Capoeta coadi, a new species of cyprinid fish from the Karun River drainage, Iran based on morphological and molecular evidences (Teleostei, Cyprinidae)

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Abstract
As presently recognized, the genus Capoeta includes 24 species, nine of which are known to occur in Iran (C. aculeata, C. capoeta, C. buhsei, C. damascina, C. fusca, C. heratensis, C. mandica, C. saadii and C. trutta) and are distributed in almost all Iranian basins except Sistan and Mashkid. Capoeta coadi sp. n. is a new species from the Karun River, southern Iran, draining into the Arvand Rud (Shatt al-Alar) which drains into the Persian Gulf. It is distinguished from all other species of Capoeta by the combination of the following characters: elongate and usually cylindrical body; 8–9 branched dorsal-fin rays; last unbranched dorsal-fin ray weakly to moderately ossified and serrated along 1/3–2/3 of its length; scales small; 70-84 in lateral line (total); 12–17 scales between dorsal-fin origin and lateral line; 9-11 scales between anal-fin origin and lateral line; 26–32 circum-peduncular scales; 10–13 gill rakers on lower limb of first gill arch; 45–47 total vertebrae; one posterior pair of barbels; bright golden-greenish or silvery body coloration in life; length of the longest dorsal-fin ray 15–22% SL; head length 23–26% SL; mouth width 7–10% SL. Capoeta coadi is also distinguished from all other congeners in the Iranian drainages by fixed diagnostic nucleotide substitutions in the mtDNA COI barcode region and cyt b. It is nested in the Capoeta damascina species complex.

Keywords
Capoeta damascina species complex, COI, Cyt b, Persian Gulf, phylogenetic relationships
Introduction

The Middle East is a transition zone between three major biogeographic units, the Palaearctic, the Afrotropical, and the Oriental realms. It served as an important crossroad of biotic exchange resulting in an outstanding biological diversity of freshwater fishes (Durand et al. 2002, Krupp et al. 2009). Lying between major drainages of the Nile in Africa to the west, the Indus in southern Asia to the east and the Caspian and Black Sea drainages to the north, the Tigris-Euphrates River drainage is the largest river system in the Middle East and has high fish diversity, especially in cyprinid fishes.

*Capoeta* Valenciennes in Cuvier and Valenciennes 1842 is an example of a cyprinid genus widely spread in the Middle East (Krupp and Schneider 1989). Being found in a wide range of habitats, species of this genus display considerable morphological variability (e.g., scale counts and colour pattern) and the extent of morphological plasticity and genetic variability remain to be determined. As a consequence, there has been considerable disagreement regarding the status of several species. However, *Capoeta* is considered monophyletic (Krupp 1985, Küçük et al. 2009).

Members of the genus *Capoeta* are cyprinids characterized by having an elongate, cylindrical body and a short dorsal fin. They have three to five unbranched and 5–9 branched dorsal-fin rays, the last unbranched ray being ossified and serrated. All species have three unbranched and 5 branched anal-fin rays. Scales are usually small. Mouth is inferior and the lower lip is covered with a horny sheath. One pair of barbels (rarely two) is present and the pharyngeal teeth are arranged in three rows. The shape of the mouth as well as the pharyngeal teeth are nearly identical in all species, which indicate their adaptation to the same mode of feeding. This combination of character states distinguishes *Capoeta* from all other cyprinids (Krupp 1985, Krupp and Schneider 1989).

As presently recognized, the genus *Capoeta* includes about 24 species (Eschmeyer and Fricke 2016) in different phylogenetic groups widely distributed in many river drainages and basins in southwestern Asia except the Arabian Peninsula (Alwan 2011, Levin et al. 2012). Levin et al. (2012) studied the phylogenetic relationships of the genus *Capoeta* based on complete mitochondrial gene for cytochrome *b* sequences obtained from 20 species from the overall range of the genus. Three main groups were detected: the Mesopotamian group (*Capoeta trutta* group), the Anatolian-Iranian group (*Capoeta damascina* group) and the Aralo-Caspian group (*Capoeta capoeta* group).

Members of the *Capoeta damascina* species group, characterized by having small scales, include *C. buhsei* Kessler, 1877, *C. caelestis* Schöter, Özuluğ & Freyhof, 2009, *C. damascina* (Valenciennes, 1842), *C. kossuwigi* Karaman, 1969, *C. saadii* ( Heckel, 1847), and *C. umbla* (Heckel, 1843) (Alwan 2011). Based on phylogenetic analyses of cytochrome c oxidase subunit I (COI) and the large subunit (LSU or 28S) ribosomal RNA gene sequences Alwan (2011) identified two main lineages within what we will refer to in this paper, as the “*C. damascina* species complex”. A western lineage is represented by *C. caelestis*, *C. damascina* and *C. umbla* and an eastern lineage represented by *C. buhsei*, *C. saadii*, and a new undescribed species.
Traditionally, *C. damascina* is recorded from Tigris, Mond, Kor, Esfahan, Dasht-e Kavir, Namak Lake, Kor River, Lake Maharlu, Persian Gulf (now Persis), Kerman-Na’in, Dasht-e Lut, Sirjan, Hormuz, and Hamun-e Jaz Murian basins in Iran (Nikol’skii 1899, Berg 1949, Kähsbauer 1964, Armantrout 1980, Rainboth 1981, Bianco and Banarescu 1982, Ghorbani Chafi 2000, Jalali et al. 2005, Esmaeili et al. 2010, Bahrami Kamangar et al. 2012). Its distribution over such wide range of isolated water bodies, raises questions regarding the status of *C. damascina*. Currently, *C. damascina* s.l. represents a complex of closely related species with high intraspecific and comparatively low interspecific variation (Alwan 2011, Levin et al. 2012). Now, three species of *Capoeta* from Iranian water bodies are recognized as being members of *C. damascina* species complex group: *C. buhsei*, *C. saadii* (Iranian populations were considered as *C. damascina*) (see Alwan 2011, Levin et al. 2012), and a new undescribed species from the Karun (Karoun) River drainage. It is described here as a new species, *Capoeta coadi*.

**Material and methods**

After anaesthesia, fishes were either fixed in 5% formaldehyde, and stored in 70% ethanol, or directly fixed in 99% ethanol (for molecular studies). Measurements were made with a digital caliper and recorded to 0.01 mm. All measurements were made point to point, and never by projections. Methods for counts and measurements follow Hubbs and Lagler (1958) and Krupp (1983). 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. The last two branched rays articulating on a single pterygiophore in the dorsal and anal fins are counted as “1½”. The holotype is included in the calculation of means and SD.

Abbreviations used: SL, standard length; HL, lateral head length.

Abbreviations used for museum collections: Zoological Museum of Shiraz University, Collection of Biology Department, Shiraz, Iran (ZM-CBSU), the Senckenberg Research Institute and Natural History Museum (SMF: Frankfurt, Germany), and the private collection of Jörg Freyhof (FSJF: Fischsammlung J. Freyhof).

**DNA extraction and PCR amplification protocol**

For DNA sequencing, specimens were directly fixed in 99% molecular grade ethanol. Mitochondrial DNA was extracted using Salt method (Bruford et al. 1992). The standard vertebrate DNA barcode region of the COI (cytochrome c oxidase subunit 1) and cytochrome *b* (*cyt b*) were amplified using primer pairs named FishF1-5’TCAACCAACCACAAAGACATTGGCAC3’ and FishR1-5’TAGACTTCTGGGTGGCCAAAGAATCA3’ (Ward et al. 2005) and L14724-
5’GTGACTTTGAAAAACCACCGTTG3’ and H15915-5’CAACGATCTCCGTTTTAGAAGAC3’ (Xiao et al. 2001) or GluF- 5’AACCACCGTTGTATTCAACTACAA3’ and H-15560 5’TAGGCRAATAGGAAR TATCA3’ (Machordom and Doadrio 2001), respectively.

