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Oxynoemacheilus fatmae, a new species from the Güzelhisar Stream in the Aegean Sea basin, Türkiye (Teleostei: Nemacheilidae)

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Abstract

Oxynoemacheilus fatmae, a new species, is found in the Güzelhisar Stream in the northern Aegean Sea basin. It is differentiated from all other species of *Oxynoemacheilus* in the northern Aegean Sea and adjacent basins by having four to eight irregularly shaped narrow black bars on the posterior part of flank, and anterior parts of the flank with a marbled pattern. *O. fatmae* is differentiated from the closest species *Oxynoemacheilus theophilii* by having 14 fixed diagnostic nucleotide substitution sites, and the pair-wise genetic distance is 2.22%. It further differs from *O. theophilii* by having a slenderer body (body at dorsal-fin origin: 15%–17% standard length [SL] vs. 17%–18%), a slenderer caudal peduncle (10%–12% SL vs. 12%–13%), a more forked caudal fin (length of middle caudal-fin lope: 16%–19% SL vs. 19%–23%), and the absence the dorsal and ventral adipose crests on the caudal peduncle behind the vertical of the posterior anal-fin base (vs. present). Three species delimitation tests (assemble species by automatic partitioning (ASAP), automatic barcode gap discovery (ABGD) and generalized mixed yule-coalescent (GMYC)) and phylogenetic analyses reinforce the validity of *O. fatmae* as a distinct species.

KEYWORDS

freshwater fish, loach, species delimitation tests, systematics

1 | INTRODUCTION

The genus *Oxynoemacheilus* is distributed in the Eastern Mediterranean, the southern Caucasus, Anatolia, Mesopotamia, and Central Iran (Kottelat, 2012). According to recent studies, this genus contains 66 valid species; 44 of them are found in Anatolia, and 36 of them are endemic in Türkiye (Bektaş et al., 2022; Çiçek et al., 2023; Turan et al., 2019; Turan, Aksu, et al., 2023; Turan, Bayçelebi, et al., 2023).

In the Aegean region, the genus *Oxynoemacheilus* does not show high diversity compared to other regions of Türkiye. A total of three valid species have been described or reported from this region. Erk'akan et al. (2007) and Erk'akan (2012) described *Barbatula germencicus*, *Barbatula mesudae*, and *Barbatula cinicus* from Büyük Menderes River. Later, Yoğurtçuoğlu et al. (2022) reported *Oxynoemacheilus mesudae* and *Oxynoemacheilus cinicus* as synonyms of *Oxynoemacheilus germencicus*. In addition to reporting *Oxynoemacheilus theophilii* from Bakır Stream, the same researchers described *Oxynoemacheilus eliasi* species from Tahtalı Reservoir. They also recorded *O. eliasi* from the Gediz and Küçük Menderes Rivers and *O. germencicus* from the Gediz River. Apart from these, an undescribed species, herein described as *Oxynoemacheilus fatmae*, was found in the Güzelhisar Stream.

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2 | MATERIALS AND METHODS

Twenty-three fish samples were collected from Güzelhisar Creek in the northern Aegean Region in October 2023. Comparative materials used in the study are from FFR, Zoology Museum, Faculty of Fisheries, Recep Tayyip Erdoğan University, Rize; and IFC-ESUF, Inland Fishes Collection, Faculty of Eğirdir Fisheries, Isparta University of Applied Sciences, Isparta. The fish samplings and experiments were approved by Recep Tayyip Erdoğan University Local Ethics Committee for Animal Experiments in the Republic of Türkiye (permit reference number 2020/4). After anaesthesia, sample fixation was initially carried out in 5% formaldehyde, followed by immersion in 70% ethanol when feasible. Alternatively, some samples were directly fixed in absolute ethanol. Measurements were performed using a dial caliper set precisely to 0.1 mm, adhering to stringent point-to-point measurement procedures outlined in the guidelines of Kottelat and Freyhof (2007).

2.1 | DNA extraction, amplification, sequencing, and molecular analysis

DNA was isolated from five specimens' fin clips via Hibrigen Genomic DNA extraction kits. DNA quality was checked on agarose gel electrophoresis. The mitochondrial genome cytochrome oxidase subunit I (*COI*) barcode region (626 bp) was amplified with the FishF1 and FishR2 (Ward et al., 2005) primer pair. PCR conditions were given in Turan, Aksu, and Kalaycı (2023), except for annealing temperature. The annealing temperature was used at 58°C for 45 s. The PCR products were visualized with a gel documentation system, and eligible PCR products were sent to Macrogen Europa Inc. (Amsterdam, Netherlands) for purification and Sanger sequencing.

