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Revalidation and redescription of *Brachymystax tsinlingensis* Li, 1966 (Salmoniformes: Salmonidae) from China

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Abstract

Brachymystax tsinlingensis Li, 1966 is revalidated and redescribed. It can be distinguished from all congeners by the following combination of characteristics: no spots on operculum; gill rakers 15–20; lateral-line scales 98–116; pyloric caeca 60–71. Unique morphological characters and genetic divergence of this species are discussed. This species has a limited distribution in several streams of the middle part of the Qinling Mountains in China. Methods for management and protection of *B. tsinlingensis* need to be re-evaluated.

Key words: *Brachymystax*, revalidation, redescription, Salmonidae, China

Introduction

The genus *Brachymystax* Günther, 1866, belonging to Salmonidae, Salmoniformes, is distributed in eastern and northern Asia with three currently recognized valid species (Froese & Pauly, 2014): *B. lenok* (Pallas, 1773), *B. tumensis* Mori, 1930, and *B. savinovi* Mitrofanov, 1959.

Li (1966) described a subspecies, *Brachymystax lenok tsinlingensis*, based on specimens collected from rivers on the eastern and southern sides of the Taibai Mountains segment of the Qinling Mountains. He pointed out that this subspecies could be distinguished from *B. lenok lenok* by lower numbers of pyloric caeca (65–75 vs. 91–111), lateral-line scales (115–127 vs. 132–175) and rakers (19–23 vs. 24–26) on the first gill arch (Li, 1966). However, because reference was made to only two specimens of *B. lenok lenok*, the validity of *B. lenok tsinlingensis* was questioned by other ichthyologists (Gao, 1980; Song & Fang, 1984; Song, 1987; Qin & Wang, 1989; Wang, 1988; Ma *et al.* 2009). Therefore, *B. lenok tsinlingensis* has been synonymized with *B. lenok lenok* (Song, 1987; Qin & Wang, 1989; Froese & Pauly, 2014). In fact, the identification of *B. lenok tsinlingensis* remains vague. For instance, Shedko (2001) synonymized it with *B. tumensis*. Kim and Park (2002) identified specimens from the South Korea as *B. lenok tsinlingensis*. The original description of *B. lenok tsinlingensis* in Chinese also has led to confusion among non-Chinese workers.

After examining paratypes of *B. lenok tsinlingensis*, additional comparative specimens, and information from other studies on mitochondrial DNA (Qi *et al.* 2009; Crete-Lafreniere *et al.* 2012; Si *et al.* 2012; Shedko *et al.* 2013), we conclude that *B. lenok tsinlingensis* is a valid species and re-describe it with more data on morphological characteristics. In addition, since the holotype is lost, herein we select a paratype as lectotype.

Materials and methods

We examined 50 specimens of *B. tsinlingensis*, including all paratypes. Comparative materials included 84 and 14

specimens of *B. lenok* and *B. tumensis*, respectively, collected from different rivers in China and Mongolia. Specimens are deposited in the National Zoological Museum, Institute of Zoology, Chinese Academy of Sciences (NZMC), Heilongjiang Fisheries Research Institute, Chinese Academy of Fishery Sciences (HFRI) and Beijing Museum of Natural History (NHB). Detailed specimen information is listed in the Description or Comparative Materials section. Specimens of *B. lenok* and *B. tumensis* from the Altai Mountain area and the Tumenjiang River, China, respectively, were examined. Although types of *B. lenok* and *B. tumensis* were not examined because their whereabouts are unknown, other specimens from both type localities were compared in the present study.

Measurements were made with a digital caliper and data recorded to 0.01 mm. Counts and measurements were taken on the left side of specimens whenever possible. Individual measurements were taken as shown in Figure 1. Photographs were taken using Canon EOS5DII and EOSM Cameras. Statistical analyses were carried out using SPSS Statistics 17.0.

