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New records of moths (Lepidoptera) from Novaya Zemlya, Arctic Russia, with a supplement of DNA barcoding data

Новые находки чешуекрылых (Lepidoptera) с архипелага Новая Земля с данными ДНК-баркодинга

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КЛЮЧЕВЫЕ СЛОВА: биогеография, арктические острова, Баренцево море, Crambidae, Geometridae.

ABSTRACT. This paper reports the first records of four moth species from Novaya Zemlya: *Eupithecia gelidata*, *Rheumaptera subhastata*, *Entephria byssata* (Geometridae), and *Udea* cf. *cacuminicola* (Crambidae). The *COI* barcode data is provided for each species. Molecular sequences suggest that the Lepidoptera fauna of Novaya Zemlya was originated recently via long-distance dispersal of widespread lineages from the mainland.

РЕЗЮМЕ. Приводятся первые находки четырех видов чешуекрылых на архипелаге Новая Земля: *Eupithecia gelidata*, *Rheumaptera subhastata*, *Entephria byssata* (Geometridae) и *Udea* cf. *cacuminicola* (Crambidae). Для каждого вида приводятся последовательности нуклеотидов гена *COI*, на основе которых можно предположить, что фауна чешуекрылых архипелага возникла недавно.

Introduction

Novaya Zemlya is one of the most inaccessible archipelagoes of the High Arctic [Potapov et al., 2018]. As a result, its fauna remains poorly known. The Lepidoptera fauna of Novaya Zemlya includes 30 species [Jacobson, 1898; Rebel, 1923; Karsholt et al., 2013; Kullberg et al., 2018], while the actual species richness seems to be at least twice this value. For example, only two geometer species (Geometridae) are known from Novaya Zemlya [Kullberg et al., 2018], whereas 4–5 species are recorded from neighboring areas, i.e. Yugorsky Peninsula and Kolguev Island [Kullberg et al., 2013, 2018].

In this paper, we present the first records of four moth species from Novaya Zemlya and discuss the putative origin of this insular fauna by means of a molecular approach.

Material and Methods

Moths were collected with an entomological net near the Bezymyannaya Bay (Novaya Zemlya, Yuzhny Island) from July 19 to July 26, 2017. Specimens were prepared using standard methods [Schauff, 2001]. They are deposited in the Russian Museum of Biodiversity Hotspots (RMBH), N. Laverov Federal Center for Integrated Arctic Research of the Ural Branch of the Russian Academy of Sciences (Arkhangelsk, Russia).

Total DNA was extracted from a single leg of each dry specimen according to standard phenol/chloroform procedures [Sambrook et al., 1989]. The mitochondrial *cytochrome c oxidase subunit I (COI)* gene was amplified and sequenced using primers LCO1490 [Folmer et al., 1994] and LepR [Hajibabaei et al., 2006]. The PCR mix contained approximately 200 ng of total cell DNA, 10 pmol of each primer, 200 ìmol of each dNTP, 2.5 ìl of PCR buffer (with 20 mmol $MgCl_2$), 0.8 units Taq DNA polymerase (SibEnzyme Ltd., Russia), and H_2O was added for a final volume of 25 ìl. Temperature cycling

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was as follows: 95°C (5 min), 30–33 cycles of 95°C (50 sec), 48°C (50 sec), 72°C (50 sec) and a final extension at 72°C (5 min). The sequencing was carried out at the facilities of the Inter-Institution Center of Group Use (Genom) (Engelhardt Institute of Molecular Biology of the Russian Academy of Sciences, Moscow) using the ABI PRISM® BigDye™ Terminator v. 3.1 reagents kit. Reaction products were analyzed using an automatic sequencer ABI PRISM® 3730 (Applied Biosystems).

We obtained new *COI* sequences from 12 specimens (Table 1). The resulting sequences were checked manually using a sequence alignment editor BioEdit v. 7.2.5 [Hall, 1999].

Results

Here we present the first records of four species of Lepidoptera for the Novaya Zemlya Archipelago with a supplement of the DNA barcoding data. In summary, 34 Lepidoptera species are recorded from Novaya Zemlya [Kullberg et al., 2018; this study].

Crambidae

Udea cf. *cacuminicola* Munroe, 1966
Fig. 1.

MATERIAL EXAMINED. Russia, Novaya Zemlya, Yuznhy Island, near the Bezymyannaya Bay, meadow-like association with *Artemisia tilesii* and *Salix lanata*, 72°50′42″N, 53°44′21″E, 19–26.XII.2017, Spitsyn leg. — 2 ex.

