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The aim of "Annals of Agrarian Science" is to overview problems of the following main disciplines and subjects: Agricultural and Biological Sciences, Biochemistry, Genetics and Molecular Biology, Engineering, Environmental Science. The Journal will publish research papers, review articles, book reviews and conference reports for the above mentioned subjects.

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# Numeral investigation of the dependence of atmospheric pollution of city with a complex relief on the direction of background wind

A. Surmava<sup>a,b</sup>, N. Gigauri<sup>a</sup>, V. Kukhalashvili<sup>b</sup>, L. Intskirveli<sup>a,\*</sup>, D. Demetrashvili<sup>b</sup>

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#### **ABSTRACT**

Dust propagation in the atmospheric air of Tbilisi city in case of western and eastern background light airs was numerically modeled and analyzed using the 3D regional model of the evolution of atmospheric processes and numerical integration of admixtures transfer-diffusion equation. The main differences and similarities appearing under conditions of background winds of both directions typical for the city are explored. The locations of high pollution areas are determined. Modeling made it possible to establish that in case of the eastern background light air the wind velocity field formed under the influence of the relief impedes dust removal from the city and forms over the large area of the city a zone with high pollution with a maximum concentration of 2-2.5 MAC. In case of the western background light air the wind velocity field promotes city atmosphere "self-purification" process and high pollution level is mainly registered during motor transport traffic "rush hour" situation. As a result of the calculations it is established that the dust propagation process conditionally runs by four stages and depends on the motor transport traffic intensity, city mains location and city micro-relief. From 6 AM to 9 AM the rapid growth of concentration takes place, from 9 AM to 6 PM – slight reduction or consistency of concentration, from 6 PM to 9 PM – concentration growth, while from 9 PM to 6 AM self-purification of the city air is observed.

*Keywords:* Atmosphere, Pollution, Numerical modeling, Concentration, Background wind, PM.

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#### INTRODUCTION

Researches [1-3] showed that the mortality rate caused by COVID-19 is relatively high at the urban territories with increased contamination of atmospheric air. As one of the reasons of rise in mortality, one may mention the spreading of COVID-19 virus hitting the dust particles, PM2.5 and PM10 [4, 5]. That is why, during viral pandemic, the study of aerosols propagation in the atmosphere of industrial centers and big cities, air purity protection and carrying-out the measures aimed to ecological safety are of special importance.

Tbilisi is an administrative center of Georgia and one of the major cities of the South Caucasus. While it isn't ranked among 500 cities worldwide mostly polluted by micro particles [6], but according to the data of National Environmental Agency of the Ministry of Environmental Protection and Agriculture of Georgia the concentrations of dust and micro particles often exceed maximum allowable concentrations [7].

Tbilisi city is a key junction point of the Great Silk Road connecting Europe and Asia, and routes connecting Russia with the Asia Minor. Many thousands of light and heavy vehicles pass through a city every day. Hundreds of thousands of cars drive about narrow and complex-shape city streets. There are no atmosphere-polluting large industrial enterprises in the city that's why micro particles emitted from cars and dust raised up from underlying surfaces are the major pollution source. A dust hitting the atmosphere, along with a settled COVID-19 virus transfers throughout a city, accumulates at the particular territories, and creates a situation favorable for spread the hazardous for health infection.

The peculiarities of dust propagation at the territories of cities with complex terrain are explored in the present article. The dependence of atmospheric air pollution by dust on wind directions is studied using numerical modeling. Two main meteorological situations – eastern and western background winds – are considered. The modeling is conducted using the regional model of atmospheric processes in Georgia and numerical integration of equations of admixtures transfer and diffusion [8 -11].