Purification and sequencing of the PCR products were conducted at Macrogen Korea Laboratories using the aforementioned primer sets.

Molecular data analyses

Data processing and sequence assembly was done in BioEdit 7.2.5 (Hall 1999); MEGA6 (Tamura et al. 2013) was used to create a DNA sequence alignment. No indications of unexpected stop-codons or nuclear copies of mitochondrial fragments occurred in any sequences. All generated DNA barcodes and cyt b were deposited in the NCBI GenBank. The most appropriate sequence evolution model for the given data was determined with Modeltest (Posada and Crandall 1998) as implemented in the MEGA6 software, treating gaps and missing data with the partial deletion option under 95% site coverage cut-off. The model with the lowest BIC (Bayesian Information Criterion) score is considered the best model to describe the substitution pattern for each gene. To explore species phylogenetic relationships, trees were generated using Maximum Likelihood analysis with 10,000 bootstrap replicates in RaxML 7.2.5 (Stamatakis 2006) under the GTR+G model of nucleotide substitution, with fast bootstrap and also Bayesian analysis (BA), using the Markov Chain Monte Carlo method (MCMC), with 6,000,000 generations under the most generalizing model (GTR+G+I) using Mr. Bayes 3.1.1 (Huelsenbeck and Ronquist 2001). Screening for diagnostic nucleotide substitutions was performed manually from the sequence alignment. As an appropriate outgroup to root the constructed phylogenetic hypothesis, we included the distantly related Cyprinus carpio.

Results

Morphological assessments

Capoeta coadi sp. n.
http://zoobank.org/4B5B0984-0C65-4B6D-97CC-31245E179D13
Figs 1–3

Holotype. ZM-CBSU Z190, 157 mm SL; Iran, Kohgiluyeh and Boyer Ahmad prov., Beshar (Bashar) River at Tale Gah village, Karun River drainage, 30°47′27″N, 51°25′13″E.

Paratypes. ZM-CBSU Z191, 6, 91–157 mm SL; same data as holotype. ZM-CBSU J520, 1, 107 mm SL; ZM-CBSU Z275, 12, 105–152 mm SL; Iran, Koh-
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giluyeh and Boyer Ahmad prov., Beshar (Bashar) River at Tale Gah village, Karun River drainage, 30°47'27"N, 51°25'13"E. 15 December 2014, G. Sayyadzadeh, R. Khaefi, A. Khajehpanah. ZM-CBSU J526, 1, 98 mm SL; ZM-CBSU J533, 1, 114 mm SL; ZM-CBSU J535, 1, 97 mm SL; ZM-CBSU J540, 1, 67 mm SL; All from Iran, Kohgiluyeh and Boyer Ahmad prov., Beshar River at Tange sorkh, Karun River drainage, 30°26'14"N, 51°45'48"E. 24 July 2011, R. Zamaneian Nejad, S. Mirgheiasi, S. Ghasemian. ZM-CBSU J444, 2, 73–90 mm SL; ZM-CBSU J447, 2, 76–111 mm SL; ZM-CBSU J450, 1, 86 mm SL; ZM-CBSU J452, 1, 107 mm SL; ZM-CBSU J459, 2, 104–120 mm SL; ZM-CBSU J464, 1, 110 mm SL; all from Iran, Kohgiluyeh and Boyer Ahmad prov., Beshar River at Mokhtar village, Karun River drainage, 30°40’31"N, 51°31’26"E. 25 May 2011, R. Zamaneian Nejad.

Additional material. ZM-CBSU 7880–7881, 2, 96.69–158.12 mm SL; Iran, Fars prov., Sepidan city, Gorgu River, a tributary of Beshar River, north of Sepidan city, Karun River drainage, 30°21.283’N, 51°45.754’E. 2006. H.R. Esmaeili, A. Teimori, M. Ebrahimian and A. Gholamhoseini. SMF 33337, 1, 48.86 mm SL; Iran, Lorestan prov., Hadi River between Zagheh and Polehoru, 33°31.138’N, 48°46.340’E. 04 March 2008. N. Alwan, K. Borkenhagen, M. Ghanbari Fardi and A. Kazemi. FSJF 2213, 11, 107.92–143.94 mm SL; Iran, Chaharmahal and Bakhtiari Prov., Sandgan River (Sandgan stream) at Sandgan, 31°15.692’N, 51°17.150’E. 19 April 2007, A. Abdoli and J. Freyhof. FSJF 2233, 2, 156.22–162.23 mm SL; Iran, Kohgiluyeh and Boyer Ahmad prov., Beshar River, 20 km northeast of Yasuj, 30°44.152’N, 51°29.522’E. 19 April 2007. A. Abdoli and J. Freyhof. SMF 30865, 1, 26.94 mm SL; Iran, Kohgiluyeh and Boyer Ahmad prov., Beshar River at Tang-e Sorkh, 30°27.680’N, 51°44.907’E. 28 November 2007, K. Borkenhagen, H. R. Esmaeili and F. Wicker (in 96% alcohol). SMF 30871, 1, 28.34 mm SL; Iran, Kohgiluyeh and Boyer Ahmad prov., Beshar River at Tang-e Sorkh, 30°27.680’N, 51°44.907’E. 28 November 2007. K. Borkenhagen, H. R. Esmaeili and F. Wicker (in 96% alcohol). SMF 30872, 1, 29.70 mm SL; Iran, Kohgiluyeh and Boyer Ahmad prov., Beshar River at Tang-e Sorkh, 30°27.680’N, 51°44.907’E. 28 November 2007, K. Borkenhagen, H. R. Esmaeili and F. Wicker (in 96% alcohol).
Figure 2. Capoeta coadi sp. n., paratypes: a ZM-CBSU Z191; 157 mm SL b ZM-CBSU Z192, 148 mm SL; Iran: Kohgiluyeh and Boyer Ahmad, Beshar River, Karun River drainage.

Capoeta coadi specimens used for molecular genetic analysis. ZM-CBSU M1275, 1, Iran, Kohgiluyeh and Boyer Ahmad prov., Beshar River at Dehno village, Karun River drainage, 30°38’55"N, 51°37’05"E. 16 January 2014. H.R. Esmaeili, G. Sayyadzadeh, H.R. Mehraban, M. Razbani. GenBank accession number: (COI: KU564296); ZM-CBSU M1447, 2, GenBank accession number: (COI: KU564297, KU564298; cytb: KU564303, KU564304) ZM-CBSU M1458, 2); Iran, Kohgiluyeh and Boyer Ahmad prov., Beshar River at Tale Gah village, Karun River drainage, 30°47’27"N, 51°25’13"E. 14 December 2013. G. Sayyadzadeh, A. Khajehpanah, R. Khaefi. GenBank accession number: (COI: KU564294, KU564295; cytb: KU564305, KU564306).

