Re-description and molecular systematics of Paraschistura delvari (Teleostei: Nemacheilidae)

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Abstract. Paraschistura delvari Mousavi-Sabet & Eagderi, 2015 was originally described based on six specimens collected from the Mond River drainage of the Persian Gulf basin. However, based on examination of types and newly collected materials, it is clear that P. delvari was mis-described. Here we re-describe P. delvari based on correct morphological characters and molecular data set and provide its distinguishing characters with the sympatric loaches, P. naumanni, P. nielseni and Oxynoemacheilus persa.

Key words: Nemacheilid loach, COI, Persian Gulf basin, Sympatricity, mis-description.

Introduction

Nemacheilid loaches are the largest group within loaches senso lato that inhabit diverse freshwater habitats of Eurasia and the northernmost parts of Africa (Kottelat 1998, 2001, 2004, Kottelat & Lim 1993, Tan 2006, Kottelat & Freyhof 2007). In Iran they are distributed in almost all drainage basins and some of them are found in sympatry (Esmaeili et al. 2010, 2014, 2017, Freyhof et al. 2014, 2015) as seen in the Mond River, Persis sub-basin of the Persian Gulf. In some localities of the Mond River drainage there are four sympatric nemacheilid species including Paraschistura delvari, P. naumanni, P. nielseni and Oxynoemacheilus persa. Researchers have problems in description of loaches due to their small size and interspecies similarity in meristic data despite interspecies dissimilarity in color patterns. So the systematic position of some genera and species of the loaches is still unsettled and many taxa are artificial assemblages (Kottelat 2004). Here we present some molecular and morphological evidences for mis-description of the newly described species P. delvari based on a pre-approved and more improved molecular character and re-describe it with a taxonomic review of other loaches in the Mond River drainage.

Material and Methods

After anesthesia by 1% clove solution, fishes were fixed in 5% formaldehyde and later stored in 70% or directly fixed in 96% ethanol and were deposited in the Zoological Museum, Collection of Biology Department, Shiraz University (ZM-CBSU). Measurements were made with dial calipers and recorded to 0.1 mm. All measurements were made point to point, never by projections. Methods for counts and measurements follow Kottelat & Freyhof (2007). Standard length (SL) was measured from the tip of the snout to the end of the hypural complex. The length of the caudal peduncle was measured from behind the base of the last anal-fin ray to the end of the hypural complex, at mid-height of the caudal-fin base. The last two branched rays articulating on a single pterygiophore in the dorsal and anal fins were counted as “1½”.

Abbreviations used: SL, standard length; HL, head length; GUC, Collection of the Ichthyology Museum, Department of Fisheries Sciences, Faculty of Natural Resources, the University of Guilan, Guilan province, Iran; NMW, Naturhistorisches Museum, Wien; ZM-CBSU, Zoological Museum of Shiraz University, Collection of Biology Department, Shiraz; VMFC, Vatandoust and Mousavi-Sabet Fish Collection, Tehran.

