Ruthenica, 2019, vol. 29, No. 2: 103-113.

http: www.ruthenica.com

"Invasion" in the Russian Arctic: is global Climate Change a real driver? A remarkable case of two nudibranch species

Irina A. EKIMOVA[1,5], Tatiana I. ANTOKHINA[2], Dimitry M. SCHEPETOV[1,3,4]

[1]*Lomonosov Moscow State University, Leninskie Gory 1-12, 119234 Moscow, RUSSIA;*
[2]*A.N. Severtsov Institute of Ecology and Evolution of Russian Academy of Sciences, Leninsky prospect 33, Moscow 119071, RUSSIA;*
[3]*N.K. Koltzov Institute of Developmental Biology of Russian Academy of Sciences, Vavilov str. 26, 119334 Moscow, RUSSIA;*
[4]*National Research University Higher School of Economics, Myasnitskaya str. 20, 101000 Moscow, RUSSIA*
[5]*Corresponding author; E-mail: irenekimova@gmail.com*

ABSTRACT. In present paper, we report the first findings of two large and conspicuous nudibranch species for Russian waters – *Goniodoris nodosa* and *Flabellina pellucida*. Both species show wide distribution in the northeastern Atlantic waters or even in the northwestern Atlantic in the case of *F. pellucida.* The morphology of both species was studied using anatomical dissections and scanning electron microscopy. Molecular diversity was assessed by sequencing of COI marker and reconstruction of haplotype networks. In the Barents Sea, both species show minor genetic differences from their European relatives and similar morphological characters. Putative explanations of new findings for Russian Arctic are discussed.

Introduction

Discussion of global Climate Change as a driver of biocenotic changes is a growing prominent trend in biodiversity studies during the last decades [Post *et al.*, 2009; Wassmann *et al.*, 2011]. At the same time, with growth of anthropogenic pressure many introduction events have been reported. Such events are numerous for aquatic ecosystems due to long-term disruption in marine communities, caused by agricultural and industrial projects and ballast waters acting as a vector of invasion [Ruiz *et al.*, 2000; Occhipinti-Ambrogi, Savini, 2003]. These two phenomena often tend to be discussed as coinciding. Contemporary methods of molecular phylogenetics give us a powerful tool to distinguish real invasions from cases of understudied fauna, and recent events from ones that occurred in relatively distant past [Leydet, Helberg, 2015]. A prominent case of such putative Climate Change role is the Barents Sea fauna expansion. This area is a keystone for Arctic biodiversity and hydrology, linking Atlantic and Arctic water masses. It is believed that global warming affects this region drastically, which possibly causes massive introduction of boreal species into the Arctic [Chan *et al.*, 2018]. Recently new species of various invertebrate taxa were recorded for the first time for the Barents Sea and consequently attributed as novel invasions: nudibranch molluscs [Martynov *et al.*, 2006; Zakharov, Jorgensen, 2017], caenogastropods [Kantor *et al.*, 2008], bivalves [Deart *et al.*, 2013], cephalopods [Golikov *et al.*, 2013; 2014], annelids [Rzhavsky *et al.*, 2011], and echinoderms [Zakharov *et al.*, 2016]. In further text, we use term "invasion" in a sense used in works concerning the Russian Arctic fauna, *i.e.* any introduction of new populations not caused by a gradual area expansion.

In the present work, we follow recent trend in updating of the Barents Sea faunistics with discovering of two conspicuous nudibranch species (Mollusca, Gastropoda) new to Russian waters and this area in particular. Dorid nudibranch *Gonidodoris nodosa* (Montagu, 1808) is reported from North-East Atlantic (north Spain, along French Atlantic coast to British Isles) to the North Sea and western Barents Sea with Varangerfjord as the northernmost location [Thompson, Brown, 1984; Eversten, Bakken, 2002; 2005] (Fig. 1A). A cladobranch, *Flabellina pellucida* (Alder et Hancock, 1843) is a widespread species, being reported from North-West Atlantic (from Massachusetts to North Canada), Mediterranean coast, North-East Atlantic (British Isles, Norway and Faeroes) with central Norway as the easternmost and northernmost location [Lemche, 1929; Kuzirian, 1979; Thompson, Brown, 1984; Eversten, Bakken, 2005] (Fig. 1B). At least for *F. pellucida* the previously known easternmost record is in about 1300 kilometers to the southwest and therefore its findings in the Barents Sea can be attributed as an invasion event. However,

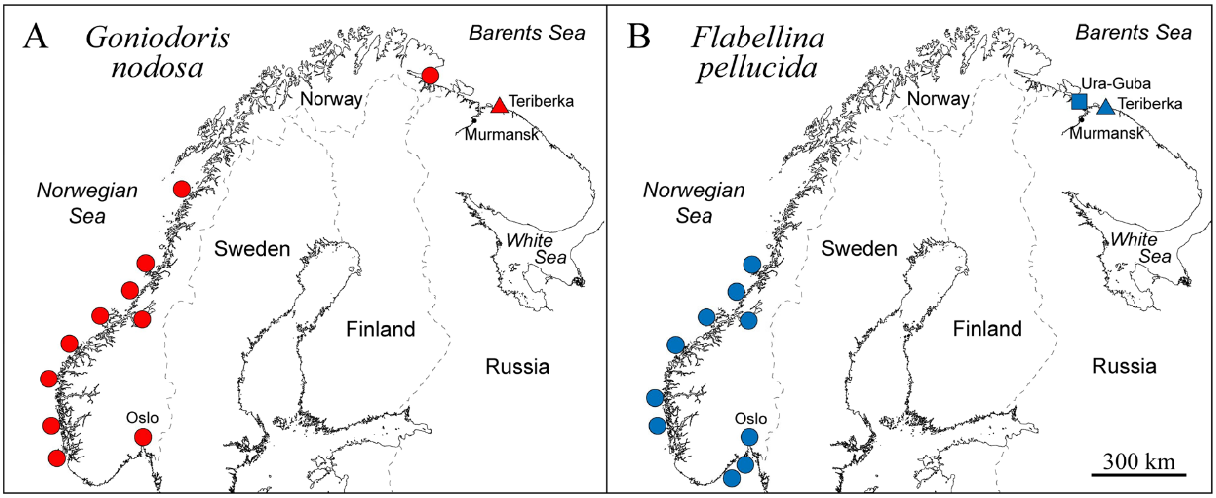

FIG. 1. Known distribution of studied species in northern Europe (circles indicate previous records, triangles and squares - original data). **A**. *Goniodoris nodosa*. **B**. *Flabellina pellucida*. Teriberka area includes Teriberka Bay and Zelenaya Inlet; Ura-Guba area incudes Ura Inlet.

