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## First survey of shallow-water Amphipoda along the Georgian Black Sea coast reveals new faunistic records and the unexpected Atlantic invader *Melita nitida*

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### Abstract

We present findings from the first survey of shallow-water amphipods conducted along the Black Sea coast in Georgia. Eight species from five families were identified, all but one being new for the Georgian fauna. Most are usual inhabitants of the Black and Mediterranean seas; however, we report the first record of the invader *Melita nitida* in this region. This species, originating from North America, was previously known in Europe only on the Atlantic and Baltic coasts. Its discovery in the Black Sea implies either jump dispersal or that its distribution is more widespread than is currently recognized. Given that the total number of species reported in Georgia is lower than that reported for the other countries that neighbour the Black Sea, we anticipate the discovery of new taxa in future surveys. Our study highlights the importance of faunistic exploration in previously overlooked regions for detecting potentially cryptic invasions and corroborating biogeographical patterns.

**Keywords:** biogeography; Amphipoda; Caucasus; diversity; invasive species.

### Introduction

The Black Sea harbours a generally impoverished biota due to significant fluctuations in salinity since its Late Miocene formation until the Holocene (Badertscher *et al.*, 2011). Its amphipod crustacean fauna is euryhaline and is comprised almost entirely of recent Mediterranean immigrants, with a few autochthonous Ponto-Caspian relicts being exceptions. Overall, the fauna comprises ca. 24 families, 49 genera and 89 species (Sezgin & Katağan, 2007; Grintsov & Sezgin, 2011). Amphipods are well known in most countries that are adjacent to the Black Sea, i.e., Bulgaria (Uzunova, 2012), Romania (Petrescu, 1994, 1998), Russia (Miloslavskaya, 1939), Ukraine (Grintsov & Sezgin, 2011; Kudrenko, 2016), and Turkey (Sezgin & Katağan, 2007). In each of these countries, dozens of species have been reported. In contrast, the coastal amphipod fauna in Georgia has remained very little explored, with only four taxa reported to date from the sea itself (*Ampithoe* sp.–Ampithoidae, *Gammarus* sp. and *Echinogammarus ischnus* (Stebbing, 1899)–Gammaridae, and *Phtisica marina* Slabber, 1769–Caprellidae) (Birstein, 1935; Komakhidze & Mazmanidi, 1998) and two reported from the brackish Paliastomi Lake and mouth of the Rioni River (*Pontogammarus robustoides*

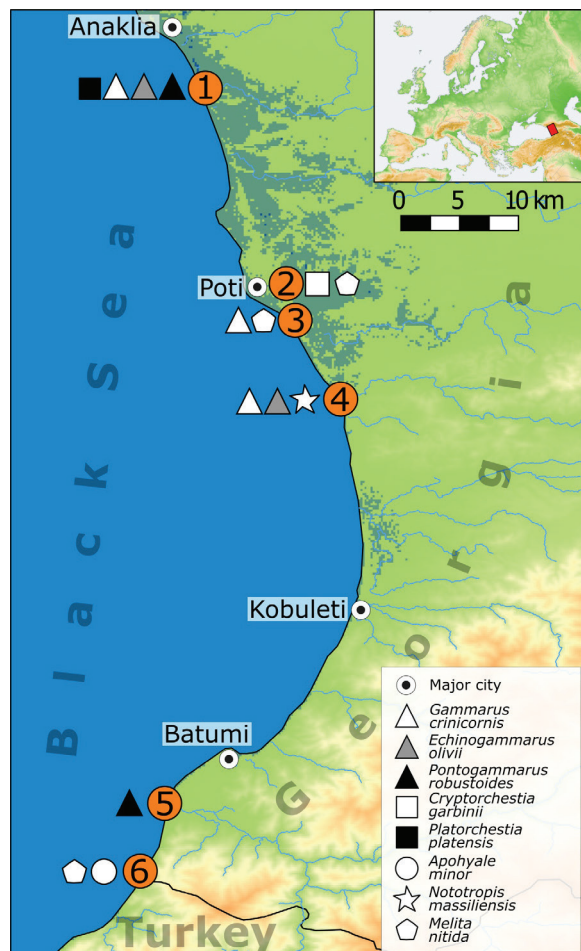
(Sars, 1894)–Gammaridae, and *Chelicorophium curvispinum* (Sars, 1895)–Corophiidae) (Derzhavin, 1924). The Georgian coast is biogeographically important as it lies in the Caucasus biodiversity hotspot and served as a long-term glacial refugium (Tarkhnishvili *et al.*, 2012; Mumladze *et al.*, 2019). Moreover, due to increasing shipping activity, the Black Sea is receiving increasing numbers of invasive species (Shiganova, 2010). As such, better knowledge of amphipod diversity along the Georgian coast will (1) help improve biodiversity estimates, (2) provide insight into the dispersal of invasive species, and (3) allow the distribution of Black Sea species to be updated.