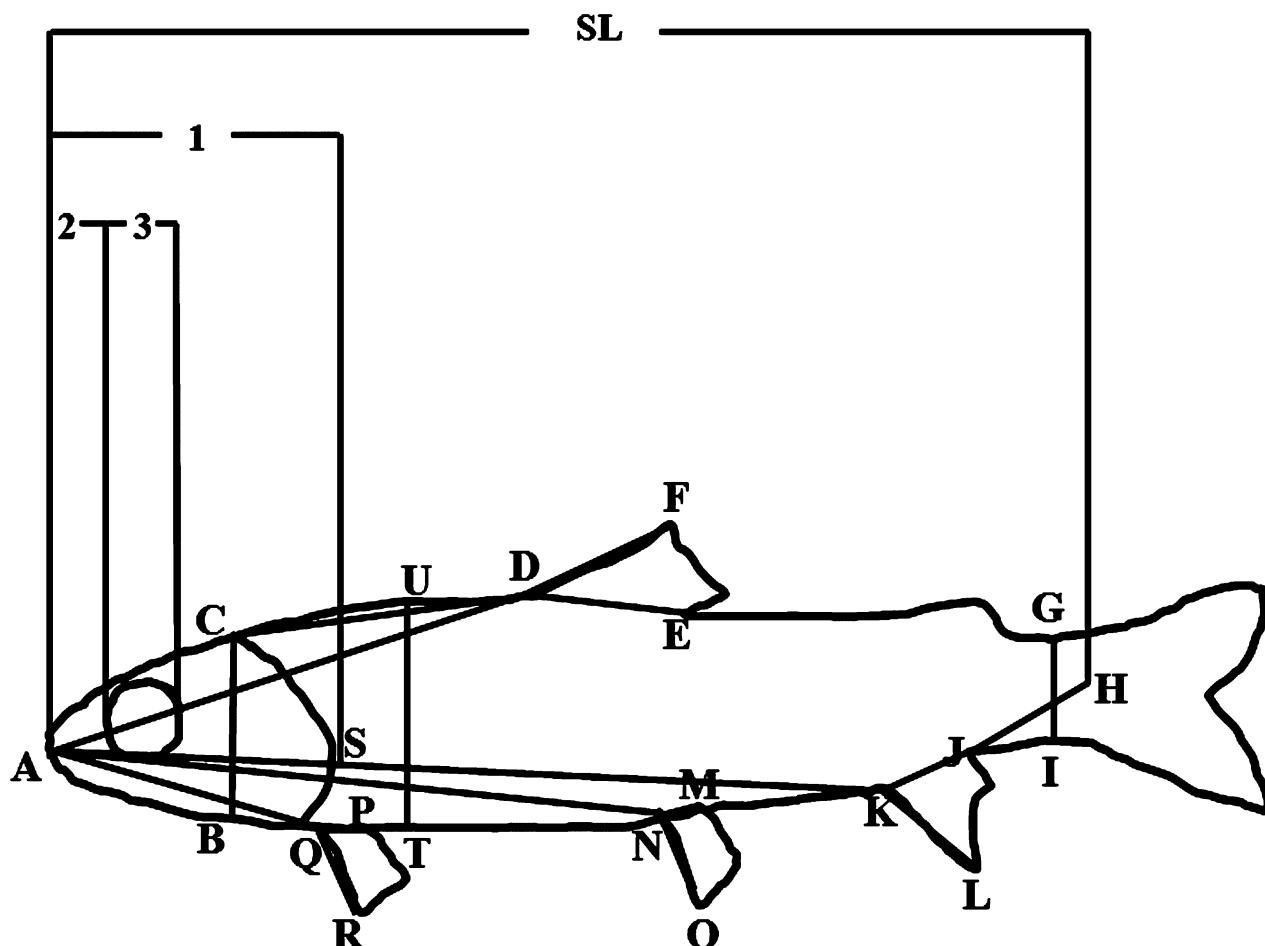


FIGURE 1. Principal measurements taken on species of *Brachymystax*. Drawing of *B. tsinlingensis*. Standard length (SL), from tip of snout to posterior end of last half-centrum (A-H); body depth, from insertion of dorsal fin to ventral midline (U-T); predorsal length, from tip of snout to insertion of dorsal fin (A-D); posterior edge of head to dorsal-fin origin (dorsal scales start here) (C-D); dorsal-fin length, from insertion of dorsal fin to tip of longest ray (D-F); dorsal-fin base length, from anterior to posterior end of dorsal-fin base (D-E); preanal length, from tip of snout to insertion of anal fin (A-K); anal-fin length, from insertion of anal fin to tip of longest ray (K-L); anal-fin base length, from anterior to posterior end of anal-fin base (K-J); prepectoral length, from tip of snout to base of pectoral-fin origin (A-Q); pectoral-fin length, from base of anterior pectoral-fin ray to tip of longest ray (Q-R); pectoral-fin base length, from anterior to posterior end of pectoral-fin base (Q-P); prepelvic length, from tip of snout to base of anterior pelvic-fin ray (A-N); pelvic-fin length, from base of anterior pelvic-fin ray to tip of longest ray (N-O); pelvic-fin base length, from anterior to posterior end of pelvic-fin base (N-M); caudal-peduncle length, from end of anal-fin base to posterior end of last half-centrum (J-H); caudal-peduncle depth, at middle of caudal peduncle (G-I); head length, from tip of snout to most posterior point of operculum (not including skin flap, 1; A-S); head depth, from nape vertically to ventral midline (C-B); head width, distance between most posterior margins of opercle; snout length, from tip of snout to anterior margin of circumorbital series (2); eye diameter, from anterior to posterior margins of circumorbital series (3), pressing slightly to find firm points; interorbital width, shortest distance between orbits across top of head; rictal barbel length, from anterior to posterior end of rictal barbel. [HALF COLUMN]

Complete-mitochondrial DNA sequences and mitochondrial cytochrome *b* gene of *Brachymystax* were downloaded from GenBank. Three species of *Hucho* were included as outgroups (Table 1). The sequences alignment was analyzed in Clustal X1.83 and edited with Seaview. Neighbor-Joining (NJ) trees were built, and pairwise genetic distance and bootstrap confidence values (% of 1000 bootstrap replicates) were calculated, using MEGA 4.1.

TABLE 1. List of species examined, with GenBank accession numbers and references.

| Species | Accession No. | Basin | Reference |
|-----------------------------------|---------------|---|-------------------------------------|
| <i>Brachymystax lenok</i> | JX262003 | Ussuri basin, Russia (Amur basin) | Shedko <i>et al.</i> 2013 |
| | JQ686730 | | Si <i>et al.</i> 2012 |
| | NC_018341 | | Si <i>et al.</i> 2012 |
| | JX960765 | Anui River, Russia (Amur basin) | Crete-Lafreniere <i>et al.</i> 2012 |
| | JX960766 | Leprindokan Lake, Russia (Lena basin) | Crete-Lafreniere <i>et al.</i> 2012 |
| <i>Brachymystax tumensis</i> | JX262002 | Amur River | Shedko <i>et al.</i> 2013 |
| <i>Brachymystax</i> sp. Korea | JQ675732 | Korea | Yu <i>et al.</i> 2013 (unpublished) |
| <i>Brachymystax tsinlingensis</i> | NC_018342 | | Si <i>et al.</i> 2012 |
| | JQ686731 | | Si <i>et al.</i> 2012 |
| <i>Brachymystax savinovi</i> | JX960767 | Amalyk Lake, Russia (Lena basin) | Crete-Lafreniere <i>et al.</i> 2012 |
| | JX960768 | Anui River, Russia (Amur basin) | Crete-Lafreniere <i>et al.</i> 2012 |
| <i>Hucho taimen</i> | JX262004 | Ussuri basin, Russia (Amur basin) | Shedko <i>et al.</i> 2013 |
| | JX960799 | Khor River, Russia (Amur basin) | Crete-Lafreniere <i>et al.</i> 2012 |
| <i>Hucho hucho</i> | JX960797 | Bol'shaya River, Russia (Baikal-Enisei basin) | Crete-Lafreniere <i>et al.</i> 2012 |
| <i>Hucho bleekeri</i> | FJ597623 | Makehe River (upper Yangtze River) | Qi <i>et al.</i> 2009 |



FIGURE 2. *Brachymystax tsinlingensis*, NZMC 51100 (H2057), lectotype, 176.24 mm SL. [HALF COLUMN]

Results

Brachymystax tsinlingensis Li, 1966 (Fig. 2, Table 2)

Brachymystax lenok tsinlingensis Li, 1966: 92 (type locality: Zhouzhi County, Taibai County, Shaanxi Province, China)

Brachymystax lenok: Song, 1987: 14 (Minxian County, Weiyuan County, Zhangxian County, Zhangjiachuan County, Gansu Province; Zhouzhi County, Shaanxi Province, China); Qin & Wang, 1989: 52 (Upper Weihe River, Gansu Province, China)

Brachymystax tumensis: Shedko, 2001: 229

Type specimens. Lectotype: NZMC 51100 (H2057), 176.24 mm SL, Yangtze River basin: Xushuihe River, Hetaoping Village, Taibai County, Shaanxi Province, China P.R., Aug. 23, 1962.

Paratypes: NZMC 51101-51103 (H2058-H2059, H2061), NZMC 51110-51112 (H2063, H2056, H2054), 170.23-207.49 mm SL, Yangtze River basin: Xushuihe River, Hetaoping Village, Taibai County, Shaanxi Province, China P.R., Aug. 23- Sept. 5, 1962; NZMC 51104-51109 (H2064-H2066, H2073-H2075), 187.52-228.53 mm SL, Yellow River basin: Heihe River, Dudumen Village, Zhouzhi County, Shaanxi Province, China P.R., Aug. 23- Sept. 5, 1962.

Other specimens. NZMC 197877-197885 (9), 63.48-170.62 mm SL, Yangtze River basin: Xushuihe River, Dajiangou, Huangboyuan Village, Taibai County, Shaanxi Province, China P.R., Nov. 20, 2012; NZMC 197886, 82.02 mm SL, Yangtze River basin: Hongshuihe River, Hetaoping Village, Taibai County, Shaanxi Province, China P.R., Nov. 21, 2012; NZMC 197888-197892 (5), 85.20-161.62 mm SL, Yangtze River basin: Hongshuihe River, Taibai County, Shaanxi Province, China P.R., Nov. 21, 2012; NZMC 197893-99 (7), 68.53- 255.56 mm SL, Yellow River basin: Xianyihe River, Xianyiguan, Caojiawan Village, Longxian County, Shaanxi Province, China P.R., Nov. 22, 2012; NZMC 197900-197910 (11), 64.65-257.30 mm SL, Yellow River basin: Guanshangou River, Guguan Village, Longxian County, Shaanxi Province, China P.R., Nov. 22, 2012; NZMC 197913-197916 (4), 154.85-178.91 mm SL, Yellow River basin: Sujiahe River, Sujiahe Village, Longxian County, Shaanxi Province, China P.R., Nov. 22, 2012.

Diagnosis. *Brachymystax tsinlingensis* can be distinguished from all congeners by the following combination of characteristics: no spots on operculum; gill rakers 15-20; lateral-line scales 98-116; pyloric caeca 60-71.

