Algae and cyanobacteria in soils polluted with heavy metals (Northwest Russia, Murmansk region)

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Abstract

The analysis of algae and cyanobacteria in Albic Podzols affected by emissions of the copper–nickel plant Pechenganikel in the forest-tundra of the Arctic region was carried out. The main pollutants contained in the emissions and entering the soil are sulfur compounds and heavy non-ferrous metals. Algae and cyanobacteria were identified in soil samples collected in three zones differing in their distance from the pollution source: the zone of strong pollution (at a distance of 3 km southwest from the source of the emission), the zone of medium pollution (5-7 km), and the zone of weak pollution (16-25 km). In total, 61 species of eukaryotic algae and 2 species of cyanobacteria were found. In the studied soils, several species of algae were found with a high frequency, apparently resistant to unfavorable natural and anthropogenic factors: *Chloromonas* sp., *Neocystis brevis*, *Parietochloris alveolaris*, *Pseudococcomyxa simplex*, *Stichococcus bacillaris*, *Interfilum terricola*, *Leptosira* cf. *obovata*, *Myrmecia bisecta*, *Nostoc muscorum*. Algae from the Chlorophyta division predominated in all soils studied. Yellow-green algae and diatoms were found only in the zone of strong pollution and were represented by a very small number of species. The presence of *Microthamnion kuetzingianum*, which is resistant to high acidity and heavy metals concentration, can be useful as an indicator of severe heavy metal contamination. Our study confirmed sensitivity of *Vischeria magna* to soil contamination with heavy metals. In long-term aspect, the species diversity of algae has increased by 35% in the soils influenced by the Pechenganikel plant in comparison with the data obtained 30 years ago, which probably indicates a certain decrease in anthropogenic load on the adjacent territories.

Key words: Arctic region, Kola Peninsula, copper–nickel plant Pechenganikel, soil pollution, biodiversity, microorganisms

DOI: 10.5817/CPR2021-2-19

Introduction

The ecosystems of the Arctic region are very vulnerable and important for scientific research and biopreservation. The Pechenganikel plant, which extracts and processes sulfide copper-nickel ores, is known to be one of the largest sources of pollution in the Arctic. The plant is split into two industrial sites in the north-

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Received February 25, 2021, *accepted* November 30, 2021.
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Acknowledgements: The work was supported by the Russian Science Foundation, project No. 20-74-00108 "Algae and cyanobacteria in technogenic substrates and contaminated soils of the Kola Arctic".

western part of the Kola Peninsula in Murmansk region near the Norwegian border. One is in the city of Zapolyarny, and the second one is in the Nikel settlement. The industrial production is adjacent to a unique nature preserve, the Pasvik, founded in 1992 for the preservation and study of the northernmost pine forests in Europe, wetlands, and waterfowl fauna, as well as for the multiple monitorings of northern ecosystems. It is located 15 km away from Nikel, in the southwest region, spanning across both sides of the state border of Russia and Norway.

Gas emissions of the Pechenganikel plant entering the soil contain sulfur compounds and heavy metals. The clear correlation between the content of the main pollutants in the soil (Cu and Ni) and the distance from the pollution source enabled the zoning based on the pollution gradient (Evdokimova et al. 2014). Three zones were identified, differing in the intensity of soil pollution. The zone of strong pollution spreads up to 3 km from the source (Zone 1). Serious disturbance of the vegetation cover in the immediate vicinity of the smelter was noted after 30 years of its operation (Kalabin 1991). The content of heavy metals in the soil of the zone is two orders of magnitude higher than in the unpolluted regions (copper and nickel content exceeded 2000 and 3000 mg/kg, respectively). Here, forests and most of the vegetation have come to halt, and the soil fertile layer is almost absent (Mülgauzen et al. 2019). The zone of medium pollution (Zone 2, Cu and Ni content reached 450 and 700 mg/kg, respectively) extends up to 15 km, and the zone of weak pollution (Zone 3, Cu and Ni content reached 100 and 180 mg/kg, respectively) up to 25-30 km in the southwest direction from the source. Thus, the northern part of the Pasvik preserve is exposed to gas emissions of the Pechenganikel plant, especially those of Ni compounds. The content of Cu and Ni in the background soil did not exceed 50 and 80 mg/kg, respectively.

