



<https://doi.org/10.11646/zootaxa.4277.1.3>

<http://zoobank.org/urn:lsid:zoobank.org:pub:58DE9D03-DE8D-4FEF-84FE-46D353AFB347>

Luciobarbus chelifensis and *L. mascarensis*, two new species from Algeria (Teleostei: Cyprinidae)

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Abstract

Cyprinids of the genus *Luciobarbus* are the most abundant and widespread fishes in most freshwater ecosystems in the Maghreb. In the Mediterranean basin of Morocco, Algeria and Tunisia, all species of *Luciobarbus*—with exception of *L. guercifensis*—are superficially very similar and are distinguished mostly by minor morphometric characters. Molecular characters distinguish all species well and nine species are recognised from the area, two of them described here. *Luciobarbus chelifensis*, from the Chelif River drainage in Algeria, is distinguished by having 41–43+1–2 lateral line scales and a very short anal fin (18–19% SL). *Luciobarbus mascarensis*, from the Macta River drainage in Algeria, is distinguished by having usually 41+1–2 lateral line scales, a long anal-fin (19–22%) and a short caudal peduncle (15–17% SL). An identification key is given for all African Mediterranean *Luciobarbus* species except for *L. callensis* and *L. rifensis*, which could not be distinguished.

Key words: North Africa, Mediterranean biodiversity hotspot, Morphology, Cytochrome b, D-loop control region

Introduction

Incomplete knowledge of biodiversity remains a stumbling block for conservation planning and even occurs within globally important Biodiversity Hotspots (Geiger *et al.* 2014). The Mediterranean area is one of these globally important Biodiversity Hotspots (Myers *et al.* 2000) and Geiger *et al.* (2014) pointed on a high number of potentially undescribed freshwater fishes in that area, including cyprinid fishes of the genus *Luciobarbus* from the Maghreb. Barbels of the genus *Luciobarbus* are widespread and species-rich in the Maghreb countries Tunisia, Algeria and Morocco. Almost all species had been described in the 19th and early 20th century and the last comprehensive reviews were published by Pellegrin (1920, 1939). In the early 21st century, African *Luciobarbus* species had been involved in several molecular studies shedding some light to the diversity of this group of fishes (Machordom & Doadrio 2001, Tsigenopoulos *et al.* 2003, Geiger *et al.* 2014, Brahimi *et al.* 2016). The results of these studies suggested the occurrence of several undescribed *Luciobarbus* species in the Maghreb. Only very recently, some *Luciobarbus* species groups have been reviewed and new species have been described (Casal-Lopez *et al.* 2015, Doadrio *et al.* 2016a, 2016b).

The above-mentioned studies suggest that six *Luciobarbus* species (Fig. 2) occur in the African rivers flowing to the Mediterranean Sea: *L. rifensis* in the Mediterranean Laou River drainage and some Atlantic rivers in Morocco, *L. guercifensis* and *L. yahyaouii* in the Moulouya River drainage and some small adjacent coastal rivers in Morocco, *L. leptogon* in the small coastal rivers around Algiers in Algeria, *L. setivimensis* in the Soummam

River drainage and some small rivers in the Bay of Bedjaia in Algeria and *L. callensis* being widespread in Tunisia west to the Kebir River drainage in eastern Algeria. Furthermore, *L. biscariensis* (Fig. 2) is widespread in the Shott Melghir basin, El Hodna (Keddara) lake basin and the Chiffa River drainage, all being endorheic waters in Algeria. Results from molecular studies (Tsigenopoulos *et al.* 2003, Geiger *et al.* 2014) indicate, that there might be one additional, undescribed *Luciobarbus* species in the Algerian Macta River drainage. During our studies, we found another undescribed species in the Algerian Chelif River drainage, not detected by previous studies. It is the aim of this study to describe these two new species.

Material and methods

Morphology. After anaesthesia, fishes were fixed in 5% formaldehyde and stored in 70% ethanol or directly fixed in 99% ethanol. Measurements were made point-to-point, with a dial caliper and recorded to 0.1 mm. Two measurements were taken over projections to the body axis: pre-dorsal length and pre-pelvic length. Other methods for measurements and all counts follow Kottelat & Freyhof (2007). Standard length (SL) is measured from the tip of the snout to the posterior extremity of the hypural complex. The length of the caudal peduncle is measured from behind the base of the last anal-fin ray to the posterior extremity of the hypural complex, at mid-height of the caudal-fin base. The first unbranched dorsal and anal-fin rays are hidden in the skin and unbranched rays were counted from X-ray pictures. The position of the dorsal-fin origin was determined from x-ray pictures and not from preserved materials directly. The last two branched rays articulating on a single pterygiophore in the dorsal and anal fins are counted as "1½". Scales in lateral series are counted along the midlateral line from the first one to touch the shoulder girdle to the last scale at the posterior extremity of the hypural complex. The scales on the caudal-fin base are separated by +. The holotype is included in the calculation of ranges, means and SD.

Abbreviations used. SL, standard length; HL, lateral head length. Collection codes: FSJF, Fischsammlung J. Freyhof, Berlin; RMCA, Royal Museum for Central Africa, Tervuren; ZFMK, Zoologisches Forschungsmuseum A. Koenig, Bonn. Authors of species names are listed in Table 1.

TABLE 1. Species names mentioned in this study, and their authors.

<i>Barbus barbatus</i> Linnaeus, 1758
<i>Luciobarbus biscarensis</i> (Boulenger, 1911)
<i>Luciobarbus callensis</i> (Valenciennes, 1842)
<i>Luciobarbus guercifensis</i> Doadrio, Perea & Yahyaoui, in Doadrio, Casal-López, Perea & Yahyaoui, 2016
<i>Luciobarbus leptopogon</i> (Schimper, 1834)
<i>Luciobarbus setivimensis</i> (Valenciennes, 1842)
<i>Luciobarbus yahyaouii</i> Doadrio, Casal-López & Perea, 2016
<i>Luciobarbus rifensis</i> Doadrio, Casal-Lopez & Yahyaoui in Casal-Lopez, Perea, Yahyaoui & Doadrio, 2015

Molecular analysis. We were able to generate 53 new mitochondrial sequences (cyt b and D-loop) for eight *Luciobarbus* species (*L. biscarensis*, *L. callensis*, *L. chelifensis*, *L. leptopogon*, *L. mascarensis*, *L. rifensis*, *L. setivimensis* and *L. yahyaouii*). From NCBI Genbank, 28 additional mitochondrial sequences from African Mediterranean *Luciobarbus* species were downloaded (Table 2). *Barbus barbatus* was used as outgroup. Polymerase chain reaction (PCR) amplification and sequencing were applied to two fragments of the mitochondrial genome: cytochrome b (cyt b) and D-loop control region, amplified using GLUDG.L (Palumbi 1996) and H16460 primers (Perdices & Doadrio 2001) for cyt b, while the D-loop was amplified using L15923 (Iguchi *et al.* 1997) and H16500 primers (Nishida *et al.* 1998). Sequences for cyt b and D-loop were obtained with a length of 1140 bp and 470 bp respectively. All sequences were visualized and edited in SEQUENCHER v.5.2.4 (Gene Codes, USA) and then compared with other GenBank sequences in order to identify any undesirable contamination. The sequences were then aligned using MAFFT v.7 (Katoh & Standley 2013) with default parameters. Alignment refinement was performed using Gblocks v.0.90b available on the Gblocks Server in order to eliminate poorly aligned positions and divergent regions (Talavera & Castresana 2007). The sequences were concatenated into a single matrix in seaview v.4.5.4 (Gouy *et al.* 2010). All the analyses were performed on the combined mtDNA dataset (1610 bp).

The concatenated was analysed in PartitionFinder v.2 (Lanfear *et al.* 2017) in order to select the best models per locus. The alignment dataset was then partitioned by gene and codon position into four: cyt b (the codons are partitioned into 1st, 2nd and 3rd positions) and D-loop. Maximum likelihood analyses (Felsenstein 1981) were implemented with GARLI v.2.1 (Bazin *et al.* 2014) using the K80+G (Kimura 1980) F81+I (Felsenstein 1981), GTR+G (Lanave *et al.* 1984; Tavare 1986; Rodriguez *et al.* 1990) and HKY+G (Hasegawa *et al.* 1985) evolutionary models for cyt b 1st, 2nd and 3rd codons positions and D-loop, respectively, as suggested by PartitionFinder v.2 (Lanfear *et al.* 2017).

TABLE 2. Sequences from GenBank used in this study. Species, localities, the number of sequenced specimens and GenBank accession numbers (cyt b) are given.

