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Unexpected finding of the invasive Baikalian amphipod *Gmelinoides fasciatus* in a cold spring of the southern Pamir Mountains

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ABSTRACT

Key-words: *alien species, inland waters, Pamir Mountains, Gmelinoides, first record*

A specimen of the Baikalian micruropodid amphipod *Gmelinoides fasciatus* was recently found in a spring brook in the Shugnansky District, Gorno-Badakhshan Autonomous Province of Tajikistan. This occurrence represents the first record confirmed with mt-*cox1* sequence data for this species from the Pamir Mountains. Reliable information on the introduction of *Gm. fasciatus* and the pattern of its distribution in the region is not yet available, but we hypothesise that the species was introduced intentionally to increase the fish productivity of Yashilkul Lake in 1973–1974. This finding of probably an isolated population shifts the southernmost limit of the species range approximately 1400 km from the Bukhtarminskoye reservoir, Eastern Kazakhstan. A general overview of the morphology, ecology, distribution and invasive history of the species is provided.

RÉSUMÉ

Découverte inattendue de l'amphipode invasif baïkalien *Gmelinoides fasciatus* dans une source froide des montagnes du sud du Pamir

Mots-clés : *espèces exotiques, les eaux intérieures, montagnes du Pamir, Gmelinoides, premier enregistrement*

Un spécimen de l'amphipode baïkalien micruropodide *Gmelinoides fasciatus* a récemment été trouvé dans un ruisseau dans le district Shugnansky, de la province autonome de Gorno-Badakhshan au Tadjikistan. Cet occurrence représente le premier enregistrement confirmé par les données de séquence mt-*cox1* pour cette espèce dans les montagnes du Pamir. Des informations fiables sur l'introduction de *Gm. fasciatus* et le schéma de sa distribution dans la région n'est pas encore disponible, mais nous supposons que l'espèce a été introduite intentionnellement pour augmenter la productivité des poissons du lac Yashilkul en 1973–1974. Cette découverte d'une population probablement isolée décale la limite méridionale de l'espèce d'environ 1400 km depuis le réservoir Bukhtarminskoye, Kazakhstan oriental. Un aperçu général de la morphologie, l'écologie, la distribution et l'histoire de la propagation de l'espèce est fourni.

INTRODUCTION

The Baikalian amphipod *Gmelinoides fasciatus* (Stebbing 1899), which derives its name from its striped colour pattern, is a small omnivorous gammaroid crustacean (Takhteev, 2000).

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Its natural range is restricted primarily to Lake Baikal and its basin (including tributaries of the lake, *e.g.*, the Selenga and Barguzin), the Angara and the Yenisei rivers (Bazikalova, 1945; Matafonov *et al*., 2005; Kamaltynov, 2009). *Gmelinoides fasciatus* shows extremely broad "ecological plasticity" and tolerance to environmental factors. The species has substantially expanded its geographical range and has become one of the most widely distributed species of continental gammarids, second in this respect to the other representative of the suborder – *Gammarus lacustris* G.O. Sars 1863. The current distribution of *Gm. fasciatus*[1] ranges from Eastern Siberia and the Amur River basin in the east to the European part of Russia, where the species has not only penetrated to the Central Moscow region (Chertoprud, 2006) but also settled in the freshwater-influenced areas of the Gulf of Finland (Berezina and Panov, 2004).

Gmelinoides fasciatus has generally been considered a desirable species for use in introductions to increase the fish productivity of lakes and reservoirs. In the late 1960's – early 1990's, at the peak period of acclimatisation activities in the Soviet Union, the species was artificially introduced into a number of lakes and reservoirs in the Asian and European parts of the U.S.S.R. According to the literature (Ioffe, 1968; Berezina, 2007) *Gm. fasciatus* was purposely introduced into the Gorkovskoe, Ozerninskoe, Novosibirskoe, Krasnoyarskoe, Iriklinskoye, Bukhtarminskoye, and Ust'-Kamenogorskoe reservoirs and into natural lakes (*e.g.*, Peipsi, Goosinoye, Bol. Eravnoe and Il'men) and into a number of medium-sized lakes in the Leningrad region. The most complete summary of this history, with a synopsis of papers published between 1960 and 2002 on the acclimatisation of *Gm. fasciatus* and its subsequent detection in both artificial and natural waters, has been presented by the following authors: Panov and Berezina (2002) and Matafonov *et al*. (2005). However, we have not found any conclusive information on the introduction of *Gm. fasciatus* into water bodies in Tajikistan other than a statement in F. Ahrorov's Ph.D. thesis referring to the implementation act: "cit. by: Ahrorov (2001). The biological basis for acclimatisation in Yashilkul Lake of the deep sor Baikalian gammarids for enrichment of the forage deep part of the lake (implementation act 24 May 1973 and 28 May 1974)." Whether these acclimatisation plans were actually implemented remains unknown, and the possible results of any resulting acclimatisation are likewise unknown.

