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MORPHOLOGICAL AND ANATOMICAL STUDY, CONSERVATION AND WISE USE OF SPHAGNUM SPECIES OF PERCOLATION MIRES OF KOLKHETI LOWLAND

A B S T R A C T

of the thesis presented to obtain academic degree in Biological Science

Specialty: Biodiversity

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Content

Introduction

Actuality of the Research Topic. The sphagnum peatlands and flora of Kolkheti are relict ecosystem formed during the tertiary ice age.The glaciation was of local importance in Kolkheti. Some glaciers descended into the valley. The melting of the glaciers took many plants with them. Since that time, the flowering plant of the northern flora (Drozera) and the species of sphagnum survived both in the mountains and in the valley of Kolkheti. (Ketskhoveli, 1960: 3, Флеров, 1929, Флеров, 1929).

Kolkheti is the only place in the warm zone in the world where where alive ombotrophic sphagnum peatlands are widespread.These peatlands occupy a special place among the peatlands of the tropical and boreal zone. Kolkheti, due to its special properties and location is considered among the peatlands of the world as a unique place in the world for the percolation peatlands with living sphagnum. (Joosten, 2003, Joosten&Clarke, 2002).

Solving environmental problems and improving air quality, which is focused on conserving peatlands and restoring degraded ones, are on the agenda of the world today. Among all the other peat-producing species, the sphagnum occupies a special place. *Sphagnum* species are an edificator. Although most of the peatlands are protected, they are still affected by the impact of infrastructure projects. Peat is mainly produced in the Kolkheti lowland by sphagnum species, so each species plays a special role in the functioning of peatlands.

Recently, pollution of the biosphere (air, soil and water) has become a global environmental threat. Wetlands and reservoirs are one of the main "repositories" of pollutants, since industrial and agricultural discharges contain a wide range of toxic substances, including fertilizers, coloring materials, pesticides, heavy metals, radionuclides and others. Biological control of water and air is especially relevant for Georgia, where infrastructure, energy, agriculture and especially the extraction of natural resources (gold) are actively developing. In addition, the use of pesticides on agricultural land has increased the probability of pollution of the biosphere by various pollutants, especially heavy metals (Tetemadze et al., 2017).

240 species of bryophytes are affected by various pollutants (pesticides, discharge of industrial waste into lakes, emissions), of which 66 species are threatened with extinction (IUCN 2015).

2019 Red List results show that 88.2% of bryophyte species (1,603 species, including 319 endangered species) should at least exist in one protected area (including National Park, 2000 nature reserve or nature reserves) as these areas are subject to protection.

EU environmental policy is based on two important issues : 1. species protection under the Birds Directive, 1979 (Directive 79/409/EEC), and 2. conservation of natural habitats under the Habitats Directive, 1992(Directive 92/43/EEC;jointly referred to as the Nature Directives) (EC 2018).

IUCN provides the following guidelines for the protection of bryophytes: Use the European Red List of Bryophytes as a scientific basis for providing information on endangered species to the regional / national list and for setting conservation priorities

Research Aim and Objectives. The aim of this research was to study the biological characteristics and chemical composition of sphagnum species of peatlands of the Kokheti lowland.

In order to achieve the aim of the present study, research activities were carried out in accordance with the following tasks:

Task 1. The study of the systematic, morphological, anatomical characteristics and ecological environment of sphagnum species spreading to percolation sphagnum peatlands of the Kolkheti lowland;

Task 2.Determination of the content of the chemical composition of sphagnum species (*Sph.palustre, Sph. Papillosum, Sph.rubelum, Sph.imbricatum, Sph.cuspidatum*);

Task 3.Production and installation of the so-called sphagnum bags "in pre-selected areas, overloaded with transport in Batumi, and their chemical analysis to determine the content of heavy metals in the air.

Scientific Novelty. For first time in Georgia, it was possible to study the morphological, anatomical and ecological characteristics of 9 sphagnum species of the percolation peatlands, and the chemical composition of 5 sphagnum species. Moreover, to determine the content of heavy metals in the air of Batumi using the so-called "sphagnum bags".

Theoretical and Practical Significance. This study is important from a theoretical and practical point of view, since the determination of anatomical, morphological and ecological characteristics facilitated the identification of similar species, as well as the results of determining the chemical composition, which can be used to determine the antiseptic nature of sphagnum species, and "sphagnum bag" as a cheap means to monitor air pollution by heavy metals can be introduced for systematic and large-scale studies in different cities.

Approbation of Work. The research results were presented on the International and National Conferences:

1. International Scientific Conference “Green Medications - By Green Technologies - For Healthy Life”, Tbilisi, 2019;
2. IX International Scientific Agroculture Symposium “AGROSYM 2018”, Jahorina, Bosnia-Herzegovina, October 3-6, 2018;
3. 2nd International Conference on the Utilisation of Wetland Plants "RRR2017Renewable Resourcesfrom Wet and RewettedPeatlands", Greifswald, Germany, 2017;
4. Conference "Future Technologies and Quality of Life", Batumi. Georgia, September 29, 2017;
5. III Scientific Conference "Biodiversity and Georgia", Georgian Botanical Garden, May 2017;
6. Black Sea River Basin Ecology, Batumi, October 2016.

3 scientific papers were published in the refereed national and international scientific journals and in conference proceedings, and 7 are accepted for the publication

The Structure of the Thesis. The work consists of the following sections: Introduction, Literature Review, Materials and Methods, Results, Discussion, Conclusions, Recommendations, References. The thesis consisted of computer-printed 155 pages in Georgian language, 28 tables, 15 diagrams and11 figures. The bibliography of the PhD thesis is consisting of 92 sources in Georgian, Russian and English languages.

In literature review 45 references are analyzed on particular topic. It gives the state of current knowledge, an overview of key findings, concepts and developments in relation to a research problem or question.

ResearchMaterialandMethodology

The place and conditions of the research. The chemical composition of the sphagnum species was determined in the LEPL laboratory of the National Bureau of Forensic Science. Levana Samkharali within the framework of the Doctoral Scholarship of the National Science Foundation of Georgia Shota Rustaveli (SRNSFG): "Study of The Ecological, Chemical and Biological Characteristics of Sphagnum Species of Kolkheti Lowlands, Bio-Indicator of Air Pollution and Prospects for Use in Medicine".

The cological, systematic and morphological characteristics of the genus Sphagnum L. were studied at the Institute of Phytopathology and Biodiversity of Batumi Shota Rustaveli State University.

The spectrum of heavy metals in the "Sphagnum bag" was determined on the basis of the laboratory of the Department of Geology of Georgian Technical University. Batumi was a study area for monitoring atmospheric air with a sphagnum bag.

Research Material. All identified sphagnum species, widespread in the Kolkheti lowland, were used as research material to determine their anatomical and morphological features and environmental characteristics. The chemical composition was determined only for 5 species (Sph.paluster, Sph. Papillosum, Sph.rubellum, Sph.imbricatum, Sph.cuspidatum). In the process of biomonitoring, the study material was a so-called "sphagnum bag" made of Sph. From papillos.

Research Methodology. Handbook of European Sphagna (Daniels and Eddy 1985) was used as a guideline for systematic, morphological and anatomical studies of sphagnum.

ABinocular microscope was used to determine the anatomical features of the sphagnum species. The loupe (18 MM-10X TRIPLET) was used while working in the field.

To study of pant communities of Sphagnum using quadrate methodes. The dimensions of the squares are 25 cm x 25 cm.

A equipment (EC -Electric conductivity) was used to study the acidity and electrical conductivity of the ecological environment.

Atmospheric air was monitored by the"sphagnum bag" method in Batumi. To detect heavy metals in samples taken from the study areas, we used the preparation of a typical test procedure for sample 139 AY-4: dry ash ANALMETH.DOC, dry ash method of "standard conditions"using graphite cuvette.

Research results and discussion

Chapter 2.Systematical, anatomical, morphological and ecological characteristics of *Sphagnum* species in the Kolkheti lowland

Section*Sphagnum L.*

There are 5 species (*Sphagnum austinii* Sull. Sph*. magellanicum* Brid, *Sph. palustre* L, *Sph. rubellum* Wilson., *Sph. papillum* Lindb.) of the section –*Sphagnum*, which are common in live sphagnum peat bogs of the Kolkheti lowland.

Sphagnum austinii. *Sphagnum austinii* (*Sphagnum imbricatum*) is a main peat-producing species, which is the rarest and most sensitive species to environmental pollution worldwide (Machutadze et al., 2009; Kolkheti et al., 2019, Dokturovski et al., 1931, Kaff. , 2000)The climate change, drainage, fires, grazing and eutrophication are the reason for its reduction.

***Sphagnum austinii* – Morphological description.**The plant is more usually, rather dense, occasionally compact so that the stem is completely hidden by the closely packed spreading branches; dull green to yellow-brown or chestnut, compact forms often very dark with some purple-brown coloration

Description: Typically, of 4, sometimes only 3 branches; 2 distally spreading branches and 1-2 pendent branches which are short.

Stem: Typical of the section, inner layers of cortexstrongly fibrillose compared to the outer layer (exposed face often lackingfibrils); outer face of cortical cells with 2-3 pores.

Branch anatomy: Spreading branches about 15 mm; pendent branches rarely exceeding12 mm; Branch leaves: short, 1.5 mm, with well-developed hyaline cells.

Unlike other species of *Sphagnum,* only *Sphagnum papillosum,* like *Sphagnum austinii,* can create greenish-brown hummocks. Hummocks are characterized by a smooth, simple surface, and thin and small branch leaves (Daniels et al., 1985). In field work, *Sphagnum austinii* Sull (*Sph. Imbricatum* Russ) is sometimes difficult to distinguish from *Sphagnum papillosum* with the naked eye. Distinctive features of these two similar species are observed at the microscopic level, on the basis of which the species is identified.One reason for this is that both species are involved in the creation of sphagnum domes. They never interact with the aquatic environment and form permanent close communities, which makes it difficult to distinguish the two species from each other.In the usual case, Sphagnum austinii and Sphagnum papillosum are sharply different species with morphological features.

Comparative anatomical description of *Sphagnum austinii and Sph. Papillosum.*During field expeditions, the species differ from each other mainly in morphological observations. The loupe was used to study the morphological features of *Sphagnum* species. However, there are exceptional cases when different species are morphologically similar to each other and they can be distinguished by anatomical features. *Sphagnum Papillosum* is sometimes morphologically very similar to *Sphagnum austinii* in low temperature and low water conditions, which makes it difficult to distinguish between the two species. The difference can only be detected by microscopic examination. On microscopic examination, the fibrils of the hyaline cells of the branch leaves are more intense in *Sph. austinii* than in *Sph. in papillosum*. The difference between *Sph. papillosum* and *Sph. asutinii* is mainly based on the height and extent of the comb fibrils. Chlorophyll cells of *Sphagnum Austinii* are more intensely colored, fibrils are sharper and more unequal in length, while chlorophyll cells *Sph. Papillosum* are lighter in color, and the fibrils are of the same length, crossing the entire hyaline cell (Fig 1).