Diagnosis. Capoeta coadi sp. n. is distinguished from all other species of Capoeta by the following combination of characters: last unbranched dorsal-fin ray weakly to moderately ossified and serrated in 1/3–2/3 of its length; scales small, 70–84 total lateral line scales (84 in holotype), 12–17 scales between dorsal-fin origin and lateral line (16 in holotype), 9–11 scales between anal-fin origin and lateral line (11 in holotype), 26–32 encircling least circumference of caudal peduncle (31 in holotype); total gill rakers 14–18 (17 in holotype), 10–13 gill rakers on lower limb of first gill arch (12 in holotype); 45–47 total vertebrae; one posterior pair of barbels; length of the longest dorsal-fin ray 14.92–21.58% SL (18.90 in holotype); head length 22.87–26.33% SL (23.76 in holotype); mouth width 7.48–9.77% SL (8.65 in holotype); bright golden-greenish or silvery body coloration in life.
Description. General body shape and appearance are shown in Figs 1–3, morphometric data in Table 1 and meristic data are summarized in Tables 2–9. Body elongate and cylindrical; predorsal body profile smoothly convex with no marked discontinuity between head and body except when a nuchal hump is present in few specimens; greatest body depth at level of dorsal-fin origin; snout rounded (in 20 specimens) or pointed (in 14 specimens) and not size dependent; mouth inferior; lips slightly fleshy, especially at the mouth corners; lower lip covered with a sharp-edged horny sheath, its anterior margin straight in adult specimens and rounded to almost crescent-shaped in juveniles, with a considerable degree of individual variation.

Dorsal-fin origin anterior to pelvic-fin origin, its outer margin usually straight to concave with 3–5 unbranched and 8–9 branched rays (3 and 8 in holotype, respectively); last unbranched dorsal-fin ray weakly to moderately ossified, flexible and soft at the tip, serrated in 1/2–2/3 of its length (Fig. 4); pectoral fins not extending to pelvic-fin base; their outer margins usually slightly convex with 16–22 rays in total (19 in holotype) (Table 2); pelvic fins not extending to anal fin base, their outer margin straight or slightly convex and blunt with 7–11 rays in total (8 in holotype) (Table 2); pelvic axillary scale present; anal fin with 3 unbranched and 5 branched rays, outer margin straight or slightly convex; caudal fin forked with 16–19 branched rays (17 in holotype) (Table 3), its tip pointed and its upper lobe often longer than lower one.

Scales small, total lateral-line scales 70–84; 12–17 scales between dorsal-fin origin and lateral line (Table 4); 9–11 scales between anal-fin origin and lateral line (Table 4); 26–32 circum-peduncle scales (Table 5); ventral midline and pectoral region covered with deeply embedded scales of reduced size; gill rakers slightly hooked, total gill rakers 14–18 (10–13 gill rakers on lower limb) of first gill arch (Table 8–9); 45–47 total vertebrae; usually one posterior pair of barbels present (very rarely two, 1 out of 51 individual); pharyngeal teeth arranged in 3 rows in the following manner: 2.3.5–5.3.2 and very similar in shape to those of C. damascina; teeth in the main row spatulate or spoon-shaped and crowns flat, narrow and curved.
Table 1. Morphometric data of *Capoeta coadi* sp. n. (holotype ZM-CBSU Z190, and 33 paratypes), *C. buhsei* and *C. saadii*.

|                      | Holotype | Paratypes (n=33) | Capoeta buhsei (n=27) | Capoeta saadii (n=20) |
|----------------------|----------|------------------|-----------------------|-----------------------|
| **Standard length (mm)** | 157.64   | 67.23–157.64     | 110.67                | 74.30–149.30          |
| **In percent of standard length** |          |                  |                       |                       |
| Head length           | 23.76    | 22.87–26.33      | 23.15                 | 21.47–25.98           |
| Body depth at dorsal-fin origin | 21.82    | 21.33–25.04      | 21.82                 | 19.78–24.55           |
| Predorsal length      | 49.07    | 47.75–53.43      | 51.59                 | 48.85–55.05           |
| Postdorsal length     | 54.13    | 54.13–63.19      | 55.24                 | 48.20–60.13           |
| Preamal length        | 72.45    | 70.22–76.14      | 74.01                 | 71.34–76.34           |
| Prepelvic length      | 53.74    | 50.22–55.90      | 53.58                 | 50.17–56.64           |
| Distance between pectoral and pelvic-fin origins | 32.42    | 27.81–32.42      | 31.30                 | 29.07–33.64           |
| Distance between pelvic and anal-fin origins | 21.48    | 19.31–23.17      | 21.12                 | 19.90–23.65           |
| Depth of caudal peduncle | 10.37    | 10.03–11.61      | 10.05                 | 8.58–10.84            |
| Length of caudal peduncle | 20.73    | 17.16–22.35      | 19.85                 | 18.64–22.01           |
| Dorsal-fin base length | 12.71    | 12.27–16.17      | 14.41                 | 11.75–15.28           |
| Anal-fin base length  | 6.78     | 6.38–8.85        | 7.39                  | 6.96–8.80             |
| Pectoral-fin length   | 17.32    | 16.68–20.46      | 18.43                 | 16.39–20.96           |
| Pelvic-fin length     | 15.05    | 14.24–16.96      | 15.61                 | 13.85–18.08           |
| Length of the longest dorsal fin ray | 18.90    | 14.92–21.58      | 19.57                 | 16.42–21.22           |
| Mouth width           | 8.65     | 7.48–9.77        | 8.63                  | 6.49–8.89             |
| **In percent of head length** |          |                  |                       |                       |
| Head depth at eye     | 56.88    | 49.05–61.87      | 54.21                 | 48.01–56.63           |
| Snout length          | 38.32    | 31.60–47.70      | 38.08                 | 32.69–38.89           |
| Postorbital distance  | 48.83    | 33.82–51.84      | 48.01                 | 47.66–56.59           |
| Interorbital width    | 40.04    | 34.62–42.81      | 38.19                 | 33.88–41.49           |
| Eye diameter          | 15.97    | 15.07–23.57      | 18.52                 | 13.91–24.44           |
| Maximum head width    | 60.53    | 51.75–66.89      | 59.60                 | 57.83–69.68           |
| Barbel length         | 15.14    | 13.30–20.20      | 16.25                 | 15.66–24.60           |
**Table 2.** Number of pectoral and pelvic fin rays in examined *Capoeta* species.

| Pectoral fin rays | Pelvic fin rays |
|-------------------|------------------|
| 13 14 15 16 17 18 19 20 21 22 |
| 7 8 9 10 11 |
| **C. buhsei** | 2 10 4 6 |
| **C. coadi** | 6 10 8 11 7 1 3 |
| **C. mandica** | 1 7 2 1 |
| **C. saadii** | 3 12 4 1 |
| **C. trutta** | 2 8 17 8 5 |

**Table 3.** Number of branched caudal fin rays in examined *Capoeta* species.

| Branched caudal fin rays | 15 16 17 18 19 20 |
|--------------------------|------------------|
| **C. buhsei** | 3 21 3 |
| **C. saadii** | 1 29 2 1 |
| **C. mandica** | 2 8 1 |
| **C. trutta** | 1 9 16 11 3 |

**Table 4.** Number of scales above (between dorsal-fin origin and lateral line) and below (between dorsal-fin origin and lateral line) lateral line in examined *Capoeta* species.

| Above lateral line | Below lateral line |
|-------------------|------------------|
| 6 7 8 9 10 11 12 13 14 15 16 17 | 5 6 7 8 9 10 11 12 13 |
| **C. buhsei** | 3 6 4 12 |
| **C. coadi** | 1 9 9 15 15 1 |
| **C. mandica** | 1 10 |
| **C. saadii** | 1 2 1 8 7 1 |
| **C. trutta** | 2 1 1 7 16 7 3 3 |

**Coloration.** Live specimens. Dorsum and sides bright golden-green or silvery, darker dorsally and lighter below the lateral line; dorsal head bright golden-green or light pink-brown; dorsal, anal and caudal fins beige to light brown with light pink to red tinge; pectoral and pelvic fins beige to light brown or golden with brown tinge on the first few rays (Fig. 3); few large black blotches present on the body of some specimens whereas small diffuse black spots are present only on the body of some juveniles (above the lateral line).