During our study, *Gm. fasciatus* was found in a cold spring in the Southern Pamir. This finding significantly extends the southern boundary of the range of habitats to which the species has artificially been introduced.

MATERIALS AND METHODS

The specimens are included in the amphipod collection made by one of the authors (DMP) in the Gorno-Badakhshan Autonomous Oblast (Figure 1) from the riparian zone of a spring brook in the Gunt River catchment area, the source of which is Yashilkul Lake. The sample was preserved in 96% ethanol. The morphological identification of the specimens was based on A.J. Bazikalova's description of *Gm. fasciatus* (see Bazikalova, 1945, p. 66, pl. 9(4), pl. 10(1)). Because a number of morphological features (Figure 2) needed clarification (*e.g.*, the armament of ghathopods 1 and 2), we further studied the specimens to obtain a detailed investigation and description.

To confirm the affinity of our specimen, a partial sequence (565nt long) of the mitochondrial cytochrome oxidase subunit 1 (*cox1*) gene was obtained with HCO2198 and LCO1490 (Folmer *et al*., 1994) primers. The annealing temperature was set to 40 °C for 20 s. To analyse the affinity of our specimen in more detail, we aligned the new *Gm. fasciatus* sequence (Genbank acc. no. HF937342) with those obtained by Gomanenko *et al*. (2005) from different Baikalian populations and conducted maximum-likelihood analyses in PAUP 4.0b10 (Swofford, 2002) using the best-fit GTR+I+Γ model (Modeltest; Posada and Crandall, 1998). Prior to the analyses, redundant sequences were removed from the alignment.

[1] The generic name (*Gmelinoides*) is abbreviated through the text for two-letter manner to convenience easy it tells from *Gammarus*.