Figure 1. Habitus of *Sphagnum austinii* (left) and *Sphagnum papillosum* (right)

Sphagnum austinii.The shape of the leaves of the transverse branch is lanceolate, is hunging inclined towards the apex, the apex is sharply pointed, the apex ends with a mustache-like cell protrusion, which Sph. does not have *Sphagnum papillosum*. The leaf shape of the *Sphgnum papillosum* branch is pitcher-like, the basal part of the leaf is wider, narrows towards the apex and is inclined from the base to the tip.

If we place the branch leaves of these two species on the slide and and carefully examine, we will see that they differ not only morphologically, but also in size.

Sphagnum magellanicum **Brid.** This species also belongs to the Sphagnum section and participates in the formation of the sphagnum dome. From the sphagnum peatlands of Kolkheti it grows only on the peatlands of Imnati. **Plant:** Robust but, typically, rather short (in size and habit rather similar toS. Papillosum. Pale green (though nearly always with at leastsome flecks of red or pink) to wine-red throughout. **Fascicles**: Mostcommonly with 4, occasionally 5, branches; spreading branches usually 2, blunt; pendent branches thin and pale.
Branch leaf: The shape is tongue-shaped, in the direction of the acumen, the ends are bent, in the third part of the leaf.

They are characterized by acute-angled hyaline cells.
Stem: Cortex often lacking fibrils or only faintly fibrillose; outer cortex with one pore per cell; internal cylinder dark purple-brown ordeep red.
Habitat: Widespread in oligotrophic mires, where it grows usually in association with S. papillosum or S. capillifolium, as broad carpets or low hummocks. It is more susceptibleto burning and drainage than S. Papillosum.
Sph.capillifolium **(Ehrh.) Hedw. var *rubellum* Wilson.**It may be associated with Sphagnum austionii and *Sphagnum papillosum* forming spagnum dome. Sph. capillifolium is rather delicate, but variable in height and compactness according to abiotic factors; capitula well developed, pale or with small to extended patches of red in its outer layers (Fig. 2).

Fig.2 . ***Sph.capillifolium*** **(Ehrh.) Hedw. var *rubellum* Wilson**

Fig 3. *Sphagnum papillum* Lindb

Fascicles: Composed of quite strongly dimorphic branches; spreading branches 2, the upper ones rather short, colorful; **Stem**: Up to 0.7 mm diameter; cortex of 3-4 layers of hyaline cells without pores; internal cylinderwell developed, pale or with small to extended patches of red in its outer layers. **Stem leaves**: lingulate; apex broadly rounded-truncatewith a rather narrow area of apical resorption, occasionally slightly inrolled; Hyaline cells strongly fibrillose in the upper half of leaf, at least on the abaxial face.

Fig. 4. *Sphagnum austinii* Sull

Sph. capillifolium is a widespread and abundant taxon, shows considerable variation in colour, habit and minor morphological characters. Plants: capitula usually more or less flat; nearly always with red to reddish brown, seldom deep crimson throughout. **Fascicles**: not closely packed. **Stem leaves**: Lingulate, fibrillose in the upper half; Branch leaves: usually narrowed above to a more or less tubular apex (appearing distinctly acute). Although this species is characterized by large variations in coloration and habitat, it can be easily distinguished during fieldwork by its coloration and distribution area.

***Sphagnum palustre* L.**

Sphagnum palustre grows in an ombotrophic environment in Kolkhetis, while everywhere it is characteristic of a minerotrophic environment.

Sphagnum paluste (Fig. 5) does not participate in the formation of the dome and spreads in the buffer zone of the peat bog along the margin, in ditches, close to the water surface.

Fig. 5 *Sphagnum palustre* L.

Plants: Robust, normally pale green or yellow-brown with a more strongly colored capitulum varying from red-brown to straw to pinkish, but never deep red, occasionally the whole plant green (in wet habitats in deep shade); the capitulum is not colored differently. **Fascicles**: Distant orrather crowded; of 3-6 dimorphic branches; spreading branches 2-3; pendent branches 1-4, pale and thin; as long as or longerthan the spreading. **Stem**: typical of the section; outer cortical cells almost always with spiral fibrils; outer faces of outer cortical cells mostly with 2-5 fairly large pores; internal cylinder dark brown;**Branch:** Branches15-25 mm long; leaf length varies from 1.5-3 mm depending on plant propagation and lighting characteristics. The less light the plant is exposed to, the larger its branch (3 mm). **Hyaline cells**: Variable in size but always relatively wide (20-30 micrometers).

Habitat: This species is widespread in mesotrophic peatlands under conditions of high acidity. This is one of the species that tolerates shade well. In the Kolkheti lowland, this species is found on almost all peat bogs (Churia, Grigoleti, Imnati, Ispani 1 and Ispani 2), and is not common on the sphagnum peatlands of Nabada.

Section *Cuspidata*

Sphagnum cuspidatum Ehrh.ex Hoffm.

Plants: Mostly medium-size, varying from rather compact terrestrial forms to very lax, free-floating; The coloration ranges from yellowish-green to orange, but is never found in red. **Fascicles**: Closely set to very distant, with 4-5, rarely 3 branches; **Stem**: distinct cortex, of 2-3 layers of moderately inflated hyaline cells; Branch leaves: narrowly lanceolate to linear (except a few at branch base); **Hyaline cells**: narrow, abaxialsurface with 1-3 pores..

Habitat: A common species of wet, oligotrophic sites, where it is often floating or submerged. It is found mostly in ombrotrophic bogs or acid fens. It may be associated with S. recurvum forma fallax or S. papillosum.

At the edges of open waters in the buffer zone of the sphagnum peat bogs of the Kolkheti lowland, we often encounter Sph .cuspidatum, namely on Ispan 2, Churia, and Imnati, which form an association with Sph. Fallax.

Sphagnum fallax

This species also belongs to the section Cuspidata. **Plant**: Medium-sized, green to mustard-brown, growing in carpets. Male plants may have a contrasting darker tip to the capitula and spreading branches. Capitula are usually stellate. Branch leaves are weakly to clearly in straight lines. Spreading and pendent branches are well-differentiated; the pendent branches are almost white, appressed to and hiding the stem, about as long as or shorter than the spreading branches.The stem leaf is more or less triangular. When dry, spreading branch leaves are conspicuously recurved.

Habitat: Very common in a wide range of permanently damp or wet habitats, including nutrient-poor to intermediate fens, and pools and runnels on bogs.

Section Subsecunda

***Sphagnum denticulatum* Brid.**

A medium to large species. Plants are green, although in Europe it is also found in yellowish-brown; capitula is rounded; branches to the capitula are relatively large and curved; Capsules are occasional; The branch leaf is large and wide and opens only in the apex part; Fascicles are of 3–4 similar looking branches, sometimes with a fifth vestigial branch. Habitat: Found in areas of high humidity, markedly acidic and nutrient-poor habitats, but does occur in habitats that are moderately enriched with nutrients. In the Kolkheti lowland it is distributed in the peatlands of Churia, Imnati, Ispan 2 and 1.

Threathened *Sphagnum* Species in the Kolkheti lowland

Red List of European Bryophytes published by IUCN in 2019 includes the status of mosses, especially the genus Sphagnum. According to this list, the status of 7 species of sphagnum spread in the Kolkheti lowland is as follows: *Sphagnum austinii*-EN, *Sphagnum capillifolium*-LC , Sphagnum cuspidatum -LC, *Sphagnum fallax*–LC, *Sphagnum palustre*– LC, Sphagnum papillosum LC, Sphagnum rubellum LC

Ecology of habitat of destribution of Sphagnum species

In order to study the ecological characteristics of the Kolkheti peatlands, the water acidity and electrical conductivity of the Kolkheti sphagnum peatlands were studied. The tool: EC -Electric conductivity was used to study.

A study of the acidity of water in Imnati Pelands showed that the pH in the upper layers of water of peat bog varies between 4.0-5.5. Electrical conductivity (EC) was also measured, ranging from 40 to 70 μS at a depth of 70-100 cm. Both indicate an ombrotrophic environment. The level of C / N content was also determined. In peat bog, at a depth of 10 cm, it ranges between 9.8–51.6, which indicates a mesotrophic and oligotrophic peat environment. The following species of sphagnum grow on Imnati peat bog: *Sphagnum palustre, Sph. rubellum, Sph. magellanicum, Sph. austinii, Sph. Papillosum.*

Sphagnum palustre and Sph. papilosum are common in the peatland. The pH of the peat was determined in the range from 4.26 to 5.65, and the conductivity (EC) of the water in the peat was from 22 to 134 μS cm^{-1}.

Chapter 3. Determination of Antibacterial substance of *Sphagnum* species

Juice producing technology. Raw plant material is cut with laboratory machine – the sizes of cut particles are 2-8 mm. the cut raw material is wrapped in linen and then put in pressure cylinder. They are separated with rust resisting steel sheets and pressed for the

purpose of receiving juice. To warm the juice, juice is put in water which has 80-85^{0}C temperature for 30 minutes and then it is cooled with water. Such changes of temperature support inactivation of ferments and protein materials. The sediment is removed with centrifugation. This way clean transparent juice is produced.

Producing watery exhaustions and water-soluble saccharide material from dry plant raw material. The exhaustion of research plant raw material is carried with hot rule at boiling water bath for 2 hours. The received exhaustions are filtered as soon as prepared. The correlation of raw material and extragen equals to 1:1 weight part. The exhaustion is divided into two parts. One part will be used in further research with the title "Watery Extract".

From the second part of exhaustion polysaccharide complex is produced for which exhaustion is thickened in vacuum-rotational apparatus at initial volume 1/10, in rarely conditions.

The sedimentation of water-soluble materials in particular polysaccharides is made by adding three times more ethyl alcohol 96%. It is added regularly in mixing conditions. The exhausted mucous sediment is put for some period to rest, the liquid upon sediment is removed with decantation and the rest mass is dried in vacuum box at 50-60^{0}C at not more than 7% humidity.