Re-description. General body features are shown in Figure 2. Morphometric and meristic characters are listed in Table 2. Body elongated, compressed. Dorsal profile rising slightly from snout tip to dorsal-fin origin, then sloping gently to adipose-fin origin, and then sloping slightly to end of caudal peduncle. Ventral profile horizontal to pelvic-fin origin, then upward clearly to caudal fin. Caudal peduncle moderately compressed. The greatest body depth at dorsal-fin origin, and least depth of caudal peduncle close to caudal-fin base.

Head blunt, top broad, flat at center. Blunt-snouted, lower jaw nearly equal to or slightly shorter than upper jaw. Teeth on the upper and lower jaws. Anterior nostril tubular and short, next to posterior nostril. Eyes large and rounded, nearer to snout tip than to trailing edge of operculum; interorbital broad. Pair of short rictal barbels, shorter than half of eye diameter. Gill opening large, reaching or extending downwards beyond pectoral-fin base. Gill rakers developed and sparse.



FIGURE 3. Live *Brachymystax tsinlingensis*, collected from the type locality. [HALF COLUMN]

TABLE 2. Biometric data for species of *Brachymystax*.

| | <i>Brachymystax tsinlingensis</i> (n=50) | <i>Brachymystax lenok</i> (n=84) | <i>Brachymystax tumanensis</i> (n=14) |
|--------------------------------|--|----------------------------------|---------------------------------------|
| Number of branched rays | | | |
| Dorsal fin | 10-11 | 9-10 | 9-10 |
| Anal fin | 9 | 8-10 | 8-10 |
| Pectoral fin | 15-16 | 14-17 | 15-16 |
| Pelvic fin | 9 | 8-9 | 9 |
| Number of countable characters | | | |
| Gill rakers | 15-20 | 17-29 | 23-25 |
| Lateral-line scales | 98-116 | 111-158 | 111-124 |
| Pyloric caeca | 60-71 | 70-111 | 55-65 |
| Circumpeduncular scales | 34-40 | 34-40 | 32-48 |
| Range(mm) | Mean±SD | Range (mm) | Mean±SD |
| Standard length | 63.48-257.3 | 147.22±53.81 | 76.21-340.36 |
| % standard length | | | |
| Body depth | 10.87-31.05 | 21.66±3.02 | 22.04-38.97 |
| Predorsal length | 38.53-59.28 | 47.78±2.83 | 36.65-62.71 |
| Head to dorsal | 15.34-37.72 | 29.48±3.32 | 25.89-44.88 |
| Dorsal-fin base length | 12.02-19.64 | 15.21±1.37 | 8.97-22.42 |
| Dorsal-fin length | 16.56-24.79 | 21.06±1.66 | 14.30-29.66 |
| Preanal length | 61.18-85.22 | 77.62±3.77 | 68.34-91.41 |
| Anal-fin base length | 8.90-13.07 | 10.71±0.86 | 7.01-12.95 |
| Anal-fin length | 14.37-20.10 | 16.87±1.16 | 12.38-23.01 |
| Prepectoral length | 21.69-33.80 | 26.10±2.98 | 17.33-30.56 |
| | | 23.83±1.88 | 22.17-27.84 |
| | | | 24.77±1.68 |

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TABLE 2. (Continued)

| | <i>Brachymystax tsinlingensis</i> (n=50) | <i>Brachymystax lenok</i> (n=84) | <i>Brachymystax tumensis</i> (n=14) |
|--------------------------|--|----------------------------------|-------------------------------------|
| Pectoral-fin base length | 3.23-6.36 | 4.47±0.83 | 2.72-4.82 |
| Pectoral-fin length | 13.80-21.12 | 17.23±1.57 | 10.93-22.33 |
| Prepelvic length | 51.84-67.65 | 56.7±10.68 | 49.06-74.08 |
| Pelvic-fin base length | 2.79-4.90 | 3.6±0.48 | 2.40-4.57 |
| Pelvic-fin length | 12.08-18.26 | 14.32±1.55 | 7.72-19.42 |
| Caudal-peduncle length | 11.34-16.95 | 14.37±1.18 | 11.50-17.69 |
| Caudal-peduncle depth | 7.96-12.54 | 9.35±0.67 | 6.47-11.55 |
| Head length | 23.28-32.09 | 26.34±1.91 | 19.00-31.90 |
| % head length | | | 24.73±1.57 |
| Head depth | 56.27-83.09 | 65.39±6.09 | 54.93-75.78 |
| Head width | 37.22-51.09 | 42.68±3.80 | 36.00-61.74 |
| Snout length | 21.28-29.76 | 25.32±1.85 | 19.84-30.82 |
| Eye diameter | 22.54-33.02 | 27.71±2.34 | 20.89-33.24 |
| Interorbital width | 20.80-31.12 | 26.37±2.05 | 20.91-31.86 |
| Upper-jaw length | 28.85-44.46 | 39.68±3.25 | 31.99-44.23 |
| Lower-jaw length | 14.48-33.18 | 25.20±4.13 | 18.46-31.32 |
| Mouth width | 22.98-45.21 | 31.03±4.82 | 15.13-34.52 |
| | | | 27.26±3.64 |
| | | | 21.51-32.06 |
| | | | 28.01±2.45 |

Dorsal-fin origin in front of vertical line of pelvic-fin origin, nearer to snout tip than to caudal-fin base; distance between snout tip and dorsal-fin origin nearly equal to distance from posterior end of dorsal-fin base to adipose-fin origin. Dorsal fin short, posterior end of base opposite to pelvic-fin origin. Pectoral fin insertion in vertical through posterior margin of operculum, pectoral fin short, not reaching to upright position of dorsal-fin origin. Pelvic-fin insertion nearer to anal-fin base than to pectoral-fin origin, opposite to third or fourth branched ray of dorsal fin, end not reaching to anus. Anal fin short, insertion nearly at midway between pelvic-fin origin and caudal-fin base, origin nearly opposite to adipose-fin origin. Adipose-fin origin nearer to caudal-fin base than to dorsal-fin base. Caudal-fin bifurcate, upper lobe equal to lower one.