#### STATEMENT OF THE PROBLEM

The 30,6x24 km² area of Tbilisi and surrounding territories is considered. In order to mathematically correctly describe the dynamic fields of atmosphere and meteorological parameters under conditions of complex terrain of the city a relief-following coordinate system (t, x, y, (t, x, y,  $\zeta = (z - \delta)/h$ )) is used. Here t is time, x and y are coordinates directed along parallel and meridian,  $\zeta$  is a vertical non dimensional coordinate,  $\delta(x,y)$  is a relief height above sea level,  $h = H - \delta$  – troposphere thickness, H(t,x,y) – tropopause height. The equation for dust concentration transfer and diffusion in the selected coordinate system will be written in the following form

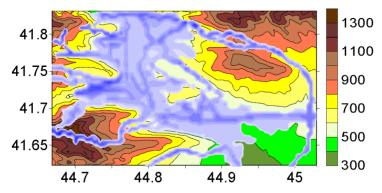
$$\frac{\partial C}{\partial t} + u \frac{\partial C}{\partial x} + v \frac{\partial C}{\partial y} + (\widetilde{w} - \frac{w_0}{h}) \frac{\partial C}{\partial \zeta} = \frac{\partial}{\partial x} \mu \frac{\partial}{\partial x} + \frac{\partial}{\partial y} \mu \frac{\partial}{\partial y} + \frac{1}{h^2} \frac{\partial}{\partial \zeta} v \frac{\partial C}{\partial \zeta} + F, \qquad (1)$$

where, C is concentration of dust; u, v, w and  $\widetilde{w}$  are wind velocity components along the x, y, z and  $\zeta$  axes, w<sub>o</sub> is dust deposition rate, F(t, x, y  $\zeta$ ) the rate of dust dissipation in the atmosphere by the source,  $\mu$  and v are coefficients of horizontal and vertical turbulence. Wind velocity components and coefficients of turbulence in the free atmosphere and surface layer of atmosphere are calculated by means of numerical integration of equations given in [9,10].

Dust propagation in the free atmosphere and surface layers of atmosphere is modeled through numerical integration of equation (1), using respective initial and boundary conditions. Numerical grid steps along the x and y axes equal to 300 and 400 m, and vertical non dimensional step in the free atmosphere is 1/31 and is equal to about 300 m. In the 100 m thick surface layer of atmosphere a vertical step varies from 0.5 to 15 m, while time step is 1 sec.

Calculations are made for 3-day period. The cases of western and eastern background light airs under dry weather conditions of June are considered. Background wind velocity equals to 1 m/sec at 100 m height above the ground, and 20 m/sec – in the tropopause (9 km altitude). Relative atmosphere humidity is 50%.

It is assumed that atmosphere is polluted by a dust originated at city mains and cities due to motor transport traffic. Its quantity changes in time and is determined according to assessment of continuous surveillance materials and transport traffic intensity. In Fig. 1 the relief of Tbilisi is shown. Pollution source distribution is marked in dark blue. They are mainly located at the central city mains and urbanized territories.



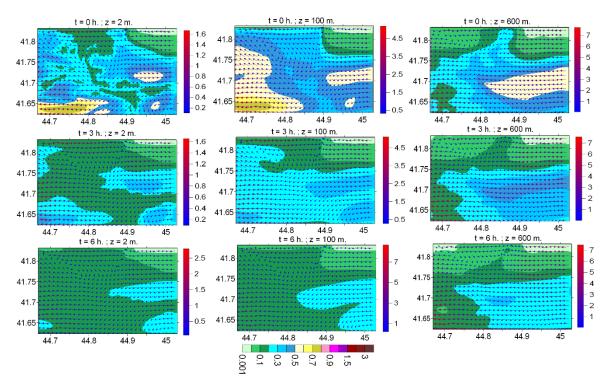
**Fig.1.** *Tbilisi city terrain heights (m) and pollution source distribution (blue zone and lines). Actual geographic coordinates are placed on the axes* 