Species	Locality	Coordinates	GenBank Nr.
<i>Luciobarbus biscarensis</i>	Abiod River, Biskra, Algeria		AY004726
<i>Luciobarbus biscarensis</i>	Illizi, Algeria		AY004724
<i>Luciobarbus callensis</i>	Abid River, Cap Bon, Tunisia	36.867286°N, 10.724574°E	AY004878
<i>Luciobarbus callensis</i>	Sarrath River, Djerissa, Tunisia	35.807531°N, 8.563658°E	AY004732
<i>Luciobarbus callensis</i>	Ain Assel reservoir, El-Taref, Algeria	36.747037°N, 8.400955°E	AF045974
<i>Luciobarbus leptopogon</i>	Keddara reservoir, Boumrdèse, Algeria	36.647275°N, 3.424412°E	AF145931
<i>Luciobarbus leptopogon</i>	Chiffa River, Blida, Algeria	36.445279°N, 2.756340°E	AY004749
<i>Luciobarbus mascarensis</i>	Tifrit River, Balloul, Algeria	34.987064°N, 0.408772°E	AY004743
<i>Luciobarbus rifensis</i>	Laou River, Derdara, Morocco		KT003926
<i>Luciobarbus rifensis</i>	Laou River, Derdara, Morocco		KT003927
<i>Luciobarbus rifensis</i>	Laou River, Derdara, Morocco		KT003928
<i>Luciobarbus rifensis</i>	Laou River, Derdara, Morocco		KT003929
<i>Luciobarbus rifensis</i>	Laou River, Derdara, Morocco		KT003930
<i>Luciobarbus rifensis</i>	Hachef River, Dar Chaoui, Morocco		KT003931
<i>Luciobarbus rifensis</i>	Hachef River, Dar Chaoui, Morocco		KT003932
<i>Luciobarbus rifensis</i>	Hachef River, Dar Chaoui, Morocco		KT003933
<i>Luciobarbus rifensis</i>	Hachef River, Dar Chaoui, Morocco		KT003934
<i>Luciobarbus rifensis</i>	Hachef River, Dar Chaoui, Morocco		KT003935
<i>Luciobarbus rifensis</i>	Loukos River, Souk Had Laghdir, Morocco		KT003936
<i>Luciobarbus rifensis</i>	Loukos River, Souk Had Laghdir, Morocco		KT003937
<i>Luciobarbus rifensis</i>	Loukos River, Souk Had Laghdir, Morocco		KT003938
<i>Luciobarbus rifensis</i>	Loukos River, Souk Had Laghdir, Morocco		KT003939
<i>Luciobarbus rifensis</i>	Loukos River, Souk Had Laghdir, Morocco		KT003940
<i>Luciobarbus setivimensis</i>	Soummam River, Algeria	36.632246°N, 4.74745°E	AY004748
<i>Luciobarbus setivimensis</i>	Aissi River, Tizi Ouzou, Algeria	36.668968° N, 4.124125°E	AY015991
<i>Luciobarbus setivimensis</i>	Aissi River, Tizi Ouzou, Algeria	36.668968° N, 4.124125°E	AY015992
<i>Luciobarbus yahyaouii</i>	Moulouya River, Boumia, Morocco	32.801789°N, 4.791778°W	AY004742
<i>Luciobarbus yahyaouii</i>	Moulouya River, Morocco	34.226706°N, 2.392343°W	AF145925
<i>Barbus barbuis</i>	Danube River, Austria		AB238965

In total, 1000 bootstraps (BT) were obtained in four independent runs, each including 250 repetitions. BT values were then summarized on the best maximum likelihood tree using SumTree (Sukumaran & Holder 2015) (run on DendroPy v.4.0.0; Sukumaran & Holder 2010). The K80+G, F81+I, GTR+G and HKY+G models were incorporated into Bayesian inference, which was performed using four runs of Metropolis-coupled Markov Chain Monte Carlo (MCMC) sampling as implemented in the MrBayes program v.3.2.5 (Ronquist *et al.* 2012), using default model parameters as starting values. The following parameter settings were used: mchains = 4; lset nst = 2, rates = gamma (for 1st cyt b codons and D-loop); lset nst = 2, rates = propinv (for 2nd cyt b codons); lset nst = 6, rates = gamma (for 3rd cyt b codons); mcmc ngen = 10,000,000; sample freq = 1,000. The trees were represented using Figtree v.1.4.2 (Rambaut 2009). The average uncorrected p-distances among African Mediterranean *Luciobarbus* species were calculated for the cyt b gene using MEGA v.6.0 (Tamura *et al.* 2013).

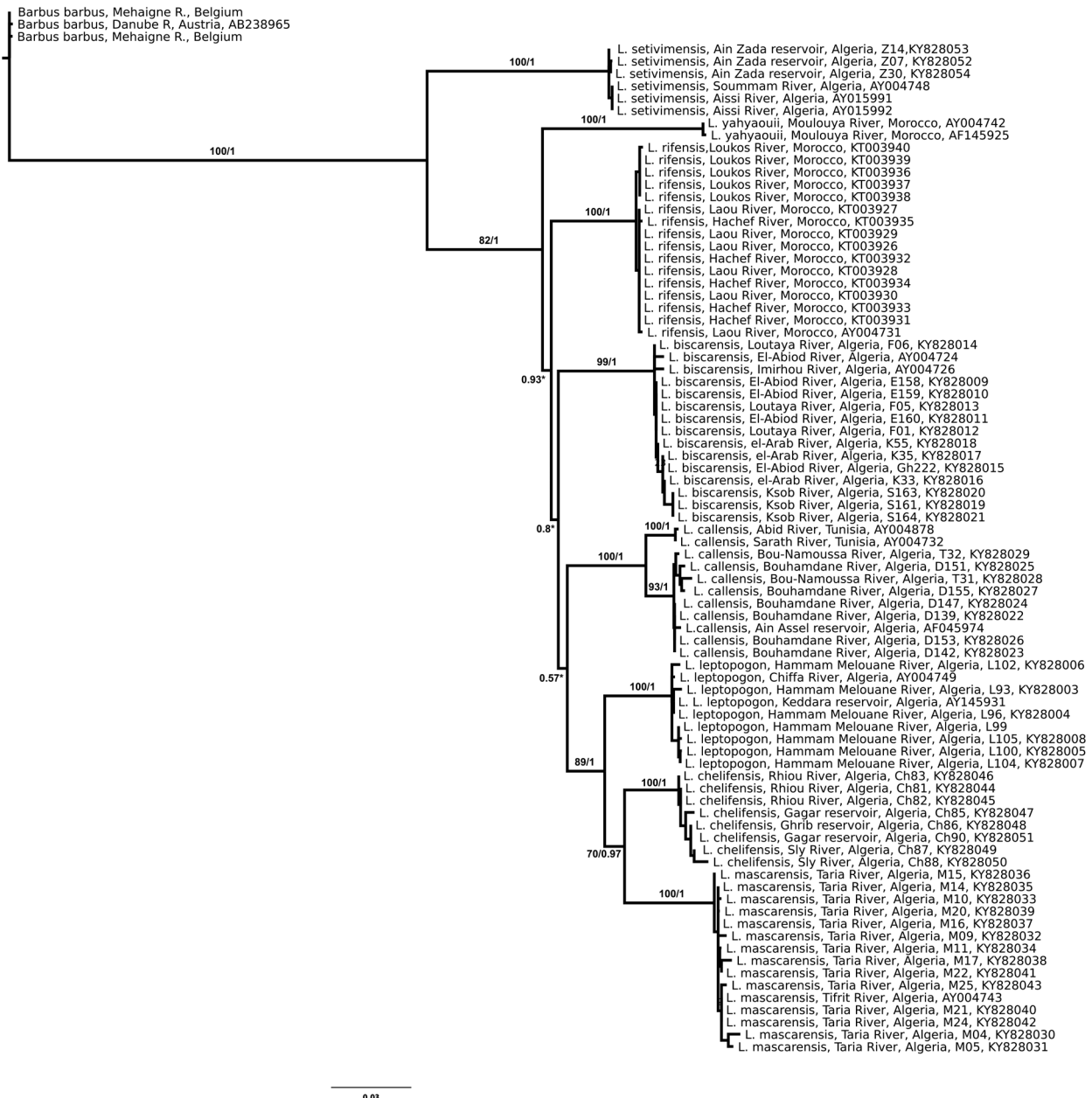


FIGURE 1. Phylogenetic relationships from NAmT (Cyt b and D-loop) of *Luciobarbus* species from the African Mediterranean basins, using Bayesian inference with posterior probability (PP); numbers left slash correspond to the bootstrap (BT) values.