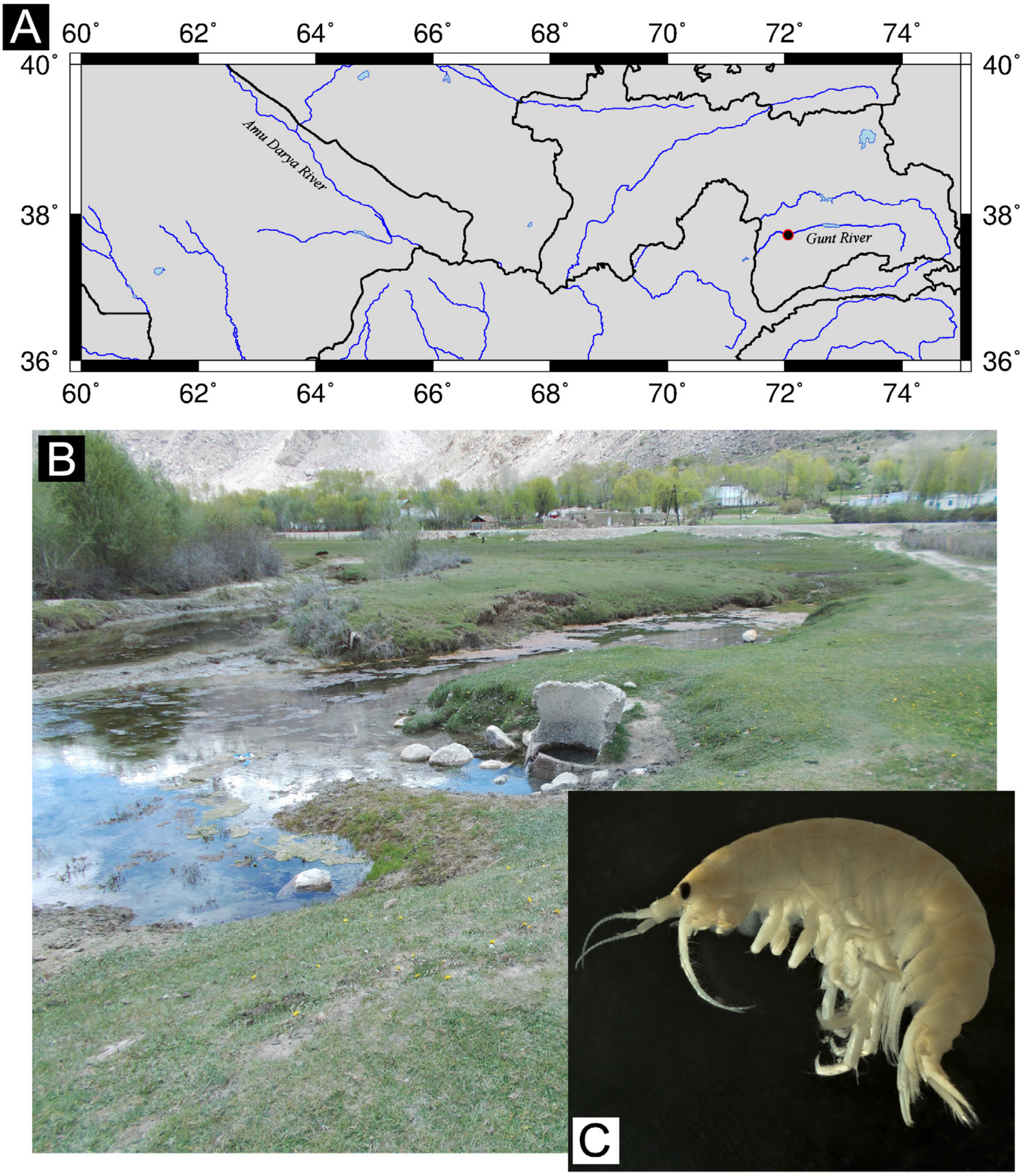

Figure 1
A – map showing location of the sampling site; B – sampling site in Charthem kishlak, GBAO, Tadjikistan; C – habitus of Gm. fasciatus (Stebbing 1899) (coloring after fixation in alcohol). (Photographs by D. Palatov and A. Palatov).

A geographical map, with the location of the sampling site, was constructed with free GMT 4.5.6.-1. GIS software.

The materials examined in this study were deposited in the Zoological Museum of the Far Eastern Federal University (FEFU), Vladivostok.

TAXONOMY

Order Amphipoda Latreille 1818
Family Micruropodidae Kamaltynov 1999[2]

[2] Takhteev (2000) described the family Carinogammaridae in which includes the genus *Gmelinoides*, whereas the smooth “micruropodids” without cuticular body formations (as, *Baicalogammarus*, Stebb.,

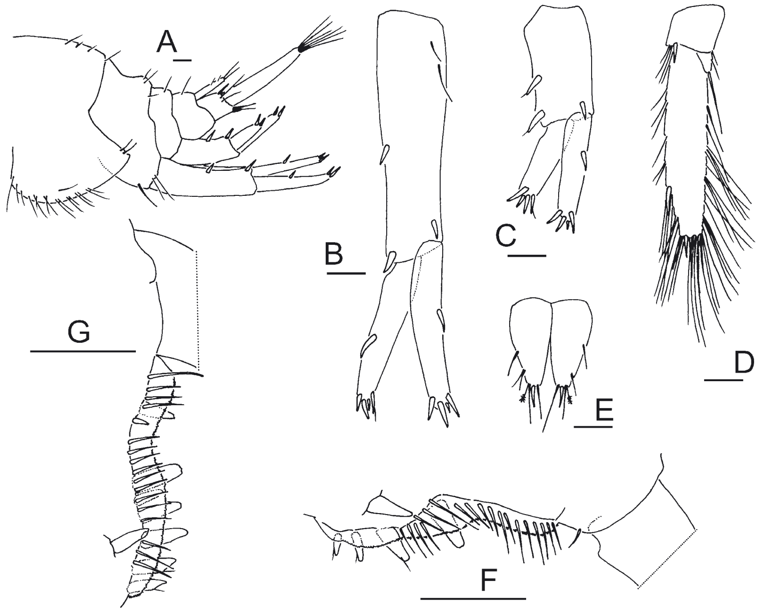

Figure 2
Gm. fasciatus (Stebbing 1899), male 9.0 ᴍᴍ*, № X38470/Cr-1467-FEFU: A – urosome, B – uropod 1, C – uropod 2, D – uropod 3, E – telson, F – gnathopod 1, palmar margin (inside view), G – gnathopod 2, palmar margin (inside view). Scale bars 0.1 mm.*