DNA extraction and PCR: Genomic DNA was extracted using Macherey & Nagel Nucleospin® Tissue kits following the manufacturer’s protocol on an Eppendorf EpMotion® pipetting-robocter with vacuum manifold. The standard vertebrate DNA barcode region of the COI (cytochrome c oxidase subunit I) was amplified using a Mi13 tailed primer cocktail including FishF2_t1 (5’TGTAAACCGACCG CCATGCATCTACTCATACAAATATGCAGC), FishB2_t1 (5’CAG GAAACGACATGACCTCAGCGAAGCACAACTCA), VF2_t1 (5’TGTAACGAGCCGGCCGTCATCAACCAACCACACAAAGAC ATGGCCAG) and FRTd_t1 (5’CAGGAACACGCTATGACACCTCAAGGCTGAACTCA), VF2_t1 (5’TGTAAACCGACCGACCGTCATCAACCAACCACACAAAGAC ATGGCCAG) and FRTd_t1 (5’CAGGAACACGCTATGACACCTCA AGGCTGAACTCA) (Ivanova et al. 2007). Sequencing of the ExoSAP-IT (USB) purified PCR product in both directions was conducted at Macrogen Europe Laboratories with forward sequencing primer M13F (5’TGTAAAACGACGGCCAGT) and reverse sequencing primer M33R-prUC (5’CAGGAAACGCTATGACACCTCAAGGCTGAACTCA) and FRTd_t1 (5’CAGGAACACGCTATGACACCTCA AGGCTGAACTCA) (Ivanova et al. 2007). Sequencing primer M13F (5’TGTAAAACGACGGCCAGT) and reverse sequencing primer M33R-prUC (5’CAGGAAACGCTATGACACCTCA AGGCTGAACTCA) and FRTd_t1 (5’CAGGAACACGCTATGACACCTCA AGGCTGAACTCA) (Ivanova et al. 2007). Sequencing primer M13F (5’TGTAAAACGACGGCCAGT) and reverse sequencing primer M33R-prUC (5’CAGGAAACGCTATGACACCTCA AGGCTGAACTCA) and FRTd_t1 (5’CAGGAACACGCTATGACACCTCA AGGCTGAACTCA) (Ivanova et al. 2007).

Molecular data analysis: We used 60 sequences from Freyhof et al. (2015) and Sayyadzadeh et al. (2016) and an additional 15 sequences in this study. Data processing and sequence assembly was done in BioEdit (Hall 1999) and the CLUSTALW algorithm (Higgins and Sharp 1988) was used to create a DNA sequence alignment. Medtest (Posada & Crandall 1998), implemented in the MEGA 7 software (Tamura et al. 2011), was used to determine the most appropriate sequence evolution model for the given data, treating gaps and missing data with the partial deletion option under 95% site coverage cutoff. We generated maximum likelihood phylogenetic trees with 10,000 bootstrap replicates in RaxML software 7.2.5 (Stamatakis 2006) under the GTR+G+I model of nucleotide substitution, with CAT approximation of rate heterogeneity and fast bootstrap to explore species phylogenetic affinities. Bayesian analyses of nucleotide sequences were run with the parallel version of MrBayes 3.1.2 (Ronquist & Huelsenbeck 2003) on a Linux cluster with one processor assigned to each Markov chain under the most generalizing model (GTR+G+I) because overparameterization apparently does not negatively affect Bayesian analyses (Huelsenbeck & Ronquist 2004).

Each Bayesian analysis comprised two simultaneous runs of four Metropolis-coupled Markov-chains at the default temperature (0.2). Analyses were terminated after the chains converged significantly, as indicated by the average standard deviation of split frequencies <0.01.

Bayesian inference of phylogeny was conducted for 6,000,000 generations. Seven hundred bootstrap replicates were used as ML branch support values. The posterior probabilities equal/higher than 0.95 and bootstrap supports equal/higher than 70% were considered as strong support values (Toussaint et al. 2015). MEGA 7 was also used to compute intra-clade and inter-clade K2P genetic distances. As appropriate out-group to root the constructed phylogenetic hypothesis we included the Cobitis avicoenae KP050525.
Results

We compared 15 COI barcode sequences of loaches from the Mond River drainage with 60 sequences from our last studies (mentioned above). Both the ML and BI phylogenetic trees were mostly similar in their topology, hence here only the BI tree including the posterior probability values from the Maximum Likelihood phylogram is presented (Fig. 1).

Figure 1. Maximum Likelihood and Bayesian phylogeny reconstructed based on 617 bp of COI 5’ end. The values beside the branches before and after a slash are BI posterior and ML bootstrap probability values, respectively.
Table 1. Diagnostic nucleotide substitutions found in mtDNA COI of loach species in the Mond River drainage (O. persa, n=12; P. delvarii, n=6; P. naumanni, n=6; P. nielseni, n=13).