РИС. 1. Известное распространение исследованных видов в водах северной Европы (круги обозначают предыдущие находки, треугольники и квадраты - оригинальные данные). **A**. *Goniodoris nodosa*. **B**. *Flabellina pellucida*. Район с.Териберки включает губы Териберская и Зелёная; район с.Ура-губы включает губу Ура.

other explanations are plausible, *e.g.* survey incompleteness, gradual area expansion or restoration of depressed native population, *etc*. Therefore, our main goal is to investigate molecular and morphological diversity of *F. pellucida* and *G. nodosa* from the Barents Sea as thoroughly as possible with available specimens.

Material and methods

Material

Material was collected in three localities of the Murmansk coast of the Barents Sea during summer and autumn seasons of 2015 and 2018: (1) Zelenaya Inlet, 25-27 Aug. 2015, 2 specimens of *F. pellucida*; (2) Teriberka Bay, 25 Aug. – 1 Sep. 2018, 4 specimens of *F. pellucida*, 2 specimens of *G. nodosa*; (3) Ura Inlet, 18 Nov. 2018, 2 specimens of *F. pellucida* (Table 1). Specimens were collected during scuba diving at depths of 2-20 m on hard bottoms. Water temperature in the collection sites ranged from +7 to +10ºC in August-September and was +5ºC in November. Each mollusc was relaxed and photographed using Sony NEX-5N camera and then fixed in 96° ethanol. Several specimens were photographed in their natural environment under water using the same camera. Voucher specimens and DNA samples are stored in the personal collection of the first author and are available by a request.

Taxonomic remarks

As discussed in several recent works the alpha-taxonomy of the family Flabellinidae *s.l.* is controversial and different generic divisions have been proposed [Korshunova *et al*., 2017a; Gosliner *et al*., 2018]. According to recent revision by Korshunova *et al.* [2017a], the species *Flabellina pellucida* has been transferred into a new genus *Carronella* Korshunova, Martynov, Bakken, Evertsen, Fletcher, Mudianta, Saito, Lundin, Schrödl et Picton, 2017. However, Gosliner *et al*. [2018] suggested retaining most of Flabellinidae *s.l.* diversity in the genus *Flabellina* McMurtrie, 1831 until a dedicated revision is undertaken. For ease of comparison and comprehension we use a stable version of Flabellinidae *s.l.* taxonomy in the present study as published in Furfaro *et al.* [2018].

Morphological analysis

The external morphology of the specimens was studied under a stereomicroscope. The buccal mass of each specimen was extracted and soaked in proteinase K solution for 2 hours at 55ºC to dissolve connective and muscle tissues, leaving only the radula and the jaws. The coated radulae were examined and photographed using scanning electron microscope EVO-40 Zeiss. The reproductive system of both species was examined using the stereomicroscope. The features of the jaws of *F. pellucida* were analyzed under the stereomicroscope and scanning electron microscope JEOL JSM.

Molecular analysis

DNA was extracted from small pieces of foot tissue using PALL™ AcroPrep 96-well plates by

Table 1. Specimens used in the present study with information on vouchers and GenBank accession numbers.

Таблица 1. Изученные особи с указанием регистрационных номеров ГенБанка.

Species	Voucher	Location	COI GB accession number	Reference
Flabellina pellucida	IEF57	Barents Sea, Russia, Zelenaya Inlet	MK533612	This study
Flabellina pellucida	IEFp10	Barents Sea, Russia, Zelenaya Inlet	MK533605	This study
Flabellina pellucida	IEFp9	Barents Sea, Russia, Teriberka Bay	MK533606	This study
Flabellina pellucida	IEFp8	Barents Sea, Russia, Teriberka Bay	MK533607	This study
Flabellina pellucida	IEFp3	Barents Sea, Russia, Teriberka Bay	MK533608	This study
Flabellina pellucida	IEFp2	Barents Sea, Russia, Teriberka Bay	MK533609	This study
Flabellina pellucida	IEIn3	Barents Sea, Russia, Ura Inlet	MK533610	This study
Flabellina pellucida	IEIn1	Barents Sea, Russia, Ura Inlet	MK533611	This study
Flabellina pellucida	ZMMU:Op-514	North Sea, Norway, Gulen	MF523349	Korshunova *et al.*, 2017
Flabellina pellucida	ZMMU:Op-513	North Sea, Norway, Gulen	MF523350	Korshunova *et al.*, 2017
Flabellina pellucida	NTNU-VM:68824	North Sea, Norway, Gulen	MG452617	Korshunova *et al.*, 2017
Flabellina pellucida	NTNU-VM:68823	North Sea, Norway, Gulen	MG452618	Korshunova *et al.*, 2017
Goniodoris nodosa	IEGn1	Barents Sea, Russia, Teriberka Bay	MK533614	This study
Goniodoris nodosa	IEGn4	Barents Sea, Russia, Teriberka Bay	MK533613	This study
Goniodoris nodosa	unknown	NE Atlantic, Spain	AF249788	Wollscheid-Lengeling *et al.*, 2001
Goniodoris nodosa	MT09684	North Sea	KR084683	Barco *et al.*, 2016

PALL Corp. [Ivanova *et al.*, 2006]. Extracted DNA was used as a template for amplification of partial cytochrome *c* oxidase subunit I (COI), using standard primers: HCO2198 (5'-TAAACTTCAGGGTGACCAAAAAATCA-3') and LCO1498 (5'-GGTCAACAAATCATAAAGATATTGG-3') [Folmer *et al.*, 1994]. Polymerase chain reactions were carried out in a 25-µL reaction volume, which included 5 µL of 5x Taq Red Buffer by *Eurogen Lab*, 0.5 µL of HS-Taq Polymerase by *Eurogen Lab,* 0.5 µL of dNTP (50 µM stock), 0.3 µL of each primer (10 µM stock), 1 µL of genomic DNA and 17.7 µL of sterile water. The amplification was performed with an initial denaturation for 1 min at 95°C followed by 35 cycles of 15 s at 95°C (denaturation), 30 s at 45°C (annealing) and 45 s at 72°C (elongation) with a final extension of 7 min at 72°C. Sequencing for both strands proceeded with the *Big Dye Terminator v3.1* sequencing kit by Applied Biosystems, the same primers as for PCR were used. Sequencing reactions were analyzed using ABI 3500 Genetic Analyser (*Applied Biosystems*) at N.K. Koltsov Institute of Developmental Biology (Moscow, Russia). All new sequences were deposited in GenBank (Table 1).

DNA barcoding, species delimitation and population analysis

Raw reads for each gene were assembled and checked for improper base-calling using Geneious-Pro 4.8.5 (Biomatters, Auckland, New Zealand). Assembled sequences were identified using the BLAST-n program on the GenBank website (http://www.ncbi.nlm.nih.gov). Original data and publicly available sequences of each species were aligned with the MUSCLE [Edgar, 2004] algorithm in MEGA7 [Kumar *et al.*, 2016] software. Sequences were translated into amino acids to verify authenticity of coding sequences. The resulting alignments were of 621 bp in the case of *F. pellucida* and of 645 bp in the case of *G. nodosa*. Haplotype networks based on four COI sequences of *G. nodosa* and 12 COI sequences of *F. pellucida* were reconstructed using PopArt software [Leigh, Bryant, 2015] with the TCS network method [Clement *et al.*, 2002].