**Preserved specimens.** Dorsum, head and sides grey or brownish-grey dorsally and beige or yellow ventrally; dorsal and caudal fins dusky grey; pectoral, pelvic and anal fins white or beige with or without grey tinge; blotches and spots well discernible (Figs 1–2).

**Sexual dimorphism.** Breeding tubercles present in both sexes, being bigger and more pronounced in males. Tubercles present on the sides of the snout but may also cover the entire body surface, on and above the lateral line with one or two tubercles per scale but not on each scale, below the lateral line especially in the area above the anal fin and on the branched anal-fin rays; tip of anal fin reaching to or beyond the vertical of the caudal-fin base in females and to about 2/3 of the caudal peduncle in males.
Figure 4. Dorsal fins of Capoeta coadi sp. n. a ZM-CBSU J 444; 73 mm SL b ZM-CBSU Z195; 104 mm SL c ZM-CBSU Z192; 148 mm SL; Iran: Kohgiluyeh and Boyer Ahmad, Beshar River, Karun River drainage, to show size-dependent variability of the last simple dorsal-fin ray serration.

Table 5. Number of circum-pendicular scales in examined Capoeta species.

|     | 19 | 21 | 22 | 23 | 24 | 25 | 26 | 27 | 28 | 29 | 30 | 31 | 32 | 33 |
|-----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|
| C. buhsei | 1  | 3  | 2  | 5  | 5  | 3  | 4  | 3 |
| C. coadi | 3  | 11 | 7  | 14 | 4  | 7  | 5  |
| C. fusca | 1  | 6  | 2  | 4  | 1  | 1  |
| C. mandica | 5  | 1  | 3  | 1  | 1  |

Table 6. Number of caudal-peduncle scales in examined Capoeta species.

|     | 10 | 11 | 12 | 13 | 14 | 15 | 16 | 17 | 18 | 19 | 20 | 21 | 22 |
|-----|----|----|----|----|----|----|----|----|----|----|----|----|----|
| C. buhsei | 6  | 3  | 7  | 9  | 1  | 1 |
| C. coadi | 1  | 1  | 2  | 8  | 15 | 2  | 3  | 1 |
| C. mandica | 4  | 2  | 4  | 1  |
| C. saadi | 2  | 1  | 10 | 6  | 1  |
| C. trutta | 1  | 1  | 2  | 3  | 5  | 14 | 8  | 1  | 4  | 1  |

Habitat and distribution. Capoeta coadi sp. n. occurs in medium-fast flowing rivers with usually gravel substrates and clear waters (Fig. 5). At the Beshar River sampling site, the river is about 25 m wide, with substrate consisting of coarse gravel and boulders, and fast-flowing and semi-transparent waters. The physicochemical parameters at the spot were: dissolved oxygen, 9.89 mg/L; total dissolved solids, 190.2 mg/L; salinity, 0.19‰; conductivity, 395 µs/cm; pH: 8.5 and water temperature 23.4 °C. It is known only from the Karun River drainage, a system that constitutes the southeastern part of the Tigris-Euphrates River system.

Etymology. The new species is named after Brian W. Coad, a well-known ichthyologist for his valuable contribution to the knowledge of freshwater fishes of Iran.

Comparative remarks. The presence of one pair of barbels in Capoeta coadi sets the species apart from C. antalyensis, C. baliki, C. banarescui, C. tinca, and C. heratensis, all of which have two pairs of barbels based on data from Turan et al. (2006a) and this study. The new species is further distinguished from C. antalyensis by the presence of serrae on the last unbranched dorsal-fin ray (vs. absence) (Fig. 4), and by number of scales between dorsal-fin origin and lateral line (12–17 vs. 10–12 in C. antalyensis).
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Figure 5. Beshar River at Taleh Gah village, Karun River drainage, type locality of C. coadi.

(Table 4), between anal-fin origin and lateral line (9–11 vs. 7), and by total number of the lateral-line scales (70–84 vs. 51–57) (Table 7). Capoeta coadi is distinguished from C. banarescui by number of scales between anal-fin origin and lateral line (9–11 vs. 8–9) (Table 4). Data for C. antalyensis and C. banarescui are from Turan et al. (2006a).

Capoeta coadi is distinguished from C. mandica, C. erhani, and C. trutta by having 10–13 gill rakers on the lower limb of the first gill arch (vs. 17–24 in C. mandica, 20–22 in C. erhani and 18–25 in C. trutta [data from Krupp 1985, Turan et al. 2008, Table 8]). The total number of gill rakers in Capoeta coadi specimens is 14–18 that is lower than in C. mandica (23–27), C. barroisi (28–30), C. turani (25–30) and C. trutta (21–31) [data from Turan et al. (2006b), Özuluğ and Freyhof (2008), and this study] Table 9. Capoeta coadi is further distinguished from C. mandica by having fewer pectoral fin rays (16–22 vs. 13–16) (Table 2). Capoeta coadi is distinguished from C. bergamae, C. capoeta and C. sieboldii by number of scales between dorsal-fin origin and lateral line (12–17 in C. coadi vs. 8–10 in C. capoeta and 9–11 in C. sieboldii); number of scales between anal-fin origin and lateral line (9–11 in C. coadi vs. 7–9 in C. bergamae, 6–10 in C. capoeta and 8–10 in C. sieboldii); total lateral line scales (70–84 in C. coadi vs. 48–66 in C. capoeta and 52–60 in C. sieboldii) [data from Banarescu 1999, Turan et al. 2006b, Tables 4, 7]. In addition to the presence of serrae on the unbranched dorsal-fin ray, Capoeta coadi is set apart from C. caelestis by the number of scales between the dorsal-fin origin and lateral line (12–17 in C. coadi vs. 10–13.5
Table 7. Number of lateral-line scales in examined Capoeta species.

|      | 58 | 59 | 61 | 62 | 63 | 64 | 65 | 66 | 67 | 68 | 69 | 70 | 71 | 72 | 73 |
|------|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|
| C. buhsei |    |    |    |    |    |    |    |    |    |    |    |    | 1  |    |    |
| C. coadi   |    |    |    |    |    |    |    |    |    |    |    | 2  | 1  | 2  | 6  |
| C. mandica | 1  | 1  | 2  | 1  | 2  | 1  | 2  | 1  |    |    |    |    |    |    |    |
| C. saadii  | 1  |    |    |    | 2  | 1  | 2  | 2  | 1  | 2  | 3  | 1  |    |    |    |
| C. trutta  | 1  |    |    |    | 2  |    | 1  | 3  | 1  | 5  | 4  | 2  | 2  | 2  |    |

|      | 74 | 75 | 76 | 77 | 78 | 79 | 80 | 81 | 82 | 83 | 84 | 85 | 87 | 89 |
|------|----|----|----|----|----|----|----|----|----|----|----|----|----|----|
| C. buhsei | 3  | 1  | 2  | 1  | 2  | 4  | 3  | 1  | 2  | 4  | 1  | 1  |    |    |
| C. coadi | 1  | 4  | 4  | 1  | 5  | 3  | 5  | 6  | 5  | 1  | 1  |    |    |    |
| C. mandica |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| C. saadii | 1  | 2  | 2  |    |    |    |    |    |    |    |    |    |    |    |    |
| C. trutta | 5  | 3  | 2  | 4  |    |    |    |    |    |    |    |    | 1  |    |    |

in C. caelestis); scales between anal-fin origin and lateral line (9–11 in C. coadi vs. 7–8 in C. caelestis); circum-peduncular scales (26–32 in C. coadi vs. 23–24 in C. caelestis) (Tables 4–5) and probably vertebral counts (45–47 in C. coadi vs. 44 in C. caelestis) [data from Schöter et al. 2009].