<table>
<thead>
<tr>
<th>Nucleotide position</th>
<th>O. persa</th>
<th>P. delvarii</th>
<th>P. naumanni</th>
<th>P. nielseni</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>C</td>
<td>A</td>
<td>G</td>
<td>T</td>
</tr>
<tr>
<td>1</td>
<td>T</td>
<td>C</td>
<td>C</td>
<td>G</td>
</tr>
<tr>
<td>2</td>
<td>T</td>
<td>C</td>
<td>C</td>
<td>T</td>
</tr>
<tr>
<td>3</td>
<td>T</td>
<td>T</td>
<td>C</td>
<td>T</td>
</tr>
<tr>
<td>4</td>
<td>T</td>
<td>A</td>
<td>T</td>
<td>A</td>
</tr>
<tr>
<td>5</td>
<td>T</td>
<td>A</td>
<td>T</td>
<td>A</td>
</tr>
<tr>
<td>6</td>
<td>T</td>
<td>A</td>
<td>T</td>
<td>A</td>
</tr>
<tr>
<td>7</td>
<td>T</td>
<td>A</td>
<td>T</td>
<td>A</td>
</tr>
</tbody>
</table>

Table 2. Estimates of evolutionary divergence (%) over sequence pairs between species found in the COI barcode region of loach species studied in the Mond River drainage.

<table>
<thead>
<tr>
<th>Species</th>
<th>O. persa</th>
<th>P. delvarii</th>
<th>P. naumanni</th>
<th>P. nielseni</th>
</tr>
</thead>
<tbody>
<tr>
<td>O. persa</td>
<td>20</td>
<td>17</td>
<td>18</td>
<td>13</td>
</tr>
<tr>
<td>P. delvarii</td>
<td>17</td>
<td>7</td>
<td>13</td>
<td>11</td>
</tr>
</tbody>
</table>

Table 1 lists the diagnostic nucleotide substitutions and Table 2 lists the average estimates of the evolutionary divergence in the 617 base pairs long mtDNA COI barcode region between the loach species from the Mond River drainage.

Key to Nemacheilid loach fishes of the Mond River drainage:

1a. No prominent black spot at base of unbranched dorsal fin-rays and sometimes on first and second branched rays; a dark brown blotch or saddle present at dorsal-fin origin, extending on dorsal fin; lateral line complete; 8½ branched dorsal fin rays.

1b. Prominent black spot at base of unbranched dorsal fin-rays and sometimes on first and second branched rays; lateral line incomplete; 6½ - 7½ branched dorsal fin rays.

2a. Male with suborbital groove.

2b. Male without suborbital groove.

3a. Distance between anus and the end tip of pelvic fin 7.2 - 10.5% SL, no distinct bars on the flank especially in small specimens.

3b. Distance between anus and the end tip of pelvic fin 4.2 - 7.1% SL, distinct bars on the flank.

Re-description of Paraschistura delvarii

Mousavi-Sabet & Eagderi, 2015 (Figs 2-7)

Material examined: VMFC PSD1-H: (Holotype), 38.0 mm SL; Iran: Fars prov.: upstream of Mond River, Mond River drainage, the Persian Gulf basin, 29°40’22” N, 52°08’57” E, 13 August 2013, H. Mousavi-Sabet & S. Eagderi. – VMFC PSD1-P1 to VMFC PSD1-P5: 5 specimens, 27.2 – 42.1 mm SL, same data as holotype. – GUIC PSD1-P6 and GUIC PSD1-P7, 2 specimens, 31.1 – 35.2 mm SL, same data as holotype.

– ZM-CBSU J3304, 9, 36.9-50.7 mm SL; Iran: Fars prov.: Firuzabad city, near Tangab dam, Qareh Aqaj River, a tributary of Mond River drainage, Persis Basin, 28°57'48.11"N 52°33'15.54"E; G. Sayyadzadeh, A. Khajeh Panah, E. Izadi and A. Danesh Nia, 18 Oct 2014. – ZM-CBSU H20358, 1, 38.2 mm SL; Iran: Fars prov.: Shiraz, Khanzenian, Qareh Aqaj River, Mond River drainage, 29°41'13.0"N, 52°02'58.6"E; G. Sayyadzadeh, H. Darvish Nia, 24 July 2014.