Results

Suborder Doridina
Family Goniodorididae H. Adams et A. Adams, 1854

Goniodoris Forbes et Goodsir, 1839
Goniodoris nodosa (Montagu, 1808)
(Figs 1A, 2, 4A)

Material examined: Barents Sea, Teriberka Bay, 69°11.298'N, 35°11.072'E, 10 m, on a vertical rock wall with bryozoans, 25.08.2018, coll. T.I. Antokhina (1 spm); Barents Sea, Teriberka Bay, 69°10.444'N, 35°07.805'E, 18 m, on a rock, 30.08.2018, coll. T.I. Antokhina (1 spm).

Description (Fig. 2). Size of adults up to 25 mm. Body elongate, oval with sparse wrinkles and lacking tubercles. Foot broad. Rhinophores perfoliated with 12-14 lamellae. Gills with bi- and tripinnate brachial leaves arranged in circle, surrounding anus. Notal edge forms distinct rim. Background color milky-white, pale beige or light pink. Numerous white dots on notum, upper parts of foot, rhinophores and oral tentacles. Radular formula: 18-21 x 1.1.0.1.1. Inner lateral teeth with wide base and elongate acute cusp. Masticatory border of cusp with 16-25 sharp denticles. Inner part of tooth base bears outstanding bulbous leaf-like process, processes of right and left inner laterals form central row. Outer lateral teeth reduced plate-like with short cusp along outer edge. Reproductive system triaulic. Ampulla large sausage-shaped, vas deferens very long with prominent prostatic part. Seminal receptaculum elongate sausage-shaped, bursa copulatrix small oval. Penis short and wide with muscular penial sheath.

Biology. Feeding upon encrusting bryozoans and ascidians. Spawning in the early spring or autumn [Thompson, Brown, 1984]. In the Barents Sea mature individuals were found in August.

Distribution. North-East Atlantic coast from the northwestern Spain to the Barents Sea [Thompson, Brown, 1984; Eversten, Bakken, 2002, 2013; this study].

Genetic barcode. GenBank accession numbers MK533613, MK533614. A BLAST-n of these sequences resulted 99% identical to sequences of *G. nodosa* from North-East Atlantic (AJ223264, KR084683). TCS haplotype network (Fig. 4A) is linear with minimum possible number of bifurcations. Closest haplotypes are distinguished by a single nucleotide substitution. The most diverse haplotypes are separated by six mutation steps.

Suborder Cladobranchia
Family Flabellinidae Bergh, 1889

Flabellina McMurtrie, 1831
Flabellina pellucida
(Alder et Hancock, 1843)
(Figs 1B, 3, 4B)

Material examined: Barents Sea, Zelenaya Inlet, 69°13.244'N, 34°48.149'E, 24 m, 25.08.2015, coll. T.I. Antokhina (1 spm); Barents Sea, Zelenaya Inlet, 69°13.221'N, 34°48.015'E, 14 m, 27.08.2015, coll. T.I. Antokhina (1 spm); Barents Sea, Teriberka Bay, 69°11.298'N, 35°11.072'E, 10 m, on a vertical rock wall with bryozoans, 25.08.2018, coll. T.I. Antokhina (1 spm); Barents Sea, Teriberka Bay, 69°11.196'N, 35°09.873'E, 10 m, on a vertical rock, 26.08.2018, coll. T.I. Antokhina (1 spm); Barents Sea, Teriberka Bay, 69°12.767', 35°03.223'E, 20 m, on a rock block, 01.09.2018, coll. T.I. Antokhina (2 spm); Barents Sea, Ura Inlet, 69°22.663'N, 33°4.618'E, 2-8 m, 18.11.2018, coll. Yu.A. Zuev (2 spm).

Description (Fig. 3). Size of adults up to 30 mm. Body moderately wide, foot moderately wide with long anterior corners. Rhinophores 1.2-1.5 times longer than oral tentacles. Cerata arranged in distinct groups, up to eight groups per row. Cerata cylindrical or finger-shaped, pointed distally. Digestive gland diverticula cylindrical, fills about ½ of ceratal volume, rarely more. Slightly pronounced notal edge under ceratal groups. Anus pleuroproctic, reproductive openings lateral, below first group of cerata. Background color translucent-white. Digestive gland diverticula in cerata from bright-orange to pale or deep reddish. Cnidosacs bright-white, covered with white opaque pigment at their base. Sometimes numerous white pigment dots present under cnidosac area. Rhinophores and oral tentacles covered by white dots distally. Masticatory process with single row of distinct denticles.