We have used the five newly generated and 27 DNA barcodes from The National Center for Biotechnology Information (NCBI GenBank) (Bektaş et al., 2022; Geiger et al., 2014; Turan, Aksu, et al., 2023; Turan, Bayçelebi, et al., 2023; Yoğurtçuoğlu et al., 2022) for the molecular data analysis. *Oxynoemacheilus bureschi* (KJ553692) was added to the analysis as an out-group taxon. Multiple sequence alignment was performed using Clustal W options (Thompson et al., 1994) in Bioedit v7.2.5 software (Hall, 1999). Sequences were submitted to NCBI GenBank with accession numbers OR948615–OR948619. Phylogenetic relationships were determined with maximum likelihood (ML), neighbor-joining (NJ), and Bayesian inference (BI) analysis using MEGA X (Kumar et al., 2018) and MrBayes 3.1.2 (Ronquist & Huelsenbeck, 2003) software. The nucleotide substitution model was estimated as the TrN + I model (Tamura & Nei, 1993) based on the Bayesian information criterion (BIC) in jModeltest v. 0.0.1 (Posada, 2008). The *p*-distance model was used in MEGA X software to estimate pair-wise genetic distances. We used three single-locus species delimitation methods: ASAP (Puillandre et al., 2021), ABGD (Puillandre et al., 2012), and GMYC (Fujisawa & Barraclough, 2013). Analysis parameters and settings were given in detail by Turan, Aksu, et al. (2023), Turan, Bayçelebi, et al. (2023).

2.2 | Collection codes

FFR, Zoology Museum, Faculty of Fisheries, Recep Tayyip Erdoğan University, Rize; IFC-ESUF, Inland Fishes Collection, Faculty of Eğirdir Fisheries, Isparta University of Applied Sciences, Isparta.

3 | RESULTS

3.1 | *O. fatmae*, sp. nov.

urn:lsid:zoobank.org:act:36BF22D2-F19A-4DA7-96F1-02AAB801CD15.

3.2 | Holotype

FFR 15654, 54 mm standard length (SL); Türkiye: İzmir Prov.: Stream Güzelhisar at Süngüllü village. 38°52′09.4″N, 27°18′33.3E″.

3.3 | Paratypes

FFR 15653, 22, 44–57 mm SL; same data as holotype.

3.4 | Material used in molecular genetic analysis

FFRDNA 15653, 3, Türkiye: İzmir Prov.: Stream Güzelhisar at Süngüllü village, 38°52′09.4″N, 27°18′33.3″ E. (GenBank accession numbers: OR948615- OR948617).

3.5 | Diagnosis

O. fatmae is distinguished from other *Oxynoemacheilus* species in the Aegean and Marmara Sea basins by the following combination of characters: a small size (maximum size 57 mm SL) (vs. at least about 70 mm SL, except *O. theophilii*), the flank with four to eight irregularly shaped narrow black bars posteriorly and a marbled pattern anteriorly (vs. more or less irregularly shaped dark brown or pale brown blocks or bars on flank in *O. germencicus*, *O. theophilii*, *O. eliasi*, *Oxynoemacheilus simavicus*, *Oxynoemacheilus angorae* in most individuals; body with vermiculated pattern in *Oxynoemacheilus marmaraensis*); a somewhat slender body and caudal peduncle; a suborbital groove in males; an axillary lobe at base of pelvic fin; and no adipose crest on lower and upper parts of caudal peduncle.

3.6 | Description

The general body shape is shown in Figures 1 and 2, and morphometric data are given in Table 1. Maximum size 57 mm SL. Body slender, body depth at dorsal-fin origin 15%–17% SL, upper profile convex at

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FIGURE 1 *Oxynoemacheilus fatmae*: FFR 15654, holotype, female, 54 mm standard length (SL); Türkiye: İzmir Prov.: Güzelhisar Stream.