> *GMELINOIDES FASCIATUS (STEBBING 1899)*

Gammarus zebra Dybowsky 1874 non *G. zebra* Rathke 1843
Brandtia fasciata Stebbing 1899, 1906; Sowinsky 1915
Micruropus sublittoralis Sowinsky 1915
Gmelinoides fasciatus: Bazikalova 1945; Gurjanova 1951; Kamaltynov 2002, 2009
Gmelinoides fasciata: Barnard and Barnard 1983

Material. ♂ 9.0 mm, X38470/Cr-1467-FEFU, ♀ 7.0 mm, 38471/Cr-1468-FEFU; Tajikistan, Gorno-Badakhshan Autonomous Oblast (GBAO), Shugnansky District, near Charthem kishlak (37.724722 N, 72.163528 E), spring brook in the Gunt River basin, 14 VI 2012, coll. D. Palatov.

Diagnosis (refined, cf. Bazikalova, 1945, p. 66; Barnard and Barnard, 1983, p. 486). Small-sized species without marked secondary sexual dimorphism. Head smooth with shallow inferior sinus, interantennal lobe short, cone-shaped. Eyes medium-sized, reniform. Body stout, robust (Figure 1C), dorsal surface of body segments smooth, bearing weak cuticular projections on last pereonite and pleonites 1–3. Antenna 1 as long as antenna 2 and comprises approximately 25% length of body. Coxal plates 1–4 deep. Gnathopods *Gammarus*-form, subchelate; gnathopod 2 slightly larger than gnathopod 1 in male. Pereopod 7 as long as pereopod 6, basipodite well developed with wing-shaped margin, margin setose with short soft setae. Coxal gills on gnathopod 2 and pereopods 3–7. Female marsupial plates 2–5 very large with long soft setae, last plate the smallest. Uropod 3 parviramous; outer ramus biarticulated, more than 3 times longer than inner, with a sets of simple setae along inner margin. Telson *Gammarus*-like, entirely cleft. Body length 7.0–15.0 mm.

Description. Male. *Gnathopod 1* and *2* (Figures 2F, 2G): palm with central concavity, cutting margin developed, bearing 4–6 strong spines on outside and 1 strong shutoff spine on inside. *Uropod 1* (Figure 2B): peduncle without basofacial spine, bearing 1 and 2 spines on inner and outer margins, respectively; inner ramus approximately 2 times shorter than peduncle with

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Crypturopus Sow., *Homocerisca* Baz., *Micruropus* Stebb.) were left in Gammaridae. Subsequent compound phylogenetic analysis by Macdonald *et al*. (2005) has not confirmed of the validity of Carinogammaridae. Kamaltynov (2009) during an extensive review resurrected the family Carinogammaridae as a monogeneric family.

1 spine on outer margin and 4 distal spines; outer ramus slightly shorter than inner, bearing 1 spine on outer margin and 4 distal spines on apex. *Uropod 2* (Figure 2C): peduncle bearing 1 and 2 spines on inner and outer margins, respectively; inner ramus as long as peduncle with 1 spine on outer margin and 4 distal spines; outer ramus unarmed, slightly shorter than inner, bearing 4 distal spines on apex. *Uropod 3* (Figure 2D): biramous; outer ramus 7 times longer than inner, biarticulate (terminal article very small, barely visible), bearing 3 spines and clusters of long stiff setae apically; inner ramus minute, scale-like with 1 long apical seta. *Body length* 7.0–9.0 mm. *Body colour* with distinct banding.

Remarks. The morphology of the palmar margin of gnathopods 1, 2 and uropods 1, 2 is described for the first time. Our observations also confirm the significant differences in the structure of gnathopods 1, 2 and uropod 3 noted by Bazikalova (1945). According to this author, the body length of adult amphipods ranges from 9.0–15.0 mm, whereas the ovigerous female that we collected had a body length of only 7.0 mm. As Mekhanikova (2000) has shown, *Gm. fasciatus* is highly polymorphic morphologically, but this variation is clearly correlated with the environmental conditions in which the species is found. The species could be genetically heterogeneous as well: mt-*cox1* sequence comparisons revealed at least four sub-groups in the Baikalian population of the species (Gomanenko *et al*., 2005, our Figure 3). However, correlations between the morphological features of the species and molecular data have not yet been found.