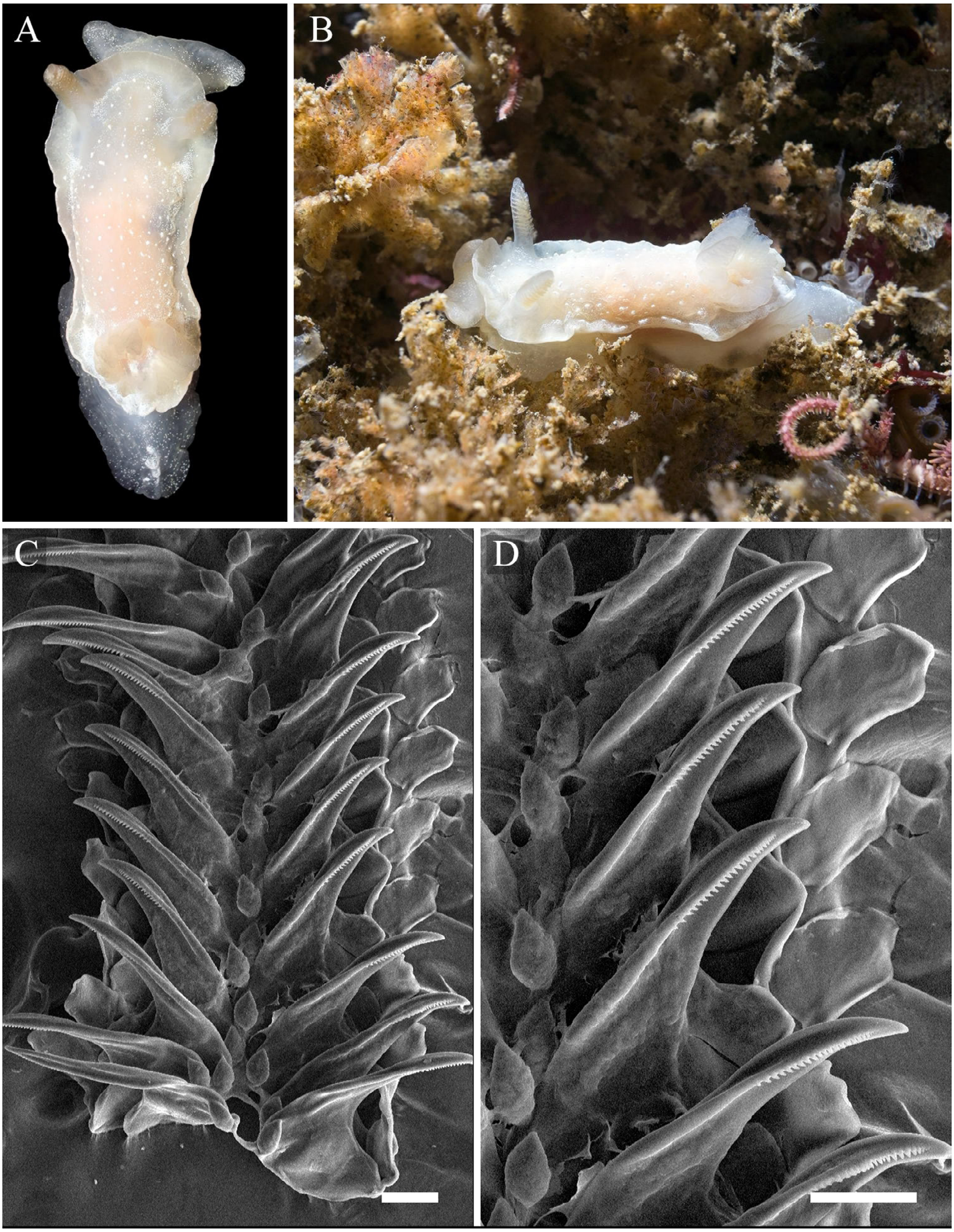

FIG. 2. *Goniodoris nodosa* from the Barents Sea. **A**. Living specimen, 37 mm length. **B**. Living specimen in natural environment. **C**. Anterior radular portion. **D**. Details of lateral and marginal teeth. Scale bars 100 µm.

РИС. 2. *Goniodoris nodosa* в Баренцевом море. **A.** Прижизненная фотография, особь 37 мм длиной. **B**. Прижизненная фотография в естественных условиях. **C**. Передняя часть радулы. **D**. Морфология латеральных и маргинальных зубов. Масштабные линейки 100 µm.

Radula triserial. Rachidian tooth elongated-triangular, bearing from seven to ten large denticles. Central cusp prominent and longer than denticles. Lateral teeth triangular with attenuated outer process and long cusp, lacking denticles. Reproductive system diaulic. Ampulla large, convoluted; vas deference long, lacking distinct prostate; two seminal receptaculum present; penis small, conical.

Biology. Feeding on various hydrozoans from genera *Tubularia* Linnaeus, 1758 and *Eudendrium* Ehrenberg, 1834. Spawning in the spring [Kuzirian, 1979; Thompson, Brown, 1984]. In the Barents Sea only mature individuals were found.

Distribution. North-West Atlantic (from Massachusetts to North Canada), Mediterranean coast, North-East Atlantic (British Isles, Norway and Faeroes), and the Barents Sea [Kuzirian, 1979; Thompson, Brown, 1984; Eversten, Bakken, 2005; present study].

Genetic barcode (Fig. 4B). GenBank accession numbers MK533605-MK533612. A BLAST-n of these sequences resulted 99-100% identical to available COI sequences of *F. pellucida* (MF523349-50; MG452617-18). TCS haplotype network (Fig. 4B) resulted in 9 haplotypes separated by 1-3 substitutions. Among them, five haplotypes are unique for the Barents Sea, two - for Norway and two haplotypes are shared for both studied localities. Network is well structured with no apparent alternative links.

Discussion

Contemporary studies of the Russian Arctic tend to lend heavily on traditional morphology with limited regard for newer methods and trends in taxonomy and biogeography. Identification approaches based on outdated initial species descriptions are often used without any modifications or updates. This isolation leads to discussion mal-synchronization with approaches used in other regions. In many cases, concepts are used in a slightly different sense, *e.g.* events that would not be considered as invasions in modern worldwide scientific community are reported as such.

We have shown that studied specimens truly belong to species *G. nodosa* and *F. pellucida,* despite minor genetic divergence between their European and Barents Sea populations. Morphologically both species are similar to their European relatives [Thompson, Brown, 1984]. These two species of nudibranch molluscs are new for the Russian fauna. Heterobranch molluscs in the Russian waters have been extensively studied for last 20 years, resulting in numerous new findings: *Melanochlamys diomedea* (Bergh, 1894) [Chaban, Martynov, 1998], *Aplysia parvula* Mörch, 1863 and *A. juliana* Quoy et Gaimard, 1832 [Martynov, Chaban, 1998], *Eubranchus pallidus* (Alder et Hancock, 1842) [Roginskaya, 1998], *Polycera quadrilineata* (O.F. Müller, 1776)*, Doto fragilis* (Forbes, 1838) and *Eubranchus tricolor* Forbes, 1838 [Martynov *et al.*, 2006], *Dendronotus orientalis* (Baba, 1932) and *Apata pricei* (MacFarland, 1966) [Martynov *et al.*, 2015]; *Dendronotus albus* MacFarland, 1966 [Korshunova *et al.*, 2016; Ekimova *et al.*, 2016], *Tritonia newfoundlandica* Valdés *et al.*, 2017 [Zakharov, Joergensen, 2017], *etc*. Also a series of comprehensive taxonomical revisions of nudibranch molluscs were carried [Martynov, Schrödl, 2009; Martynov *et al.*, 2009; Cella *et al.*, 2016; Ekimova *et al.*, 2015; Martynov, Korshunova, 2015, 2017; Korshunova *et al.*, 2016; 2017a,b,c; 2018]. Despite continuous and dedicated studies of the Russian fauna by experienced malacologists, new findings of two large and conspicuous species in one of the most studied Russian sea suggest vacancy of our knowledge regarding heterobranch fauna. At least *F. pellucida* demonstrates a stable population in the Barents Sea, being found during two seasons in three different localities. Further complications arise from deficiency of a regional biodiversity and barcoding research programs. While different regions of North Atlantic are thoroughly studied (*e.g.* North Sea – Raupach *et al.* [2015], Barco *et al.* [2016]; Canada – Radulovici *et al.* [2009], Layton *et al.* [2014]) only several parts of Russian Arctic seas and limited taxa went under such scrutiny.