Studying antibacterial activity of research subjects. The research of antibacterial activity of subjects was carried in Giorgi Eliava Bacteriophage, Microbiology and Virology Institute. The evaluation of antibacterial effect was carried via standard protocol using Disc Diffusion Method – DDM and agar well diffusion methods. Antibacterial activity will be measured according to existence or non-existence of inhibitory zone and determination of Minimal Inhibitory Concentration –MIC. In-vitro evaluation of antimicrobial activity was carried on the following crops towards certain set of strains: Klebsiella spp, E-coli, *Pseudomonas aeruginosa, Streptococcus spp, Enterococcus spp, Shigella spp, Salmonella spp, Enterobacter spp, Proteus spp, Staphylococcus aureus*. 2% BH in Agar cup bacterial greenway was done – 1 ml bacterial crop was put and separated at whole cup. After the cup was dried 10 mkg research subject was put on it. The cups were placed in thermostat during 18-24 hours for incubation.

Antibacterial Activity. The used bacterial strain list with sensitivity results is given under Table N 1. *Sphagnum papilosum* -Juce; *Sphagnum palustre*- juce; *Sphagum rubellum* – Polysaccharides; *Sphagnum papilosum* – Polysaccharides; *Sphagnum papilosum* -waterly extract; *Sphagnum imbricatum*-waterly extract; *Sphagnum palustre* - Polysaccharides;

Table 1

Results of the study of antibacterial action

Strain	SumpleN							
	1	2	3	4	5	6	7	8
Streptococcus pyogenes			2+					
Escherichia coli								1+
Enterobacter cloacae						1+		

Salmonella typhimurium							4+	
Klebsiella pneumoniae								
Proteus vulgaris							4+	
Shigella flexneri							4+	4+
Enterococcus faecalis								
Staphylococcus aureus							4+	
Pseudomonas aeruginosa								

According to carried research antiseptic substances of *Sphagnum rubellum, Sphagnum austinii* and *Sphagnum papilosum* are stipulated by polymeric substances similar to spectin – Sphagnan (Table N 1 Sample N 3 and N 6). From research subjects *Sphagnum papillosum* watery extract has wide spectrum and expressed bactericide activity. The results of antibacterial activity research make vivid the prospect of usage sphagnum in medical practice.

Chapter 4.Determination of the Chemical Composition of *Sphagnum* Species of the Percolation mires of the Kolkheti Lowland

The chemical composition of 5 sphagnum species (*Sphagnum palustre, Sph. papillosum, Sph.capiliifolium, Sph. austinii, Sph. cuspidatum*) of the unique relict percolation peat bog of Ispan 2 was determined. The content of biologically active substances in the species was determined by gas chromatography-mass spectrometry (GC-MS / MS). It was found that sphagnum contains biologically active substances, such as: esters - palmitic acid, oleic acid, carbohydrates - glucofuranose, ribofuranose and phenolic compounds.

Research was carried out on 5 Sphagnum species: *Sph.paluster, Sph. Papillosum, Sph.rubelum, Sph.imbricatum, Sph.cuspidatum*. Therefore, *Sph. austinii* is a rare species in the world that is sensitive to air pollution (Allard, 2017), preserved by the pristine percolation peatland of Spain 2. Determination of their chemical composition and concern for conservation is crucial to the conservation of species.

The research objects were prepared in February 2016. The material was taken from different zones (dome and buffer) of the Ispani-2 percolation peat bog of the Kolkheti lowland. The test material was kept at room temperature for 24 hours, then dried in an incubator (Thermo Scientific Heraeus) at 30-70°C. The dried sphagnum was broken into pieces, of which 2 g were placed in a container and methanol was added.The containers were placed in an ultrasonic bath (mrc) at 30 ° C for 30 min. Re-extraction was carried out after extraction. Agilent Technologies 7000 GC / MS / MS Triple Quad was used to analyze methanol extracts. 1 μl of extract is injected into the injector. The oven is heated from 250^0C to 310^0C. The carrier gas is helium. The tube diameter is 60 m × 0.25 mm (TX-1701).The retention time of ions in a tube depends on its physical properties. Multilateral processing of the material was carried out after the development of the mass spectrum.

16 biologically active substances were detected in 5 species of Sphagnum (Sphagnum austinii Sull. (Sphagnum imbricatum ssp. Austinii), Sph. papilosum Lindb., Sph. palustre L., Sph.cuspidatum Ehrh.ex Hoffm., Sph. rubellum Wilson.) by using the gas chromatography -

Agilent Technologies 7000 GC/MS/MS Triple Quad. Qualitative difference in the spectrum of substances was found in the studied objects (Diagram 1). Esther Cis-9-Octadecenoil Acid - oleic acid, retention time 12.53 and Cis-9-Palmitoleic Acid - palmitic acid, retention time 11.67 (Table 1, Figure 3) and also, Dimethyl 1,4-cyclohexanedicorboxylate, retention time 10.32 were identified in all five species.

Phenolic compound - Methyl 4-O-benzyl-beta-D-xylopiranoside was observed only in N4 sample, retention time 11.16 (Table 1 and Diagram 2). Polosaccharide 1,2-Benzenedicarboxylic acid revealed in all five samples, retention time 9.5 (Diagram 1 and 4). Polysaccharide Alpha-D-glucopyranoside was detected in all five species, retention time 14.36(Diagram 4).

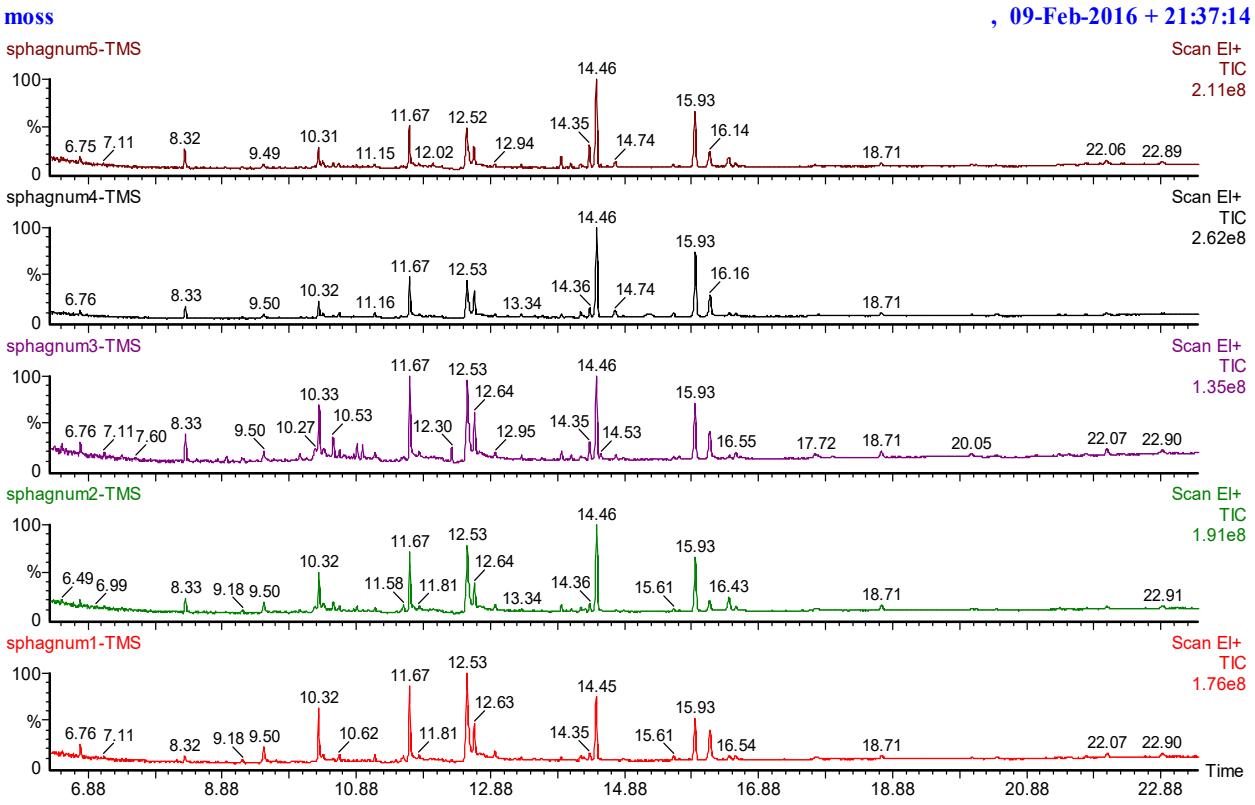
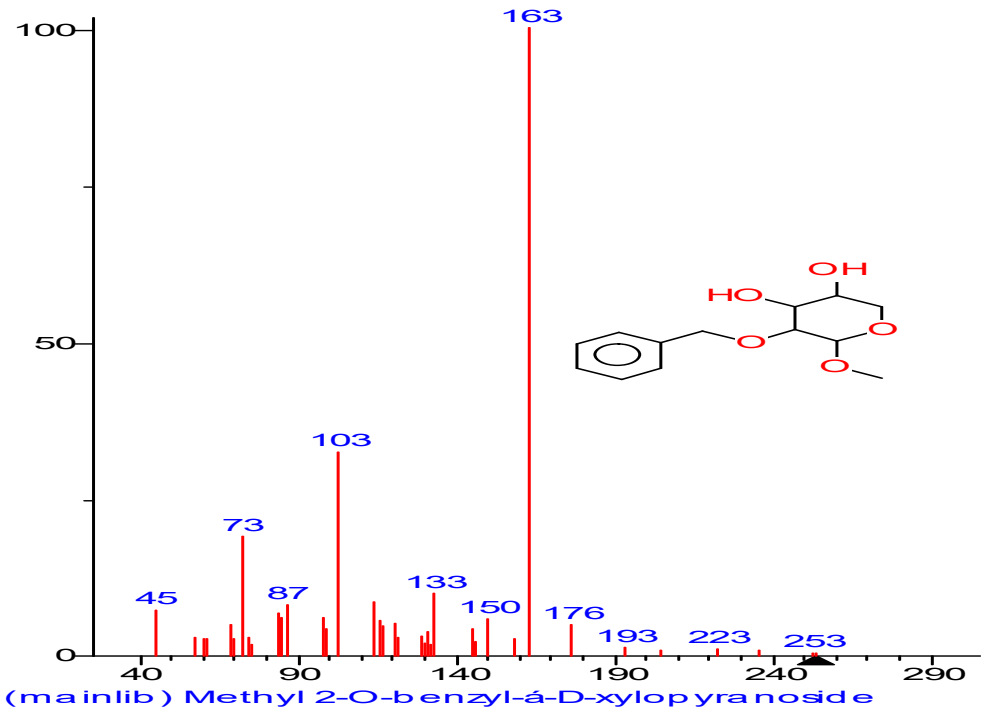


Diagram 1. *Sphagnum L.* (The sample *Sphagnum* 1 corresponds to *Sph.rubelum*, the sample *Sphagnum* 2 corresponds to *Sph. papillosum,* the sample *Sphagnum* 3 corresponds to *Sph.palustre,* the sample *Sphagnum* 4 corresponds to *Sph. cuspidatum* and the sample sphagnum 5 corresponds to *Sph. imbricatum)* A range of substances of *Sphagnum* moss detected by using GC-MS/M. Figures on the diagram describe the retention time of the listed substances as indicated in Table 1.