It is distinguished from C. damascina by having 11–13, modally 13, gill rakers on the lower limb of the first gill arch (vs. 12–18, modally 14–15) (Alwan 2011, Table 8). Capoeta coadi is clearly distinguished from C. ekmekciae by number of scales between dorsal-fin origin and lateral line (12–17 in C. coadi vs. 9–10 in C. ekmekciae); number of scales between anal-fin origin and lateral line (9–11 in C. coadi vs. 6–7 in C. ekmekciae) (Table 4); number of lateral line scales (70–84 in C. coadi vs. 55–61 in C. ekmekciae) [data from Turan et al. 2006b; Alwan 2011].

Capoeta coadi is distinguished from C. kosswigi by total number of gill rakers (Table 9): 14–18 in C. coadi vs. 19–28 in C. kosswigi (see Karaman 1969; Turan et al. 2006b; Turan 2008).

Capoeta coadi is distinguished from C. mauricii and C. pestai by having a weaker, thinner and less ossified last unbranched dorsal-fin ray in juveniles and adults and fewer scales between dorsal-fin origin and lateral line (12–17 in C. coadi vs. 18–22 in C. mauricii and 16–19 in C. pestai [data from Özuluğ and Freyhof 2008, Küçük et al. 2009]). It is further distinguished from C. pestai by the absence of spots on the body except in juveniles (vs. presence of many on the body [see Özuluğ and Freyhof 2008, Küçük et al. 2009]). Capoeta coadi is distinguished from C. umbla by total number of lateral line scales (70–84 vs. 86–104), number of scales between dorsal-fin origin and lateral line (12–17 vs. 18–24), number of scales between anal-fin origin and lateral line (9–11 vs. 11.5–15.5), and circum-pendicular scales (26–32 vs. 32–39) (see Alwan 2011, Tables 4–7).

Compared to other Iranian species of Capoeta, C. coadi has more scales and fewer gill rakers than C. aculeata (number of scales between dorsal-fin origin and lateral line:}
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Table 8. Gill rakers on the lower limb of the first gill arch in studied Capoeta species.

| GR | 8 | 9 | 10 | 11 | 12 | 13 | 17 | 18 | 19 | 20 | 22 | 24 |
|----|---|---|----|----|----|----|----|----|----|----|----|----|
| C. buhsei | 2 | 19 | 6 |
| C. coadi | 1 | 9 | 19 | 20 |
| C. mandica | | | 1 | 2 | 3 | 1 | 3 | 1 |

Table 9. Number of total gill rakers on the first gill arch in examined Capoeta species.

|  | 12 | 13 | 14 | 15 | 16 | 17 | 18 | 21 | 22 | 23 | 24 | 25 | 26 | 27 | 28 | 31 |
|---|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|
| C. buhsei | 10 | 13 | 6 |
| C. coadi | 1 | 7 | 14 | 6 | 5 |
| C. mandica | | | 2 | 2 | 2 | 1 | 4 |
| C. saadii | 1 | 9 | 6 | 1 | 2 | 1 |
| C. trutta | | | 1 | 1 | 9 | 11 | 7 | 5 | 4 | 1 | 1 |

12–17 vs. 6–10; number of scales between anal-fin origin and lateral line: 9–11 vs. 5–8; circum-peduncular scales: 26–32 vs. 13–23; total number of lateral line scales: 70–84 vs. 36–52; caudal peduncle scales: 14–18 vs. 10–12; gill rakers on the lower limb of the first gill arch: 10–13 vs. 15–18 [data from Coad and Krupp 1994] and this study (Tables 4–9)). *Capoeta coadi* is distinguished from *C. fusca* by more total vertebrae (45–47 vs. 44), and more total lateral-line scales (70–84 vs. 40–62) (see Coad 2008, Johari et al. 2009).

*Capoeta coadi* differs from its sister species (see Figs 6–7), *C. buhsei* in having more gill rakers on lower limb of first gill arch (10–13 vs. 8–10), more gill rakers on the whole first gill arch (14–18 vs. 12–14, see Tables 8–9) and by depth of caudal peduncle in percent of standard length (10.03–11.61 vs. 8.58–10.84). *Capoeta coadi* is distinguished from another closely related species, *C. saadii* by having more scales below the lateral line (9–11 vs. 6–10, modally 9) (Table 4) and more circum-pendicular scales (26–32 vs. 23–28, modally 25–26) [data from Alwan (2011)].

Molecular phylogenetic assessments

We generated COI barcode and *cyt b* sequences for a total of 76 and 61 *Capoeta* specimens, respectively (Tables 10–11). Two phylogenetic approaches including Maximum Likelihood and Bayesian analyses for species of *Capoeta* are given in Figs 6–7. Tables 12–13 provide the diagnostic nucleotide substitutions found in the mtDNA COI barcode region and *cyt b*, respectively.

For inter-specific differences, the greatest pairwise genetic divergence between *C. coadi* and its congeners was found to be 6.5 by *C. erhani* and lowest by *C. buhsei* (0.4) for COI and greatest 9.7 by *C. mandica* and lowest (1.5) by *C. buhsei* for *cyt b* (Tables 14–15).
Figure 6. Bayesian tree inferred from cyt b. Numbers left of the slash, indicate the posterior probabilities of the Bayesian analysis, using MrBayes, while numbers right of the slash are the bootstrap support for 10,000 replicates in the Maximum Likelihood tree, using RaxML. Asterisks (*) indicate less than 50% Maximum Likelihood support for the node.
Figure 7. Bayesian tree inferred from COI. Numbers left of the slash, indicate the posterior probabilities of the Bayesian analysis, using MrBayes, while numbers right of the slash are the bootstrap support for 10,000 replicates in the Maximum Likelihood tree, using RaxML. Asterisks (*) indicate less than 50% Maximum Likelihood support and (-) indicates less than 0.50 Bayesian posterior probabilities for the node.
Table 10. List of species used for molecular analysis for \textit{cyt b} (*present study, the ones without * are obtained from GenBank). \textit{Cyprinus carpio} was considered as outgroup.