At the end of 2020, after 74 years of continuous operation, the smelter of the Pechenganikel plant located in Nikel was shut down due to the technological and environmental standards which were classified as outdated. As a result, harmful emissions were no longer released into the atmosphere near the Russian-Norwegian border. However, the damaged ecosystems of the Far North, where the processes of energy and mass transfer are especially slow, are extremely sensitive to anthropogenic impacts and need a long recovery period. The soils have low biogenicity and, consequently, low self-purification potential. Natural decontamination of soils polluted by metallurgical dusts can take centuries after the cessation of pollutant emissions (Barcan and Kovnatsky 1998).

Soil algae are promising as possible bioindicators in the soil monitoring system, since they possess a number of advantages over other soil organisms. They are quite easily identified into species, which makes it possible to compare the algal flora of different soils. Moreover, they are sensitive to changes in habitat conditions and respond to these changes in a way similar to that of higher plants. The response of soil algae to heavy metal pollution has been described in a number of research papers (Trzcińska and Pawlik-Skowrońska 2008, Kalinowska et al. 2008, Cabala et al. 2011, Song et al. 2014, Novakovskaya and Patova 2007, Maxwell 1991, Kalinowska and Pawlik-Skowrońska 2008, Rahmonov et al. 2015, Gaisina and Khaibullina 2007, Safiullina et al. 2009, Kabirov et al. 2017, Gornostaeva 2015).

In ecosystems degrading under the influence of anthropogenic factors, the intensity of soil algae development increases, which is one of the ways to maintain the stability of the autotrophic component of terrestrial ecosystems. The aim of the research was to study the algocenoses of soils along the southwest direction of pollution gradient from the Pechenganikel plant towards the Pasvik nature reserve.

Material and Methods

The field studies were performed on stationary monitoring plots in the aerial pollution gradient in the areas located southwest from the Pechenganikel plant, including the territory of the Paskvik reserve. The soils of the monitored plots are Albic Podzols developed from sandy moraine deposits with a high content of boulders. The soil samples for the algological analyses were collected from four zones differing in the intensity of the soil pollution: the zone of strong pollution (Zone 1: up to 0.1 km of the pollution source, at a distance of 2, 3 km to the southwest from the pollution source), the zone of medium pollution (Zone 2: 5, 7 km), the zone of weak pollution (Zone 3: 16, 25 km) and unpolluted or background (control) zone (40, 50 km) (Fig.1, Table 1). Five individual soil samples were taken aseptically from each plot from the organic horizons to a depth of 5 cm.

For the analysis of species diversity, strains were isolated using Z8 medium for cyanobacteria and Bold 3N liquid medium for algae and in Petri dishes with the same agarized culture media (Gaisina et al. 2008, Kotai 1972). Species were identified by morphological characteristics using a microscope Olympus CX 41 (Japan) with the camera Jenoptic ProgRes CT3 (Germany). For species identification a number of guides were used (Andreeva 1998, Ettl and Gärtner 2014, Komárek 2013). The species names and taxonomic affiliation were clarified by the electronic database AlgaeBase [1]. The similarity of the algae communities was analyzed using Sørensen-Chekanovsky coefficient (for average distance) in the GRAPHS program (Novakovskii 2004). The frequency of occurrence of algae species was calculated based on the following equation (Kondrat'eva and Kovalenko 1975, Kurakov 2001): B = (a/A) × 100, where B is occurrence (%); a is the number of samples containing the certain species, and A is the total number of the samples.

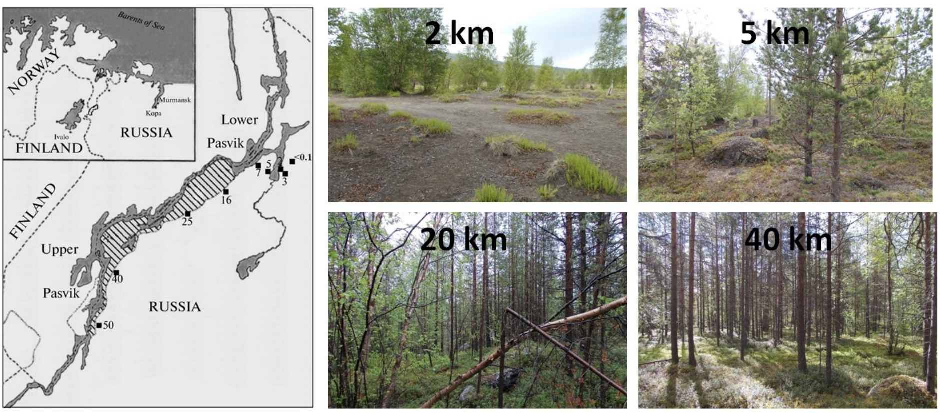

Fig. 1. Map (and photo) of the investigated area and sampling plots. The hatching shows the territory of the Pasvik reserve; the figures designate the distance from the Pechenganikel (km) plant.