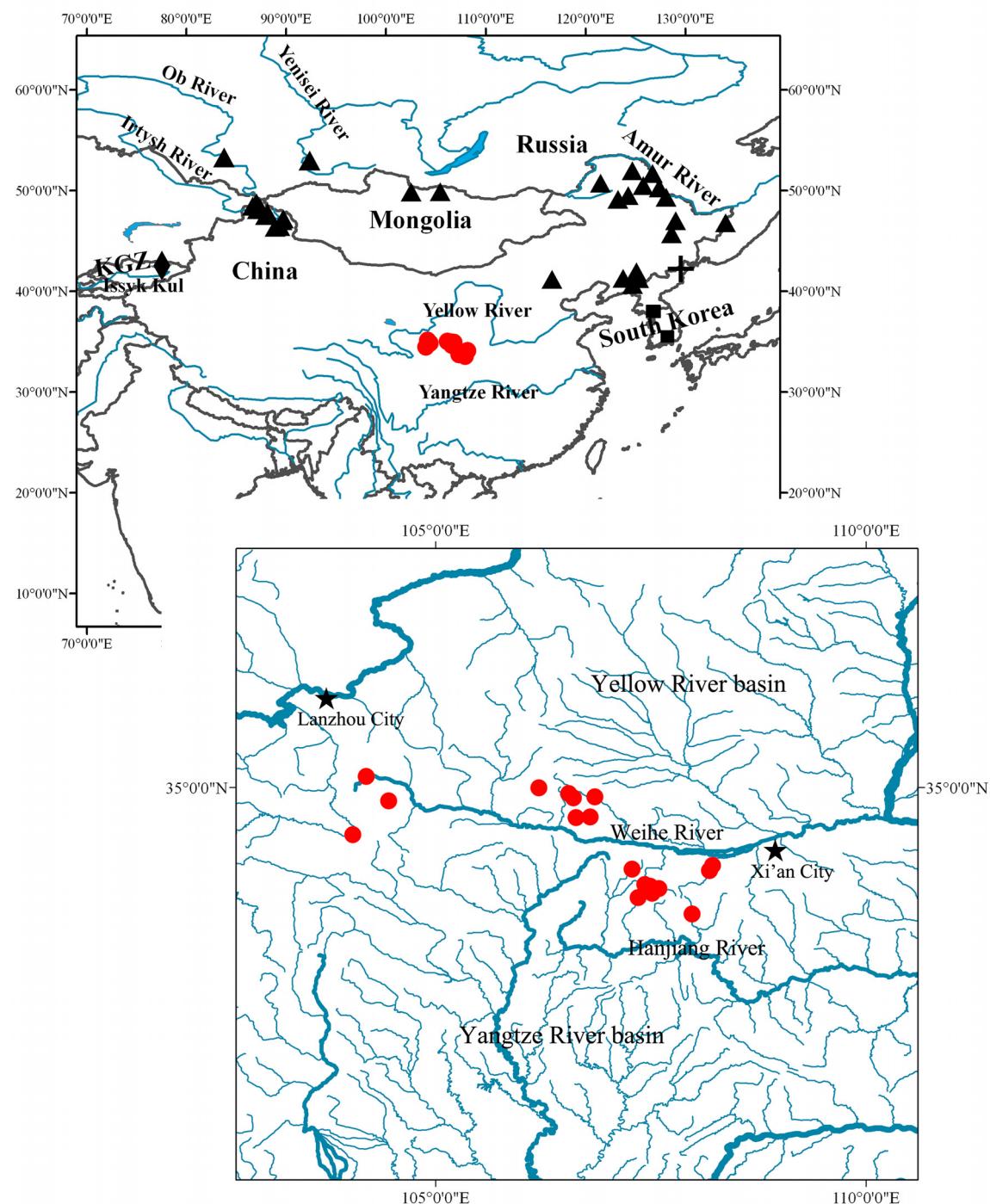


FIGURE 4. Distribution of *Brachymystax* in China and neighbor areas, above: Distribution; below: Collect locality of *B. tsinlingensis*. (Dot, *B. tsinlingensis*; triangle, *B. lenok*; diamond, *B. savinovi*; cross, *B. tumensis*; square, *Brachymystax* sp.). [HALF COLUMN]

Lateral line complete, almost straight. Body covered by small scales. Lateral-line scales 98-116, rows of scales above lateral line 20-28, below lateral line 20-28. Predorsal scales 47-70, regularly arranged. Circumpeduncular scales 34-40. Gill rakers 15-20. Pyloric caeca 60-71. Air bladder with one chamber, larger than eye diameter, with thin membrane and slim vessel connecting to intestine.

Coloration. Live individual (Fig. 3): body generally brownish, back dark brownish, light yellowish on sides and whitish on belly. A number of black oval spots with light pinkish edges scattered on back and sides except on operculum; 7-8 dark spots on the bases of dorsal fin and adipose fin. Around 10 light blackish bars on side of body. Orange on the edges of pectoral fin, pelvic fin, anal fin and caudal fin.

Preserved specimens (Fig. 2): lectotype specimen had been fixed and preserved in formalin since 1962, and then transferred to alcohol in 2005. Body dark brownish, back darker and belly lighter. Spots on back and sides; bars on sides. All fins light grayish.

Distribution. This species is only found in parts of the Taibai Mountains segment of the Qinling Mountains, including Heihe, Xushuihe and Qianhe, which are located in the Yangtze and Yellow River basins in China (Fig. 4). Although reported in South Korea, the identification of this population needs to be confirmed.

Habitat. *Brachymystax tsinlingensis* inhabits mountainous rivers of 900-2300 m above sea level and seldom is found in the rivers on the plain. Substrates are composed of gravel and larger stones. This species lives mainly in two environments, shallow areas of streams (0.3-0.5 m) and deep pools (>15 m) which alternate with one another in rivers. During the breeding season, *B. tsinlingensis* spawns at shallows. Stream water is typically clear, has high transparency and is slightly alkaline, with a velocity 0.2-0.5 m/s. Shore vegetation is prolific. This species is typically found syntopically with *Phoxinus lagowskii*, *P. oxycephalus*, *Paracobitis variegatus*, *Pseudorasbora parva*, *Triphlophysa sellaefer*, *T. shaanxiensis*, *T. robusta* and *T. dalaica*, based on our investigations from 2012-2013.

Etymology. The name of this species, *tsinlingensis*, is derived from the name of the area in the Qingling Mountains where the species was collected.



FIGURE 5. Comparison of spots on opercles of (a) *Brachymystax tsinlingensis*, (b) *B. tumensis*, and (c) *B. lenok*.

Discussion

Both *B. tsinlingensis* and *B. tumensis* lack spots on the operculum, distinguishing them from *B. savinovi* and *B. lenok* (Fig. 5). In addition, *B. tsinlingensis* differs from *B. lenok* by having a lower number of lateral-line scales (98-116 vs. 111-158; Fig. 6) and pyloric caeca (60-71 vs. 70-111; Table 2). Li (1966) reported that the major differences between *B. tsinlingensis* and *B. lenok* were that the former has fewer pyloric caeca (65-75 vs. 91-111) and lateral-line scales (115-127 vs. 132-175). Our results reflected similar divergences between the two species. *Brachymystax tsinlingensis* differs from *B. tumensis* by having fewer lateral-line scales (98-116 vs. 111-124; Fig. 6) and gill rakers (15-20 vs. 23-25; Fig. 7), and more pyloric caeca (60-71 vs. 55-65; Table 2).