Gmelinoides fasciatus was collected in a small spring brook (approximately 3 m wide) located in a low gully in the bushy Gunt River floodplain. The main abiotic factors in the brook: flow velocity $\sim$0$-$0.01 m$\cdot$s^{-1}, depth $\sim$0.1$-$0.5 m, water temperature –10 °C. The species was collected from grass roots approximately 70 m from the brook source in the riparian zone. The amphipods were absent from the spring source. All areas of the bottom of the spring brook were thickly overgrown with dense clumps of green filamentous algae, which may indicate an increased mineralisation of the spring. The brood pouch of the female contained 29 eggs.

Under natural conditions, the indigenous Baikalian populations of *Gm. fasciatus* occur under a wide range of water temperatures, but the species is generally thermophilic. *Gmelinoides fasciatus* has been found in cold (4.0–8.5 °C) springs in water bodies surrounding Lake Baikal as well as in several thermal Baikalian springs with temperatures close to 29 °C (Takhteev, 2009). It has been shown experimentally that the final selected thermopreferendum (FST or final thermopreferendum) for these crustaceans is in a warmer range, 17–18 °C (Timofeyev *et al*., 2001). New data on heat-shock proteins (HSP) indicate that the species is a thermophilic relict (Protopopova *et al*., 2011). This evidence additionally furnishes an excellent demonstration that *Gm. fasciatus* is adapted to a wide temperature range, from 0 to +30 °C (see Baikalogy, 2012, p. 872).

The spring brook near Charthem kishlak supports a relatively impoverished invertebrate fauna. After *Gm. fasciatus*, the most significant subdominant was the mayfly *Ameletus alexandrae* Brodsky 1930. *Baetis* (*Rhodobaetis*) ex gr. *oreophilus*, unidentified caddisflies of the families Lepidostomatidae and Limnephilidae, chironomids (the most numerous of which was the crenophilous *Pseudodiamesa*), the predatory dytiscid beetle *Agabus* and the lymnaeid gastropod *Radix* were also found in the spring, in addition to a common gammarid amphipod, *G. lacustris*. The observed density of this species was notably low, only 3 individuals, in contrast to 150 specimens of *Gm. fasciatus*.

Based on its life form, *Gm. fasciatus* is a benthic phytophilous-omnivorous-fossorial species (Takhteev, 2000), attributed in the same publication to the “variable-modes group”. Generally, alien species are extremely abundant in the environments that they invade, and they either significantly depress indigenous crustaceans or completely exclude them: *e.g.*, in the Volga River and its reservoirs it suppresses the Ponto-Caspian taxa, but in the other regions it supresses *G. lacustris* (Kamaltynov, 2001, 2009). *Gm. fasciatus* has most likely excluded the Caspian species *Obesogammarus obesus* (G.O. Sars 1894) from the Gorkovskoe reservoir, where it formerly lived (Mordukhai-Boltovskoi, 1979), whereas it notably reduced the distributional area and decreased the density of both *G. lacustris* and *Asellus aquaticus* (Linnaeus 1758) in Lake Ladoga (see Berezina *et al*., 2009, p. 405).