Table 2

Biologically active substances detected in Sphagnum species using GC-MS/MS.

	Retention time	Chemical name	Common name	Molecular weight
1	8,32	Malic acid	Malic acid	134,08744
2	9,5	1,2- Benzenedicarboxylic acid	Phthalic acid	166.13084
3	10,32	Dimethyl 1.4-cyclohexanedicorboxylate	Benzenedicarboxylic acid	200,23
4	10,53	Glucofuranosa	Glucofuranose	220.21974
5	10.62	Myristic acid	Tetradecane acid	228.37
6	10.88	D-xylose	Xylose	150.1299
7	11.16	Methyl 4-O-benzyl-beta-D-xylopiranoside	Ribofuranosis	254.27
8	11,24	Ascorbic Acid	Vit-C	176.12412
9	11.67	Cis-9-Palmitoleic Acid	Palmitic acid	326
10	12,53	Cis-9- Octadecenoil Acid	Oleic acid	282,46136
11	13,93	3-O-Alpha-D-glucopyranosyl-D-fructose	Turanose	342,29648
12	14,0	Arabinose	Arabinosa	150,1299
13	14,23	Glycerol-3-palmitate	Monopalmitic acid	330,50262
14	14,36	Alpha-D-glucopyranoside	Glucopyranosis	918
15	14.35	Ribitol	Ribitol	152,14578
16	16.55	1-(4-Methyl-6-methoxy-2-quinolyl)1'-phenyl-3,3'-dimethyl-(4,5'-bipyrazol)-5-ol		425

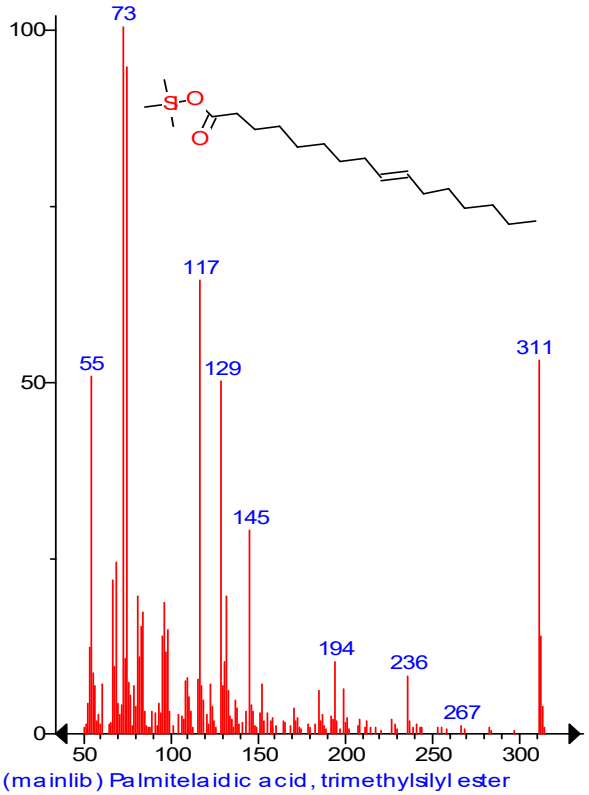


Name: Methyl 2-O-benzyl-á-D-xylopyranoside

Formula: $C_{13}H_{18}O_5$

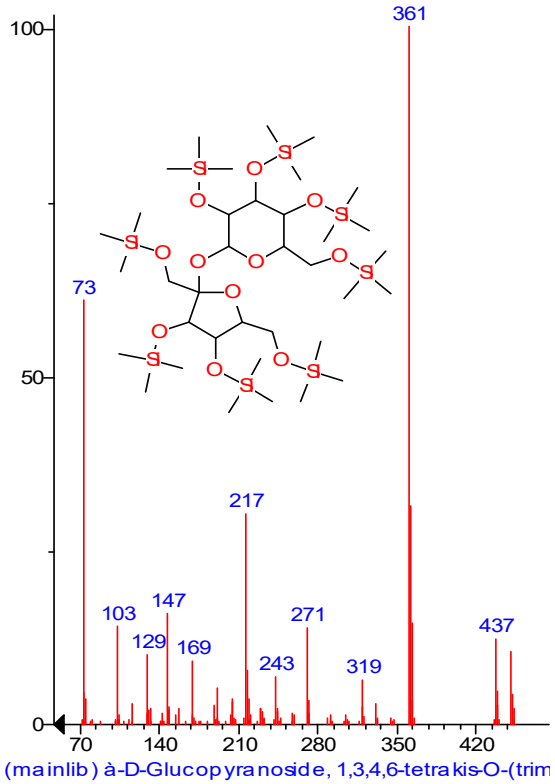
MW: 254

Diagram 2. The phenol compound Methyl 2-O-benzyl-á-D-xylopyranoside detected in *Sphagnum cuspidatum* using gas chromatography (GC-MS/MS).



Name:Palmitelaidic acid, trimethylsilyl ester
Formula: $C_{19}H_{38}O_2Si$
MW: 326

Diagram 3. The ester Cis-9-Palmitoleic Acid detected in *Sphagnum* species of the percolation bog using gas chromatography (GC-MS/MS)



Name:a-D-Glucopyranoside
Formula: $C_{36}H_{86}O_{11}Si_8$
MW: 918

Diagram 4. The Alpha-D-glucopyranoside polysaccharide detected in *Sphagnum* species of the percolation bog using gas chromatography (GC-MS/MS)

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atmosphere under conditions of high precipitation.

During the study period: average precipitation was 40.81 (mm), and the temperature was 11.5 ˚C in November 2016. In February 2017, average precipitation was 5.5 (mm) and the temperature was 13.8 ˚C. In May 2017, precipitation was 74 (mm)), and temperature was 16.72 ˚C, precipitation in August 2017 was 56.96 (mm), and temperature - 27 C˚ (Diagram 5).

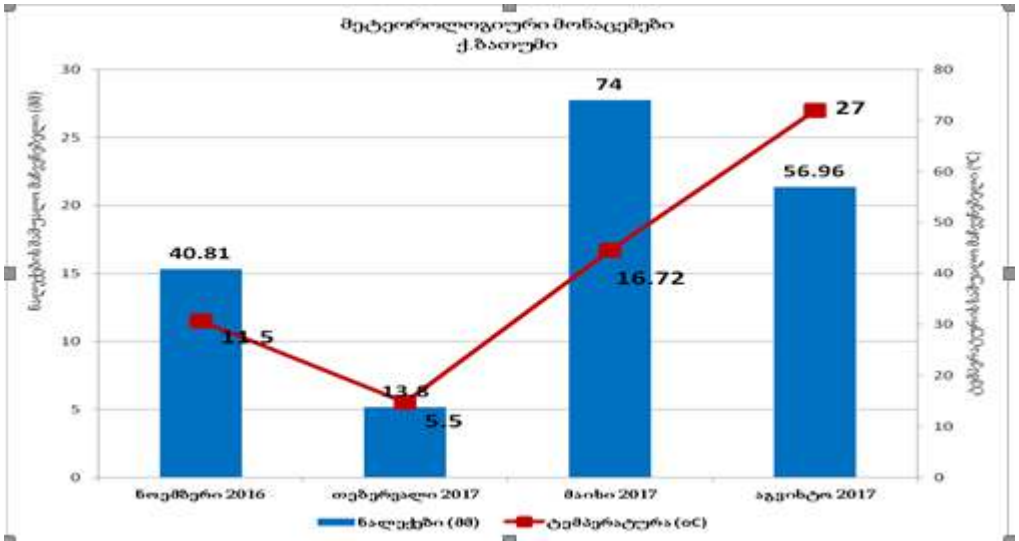

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Diagram 5. Meteorological parameters - average precipitation and temperature data for the study period

The result of Batumi biomonitoring research

On 2nd of November 2016, sphagnum bags were installed at four pre-selected study sites. On 17 th of November, samples were removed, packaged, and prepared for further laboratory testing. Atomic adsorption studies on 36 heavy metals were conducted for samples taken from all four sites. The range of different heavy metals was found in the samples depending on the area and time of year.

5.1.The Result of the Study, Autumin 2017

The study showed that the sample of the sphagnum bag installed on Gogoli street in November did not contain the following 8 heavy metals: As, Sr, Mo, Sn, Sb, Ba, Ta, Pt, but 9 elements were in such a small amount that its percentage was equal to zero. High content of the following metals: Mg, Al, Si, P, S, K, Ca, Ti, Cr, Mn, Fe, Ni, Cu, Zn, Zr, Ag, Pb was observed.From the listed 3 toxic element ofUnited Nation highest concentrationrecorded

Cd - 2.33 (μg/ml), Hg 6.64 μg/ml, Pb 8.25398 (μg/ml).Chromium (Cr) is highly toxic and carcinogenic (Tchounwou et al., 2001), the level of which in the sample is 1.7324 μg / ml. (Table 1 and Diagram 1).

Table 3

The result of the AAS analysis of samples taken from Gogoli Street in November 2016

Name of sample	Gogoli Street XI/16	20.11.16
Voltage	9(KV)	
Flow	150(μA)	
Elements	Concentration (μg / ml)	Showed in %
Mg	12.15	1.31012
Al	25.05	1.016039
Si	386.15	6.49371
P	45.02	0.21922
S	114.61	1.57203
K	549.83129	4.50258
Ca	861.75	6.85041
Ti	8.28889	0.0.20534
V	0.59966	0
Cr	1.7324	0.0115
Mn	25.49034	0.5285
Fe	469.32176	2.87001
Co	2.13394	0
Ni	2.07586	0.00661

Cu	21.40651	0.0261
Zn	72.42371	0.25331
As	0	0
Zr	24.32611	0.01432
Sr	0	0
Nb	1.04094	0
Mo	0	0
Ag	2.99841	0
Cd	2.33	0
Sn	6.48	0
Sb	0	0
W	2.68817	0
Au	1.21736	0
Pb	11.51203	0.02334
Ba	0.48	0
Hf	0	0
Ta	0	0
Hg	6.64	0
Bi	0.01	0
Th	1.19	0
U	2.81	0
Pt	0	0

During the atomic adsorption studiesof sphagnum bags taken from Tbel Abuseridze Street, it was found that 17 studied heavy metals were not detected or were equal to zero in percentage.Among them, Cd and Hg are highly toxic. However, higher levels of Pb (10.46483 μg / ml) and Cr (2.98128 μg / ml) were observed compared to Gogol Street (Table 2 and Diagram 1). Tbel Abuseridze Street is characterized by high-rise buildings, congested transport and low ventilation, which led to the accumulation of lead and chromium with a relatively high concentration in the area.