Results

The Bayesian (BI) and the Maximum Likelihood (ML) analysis generated identical topologies in the clades recovered, but did differ in the support for the recovered groups. *Luciobarbus biscarensis*, *L. callensis*, *L. chelifensis*, *L. leptopogon*, *L. mascarensis*, *L. rifensis*, *L. setivimensis* and *L. yahyaouii* each correspond to one of the molecular groups in our analysis (Fig. 1). *Luciobarbus callensis* is separated into two groups, indicating, that an additional undescribed species might occur in the east of Algeria. Except for *L. setivimensis*, the largest molecular distance was found between *L. yahyaouii* and *L. chelifensis* ($d = 6.3\%$) and lowest differentiation between *L. mascarensis* and *L. chelifensis* ($d = 2.9\%$).

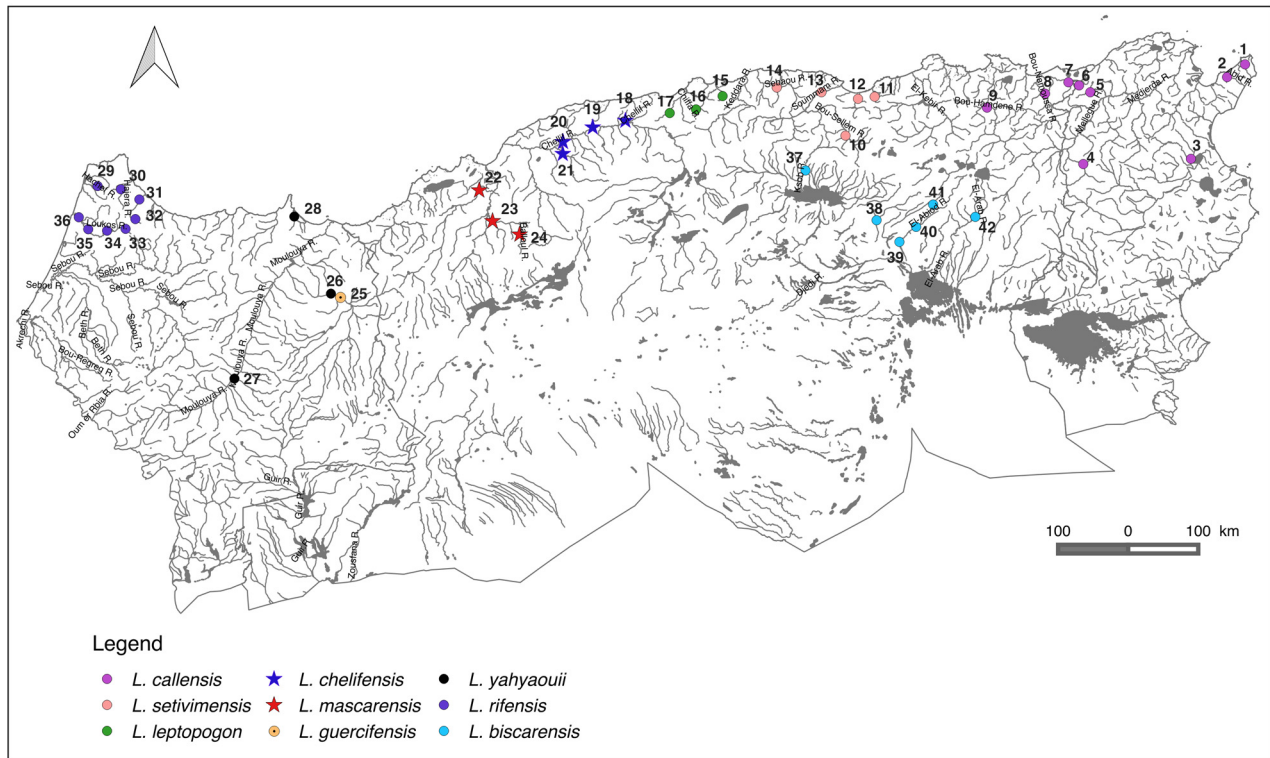


FIGURE 2. Records of *Luciobarbus* species examined in this study. Numbers correspond to materials examined listed below.

Key to species of *Luciobarbus* in the African Mediterranean and Sahara basins in Central Algeria and Tunisia

- 1a Barbels thick, rostral-barbel origin close to tip of snout. *L. guercifensis*
- 1b Barbels thin, rostral-barbel origin clearly behind tip of snout. 2
- 2a Usually 49–51+1–3 lateral line scales; $7\frac{1}{2}$ – $8\frac{1}{2}$ scale rows below lateral line; dorsal-fin origin behind of vertical of pelvic-fin origin. *L. biscarensis*
- 2b Usually 41–47+1–3 lateral line scales; $4\frac{1}{2}$ – $6\frac{1}{2}$ scale rows below lateral line; dorsal-fin origin in front or above vertical of pelvic-fin origin. 3
- 3a Last unbranched dorsal-fin ray serrated along $\frac{2}{3}$ of its length; anal-fin length 19–22% SL. 4
- 3b Last unbranched dorsal-fin ray serrated along almost its entire length; anal-fin length 16–19% SL. 7
- 4a Predorsal length 46–52% SL. 5
- 4b Predorsal length 52–58% SL. 6
- 5a Interorbital distance 36–39% HL; 41–43+1–2 lateral line scales. *Luciobarbus leptopogon*
- 5b Interorbital distance 40–44% HL; 43–47+1–3 lateral line scales. *Luciobarbus setivimensis*
- 6a Distance between pelvic and anal-fin origins 21–24% SL; caudal peduncle 1.3–1.5 times longer than deep; usually 5, very rarely 4 unbranched dorsal-fin rays. *Luciobarbus yahyaouii*
- 6b Distance between pelvic and anal-fin origins 24–29% SL; caudal peduncle 1.2–1.3 times longer than deep; 4 unbranched dorsal-fin rays. *Luciobarbus mascarensis*
- 7a Anal-fin base length 8–10% SL; 41–43+1–2 lateral line scales. *Luciobarbus chelifensis*
- 7b Anal-fin base length 6–8% SL; 43–47+1–3 lateral line scales. *Luciobarbus callensis* / *Luciobarbus rifensis*

***Luciobarbus chelifensis*, new species**

(Figs. 3–5)

Holotype. RMCA 2016-024-P-0010, 128 mm SL; Algeria: Chelif prov.: Oued Sly west of Ouled Ben Abdelkader village, 36.026609°N 1.266014°E; A. Brahim, 23 Jun 2016.

Paratypes. RMCA 2016-024-P-0011-0016, 6, 102–152 mm SL; same data as holotype.

Material for molecular analyses. RMCA DNA-2016-024-P-Ch81, RMCA DNA-2016-024-P-Ch82, RMCA DNA-2016-024-P-Ch83, Algeria: Relizane prov.: Oued Rhiou at Sidi Abed village, 36.009889°N 0.992520°E (GenBank accession numbers: Cyt b: KY828044, KY828045, KY828046; D-loop: KY828098, KY828099, KY828100).—RMCA DNA-2016-024-P-Ch85, RMCA DNA-2016-024-P-Ch90, Algeria: Relizane prov.: Gagar Dam at Ouled Taieb village, 35.908391°N 1.008624°E (GenBank accession numbers: Cyt b: KY828047, KY828051; D-loop: KY828101, KY828097).—RMCA DNA-2016-024-P-Ch87, RMCA DNA-2016-024-P-Ch88, Algeria: Chelif prov.: Oued Sly west of Ouled Ben Abdelkader village, 36.026609°N 1.266014°E (GenBank accession numbers: Cyt b: KY828049, KY828050; D-loop: KY828103, KY828096).—RMCA DNA-2016-024-P-Ch86, Algeria: Ain Defla prov.: Ghrib reservoir at Oued Chorffa, 36.140553°N 2.563226°E (GenBank accession numbers: Cyt b: KY828048; D-loop: KY828102).