Another explanation of new findings of invertebrates in the Barents Sea is a result of most recent invasions or gradual area expansion, both promoted by Climate Change. Most of these “invasions” were indicated among gastropod molluscs [Martynov *et al.*, 2006; Kantor *et al.*, 2008; Zakharov, Jorgensen, 2017], but also shown for cephalopods [Golikov *et al.*, 2013], bivalves [Deart *et al.*, 2013] and annelids [Rzhavsky *et al.*, 2011]. However, in all cases no valid proves of this concept were presented, and link to climate is quite speculative. Some authors proposed different explanations for similar findings, which were based only on fragmentary studied biology of species and temperature fluctuations in studied area [Martynov *et al.*, 2006; Golikov *et al.*, 2013]. In recent papers by Nekhaev [2011, 2013, 2016] this discontinuity was discussed with emphasis on alternative hypotheses that could possibly explain observed data. Nekhaev [2016] illustrated that new findings may be explained not by real fauna area expansions, but by researcher’s biases rising from misguided sense of faunistic study completeness. In all priory published cases the fact of invasion is based on absence of previous records in studied area and combined with data on temperature fluctuations in the Barents Sea during last 20 years. Thereby, possible natural processes as local extinctions or population dynamics are often ignored. In addition, in most papers no molec-

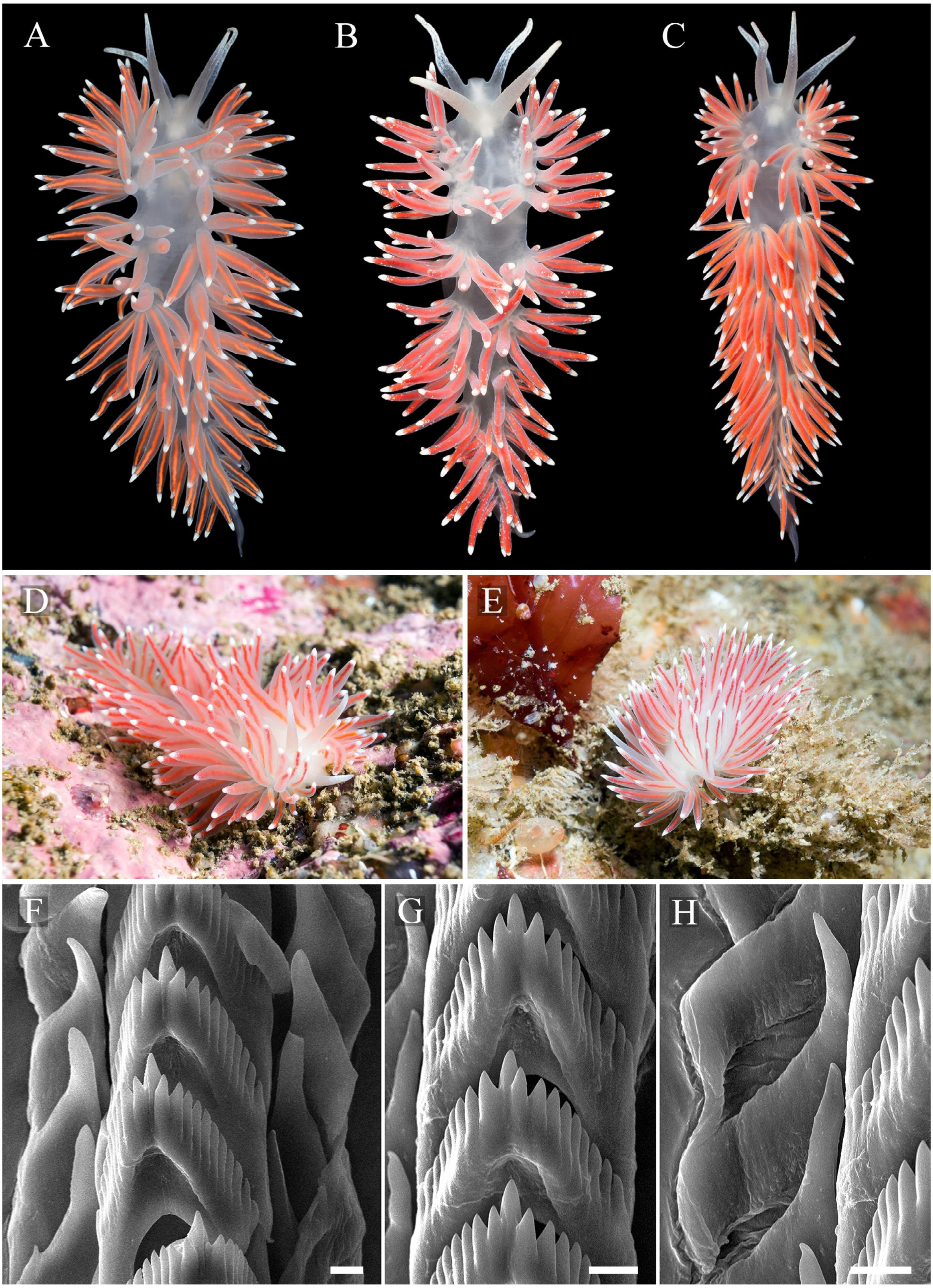

FIG. 3. *Flabellina pellucida* from the Barents Sea. **A**. Living specimen, 23 mm length. **B**. Living specimen, 32 mm length; **C.** Living specimen, 39 mm length. **D**, **E**. Living specimen in natural environment. **F**. Anterior radular portion. **G**. Details of rachidian teeth. **H**. Derails of lateral teeth. Scale bars 10 µm.

РИС. 3. *Flabellina pellucida* в Баренцевом море. **A.** Прижизненная фотография, особь 23 мм длиной. **B**. Прижизненная фотография, особь 32 мм длиной. **C**. Прижизненная фотография, особь 29 мм длиной. **D**, **E**. Прижизненные фотографии в естественных условиях. **F**. Передняя часть радулы. **G**. Морфология центральных зубов. **H**. Морфология латеральных зубов. Масштабные линейки 10 µm.