Table 4

The result of the AAS analysis of samples taken from TbelAbuseridzein November 2016

Name of sample	TbelAbuseridze XI/16	20.11.16
Voltage	9(KV)	
Flow	150(μA)	
Elements	Concentration (μg / ml)	Showed in %
Mg	7.58273	0.80389
Al	24.51883	1.13473
Si	383.8782	6.47472
P	20.39734	0.07101
S	76.88768	1.0389
K	112.41237	0.90152
Ca	1145.70245	9.85005
Ti	10.44676	0.2703
V	1.30535	0
Cr	2.98128	0.02494
Mn	27.72905	0.54338
Fe	627.96027	3.78859
Co	4.15372	0

Ni	5.13411	0.04135
Cu	24.5052	0.05078
Zn	128.35247	0.52758
As	0	0
Zr	25.76621	0.01528
Sr	0	0
Nb	3.32539	0
Mo	0	0
Ag	25.695	0.06662
Cd	3	0
Sn	1.66	0
Sb	0.39	0
W	2.39621	0
Au	1.41265	0
Pb	10.46483	0.01926
Ba	0.25	0
Hf	2.18864	0
Ta	0	0
Hg	3.71	0
Bi	0.07	0
Th	0.54	0
U	0.84	0
Pt	0	0

The following content of the four most toxic elements: Cd (3,43 μg / ml), Hg (4,27 μg / ml), Pb (8,58129 μg / ml), Cr (1,45691 μg / ml) were detected from the samples of the sphagnum bag installed on Tamar Mepe Avenue. However, the contents of Cd (3,43 μg / ml) and Hg (4,27 μg / ml) are so insignificant that their percentage is zero. Pb (8.58129 μg / ml) is lower than the rate found in the samples of Tbel Abuseridze and Gogoli street. Cr (1,45691 μg / ml) content also lags behind the results of analysis of samples from Tbel Abuseridze and Gogol streets (Table 3 and Diagram 1).

It should be noted that Tamar Mepe Avenue is characterized by high ventilation, and there are no high-rise buildings in the installation area of samples which is an obstructive factor for the accumulation of dust, exhaust and trace elements.

Table 5

AAS analysis of samples of "sphagnum bag" taken from Tamar Mepe Avenue in November 2016

Nameofsample	Tamar Mepe Avenue XI/16	20.11.16
Voltage	9(KV)	
Flow	150(μA)	
Elements	Concentration (μg / ml)	Showedin %
Mg	12.82	1.38
Al	31.32	1.46334
Si	436.17	7.05388
P	24.38	0.09135
S	64.6	0.86523
K	140.30637	1.10202
Ca	1656.54	15.64724
Ti	10.04552	0.25822
V	1.87537	0

Cr	1.45691	0.00854
Mn	26.59321	0.53583
Fe	795.45773	4.84127
Co	5.75837	0.00277
Ni	2.9255	0.01626
Cu	22.91152	0.03824
Zn	108.43052	0.42954
As	0	0
Zr	40.97229	0.02546
Sr	0	0
Nb	1.2774	0
Mo	0	0
Ag	13.20702	0.02685
Cd	3.43	0
Sn	0	0
Sb	1.78	0.00178
W	1.86786	0
Au	1.69002	0
Pb	8.58129	0.0143
Ba	0	0
Hf	1.98453	0
Ta	0	0
Hg	4.27	0
Bi	0.27	0
Th	0.58	0
U	1.12	0
Pt	0	0

The following spectrum of heavy metals: Mg, Al, Si, P, S, K, Ca, Ti, Cr, Mn, Fe, Ni, Cu, Zn, Zr, Ag, Pb were detected in the samples taken from Kakhabri Street. Although, this area is characterized by high traffic intensity. The lead, mercury and chromium content is lower than the concentration found in the sphagnum bags of the other study objects and it is also close to the index of samples from Tamar Mepe Avenue. These two areas are characterized by a high rate of ventilation and low buildings. Notably, the highest Cd rate (3.96115 μg / ml) was observed in the Kakhabari street samples compared to all sites.

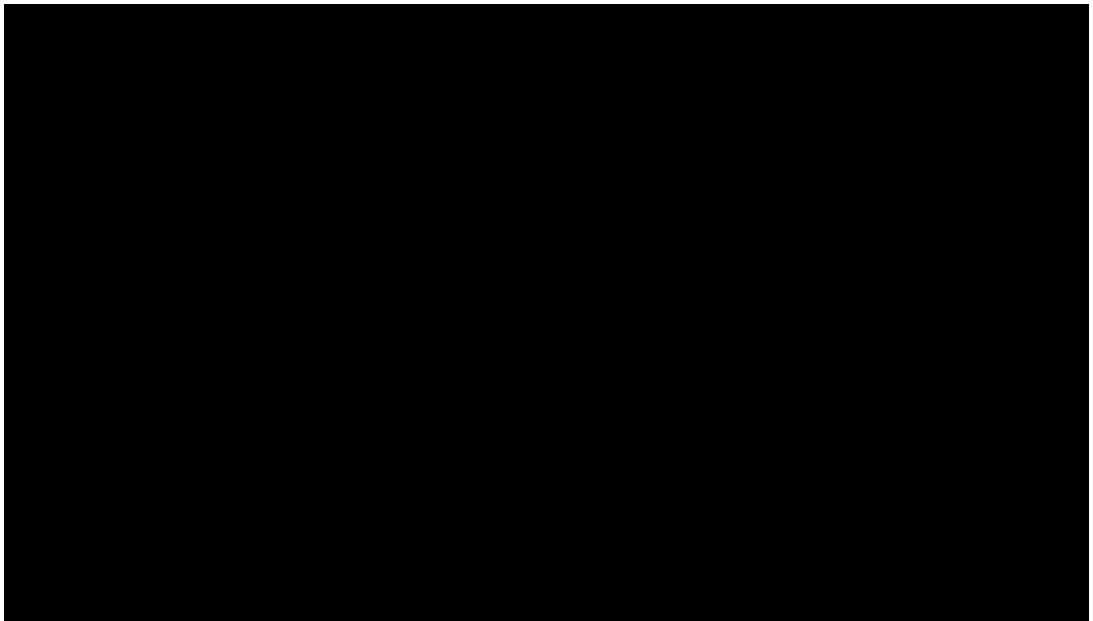

Diagram 6. Comparison of the results of the study of Cd, Hg, Pb, Cr in samples taken at different sites in November 2016

Table 6

AAS analysis of samples of "sphagnum bag" taken from Kakhaberi Street in November 2016

Name of sample	Kakhaberi Street XI/16	20.11.16
Voltage	9(KV)	
Flow	150(μA)	
Elements	Concentration (μg / ml)	Showedin %
Mg	6.68	0.70383
Al	21.99	1.01254
Si	338.48	6.00937
P	18.87	0.06441
S	73.01	0.98409
K	118.3408	0.94413
Ca	1137.35	9.76181
Ti	8.26359	0.20458
V	1.40131	0
Cr	1.17602	0.00552
Mn	28.23333	0.54673
Fe	589.19399	3.56412
Co	4.25677	0
Ni	3.59424	0.02386
Cu	22.15016	0.0321
Zn	126.4034	0.51799
As	0	0
Zr	17.42527	0.0097
Sr	0	0
Nb	1.30059	0
Mo	0	0
Ag	13.78778	0.0287

Cd	3.96115	0
Sn	0	0
Sb	0	0
W	2.53282	0
Au	1.25481	0
Pb	8.25398	0.01328
Ba	0	0
Hf	1.99147	0
Ta	0	0
Hg	2.4307	0
Bi	0.10003	0
Th	0.39011	0
U	0.38011	0
Pt	0	0

5.2. The Result of the Study, Winter 2017

On February 5, 2017, the atomic adsorption studiesof the "sphagnum bag" taken from four research sites in Batumi revealed the total range of heavy metals for all four sites, which consists of 17 elements: Mg, Al, Si, P, S, K, Ca, Ti, Cr, Mn, Fe, Ni, Cu, Zn, Zr, Ag, Pb. However, in the samples installed on Kakhaberi Street, 2 different elements were found: Sb and Co, and only Co was found on Tamar Mepe Avenue (Table 5, 6,7,8).

The highest rates of Pb and Cr were observed in the sample of Gogoli Street from four toxic elements (Cd, Hg, Pb, Cr). Data on Kakhaberi Street and Tamar Mepe Avenue lag behind this. The lowest content is in the sample of Tbel Abuseridze.

The highest content of Hg was found in the sample from Gogoli Street. The concentration of samples from Tamar Mepe Avenue, Kakhabri and Abuseridze streets is constantly lagging behind this. A small amount of Cd was observed in the sample of Gogol Street, and the maximum number was observed in the sample of Kakhabri Street. The indicators of Abuseridze street and Tamar Mepe Avenue lag behind this (Diagram 2).