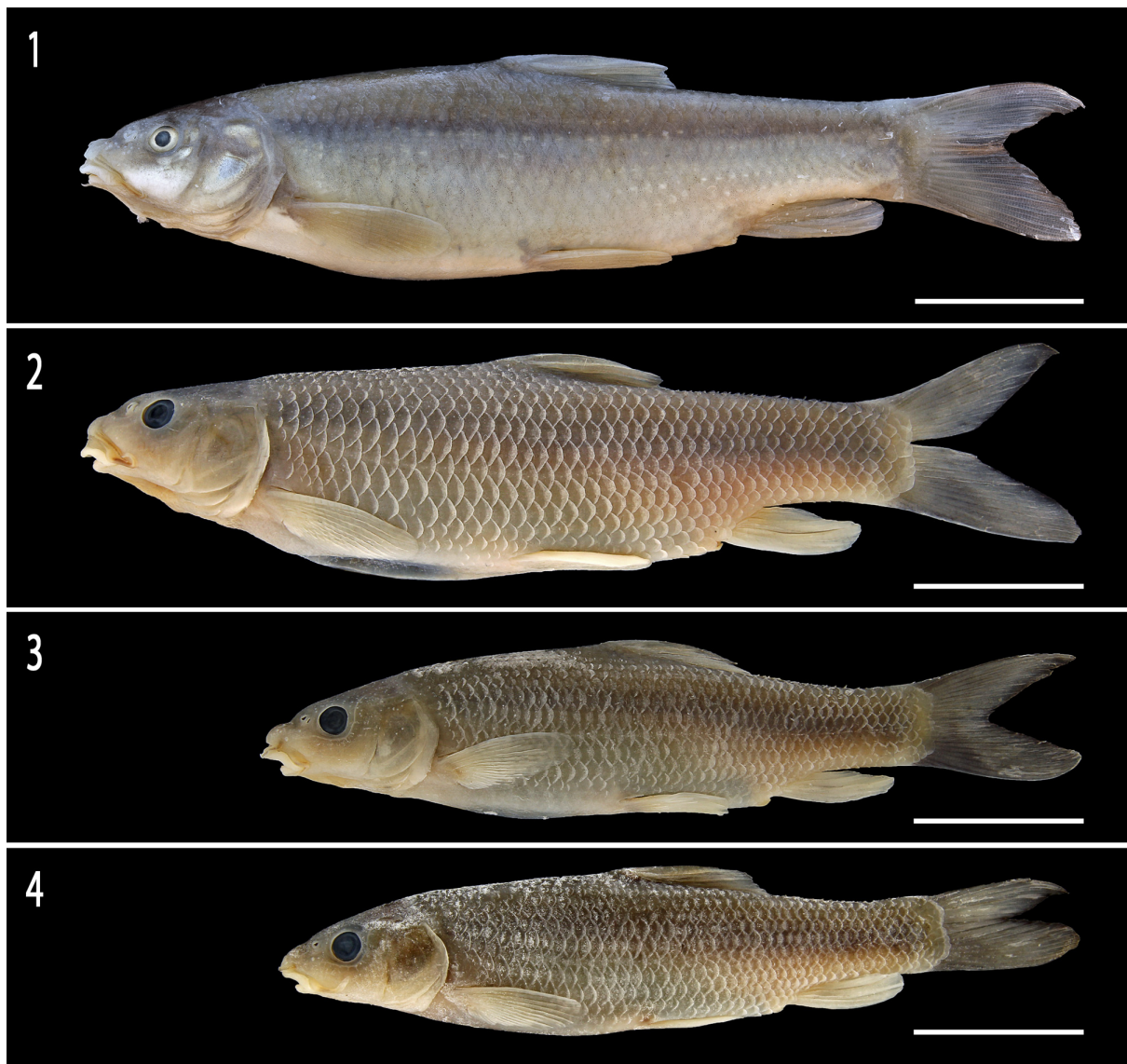


FIGURE 3. *Luciobarbus chelifensis*, **1**, RMCA 2016-024-P-0010, holotype (before preservation), 129 mm SL; **2**, RMCA 2016-024-P-0013, paratype, 126 mm SL; **3**, RMCA 2016-024-P-0015, paratype, 107 mm SL; **4**, RMCA 2016-024-P-0016, paratype, 102 mm SL; Algeria: Oued Sly west of Ouled Ben Abdelkader. Scale bars = 30 mm.

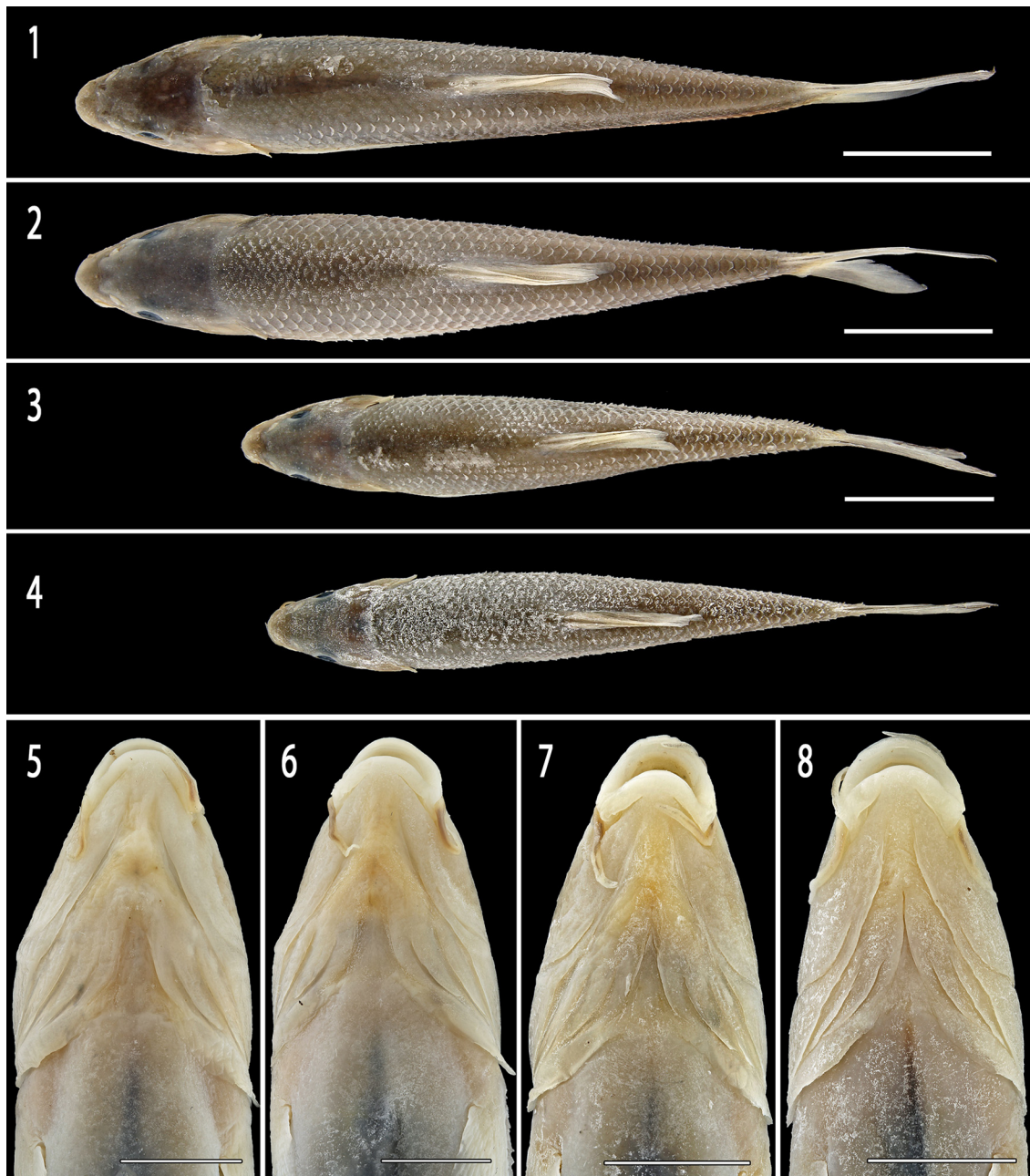


FIGURE 4. *Luciobarbus chelifensis*, 1+5, RMCA 2016-024-P-0010, holotype, 129 mm SL; 2+6, RMCA 2016-024-P-0013, paratype, 126 mm SL; 3+7, RMCA 2016-024-P-0015, paratype, 107 mm SL; 4+8, RMCA 2016-024-P-0016, paratype, 102 mm SL; Algeria: Oued Sly west of Ouled Ben Abdelkader. Scale bars: 1–4 = 30 mm; 5–8 = 10 mm.



FIGURE 5. *Luciobarbus chelifensis*, not preserved, about 150 mm SL; Algeria: Oued Sly west of Ouled Ben Abdelkader.