Zone	Distance from the pollution source, km	Coordi-nates	Thickness of the O horizon, cm	Characteristics of the plot	Cu, mg/kg	Ni, mg/kg
Strong pollution	up to 0.1	69.4108 N 30.2488 E	Soil is eroded	No vegetation	-	-
	2	69.4075 N 30.1902 E	Soil is eroded	Birch forest with rare groups of horsetail on the eroded soil	2615	2083
	3	69.3955 N 30.1783 E	Soil is eroded	Cowberry pine forest with birch	1587	2143
Medium pollution	5	69.3925 N 30.1013 E	0–11	Dwarf shrub pine forest with birch. Ground cover: crowberry > cowberry	447	696
	7	69.4097 N 30.0566 E	0–5 (3)	Dwarf shrub pine forest. Ground cover: blueberry > cowberry > crowberry	426	664
Weak pollution	16	69.3625 N 29.8744 E	0–10	Dwarf shrub pine forest with birch. Ground cover: cowberry > crowberry > ledum	99	182
	25	69.3197 N 29.6722 E	0–3	Cowberry pine forest with birch. Ground cover: cowberry > ledum > lichens	110	160
Unpolluted	40	69.2266 N 29.3586 E	0–10 (15)	Dwarf shrub–green moss pine forest. Ground cover: bilberry > ledum > cowberry, green mosses	46	74
	50	69.1322 N 29.2755 E	0–5	Lichen–dwarf shrub pine forest. Ground cover: blueberry > bilberry> crowberry, lichens	26	22

Table 1. Sampling plots and characterization of the areas along the pollution gradient from the Pechenganikel plant (according to Evdokimova et al. 2014).

Results and Discussion

In total, 63 species of algae and cyanobacteria were found in the studied soils (Table 2). The highest species diversity of microphototrophs (25 species) was found 50 km away from the Pechenganikel plant in the southwest direction. Only 7 species were found in the heavily disturbed soil at the epicenter of pollution. In the remaining plots, 14 to 22 taxa were found, whereas no clear relationship between the abundance of soil algal species and the distance from the source of emissions was established. Overall, 34 species of algae were identified in the soils within the Zone 1, 26 species in the Zone 2, and 29 in the Zone 3. In control zone, 31 species in were identified. Regarding the number of species, algae from the Chlorophyta division predominated in all soils, accounting for more than 68% of the total number of taxa (Fig. 2).

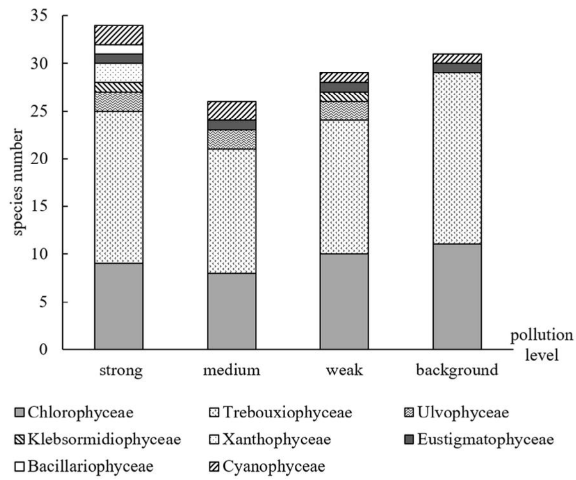

Fig. 2. Relative contribute of main algae groups (at the class level) to total abundance.