#### 3. RESULTS OF NUMERICAL MODELING

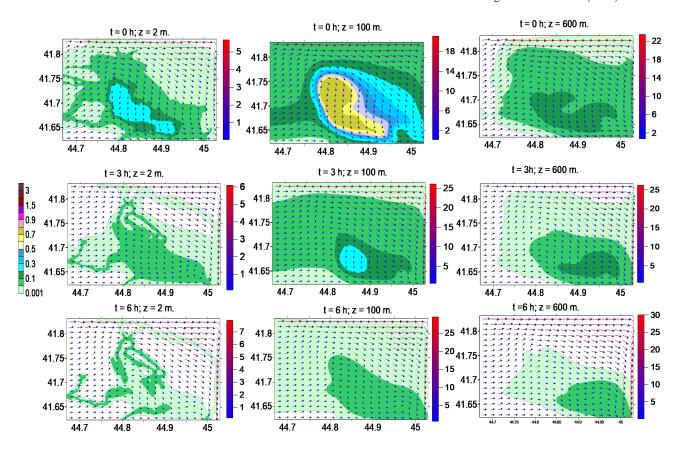
In Fig. 2 and 3 there is shown the spatial distribution of dust concentration and wind velocity at 2, 100 and 600 m heights from the earth surface for t = 0, 3 and 6 h in case of eastern and western background light airs, obtained via calculations. Concentration is given in units of one-off maximum allowable concentration (MAC =  $0.5 \text{ mg/m}^3$ ). It is seen from Fig. 2 and 3 that dust concentration spatial distributions, in case of eastern and western background light airs have both similar properties and significant distinguishing signs. Among similar properties are: concentration values starting with t = 0 h gradually reduce and reach minimum magnitude, when t = 6 h; maximum value of concentration equals to 0.7 MAC and is obtained in the surface layer of atmosphere, when t = 3 h; concentration values at the upper limit of the surface layer of atmosphere (at 100 m height from the earth surface) are higher or equal to those obtained at 2 and 100 m altitudes above the ground; the dust quantity taken out from the modeling area by dynamic and diffusion processes surpasses the quantity dissipated by vehicles, and due to this fact the process of atmosphere self-purification takes place in the time interval from t = 0 to 6 h. Distinguishing signs are: pollution level at the central and densely populated urbanized territories in case of eastern background light air is less than that of peripheral

part. On the contrary, in case of western background light air a maximally polluted zone is obtained at the central and densely populated urbanized territories; the horizontal diffusion process and related dust transfer process are more intense in case of eastern background light air.

The differences in spatial distribution concentration result from the influence of complex terrain. In case of eastern background light air, high mountains located in the western part of modeling area inhibit air flows free movement to the west and impede dust removal from city territory (Fig. 1, Fig. 2). At the same time, the relief generates a localscale anticyclonic swirl, which cause dust relocation from the central part to peripheral areas by means of divergent velocity field. As a result, the dust concentration in the surface layer of the atmosphere and in the vicinity of main pollution sources is less than concentration obtained in the surroundings of recreational zones and sources with less pollution capacity. In case of western background light air there is a lowland territory in the south-eastern part of the city and there are no high orographic obstacles (Fig. 1). At that, the relief causes the wave disturbance of wind velocity only. As a consequence, a dust available in the air easily moves out of main pollution zones, its concentration is getting smaller in the central and peripheral parts of the city and remains slightly high along some city mains (Fig. 3).



**Fig. 2.** Distribution of wind velocity (m/s) and dust concentration (MAC) in case of eastern background light air, when t = 0, 3 and 6 h at 2, 100 and 600 m height from the earth surface