| Species      | Accession Number | Locality                                                                 |
|--------------|------------------|--------------------------------------------------------------------------|
| \textit{C. aculeata} | JF798267         | Stream Sangan, Karun River basin, Tigris basin, Iran                     |
| \textit{C. aculeata} | JF798264         | Sevah River, Kor basin, Iran                                             |
| \textit{C. aculeata} | JF798266         | Beshar River, Karun basin, Tigris basin, Iran                           |
| \textit{C. aculeata} | JF798265         | Sevah River, Kor basin, Iran                                             |
| \textit{C. angorea}  | JF798268         | Pozanti River, Mediterranean Sea basin, Turkey                          |
| \textit{C. antalyensis} | JF798269       | Boga Cayi River, Mediterranean Sea basin, Turkey                        |
| \textit{C. baliki}   | JF798272         | Kizilirmak River, Black Sea basin, Turkey                               |
| \textit{C. baliki}   | JF798273         | Biggest tributary of Kurtbog‘azi dam lake, Sakarya River basin, Turkey  |
| \textit{C. baliki}   | JF798275         | Stream Cakirca, Lake Iznik basin, Turkey                                |
| \textit{C. baliki}   | GQ424019         | Unknown                                                                  |
| \textit{C. baliki}   | GQ424020         | Unknown                                                                  |
| \textit{C. baliki}   | JF798271         | Kizilirmak River, Black Sea basin, Turkey                               |
| \textit{C. banarescui}| GQ423987        | Unknown                                                                  |
| \textit{C. banarescui}| GQ423988        | Unknown                                                                  |
| \textit{C. bergamae} | JF798282         | Bakacak stream, Marmara Sea basin, Turkey                               |
| \textit{C. bergamae} | JF798280         | Bakircay River, Turkey                                                  |
| \textit{C. bergamae} | JF798281         | Stream Guzelhisar, Aegean Sea basin, Turkey                             |
| \textit{C. buhsei}   | JF798283         | Taghra Rud stream, Namak Lake basin, Iran                               |
| \textit{C. buhsei}   | KU312369         | Kordan River, Namak Lake basin, Karaj, Iran                             |
| \textit{C. caelestis} | JF798336         | Ilica stream, Gulf of Antalya, Mediterranean Sea basin, Turkey          |
| \textit{C. caelestis} | JF798286         | Goksu River, Mediterranean Sea basin, Turkey                            |
| \textit{C. caelestis} | JF798287         | Kargi Cayi River, Mediterranean Sea basin, Turkey                       |
| \textit{C. coidi}    | KU564303         | Beshar River, Tigris River basin, Iran                                  |
| \textit{C. coidi}    | KU564304         | Beshar River, Tigris River basin, Iran                                  |
| \textit{C. coidi}    | KU564305         | Beshar River, Tigris River basin, Iran                                  |
| \textit{C. coidi}    | KU564306         | Beshar River, Tigris River basin, Iran                                  |
| \textit{C. damascina}| JF798309         | Karadut River, Euphrates basin, Turkey                                  |
| \textit{C. damascina}| JF798303         | Stream Arsuz, Iskenderun Gulf basin, Mediterranean Sea, Turkey          |
| \textit{C. damascina}| JF798308         | Yocaltı River, Turkey                                                   |
| \textit{C. damascina}| JF798306         | Spring İncesu, Orontes basin, Mediterranean Sea, Turkey                 |
| \textit{C. damascina}| JF798307         | Yocaltı River, Mediterranean Sea basin, Turkey                          |
| \textit{C. ekmeckiae}| GQ424027         | Unknown                                                                  |
| \textit{C. heratensis}| JF798319         | Keltechinar River, Turkmenistan                                         |
| \textit{C. heratensis}| JF798318         | Yanbash River, Turkmenistan                                             |
| \textit{C. heratensis}| JF798317         | Yanbash River, Turkmenistan                                             |
| \textit{C. heratensis}| JF798316         | Murgab River, Turkmenistan                                             |
| \textit{C. kousugi}  | JF798322         | Deli Cayi River, Van Lake basin, Turkey                                 |
| \textit{C. kousugi}  | JF798323         | Deli Cayi River, Van Lake basin, Turkey                                 |
| \textit{C. mandica}  | KU564307         | Ghare Aghaj River, Mond River basin, Khaneh Zanian, Iran                |
| \textit{C. mandica}  | KU564308         | Ghare Aghaj River, Mond River basin, Khaneh Zanian, Iran                |
| \textit{C. mandica}  | KU312375         | Ghare Aghaj River, Mond River basin, Khaneh Zanian, Iran                |
Table 11. List of species used for molecular analysis for COI (*present study, the ones without * are obtained from GenBank). *Cyprinus carpio* was considered as outgroup.
| Species        | Accession num. | Locality                                           |
|---------------|----------------|---------------------------------------------------|
| *C. buhsei*    | KU564293       | Roudbar River, Kavir basin, Iran                  |
| *C. caelestis* | KJ552856       | Göksu, Turkey                                     |
| *C. caelestis* | KJ553237       | Ilica, Turkey                                     |
| *C. caelestis* | KJ553301       | Göksu, Turkey                                     |
| *C. damascina* | KJ553080       | Arsus, Turkey                                     |
| *C. damascina* | KJ553043       | Orontes, Turkey                                   |
| *C. damascina* | KJ552896       | Orontes, Turkey                                   |
| *C. damascina* | KJ553272       | Orontes, Turkey                                   |
| *C. damascina* | KJ552846       | Orontes, Turkey                                   |
| *C. damascina* | KJ552874       | Ceyhan, Turkey                                    |
| *C. damascina* | KJ553027       | Ceyhan, Turkey                                    |
| *C. damascina* | KJ553194       | Ceyhan, Turkey                                    |
| *C. damascina* | KJ552763       | Ceyhan, Turkey                                    |
| *C. damascina* | KJ552939       | Jordan River Drainage, Syria                     |
| *C. damascina* | KJ553216       | Orontes, Syria                                    |
| *C. damascina* | KJ553089       | Orontes, Turkey                                   |
| *C. erhani*    | KJ552767       | Ceyhan, Turkey                                    |
| *C. erhani*    | KJ552087       | Ceyhan, Turkey                                    |
| *C. erhani*    | KJ552806       | Ceyhan, Turkey                                    |
| *C. erhani*    | KJ553067       | Ceyhan, Turkey                                    |
| *C. mandica*   | KU564301       | Ghare Aghaj River, Mond River basin, Khanah Zanian, Iran |
| *C. mandica*   | KU564302       | Ghare Aghaj River, Mond River basin, Khanah Zanian, Iran |
| *C. mandica*   | KU312368       | Ghare Aghaj River, Mond River basin, Khanah Zanian, Iran |
| *C. pestai*    | KJ553304       | Egirdir, Turkey                                   |
| *C. pestai*    | KJ553138       | Egirdir, Turkey                                   |
| *C. pestai*    | KJ552113       | Egirdir, Turkey                                   |
| *C. pestai*    | KJ552841       | Egirdir, Turkey                                   |
| *C. pestai*    | KJ552818       | Egirdir, Turkey                                   |
| *C. tinca*     | KJ553229       | Simav, Turkey                                     |
| *C. tinca*     | KJ553168       | Simav, Turkey                                     |
| *C. trutta*    | KU312352       | Karkheh River, Tigris River basin, Seymareh, Iran  |
| *C. trutta*    | KU312351       | Gavi River, Tigris River basin, Illam, Iran       |
| *C. turani*    | KJ553224       | Ceyhan Nehri, Turkey                              |
| *C. saadii*    | KU312358       | Saadii Tomb Spring, Maharlou basin, Iran          |
| *C. saadii*    | KU312395       | Spring Pirbanoo, Maharlou basin, Iran             |
| *C. saadii*    | KU312360       | Helleh River, Helleh basin, KohmarSorkhi, Iran    |
| *C. saadii*    | KU312361       | Helleh River, Helleh basin, KohmarSorkhi, Iran    |
| *C. saadii*    | KU564299       | Kor River, Kor basin, Kamfiruz, Iran              |
| *C. saadii*    | KU564300       | Kor River, Kor basin, Kamfiruz, Iran              |
| *C. saadii*    | KU312359       | Kor River, Kor basin, Kamfiruz, Iran              |
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Table 12. Diagnostic nucleotide substitutions found in mtDNA COI barcode region of Capoeta species. Nucleotide position relative to Cyprinus carpio complete mitochondrial genome.