TABLE 3. Morphometric data of *L. chelifensis* (holotype RMCA 2016-024-P-0010; paratypes RMCA 2016-024-P-0011-0016, n=6; non-preserved individuals, n=19). The holotype is included in the calculation of ranges, means and SD.

	holotype	holotype & paratypes		
		range	mean	SD
Standard length (mm)	128	102–296		
In percent of standard length				
Body depth at dorsal-fin origin	27.3	24.8–31.3	27.6	1.3
Head length	24.3	22.4–26.4	24.2	1.2
Pre-dorsal length	52.3	49–54.2	51.6	1.1
Pre-pelvic length	54.7	50.3–56.0	53.4	1.6
Pre-anal length	78.1	73.8–79.3	77.3	1.6
Dorsal fin base length	14.1	12.8–15.2	13.6	0.6
Anal fin base length	9.4	8.5–9.9	9.0	0.4
Anal fin length	18.2	17.6–18.5	18.1	0.3
Pelvic fin length	15.6	14.1–16.2	15.2	0.6
Pectoral fin length	20.3	17.4–20.7	19.0	0.9
Caudal peduncle length	16.4	14.0–18.3	15.7	1.3
Caudal peduncle depth	12.2	11.2–12.7	11.9	0.5
Distance between dorsal and anal-fin origins	35.2	32.9–38.3	35.6	1.3
Distance between pectoral and pelvic-fin origins	29.9	25.5–31.3	29.2	1.5
Distance between pelvic and anal-fin origins	23.4	20.6–27.6	23.9	1.2
In percent of head length				
Snout length	35.6	27–37	32.2	2.9
Eye diameter	15.0	14–18	16.3	1.0
Post-orbital length	49.4	47–55	51.5	2.6
Interorbital distance	36.7	36–40	37.9	0.8
Pectoral-fin length	81.9	70–85	78.7	4.2
Caudal peduncle depth	49.1	46–52	49.2	1.2

Diagnosis. *Luciobarbus chelifensis* is distinguished from other *Luciobarbus* species from the African Mediterranean basins by a combination of characters, none of them unique. It is distinguished from all other species, except *L. leptopogon*, by having 41–43+1–2 lateral line scales (vs. 49–51+1–2 in *L. biscarensis*, 43–47+1–2 in *L. callensis*, 43 to 45+1–3 in *L. rifensis* and *L. setivimensis*, 42–45+1–2 in *L. yahyaouii*). The new species also has a very short anal fin (anal-fin length 18–19% SL vs. 19–22 in *L. leptopogon* and *L. setivimensis*, 19–21 in *L. yahyaouii*). *Luciobarbus chelifensis* is also distinguished from *L. leptopogon* by having a smaller eye (14–16% HL vs. 16–20), a greater body depth at the dorsal-fin origin (25–31% SL vs. 20–27), a shorter pelvic-fin length (14–16% SL vs. 16–20) and the last unbranched dorsal-fin ray being serrated along almost its entire length (vs. last unbranched dorsal-fin ray serrated along 2/3 of its length). It is further distinguished from *L. biscarensis* by having 4 unbranched dorsal-fin rays (vs. 5), 7½ scale rows between the lateral line and the dorsal-fin origin (vs. 8½–9½), 4½–5½ scale rows between the lateral line and the pelvic-fin origin (vs. 7½–8½) and the dorsal-fin origin being situated above the pelvic-fin origin (vs. behind). *Luciobarbus chelifensis* is further distinguished from *L. callensis* and *L. rifensis* by having a wider anal-fin base (8–10% SL vs. 5–8 in *L. callensis* and *L. rifensis*) and from *L. setivimensis* by the last unbranched ray being serrated along almost its entire length (vs. last unbranched ray serrated along 2/3 of its length) and having a narrower interorbital distance (36–40% HL vs. 40–44). The new species is also distinguished from *L. yahyaouii* by having always 4 unbranched dorsal-fin rays (vs. usually 5), the last unbranched dorsal-fin ray being serrated along almost its entire length (vs. serrated along 2/3 of its length) and having a shorter pelvic-fin length (14–16% SL vs. 16–19).

Description. See Figures 3–5 for general appearance. Morphometric data are given in Table 3 and meristic data are given in Table 5 and 6. A medium-sized species with a moderately long head. Body deepest at dorsal-fin origin or about midline between nape and dorsal-fin origin. Depth decreasing continuously towards caudal-fin base. Greatest body width in front of dorsal-fin base. Caudal peduncle compressed, 1.2–1.5 times longer than deep. Section of head roundish, flattened on ventral surface. Snout rounded. Mouth inferior. Dorsal-fin origin situated above pelvic-fin origin. Anal-fin origin slightly behind vertical of middle between dorsal and caudal-fin origins. Anal fin not reaching caudal-fin base. Pectoral fin reaching approximately 60–75% of distance from pectoral-fin origin to pelvic-fin origin. Pelvic fin reaching vertical of tip of last dorsal-fin ray when folded down. Pelvic fin reaching to a short distance in front of anus. Posterior dorsal-fin margin straight or slightly convex. Posterior pectoral and anal-fin margins convex. Caudal fin forked with rounded lobes of equal length. Largest known specimen 152 mm SL, but expected to grow much larger.

Dorsal fin with 4 unbranched and 8½ branched rays, last unbranched ray serrated along almost its entire length. Anal fin with 3 unbranched and 5½ branched rays. Pectoral-fin with 15–18 rays (mode 16) and pelvic fin with 7–8 (mode 8) rays. Lateral line with 41–43 scales on flank and 1–2 scales on caudal-fin base. Between dorsal-fin origin and lateral line 7½ scale rows and 4½–5½ (mode 4½) scale rows between pelvic-fin origin and lateral line. Pharyngeal teeth in three rows: 4+3+2.

Coloration. Whitish golden on belly, golden to greenish-grey on flank and back. A wide and indistinct bronze stripe along lateral line. Paired fins and anal fin yellowish or orange. Dorsal and caudal fins grey.

Etymology. *Luciobarbus chelifensis* is named for the Chelif River. A noun in genitive, indeclinable.

Distribution. *Luciobarbus chelifensis* was found in the Chelif River drainage in northwestern Algeria. See Figure 2 (18–21) for the distribution of African Mediterranean *Luciobarbus* species.

***Luciobarbus mascarensis*, new species**

(Figs. 6–8)

Holotype: RMCA 2016-024-P-0017, 126.1 mm SL; Algeria: Mascara prov.: Oued Taria north of Meftah Sidi Boubekeur, 35.109848°N, 0.0673°E; A. Brahimi, A. El-Najar & M. Kadjame, 1 May 2016.

Paratypes. RMCA 2016-024-P-0018-0026, 9, 66–188 mm SL; same data as holotype.—FSJF 3283, 4, 75–121 mm SL; Algeria: Oued el Hammam upriver of Hacine, 35.444265°N 0.035043°E.

Material for molecular analyses. RMCA DNA-2016-024-P-M09-M22; Algeria: Mascara prov.: Oued Taria north of Meftah Sidi Boubekeur, 35.109848°N, 0.0673°E (GenBank accession numbers: Cyt b: KY828030, KY828031, KY828032, KY828033, KY828034, KY828035, KY828036, KY828037, KY828038, KY828039, KY828040, KY828041, KY828042, KY828043; D-loop: KY828082, KY828083, KY828084, KY828085, KY828086, KY828087, KY828088, KY828089, KY828090, KY828091, KY828092, KY828093, KY828094, KY828095).

Diagnosis. *Luciobarbus mascarensis* is distinguished from other *Luciobarbus* species from the African Mediterranean basin except *L. chelifensis* and *L. leptopogon*, by having usually 41+1–2 lateral line scales (vs. 49–51+1–2 in *L. biscarensis*, 43–47+1–2 in *L. callensis*, 43–45+1–3 in *L. rifensis* and *L. setivimensis*, 42–45+1–2 in *L. yahyaouii*) (Table 6). Only *L. chelifensis* and *L. leptopogon* have regularly 41 lateral-line scales on the flank. *Luciobarbus mascarensis* is distinguished from *L. chelifensis* by having a longer anal-fin (19–22% SL vs. 18–19) and the dorsal fin serrated at 2/3 of its length (vs. serrated along almost its entire length) and from *L. leptopogon* by having a deeper body (body depth at dorsal-fin origin 26–31% SL vs. 20–27) and a wider caudal peduncle (caudal peduncle depth 74–84% caudal peduncle length vs. 60–74). *Luciobarbus mascarensis* is also distinguished from *L. biscarensis* by having 7½ scale rows between the lateral line and the dorsal-fin origin (vs. 8½–9½), 4½–5½ scale rows between the lateral line and the pelvic-fin origin (vs. 7½–8½), 4 unbranched dorsal-fin rays (vs. 5), the dorsal-fin origin situated in front or above the pelvic-fin origin (vs. behind), a longer anal-fin (19–22% SL vs. 17–20), a short caudal peduncle (caudal peduncle length 15–17% SL vs. 16–19). It is further distinguished from *L. callensis* and *L. rifensis* by having a longer anal-fin (19–22% SL vs. 17–19 in *L. callensis*, 16–19 in *L. rifensis*), a wider anal-fin base (7–10% SL vs. 6–8 in *L. rifensis*) and a wider caudal peduncle (caudal peduncle depth 74–84% caudal peduncle length vs. 60–78 in *L. rifensis*). *Luciobarbus mascarensis* is also distinguished from *L. setivimensis* by having a more narrow interorbital distance (36–40% HL vs. 40–44). The new species is also distinguished from *L.*

yahyaouii by having 4 unbranched dorsal-fin rays (vs. usually 5), a greater distance between the pelvic and anal-fin origins (24–28% SL vs. 21–24) and a shorter caudal peduncle (caudal peduncle length 15–17% SL vs. 16–19.4).