### Material and Methods

Material was collected from six localities along the Georgian Black Sea coast between the 2<sup>nd</sup> and 5<sup>th</sup> of August 2019 (see Results) (Table 1, Fig. 1). We aimed to cover a broad diversity of habitats, including seashores, river mouths and adjacent saline lagoons. Amphipods were collected with standard hand nets (0.5 mm mesh size) and fixed in 96% ethanol in the field. Specimens were examined under a Nikon SMZ1000 stereomicro-

**Table 1.** Information on localities, habitats and taxa collected.

No.	Locality	Lat.	Lon.	Date	Habitat	Species	N
1	Tikori, mouth of Churia River	42.334	41.611	05.08.2019	Sublittoral, sand and vegetation Depth: 0.1–1 m	<i>G. crinicornis</i> <i>E. olivii</i> <i>P. robustoides</i>	1 ♂ 2 ♀, 1 ♂ 3 ♀, 4 ♂
					Supralittoral, sand, pebbles, driftwood	<i>Pl. platensis</i>	4 ♀, 1 ♂
2	Poti, Paliastomi Lake shore	42.132	41.697	03.06.2018	Supralittoral, under rocks	<i>C. garbinii</i>	3 ♀, 7 ♂, 5 juv.
				04.08.2019	Sublittoral, wooden pier, barnacles Depth: 0.5 m	<i>M. nitida</i>	12 ♀, 9 ♂
3	Poti, outflow of Paliastomi Lake	42.095	41.707	04.08.2019	Sublittoral, algae covered rocks Depth: 0.1–0.5 m	<i>G. crinicornis</i> <i>M. nitida</i>	4 ♀, 3 ♂, 1 juv. 11 ♀, 9 ♂, 1 juv.
4	Magnetiti, Black Sea shore	42.015	41.752	04.08.2019	Sublittoral, sand/algae growing on shipwreck Depth: 0.1–1.2 m	<i>G. crinicornis</i> <i>E. olivii</i> <i>N. massiliensis</i>	2 ♂ 14 ♀, 11 ♂, 1 juv. 19 ♀, 18 ♂
5	Batumi, mouth of Chorokhi River	41.602	41.573	03.08.2019	Sublittoral, sand and vegetation Depth: 0.1–1 m	<i>P. robustoides</i>	24 ♀, 21 ♂, 7 juv.
6	Kvariati, Black Sea shore	41.533	41.549	02.08.2019	Eulittoral, algae on boulders Depth: 0 m	<i>A. minor</i>	17 ♀, 22 ♂
					Sublittoral, under stones Depth: 0.1–0.3 m	<i>M. nitida</i>	4 ♀, 3 ♂, 3 juv.



**Fig. 1:** Map of the sampling localities along the Black Sea coast in Georgia. Symbols represent families, and shades represent species (triangles: Gammaridae, squares: Talitridae, circles: Hyalidae, stars: Atylidae, pentagons: Melitidae).