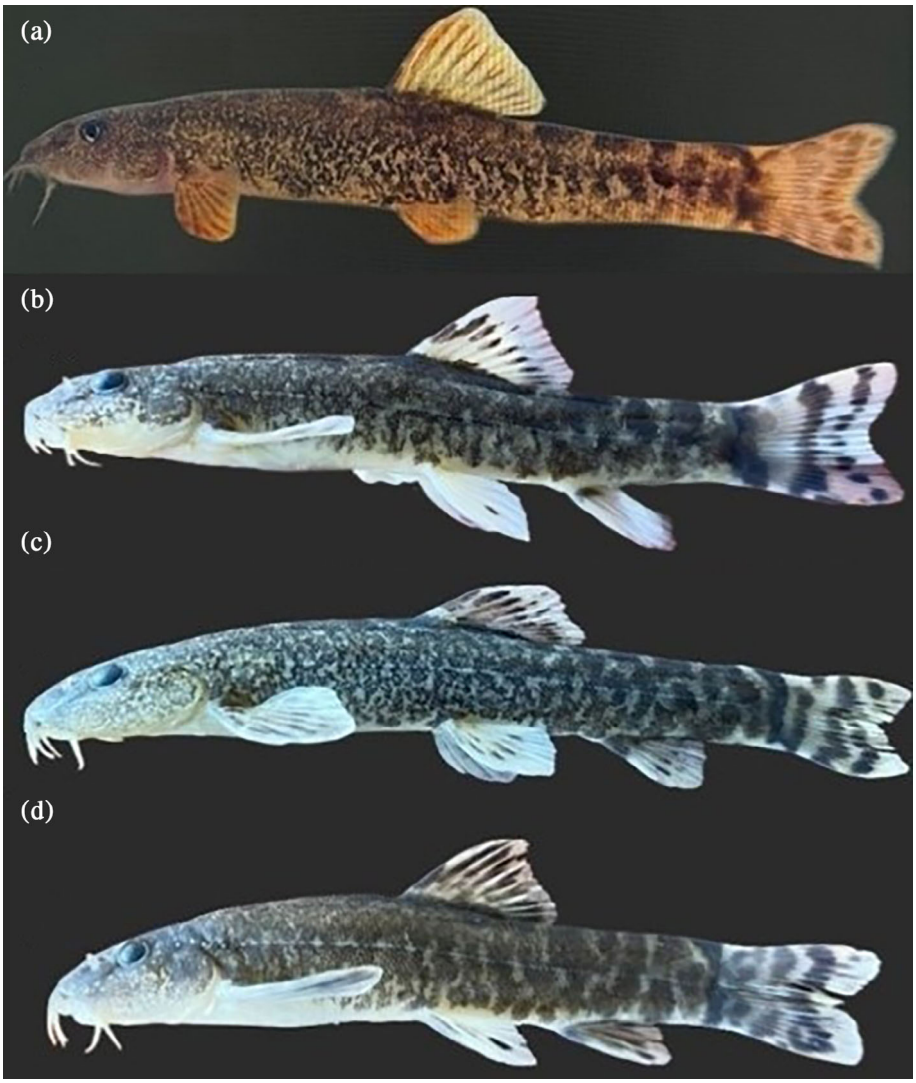

FIGURE 2 *Oxynoemacheilus fatmae*: FFR 15653, paratypes. (a) Female, 54 mm standard length (SL); (b) male, 4 mm SL; (c) female, 52 mm SL; (d) male, 43 mm SL; Türkiye: İzmir Prov.: Güzelhisar Stream.

the predorsal area and straight or slightly concave at postdorsal area. Head long, length 23%–27% SL, slightly pointed, not flattened on the ventral and dorsal surfaces. The caudal peduncle somewhat slender, length 17%–20% SL, and 1.5–1.8 greater than its depth. No adipose crest on lower and upper parts of caudal peduncle. Presence of an axillary lobe at the base of the pelvic fin. Pectoral fin moderately long,

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TABLE 1 Morphometric data of *Oxynoemacheilus fatmae* (holotype FFR 15654 and paratype FFR 15653, $n = 19$) and *Oxynoemacheilus theophilii* (FFR 1553, 8$n = 14$).

	O. fatmae ($n = 19$)			*O. theophilii* ($n = 14$)	
Morphological characters	**H**	**Range (mean)**	**SD**	**Range (mean)**	**SD**
Standard length (mm)	54	44–57		40–56	
In percentage of standard length					
Head length	24.1	23.4–27.0 (25.2)	0.9	23.6–25.7 (24.6)	0.7
Body depth at dorsal-fin origin	16.3	15.1–17.3 (16.4)	0.7	16.8–19.0 (18.0)	0.7
Body width at dorsal-fin origin	11.4	10.1–13.8 (11.7)	0.6	11.6–14.3 (12.7)	0.6
Predorsal length	51.6	49.2–53.8 (51.1)	1.1	46.7–53.9 (51.2)	1.8
Postdorsal length	34.6	32.4–38.2 (34.7)	1.6	32.6–37.1 (35.4)	1.5
Pre-anal length	75.7	73.1–77.3 (75.4)	1.1	75.6–79.5 (76.5)	1.1
Prepelvic length	50.7	49.7–55.0 (51.2)	1.4	51.6–54.6 (52.6)	1.0
Distance between pectoral- and pelvic-fin origins	28.4	26.7–32.3 (29.8)	1.4	26.7–32.2 (29.1)	1.7
Distance between pelvic- and anal-fin origins	22.9	21.7–25.0 (23.1)	0.9	20.6–32.2 (29.1)	1.7
Depth of caudal peduncle	11.4	10.2–11.5 (11.1)	0.4	11.9–14.3 (12.8)	0.7
Length of caudal peduncle	17.0	17.0–19.8 (18.2)	0.7	15.2–18.8 (17.0)	1.2
Dorsal-fin depth	20.1	17.3–23.4 (20.2)	1.7	17.6–24.4 (20.9)	1.6
Anal-fin base length	17.4	15.0–20.0 (17.2)	1.4	15.8–20.0 (18.0)	1.2
Pectoral-fin length	22.1	19.4–24.8 (22.0)	1.6	20.1–23.7 (21.9)	1.1
Pelvic-fin length	17.3	14.1–18.4 (17.1)	1.1	15.7–19.9 (17.7)	1.4
Caudal-fin length	23.6	22.1–27.3 (24.9)	1.2	22.8–29.2 (25.8)	1.9
Middle lop of caudal-fin length	18.1	16.1–19.2 (17.9)	1.0	18.9–22.8 20.3)	1.2
In percentage of head length					
Head depth at eye	40.9	35.4–43.8 (40.1)	2.5	38.3–48.6 (43.4)	3.0
Snout length	42.0	36.6–44.8 (41.6)	2.9	34.5–44.69 (39.3)	2.6
Eye diameter	16.8	19.3–26.1 (23.1)	2.4	16.2–25.5 (19.6)	2.6
Postorbital distance	48.9	40.0–57.1 (46.5)	4.4	47.5–62.3 (53.2)	5.2
Maximum head width	54.2	46.9–58.3 (53.7)	2.3	52.9–64.5 (58.4)	3.7
Interorbital width	26.4	21.4–26.4 (24.1)	1.6	19.7–28.6 (24.5)	2.8
Length of inner rostral barbel	28.2	19.6–31.9 (26.4)	2.9	16.3–32.2 (24.7)	4.9
Length of outer rostral barbel	33.6	27.2–37.6 (32.2)	4.1	26.1–40.2 (31.2)	4.8
Length of maxillary barbel	24.7	26.2–36.4 (30.4)	3.1	20.8–37.5 (29.6)	4.2
In percentage of caudal peduncle length					
Depth of caudal peduncle	1.5	1.5–1.82 (1.6)	1.0	1.2–1.5 (1.3)	1.1
In percentage of body depth at dorsal-fin origin					
Caudal peduncle depth	70.0	62–75 (68)	3.5	65.1–83.0 (71.2)	4.7
In percentage of length of caudal-fin					
Length of middle caudal-fin lope	67	63–80 (72)	4.7	73.3–83.1 (78.4)	3.4