FIGURE 6. *Luciobarbus mascarensis*, **1**, RMCA 2016-024-P-0017, holotype (before preservation), 129 mm SL; **2**, RMCA 2016-024-P-0021, paratype, 123 mm SL; **3**, RMCA 2016-024-P-0024, paratype, 120 mm SL; **4**, RMCA 2016-024-P-0025, paratype, 93 mm SL; Algeria: Oued Taria north of Meftah Sidi Boubekeur. Scale bars = 30 mm.

TABLE 6. Frequencies of lateral-line scales on flank in *Luciobarbus* species examined for this study.

Species	N	Scales on the lateral line										
		41	42	43	44	45	46	47	48	49	50	51
<i>L. biscarensis</i>	26									8	2	16
<i>L. callensis</i>	34			2	5	16	9	2				
<i>L. chelifensis</i>	15	4	9	2								
<i>L. leptopogon</i>	18	5	8	5								
<i>L. mascarensis</i>	15	14		1								
<i>L. rifensis</i>	19			10	6	3						
<i>L. setivimensis</i>	30			6	9	5	8	2				
<i>L. yahyaouii</i>	19		6	7	4	2						

Description. See Figures 6–8 for general appearance. Morphometric data are given in Table 4 and meristic data are given in Table 5 and 6. Middle sized and stout species, with a moderately long head. Dorsal profile with a well-marked discontinuity between head and back. Body deepest at about midline between nape and dorsal-fin origin. Depth decreasing continuously towards caudal-fin base. Greatest body width between pectoral and dorsal-

fin origins. Caudal peduncle compressed, 1.2–1.3 times longer than its deep. Section of head roundish, flattened on ventral surface. Snout blunt. Mouth inferior. Dorsal-fin origin in front or almost above pelvic-fin origin. Anal-fin origin slightly behind vertical of middle between dorsal and caudal-fin origins. Anal fin reaching to caudal-fin base. Pectoral fin reaching approximately 59–77% of distance between pectoral-fin origin and pelvic-fin origin. Pelvic fin not reaching vertical of tip of last dorsal-fin ray when folded down. Pelvic fin reaching to a short distance in front of anus. Posterior dorsal-fin margin straight or slightly convex. Posterior pectoral-fin margin convex. Posterior anal-fin margin convex. Caudal fin forked with rounded lobes of equal size. Largest known specimen 140 mm SL, but expected to grow much larger.

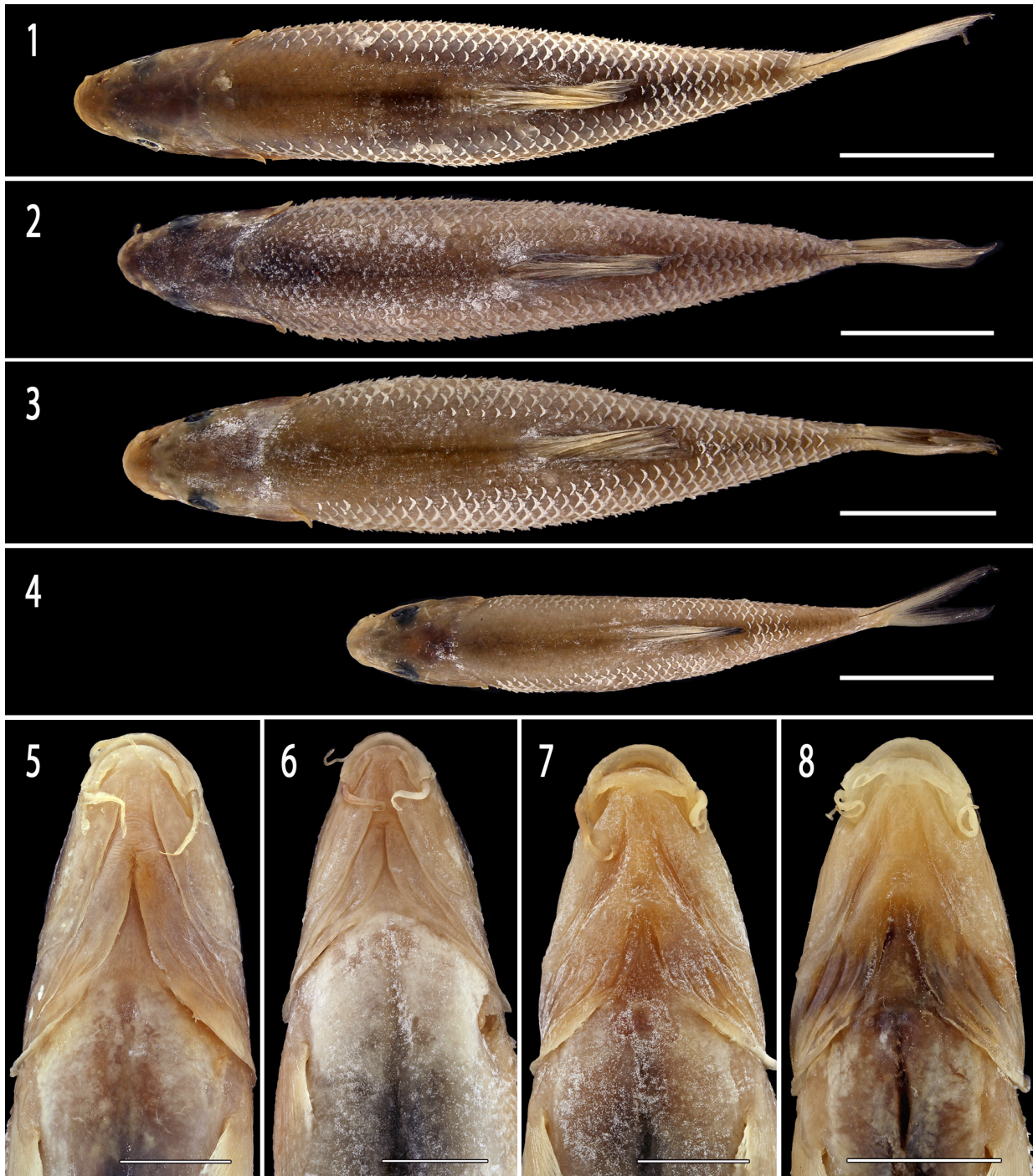


FIGURE 7. *Luciobarbus mascarensis*, **1+5**, RMCA 2016-024-P-0017, holotype (before preservation), 129 mm SL; **2+6**, RMCA 2016-024-P-0021, paratype, 123 mm SL; **3+7**, RMCA 2016-024-P-0024, paratype, 120 mm SL; **4+8**, RMCA 2016-024-P-0025, paratype, 93 mm SL; Algeria: Oued Taria north of Meftah Sidi Boubekeur. Scale bars: 1–4 = 30 mm; 5–8 = 10 mm.