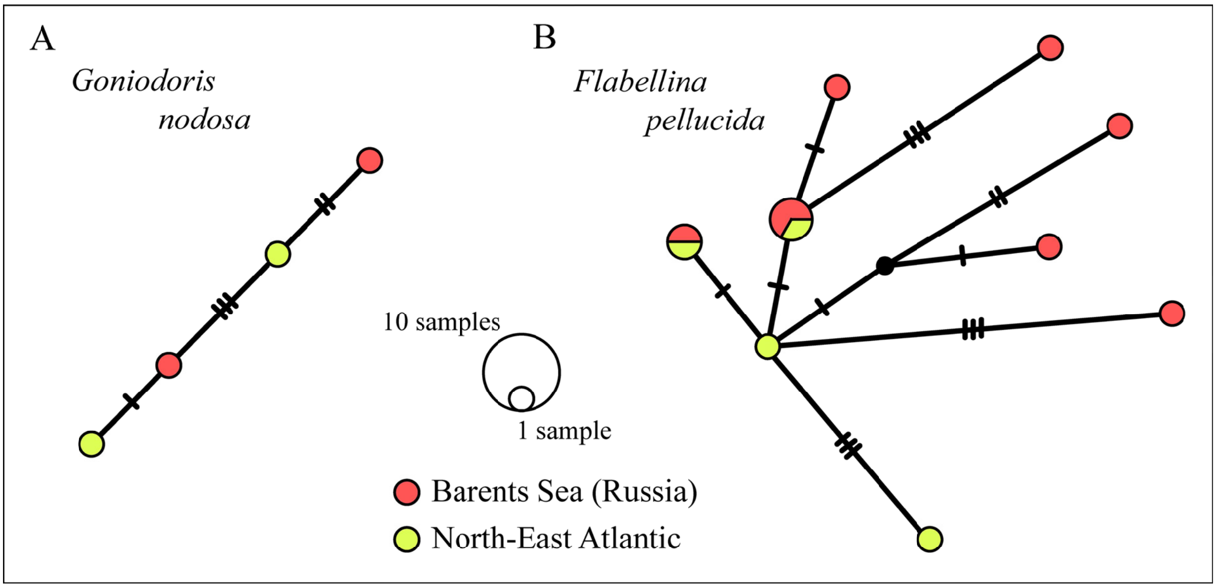

FIG. 4. COI haplotype networks of *Goniodoris nodosa* and *Flabellina pellucida* produced with TCS method in PopART. **A.** *Goniodoris nodosa.* **B.** *Flabellina pellucida.* Geographic region of each haplotype was found is coded by color. The relative size of circles is proportional to the number of sequences of that same haplotype.

РИС. 4. Сеть гаплотипов *Goniodoris nodosa* и *Flabellina pellucida*, реконструированная по фрагменту гена COI при помощи метода TCS в программе PopART. **A.** *Goniodoris nodosa.* **B.** *Flabellina pellucida.* Географическая приуроченность каждого гаплотипа отмечена цветом. Относительный размер кругов пропорционален количеству последовательностей, относящихся к одному гаплотипу.

ular data linking new findings to putative ancestral populations is available.

Our preliminary molecular results on *F. pellucida* population structure indicate that most likely Barents Sea populations were not formed in a single recent invasion event as they demonstrate the higher diversity than Norway populations (Fig. 4B). Unique Barents Sea haplotypes are unrelated to haplotypes found in both sampling locations, thus excluding possibility of a single bottleneck event. In the case of *G. nodosa,* limited molecular data unable us to study its population structure, however they also demonstrate similar pattern having 1-3 substitutions between observed haplotypes with no specimens having identical sequences (Fig 4A). Limited sample size in both cases does not allow us to distinguish between possible dispersal scenarios, *i.e.* historic migration or recent gradual area expansion. Further application of population genetics methods should bring light upon dispersal scenarios leading to contemporary situation. In the case of the actual recent invasion, we should expect to find few haplotypes common in ancestral population as seen in Rius *et al.* [2015]. If it is not an invasion, but a gradual area expansion (even most recent one) we would observe Barents Sea fauna as a part of neighbor populations with a possible minor drop in haplotypic richness [Cherneva *et al*., 2018]. In addition, not all bottleneck events presenting similar population structure can be attributed to the latest species dispersal. Glacial driven invasions, combined with relatively low mutation rates will give similar diversity [Maggs *et al*., 2008; Cherneva *et al*., 2018]. In other cases, we should expect to see a defined population structure independent in different localities or a panmixture [Genelt-Yanovskiy *et al.,* 2019].

Acknowledgements

We are grateful to Ivan Nekhaev, who provided several specimens for this study and to Yuri Zuev for help with sampling during field trip. We are thankful to Alexander Gulimchuk and Igor Konovalov for organizing dives and their assistance in Teriberka Bay. Valentina Tambovceva is thanked for assistance with Sanger sequencing. Two anonymous reviewers are owed special thanks for constructive criticism that improved the manuscript. We also want to acknowledge the staff of the Cooperative Far Eastern Center of Electron Microscopy and for providing SEM facilities. Molecular study was conducted using equipment of the Core Centrum of Institute of Developmental Biology RAS. This study was supported by Russian Science Foundation grant no. 18-74-00062 to IE (morphological and molecular studies). The collection of material in Ura Inlet was supported by Grant Council of the President of Russia (grant No. MK-4797.2018.4).

References

Barco A., Raupach M.J., Laakmann S., Neumann H., Knebelsberger T. 2016. Identification of North Sea molluscs with DNA barcoding. *Molecular Ecology Resources*, 16(1): 288–297.

Cella K., Carmona L., Ekimova I., Chichvarkhin A., Schepetov D., Gosliner, T.M. 2016. A radical solution: the phylogeny of the nudibranch family Fionidae. *PloS One*, 11(12): e0167800.

Chaban E.M., Martynov A.V. 1998. *Melanochlamys diomedea* (Bergh, 1893) (Opisthobranchia: Aglajidae), a new genus and species in the fauna of Russia. *Ruthenica, Russian Malacological Journal*, 8(2): 147–152 [In Russian].

Chan F.T., Stanislawczyk K., Sneekes A.C., Dvoretsky A., Gollasch S., Minchin D., David M., Jelmert A., Albretsen J., Bailey S.A. 2018. Climate change opens new frontiers for marine species in the Arctic: Current trends and future invasion risks. *Global Change Biology*, 25(1): 25–38. https://doi.org/10.1111/gcb.14469

Cherneva I.A., Chernyshev A.V., Ekimova I.A., Polyakova N.E., Schepetov D.M., Turanov S.V., Neretina T.V., Chaban E.M., Malakhov V.V. 2018. Species identity and genetic structure of nemerteans of the "*Lineus ruber-viridis*" complex (Muller, 1774) from Arctic waters. *Polar Biology*. https://doi.org/10.1007/s00300-018-2438-7

Clement M., Snell Q., Walke P., Posada D., Crandall K. 2002. TCS: estimating gene genealogies. *Proceedings 16th International Parallel and Distributed Processing Symposium*, 2: 184.

Deart Y.V., Frolov A.A., Manushin I.E. 2013. Bivalves *Abra prismatica* (Montagu, 1808) and *Gari fervensis* (Gmelin, 1791) – species new to the fauna of the Russian sector of the Barents Sea. *Russian Journal of Biological Invasions*, 4(3): 139–148.

Edgar R.C. 2004. MUSCLE: multiple sequence alignment with high accuracy and high throughput. *Nucleic Acids Research*, 32(5): 1792–1797.

Ekimova I., Korshunova T., Schepetov D., Neretina T., Sanamyan N., Martynov A. 2015. Integrative systematics of northern and Arctic nudibranchs of the genus *Dendronotus* (Mollusca, Gastropoda), with descriptions of three new species. *Zoological Journal of the Linnean Society*, 173(4): 841–886.

Ekimova I., Valdés Á., Schepetov D., Chichvarkhin A. 2016. Was Gordon Robilliard right? Integrative systematics suggest that *Dendronotus diversicolor* (multicolor frond-aeolis) is a valid species. *Canadian Journal of Zoology*, 94(11): 793–799.

