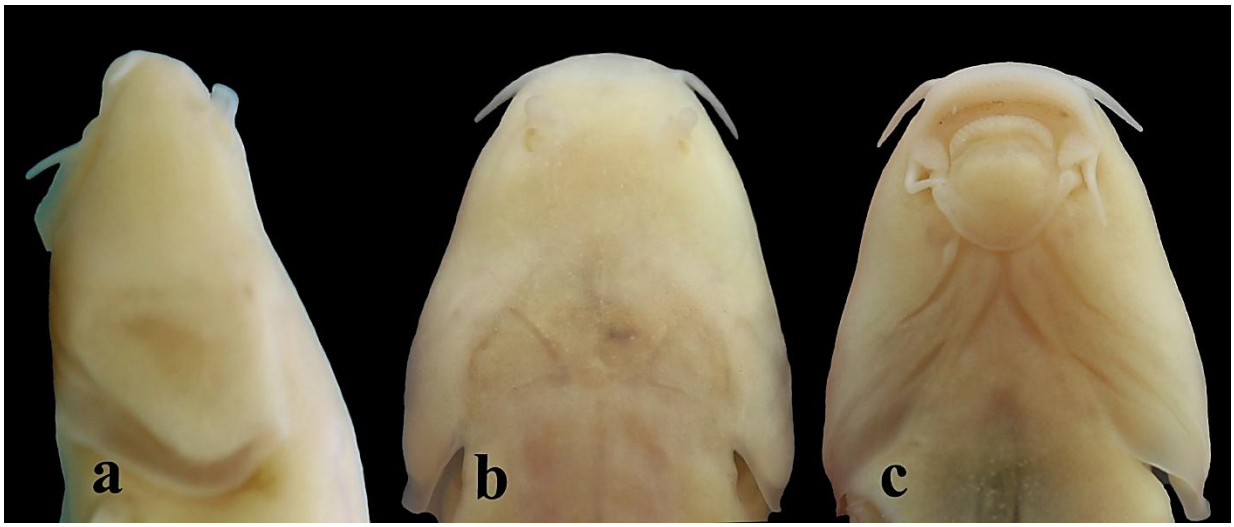

Figure 2. *Garra lorestanensis*, VMFC GL-H, holotype, 55 mm SL; Iran: Loven Cave.

Figure 3. *Garra lorestanensis*, VMFC GL-H, holotype, 55 mm SL; Iran: Loven Cave.

posteriorly especially in caudal peduncle region. Body deepest at or slightly in front of dorsal-fin base, depth decreasing towards caudal-fin base. Greatest body width at or slightly behind of pectoral-fin base, body almost equally wide until dorsal-fin origin. Head relatively large, and deeply depressed. Dorsal head profile rising gently from the tip of snout, slightly convex, sharply continuous with dorsal body profile from about middle between tip of snout and nape to about middle between nape and dorsal-fin origin. Ventral profile slightly

concave in pectoral-pelvic contour, and more or less straight from pelvic to anal-fin origin. Caudal peduncle relatively shallow (caudal peduncle depth 9.5–11.5% SL). Caudal peduncle length 1.4-1.8 times longer than its depth. Lateral line complete, with 28-35 pores. Body naked. Pharyngeal teeth in three rows with a formula of 3.4.5-5.4.3 in three studied specimens. 10-12 total gill rakers on the first branchial arch, in five studied specimens. Snout roundish; transverse lobe with sparsely small tubercles, shallow transverse groove between transverse lobe and proboscis in larger specimens, no obvious transverse groove in small individuals. Proboscis with sparsely small sized tubercles. Proboscis not (commonly in small individuals) or only slightly (in larger specimens) elevated from depressed rostral surface. Scattered small sized tubercles on lateral surface of snout reaching to posterior nostril in larger specimens. Depressed rostral surface normally without tubercles, anterior arm of depressed rostral surface not reaching to base of rostral barbel, separating transverse lobe from lateral surface. Commonly no obvious groove between transverse lobe and lateral surface. No obvious head tubercles in small individuals. Mouth surrounded by two pairs of barbels; rostral barbel anterolaterally located, maxillary barbel at corner of mouth, shorter than rostral barbel. Rostral cap poorly developed, fimbriate, papillate on ventral surface. Upper lip present, upper jaw almost covered by rostral cap. Disc elliptical, longer than wide and narrower than head width through roots of maxillary barbel; papillae on antero-median fold; well-developed groove between antero-median fold and central callous-pad narrow and deep, scattered small sized papillae on latero-posterior flap; surface of central callous pad with sparsely arranged small papillae.

