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First record of Pontian Monkey Goby, *Neogobius fluviatilis* (Pallas, 1814) in the Evros River (Greece); Is it an alien species?

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Abstract

The Pontian Monkey Goby, *Neogobius fluviatilis* (Pallas, 1814), was recorded for the first time in Greece in August 2011. Eight specimens were collected in the Greek-Turkish section of the Evros River, 65 kilometers upstream of its river-mouth. Although the species has been recently discovered in the Tundza, a tributary of the Evros in Bulgaria, it has never before been found in the Evros' main stem. Although the lower Evros has been poorly researched by ichthyologists, it is unlikely that a conspicuous medium-sized fish would go unnoticed in this river; and, it is therefore suggested to be a probable alien. However, since the Evros basin has had geological connections to the Marmara Sea and Black Sea in the past and it is immediately adjacent to native populations of *N. fluviatilis*, the species status is categorized as questionable until genetic and morphological studies are completed.

Keywords: Neogobius fluviatilis; Greece; Bulgaria; Turkey; Evros; Maritsa; Meriç; Alien.

Introduction

A recent electrofishing expedition revealed the existence of the Pontian Monkey Goby *Neogobius fluviatilis* (Pallas, 1814) on the lower main stem of the river Evros-Maritsa-Meriç (hereafter referred as Evros, the river's classical Greek name). This is the first record of the species in Greece and its second published documentation in lotic environments in this river basin. Although many river reaches have been recently surveyed in the Bulgarian section of the Evros, the species was recently found only in one section of the Bulgarian Tundza tributary (STEFANOV *et al.*, 2008), where it is well established and provisionally considered as non-native.

The native distribution of *N. fluviatilis* is known to encompass many Black Sea river basins and parts of the Caspian basin including the lower parts of associated lot-

ic systems (Dnieper, Dniester, Danube, Don and Volga rivers), and the Sea of Marmara basins (KOTTELAT & FREYHOF, 2007). According to NEILSON & STEPIEN (2011) certain Caspian populations are currently considered a different species, Neogobius pallasi. Moreover, N. fluviatilis is notorious for its remarkable East-to-West invasions within Europe in recent decades. Assisted by river navigation channel creation and accidental introductions the species begun a rapid expansion in the rivers of the northern Black Sea since 1970 (COP et al., 2005). By the first years of this century alien populations of the species have been found far up the Danube in Germany, throughout the main stem of the Vistula in Poland, and more recently as far west as the Rhine in the Netherlands (VAN KESSEL et al., 2009).

The new distributional locality and specimens

The Evros river remains one of the poorest studied large river basins in the Balkans in terms of its ichthyofauna, and this is especially true of its lower section where several new discoveries have recently been made (KOTTELAT & ECONOMIDIS, 2006; FREYHOF et al., 2008; ZOGARIS et al., 2009a; KOUTRAKIS et al., 2011). Although the Bulgarian section was rather well explored by the 1960's (RUSSEV, 1966), the Greek tributaries of the Evros were first surveyed for fish in 1970-72 by ECONOMIDIS (1974). After that, the wider border area was heavily militarized following worsening Greek-Turkish relations since the 1974 Cyprus conflict. Access to the 200 kilometer border zone of the Evros river in Greece and in Turkey is strictly restricted and very few fish surveys have been conducted. Therefore, it is possible that even native fish species or longestablished aliens may have gone unnoticed in the lower Evros.

Members of the Institute of Inland

Waters-HCMR (IIW-HCMR) recently begun electrofishing surveys and have now widely sampled the Evros' Greek tributaries. Between 2009-2011, fifty-five river sites have been surveyed for their ichthyofauna within Greece's section of the Evros river basin, following the rapid quantitative sampling protocol (described in ECONOMOU et al., 2007). During one of these surveys, on the 25th of August 2011, eight specimens of N. fluviatilis were caught on a one-bank 500 meter longitudinal transect of the main stem of the Evros using a Smith-Root 24 volt back-pack electrofisher (battery output voltage: 50 to 990V; 400W maximum continuous). The location of the sampling site (40°) 58' 35 N, 26°19'40 E) is 65 river kilometers from the Evros' river-mouth between the villages of Tychero and Gemisti at an elevation of about 7 meters above sea level. Eighteen fish species were caught at this site including the following aliens in descending order of abundance: Carassius gibelio (68 individuals), Pseudorasbora parva (13), Lepomis gibbosus (6), Gambusia holbrooki (5). The only other goby species present was the native Proterorhinus semillunaris (25 individuals). Neogobius fluviatilis inhabited sand and fine gravel substrates in the main channel of the river near its shoreline at depths ranging from 30 to 60 cm. The length of the collected specimens ranged between 58 - 71 mm Sl. Five of the collected specimens were preserved in alcohol (80% solution) and three were transported alive to an aquarium (Fig.1).