Material used in the molecular genetic analysis: ZM-CBSU M1308, M1310; Iran: Fars prov.: Firuzabad, near Tangab dam, Qareh Aqaj River, Mond River drainage, 28°57'48.11"N 52°33'15.54"E (GenBank accession number: KY808477, KY808478). – ZM-CBSU M1315; Iran: Fars prov.: Shiraz, Khanzenian, Qareh Aqaj River, Mond River drainage, 29°41'13.0"N, 52°02'58.6"E (GenBank accession number: KY808479).

Diagnosis: Paraschistura delvarii is distinguished from all other species of Paraschistura in Iran except P. naumanni by having the pelvic-fin origin situated below or slightly in front of the vertical of the dorsal-fin origin (vs. behind dorsal-fin origin in other species). Paraschistura delvarii is distinguished from P. naumanni by having; distance between anus and the end tip of pelvic fin 7.2-10.5% SL (vs. 4.2-7.1% SL) and no distinct bar on the flank especially in small specimens (vs. presence). It is distinguished from P. abdolii, P. cristata, P. kessleri and P. turcmenica by having the body fully covered by scales (vs. scales absent on the back and on the flank in front of the dorsal-fin origin in P. abdolii and P. cristata and scales completely absent in P. kessleri and P. turcmenica). Paraschistura delvarii is distinguished from P. alta by having a deeply forked caudal fin (vs. emarginated) and from P. cristata by the absence of a long dorsal adipose crest (vs. presence) and an incomplete lateral line (vs. complete). Paraschistura delvarii is distinguished from P. bampurensis and P. hormuzensis by the absence of a suborbital flap in males (vs. presence) and from P. nielseni by the absence of a suborbital groove in males (vs. presence).

Among loach fishes studied for molecular characters in the Mond River drainage, P. delvarii is characterized by 9 fixed diagnostic nucleotide substitutions in the mtDNA COI barcode region studied (Table 1) and a K2P nearest-
neighbour distance of 7% to *P. naumanni* (Table 2).

**Description:** For general appearance see Figures 2-7; morphometric data are provided in Table 3. Small sized, moderately elongate species with short head. Body deepest at or slightly in front of dorsal-fin origin, depth moderately decreasing towards caudal-fin base. No hump at nape. Greatest body width at pectoral-fin base. Section of head roundish, flattened on ventral surface. Caudal peduncle compressed laterally, 1.2-1.9 (mean 1.5) times longer than deep. Pectoral fin reaching approximately 43-55% of distance from pectoral-fin origin to pelvic-fin origin. Pelvic axillary lobe ovoid, fully attached to body or absent. Pelvic-fin origin below or slightly in front of vertical of dorsal-fin origin. Pelvic fin reaching to a point about 2.5-3.0 eye diameter
Table 3. Morphometric data of *Paraschistura delvarii* ZM-CBSU J3304, n=9.