The predominance of algae from the Chlorophyta division, their low diversity in soils in coniferous forests was previously reported by several authors (*e.g.* Aleksakhina and Shtina 1984, Evdokimova and Mozgova 2001, Novakovskaya and Patova 2007, Kabirov 1991, Korneykova et al. 2017). The factors limiting the development of algae in such soils include the unfavorable effect of coniferous leaf litter, low soil pH values, and poor light penetration. Coniferous litter is poor in nitrogen, contains algostatic substances, has an acidic reaction, and among the heterotrophs decomposing such litter there are algae antagonists (Kabirov 1991). At the same time, several algae species were found in the studied soils, which showed resistance to unfavorable natural and anthropogenic factors (the frequency of occurrence is more than 77%). These included *Chloromonas* sp., *Neocystis brevis*, *Parietochloris alveolaris*, *Pseudococcomyxa simplex*, *Stichococcus bacillaris*. In addition to these species, a high frequency of occurrence (55-65%) was found for *Interfilum terricola*, *Leptosira* cf. *obovata*, *Myrmecia bisecta*, *Nostoc muscorum*.

Species	Pollution level			
	strong	***medium***	***weak***	***bg***
Phylum Chlorophyta, Class Chlorophyceae				
Bracteacoccus minor (Schmidle ex Chodat) Petrová	+		+	
Bracteacoccus sp.		+		
Chlamydocapsa lobata Broady	+			
Chlamydomonas sp.1	+		+	
Chlamydomonas sp.2				+
Chlamydomonas sp.3	+		+	
Chlorococcum sp.		+		+
Chloromonas sp.	+	+	+	+
Coelastrella sp.	+			
Coenochloris signiensis (Broady) Hindák		+	+	+
Dictyochloris fragrans Vischer				+
Eutetramorus tetrasporus Komárek		+	+	
Gloeococcus minutissimus J.M.King		+		
Lobosphaeropsis lobophora (V.M.Andreeva) Ettl & Gärtner			+	
Monoraphidium sp.				+
Neocystis brevis (Vischer) Kostikov & Hoffmann	+	+	+	+
Neocystis sp.	+			+
Neospongiococcum sp.			+	
Palmellopsis sp.				+
Radiococcus sp.				+
Radiosphaera negevensis Ocampo-Paus & Friedmann		+	+	+
Tetracystis sp.	+			
Class Trebouxiophyceae				
Asterochloris sp.		+		+
Chlorella sp.				+
Chloroidium angusto-ellipsoideum (Hanagata & Chihara) Darienko, Gustavs, Mudimu, Menendez, Schumann, Karsten, Friedl & Proschold	+	+	+	
Chloroidium saccharophilum (W.Krüger) Darienko & al.	+			+
Chloroidium sp.	+			
Choricystis minor (Skuja) Fott	+			+
Coccomyxa sp.1				+
Coccomyxa sp.2			+	+
Coccomyxa sp.3	+			
Desmococcus olivaceus (Persoon ex Acharius) J.R.Laundon	+			
Deuterostichococcus allas (Reisigl) Pröschold & Darienko				+
Diplosphaera chodatii Bialosuknia		+		
Elliptochloris bilobata Tschermak-Woess			+	+

Elliptochloris reniformis Darienko & Pröschold			+	
Elliptochloris sp.			+	+
Elliptochloris subsphaerica (Reisigl) Ettl & Gärtner			+	+
Leptosira cf. *obovata* Vischer	+	+	+	+
Microthamnion kuetzingianum Nägeli ex Kützing	+			
Muriella terrestris J.B.Petersen	+			
Myrmecia bisecta Reisigl	+	+		+
Myrmecia cf. *astigmatica* Vinatzer	+	+		
Parietochloris alveolaris (Bold) Shin Watanabe & G.L.Floyd	+	+	+	+
Parietochloris ovoidea T.I.Mikhailyuk & E.M.Demchenko	+			+
Pseudochlorella pringsheimii (Shihar & Krauss) Darienko & al.		+		
Pseudococcomyxa simplex (Mainx) Fott	+	+	+	+
Pseudococcomyxa sp.1	+		+	+
Pseudococcomyxa sp.2		+	+	+
Pseudococcomyxa sp.3		+	+	
Pseudostichococcus monallantoides var. exiguus (Gerneck) Pröschold & Darienko		+	+	
Stichococcus bacillaris Nägeli	+	+	+	+
Class Ulvophyceae				
Fernandinella cf. *semiglobosa* (F.E.Fritsch & R.P.John) Škaloud & Leliaerta	+			
Interfilum massjukiae Mikhailyuk, Sluiman, Massalski, Mudimu, Demchenko, Friedl & Kondratyuk		+	+	
Interfilum terricola (J.B.Petersen) Mikhailyuk & al.	+	+	+	
Phylum Charophyta, Class Klebsormidiophyceae				
Klebsormidium flaccidum (Kützing) P.C.Silva, K.R.Mattox & W.H.Blackwell	+		+	
Phylum Ochrophyta, Class Xanthophyceae				
Botrydiopsis eriensis J.W.Snow	+			
Xanthonema exile (Klebs) P.C.Silva	+			
Class Eustigmatophyceae				
Monodopsis subterranea (J.B.Petersen) D.J.Hibberd	+	+	+	
Vischeria magna (J.B.Petersen) Kryvenda, Rybalka, Wolf & Friedl				+
Class Bacillariophyceae				
Eunotia sp.	+			
Phylum Cyanobacteria, Class Cyanophyceae				
Nostoc muscorum C.Agardh ex Bornet & Flahault	+	+		+
Stenomitos kolaensis Shalygin, Shalygina & Johansen	+	+		