| Species       | 6545 | 6620 | 6665 | 6818 | 6845 | 6875 | 6905 | 6936 | 6995 | 7076 | 7088 |
|---------------|------|------|------|------|------|------|------|------|------|------|------|
| C. buhsei     | C    | A    | A    | T    | A    | G    | T    | G    | G    | C    | A    | G    | T    |
| C. caelestis   | T    | G    | C    | T    | G    | A    | G    | T    | G    | A    | C    | G    | A    | G    | C    |
| C. coadi      | C    | A    | T    | A    | G    | T    | C    | G    | G    | A    | C    | C    | A    | A    | G    | T    |
| C. damascina  | T    | G    | T    | G    | A    | C    | A    | A    | G    | A    | G    | A    | C    | T    | G    | A    | C    |
| C. saadii     | T    | A    | A    | C    | G    | G    | T    | C    | A    | A    | G    | A    | A    | C    | C    | T    | A    | G    | T    |

Table 13. Diagnostic nucleotide substitutions found in cyt b of Capoeta species. Nucleotide position relative to Cyprinus carpio complete mitochondrial genome.

| Species       | 15430 | 15451 | 15457 | 15463 | 15472 | 15526 | 15550 | 15561 | 15570 | 155814 | 155815 | 155825 | 16011 | 16027 | 16039 | 16045 | 16063 |
|---------------|-------|-------|-------|-------|-------|-------|-------|-------|-------|--------|--------|--------|-------|-------|-------|-------|-------|
| C. buhsei     | C     | G     | T     | A     | A     | G     | G     | T     | G     | G     | G     | C      | C     | A     | G     | C     |
| C. caelestis   | T     | A     | C     | A     | G     | G     | A     | A     | C     | A     | A     | G     | T     | T     | G     | A     | T     |
| C. coadi      | T     | G     | C     | A     | G     | A     | G     | A     | C     | G     | T     | G     | G     | C     | C     | A     | G     | C     |
| C. damascina  | T     | A     | C     | G     | A     | A     | A     | T     | A     | T     | G     | A     | C     | T     | G     | A     | T     |
| C. saadii     | T     | G     | C     | G     | A     | A     | A     | T     | A     | T     | G     | A     | C     | T     | G     | A     | T     |

The two different phylogenetic analyses produced similar topologies. Both analyses produced a tree with 3 major clades (Figs 6–7). These included Clade I) C. antalyensis, C. baliki, C. banarescui, C. boga, C. buhsei, C. caelestis, C. coadi, C. damascina (C. angorae is a synonym [Alwan 2011]), C. kosswigi, C. mauricii, C. pestai, C. saadii, C. sieboldii, and C. tinca, Clade II) C. aculeata, C. ekmeckiae, and C. heratensis, and Clade III) C. barroisi, mandica, C. trutta, and C. turani.

The Iranian members of the C. damascina species complex, clustered together and formed the sister group to the other members in the complex. In these trees, samples of the Capoeta coadi, from Beshar River in Tigris River basin, form a well-supported monophyletic group, sister to C. buhsei in clade I.
Discussion

Based on morphological and molecular results, *C. saadii* and *C. coadi* are distinct species in the *Capoeta damascina* species complex group formerly known as *C. damascina* in Iranian water bodies. Phylogenetic analyses recovered three main groups inside the genus *Capoeta*: the Mesopotamian group (*C. trutta* group), the Anatolian-Iranian group (*C. damascina* group) and the Aralo-Caspian group (*C. capoeta* group) which is in agreement with Levin et al. (2012). The genus *Capoeta* is monophyletic (Levin et al. 2012). Based on the previous published data, the *Capoeta damascina* species complex group diverged from the *C. capoeta* group about 9.1 MYA (95% CI: 6.4–10.9) in the Tortonian period (Levin et al. 2012). Iranian members of the *C. damascina* group (*buhsei, saadii and coadi*) formed a clade sister to other *C. damascina* species complex group members.

The populations of *Capoeta* from the Karun River drainage have long been considered as *C. damascina* (Esmaeili et al. 2010). However, it has been proposed that *C. damascina* might be restricted to the Damascus area in Syria. Most Iranian populations, referred to *C. damascina*, including Karun River population have been considered as *C. saadii* (Heckel, 1847) (Teimori et al. 2016). *Capoeta saadii* was originally described from Persepolis, Pulwar (Sivand) River, Kor River basin, Ruins, northeast of Shiraz,
Table 15. Mean genetic distance for COI gene between *Capoeta* species.

|                | C. trutta | C. heratensis | C. buhsei | C. coadi | C. saadii | C. caelestis | C. damascina | C. barroisi | C. bergmae | C. tinca | C. erhani | C. angore | C. antalyensis | C. mauricii | C. mandica |
|----------------|-----------|---------------|-----------|----------|-----------|-------------|--------------|-------------|-------------|----------|-----------|-----------|----------------|-------------|------------|
| *C. trutta*    |           |               |           |          |           |             |              |             |             |          |           |           |                |             |            |
| *C. heratensis*| 7.15      |               |           |          |           |             |              |             |             |          |           |           |                |             |            |
| *C. buhsei*    | 6.02      | 5.10          |           |          |           |             |              |             |             |          |           |           |                |             |            |
| *C. coadi*     | 6.01      | 5.23          | 0.44      |          |           |             |              |             |             |          |           |           |                |             |            |
| *C. saadii*    | 6.22      | 5.16          | 1.12      | 1.42     |           |             |              |             |             |          |           |           |                |             |            |
| *C. pestai*    | 5.82      | 5.07          | 3.83      | 3.60     | 3.82      |             |              |             |             |          |           |           |                |             |            |
| *C. caelestis* | 6.03      | 4.54          | 2.61      | 3.10     | 2.88      | 4.01        |              |             |             |          |           |           |                |             |            |
| *C. damascina* | 5.65      | 4.31          | 2.56      | 3.05     | 2.49      | 3.65        | 1.24         |             |             |          |           |           |                |             |            |
| *C. barroisi*  | 0.57      | 6.72          | 5.99      | 6.52     | 6.17      | 6.29        | 5.93         | 5.52        |             |          |           |           |                |             |            |
| *C. bergmae*   | 6.56      | 4.87          | 3.95      | 3.81     | 3.98      | 4.33        | 3.64         | 3.45        | 6.75        |          |           |           |                |             |            |
| *C. tinca*     | 4.39      | 5.32          | 4.50      | 4.77     | 4.49      | 5.02        | 4.44         | 4.01        | 4.21        | 4.47     |           |           |                |             |            |
| *C. erhani*    | 0.94      | 6.74          | 6.03      | 6.55     | 6.20      | 6.18        | 5.83         | 5.27        | 0.99        | 6.35     | 4.18      |           |                |             |            |
| *C. angore*    | 6.37      | 4.68          | 3.28      | 3.41     | 3.02      | 3.97        | 1.91         | 0.74        | 6.27        | 3.78     | 4.46      | 5.97      |                |             |            |
| *C. antalyensis*| 5.25     | 4.15          | 2.58      | 2.71     | 2.92      | 2.91        | 2.42         | 2.76        | 5.34        | 2.73     | 3.64      | 5.25      | 3.08          |               |            |
| *C. mauricii*  | 5.82      | 5.07          | 3.83      | 3.60     | 3.82      | 0.00        | 4.01         | 3.65        | 6.29        | 4.33     | 5.02      | 6.18      | 3.97          | 2.91         |            |
| *C. mandica*   | 0.42      | 7.31          | 6.22      | 6.34     | 6.39      | 5.99        | 6.20         | 5.83        | 0.73        | 6.54     | 4.58      | 1.18      | 6.54          | 5.43         | 5.99        |
| *Cyprinus carpio* | 15.32   | 14.89         | 15.13     | 14.97    | 14.23     | 15.85       | 16.32        | 15.39       | 15.53       | 15.57    | 15.45     | 16.06     | 15.34         | 15.61        | 15.85       | 15.87      |
Iran. It was considered as a synonym of *C. damascina* (Esmaeili et al. 2010) and as a valid species by Bianco and Bănărescu (1982), by Levin et al. (2012) and by Teimori et al. (2016). Based on morphological and molecular results presented here, *C. saadii* is a valid species closely related to *C. buhsei* (as proposed by Bianco and Bănărescu (1982) and to *C. coadi* yet is diagnosed from these species and from *C. damascina* (see Alwan 2011). *Capoeta saadii* is the least known species of the genus. It is not mentioned in the revision of the genus by Karaman (1969) who had no specimens available, but its position within the genus *Capoeta* and its close phylogenetic relationship to *C. coadi* and *C. buhsei* were demonstrated using many fresh specimens at our disposal, mostly from type localities.