length 19%–25% SL. Pelvic-fin length almost reaching to anus, its length 14%–18% SL. Anal fin short, length 15%–20% SL. The caudal fin short, slightly forked, and its length 1.3–1.6 times the length of the middle lope. The body with small scales. The lateral line complete. Presence of a slightly distinct suborbital groove in males. Mouth small with developed lips. The lower lip with a distinct median interruption. The upper lip with a median incision. Interorbital distance 21%–26% head length (HL). Barbels short, outer rostral 27%–38% HL, inner rostral 20%–32% HL, maxillary barbel 26%–36%.

Dorsal fin with III–IV simple 8 branched; pectoral fin with I simple 9–10 branched; pelvic-fin with I simple 7–6 branched; and anal-fin with III simple with 5 branched rays. Free edge of dorsal fin straight or slightly convex. Free edge of pectoral fin straight or slightly convex. Free edge of anal fin straight or slightly convex. Free

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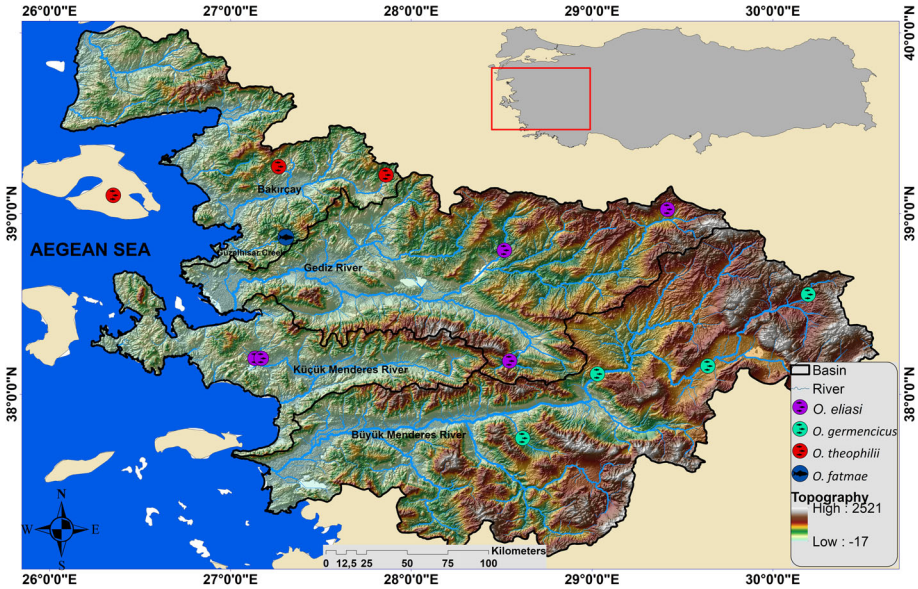

FIGURE 3 Distribution of *Oxynoemacheilus* species in Aegean Sea basin, Anatolia.

margin of pelvic fin straight. The caudal fin slightly forked and lopes slightly pointed.

3.7 | Sexual dimorphism

The pectoral fin in males is longer than that in females. In males, the pectoral-fin length was measured between 22% and 26% of SL, whereas in females, it ranged between 20% and 23% of SL.