Table 7

AAS analysis of samples of "sphagnum bag" taken from Gogoli Street in February 2017

Nameofsample	Gogoli Street II/17	22.02.17
Voltage	9 (KV)	
Flow	150 (μA)	
Elements	Concentration (μg / ml)	Showedin %
Mg	12.15	1.31012
Al	25.05	1.16039
Si	386.15	6.49371
P	45.02	0.21922
S	114.61	1.57203
K	549.83129	4.50258
Ca	861.75	6.85041
Ti	8.28889	0.20534
V	0.59966	0
Cr	1.7324	0.0115
Mn	25.49034	0.5285
Fe	469.32176	2.87001

Co	2.13394	0
Ni	2.07586	0.00661
Cu	21.40651	0.0261
Zn	72.42371	0.25331
As	0	0
Zr	17.42527	0.0097
Sr	0	0
Nb	1.04094	0
Mo	0	0
Ag	2.99841	0.0287
Cd	2.33	0
Sn	6.48	0
Sb	0	0
W	2.68817	0
Au	1.21736	0
Pb	11.60655	0.02377
Ba	0.48	0
Hf	0	0
Ta	0	0
Hg	6.64	0
Bi	0.01	0
Th	1.19	0
U	2.81	0
Pt	0	0

Table 8

AAS analysis of samples of "sphagnum bag" taken from Abuseridze Street in February 2017

Name of sample	Abuseridze Street II/17	22.02.17
Voltage	9(KV)	
Flow	150(μA)	
Elements	Concentration (μg / ml)	Showedin %
Mg	7.58	0.80358
Al	14.25	0.63857
Si	216.22	3.95326
P	30.62	0.13001
S	95.12	1.29658
K	464.11445	3.72969
Ca	681.53	4.94823
Ti	4.44268	0.08956
V	0.35094	0
Cr	1.05544	0.00422
Mn	14.77386	0.43991
Fe	223.51748	1.83544
Co	0	0
Ni	2.85868	0.0155
Cu	25.42102	0.05788
Zn	63.65958	0.2138
As	0	0
Zr	6.66704	0.00249

Sr	0	0
Nb	1.1203	0
Mo	0	0
Ag	17.57089	0.04075
Cd	3.13	0
Sn	5.05	0
Sb	0	0
W	0	0
Au	1.98212	0
Pb	6.00955	0.00626
Ba	1.17	0
Hf	2.55377	0
Ta	0	0
Hg	2.32	0
Bi	0.06	0
Th	0.55	0
U	1.08	0
Pt	0	0

Table 9

AAS analysis of samples of "sphagnum bag" taken from Kakhaberi Street in November 2016

Name of sample	Kakhaberi Street II/2017	22.02.2017
Voltage	9 (KV)	
Flow	150 (μA)	
Elements	Concentration (μg / ml)	Showedin %
Mg	12.82	1.38
Al	31.32	1.46334
Si	436.17	7.05388
P	24.38	0.09135
S	64.6	0.86523
K	140.30637	1.10202
Ca	1656.54	15.64724
Ti	10.04552	0.25822
V	1.87537	0
Cr	1.45691	0.00854
Mn	26.59321	0.53583
Fe	795.45773	4.84127
Co	5.75837	0.00277
Ni	2.9255	0.01626
Cu	22.91152	0.03824
Zn	108.43052	0.42954
As	0	0
Zr	40.97229	0.02546
Sr	0	0
Nb	1.2774	0
Mo	0	0

Ag	13.20702	0.02685
Cd	3.43	0
Sn	0	0
Sb	1.78	0.00178
W	1.86786	0
Au	1.69002	0
Pb	8.58129	0.0143
Ba	0	0
Hf	1.98453	0
Ta	0	0
Hg	4.27	0
Bi	0.27	0
Th	0.58	0
U	1.12	0
Pt	0	0

Table 10

AAS analysis of samples of "Sphagnum bag" taken from Tamar Mepe Avenue in February 2017

Name of sample	Tamar Mepe Avenue II/17	22.02.17
Voltage	9 (KV)	
Flow	150 (μA)	
Elements	Concentration (μg / ml)	Showedin %
Mg	9.11	0.97317
Al	22.28	1.02656
Si	298.24	5.37835
P	36.45	0.16613
S	107.86	1.47664
K	410.128	3.24291
Ca	790.66	6.09942
Ti	7.71955	0.1882
V	0.61815	0
Cr	1.35855	0.00748
Mn	20.77285	0.49715
Fe	464.05574	2.83952
Co	1.68813	0
Ni	2.46302	0.01101
Cu	21.67064	0.02823
Zn	68.00236	0.23338
As	2.54088	0.02171
Zr	20.99301	0.01208
Sr	0	0
Nb	1.51753	0

Mo	0	0
Ag	5.62232	0
Cd	3.04	0
Sn	0	0
Sb	3.28	0.00757
W	0	0
Au	1.26287	0
Pb	7.54254	0.01105
Ba	0.35	0
Hf	1.88266	0
Ta	0	0
Hg	5.79	0
Bi	0.15	0
Th	1.13	0
U	1.79	0
Pt	0	0

Diagram 7. Comparison of Cd, Hg, Pb, Cr study results in samples taken at different sites in February 2017

5.3. The Result of the Study, Summer 2017

A range of 17 common heavy metals was detected at all four test sites as a result of the analysis of the samples of "sphagnum bags" taken in May 2017. Only in the sample of Abuseridze Street, one more element was observed - As.

Kakhaberi stree sanples 3,53 (μg/ml), 0,3(μg/ml)-less thet at Tamar Mefe av 3,23 (μg/ml), at Gogoli street - 2,73 (μg/ml) and less concentration was at TbelAbuseridze street 2,071 (μg/ml).From four toxic element (Cd, Hg, Pb, Cr), high concentration of Cd was fixed at Tamar Mefe av 3,87 (μg/ml), less the 0,43(μg/ml)- at street - 3,44 (μg/ml), Tbel Abuseridze -

2,88 (μg/ml) and at Gogolistre2,05189 (μg/ml).The highest contents of Hg (5.99552 μg / ml) and Pb (10.274545 μg / ml) were observed in Gogoli street sample, and Cr with the highest concentration (1.66566 μg / ml) was detected in the samples of Kakhaberi site in the spring of 2017. It should be noted that the lead content was found to be low in areas with high levels of ventilation, such as Tamar Mepe Avenue and Kakhaberi Street (Table 9, 10, 11, 12 and Diagram 3).

Table 11

AAS analysis of samples of "sphagnum bag" taken from Gogoli Street in May 2017

Name of sample	Gogoli Street V/17	23.05.17
Voltage	9 (KV)	
Flow	150 (μA)	
Elements	Concentration (μg / ml)	Showedin %
Mg	11.23	1.20815
Al	33.24	1.55611
Si	514.67	8.35261
P	44.16	0.21389
S	72.59	0.97816
K	410.53238	3.24656
Ca	1307.67	11.56105
Ti	10.21513	0.26333
V	1.25011	0
Cr	1.47466	0.00873
Mn	23.08947	0.51255
Fe	590.66873	3.57266
Co	2.30102	0
Ni	2.52201	0.01168
Cu	19.43935	0.01022
Zn	74.67239	0.26345
As	0	0
Zr	30.96316	0.01876
Sr	0	0
Nb	1.30603	0
Mo	1.28303	0
Ag	11.14667	0.01964
Cd	2.05189	0
Sn	1.20111	0
Sb	0	0
W	1.90506	0
Au	0.83094	0
Pb	10.27545	0.00196

Ba	0.18017	0
Hf	0	0
Ta	0	0
Hg	5.99552	0
Bi	0.1101	0
Th	0.74068	0
U	1.80166	0
Pt	0	0

Table 12

AAS analysis of samples of "sphagnum bag" installed on Abuseridze Street in May 2017

Name of sample	Abuseridze Street V/17	23.05.17
Voltage	9 (KV)	
Flow	150 (μA)	
Elements	Concentration (μg / ml)	Showedin %
Mg	10.55	1.13278
Al	32.96	1.54258
Si	502.76	8.15556
P	45.55	0.2225
S	71.93	0.96883
K	309.36477	2.33435
Ca	1427.01	12.95455
Ti	9.34817	0.23723
V	0.7157	0
Cr	1.27048	0.00654
Mn	22.59063	0.50923
Fe	607.69863	3.67127
Co	2.25084	0
Ni	3.30347	0.02056
Cu	19.28735	0.00899
Zn	94.69748	0.36195
As	1.64776	0.00257
Zr	26.62365	0.01585
Sr	0	0
Nb	2.41663	0
Mo	0	0
Ag	11.5266	0.02116
Cd	2.88	0
Sn	3.13	0
Sb	0	0
W	1.663	0
Au	0.84653	0

Pb	8.01699	0.01254
Ba	0.88	0
Hf	0	0
Ta	0	0
Hg	4.59	0
Bi	0.1	0
Th	0.51	0
U	1.25	0
Pt	0	0

Table 13

AAS analysis of samples of "sphagnum bag" installed on Kakhaberi Street in May 2017

Name of sample	Kakhaberi Street V/2017	23.05.17
Voltage	9 (KV)	
Flow	150 (μA)	
Elements	Concentration (μg / ml)	Showedin %
Mg	5.35	0.55641
Al	25.01	1.15846
Si	310.69	5.57358
P	17.81	0.05984
S	49	0.64476
K	158.15134	1.2303
Ca	1127.35	9.65617
Ti	8.74748	0.21915
V	0.80964	0
Cr	1.66566	0.01079
Mn	15.72373	0.46042
Fe	523.75434	3.18519
Co	2.80884	0
Ni	2.723	0.01396
Cu	20.62267	0.01977
Zn	79.9947	0.28959
As	0	0
Zr	12.05912	0.0061
Sr	0	0
Nb	0.8865	0
Mo	0	0
Ag	12.38261	0.02422
Cd	3.44	0
Sn	1.73	0
Sb	0.13	0
W	2.21892	0
Au	1.13458	0

Pb	4.99529	0.00308
Ba	1.79	0
Hf	2.41716	0
Ta	0	0
Hg	2.34	0
Bi	0.05	0
Th	0.34	0
U	0.32	0
Pt	0	0

Table 14

AAS analysis of samples of "Sphagnum bag" installed on Tamar Mepe Avenue in May 2017

Name of sample	Tamar Mepe Avenue V/17	23.05.17
Voltage	9 (KV)	
Flow	150 (μA)	
Elements	Concentration (μg / ml)	Showedin %
Mg	9.81	1.05076
Al	14.04	0.62842
Si	227.53	4.19254
P	31.41	0.1349
S	124.73	1.71506
K	228.99447	1.73953
Ca	1983.4	19.67444
Ti	8.1426	0.20094
V	0.51529	0
Cr	2.01919	0.01459
Mn	15.03135	0.44547
Fe	309.44783	2.14022
Co	2.1727	0
Ni	3.20255	0.01396
Cu	21.469	0.0266
Zn	60.2086	0.19825
As	0	0
Zr	16.69807	0.00921
Sr	0	0
Nb	0.74274	0
Mo	0	0
Ag	27.72784	0.07309
Cd	3.87	0
Sn	2.57	0
Sb	0.09	0
W	0	0
Au	1.83091	0

Pb	4.6365	0.00196
Ba	0	0
Hf	1.87654	0
Ta	0	0
Hg	1.93	0
Bi	0.07	0
Th	0.24	0
U	0.68	0
Pt	0	0

Diagram 8. Comparison of the results of studies of Cd, Hg, Pb, Cr in samples taken at different sites in May 2017

5.4 The Result of the Summer Research in 2017

As a result of the analysis of the "sphagnum bag" taken from the test sites in August 2017, a spectrum of 18 heavy metals was detected in three sites, and as a result of the analysis of Kahaberi street, less than one element was detected.