<table>
<thead>
<tr>
<th>Standard length (mm)</th>
<th>Min</th>
<th>Max</th>
<th>Mean</th>
<th>SD</th>
</tr>
</thead>
<tbody>
<tr>
<td>In percent of standard length</td>
<td>36.8</td>
<td>50.6</td>
<td>52.4</td>
<td>1.0</td>
</tr>
<tr>
<td>Head length</td>
<td>21.2</td>
<td>23.3</td>
<td>22.2</td>
<td>0.7</td>
</tr>
<tr>
<td>Body depth at dorsal-fin origin</td>
<td>13.1</td>
<td>16.4</td>
<td>14.5</td>
<td>1.1</td>
</tr>
<tr>
<td>Body width at dorsal-fin origin</td>
<td>9.9</td>
<td>13.0</td>
<td>11.2</td>
<td>1.0</td>
</tr>
<tr>
<td>Predorsal length</td>
<td>52.2</td>
<td>55.6</td>
<td>54.5</td>
<td>1.2</td>
</tr>
<tr>
<td>Postdorsal length</td>
<td>35.3</td>
<td>40.4</td>
<td>37.2</td>
<td>1.5</td>
</tr>
<tr>
<td>Prepelvic length</td>
<td>52.6</td>
<td>55.8</td>
<td>54.2</td>
<td>1.3</td>
</tr>
<tr>
<td>Preanal length</td>
<td>73.0</td>
<td>81.7</td>
<td>79.7</td>
<td>2.6</td>
</tr>
<tr>
<td>Distance between pectoral and pelvic-fin origins</td>
<td>30.2</td>
<td>33.1</td>
<td>31.4</td>
<td>1.0</td>
</tr>
<tr>
<td>Distance between pelvic and anal-fin origins</td>
<td>23.4</td>
<td>26.9</td>
<td>25.4</td>
<td>1.2</td>
</tr>
<tr>
<td>Depth of caudal peduncle</td>
<td>8.7</td>
<td>11.2</td>
<td>10.2</td>
<td>0.7</td>
</tr>
<tr>
<td>Length of caudal peduncle</td>
<td>13.7</td>
<td>19.1</td>
<td>15.6</td>
<td>1.7</td>
</tr>
<tr>
<td>Dorsal-fin depth</td>
<td>11.3</td>
<td>17.8</td>
<td>14.4</td>
<td>1.7</td>
</tr>
<tr>
<td>Pectoral fin length</td>
<td>13.3</td>
<td>16.7</td>
<td>15.4</td>
<td>1.0</td>
</tr>
<tr>
<td>Pelvic fin length</td>
<td>12.6</td>
<td>14.7</td>
<td>13.6</td>
<td>0.6</td>
</tr>
</tbody>
</table>

| In percent of head length | 51.6 | 63.0 | 56.2 | 3.6  |
| Head depth at nape       | 44.3 | 53.5 | 49.5 | 2.9  |
| Head depth at eye        | 37.3 | 41.5 | 39.0 | 1.7  |
| Snout length             | 14.1 | 16.3 | 15.1 | 0.8  |
| Eye diameter             | 46.9 | 51.1 | 49.5 | 1.4  |
| Postorbital distance     | 59.6 | 70.0 | 64.7 | 3.3  |
| Maximum head width       | 29.1 | 32.8 | 31.1 | 1.3  |

in front of anus. Anal-fin origin about one eye diameter behind anus. Anal-fin origin at vertical or behind middle between dorsal- and caudal-fin origins. Very short and shallow dorsal and ventral adipose keels on caudal peduncle in some individuals. Margin of dorsal fin straight or slightly convex. Caudal fin emarginated. Largest known specimen 51 mm SL. Dorsal fin with 6-7½ branched rays. Anal fin with 5½ branched rays. Caudal fin with 15-16 branched rays. Pectoral fin with 10-11 and pelvic fin with 6-7 branched rays. Back and flank covered by embedded scales especially in post-dorsal part and few in pre-dorsal. Lateral line incomplete, reaching to a point slightly in front of dorsal-fin origin or below dorsal-fin base. Anterior nostril opening at the end of a low, pointed and flap-like tube. Nostrils separated by a narrow space, anterior nostril slightly overlapping (or not in some individuals) posterior nostril when folded backwards. No suborbital flap or groove. Mouth small, strongly arched (Figs 3, 5). Lips moderately thick, with many deep furrows. A median interruption in lower lip. Upper lip with median incision. Barbels short, inner rostral barbel not reaching to base of maxillary barbel; outer one reaching to base of maxillary barbel. Maxillary barbel reaching vertical of anterior part or middle eye.

**Coloration:** Body is cream yellow with dark to pale brown marbled pattern on flank and back. There are a series of connected blotches on flank, most obvious as bars post-dorsally. Post-dorsal with 3-7½ branched dark-brown bars irregularly shaped and set (indistinct bars in small individuals) as wide as or wider than interspaces. An obvious black spot at the base of unbranched and first branched dorsal-fin rays. An obvious black bar at caudal-fin base. Upper part of head, opercula and snout covered by dark-brown small blotches, cheeks pale with few dark-brown dots on top. Dorsal and caudal fins with black spots and stripes on rays, pectoral fin with few dark-brown or black spots, anal and pelvic fins hyaline.