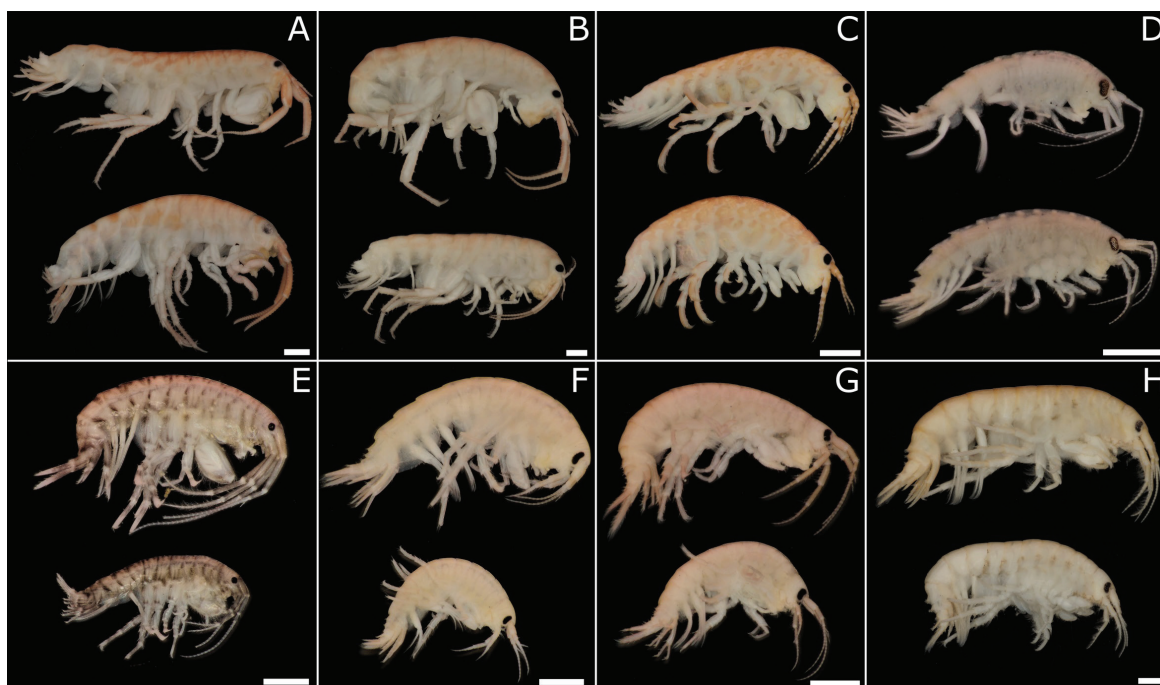
scope and identified using appropriate keys (Ruffo *et al.*, 1993; Grintsov & Sezgin, 2011; Krapp-Schickel & Sket, 2015). Photographs were taken with a Nikon D90 camera equipped with an AF Tamron 70-300 mm F/4-5.6 lens, a Raynox DCR-250 macro attachment, and a Delta DRF-14 flash.

## Results

Eight species belonging to five families were identified (Figs. 2, S1): Gammaridae: *Echinogammarus olivii* (H. Milne Edwards, 1830), *Gammarus crinicornis* Stock, 1966 and *Pontogammarus robustoides* (Sars, 1894); Talitridae: *Cryptorchestia garbinii* Ruffo, Tarocco & Latella, 2014 and *Platorchestia platensis* (Krøyer, 1845); Hyalidae: *Apohyale minor* (Chevreux & Fage, 1925); Melitidae: *Melita nitida* S.I. Smith in Verrill, 1873; and Atylidae: *Nototropis massiliensis* (Bellan-Santini, 1975). All species except *P. robustoides* are herein reported for the first time for the fauna of Georgia. Detailed results are presented in Table 1.