Evertsen J., Bakken T. 2002. Heterobranchia (Mollusca, Gastropoda) from northern Norway, with notes on ecology and distribution. *Fauna Norvegica*, 22: 15–22.

Evertsen J., Bakken T. 2005. Nudibranch diversity (Heterobranchia, Gastropoda) along the coast of Norway. *Fauna Norvegica*, 25: 1–37.

Evertsen J., Bakken T. 2013. Diversity of Norwegian Sea slugs (Nudibranchia): new species to Norwegian coastal waters and new data on distribution of rare species. *Fauna Norvegica*, 32: 45–52.

Folmer O., Black M., Hoeh W., Lutz R., Vrijenhoek R. 1994. DNA primers for amplification of mitochondrial cytochrome c oxidase subunit I from diverse metazoan invertebrates. *Molecular Marine Biology and Biotechnology*, 3: 294–299.

Furfaro G., Salvi D., Mancini E., Mariottini P. 2018. A multilocus view on Mediterranean aeolid nudibranchs (Mollusca): Systematics and cryptic diversity of Flabellinidae and Piseinotecidae. *Molecular Phylogenetics and Evolution*, 118: 13–22.

Genelt-Yanovskiy E., Nazarova S., Tarasov O., Mikhailova N., Strelkov P. 2019. Phylogeography of the temperate marine bivalve *Cerastoderma edule* (Linnaeus, 1758) (Bivalvia: Cardiidae) in the Subarctic: Unique diversity and strong population structuring at different spatial scales. *Journal of Zoological Systematics and Evolutionary Research,* 57(1), 67–79. https://doi.org/10.1111/jzs.12231

Golikov A.V., Sabirov R.M., Lubin P.A., Jørgensen L.L. 2013. Changes in distribution and range structure of Arctic cephalopods due to climatic changes of the last decades. *Biodiversity*, 14(1): 28–35.

Golikov A.V., Sabirov R.M., Lubin P.A., Jørgensen L.L., Beck, I.M. 2014. The northernmost record of *Sepietta oweniana* (Cephalopoda: Sepiolidae) and comments on boreo-subtropical cephalopod species occurrence in the Arctic. *Marine Biodiversity Records*, 7: e58. https://doi.org/10.1017/S1755267214000645

Gosliner T.M., Behrens D.W., Valdés A. 2018. *Nudibranch & Sea Slug identification: Indo-Pacific.* 2nd Edition. New World Publications, 452 p.

Ivanova N.V., deWaard J., Hebert P.D.N. 2006. An inexpensive, automation-friendly protocol for recovering high-quality DNA. *Molecular Ecology Notes*, 6: 998–1002.

Kantor Yu.I., Rusyaev S.M., Antokhina T.I. 2008. Going eastward – climate changes evident from gastropod distribution in the Barents Sea. *Ruthenica, Russian Malacological Journal*, 18(2): 51–54.

Korshunova T., Sanamyan N., Zimina O., Fletcher K., Martynov A. 2016. Two new species and a remarkable record of the genus *Dendronotus* from the North Pacific and Arctic oceans (Nudibranchia). *ZooKeys*, 630: 19–42.

Korshunova T., Martynov A., Bakken T., Evertsen J., Fletcher K., Mudianta I.W., Saito H., Lundin K., Schrodl M., Picton B. 2017a. Polyphyly of the traditional family Flabellinidae affects a major group of Nudibranchia: aeolidacean taxonomic reassessment with descriptions of several new families, genera, and species (Mollusca, Gastropoda). *ZooKeys*, 717: 1–139.

Korshunova T., Martynov A., Picton B. 2017b. Ontogeny as an important part of integrative taxonomy in tergipedid aeolidaceans (Gastropoda: Nudibranchia) with a description of a new genus and species from the Barents Sea. *Zootaxa*, 4324(1): 1–22.

Korshunova T., Zimina O., Martynov A. 2017c. Unique pleuroproctic taxa of the nudibranch family Aeolidiidae from the Atlantic and Pacific Oceans, with description of a new genus and species. *Journal of Molluscan Studies*, 83(4): 409–421.

Korshunova T., Fletcher K., Lundin K., Picton B., Martynov A. 2018. The genus *Zelentia* is an amphiboreal taxon expanded to include three new species from the North Pacific and Atlantic oceans (Gastropoda: Nudibranchia: Trinchesiidae). *Zootaxa*, 4482(2): 297–321.

Kumar S., Stecher G., Tamura K. 2016. MEGA7: molecular evolutionary genetics analysis version 7.0 for bigger datasets. *Molecular Biology and Evolution*, 33(7): 1870–1874.

Kuzirian A.M. 1979. Taxonomy and biology of four New England coryphellid nudibranchs (Gastropoda: Opisthobranchia). *Journal of Molluscan Studies*, 45(3): 239–261.

Layton K.K., Martel A.L., Hebert P.D. 2014. Patterns of DNA barcode variation in Canadian marine molluscs. *PLoS One*, 9(4): e95003.

Leigh J.W., Bryant D. 2015. PopART: Full-feature software for haplotype network construction. *Methods in Ecology and Evolution*, 6(9): 1110–1116.

Lemche H., 1929. Gastropoda Opisthobranchiata. *Zoology of the Faroes*, 52: 1–35.

Leydet K.P., Hellberg M.E. 2015. The invasive coral *Oculina patagonica* has not been recently introduced to the Mediterranean from the western Atlantic. *BMC Evolutionary Biology*, 15(1): 79.

Maggs C.A., Castilho R., Foltz D., Henzler C., Jolly M.T., Kelly J., ... Viard F. 2008. Evaluating signatures of glacial refugia for North Atlantic benthic marine taxa. *Ecology*, 89(sp11): S108–S122.

Martynov A.V., Chaban E.M. 1998. *Aplysia parvula* Guilding in Mörch, 1863 and *A. juliana* Quoy et Gaimard, 1832 from the shoaling water of the Moneron Island is the first find of the representatives of the family Aplysiidae (Opisthobranchia, Anaspidea) in the Russian fauna. *Ruthenica, Russian Malacological Journal*, 8(1): 17–28.

Martynov A., Korshunova T. 2015. A new deep-sea genus of the family Polyceridae (Nudibranchia) possesses a gill cavity, with implications for the cryptobranch condition and a 'Periodic Table' approach to taxonomy. *Journal of Molluscan Studies*, 81(3): 365–379.

Martynov A., Korshunova T. 2017. World's northenmost and rarely observed Nudibranchs: three new Onchidoridid species (Gastropoda: Doridida) from Russian seas. *Zootaxa*, 4299(3): 391–404.

Martynov A.V., Korshunova T.A., Savinkin O.V. 2006. Shallow-water opistobranch molluscs of the Murman coast of the Barents Sea, with new distributional data and remarks on biology. *Ruthenica, Russian Malacological Journal*, 16(1-2): 59–72.