Table 1. Morphometric data of *Garra lorestanensis* sp. nov. (n = 6). H, holotype.

Morphometric character	H	min	max	mean	SD
Standard length (mm)	55	27.2	58		
In percent of standard length					
Head length	23.2	22.8	26.7	25.1	1.3
Body depth at dorsal-fin origin	19.2	17.6	20.8	19.9	1.1
Prepectoral length	22.9	22.3	28.2	25.6	1.9
Predorsal length	50.1	49.9	53.4	50.7	0.9
Postdorsal length	33.4	32.2	35.9	33.8	0.8
Preanal length	73.7	72.4	75.5	74.0	0.7
Prepelvic length	54.9	53.7	56.8	55.4	0.6
Distance between pectoral and pelvic-fin origins	32.1	30.6	33.9	32.2	1.0
Distance between pelvic and anal-fin origins	18.9	18.0	20.2	19.1	0.5
Distance between vent and anal-fin origin	5.4	3.9	5.5	4.9	0.4
Depth of caudal peduncle	11.1	9.5	11.5	10.6	0.9
Length of caudal peduncle	15.2	15.0	17.5	16.0	1.1
Dorsal-fin depth	22.7	19.2	23.3	21.9	1.6
Anal-fin base length	9.8	8.8	10.4	9.5	0.5
Pectoral-fin length	18.5	17.9	20.3	18.9	1.3
Pelvic-fin length	16.0	15.2	17.1	16.3	1.0
In percent of head length					
Head depth at nape	67.6	64.1	67.7	66.5	1.7
Maximum head width	78.8	73.8	82.5	79.6	2.1
Inter-nasal width	30.5	27.1	32.0	29.7	1.9

Dorsal fin with 3 simple and 7½ (4) or 8½ (2) branched rays. Anterior dorsal-fin origin located mid dorsum, or slightly posterior. Pelvic fin with 1 simple and 6-7 branched rays. Pelvic-fin origin behind a vertical of dorsal-fin origin, about a vertical of mid dorsal-fin base. Pectoral fin with 1 simple and 13-14 branched rays. Pectoral

fin reaching approximately 55-60% of distance from pectoral-fin origin to pelvic-fin origin. Anal fin with 5½ branched rays. Margin of dorsal and anal fins straight or slightly concave. Caudal fin with 9+8 branched rays. Caudal fin distinctly forked; tip of lobes pointed.

Coloration: In live specimens (Fig. 3) body is pinkish to red from the blood showing through the skin, although this species is almost entirely unpigmented. The gill filament area is bright red. The skin over the brain semi-transparent, so that the brain can be seen as a dark spot. The intestine filled with a darkish content is visible through a semi-transparent body wall (Fig. 4). In preserved specimens body is yellowish-white. All fins are hyaline (in both live and preserved specimens).