DNA BARCODING. A large number of species of *Udea itysalis* sp. group refers to lineages that have a genetic distance of less than 2% for *COI*, while the part of taxa is paraphyletic. It is likely that most of the currently accepted nominal taxa would represent synonyms. The specimens from Novaya Zemlya belong to *U. cacuminicola* Munroe, 1966 having a rather shallow genetic divergence from other taxa in the species group. The closest sequences from Canada (Yukon) and the USA (Colorado) differ by 0.22–0.89%, the sequence of "*Udea costalis*" (incorrect identification) from Russia (Primorsky Krai) differs by 0.45%. From *Udea itysalis* on Kolguev Island (Nenets Autonomous District, Russia), it differs by 1.57%.

Geometridae

Eupithecia gelidata Möschler, 1860
Fig. 2.

MATERIAL EXAMINED. Russia, Novaya Zemlya, Yuznhy Island, near the Bezymyannaya Bay, 72°49′09″N, 53°47′33″E, 21.XII.2017, Spitsyn leg. — 1 ex.

DNA BARCODING. We sequenced a specimen from Novaya Zemlya having a widespread haplotype that is known from Greenland to Norway and Italy.

Rheumaptera subhastata (Nolcken, 1870)
Fig. 3.

MATERIAL EXAMINED. Russia, Novaya Zemlya, Yuznhy Island, near the Bezymyannaya Bay, 72°49′09″N, 53°47′33″E, 21.XII.2017, Spitsyn leg. — 1 ex.

DNA BARCODING. A sequence from Novaya Zemlya differs by one substitution from the *COI* haplotype common in Canada and by 2–3 substitutions from the series of the nearest haplotypes from Norway, Finland, Sweden, Austria, Germany, Canada, and the USA.

Entephria byssata (Aurivillius, 1891)
Fig. 4.

MATERIAL EXAMINED. Russia, Novaya Zemlya, Yuznhy Island, near the Bezymyannaya Bay, meadow-like association with *Artemisia tilesii* and *Salix lanata*, 72°50′42″N, 53°44′21″E, 19-26.XII.2017, Spitsyn leg. — 6 ex; tundra with *Astragalus alpinus*, 72°51′10″N, 53°42′48″E, 19–26.XII.2017, Spitsyn leg. — 6 ex; tundra with *Hedysarum arcticum*, 72°52′24″N, 53°38′48″E, 19–21.XII.2017, Spitsyn leg. — 15 ex; tundra with *Astragalus alpinus*, 72°48′43″N, 53°50′28″E, 23.XII.2017, Spitsyn leg. — 3 ex; tundra with *Hedysarum arcticum*, 72°52′41″N, 53°37′49″E, 23.XII.2017, Spitsyn leg. — 7 ex; associations with *Astragalus alpinus* on the

Table 1. List of new *COI* sequences for moths obtained under this study.
Таблица 1. Список новых последовательностей *COI*, полученных в ходе настоящего исследования.

Species	NCBI's GenBank acc. no.	Voucher no.	Specimen locality
Udea cf. *cacuminicola*	MN700905	Sph 705	Russia: Novaya Zemlya
Udea cf. *cacuminicola*	MN700906	Sph 706	Russia: Novaya Zemlya
Eupithecia gelidata	MN700907	Sph 707	Russia: Novaya Zemlya
Rheumaptera subhastata	MN700908	Sph 708	Russia: Novaya Zemlya
Entephria byssata	MN700909	Sph 709	Russia: Novaya Zemlya
E. byssata	MN700910	Sph 710	Russia: Novaya Zemlya
E. byssata	MN700911	Sph 711	Russia: Novaya Zemlya
E. byssata	MN700912	Sph 712	Russia: Novaya Zemlya
E. byssata	MN700913	Sph 713	Russia: Novaya Zemlya
E. byssata	MN700914	Sph 714	Russia: Novaya Zemlya
E. byssata	MN700915	Sph 715	Russia: Novaya Zemlya
E. byssata	MN700916	Sph 716	Russia: Novaya Zemlya

canyon slope, 72°50′15″N, 53°22′41″E, 23.XII.2017, Spitsyn leg. — 1 ex; tundra with *Hedysarum arcticum*, 72°48′36″N, 53°50′24″E, 23.XII.2017, Spitsyn leg. — 1 ex.

DNA BARCODING. We identified three haplotypes of this species on Novaya Zemlya, two of which are found in Scandinavia.

REMARK. Previously, this species (as *Entephria punctipes*) was listed in the fauna of the Novaya Zemlya Archipelago based on an incorrectly identified specimen of *Psychophora sabini* [Sumakow, 1912], and it was subsequently excluded from the list of the Lepidoptera species of Novaya Zemlya [Kullberg et al., 2018]. *Entephria punctipes* is considered as the Nearctic taxon at the present time [Aarvik et al., 2017]. Here, we present the first record of *Entephria byssata* from Novaya Zemlya that was confirmed by morphological characters and DNA barcoding.