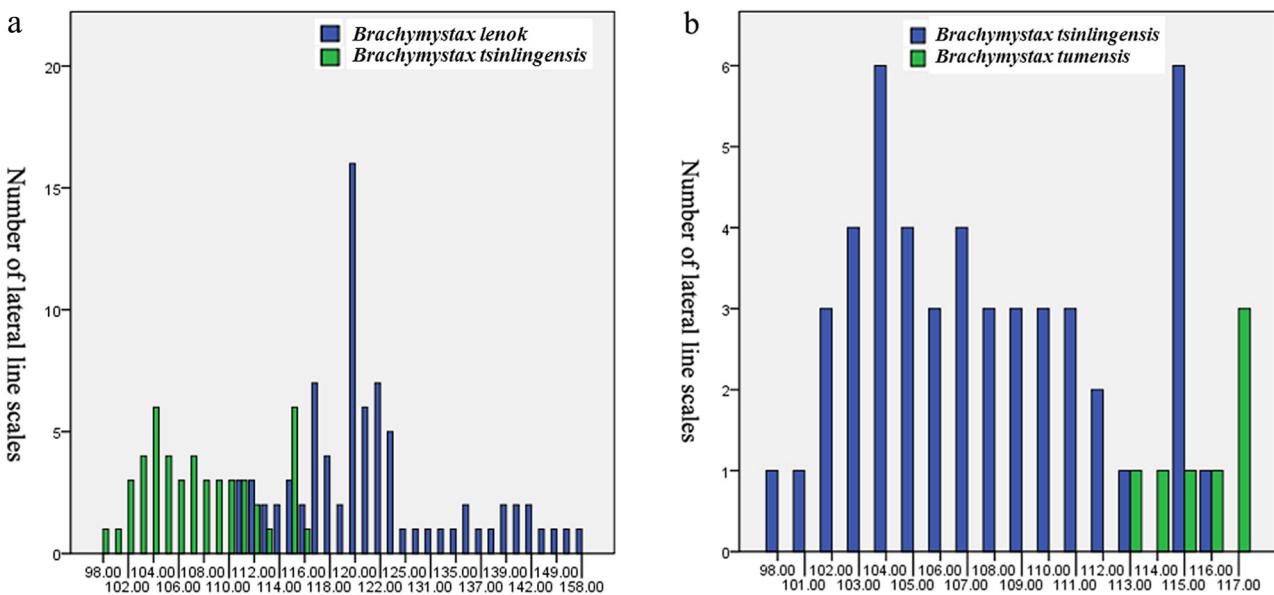


FIGURE 6. Number of lateral-line scales in (a) *Brachymystax tsinlingensis* and *B. lenok*, and in (b) *B. tsinlingensis* and *B. tumensis*.

Song & Fang (1984), Song (1987), Wang (1988) and Qin & Wang (1989) argued about the validity of *B. tsinlingensis* because they believed that characters such as lateral-line scales, gill rakers and pyloric caeca show continuous variation among populations. Although there was a narrow overlap, the data provided by these studies clearly show that differences in numbers of lateral-line scales, gill rakers and pyloric caeca separate *B. tsinlingensis* and *B. lenok* (Table 3). Also, these authors did not consider the differences in numbers of spots on the operculum.

Data from complete-mitochondrial DNA sequences and mitochondrial cytochrome *b* gene also supported the validity of *B. tsinlingensis* as an independent species. On the basis of a NJ tree (Fig. 8), *B. tsinlingensis* forms a monophyletic group, with an independent evolutionary history. *Brachymystax tsinlingensis* has a significant genetic divergence from *B. lenok* (0.020-0.022), *B. savinovi* (0.032) and *B. tumensis* (0.034), respectively. Pairwise genetic distance (Table 4) is larger than intraspecific genetic divergences of *B. tsinlingensis* (0.000), *B. lenok* (0.001-0.002) and *B. savinovi* (0.003).

TABLE 3. Meristic characters of *Brachymystax tsinlingensis* and main geographical populations of *B. lenok*.

| <i>Brachymystax tsinlingensis</i> | | <i>Brachymystax lenok</i> | | | |
|-----------------------------------|--------|-----------------------------|--------------|---------------|------------|
| | | Haihe River | IrtySh River | Selenga River | Amur River |
| Lateral-line scales | 98-116 | 120-131 (Wang, 1988 & 1993) | 136-142 | 122-158 | 111-149 |
| Gill rakers | 15-20 | 19-20 (Wang, 1988 & 1993) | 19-24 | 23-26 | 17-29 |
| Pyloric caeca | 60-71 | 63-68 (Wang, 1988) | 70-74 | 74-87 | 70-111 |

A previous study based on the isozymes LDH, MDH, ADH, GDH and EST (Qin & Wang, 1989), showed overlap between *B. tsinlingensis* and *B. lenok*. However, mtDNA data are considered more efficient in delineating fish species (Meyer, 1993; Hillis *et al.* 1996) and has been broadly used in taxonomic studies of Salmonidae (Shedko *et al.* 1996; Hansen *et al.* 2003; Xia *et al.* 2006; Froufe *et al.* 2008; Crete-Lafreniere *et al.* 2012; Si *et al.* 2012; Shedko *et al.* 2013; Liu *et al.* 2014).

The distribution of *B. tsinlingensis* in China is limited (Fig. 4) to the southernmost boundary of all species of the genus (Wang, 1990). *Brachymystax savinovi* has been recorded in rivers from Kazakhstan to the Amur basin (Mitrofanov, 1959; Alekseyev *et al.* 2003). *Brachymystax tumensis* is restricted to the Tumenjiang River, which is located in the area bordering China, North Korea and Russia. *Brachymystax lenok* has a broad distribution in China, but basically is found north of the 40-degree latitude.

Table 4. Pairwise genetic distances of *Brachymystax tsinlingensis* compared to all congeners and outgroups.

| Species | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 | 14 | 15 |
|-----------------------------------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|----|
| 1. <i>Brachymystax savinovi</i> 1 | | | | | | | | | | | | | | | |
| 2. <i>B. savinovi</i> 2 | 0.003 | | | | | | | | | | | | | | |
| 3. <i>B. tumensis</i> | 0.003 | 0.002 | | | | | | | | | | | | | |
| 4. <i>B. tsinlingensis</i> 1 | | 0.032 | 0.032 | 0.034 | | | | | | | | | | | |
| 5. <i>B. tsinlingensis</i> 2 | | 0.032 | 0.032 | 0.034 | 0.000 | | | | | | | | | | |
| 6. <i>Brachymystax</i> sp. Korea | 0.031 | 0.031 | 0.033 | 0.003 | 0.003 | | | | | | | | | | |
| 7. <i>B. lenok</i> 4 | 0.032 | 0.031 | 0.031 | 0.020 | 0.020 | 0.019 | | | | | | | | | |
| 8. <i>B. lenok</i> 5 | 0.032 | 0.031 | 0.031 | 0.020 | 0.020 | 0.019 | 0.000 | | | | | | | | |
| 9. <i>B. lenok</i> 3 | 0.031 | 0.031 | 0.031 | 0.020 | 0.020 | 0.019 | 0.001 | 0.001 | | | | | | | |
| 10. <i>B. lenok</i> 1 | 0.033 | 0.032 | 0.032 | 0.022 | 0.022 | 0.020 | 0.002 | 0.002 | 0.001 | | | | | | |
| 11. <i>B. lenok</i> 2 | 0.033 | 0.032 | 0.032 | 0.022 | 0.022 | 0.020 | 0.002 | 0.002 | 0.001 | 0.000 | | | | | |
| 12. <i>Hucho taimen</i> 1 | 0.060 | 0.060 | 0.059 | 0.060 | 0.060 | 0.059 | 0.059 | 0.058 | 0.058 | 0.060 | 0.060 | | | | |
| 13. <i>H. taimen</i> 2 | 0.060 | 0.060 | 0.059 | 0.060 | 0.060 | 0.059 | 0.059 | 0.058 | 0.058 | 0.060 | 0.060 | 0.060 | | | |
| 14. <i>H. hucho</i> | 0.065 | 0.065 | 0.065 | 0.063 | 0.063 | 0.062 | 0.064 | 0.064 | 0.065 | 0.066 | 0.066 | 0.068 | 0.008 | | |
| 15. <i>H. bleekeri</i> | 0.061 | 0.061 | 0.059 | 0.060 | 0.060 | 0.059 | 0.060 | 0.060 | 0.060 | 0.061 | 0.061 | 0.011 | 0.009 | 0.016 | |