3.8 | Colouration

Body color is grayish in both live and preserved individuals. Small brown mottling on the top of the head and cheeks, no pigmentation on the ventral side of the head in live individuals. The dorsal-fin origin with a small, roundish, black blotch. The flank with four to eight irregularly shaped narrow black bars posteriorly, with a marbled pattern anteriorly. Two to four blotches on the back in front of the dorsal fin. Three or four irregularly shaped blotches on the upper part of the caudal peduncle in most individuals. A vertically elongated black spot on the caudal-fin base. Dorsal and caudal fins with two to three irregularly shaped black bands on rays. Anal, pelvic, and pectoral fins yellowish, with numerous small black spots on the rays.

3.9 | Distribution

O. fatmae was found in the Güzelhisar Stream drainage in the northern Aegean Sea basin (Figure 3).

3.10 | Etymology

The species is named after Fatma Aksu, the second author's wife.

3.10.1 | Comparison with other *Oxynoemacheilus* species in Aegean and Marmara basins

O. fatmae differs from *O. theophilii* (Bakır Stream, Aegean basin), *O. germencicus* (Büyük Menderes and Gediz rivers, Aegean basin), *O. eliasi* (Tahtalı Reservoir and Gediz River, Aegean basin), *O. marmaraensis* (Susurluk River, Marmara Sea basin), *O. simavicus* (Susurluk River and Marmara Sea basin), and *O. angorae* (Susurluk River, Marmara Sea, and Black Sea basin) by having four to eight irregularly shaped narrow black bars commonly on the posterior part of flank, and anterior parts of the flank with a marbled pattern (vs. more or less irregularly shaped dark brown or pale brown blocks or bars on the flank in *O. germencicus*, *O. theophilii*, *O. eliasi*, *O. simavicus*, and *O. angorae* in most individuals; body with vermiculated pattern in *O. marmaraensis*). It further differs from *O. theophilii* and *O. eliasi* by having a slenderer body (body at dorsal-fin origin: 15%–17% SL vs. 17%–18% in *O. theophilii*, 19%–22% in *O. eliasi*), a slenderer caudal peduncle (10%–12% SL, vs. 12%–13% in *O. theophilii* and in *O. eliasi*). *O. fatmae* is also distinguished from *O. theophilii* in having a shorter middle caudal-fin lope (length of middle caudal-fin lope: 16%–19% SL vs. 19%–23%), fewer irregularly shaped black bars on flank (4–8 vs. 8–13), and the absence of dorsal and ventral adipose crests on the caudal peduncle behind the vertical of the posterior anal-fin base (vs. present). It further differs from *O. angorae* by a shorter postdorsal distance (32%–38% SL vs. 38%–41%) and a slender head (head depth through eye 34%–44% SL vs. 47%–53%). It further differs from *O. simavicus* by having a deeper caudal peduncle (10%–12% SL, 6–10) and a longer head (24%–27% SL, 19–22) and the general body color and pattern. It further differs from *O. marmaraensis* by having an axillary lobe at the base of the pelvic fin (vs. absent), the presence of a median incision in the upper lip (vs. absent), and a slenderer caudal peduncle (depth times in its length, 1.5–1.8 vs. 1.2–1.6).

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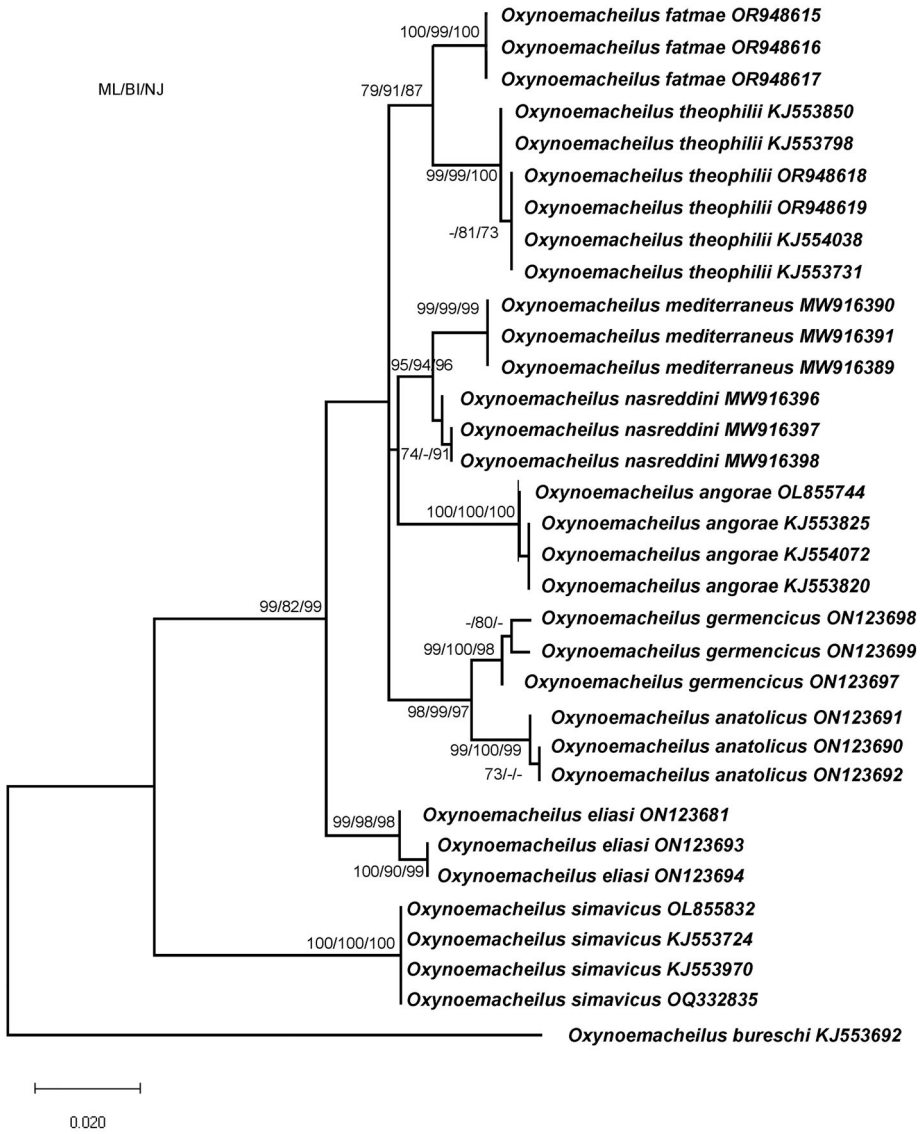