N. fluviatilis is easily identified from other large freshwater gobiid species by the following attributes (PINCHUK *et al.*, 2003; KOTTELAT & FREYHOF, 2007): (1) the second dorsal fin is uniformly lowering down from the first rays to the last ones; (2) the first branched ray of the second dorsal fin is about twice as long as the penultimate ray;



Fig. 1: First Neogobius fluviatilis (Pallas, 1814) caught in Greece (Photo: S. Zogaris, 27.08.2011; ex situ at HCMR-Anavissos).

and (3) the lack of a black spot in the posterior part of the first dorsal fin. Also, the width of its fairly large head is equal to or a bit greater than the height of the head, and terminates in a leaf-shaped snout. Morphologically the specimens of the Evros are immediately identified as *N. fluviatilis*, as they show all the above characters. However no complete morphological or comparative genetic work has ever been published on samples from the Evros.

Distribution in adjacent basins of Bulgaria and Turkey

Regular sampling by the Institute of Biodiversity and Ecosystem Research (IBER) in Bulgaria has verified that the species is fairly widespread in the whole Bulgarian Danube section, the lower section of some Danube tributaries, and several Black Sea coastal lakes and rivers of the country (Fig 2). It has also been recorded in the Black Sea shore in sandy bottoms and shallow waters (1-2m depth) such as in Butamia Bay (42° 03' 24 N, 27° 59' 16 E) near Sinemorets village. Despite this, the species is presumably absent from several sites, such as Bulgaria's Northern Black Sea lakes Shabla-Ezerets complex, Durankulak and near Varna. The Bulgarian populations outside the Danube/Black Sea area are provisionally considered as non-native (POLACIK et al., 2008). The only known populations within the Evros basin are in the upper section of



Fig. 2: Recently documented *Neogobius fluviatilis* distribution in Bulgaria and NW Turkey (data from IBER surveys and available bibliography). Where specific location information is not known, but presence has been confirmed in the basin, a question mark symbol (?) is shown. The new site in the Evros is shown with a five-point star symbol.

the Tundja River $(42^{\circ} 03' 48 \text{ N}, 26^{\circ} 30' 18 \text{ E})$, where the species was first collected in 2006; and an unconfirmed population in a reservoir within the Sazliika tributary of the Evros $(42^{\circ} 18' 12 \text{ N}, 25^{\circ} 53' 45 \text{ E})$ (STEFANOV *et al.*, 2008).

Our observations of *N. fluviatilis* nearly on the Greek-Turkish border and its establishment in a large section of the Bulgarian Tundza (180 river kilometers upstream of our new find), suggest that the species is most likely established in Eastern Thrace, within Turkish territory. However, we have found no published records of the species in the Turkish tributaries of the Evros (ERKAKAN, 1983; BALIK, 1985; ILHAN, A., *pers. com.*). Furthermore although the species is known to occupy a wide area, including rivers and wetlands on the southern shores of the Sea of Marmara (KOTTELAT & FREYHOF, 2007), specific geographic documentation is presumably scarce in the published literature (SARI *et al.*, 2006; ÖZULUĞ *et al.*, 2007; SASI & BERBER, 2010) (Fig. 2).