**Distribution:** *Paraschistura delvarii* is known from the Qareh Aqaj River, a tributary of the Mond River drainage, Persis sub-basin which drains into the Persian Gulf.

*Paraschistura naumanni* Freyhof, Sayyadzadeh, Esmaeili, Geiger 2015 (Figs 8-9)

**Material examined:** See Freyhof et al. (2015).

**Material used in the molecular genetic analysis:** ZM-CBSU M1309; Iran: Fars prov.: Firuzabad, near Tangab dam, Qareh Aqaj River, Mond River drainage, 28°57‘48.11”N 52°33‘15.54”E (GenBank accession number: KY808480).

Figure 8. *Paraschistura naumanni*, ZM-CBSU J2941, holotype, 35 mm SL; Iran: Golabi spring.

Figure 9. *Paraschistura naumanni*, paratypes; caudal region, Golabi spring; a, ZM-CBSU J2948, 48 mm SL; b, ZM-CBSU J2944, 38 mm SL; c, ZM-CBSU J2941, 35 mm SL.

**Diagnosis:** *Paraschistura naumanni* is distinguished from the other loach species in the Mond River drainage except *P. delvarii* by having the pelvic-fin origin situated below or slightly in front of the vertical of the dorsal-fin origin (vs. behind dorsal-fin origin). *Paraschistura naumanni* is distinguished from *P. delvarii* by having: distance between anus...
and the end tip of pelvic fin 4.2-7.1% SL (vs. 7.2-10.5% SL) and having distinct bars on the flank (vs. absence). It is distinguished from P. nielseni and O. persa by the absence of a suborbital groove in males (vs. presence).

Among loach fishes studied for molecular characters in the Mond River drainage, P. naumanni is characterized by four fixed diagnostic nucleotide substitutions in the mtDNA COI barcode region studied (Table 1) and a K2P nearest-neighbour distance of 7% to P. delvarii (Table 2).

**Paraschistura nielseni** (Nalbant & Bianco, 1998) (Fig. 10)

**Material examined:** ZM-CBSU H2039, 12, 23-46.8 mm SL; Iran: Fars prov.: Shiraz, Khanzenian, Qareh Aqaj River, Mond River drainage, 29°41′13.0″N, 52°02′58.6″E; G. Sayyadzadeh, H. Darvish Nia, 24 July 2014. —ZM-CBSU H2073, 15, 34.5-53.9 mm SL; Iran: Fars prov.: Shiraz, Qareh Aqaj River, Mond River drainage, 29°10′55.10″N 52°41′32.80″E; G. Sayyadzadeh, H. Darvish Nia, M. Masoudi, 22 July 2014.

**Remarks:** In a review of the genus Paraschistura in Iran, it was mentioned that there is an undescribed species in the Mond River drainage without giving a description (Freyhof et al. 2015), so Mousavi-Sabet & Eigderi (2016) referred to this molecular data and described it as P. delvarii. But we could not match our data with the description of P. delvarii; for example, they gave the presence of sub-orbital groove in male (vs. absence in our materials). We checked all of the type specimens in Gilan University, none of them have a sub-orbital groove. It seems that Mousavi-Sabet & Eigderi (2016) confused P. delvarii with P. nielseni or even with O. persa as all of them are sympatric. Our data shown that these three loach species, with P. naumanni, are found in the same localities in the Mond River (Figs 13-14).

**Orthrias farsicus** Nalbant and Bianco, 1998

**Material used in the molecular genetic analysis:** ZM-CBSU M1543; Iran: Fars prov.: Shiraz, Khanzenian, Qareh Aqaj River, Mond River drainage, 29°41′13.0″N, 52°02′58.6″E (GenBank accession number: KY808481). —ZM-CBSU M1312, M1313, M1314, M1330; Iran: Fars prov.: Kavar, Qareh Aqaj River, Mond River drainage, 29°10′55.10″N 52°41′32.80″E; G. Sayyadzadeh, H. Darvish Nia, M. Masoudi, 23 July 2014. —ZM-CBSU H1852, 17, 34-56mm SL; Fars prov.: Ghadamgah spring at Dorudzan, Kor basin, 30°14′19.65″N 52°22′23.3″E. —ZM-CBSU H1869, 98, 24-71mm SL; Iran: Fars prov.: Archin Qant at Safashahr, Kor basin, 30°36′16.9″N 52°56′40.1″E.