FIGURE 4 Maximum likelihood tree based on mitochondrial cytochrome oxidase subunit I (*COI*; 626 bp) gene sequences of *Oxynoemacheilus* spp. maximum likelihood and Bayesian inference and neighbor-joining analyses resulted in congruent trees. Bootstrap values are shown above nodes on tree if 70% or higher.

TABLE 2 Pair-wise distance values based on cytochrome oxidase sequences of *Oxynoemacheilus* species.

Species	*O. fatmae*	*O. theophilii*	*O. nasreddini*	*O. angorae*	*O. mediterraneus*	*O. eliasi*	*O. germencicus*	*O. anatolicus*
Oxynoemacheilus fatmae								
Oxynoemacheilus theophilii	0.022							
Oxynoemacheilus nasreddini	0.027	0.029						
Oxynoemacheilus angorae	0.033	0.042	0.030					
Oxynoemacheilus mediterraneus	0.034	0.036	0.010	0.033				
Oxynoemacheilus eliasi	0.035	0.037	0.030	0.040	0.034			
Oxynoemacheilus germencicus	0.038	0.036	0.030	0.041	0.033	0.041		
Oxynoemacheilus anatolicus	0.041	0.040	0.029	0.043	0.033	0.038	0.019	
Oxynoemacheilus simavicus	0.078	0.081	0.077	0.080	0.078	0.071	0.078	0.075

3.11 | Molecular distinctiveness of *O. fatmae*

COI barcode region sequences were analysed in nine *Oxynoemacheilus* species distributed in the northern Aegean. Species were divided into two main clades in all the phylogenetic analyses supported by high bootstrap values. The first clade consisted of *O. fatmae*, *O. theophilii*, *Oxynoemacheilus mediterraneus*, *Oxynoemacheilus nasreddini*, *O. angorae*, *O. germencicus*, *O. eliasi*, and *Oxynoemacheilus anatolicus*.

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The second clade consisted of *O. simavicus*; *O. fatmae* constituted a highly supported clade sister to *O. theophilii* (Figure 4). *p*-Distance between species ranged from 1.00% (*O. mediterraneus* and *O. nasreddini*) to 8.10% (*O. theophilii* and *O. simavicus*) (Table 2). *O. fatmae* differs from its most closely related congener, *O. theophilii*, by 14 nucleotide substitutions and the *p*-distance of 2.22% (Tables 2 and 3).

In the ASAP analysis, we found eight operational taxonomic units (OTU). ASAP's best partition (score = 2.50) results from a *p*-distance threshold of 0.012372. However, the ABGD and GMYC determined nine clusters. The likelihoods of null models and GMYC were 222.5873 and 225.1735, respectively. The GMYC analysis was represented by eight ML entities (CI: 6–10).