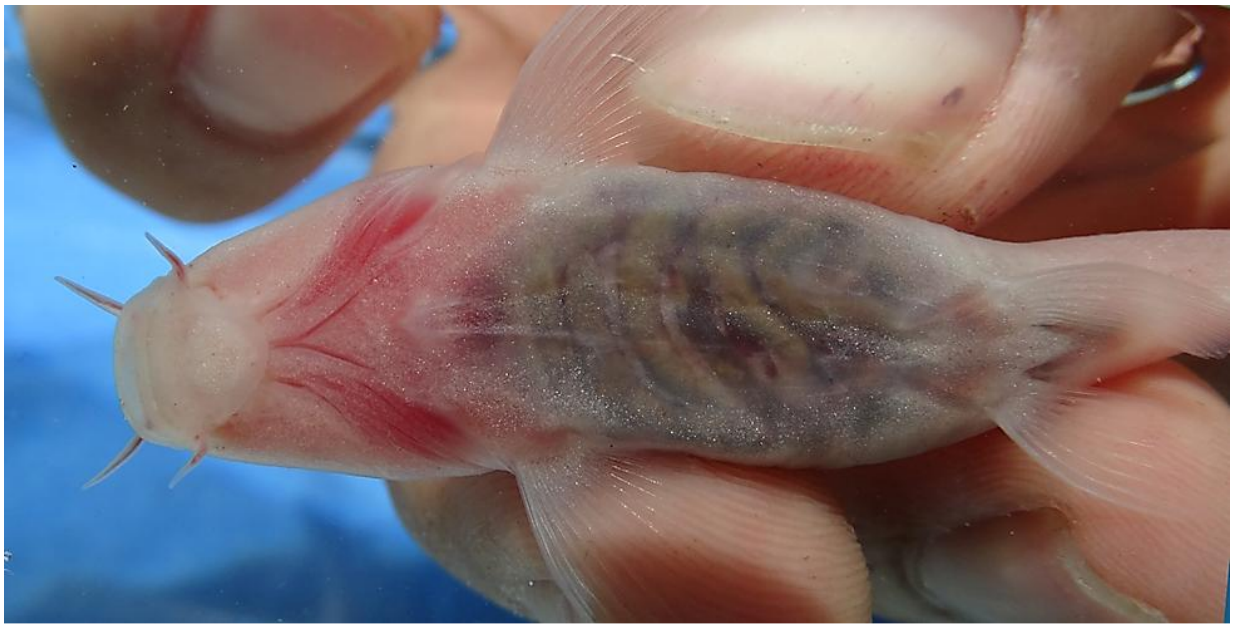

Figure 4. *Garra lorestanensis*, VMFC GL-H, holotype, 55 mm SL; Iran: Loven Cave.