N. fluviatilis is often one of the most abundant fish species in its native range (KOTTELAT & FREYHOF, 2007) and this has been confirmed in the Bulgarian Black Sea basins. Survey data from recent IBER surveys in Bulgaria always showed higher biomass of *N. fluviatilis* in comparison with other gobies, including the invasive *Neogobius melanostomus* when more than one species are established at the same site. Only in the case of Tundza and Evros does *N. fluviatilis* show lower abundance in comparison with *Proterorhinus semillunaris*. The abundance of *N. fluviatilis* in the Tundza is 4-10 specimens per 100 m river transect using electrofishing (perhaps similar to the situation in the new site on the lower Evros). The fact that *N. fluviatilis* tends to inhabit primarily the lower courses of rivers including the main stem of the Danube (unlike *Neogobius gymnotrachelus* which is also met in upland cool-water locations), indicates the species' preference for larger lotic waters and even slightly brackish water.

Are the Evros' Pontian Monkey Gobies alien?

In our opinion, the newly recorded population of N. fluviatilis is probably not native to the Evros basin, but there is currently no reliable evidence to prove that it was definitely absent from this river in the past. Based on the species' conspicuous behavior and rather large size, it is unlikely that it would go unnoticed by past researchers, if it was native. The most probable hypothesized vector for invasion is that it may have been transferred accidently through fish stocking practices to the Tundza or to Bulgarian artificial reservoirs, and has now established localized river populations. Fish stocking and associated translocations are frequent in the Bulgarian part of the basin and several freshwater organisms have infiltrated to Greece through trans-boundary rivers, especially from Bulgaria (ECONOMIDIS et al., 2000; ZENETOS et al., 2009).

However, caution is needed in accepting an alien status for *N. fluviatilis* in the Evros due to the close proximity of this river to the species' native distribution range (i.e. Sea of Marmara). Moreover, if the Evros and the Sea of Marmara basins are located within a common biogeographical ecoregional area should the alleged non-native be called an alien or a translocated species? Although there is still some controversy regarding regional biogeographical boundaries (ZOGARIS *et al.*, 2009b), the 'Thrace Freshwater Ecoregion' as defined in ABELL *et al.* (2008) includes the entire Marmara basin and the wider northeastern Aegean basins (Evros, Nestos and Strymon). In fact, this broad freshwater biogeographical ecoregion delineation seems to be supported by evidence from regional geological history, creating conditions that may challenge the alien status of the species within the Evros.

The Evros basin has been affected several times by influences of Black Sea/Marmara waters and associated biota since the time it is postulated to have had its rivermouth in the Sea of Marmara, 1.5 million years ago (OKAY & OKAY, 2002). Furthermore, in the Late Pliocene, the Tundza tributary is considered to have been independently flowing directly into the Black Sea and later became a branch of the Evros due to a river-capture event (ANGELOVA, 2003; STEFANOV et al., 2008). During the Pleistocene, the biogeographic relationship between the Evros and the Black Sea/Sea of Marmara biota is said to have had a direct connection due to the glacially-affected outflow of Black Sea water through the Dardanelles to the northern Aegean land-mass. This hypothesis, known as the Aegeopotamos, proposes that the Black Sea freshwater flood outflow had direct confluences with the Thracian rivers during this geological event (KOSSWIG & BATTALGIL, 1943; BANARESCU, 2004). This unique geological history may explain the native status of several Pontic and Anatolian species in the Evros, including the native population of Ponto-Caspian Proterorhinus semillunaris and the presence of the rare cobitid, Cobitis puncticulata, formerly considered unique to the Sea of Marmara wetlands, but recently also discovered in the Evros tributary at Lira in Greece (FREYHOF et al., 2008).

All the above suggest that it is best to

provisionally consider N. fluviatilis in the Evros as "questionable" with respect to its alien status, since the origin of the Evros specimens have not been genetically inspected. A questionable status (as per ZENETOS et al., 2010) may be characterized by insufficient or unverified information to confirm alien status. It is well known that labeling a species as 'alien' requires extreme caution (CARLTON, 1996) and must include specific biogeographical considerations (COP et al., 2005). The difficulty of interpretation is greater when the alleged alien is discovered immediately next to its native range, within a common ecoregion, and where historic species distribution data are scarce or non-existent. A genetic and morphologic comparison among N. fluviatilis from the Evros, Danube, Marmara and Black Sea basins would be appropriate for the clarification of the Evros specimens' phylogeny and population origins. Further surveys and monitoring are urgently needed within the Evros river basin, in order to assess potential "invasive" behavior of the species; including range alterations, population trends, and habitat-use in both lotic and transitional waters.

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