Table 2. Species diversity of algae and cyanobacteria in the soils affected by aerial emissions from the Pechenganikel plant. *Note*: bg: back-ground.

Only unicellular coccoid green algae from the Trebouxiophyceae class of the genera *Chloroidium*, *Myrmecia*, *Parietochloris*, *Pseudococcomyxa* were found in the heavily disturbed soils right next to the Pechenganikel plant (Zone 1). The survival of Trebouxiophyceae in an extreme environment is probably associated with the accumulation of organic osmolytes, such as polyols, which are considered to be effective stress metabolites with multiple protective functions in the microalga cell (Gustavs et al. 2011, Holzinger and Karsten 2013). No species specific exclusively to this area has been identified. All the species were found also in Zone 2 and 3. Yellow-green and eustigmatophytic algae, diatoms, and cyanobacteria were found 2-3 km away from Pechenganikel. However, they showed only low diversity: 1–2 species from each group. Yellow-green and eustigmatophytic algae were represented by the typical soil species *Botrydiopsis eriensis*, *Xanthonema exile*, *Monodopsis subterranea*. *Stenomitos kolaensis* and *Nostoc muscorum* were found among cyanobacteria, and the only species of diatom algae belonged to the genus *Eunotia*. Green algae still predominated. They were represented by three classes - Trebouxiophyceae, Chlorophyceae, and Ulvophyceae. We consider *Microthamnion kuetzingianum* to be an interesting find. In the soils of the Murmansk region, this species has been previously found only in the mountain-tundra belt of Mount Vudyavrchorr (Shtina and Roizin 1966). In addition, the drainage water of the Terrasa copper-nickel ore deposit (67.8881 N, 32.9144 E) near the city of Monchegorsk was found to contain *M. kuetzingianum* (unpublished data). There is evidence that this species is resistant to the increased acidity of the environment, high concentrations of metals, and is potentially capable of extracting toxicants from the substrate (Bray 2007, Foster 1982, Lampkin and Sommerfeld 1982, Vara Prasad and de Oliveira Freitas 2003, Płachno et al. 2015). Thus, its presence in the studied soil may be useful as an indicator, showing strong contamination with heavy metals and low soil pH.

The soil of Zone 2 is characterized by the absence of yellow-green algae and diatoms in the composition of algocenoses. Further along the gradient, we also did not find any representatives of these groups of phototrophs. In the Zone 3, the genus *Elliptochloris* appeared among green algae, which included four species at once. The genus is widespread in the soils of the Murmansk region. Its presence may indirectly indicate the relative well-being of the ecosystem in relation to anthropogenic load due to a decrease in the content of heavy metals with distance from the source of pollution. A specific feature of the background (control) area was the presence of the eustigmatophytic alga *Vischeria magna* in the soil. This species is very often encountered in soils around the world as well as in the Murmansk region. The species was found there in virgin tundra and forest soils (Shtina and Roizin 1966, Korneykova et al. 2017, 2018). The species is reported for the soils contaminated by the emissions from the Kandalaksha aluminum plant (Redkina et al. 2020b), as well as in the overgrowth on the surface of technogenic substrates (Redkina et al. 2020a, Davydov and Redkina 2021). It was shown earlier that populations of *V. magna* are highly resistant to surfactants, radioactive contamination, and are sensitive to soil contamination with heavy metals, oil, and oil products (Kabirov and Safiullina 2008). The results of our studies confirm the indicative role of *V. magna* in classification of the soil pollution with heavy metals. Therefore, the species could be used for the zoning of lands exposed to anthropogenic pollution and assessing the toxicity of the soil cover.