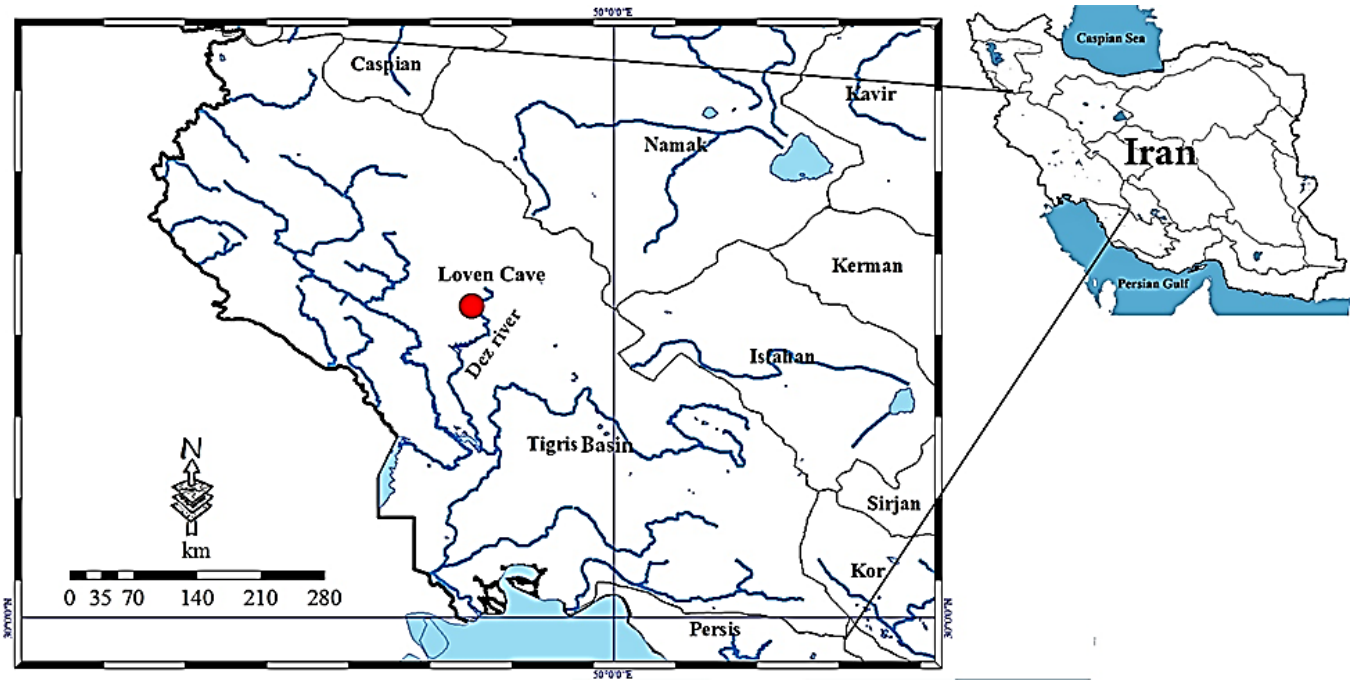

Figure 5. Map of Iranian portion of the Tigris River basin, showing the Loven Cave, the type locality of *Garra lorestanensis* sp. n. in Lorestan Province, south-west Iran.

Distribution: *Garra lorestanensis* sp. nov., is known from the Loven Cave, the natural outlet of a subterranean limestone system of the Zagros Mountains in the Ab-e Sirum or Ab-e Serum Valley near Tang-e Haft railway station, the Tigris River drainage, the Persian Gulf basin, Lorestan Province, southwestern Iran (Fig. 5). There is a small pool (about 2 m across narrowing rapidly inside) and flowing exit stream lower down the gorge (only at the beginning of spring for 40-60 days), about 50 m away from the main locality (Fig. 6). The stream falls over a high waterfall (about 10 m) so the locality is isolated from the local fishes in the main river. Also, there is an additional record for the subterranean cyprinid with mental disc, which is described here as new species (*Garra lorestanensis* sp. nov.), from the Simarreh River drainage, about 131 km away from the type locality in a direct line (Mahjoorazad and Coad 2009).

Figure 6. Loven Cave, type locality of *Garra lorestanensis* sp. n.

Etymology: The species name *lorestanensis*, treated as an adjective, is derived from Lorestan Province which including the Loven Cave, the type locality of the new species.

Remarks: The presence of subterranean cave-fish in Asia was firstly reported by two Danish biologists (Bruun and Kaiser) in 1948 from Loven Cave in the Tigris River drainage in Iran. Bruun and Kaiser came to Iran along with a Danish company which builds the southwestern railway in the country. They found the cavefish in a cave pool near Tange-Haft railway station in Lorestan Province, and after five years they described it, as *Iranocypris typhlops* (*G. typhlops*) (Smith 1953; Mousavi-Sabet 2012; Coad 2015). After twenty-two years, John Anthony Smith (the Oxford University) tried to find the fish, but he could not find the locality. Because he went to a wrong region in Kerman Province, in central Iran, and after three months searching, he could not find the fish. Later, Smith came back to Iran after twenty-six years. In 1976, he succeeded to find the correct locality and catch a different kind of cave fish from the same cave, later the specimens were described as the second cave fish, *Nemacheilus smithi* (*P. smithi*) from Iran by Greenwood (1976) (Smith 1953; Mousavi-Sabet 2012; Coad 2015). Further taxonomic studies on the Iranian subterranean fishes by Sargeran et al. (2008) revealed that there are two different types of the cyprinid species, based on presence or absence of the mental disc. Thereafter, Hashemzadeh-Segherloo et al. (2012) reported that the mean genetic distance between the two forms (with and without mental disc) of *G. typhlops* is significantly higher than generally reported for intraspecific divergence in freshwater fishes. Our present comparative morphological and osteological results between the two mentioned types of the fish supported the previous reports (Sargeran et al. 2008; Hashemzadeh-Segherloo et al. 2012), and suggested that these specimens represent two distinct taxa, which a new species is described here as *Garra lorestanensis* based on specimens with mental disc.

Garra lorestanensis sp. nov. differs from all the epigean members of the genus *Garra* by lacking pigment and eyes (vs. presence of eyes and pigments).Additionally *G. lorestanensis* sp. nov. is distinguished from all the congeners by a K2P nearest-neighbor distance of 3.8% to *G. typhlops* and 4.2% to *G. rufa* (suggested by Hashemzadeh-Segherloo et al. 2012 and Sayyadzadeh et al. 2015).