4 | DISCUSSION

O. fatmae is described from the headwaters of Güzelhisar Stream, a costal stream between Bakır Stream and Gediz River. Stoumboudi et al. (2006) described *O. theophilii* (Figure 5) from Lesbos Island in Greece based on three individuals. They reported two considerable key characters: caudal peduncle length 1.15–1.21 time its depth and the body with the marbled pattern. Later, Yoğurtçuoğlu et al. (2022) reported that *O. theophilii* from the Bakır Stream genetically (*COI*) overlapped with the Lesbos materials and also gave some morphological data of *O. theophilii* from Bakır Stream. For example, depth of the caudal peduncle 87%–97% of body depth at anterior-most dorsal-fin base; depth of the caudal peduncle 1.3–1.6 times in length; middle caudal-fin ray 71%–80% of the length of longest upper caudal-fin ray. Yoğurtçuoğlu et al. (2022) used the body depth almost uniform between dorsal- and caudal-fin bases, and the depth of the caudal peduncle 87%–99% of the body depth at anterior-most dorsal-fin base as key characters. We obtained some morphometric data belonging to *O. theophilii* (FFR 15538, $n = 14$, 40–56 mm SL) from Bakır Stream. Our morphometric data from *O. theophilii* are as follows: depth of the caudal peduncle is 65%–83% of the body depth at the anterior-most dorsal-fin base; the depth of the caudal peduncle 1.2–1.5 times in its length; and the middle caudal-fin ray is 73%–83% of the length of longest upper caudal-fin ray. The new species *O. fatmae* is closely related to *O. theophilii* based on the genetic dataset, with a genetic distance of 2.22% between the two species. Although the genetic divergence is not significant enough to support the distinctiveness, the two species are clearly distinguished from each other morphologically (see comparison section above).

O. fatmae is easily distinguished from *O. germencicus* (FFR 1523, $n = 7$, 52–58 mm SL; FFR 1528, $n = 12$, 39–56 mm SL; IFC-ESUF 19-0026, $n = 11$, 52–60 mm SL; FFR 1397, $n = 10$, 47–63 mm SL), *O. theophilii* (FFR 15538, $n = 14$, 40–56), and *O. eliasi* (FFR 15658, $n = 7$, 38–41 mm SL; IFC-ESUF 19-0015, $n = 11$, 44–65 mm SL; IFC-ESUF 19-0016, $n = 7$, 47–59 mm SL) by the body color and pattern. However, it cannot be clearly separated from *O. germencicus* using morphometric data. Similarly, *O. eliase* and *O. theophilii* cannot be differentiated from *O. germencicus*. This is because *O. germencicus* might show high morphological variation, or the genus *Oxynoemacheilus* might contain one or more species in the Büyük Menderes. To solve this problem, future studies should involve a larger number of specimens from several localities collected and compared using both morphological and morphometric methods in the basin.

4.1 | Comparative materials

For *O. theophilii*, *O. marmaraensis*, and *O. simavicus*, see Yoğurtçuoğlu et al. (2022), Turan, Aksu, et al. (2023), and Turan, Bayçelebi, et al. (2023).

4.2 | Additional materials

O. theophilii: FFR 15538, 5, 27–36 mm SL; Turkey: İzmir Prov.: Çağlayan Stream, a tributary of Bakır River 15 km east of Bergama, 39°27′12.2″ N, 27°00′30.9″ E.

TABLE 3 List of the variable nucleotide substitutions in the 626-bp-long mtDNA cytochrome oxidase subunit (COI) barcode region.

Species	GenBank number	Variable nucleotide positions													
				1	2	2	3	3	3	4	4	5	5	6	6
		5	6	9	3	8	0	4	7	3	8	0	8	1	4
		5	1	3	5	3	7	9	3	0	7	9	3	6	6
Oxynoemacheilus fatmae	OR948615	T	C	G	G	T	A	G	A	G	T	A	G	C	A
O. fatmae	OR948616														
O. fatmae	OR948617														
Oxynoemacheilus theophilii	OR948618	C	T	A	C	C	G	A	G	A	C	G	A	T	G
O. theophilii	OR948619	C	T	A	C	C	G	A	G	A	C	G	A	T	G
O. theophilii	KJ554038	C	T	A	C	C	G	A	G	A	C	G	A	T	G
O. theophilii	KJ553731	C	T	A	C	C	G	A	G	A	C	G	A	T	G
O. theophilii	KJ553850		T	A	C	C	G	A	G	A	C	G	A	T	G
O. theophilii	KJ553798		T	A	C	C	G	A	G	A	C	G	A	T	G

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FIGURE 5 *Oxynoemacheilus theophilii*: FFR 15538. (a) Female, 56 mm standard length (SL); (b) male, 48 mm SL; (c) male, 45 mm SL; Türkiye: İzmir Prov.: Bakır Stream.