From four toxic element (Cd, Hg, Pb, Cr), high concentration of Cd was fixed at Kakhaberi stree sanples 3,53 (μg/ml), 0,3(μg/ml)-less thet at Tamar Mefe av 3,23 (μg/ml), at Gogoli street - 2,73 (μg/ml) and less concentration was at TbelAbuseridze street 2,071 (μg/ml).The highest concentrations of Hg (4.23 μg / ml) were found in the samples of Tamar Mepe Avenue, Cr (2,7532 μg / ml) in the samples of Abuseridze Street, and Pb (10,27545μg / ml) in the samples of Abuseridze (Table 13,14,15, 16 and Diagram 4).

Table 15

Results of AAS analysis of "sphagnum bags" installed on Gogoli Street

Name of sample	Gogoli Street VIII/17	25.08.17
Voltage	9 (KV)	

Flow	150 (μA)	
Elements	Concentration (μg / ml)	Showedin %
Mg	21,36	2,29588
Al	14.4	0.64582
Si	350,14	6,19222
P	33,37	0,14704
S	112,85	1,54716
K	533,3534	4,35401
Ca	779,71	5,98375
Ti	6,09387	0,13926
V	1,02824	0
Cr	1,12709	0,00499
Mn	24,38977	0.52119
Fe	279,37113	2,03355
Co	0,53224	0
Ni	1,98363	0,00556
Cu	21,5092	0.02692
Zn	55,32871	0,17625
As	1,79901	0,00581
Zr	10,20922	0,00486
Sr	0	0
Nb	1,27055	0
Mo	0	0
Ag	9,88667	0,0146
Cd	2,73	0
Sn	0,57	0
Sb	0,47	0
W	0	0
Au	1,87413	0
Pb	7,91705	0,01222
Ba	0	0
Hf	0	0
Ta	0	0
Hg	4,02	0
Bi	0,15	0
Th	0.87	0
U	2,33	0
Pt	0	0

Table 16

Results of AAS analysis of "sphagnum bags" installed on TbelAbuseridze Street

Name of sample	TbelAbuseridze Street VIII/17	25.08.17
Voltage	9 (KV)	
Flow	150 (μA)	
Elements	Concentration (μg / ml)	Showedin %
Mg	12,23	1,31448
Al	11,35	0.50848
Si	223	3,94264
P	31.52	0,13868
S	110,85	1,51864
K	415,68	3,39194

Ca	680,27	5,21767
Ti	5,2691	0,12039
V	0,02321	0
Cr	2,7532	0,01211
Mn	25,6845	0.54862
Fe	348,4231	2,533
Co	0,42311	0
Ni	1,23568	0,00345
Cu	21,5092	0.02692
Zn	62,2458	0,19794
As	2,0237	0,00651
Zr	12,2457	0,00582
Sr	0	0
Nb	1,22012	0
Mo	0	0
Ag	8,8578	0,01307
Cd	2,071	0
Sn	0,42	0
Sb	0,71	0
W	0	0
Au	1,32871	0
Pb	10,2578	0,01579
Ba	0	0
Hf	0	0
Ta	0	0
Hg	3,76	0
Bi	0	0
Th	0.78	0
U	1,64	0
Pt	0	0

Table 17

Results of AAS analysis of "sphagnum bags" installed on Kakhaberi Street

Name of sample	Kakhaberi Street VIII/17	25.08.17
Voltage	9 (KV)	
Flow	150 (μA)	
Elements	Concentration (μg / ml)	Showedin %
Mg	6,28	0,66568
Al	20,55	0.92146
Si	320,69	5,75297
P	18,9	0,63502
S	52,2	0,68686
K	221,2574	1,72120
Ca	1215,23	10,4088
Ti	7,8341	0,19626
V	0,702362	0
Cr	1,502311	0,009731
Mn	17,72321	0,51896
Fe	507,7234	3,08769
Co	2,51832	0
Ni	1,97263	0,00545

Cu	19,23441	0,00958
Zn	80,6792	0,29206
As	0	0
Zr	10,20922	0,00486
Sr	0	0
Nb	1,14023	0
Mo	0	0
Ag	11,23861	0,02198
Cd	3,53	0
Sn	1,22	0
Sb	0,17	0
W	1,29127	0
Au	1,12145	0
Pb	7,78705	0,00480
Ba	1,24	0
Hf	0	0
Ta	0	0
Hg	3,05	0
Bi	0,11	0
Th	0.23	0
U	0,33	0
Pt	0	0

Table 18

Results of AAS analysis of "sphagnum bags" installed on Tamar Mepe Avenue

Name of sample	Tamar Mepe Avenue VIII/17	25.08.17
Voltage	9 (KV)	
Flow	150 (μA)	
Elements	Concentration (μg / ml)	Showedin %
Mg	12,77	1,37898
Al	32,31	1,50959
Si	422,11	6,82649
P	31.52	0,13868
S	57,23	0,76651
K	135,30678	1,06275
Ca	1780,23	16,8155
Ti	8,03521	0,20654
V	1,88237	0
Cr	1,55247	0,0091
Mn	24,6835	0.49735
Fe	882,44721	5,3707
Co	4,23137	0,002
Ni	1,2245	0,0068
Cu	21,5092	0.02692
Zn	102,2218	0,40494
As	0	0
Zr	39,23147	0,02437
Sr	0	0
Nb	1,20012	0
Mo	0	0

Ag	8,8578	0,01307
Cd	3,23	0
Sn	0	0
Sb	0,52	0
W	0	0
Au	1,42131	0
Pb	8,87259	0,01478
Ba	0	0
Hf	1,23543	0
Ta	0	0
Hg	4,23	0
Bi	0,24	0
Th	0.52	0
U	1,17	0
Pt	0	0

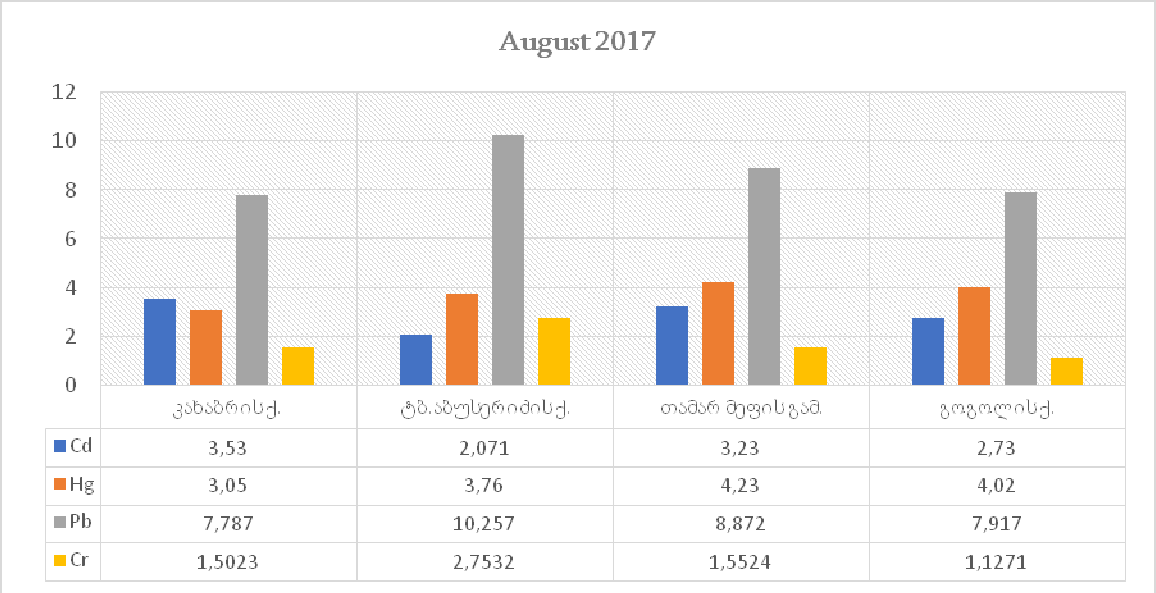

Diagram 9.Comparison of the survey results of Cd, Hg, Pb, Cr in samples taken from different sites in August 2017

To create an overall picture, we calculated the total concentrations of the four most toxic elements in the samples taken from the study area and included them in Diagram 9 for comparison.

From the diagram 10 it is visible that content of Cd is almost constant, nut little bit upper them content from Outumn and Springყველა The maximum rate of Hg was observed in winter, relatively less rate in autumn. It has the lowest rate in spring and summer. Pic of highest content in Outumn of Pb, lowest in Spring.Cr was detected at similar concentrations for three seasons, with only significantly lower rates observed in winter.

The following concentrations of heavy metals were detected in the samples taken in February 2017: Pb (8.43 μg / ml), Cd (2,98 μg / ml), Cr (1,4 μg / ml) Hg (4,75 μg / ml). It should be noted that the highest concentrations of mercury were observed in winter.

Samples taken in May 2017 showed the lowest levels of heavy metals: Pb (6.98 μg / ml), Cd (3.06 μg / ml), Cr (1.61 μg / ml), Hg (3.71 μg / ml). In May, the average monthly precipitation was 74 (mm).

In August 2017, the following values of heavy metals were found in the samples: Pb (8.71 μg / ml), Cd (2.89 μg / ml), Cr (1.73 μg / ml), Hg (3, 76 μg / ml) (Diagram 10).

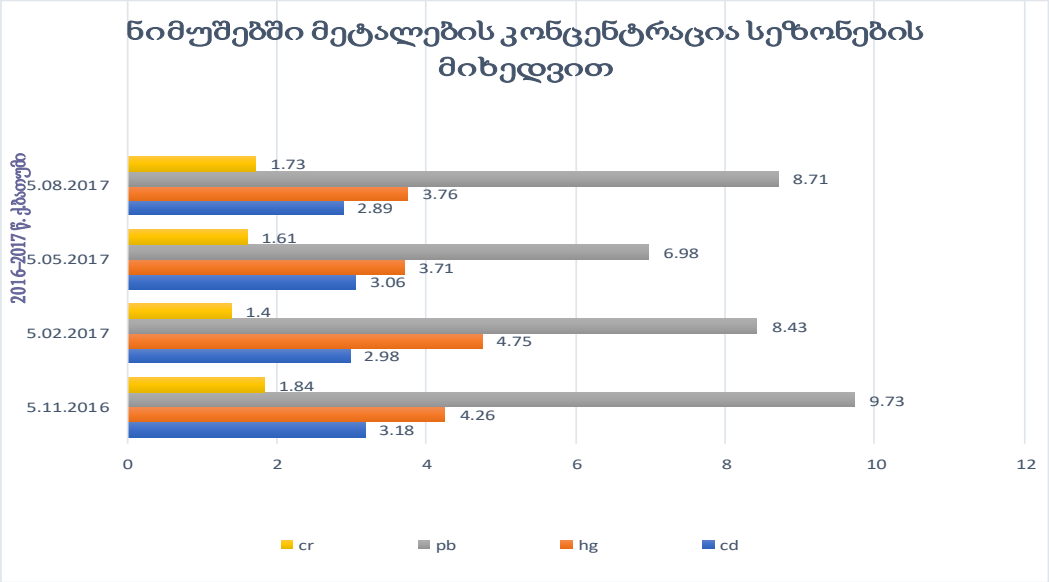

Diagram 10. Concentration of elements (Cd, Hg, Pb, Cr)by seasons

5.3. Comparison of the results obtained with the maximum allowable concentration (MAC)

It should be noted that according to the law of Environmental Quality Standards approved by the decree of the Ministry of Labour, Health and Social Affairs of Georgia (Decrees Nos. 38/N of 24.02.2003), the average daily maximum concentration of cadmium (Cd) is 0.0003 (mg / m³) (hazard class 1), mercury (Hg) - 0.0003 (mg / m³) (hazard class 1), lead (Pb) - 0.0003 (mg / m³) (hazard class 1), chromium (Cr) -0, 0015 (mg / m³) (hazard class 1). If we convert the results of the study to mg / l, we get the following (Table 18)