Figure 7. *Garra typhlops*, VMFC GT01, 41 mm SL; Iran: Loven Cave.

Garra lorestanensis sp. nov. is further distinguished from the subterranean *G. typhlops* (Fig. 7) which is sympatricaly find in the same locality, by having mental disc (vs. absence of mental disc; Fig. 8), longer intestine, and either a single chambered or bipartite swimbladder (vs. shorter intestine and bipartite swimbladder), which was previously reported too by Sargeran et al. (2008). A detailed osteological characteristics of *G. lorestanensis* sp. nov. has been provided by Jalili and Eagderi (2014) identified as *I. typhlops*. *Garra lorestanensis* sp. nov. can be osteologically distinguished from *G. typhlops* by reduced preorbital bones that enclose the infraorbial canal (vs. absent in *G. typhlops*; Fig. 9); the posterior pharyngeal process of the basioccipital is broad directing horizontally with a vertical ridge on its ventral face in *G.*

lorestanensis (vs. this process is directed vertically with lateral ridges in *G. typhlops*); commonly 35 vertebra in *G. lorestanensis* (vs. commonly 34 vertebrae in *G. typhlops*; Fig. 9); wider haemal spine of the fourth fused vertebra of the Weberian apparatus in *G. lorestanensis* (vs. narrow in *G. typhlops*; Fig. 9); small PU2 with a short neural spine in *G. lorestanensis* (vs. PU2 of the caudal skeleton is well-developed with a long neural spine in *G. typhlops*; Fig. 10).

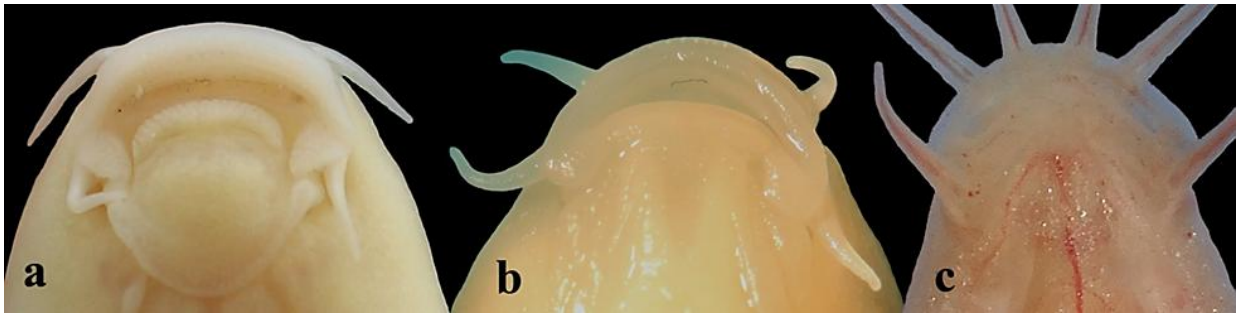

Figure 8. Head (ventral view) of: (a) *Garra lorestanensis*, VMFC GL-H, holotype, 55 mm SL; (b) *Garra typhlops*, VMFC GT01, 41 mm SL; (c) *Paracobitis smithi* VMFC PS1410, 44 mm SL. Iran: Loven Cave.

Figure 9. Cleared and stained skeletal structure of: (a) Lateral view of *Garra lorestanensis*, VMFC GL-NT1, none type, 74.0 mm SL; (b) Ventral view of *Garra lorestanensis*, VMFC GL-NT1, none type, 74.0 mm SL; (c) Lateral view of *Garra typhlops*, VMFC GT08, 42.0 mm SL.

Garra lorestanensis sp. nov. is further distinguished from the subterranean *G. widdowsoni* found in the Euphrates basin, by having a naked body (body fully covered by scales or scales restricted to lateral midline).

Garra lorestanensis sp. nov. is easily distinguished from the sympatric subterranean loach, *Paracobitis smithi* (Fig. 11) by having two pairs of barbels (vs. three pairs; Fig. 8), presence of mental disc (vs. absence), and absence of adipose keel (vs. presence of a weak adipose keel).