TABLE 4. Morphometric data of *L. mascarensis* (holotype RMCA 2016-024-P-0017; paratypes RMCA 2016-024-P-0018-0026, n=9; non-preserved individuals, n=15). The holotype and paratypes are included in the calculation of ranges, means and SD.

	holotype	holotype & paratypes		
		range	mean	SD
Standard length (mm)	126.1	63.5–185		
In percent of standard length				
Body depth at dorsal-fin origin	26.2	26.2–30.6	28.6	1.1
Head length	25.8	24.4–28.0	26.1	0.8
Pre-dorsal length	54.7	52.5–58.3	54.6	1.4
Pre-pelvic length	54.9	50.0–58.3	54.4	1.7
Pre-anal length	80.9	73.3–85.1	79.9	1.9
Dorsal fin base length	12.4	12.1–15.3	13.8	0.8
Anal fin base length	7.5	6.6–9.6	7.9	0.6
Anal fin length	19.7	19.1–21.7	20.1	0.7
Pelvic fin length	16.0	14.3–19.4	15.8	1.2
Pectoral fin length	18.7	16.4–21.3	18.5	1.3
Caudal peduncle length	15.8	15.2–17.1	16.3	0.5
Caudal peduncle depth	11.8	11.8–13.3	12.6	0.4
Distance between dorsal and anal-fin origins	34.9	32.8–38.1	34.9	1.3
Distance between pectoral and pelvic-fin origins	34.9	25.8–31.4	28.3	1.4
Distance between pelvic and anal-fin origins	27.0	24.1–28.2	25.6	1.2
In percent of head length				
Snout length	31.8	26–38	32.3	2.8
Eye diameter	21.3	15–28	21.8	2.7
Post-orbital length	46.9	44–48	45.9	0.7
Interorbital distance	39.7	36–40	38.8	0.8
Pectoral-fin length	72.5	61–80	70.8	5.2
Caudal peduncle depth	45.8	46–53	48.5	1.2

TABLE 5. Frequencies of meristic characters: unbranched dorsal-fin rays and branched fin rays of *Luciobarbus* species examined for this study.

Species	N	unbranched dorsal-fin rays			N	branched dorsal-fin rays			branched pelvic-fin rays	
		3	4	5		7½	8½	9½	7	8
<i>L. biscarensis</i>	26			26	64	5	59		22	42
<i>L. callensis</i>	34		19	4	60		60			60
<i>L. chelifensis</i>	15		15		26		26		2	24
<i>L. leptopogon</i>	18		18		39		39		1	38
<i>L. mascarensis</i>	15		13		29		29		5	24
<i>L. rifensis</i>	19		13	6	19		19			19
<i>L. setivimensis</i>	30	8	22		57		57		4	53
<i>L. yahyaouii</i>	19		1	18	29		27	2	4	25

Dorsal fin with 4 unbranched and 8½ branched rays, last unbranched ray ossified and serrated at 2/3 of its length. Anal fin with 3 unbranched and 5½ branched rays. Pectoral-fin with 13–16 (mode 15) and pelvic fin with 7–8 (mode 8) rays. Lateral line with 41 (n=14) and 43 (n=1) scales on flank and 1–2 scales on caudal-fin base.

Between dorsal-fin origin and lateral line $7\frac{1}{2}$ scale rows and $4\frac{1}{2}$ – $5\frac{1}{2}$ (mode $5\frac{1}{2}$) scale rows between pelvic-fin origin and lateral line. Pharyngeal teeth in three rows: 4+3+2.

Coloration. Body yellowish brown in life and preserved individuals without colour pattern. Head plain brown, cheeks and ventral head and body whitish. Fins hyaline, usually with yellowish rays.

Etymology. *Luciobarbus mascarensis* is named for the Mascara region in western Algeria. A noun in genitive, indeclinable.

Distribution. *Luciobarbus mascarensis* was found in streams in the Macta River drainage in northwestern Algeria. See Figure 2 (22–24) for the distribution of African Mediterranean *Luciobarbus* species.



FIGURE 8. *Luciobarbus mascarensis*, FSJF 3283, 121 mm SL; Algeria: Oued el Hammam upriver of Hacine.

Discussion

While we made considerable efforts to detect external morphological characters usable to identify the different *Luciobarbus* species from the African Mediterranean basin, we found this exercise frustrating. Most species are very similar in their morphometric and meristic characters and almost all morphometric and meristic ranges overlap and larger overlaps are expected, if larger series of specimens would be examined. Only *L. guercifensis* and *L. biscarensis* are well distinguished from the other species. We found no character to distinguish *L. rifensis* from *L. callensis* and both might be real cryptic species. Casal-Lopez *et al.* (2015) and Doadrio *et al.* (2016a, 2016b) also found very few morphological characters to distinguish their new species and they base their diagnosis largely on molecular characters. With regard to the paucity of morphological characters, the inclusion of molecular genetic characters into standard taxonomic works in *Luciobarbus* is of great value.

The literature is full of studies, where molecular trees and morphological species do not fit together (see for example Astrin *et al.* 2012 and citations herein). Molecular characters, or more correctly molecular distances, must be handled with great care in taxonomy as there are no agreed molecular distances for species level delimitations. That means, that "little" or "great" molecular distances are not defined and there is no "species level distance". Naturally, the same is true for morphological characters, which need a certain experience in handling.

Many authors failed to find the so called "barcode-gap", especially if young and allopatric species are involved (see Geiger *et al.* 2014 for an example of freshwater fishes). While there is a trend to reject morphologically "well" distinguished species in the case of "little" molecular distances, the term cryptic species is misused to name molecular lineages as own species without appropriate morphological comparative studies. Great care has to be taken and we should allow certain molecular differences between populations of geographically widely distributed species occurring in obviously isolated populations. While this is not the place to discuss this topic in depth, we ask for great care not to describe each population making a slightly different clade in a phylogenetic tree as a different, cryptic, species. Such cases are within the greyzone between species and populations and need wise treatments. Recognising, usually young species which are morphologically "well" distinguished but show "little" molecular differences should not open the door to describe just all populations with little molecular differences, also those lacking morphological differences, as cryptic species.

TABLE 7. Morphometric data of *L. biscarensis* (FSJF 3286, n=31; FSJF 3279, n=9; non-preserved individuals, n=24); *L. callensis* (FSJF 3014, n=6; FSJF 3011, n=4; FSJF 3017, n=20; FSJF 3008, n=9; non-preserved individuals, n=21); *L. leptopogon* (FSJF 3284, n=9; RMCA-2016-024-P-0001-0009: n=9; non-preserved individuals, n=21).

	<i>L. biscarensis</i>			<i>L. callensis</i>			<i>L. leptopogon</i>		
	range	mean	SD	range	mean	SD	range	mean	SD
Standard length (mm)	50–305			44–334			76–175		
In percent of standard length									
Body depth at dorsal–fin origin	21.1–28.8	25.3	2.0	22.0–30.9	26.5	2.3	20.2–27.0	24.4	2.0
Head length	25.5–28.8	26.8	0.7	24.3–29.8	26.4	1.2	23.9–28.3	25.9	0.7
Pre–dorsal length	51.9–57.4	54.4	1.2	49.4–55.2	52.0	1.3	48.8–52.6	51.1	0.9
Pre–pelvic length	50.0–56.5	54.3	1.4	50.6–56.9	53.0	1.3	51.1–57.1	54.1	1.4
Pre–anal length	73.0–81.2	77.2	1.9	73.5–81.4	77.1	1.7	73.6–81.6	78.1	2.2
Dorsal fin base length	11.3–15.4	13.3	1.0	12.1–15.8	13.9	0.9	13.9–15.8	14.6	0.6
Anal fin base length	6.0–8.6	7.5	0.5	6.5–8.4	7.3	0.5	6.8–10.2	8.4	1.0
Anal fin length	17.3–20.0	18.7	0.6	16.9–19.0	17.9	0.4	19.3–21.9	20.5	0.6
Pelvic fin length	12.3–17.8	15.3	1.3	13.9–20.5	16.4	1.5	16.3–20.0	17.8	1.1
Pectoral fin length	16.1–21.2	18.3	1.1	17.7–23.1	20.4	1.2	17.9–22.9	20.2	1.3
Caudal peduncle length	16.5–19.3	17.6	0.7	15.1–20.5	17.3	0.9	15.0–19.5	17.3	1.0
Caudal peduncle depth	11.0–13.3	11.8	0.7	11.0–14.1	12.6	0.5	10.5–12.9	11.8	0.5
Distance between dorsal and anal–fin origins	27.3–36.4	31.3	2.0	29.7–39.3	34.4	1.7	30.1–38.2	33.7	1.7
Distance between pectoral and pelvic–fin origins	21.8–30.2	27.4	1.6	22.5–29.3	26.6	1.4	25.6–31.2	28.2	1.6
Distance between pelvic and anal–fin origins	19.0–27.1	23.0	1.7	20.4–27.7	24.1	1.7	20.6–26.9	24.0	1.6
In percent of head length									
Snout length	30–39	34.5	2.2	24–41	33.8	4.8	29–34	32.1	1.2
Eye diameter	9–21	15.6	4.0	11–28	16.6	5.0	17–24	20.6	1.5
Post–orbital length	44–53	49.9	2.7	44–50	47.6	1.0	45–50	47.4	0.9
Interorbital distance	31–39	35.5	1.9	32–42	36.9	1.9	36–39	38.0	0.9
Pectoral–fin length	61–78	68.2	4.1	69–84	77.2	3.3	63–90	78.0	5.7
Caudal peduncle depth	41–47	43.8	1.7	40–53	47.8	2.8	41–59	45.7	2.1

Comparative material

Numbers in brackets correspond to Figure 2.