**Comparative materials used in morphological and molecular phylogenetic analyses**

**Morphological analyses**

*Capoeta buhsei*: ZM-CBSU Z218-229, 12, 104-149 mm SL; Iran, Semnan prov., Kavir basin, Hableh Rud at Gamarsar, 35°18′06″N, 52°24′57″E. 21 August 2011. H.R. Esmaeili, G. Sayyadzadeh, A. Gholamifard, R. Zamaniannejad. ZM-CBSU Z260-274, 15, 88–130 mm SL; Iran, Albourz prov., Kordan River at Karaj, 35°57′12″N, 56°50′18″E. 5 July 2014. M. Masoudi, R. Khaefi. H.R. Mehraban.

*Capoeta fusca*: ZM-CBSU Z197-211, 15, 50–78 mm SL; Iran, south Khorasan prov., Sharifabad Qanat at Birjand, 33°58′08″N, 59°17′03″E. 29 August 2011. H.R. Esmaeili, G. Sayyadzadeh, A. Gholamifard, R. Zamaniannejad.

*Capoeta mandica*: ZM-CBSU Z230-234, 5, 82–130 mm SL; Iran, Fars prov., Qareh Aghaj River at Khaneh Zanian, 29°41′13″N, 52°05′58″E. 30 May 2015. H. Zareian, A. Gholamhosseini, G. Sayyadzadeh. ZM-CBSU Z212-217, 6, 83-118 mm SL; Iran, Fars prov., Qareh Aghaj River at Kavar, 29°10′55″N, 52°41′32″E. 27 February 2015. G. Sayyadzadeh, M. Masoudi.

*Capoeta saadii*: ZM-CBSU Z136-146, 11, 78-121 mm SL; ZM-CBSU 2504, 1, 82 mm SL; ZM-CBSU 2508, 1, 69 mm SL; ZM-CBSU 2520-2521, 2, 51-62 mm SL; ZM-CBSU 2524-2528, 5, 113-231 mm SL; Iran, Fars prov., Ghamargah spring, Doroodzan, 30°15′11″N, 54°25′32″E. 21 December 2003. H.R. Esmaeili, Biglari.

*Capoeta trutta*: ZM-CBSU E100-123, 24, 50-149 mm SL; Iran, Kermanshah prov., Gamasbi River, 34°23′31″N, 47°42′57″E. 27 September 2007. A. Teimori, A. Gholamhosseini, M. Ebrahimi, A. Gholamifard; ZM-CBSU C453-463, 11, 67-177 mm SL; ZM-CBSU C474-477, 4, 67–75 mm SL; ZM-CBSU C481, 76 mm SL; all from Iran, Khuzestan prov., Maroon River at Aghajari, 30°44′52″N, 49°54′59″E. 21 March 2008. H. Zareian.
Molecular phylogenetic analyses

*Capoeta buhsei*; ZM-CBSU M1299-1300, 2, Iran, Albourz prov., Kordan River at Karaj, 35°57'12"N, 56°50'18"E. 5 July 2014. M. Masoudi, R. Khaefi. H.R. Mehraban. GenBank accession number: (COI: KU312349, KU312350; cytb: KU312369, KU312370); ZM-CBSU M1289-1290, 2, Iran: Semnan Prov., Kavir basin, Roudbar River at Mehdishahr, 35°37'56"N, 53°20'41"E. 30 August 2011. H.R. Esmaeili et al., GenBank accession number: (COI: KU564292, KU564293).

*Capoeta heratensis*; ZM-CBSU M813-815, 3, Iran, Razavi Khorasan prov., Gilas spring, 36°36'55"N, 59°20'17"E. 25 August 2011. H.R. Esmaeili et al. GenBank accession number: (COI: KU564288, KU564289, KU564290). ZM-CBSU M816, 1, Iran, Razavi Khorasan prov., Bezangan Lake, Tedzen basin. 36°17'03"N, 60°24'18"E. 25 August 2011. H.R. Esmaeili et al. GenBank accession number: (COI: KU564291).

*Capoeta mandica*: ZM-CBSU M1433-1435, 3, Iran, Fars prov., Qareh Aghaj River at Khaneh Zanian, 29°41'13"N, 52°05'58"E. 30 May 2015. H. Zareian, A. Gholamhosseini, G. Sayyadzadeh. GenBank accession number: (COI: KU564301, KU564302, KU312368; cytb: KU564307, KU564308, KU312375).

*Capoeta saadii*: ZM-CBSU M1426-1427, 2, Iran: Fars prov. Kor River, at Kamfiroz, 30°25'2"N, 52°8'59"E. H. Zareian. 24 October 2015. GenBank accession number: (COI: KU564299, KU564300; cytb: KU564312, KU564313). ZM-CBSU M1421, ZM-CBSU1422-1425, 3, Iran, Fars prov., Qareh Aghaj River at Firuzabad, 28°41'31"N, 52°27'43"E. 25 April 2015. H. Zareian. GenBank accession number: (cytb: KU564309, KU564310, KU564311). ZM-CBSU M157, 1, Iran, Fars prov., Shiraz, Saadii Tomb, Maharlou basin, 29°37.348"N, 52° 34.934'E. R. Khaefi, 2009. GenBank accession number: (COI: KU312358). ZM-CBSU M825, M831, 2, Iran, Fars prov., Helleh River, Helleh basin, KohmarhSorkhi, S. Mirgheiasi, S. Ghasemian. 29°23'39"N, 52°09'49"E. GenBank accession number: (COI: KU312361, KU312360; cytb: KU312373). ZM-CBSU M822, 1, Iran, Fars prov., Qareh Aghaj River at Firuzabad, 29°07'34"N, 52°51'24"E. GenBank accession number: (cytb: KU564310). FSJF DNA-18 Iran: Fars prov.: spring Pirbanoo about 10 km south of Shiraz, 29°31'08"N, 52°27'55"E GenBank accession number: (COI: KU312395). FSJF DNA-22; Iran: Fars prov.: River Kor about 73 km north of Shiraz, 30°11'37"N, 52°27'56"E. GenBank accession number: (COI: KU312359).

*Capoeta trutta*: ZM-CBSU M583, 1, Iran: Ilam prov.; Gavi River at Mehran, H.R. Esmaeili, 13 November 2012, 33°39'18"N, 47°02'14"E. GenBank accession number: COI: KU312351; ZM-CBSU M593, 1, Iran, Ilam prov.; Seymareh River, H.R. Esmaeili, 13 November 2012, 33°38'17"N, 47°01'30"E. GenBank accession number: COI: KU312352.
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