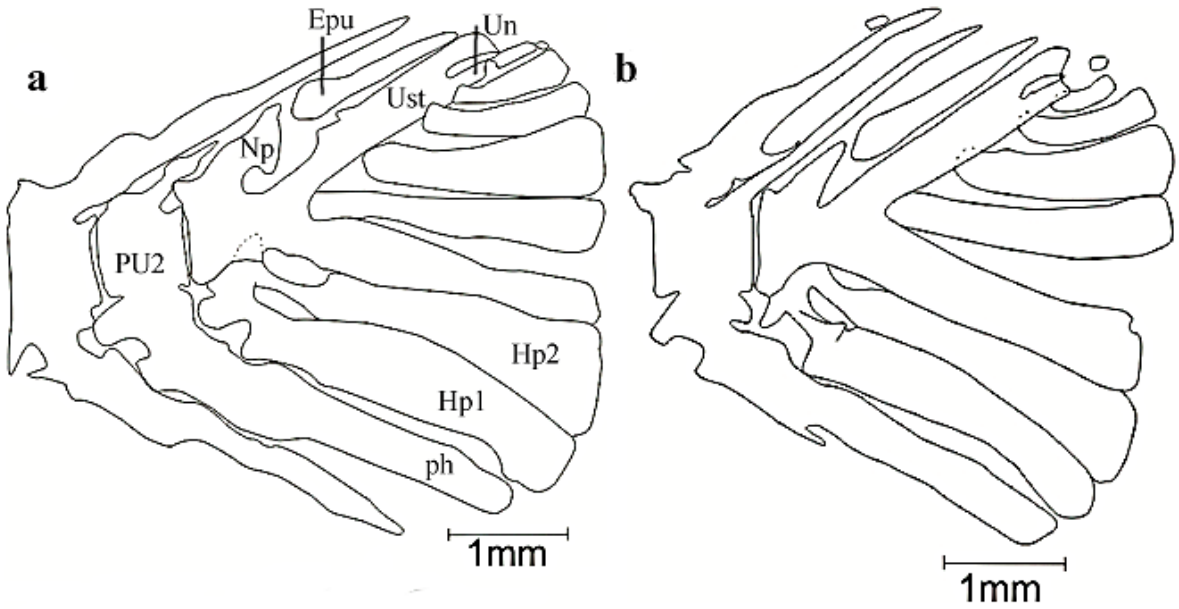

Figure 10. Caudal skeleton of: (a) *Garra lorestanensis* VMFC GL-NT1, none type, 74.0 mm SL; (b) *Garra typhlops* VMFC GT08, 55.8 mm SL. Epu: epural, Hp: hypurals, Np: neural process, Ns: neural spine, Ph: parhypurale, PU2: second Preural centra, Un: uroneural, and Ust: pleurostile.

Figure 11. *Paracobitis smithi*, VMFC PS1410, 44 mm SL; Iran: Loven Cave.

Comparative material

Garra typhlops: VMFC GT01, 1 specimen, 41 mm SL, Iran, Lorestan prov.: Loven Cave, the Tigris River drainage, the Persian Gulf basin, 33°04'39"N 48°35'33"E, 23 April 2014, H. Mousavi-Sabet, A. Jouladeh-Roudbar & S. Vatandoust. VMFC GT02 to VMFC GT07, 6 specimens, 33.2 -64.0 mm SL, Iran, Lorestan prov.: Loven Cave, the Tigris River drainage, the Persian Gulf basin, 33°04'39"N 48°35'33"E, 17 June 2012, S. Eagderi.

Material for osteological examination: VMFC GT08 to VMFC GT10, 3 specimens, 42.0-66.0 mm SL, Iran, Lorestan prov.: Loven Cave, the Tigris River drainage, the Persian Gulf basin, 33°04'39"N 48°35'33"E, 17 June 2012, S. Eagderi.

Paracobitis smithi: VMFC PS1410 to VMFC PS1411, 2 specimens, 41-44 mm SL, Iran, Lorestan Prov.: Loven Cave, the Tigris River drainage, the Persian Gulf basin, 33°04'39"N 48°35'33"E, 23 April 2014, H. Mousavi-Sabet, A. Jouladeh-Roudbar & S. Vatandoust.

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