**Material used in the molecular genetic analysis:** ZM-CBSU M1543; Iran: Fars prov.: Shiraz, Khanzenian, Qareh Aqaj River, Mond River drainage, 29°41′13.0″N, 52°02′58.6″E (GenBank accession number: KY808481). —ZM-CBSU M1312, M1313, M1314, M1330; Iran: Fars prov.: Kavar, Qareh Aqaj River, Mond River drainage, 29°10′55.10″N 52°41′32.80″E; G. Sayyadzadeh, H. Darvish Nia, M. Masoudi, 23 July 2014. —ZM-CBSU H1852, 17, 34-56mm SL; Fars prov.: Ghadamgah spring at Dorudzan, Kor basin, 30°14′19.65″N 52°22′23.3″E. —ZM-CBSU H1869, 98, 24-71mm SL; Iran: Fars prov.: Archin Qant at Safashahr, Kor basin, 30°36′16.9″N 52°56′40.1″E.

**Diagnosis:** Paraschistura nielseni is distinguished from the other loach species in the Mond River drainage except O. persa by having a suborbital groove in males (vs. absence). It is distinguished from O. persa by having a prominent black spot at base of unbranched dorsal-fin rays and sometimes on first and second branched rays (vs. absence); lateral line incomplete (vs. complete) and 6½-7½ branched dorsal fin rays (vs. 8¼). For distinguishing from the other congener see Freyhof et al. (2015).

Among loach fishes studied for molecular characters in the Mond River drainage, P. nielseni is characterized by 16 fixed diagnostic nucleotide substitutions in the mtDNA COI barcode region studied (Table 1) and a K2P nearest-neighbour distance of 11% to P. naumanni (Table 2).

**Remarks:** In a review of the genus Paraschistura in Iran, it
accession number: KY808487, KY808488, KY808489). — ZM-CBSU M1669; Iran: Fars province: Safashahr, Archin Qanat, Kor River basin, 30°36’16.9’’N, 52°56’40.1’’E (GenBank accession number: KY808490). — ZM-CBSU M1671; Iran: Fars province: Beiza, Sarab spring, Kor River basin, 29°55’28.95’’N, 52°26’42.77’’E (GenBank accession number: KY808491).

**Diagnosis:** *Oxynoemacheilus persa* belongs to a group of *Oxynoemacheilus* having a suborbital groove in males and elongated body. It is distinguished from the other loach species in the Mond River drainage by its prominent black spot at base of unbranched dorsal fin-rays and sometimes on first and second branched rays (vs. presence), lateral line complete (vs. incomplete) and 8½ branched dorsal fin rays (vs. 6½ -7½). It is also diagnosed from the other loach species in the Mond River drainage by 40 fixed (Table 1) diagnostic nucleotide substitutions in the mtDNA COI barcode region, and a K2P nearest-neighbour distance of 17% to *P. natunani* (Table 2).

**Remarks:** According to Heckel (1847) the type locality of *Cobitis Persa* is "Quellen um Persepolis". Kähsbauer (1964) reports a syntype of this species in the Naturhistorisches Museum Wien under NMW 48567 (Fig. 12). This specimen is in poor condition and not readily comparable to fresh material (Coad 2017).

*Oxynoemacheilus farsicus* (Nalbant & Bianco 1998) is the other loach species which has been described from the Kor River basin, but Freyhof et al. (2011) couldn’t find any differences between it and *O. persa*, so they considered it as a synonym of *O. persa*. We also didn’t find any diagnostic character to distinguish populations oxynoemacheilid fishes from the Kor River, Lake Maharlu and Mond River basins (see Figure 1, *O. persa* clade). Therefore, we treat *O. farsicus* as a synonym of *O. persa*.

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**References**