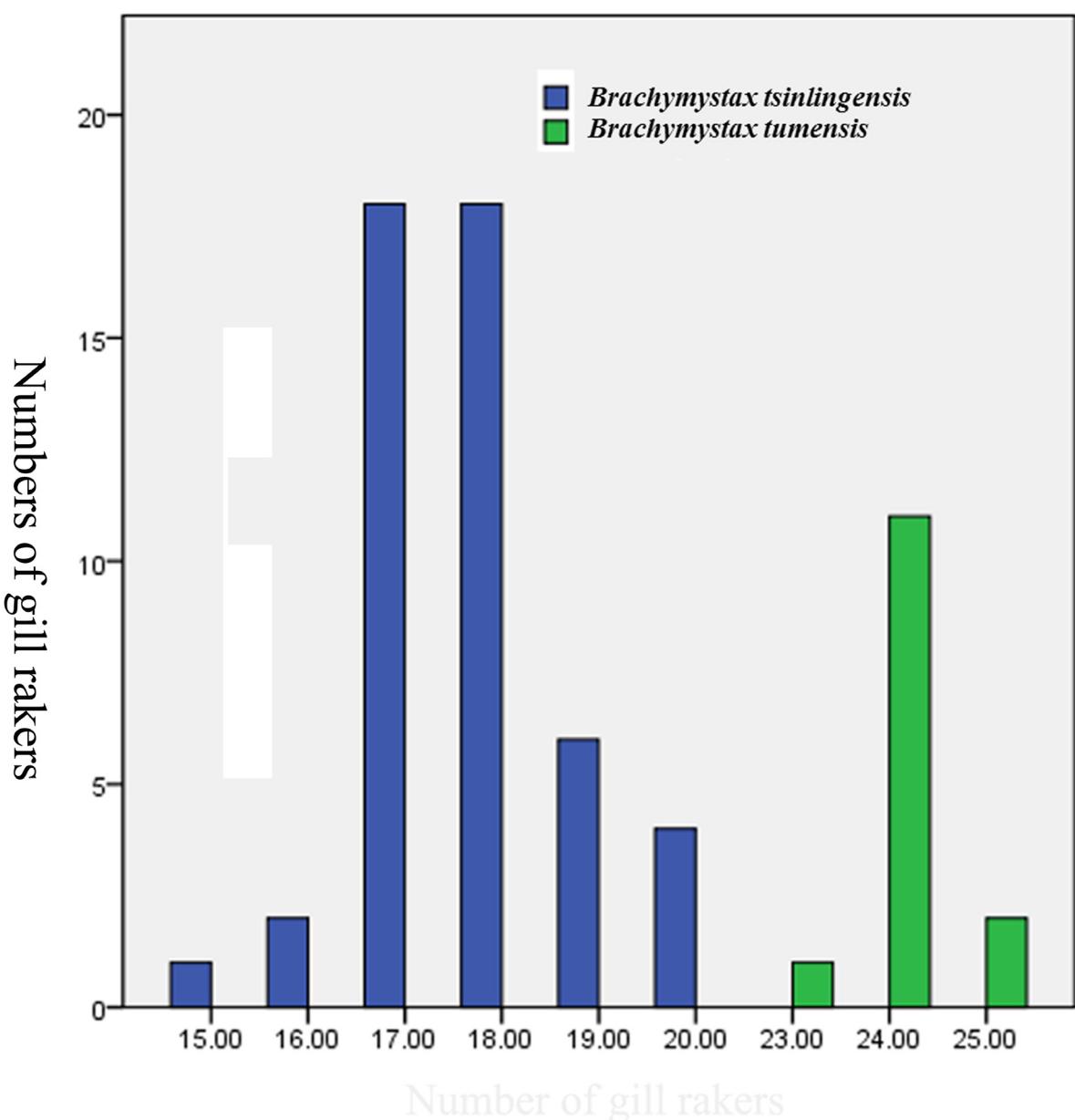


FIGURE 7. Numbers of gill rakers in *Brachymystax tsinlingensis* and *B. tumensis*. [HALF COLUMN]

“*Brachymystax tsinlingensis*” has also been recorded in South Korea (Choi *et al.* 2003; Jang *et al.* 2003; Yoon *et al.* 2014), and the genetic divergence between *B. tsinlingensis* and *Brachymystax* sp. Korea also showed a close relationship between these populations (Fig. 8). However, based on photographs of live individuals (Kim & Park, 2002), Korean specimens have spots on the operculum similar to those of *B. lenok*. Since we were not able to check the specimens applied in those works, the taxonomic position of the Korean population needs further investigation. The habitat of *B. tsinlingensis* is also significantly different from that of other congeners (Fig. 9). *Brachymystax lenok*, *B. tumensis* and *B. savinovi* inhabit river plains, large rivers and their tributaries such as Amur River, Tumenjiang River and Irtysh River (Mitrofanov, 1959; Güther, 1866; Mori, 1930). However, *B. tsinlingensis* inhabits rivers at higher altitude in the Qinling Mountains.

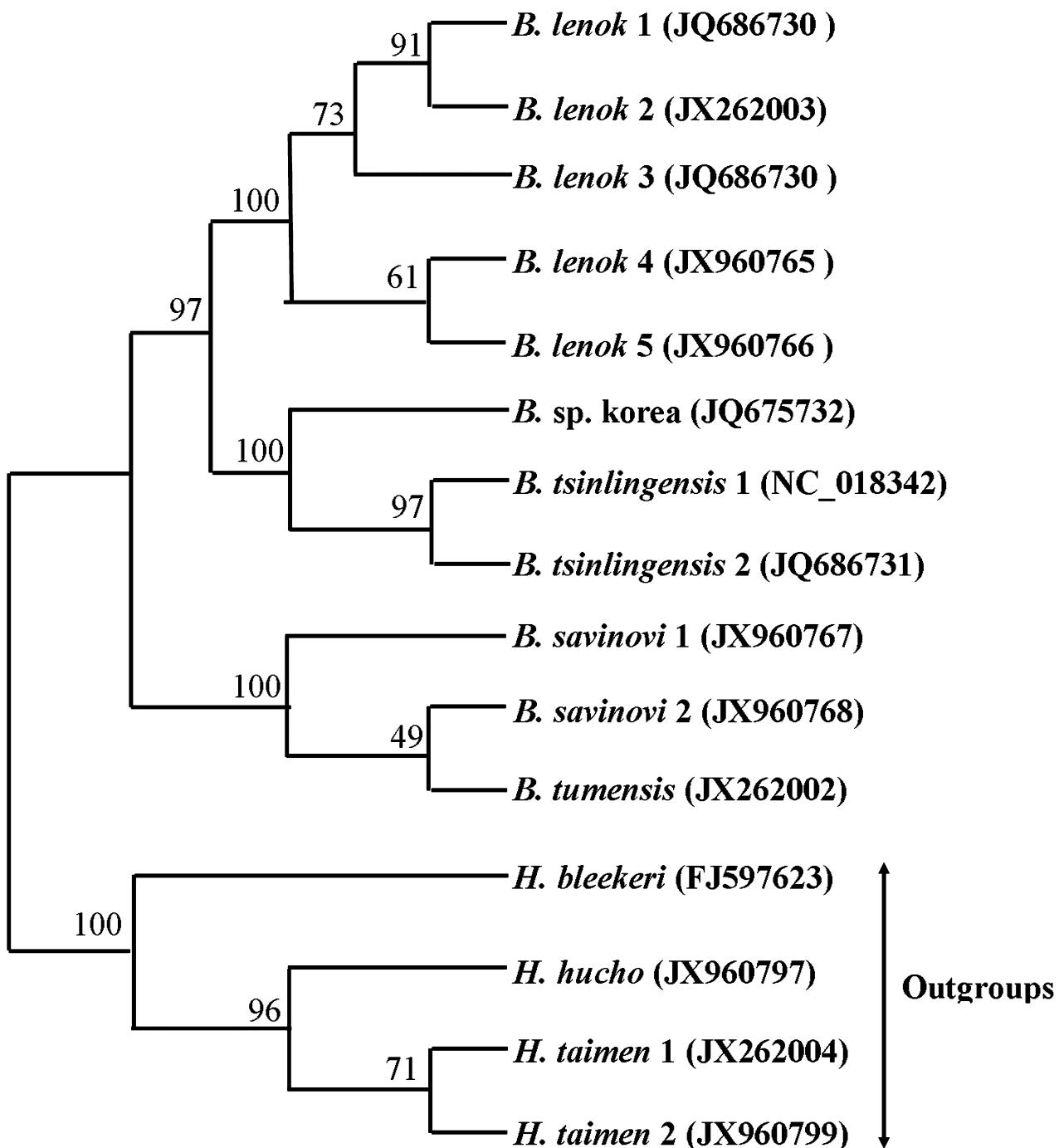


FIGURE 8. Neighbor-Joining tree obtained as a result of analysis of mtDNA data on species of *Brachymystax* and *Hucho*. Figures above branches are bootstrap confidence values (% of 1000 bootstrap replicates). Numbers in brackets refer to GeneBank Accession numbers. [HALF COLUMN]