Luciobarbus biscarensis: FSJF 3286, 31, 69–190 mm SL; Algeria: Biskra prov.: Oued el Abiod above damlake at Oued Lahbal, 34.896365°N 5.91696°E (40).—FSJF 3279, 9, 50–122 mm SL; Algeria: Biskra prov.: Oued Loutaya at Manbae, 35.094477°N 5.586152°E (38).

Luciobarbus callensis. FSJF 3014, 6, 35–97 mm SL; Tunisia: Oued el Abid about 2 km west of Erritiba, 36.867286°N 10.724574°E (2).—FSJF 3011, 4, 36–94 mm SL; Tunisia: Oued Ghezala 1 km south of Fernana, 36.643868°N 8.699214°E (5).—FSJF 3017, 20, 60–210 mm SL; Tunisia: Kébir River below Zouitina reservoir, 36.732208°N 8.529498°E (6).—FSJF 3008, 9, 71–145 mm SL; Tunisia: Oued Zahzah 2 km south of Bechechema, 35.821259°N 10.159296°E (3).

Luciobarbus leptopogon: FSJF 3284, 9, 97–128 mm SL; Algeria: stream Meliji at Hammam Melouane, 36.489275°N 3.050077°E (15).—RMCA-2016-024-P-0001-0009: 9, 76–133 mm SL; Algeria: Blida prov.: Oued Hammam Melouane, 36.4829°N, 3.0414°E (16).

Luciobarbus mascarensis: FSJF 3283, 4, 74–121 mm SL; Algeria: Oued el Hammam upriver of Hacine, 35.444265°N 0.035043°E (22).

Luciobarbus rifensis: FSJF 3335, 19, 80–208 mm SL; Morocco: Laou River near Chefchaouene, 35.168483°N 5.314508°W (31–32).

Luciobarbus setivimensis: FSJF 3289, 15, 66–156 mm SL; Algeria: Oued Zitouna at Tala Khaled, 36.634327°N 5.216879°E (12).—FSJF 3297, 13, 132–225 mm SL; Algeria: Oued Boughzazene at Boughzazene, 36.614407°N 5.361913°E (11).—FSJF 3292, 19, 66–156 mm SL; Algeria: Oued Remila at inflow into Oued Soummam, 36.632246°N 4.74745°E (13).

Luciobarbus yahyaouii: FSJF 3331, 10, 75–166 mm SL; Morocco, Mar chica, Small wadi on road to Kariat Arkmane, 35.052146°N -2.876565°W (28).—FSJF 3313, 19, 67–187 mm SL; Morocco: Oued Za near Guefait, Moulouya, 34.226706°N 2.392343°W (29).



FIGURE 9. *Luciobarbus* species from the African Mediterranean basin. From the top. *Luciobarbus biscariensis*, not preserved, 300 mm SL; Algeria: Oued El-Aboid above reservoir; *Luciobarbus callensis*, FSJF 3017, 250 mm SL; Tunisia: river below Zouitina reservoir; *Luciobarbus guercifensis*, FSJF 3311, 140 mm SL; Morocco: Oued Za near Guefait.



FIGURE 10. *Luciobarbus* species from the African Mediterranean basin. From the top. *Luciobarbus leptopogon*, FSJF 3284, 120 mm SL; Algeria: stream Meliji at Hammam Melouane; *Luciobarbus rifensis*, FSJF 3335, 220 mm SL; Morokko: Oued Laou near Chefchaouene; *Luciobarbus setivimensis*, FSJF 3292, 200 mm SL; Algeria: Oued Remila at inflow into Oued Soummam; *Luciobarbus yahyaouii*, FSJF 3313, 160 mm SL; Morocco: Oued Za near Guefaït.

Material used for molecular analyses

Luciobarbus biscarensis. F01, F05, F06, Algeria: Biskra prov.: Loutaya River at Manbae, 35.094477°N 5.586152°E (38) (GenBank accession number: cyt b: KY828012, KY828013, KY828014; D-loop: KY828070, KY828071, KY828072).—E158, E159, E160, Gh222, Algeria: Biskra prov.: Oued el Abiod above damlake at Oued Lahbal, 34.867736°N 5.926079°E (41) (GenBank accession number: cyt b: KY828009, KY828010, KY828011, KY828015; D-loop: KY828067, KY828068, KY828069, KY828073).—S161, S163, S164, Algeria: M°sila prov.: Ksob River, 35.840216°N 4.572089°E (37) (GenBank accession number: cyt b: KY828019, KY828020, KY828021; D-loop: KY828064, KY8280645, KY828066).—K33, K35, K55, Algeria: Khenchela prov.: El-Arab River at Babar, 35.166192°N 7.020921°E (42) (GenBank accession number: cyt b: KY828016, KY828017, KY828018; D-loop: KY828061, KY828062, KY828063).

Luciobarbus callensis. T31, T32, Algeria: El-Taref prov.: Oued Bou-Namoussa in Seybouse drainage, 36.621504°N 8.057990°E (8) (GenBank accession number: cyt b: KY828028, KY828029; D-loop: KY828074, KY828075).—D139, D142, D147, D151, D153, D155, Algeria: Guelma prov.: Oued Bouhamdane at Hammam Debagh drainage, 36.468478°N 7.229560°E (9) (GenBank accession number: cyt b: KY828022, KY828023, KY828024, KY828025, KY828026, KY828027; D-loop: KY828076, KY828077, KY828078, KY828079, KY828080, KY828081).

Luciobarbus leptopogon. L93, L96, L99, L100, L102, L104, L105, Algeria: Blida prov.: Oued Hammam Melouane, 36.4829°N, 3.0414°E (16) (GenBank accession number: KY828003

KY828004, KY828005, KY828006, KY828007, KY828008; D-loop: KY828055, KY828056, KY828057, KY828058, KY828059, KY828060).

Luciobarbus setivimensis. Z07, Z14, Z30, Algeria: Bordj-Bou-Arreidj prov.: Ain Zada Dam at Ain Taghrout, 36.151209°N 5.159443°E (10) (GenBank accession number: cyt b: KY828052, KY828053, KY828054; D-loop: KY828104, KY828105, KY828106).

Barbus barbatus. B01, B02, Belgium: Moha prov.: Mehaigne River, Meuse Basin, 50.5451°N 5.1164°E. River drainage.

Materials examined but not preserved.

Luciobarbus biscarensis: 24, 242–308 mm SL; Algeria: Khenchela prov.: Oued el Arab at Babar, 35.166192°N 7.020921°E (42).

Luciobarbus callensis: 2, 313–334 mm SL; Algeria: El-Taref prov.: Oued Bou-Namoussa in Seybouse drainage, 36.621504°N 8.057990°E (8).—19, 205–292 mm SL; Algeria: Guelma prov.: Oued Bouhamdane at Hammam Debagh drainage, 36.468478°N 7.229560°E (9).

Luciobarbus leptopogon: 21, 82–175 mm SL; Algeria: Blida prov.: Oued Hammam Melouane, 36.4829°N, 3.0414°E (16).

Luciobarbus setivimensis: 10, 187–363 mm SL; Algeria: Bordj-Bou-Arreidj prov.: Ain Zada Dam at Ain Taghrout, 36.151209°N 5.159443°E (10).

Acknowledgements

We thank all of those who contributed to this research: Nacer Tarai (Biskra University, Algeria), Mokhtar Kadjam (Director of Ouizert Dam, Mascara, Algeria), Abdelkader El-Najar, Fatah Boujamaa and Mourad Boujamaa (Algeria), Kai Borkenhagen (Büsum). We are also grateful for the assistance provided by Güse Serkan and Sebastian Hüllen (ZFMK). Emmanuel Vreven, Miguel Parrent, Jonathan Brecko and Aurore Mathys (RMCA) are thanked for their time and the pictures. We also thank Didier Van den Spiegel and Rudy Jocqué (RMCA) for their pertinent comments. Lastly, we thank Victoria Grace (www.english-publications.com) for editing the manuscript. This study was supported by the FREDIE project, supported by the Leibniz Association Joint Initiative for Research and Innovation (SAW). It is publication number BRC 390 of the Biodiversity Research Center, Université Catholique de Louvain, Belgium.

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