## Discussion

Our study is the first survey dedicated to the exploration of shallow-water amphipods along the Black Sea coast in Georgia. We identified seven new species for the fauna of this country. Based on the present data and published data, the Georgian Black Sea coastal fauna consists of 12 species from 8 distantly related families, revealing a high degree of phylogenetic diversity (Copilaș-Ciocianu *et al.*, 2020).



**Fig. 2:** Species sampled in this study. Males are above, and females are below. (A) *Platorchestia platensis*, (B) *Cryptorchestia garbinii*, (C) *Apohyale minor*, (D) *Nototropis massiliensis*, (E) *Melita nitida*, (F) *Gammarus crinicornis*, (G) *Echinogammarus olivii*, (H) *Pontogammarus robustoides*. Scale bar = 1 mm.

Biogeographically, the species composition is similar to that of the other countries that border the Black Sea. The autochthonous Ponto-Caspian elements are represented by *C. curvispinum*, *E. ischnus* and *P. robustoides* (Derzhavin, 1924; Birstein, 1935). In the current study, we identified the last species from two new sites but did not collect *C. curvispinum* and *E. ischnus*. These three species are widely distributed throughout the Ponto-Caspian realm and are successful invaders throughout freshwater and brackish water habitats in Europe (Arbačiauskas *et al.*, 2011). Intriguingly, we did not encounter *C. curvispinum* and *P. robustoides* in the brackish Paliastomi Lake, where both have been previously reported. The only amphipod sampled was *M. nitida*, suggesting an increase in salinity. It is possible that the Ponto-Caspian species retreated towards the less saline river inflows. All of the remaining species that are known to inhabit the Georgian coastline are recent immigrants, being either typical Mediterranean/Atlantic elements (Ruffo *et al.*, 1993) or cosmopolitan species.

Our most important finding is the discovery of *M. nitida*, a species never before reported from the Mediterranean or Black Sea. A native of the North American Atlantic coast, it was introduced to Europe by shipping, where it currently inhabits the Atlantic and Baltic shores (Normant-Saremba *et al.*, 2017). Its occurrence in Georgia is puzzling; it suggests either jump dispersal or a broader-than-recognized, cryptic distribution in the Mediterranean area. We found this species at three locations, ranging from the sea to the brackish Paliastomi Lake and its outflow, in accordance with its resilience to salinity changes (Normant-Saremba *et al.*, 2017). Males, females and juveniles were collected, indicating the existence of viable populations. A very similar species, *M. mirzajani*

*ni* Krapp-Schickel & Sket, 2015, was recently described from the Bandar-e Anzali port on the Caspian Sea in Iran (Krapp-Schickel & Sket, 2015). Its recent appearance but unknown origin is intriguing. Both *M. nitida* and *M. mirzajani* dwell in similar habitats of decreased salinity and high anthropogenic influence. They are also very similar morphologically, the main difference involving the setosity of the flagellum of the second antenna in males. Given these similarities, we suggest that *M. mirzajani* might be a junior synonym of *M. nitida*. Future studies employing DNA barcoding and detailed morphological analyses are needed to clarify this issue.

Although our study significantly increased the number of coastal species in Georgia from 5 to 12, this number is considerably lower than those for other Black Sea neighbouring countries and probably represents a fraction of the total diversity. However, the eastern Black Sea shelf is the narrowest part of the shelf and slopes abruptly below the 200 m depth limit of the oxygenated water level (Barale, 2008). Given these limiting conditions, the eastern coast might indeed harbour a reduced diversity. Nevertheless, it is clear that more species await discovery, and we emphasize the need for further research.

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## Supplementary Data

The following supplementary information is available for the article online:

**Fig. S1:** Characteristics of *Melita nitida*, collected from the outflow of Paliastomi Lake. (A) pereopod 7, (B) uropod 3, (C) gnathopod 2, (D) antenna 2, (E) urosome. Scale bar = 0.2 mm.