In the past 30 years, populations of *B. tsinlingensis* have declined mainly due to environmental changes, human activities and overfishing (Ren and Liang, 2004; Hou, 2009; Zhao and Zhang, 2009). This species has been listed in the Redbook (Wang & Xie, 2004) and is considered to be an endangered species (Yue & Chen, 1998; Zhao and Zhang, 2009). It is protected in aquatic wildlife natural reserves and through enhancements of resources. There are four National Aquatic Wildlife Natural Reserves at Shannxi and Gansu provinces specially to protect this species. However, there are plans by the local government to introduce *B. lenok* from the Amur River basin to recover *B. tsinlingensis* populations and increase resources. On the basis of our study, *B. tsinlingensis* is a valid and

independent species, and introductions of a separate closely related species is not an advisable measure for conservation.



FIGURE 9. Comparison of habitats of (a) *Brachymystax tsinlingensis* (a) and (b) congeners.

Comparative materials

***Brachymystax lenok*:** NZMC76240-41 (2), 76.21-80.40 mm SL, Amur River basin: Raohe River, Heilongjiang Province, China P.R., Jul., 1961; NZMC165294-97 (4), 160.89-229.98 mm SL, Amur River basin: Yichun City, Heilongjiang Province, China P.R., May, 2002; NZMC16594, 228.7 mm SL, Amur River basin: Heihe River, Heilongjiang Province, China P.R., May, 2002; NZMC198987-91 (5), 292.87-340.36 mm SL, Amur River basin: Wusulijiang River, Haiqing Villages, Zhuaji Town, Fuyuan County, Jiamusi City, Heilongjiang Province, China P.R., Oct. 20, 2013; NZMC5113, 43232-35 (4), 126.36-212.36 mm SL, Amur River basin: Songhua River, Heilongjiang Province, China P.R.; NZMC28807-08 (2), 157.84-180.30 mm SL, Amur River Basin: Dailing Town, Tangyuan County, Heilongjiang Province, China P.R.; NZMC50172, 50174, 50176-77 (2), NK641035, 122.58-314.38 mm SL, Koktokay, Fuyun County, Xinjiang Province, China P.R.; NZMC188998-99 (2), 188961-63 (3), 187.52-466.45 mm SL, Zelter River, Tusheng County, Selenge Province, Mongolia, Jul. 15-16, 2010; NZMC188792-95 (4), 80.37-119.73 mm SL, Harry Wagner County, Durkheim province, Mongolia, Jul. 12, 2010; NZMC188822-23 (2), 122.32-168.47 mm SL, Eoroo River, Selenge Province, Mongolia, Jul. 19, 2010; NZMC188942-46 (5), Egyn River, Durkheim province, Mongolia, Jul. 9-11, 2010; HFRI 81340-42 (3), 234-261.25 mm SL, Nenjiang River basin: Woduhe River, Heilongjiang Province, China P.R., Jan., 1981; HFRI 821705-09 (5), HFRI 820124, 135.51-238.03 mm SL, Amur River basin: Huma County, Heilongjiang Province, China P.R., May, 1982; HFRI 198207, 209.64 mm SL, Amur River basin: Huma County, Heilongjiang Province, China P.R., May, 1982; HFRI 20130701-10 (10), 195.96-228.04 mm SL, Amur River basin: Genhe River, Inner Mongolia Province, China P.R., Jul., 2013; HFRI 8900-8901 (2), 193.67-224.80 mm SL, Nenjiang River Basin: Woduhe River, Heilongjiang Province, China P.R., Jan., 1981; HFRI 201101-06 (6), 204.71-265.73 mm SL, Nenjiang River basin: Nuominhe River, Heilongjiang Province, China P.R., Apr. 21, 2011; HFRI 20070901-03 (3), 277.77-293.64 mm SL, Koktokay, Fuyun County, Xinjiang Province, China P.R., Sept. 12, 2007; HFRI 20070501-10 (10), 208.28-295.51 mm SL, Irtysh River basin: Burqin River, Burqin County, Xinjiang Province, China P.R., May, 2007; NHBH 01-03 (3), 122-228 mm SL, Haihe River basin: Honhtang Town, Fengning County, Beijing, China P.R..

Brachymystax tumensis NZMC 64781-86 (6), NZMC 71294-301 (8), 112.86-133.45 mm SL, Tumenjiang River, Heilongjiang Province, China P.R., 1977.