Table 19

The results of the study converted to mg / l

mg / l				
Metals / Period	Cr	Pb	Hg	Cd
2017 VIII	0.00173	0.00871	0.00376	0.00289
2017 V	0.00161	0.00698	0.00371	0.00306
2017 II	0.0014	0.00843	0.00475	0.00298
2016 XI	0.00184	0.00973	0.00426	0.00318

It is known that 1 mg / L = 1 ppm and this data can be converted to mg / m^3 using the following formula developed by the Environmental Protection Agency (EPA) (Terrie et al., 2006) :

Y mg/m³ = (X ppm)(atomic weight) /24.45. Where, ppm - is the metal content expressed in the sample in mg / l, atomic weight - atomic weight of the metal, 24.45 - constant value. Then, using the above formula and taking into account the atomic weight of the respective metals (atomic weight Cr = 51.9961; atomic weight Pb = 207.2; atomic weight Hg = 200 592; atomic weight Cd = 112 411) (Michael et al. , 2013: 11), we obtain the weighted concentration of heavy metals (1 m3), which is as follows (table 19).

Table 20

The results of the study converted to mg/m³

მგ/მ³				
Metals / Duration	Cr	Pb	Hg	Cd
2017 VIII	0.00368	0.0738	0.03085	0.01328
2017 V	0.00342	0.0591	0.03043	0.01406
2017 II	0.00297	0.0714	0.03896	0.01370
2016 XI	0.00391	0.0824	0.03494	0.01462
ზღვ	0.0015	0.0003	0.0003	0.0003

Comparing the results of the study and the maximum allowable concentration, it was found that the content of chromium (Cr) exceeds the maximum allowable concentration for all seasons by an average of 2.33 times, the content of lead (Pb) exceeds 236.26 times, mercury (Hg)) 112, 65 times, cadmium (Cd) 46.38 times (Table 18. Diagram 11.).

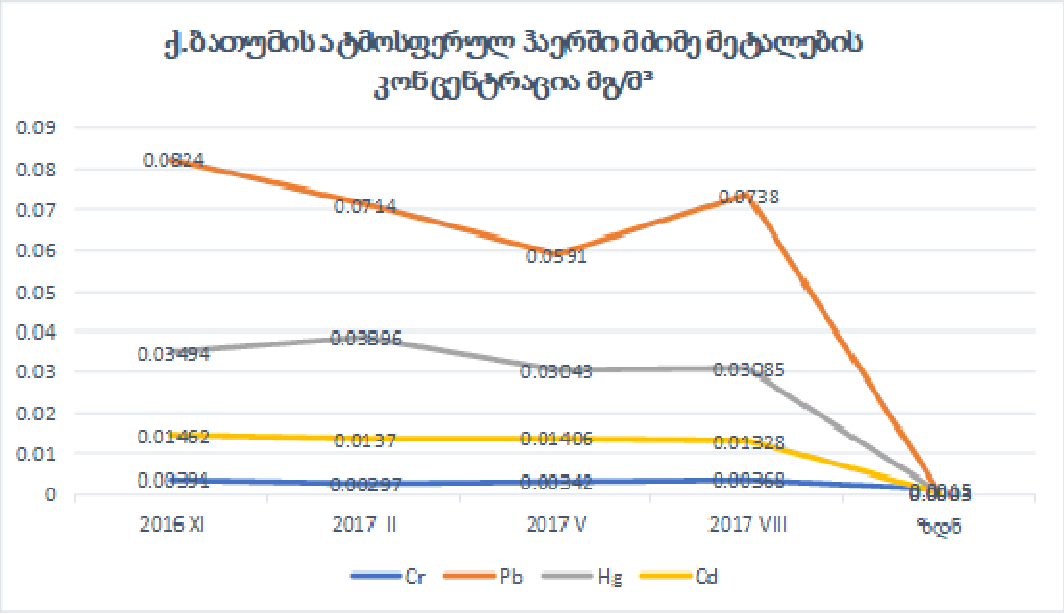

Diagram 11. Comparison of the concentration with the maximum allowable concentration observed during active biomonitoring in the atmospheric air of Batumi in November 2016, February, May and August 2017

Conclusions

According to research:

1. A systematic, anatomical-morphological, bio-ecological and phytochemical study of sphagnum species of the Kolkheti lowland were carried out;

2. Visualization of less visible species signs to the naked eye was performed using a binocular microscope;
3. As a result of the study of the acidity of Imnati and Ispan water, it was found that the pH in the upper layers of water in peatlands is 4.0-5.5, and the electrical conductivity at a depth of 70-100 cm is in the range of 40-70 μS. The obtained results prove that the percolation peatlands of Kolkheti (Imnati, Ispan) are ombrotrophic. In addition, the ecological characteristic "constant acidic aquatic environment" provides an important habitat for the sphagnum peatlands of Kolkheti;
4. The content of biologically active substances in 5 sphagnum species of the percolation bog Ispani 2 of the Kolkheti lowlands has been studied by gas chromatography–mass spectrometry (GC-MS). As a result of research, 16 biologically active substances of 3 classes (esters, phenolic compounds and polysaccharides) were identified;
5. Samples taken from different zones of the peat bog (buffer and dome zones) showed a qualitatively similar composition of esters and polysaccharides. There was a difference in the content of phenolic compounds, namely: the phenolic compound Methyl 2-O-benzyl-á-D-xylopyranoside was found only in Sphagnum cuspidatum, which is adapted to high water levels in the buffer zone;
6. In the area of Batumi, during all four seasons of the year, biomonitoring study of atmospheric air pollution with heavy metals was carried out using the so-called "sphagnum bags" of sphagnum species;
7. Studies have shown that the most favorable average precipitation for adsorption of heavy metals from atmospheric air is 40.81 mm, and the temperature is 11.5 ° C for the "sphagnum bag". During the seasonal survey in Batumi, the highest concentration of heavy metals in the atmospheric air was observed in autumn;
8. According to studies conducted in Batumi air in 2016-2017, the average concentration of chromium (Cr) exceeds the maximum permissible concentration by 2.33 times, the content of lead (Pb) exceeds 236.26 times, mercury (Hg) by 112.65 times cadmium (CD) 46.38 times;
9. The cheapest research method is the study and biomonitoring of atmospheric air pollution by heavy metals using the so-called “sphagnum bag”.

Recommendations:

1. A systematic inventory of sphagnum species is recommended.
2. To obtain biologically active substances (phenols), it is recommended to use Sphagnum cuspidatum, which is common in the buffer zone.
3. It will be recommended to introduce a “sphagnum bag” as a bio-indicator of atmospheric air pollution by heavy metals in other cities and regions where major infrastructure projects are being implemented (for example, Kulevi, Anaklia).

List of Publications:

1. NatelaTetemadze, Alyosha Bakuridze, MalkhazJokhadze, IzoldaMachutadze, 2018, Peculiarities of the composition of acids in Sphagnum species of the percolation bog of the Kolkheti lowland, Annals of Agrarian Sciencevol. 16Npp.222-225https://www.sciencedirect.com/science/article/pii/S1512188718300940

2. Natela TETEMADZE, Izolda MACHUTADZE, Alyosha BAKURIDZE, 2018, Active biomonitoring of air pollution levels using the Bioindicator – Sphagnum Baghttp://rewbc.ni.ac.rs/wp-content/uploads/2016/12/Symposium_programme_2018-Copy.pdfp.61 p561
3. Tetemadze N, Matchutadze I. 2017, Species of Sphagnum as bio indicators of atmospheric air pollution, International Scientific Conference Future Technologies and Quality of Lifep.153**https://tsmu.edu/lifeconference2017/ABSTRACTS.pdf**
4. Tuula Larmola, Eeva-Stiina Tuittila, Marja Tiirola, Hanny Nykanen, Pertti J. Martikainen, Kim Yrjala, Tero Tuomivirta, Hannu Fritze "The role of Sphagnum mosses in the methane cycling of a boreal mire" 2010. Dol:1.1890/09-1343.1 A PUBLICATION OF ECOLOGICAL SOCIETY OF AMERICA.

5. Matchutadze, Tetemadze N., Tsertsvadze A., Tsinaridze M., Memarne Q., Abuladze I., 2017., Colchis –longterm development plan, Conference week "Renewable Resources from Wet and Rewetted Peat lands" abstracts book.

6. Matchutadze I., Goradze R., Goradze I., Tetemadze N., Cheishvili T., Memarne Q., 2020, Unique Habitats of Kolkheti (West Georgia): Threats, Conservation and Wise Use, The 6th International EcoSummit Congress - EcoSummit 2021 – Building a sustainable and desirable future: Adapting to a changing land and sea-scape, will take place at The Gold Coast Convention Centre, Gold Coast, Australia, from 14th – 18th June 2021. https://www.journals.elsevier.com/water-research/conferences/6th-international-ecosummit-congress-ecosummit-2020ElsevierCiteScore: 14.5 **i**Impact Factor: 9.130 **i**Source Normalized Impact per Paper (SNIP): 2.542 SCImago Journal Rank (SJR): 2.932
7. Matchutadze I., Goradze R., Goradze I., Tetemadze N., Cheishvili T., Memarne Q., 2020, Habitat and species biodiversity of Kolkheti Lowland (Georgia), The 6th International EcoSummit Congress - EcoSummit 2021 – Building a sustainable and desirable future: Adapting to a changing land and sea-scape, will take place at The Gold Coast Convention Centre, Gold Coast, Australia, from 14th – 18th June 2021. https://www.journals.elsevier.com/water-research/conferences/6th-international-ecosummit-congress-ecosummit-2020ElsevierCiteScore: 14.5 **i**Impact

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