Discussion

We can conclude that the studied species of Lepidoptera immigrated to Novaya Zemlya after the Last Glacial Maximum. Most of the species share widespread haplotypes or singletons. Similar biogeographic patterns are common for a number of other Lepidoptera in the Northern Palearctic [Bolotov et al., 2015; Spitsyn et al., 2020]. Studies of other animal groups of the fauna of Novaya Zemlya revealed that a number of taxa have endemic haplotypes and divergent lineages from this archipelago [Potapov et al., 2018; Makhrov et al., 2019]. Novaya Zemlya was suggested as a putative refugium for these groups [Potapov et al., 2018; Makhrov et al., 2019]. However, other taxa are represented by widespread haplotypes, e.g. reindeer [Kvie et al., 2016] and moths [this study].

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Competing interests. The authors declare no competing interests.

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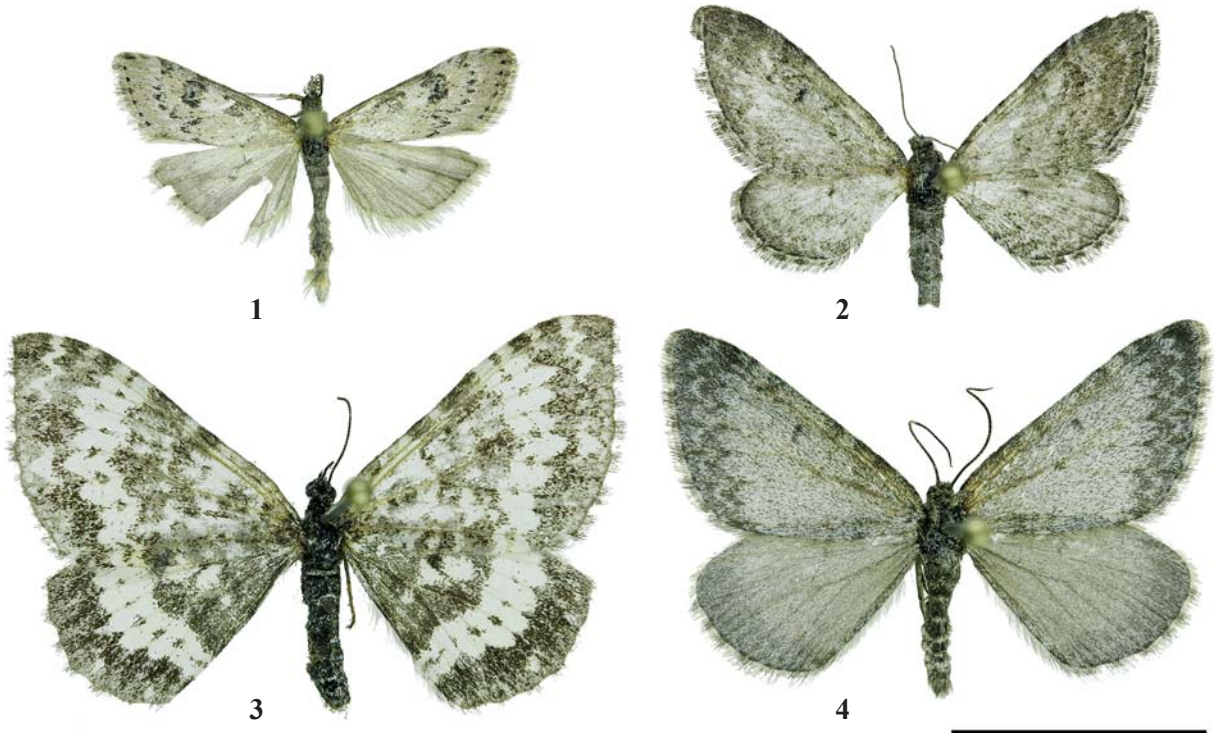

Figs 1–4. Moth species from Novaya Zemlya: 1 — *Udea* cf. *cacuminicola* Munroe, 1966; 2 — *Eupithecia gelidata* Möschler, 1860; 3 — *Rheumaptera subhastata* (Nolcken, 1870); 4 — *Entephria byssata* (Aurivillius, 1891). Scale bar = 10 mm. (Photos: Vitaly M. Spitsyn).

Рис. 1–4. Чешуекрылые архипелага Новая Земля: 1 — *Udea* cf. *cacuminicola* Munroe, 1966; 2 — *Eupithecia gelidata* Möschler, 1860; 3 — *Rheumaptera subhastata* (Nolcken, 1870); 4 — *Entephria byssata* (Aurivillius, 1891). Масштабная линейка = 10 мм. (Фото: В.М. Спицын).

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