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References

- Alekseev, S.S., Kirillov, A.F. & Samusenok, V.P. (2003) Distribution and morphology of the sharp-snouted and the blunt-snouted lenoks of the genus *Brachymystax* (Salmonidae) of east Siberia. *Voprosy Ikhtiologii*, 43, 311–333. [In Russian]
- Choi, E.J., Jin, D.H. & Sohn, Y.C. (2003) Gonadotropins in the Manchurian trout, *Brachymystax lenok tsinlingensis*. *Fish Physiology and Biochemistry*, 28, 89–90.
<http://dx.doi.org/10.1023/B:FISH.0000030483.11026.e5>
- Crete-Lafreniere, A., Weir, L.K. & Bernatchez, L. (2012) Framing the salmonidae family phylogenetic portrait: a more complete picture from increased taxon sampling. *PLoS ONE*, 7 (10), E46662.
<http://dx.doi.org/10.1371/journal.pone.0046662>
- Froese, R. & Pauly, D. (2014) FishBase. World Wide Web electronic publication. Available from www.fishbase.org (accessed February 2014)
- Froufe, E., Alekseyev, S., Alexandrino, P. & Weiss, S. (2008) The evolutionary history of sharp- and blunt-snouted lenok (*Brachymystax lenok* (Pallas, 1773) and its implications for the paleo-hydrological history of Siberia. *BMC Evolutionary Biology*, 8, 1–40.
<http://dx.doi.org/10.1186/1471-2148-8-40>
- Gao, X.Z. (1980) Study on distribution and its characteristics of *Hucho* and *Brachymystax* in China. *Shannxi Fisheries*, 1, 23–30. [In Chinese]
- Günther, A. (1866) *Catalogue of fishes in the British Museum. Vol. 6. Catalogue of the Physostomi, containing the families Salmonidae, Percopsidae, Galaxidae, Mormyridae, Gymnarchidae, Esocidae, Umbridae, Scombrésocidae, Cyprinodontidae, in the collection of the British Museum*. Trustees of the London Museum, London, pp. 162–164.
- Hansen, H., Bachmann, L. & Bakke, T.A. (2003) Mitochondrial DNA variation of *Gyrodactylus* spp. (Monogenea, Gyrodactylidae) populations infecting Atlantic salmon, grayling, and rainbow trout in Norway and Sweden. *International Journal for Parasitology*, 33, 1471–1478.
[http://dx.doi.org/10.1016/S0020-7519\(03\)00200-5](http://dx.doi.org/10.1016/S0020-7519(03)00200-5)
- Hillis, D.M., Moritz, C. & Mable, B.K. (1996) *Molecular Systematics (second edition)*. Sinauer Associates, Inc. Publishers Sunderland, Massachusetts.
- Hou, F. (2009) *Study on the conservation biology of Brachymystax lenok tsinlingensis in Gansu Province*. Master's Dissertation of Shaanxi Normal University. [In Chinese].
- Jang, M.H., Lucas, M.C. & Joo, G.J. (2003) The fish fauna of mountain streams in South Korean national parks and its significance to conservation of regional freshwater fish biodiversity. *Biological Conservation*, 114, 115–126.
[http://dx.doi.org/10.1016/S0006-3207\(03\)00016-8](http://dx.doi.org/10.1016/S0006-3207(03)00016-8)
- Kim, I.S. & Park, J.Y. (2002) *Freshwater Fishes of Korea*. Kyo-Hak Publishing Co., Seoul.
- Li, S.C. (1966) On a new subspecies of fresh-water trout, *Brachymystax lenok tsinlingensis*, from Taipaishan, Shenxi, China. *Acta Zootaxonomica Sinica*, 3, 92–94. [In Chinese]
- Liu, H.X., Li, Y., Liu, X.L., Xiong, D.M., Wang, L.X., Zou, G.W. & Wei, Q.W. (2014) Phylogeographic structure of *Brachymystax lenok tsinlingensis* (Salmonidae) populations in the Qinling Mountains, Shaanxi, based on mtDNA control region. *Mitochondrial DNA*.
<http://dx.doi.org/10.3109/19401736.2013.865168>
- Ma, B., Jiang, Z.F. & Huo, T.B. (2009) Study on the taxonomic status of species of *Brachymystax* in Heilongjiang River and Tumen River systems based on mitochondrial control region sequence. *Acta Zootaxonomica Sinica*, 34, 500–506. [In Chinese]
- Meyer, A. (1993) Evolution of mitochondrial DNA in fishes. In: Hochachka, P. & Mommsen, T.P. (Eds.), *Biochemistry and Molecular Biology of Fishes*. Vol. 2. Elsevier Science Publishers, Amsterdam.
- Mitrofanov, V.P. (1959) On the taxonomy of *Brachymystax lenok* of the Lake Marka-Kul. *Sbornik Rabot po Ikhtiologii Gidrobiologii*, 2, 267–275. [In Russian]

- Mori, T. (1930) On the fresh water fishes from the Tumen River, Korea, with description of new species. *Journal of the Chosen Natural History Society*, 11, 39–49.
- Pallas, P.S. (1773) *Reise durch verschiedene Provinzen des russischen Reichs*. Zweyter Theil. Kayserlichen Academie der Wissenschaften, St. Petersburg, 760 pp.
- Qi, D.L., Zhao, Y., Yang, C., Shen, Z.X. & Tang, W.J. (2009) Cloning of mitochondrial cytochrome *b* gene of *Hucho bleekeri* and its phylogenetic relationship in subfamily Salmoniae. *Sichuan Journal of Zoology*, 28, 805–809. [In Chinese]
- Qin, S.Z. & Wang, S.A. (1989) Studies on the subspecies of *Brachymystax lenok* (Pallas), China. *Salmon Fishery*, 2, 52–61. [In Chinese]
- Ren, J. & Liang, G. (2004) Resource survey report of *Brachymystax lenok tsinlingensis* in Qianhe River Valleys of Qinling Mountains. *Journal of Shaanxi Normal University*, 32, 165–168. [In Chinese]
- Shedko, S.V. (2001) List of cyclostomes and fishes from freshwaters of Primorye. V.Y. *Levanidov's Biennial Memorial Meetings*, 1, 229–249. [in Russian]
- Shedko, S.V., Ginatulina, L.K., Parpura, I.Z. & Ermolenko, A.V. (1996) Evolutionary and taxonomic relationships among far-eastern salmonid fishes inferred from mitochondrial DNA divergence. *Journal of Fish Biology*, 49, 815–829.
<http://dx.doi.org/10.1111/j.1095-8649.1996.tb00081.x>
- Shedko, S.V., Miroshnichenko, I.L. & Nemkova, G.A. (2013) Phylogeny of salmonids (Salmoniformes: Salmonidae) and its molecular dating: analysis of mtDNA data. *Russian Journal of Genetics*, 49, 623–637.
<http://dx.doi.org/10.1134/S1022795413060112>
- Si, S., Wang, Y., Xu, G., Yang, S., Mou, Z. & Song, Z. (2012) Complete mitochondrial genomes of two lenoks, *Brachymystax lenok* and *Brachymystax lenok tsinlingensis*. *Mitochondrial DNA*, 23, 338–340.
<http://dx.doi.org/10.3109/19401736.2012.690749>
- Song, S.L. (1987) Salmonidae. In: Institute of Zoology, Shaanxi Province, Institute of Hydrobiology, Chinese Academy of Sciences & Lanzhou University (Eds.), *Fishes in Qinling Mountain Area*. Science Press, Beijing, pp. 14–15. [In Chinese]
- Song, S.L. & Fang, S.M. (1984) Discussion of the subspecies of Salmonidae fishes, *Brachymystax lenok tsinlingensis* Li, from Shannxi, China. *Journal of Lanzhou University (Natural Sciences Edition)*, 20, 92–95. [In Chinese]
- Wang, H.Y. (1988) Research of *Brachymystax* and *B. lenok* (Pallas) from Northern area of Hebei. *Salmon Fishery*, 1, 16–25. [In Chinese]
- Wang, H.Y. (1993) *Fishes and amphibians of Beijing*. Beijing Press, Beijing, pp. 22–26. [In Chinese]
- Wang, S. & Xie, Y. (2004) *China Species Red List. Vol. 1. Red list*. Higher Education Press, Beijing, 160 pp. [In Chinese]
- Wang, S.A. (1990) Biological feature and distributive change of fine scale fish in north China. *Salmon Fishery*, 3, 39–45. [In Chinese]
- Xia, Y.Z., Sheng, Y. & Chen, Y.Y. (2006) DNA sequence variation in the mitochondrial control region of lenok (*Brachymystax lenok*) populations in China. *Chinese Biodiversity*, 14, 48–54. [In Chinese]
<http://dx.doi.org/10.1360/biodiv.050189>
- Yoon, J.D., Kim, J.H., Jo, H.B., Yeom, M.A., Heo, W.M., Joo, G.J. & Jang, M.H. (2014) Seasonal habitat utilization and movement patterns of the threatened *Brachymystax lenok tsinlingensis* in a Korean river. *Environmental Biology of Fishes*.
<http://dx.doi.org/10.1007/s10641-014-0254-7>
- Yue, P.Q. & Chen, Y.Y. (1998) *China Red Data Book of Endangered Animals, Pisces*. Science Press, Beijing, pp. 107–109. [in Chinese]
- Zhao, Y.H. & Zhang, C.G. (2009) Threatened fishes of the world: *Brachymystax lenok tsinlingensis* Li, 1966 (Salmonidae). *Environmental Biology of Fishes*, 86, 1–112.
<http://dx.doi.org/10.1007/s10641-008-9337-7>