Martynov A.V., Korshunova T.A., Sanamyan N.P., Sanamyan K.E. 2009. Description of the first cryptobranch onchidoridid *Onchimira cavifera* gen. *et* sp. nov. and of three new species of the genera *Adalaria* and *Onchidoris* (Nudibranchia: Onchidorididae) from Kamchatka waters. *Zootaxa*, 2159: 1–43.

Martynov A.V., Sanamyan N.P., Korshunova T.A. 2015. New data on the opisthobranch molluscs (Gastropoda: Opisthobranchia) of waters of Commander Islands and Far-Eastern seas of Russia. In: *Conservation of biodiversity of Kamchatka and coastal waters. Proceedings of XV international scientific conference Petropavlovsk-Kamchatsky*. Kamchat Press, Petropavlovsk-Kamchatsky: 55–69.

Martynov A.V., Schrödl M. 2009. The new Arctic side-gilled sea slug genus *Boreoberthella* (Gastropoda, Opisthobranchia): Pleurobranchoidean systematics and evolution revisited. *Polar Biology*, 32(1): 53–70.

Nekhaev I.O. 2011. Two species of parasitic molluscs new for Russian seas. *Ruthenica, Russian Malacological Journal*, 21(1): 69–72.

Nekhaev I.O. 2013. The first record of *Alvania punctura* from Russian waters (Gastropoda: Rissoidae). *Marine Biodiversity Records*, 6: 1–3.

Nekhaev I.O. 2016. Newly arrived or previously overlooked: is there evidence for climate-driven changes in the distribution of molluscs in the Barents Sea? *Biodiversity and Conservation*, 25(5): 807–825.

Occhipinti-Ambrogi A., Savini D. 2003. Biological invasions as a component of global change in stressed marine ecosystems. *Marine Pollution Bulletin*, 46(5): 542–551.

Post E., Forchhammer M.C., Bret-Harte M.S., Callaghan T.V., Christensen T.R., Elberling B., Fox A.D., Gilg O., Hik D.S., Høye T.T., Ims R.A. 2009. Ecological dynamics across the Arctic associated with recent climate change. *Science*, 325(5946): 1355–1358.

Raupach M.J., Barco A., Steinke D., Beermann J., Laakmann S., Mohrbeck I., Neumann H., Kihara T.C., Pointner K., Radulovici A., Segelken-Voigt A., Wesse C., Knebelsberger T. 2015. The application of DNA barcodes for the identification of marine crustaceans from the North Sea and adjacent regions. *PloS One*, 10(9): e0139421.

Radulovici A.E., Sainte-Marie B., Dufresne F. 2009. DNA barcoding of marine crustaceans from the Estuary and Gulf of St Lawrence: a regional scale approach. *Molecular Ecology Resources*, 9: 181–187. https://doi.org/10.1111/j.1755-0998.2009.02643.x

Rius M., Turon X., Bernardi G., Volckaert F.A., Viard F. 2015. Marine invasion genetics: from spatio-temporal patterns to evolutionary outcomes. *Biological Invasions*, 17(3): 869–885.

Roginskaya I.S. 1998. The records of *Eubranchus pallidus* (Alder et Hancock, 1842) (Nudibranchia, Eubranchidae) from Aynovskye Islands (West Murman) (Barents Sea, Kandalaksha State Preserve). *Opisthobranch Newsletter*, 24(9): 39.

Ruiz G.M., Fofonoff P.W., Carlton J.T., Wonham M.J., Hines A.H. 2000. Invasion of coastal marine communities in North America: apparent patterns, processes, and biases. *Annual Review of Ecology and Systematics*, 31(1): 481–531.

Rzhavsky A.V., Deart Y.V., Britayev T.A. 2011. Sublittoral hard-bottom communities of Kola Peninsula Inlets (Barents Sea) - current state and long-term changes. Travizi A. *et al.* (eds.) Abstracts of 46-th European Marine Biology Symposium, Rovinj, Croatia, September 12-16, 2011: 51.

Thompson T.E., Brown G.H. 1984. *Biology of opisthobranch molluscs.* Vol. 2. Ray Society, London, 229 p.

Wassmann P., Duarte C.M., Agusti S., Sejr M.K. 2011. Footprints of climate change in the Arctic marine ecosystem. *Global Change Biology*, 17(2): 1235–1249.

Wollscheid-Lengeling E., Boore J., Brown W., Wagele H. 2001. The phylogeny of Nudibranchia (Opisthobranchia, Gastropoda, Mollusca) reconstructed by three molecular markers. *Organisms Diversity & Evolution*, 1 (4): 241–256.

Zakharov D.V., Anisimova N.A., Stepanenko A.M. 2016. First Record of the Sea Star *Porania pulvillus* (O.F. Muller, 1776) in Russian Part of the Arctic. *Russian Journal of Biological Invasions*, 7(4): 321–323 [In Russian].

Zakharov D.V., Jorgensen L.L. 2017. New species of the gastropods in the Barents Sea and adjacent waters. *Russian Journal of Biological Invasions*, 10(2): 38–45 [In Russian].

●

“Виды-вселенцы” Российской Арктики: является ли глобальное потепление реальной причиной? Примечательная находка двух видов голожаберных моллюсков

Ирина А. ЕКИМОВА[1,5], Татьяна И. АНТОХИНА[2], Дмитрий М. ЩЕПЕТОВ[1,3,4]

[1]*Московский государственный университет им. М.В. Ломоносова, Ленинские горы 1-12, 119234 Москва, РОССИЯ;*

[2] *Институт проблем экологии и эволюции им. А.Н. Северцова РАН, 119071 Москва, Ленинский проспект, 33, РОССИЯ;*

[3]*Институт биологии развития им. Н.К. Кольцова РАН, ул. Вавилова 26, 119334 Москва, РОССИЯ;*

[4]*Национальный исследовательский университет Высшая школа экономии, ул. Мясницкая 20, 101000 Москва, РОССИЯ*

[5]*Автор-корреспондент; E-mail: irenekimova@gmail.com*

РЕЗЮМЕ. Два крупных вида голожаберных моллюсков – *Goniodoris nodosa* и *Flabellina pellucida* – впервые отмечены для фауны России. Оба вида широко распространены в Северо-Восточной Атлантике, а вид *F. pellucida* также зарегистрирован в водах Северо-Западной Атлантики. Были проведены анатомические исследования представителей обоих видов, а также исследования с помощью сканирующего электронного микроскопа. Молекулярные различия были оценены при помощи секвенирования митохондирального маркера COI и реконструкции сетей гаплотипов. Баренцевоморские популяции обоих видов демонстрируют небольшие генетические отличия от европейских популяций, однако идентичны морфологически. Обсуждены возможные причины новых находок беспозвоночных в водах Российской Арктики.