Concluding Remarks

The study of algae in the zone of influence of the metallurgical industry in the Kola North began in the 80s of the 20th century. Observations of the effect of heavy metals were carried out in laboratory and field experiments on the cultivated Al-Fe-humus podzolic soil. During the introduction of the initially uncontaminated soil into the zone of technogenic impact, rapid depletion of the species composition of the soil algal flora was observed due to the disappearance of yellow-green algae and cyanoprokaryotes (Evdokimova et al. 1984, Evdokimova 1995). In further laboratory experiments, air pollution of the soil with acidic compounds was excluded and the effects of copper and nickel on soil microorganisms were separated (Evdokimova et al. 1984). Tolerant, sublethal, and lethal doses of copper and nickel were identified for pure cultures of some green algae (Evdokimova et al. 1988).

Simultaneously, another group of specialists studied the disturbance of forest ecosystems in the zones of influence of metallurgical plants of the Kola Peninsula. In these studies focused on soil algaes, samples of soil were taken in dwarf shrub-green moss spruce forests near the Severonikel plant and in dwarf shrub-green moss pine forests in the vicinity of the Pechenganikel plant (Kabirov 1993, Chernenkova et al. 1995, Kabirov 1997). Due to the repeated sampling of the soil in the vicinity of the Severonickel plant from 2005 to 2008 at the test plots of the impact and buffer zones, it was possible to establish the features of the process of the restorative succession, which occurs as a result of the decrease of emissions from the metallurgical plant (Chernenkova et al. 2011). The number of algal species and the degree of their development experienced a sharp increase in comparison with the samples taken in 1982. In addition, there was a significant difference between the taxonomic composition of algocenoses in 1982 and 2008, a change in dominant species, and the appearance of yellow-green algae in the impact zone. According to Kabirov, these facts prove positive dynamics in the state of disturbed ecosystems (Chernenkova et al. 2011).

In the zone of influence of the Pechenganikel plant, Kabirov discovered 40 species of algae, mainly representatives of the Chlorophyta division (Chernenkova et al. 1995). Cyanobacteria and diatoms were not identified. A gradual decrease in species diversity correlated with the increase of the distance from the source of pollution - 27, 23, and 15 species were found in the zones of maximum, medium, and low pollution, respectively. Comparison of Kabirov's data with the results of our studies revealed a number of changes in the state of algocenoses after 30 years. Thus, the composition of the algal groupings studied by Kabirov in the zones of strong and medium pollution was numerically dominated by the representatives of the Chlorophyceae class (73%), mostly by species of the genus *Chlamydomonas*, whereas the classes Chlorophyceae and Trebouxiophyceae made an equal contribution to the total biodiversity of algae only in the weak polluted soil. According to our data, species of the Trebouxiophyceae class currently dominate in the same zones, with representatives of Chlorophyceae accounting for 27-42% of the total number of species. In addition, yellow-green, eustigmatophytic and diatom algae, as well as cyanobacteria appeared in the communities of the impact zone. In general, the species diversity of algae has increased by 35% in the soils influenced by the Pechenganikel plant in comparison with the data published previously. The described changes in the composition of the algal groups probably indicate a certain decrease in the anthropogenic load on the territories adjacent to the Pechenganikel plant. Therefore, follow-up studies of soil algae in the above-mentioned areas after the closure of the smelter of the Pechenganikel seem to be necessary in order to evaluate the process of the restorative succession in soils previously disturbed by industrial emissions.

The strains of algae and cyanobacteria isolated from the studied soils are included into the Herbarium of Institute of the Industrial Ecology Problems of the North of Kola Science Center of the Russian Academy of Sciences which is registered in the International Catalogue of World Herbaria (herbarium code INEP).

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[1] the electronic database AlgaeBase (http://www.algaebase.org)