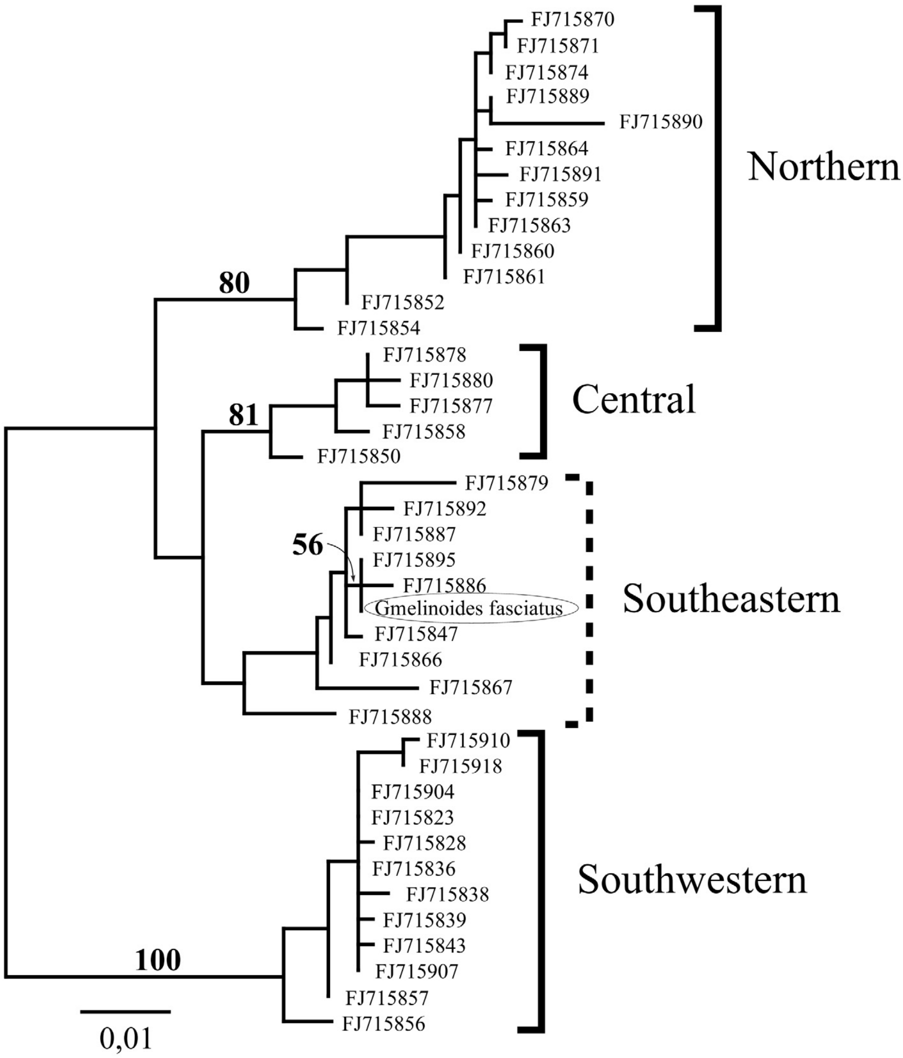

Figure 3

ML phylogenetic tree (GTR+I+Γ model) based on HF937342 mt-cox1 nucleotide sequence comparisons. Data set includes 39 sequences obtained from Gm. fasciatus Baikalian populations and one from the specimens found near Charthem kishlak (southern Pamir Mountains (Tajikistan). Clades designation followed Gomanenko et al. (2005). The numbers at the branches indicates ML bootstrap value support (100 replications).

The amphipod fauna of Tajikistan is still poorly investigated. Only fragmentary data about its composition are available. Nevertheless, its high level of endemicity is evident. In particular, the following species are known from cold springs in Tajikistan: *Comatogammarus ferghanensis* (Martynov et Behning 1948) – Darvaz mountain ridge, northwards from Kalai-Humb; *Barnardiorum shadini* (Birstein 1948) – Saradzhou River, tributary of the Varzob River, springs near Hodza-obigarm, Obisafet, Kvaka; and *Tadzocrangonyx schizurus* (Birstein 1948) – Hissar mountain ridge (exact location unknown). Considering the great ease with which *Gm. fasciatus* can exclude native species from the biotic environment, this invasive species poses a definite risk to not only endemic crenophilous Tajikistan species but also several species of *Sarothrogammarus* from Afghanistan associated with the Panj River basin. Therefore, future thorough studies of *Gm. fasciatus* in the Pamir region are much necessary.

Our phenotypic data (Figures 1C, 2A–2G) and the results of our phylogenetic analyses (Figure 3) confirm the occurrence of *Gm. fasciatus* in the Southern Pamir Mountains. BLAST search (blast.ncbi.nlm.nih.gov) revealed that our sequence is identical to *Gm. fasciatus* sequences in the GenBank (FJ756328, FJ715895, FJ715896, and FJ715849). According to the tree obtained (Figure 3) our specimen from Pamir was a member of "Southeastern" cluster having no bootstrap support.

The former southernmost limit of the range of the species was previously represented by the Bukhtarminskoye reservoir, Eastern Kazakhstan (Ioffe, 1974). Given the significant rate of *Gm. fasciatus* range expansion (ca. 24-35 km/per year during the increasing phase of population outbreaks (Matafonov *et al*., 2005)), it is highly likely that this species will be found in freshwater ecosystems in the western part of Central Asia and in the middle reaches or tributaries of the Amu Darya River.

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