O. eliasi: FFR 15658, 7, 38–41 mm SL; Turkey: İzmir Prov.: inlet of Tahtalı Reservoir, under Şaşal bridge, 38°11′56.4″ N, 27°10′13.8″ E.—IFC-ESUF 19-0015, 14, 43–66 mm SL; Türkiye: Manisa Prov.: Derbent Stream, Gediz River, 38°11′3.43″ N, 28°32′37.65″ E.—IFC-ESUF 19-0016, 91, 38–62 mm SL; Türkiye: Manisa Prov.: Derbent Stream, Gediz River, 38°11′3.43″ N, 28°32′37.65″ E.—IFC-ESUF 19-0021, 7, 42–53 mm SL; Türkiye; Manisa Prov.: Demirci Stream, Gediz River, 38°47′48.36″ N, 28°30′52.48″ E.—IFC-ESUF 19-0022, 15, 37–74 mm SL; Türkiye: Manisa Prov.: Derbent bridge, Gediz River, 39°01′23.50″ N, 29°25′02.23″ E.—IFC-ESUF 19-0011, 5, 41–58 mm SL; Türkiye: Kütahya Prov.: Bahçeler Creek, Gediz River, 38°58′36.14″ N, 29°23′43.66″ E.—IFC-ESUF 19-0012, 3, 44–48 mm SL; Türkiye: Manisa Prov.: Gediz River, 38°38′23.92″ N, 27°32′37.74″ E.—IFC-ESUF 19-0014, 5, 37–55 mm SL; Türkiye: Manisa Prov.: Gördük Stream, Gediz River, 39°02′55.19″ N, 27°55′39.27″ E.—IFC-ESUF 19-0017, 2, 46–61 mm SL; Türkiye: Kütahya Prov.: Bahceler Creek, Gediz River, 38°58′36.14″ N, 29°23′43.66″ E.—IFC-ESUF 19-0018, 3, 35–37 mm SL; Türkiye: Manisa Prov.: Gördük Stream, Gediz River, 39°02′55.19″ N, 27°55′39.27″ E.—IFC-ESUF 19-0023, 1, 54 mm SL; Türkiye; Manisa Prov.: Akpınar Spring, Gölmarmara Lake, 38°42′04.40″ N, 27°58′07.97″ E.—IFC-ESUF 19-0024, 1, 43 mm SL; Türkiye: Kütahya Prov.: Gümüşlü DSI Regl., Gediz River, 38°58′18.76″ N, 29°28′01.56″ E.—IFC-ESUF 19-0025, 9, 40–50 mm SL; Türkiye: Manisa Prov.: Yurtbaşı bridge, Gediz River, 38°36′16.19″ N, 28°48′54.68″ E.—IFC-ESUF 19-0028, 1, 48 mm SL; Türkiye: Manisa Prov.: Demirci Stream, Gediz River, 38°47′48.36″ N, 28°30′52.48″ E.

O. germencicus: FFR 1523, 7, 52–58 mm SL; Turkey: Denizli Prov.: Aksu Stream 4 km north of Honaz, 37°47′21.1″ N, 29°15′40.0″ E.—FFR 1508, 22, 35–65 mm SL; Turkey: Muğla Prov.: Çine Stream a tributary of Adnan Menderes Reservoir 8 km south of Çine, 37°32′33.7″ N, 28°03′44.6″ E.—FFR 1528, 12, 39–56 mm SL; Turkey: Denizli Prov.: Suçıkan Stream, a tributary of Lake Işıklı 1 km north of Çıtak, 38°09′20.9″ N, 29°38′14.3″ E.—FFR 1530, 61, 28–68 mm SL; Turkey: Uşak Prov.: Banaz River, 8 km north of Sivaslı, 38°33′00.4″ N, 29°37′12.7″ E.—FFR 1597, 10, 47–63 mm SL; Turkey: Aydın Prov.: Karacasu Stream, Büyük Menderes River.—IFC-ESUF 19-0006, 10, 27–60 mm SL; Türkiye: Denizli Prov.: Cindere Reservoir, Büyük Menderes River, 38°06′45.47″ N 29°01′47.65″ E.—IFC-ESUF 19-0007, 14, 33–56 mm SL; Türkiye: Denizli Prov.: Çıtak Bridge, Büyük Menderes River, 38°09′23.69″ N, 29°38′24.29″ E.—IFC-ESUF 19-0009, 9, 54–64 mm SL; Türkiye: Afyonkarahisar Prov.: Karadirek Stream, Büyük Menderes River, 38°33′08.29″ N, 30°11′45.52″ E.—IFC-ESUF 19-0026, 11, 52–61 mm SL; Türkiye: Aydın Prov.: Dandalas Stream, Büyük Menderes River. 37°45′26.00″ N, 28°36′58.53″ E.—IFC-ESUF 19-0010, 7, 36–67 mm SL; Türkiye: Denizli Prov.: Işıklı Lake canal, Büyük Menderes River, 38°16′22.89″ N, 29°54′23.64″ E.—IFC-ESUF 19-0019, 6, 51–65 mm SL; Türkiye: Aydın Prov.: Şirindere Stream, Büyük Menderes River, 37°55′41.87″ N, 27°46′39.37″ E.—IFC-ESUF 19-0020, 7, 46–61 mm SL; Türkiye: Uşak Prov.: Banaz Stream, Büyük Menderes River, 38°31′48.46″ N, 29°36′43.56″ E.—IFC-ESUF 19-0027, 4, 38–40 mm SL; Türkiye: Denizli Prov.: Yenicekent DSI Regl., Büyük Menderes River, 38°02′15.45″ N, 28°57′47.50″ E.

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AUTHOR CONTRIBUTIONS

DT designed the study plan, conducted fieldwork, and wrote the manuscript draft. SA and SSG conducted fieldwork and performed morphological analysis. GK performed the molecular genetic study and wrote part of the manuscript. All authors read, edited, and approved the final version of the manuscript.

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