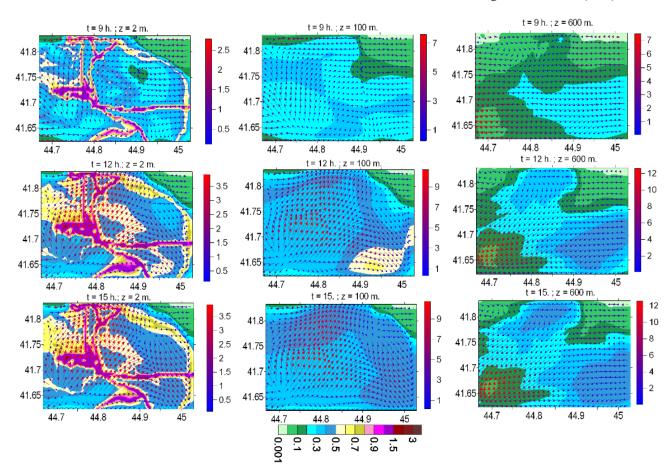


**Fig. 3**. Distribution of wind velocity (m/s) and dust concentration (MAC) in case of western background light air, when t = 0, 3 and 6 h at 2, 100 and 600 m height from the earth surface

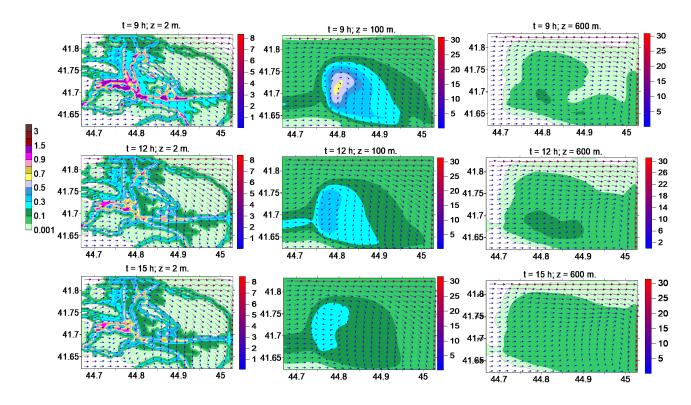
In the process of numerical modeling the intensity of transport traffic is changed during day: from 6 AM to 9 AM the motor transport traffic intensity increases, from 9 AM to 11 AM intensity is constant and corresponds to the first "rush-hour" situation, from 11 AM to 5 PM period, traffic intensity slightly reduces. Along with traffic intensity change the quantity of dust hitting the atmosphere changes, as well, and this dust extends over the whole city and its adjusting territories by means of advective, convective and turbulent diffusive mechanisms. Dust spatial distributions obtained in this period through modeling in case of eastern and western light airs are shown in Fig. 4 and 5, respectively. It is clearly seen from the figures that in the lower part of the atmospheric boundary layer the dust propagation process has a qualitative difference. In case of western background wind, the mountains encircling the city from three sides, impede free movement of background flow to the west and together with underlying surface thermobaric field change develop local convergence and divergence zones of wind velocity, which are distributed in such a way that form the enclosed area of motion.

Respectively, the dust taking-out from city territories inhibits and the level of city pollution increases (Fig. 4). Concentration growth takes place up to the midday. In the interval of t=12-15 h a quasistationary state of the dust concentration is established. This time the concentration is high at main highways of every district of the city and its value varies within a range of 1-1.5 MAC. The level of pollution at the urbanized territory adjacent to the central part of the city is within 0.5-0.8 MAC in average.

Free taking-out of dust takes place in case of western background wind. As a result, from 6 AM to 9 AM time interval, on separate sections of highways located in the central and south-eastern parts of the city a relatively slight growth of dust concentration is obtained (Fig. 5). After 9 AM the level of city pollution drops until the second "rush-hour" (6 PM). From the midday to 6 PM the dust pollution level is non-uniformly distributed and 1 MAC concentration is obtained in the surroundings of several crossroads. Dust pollution level at 100 and 600 m height above the ground increases together with the rise of ground-level dust concentration.



**Fig. 4.** Distribution of wind velocity (m/s) and dust concentration (MAC) in case of eastern background light air, when t = 9, 12 and 15 h at 2, 100 and 600 m height from the earth surface

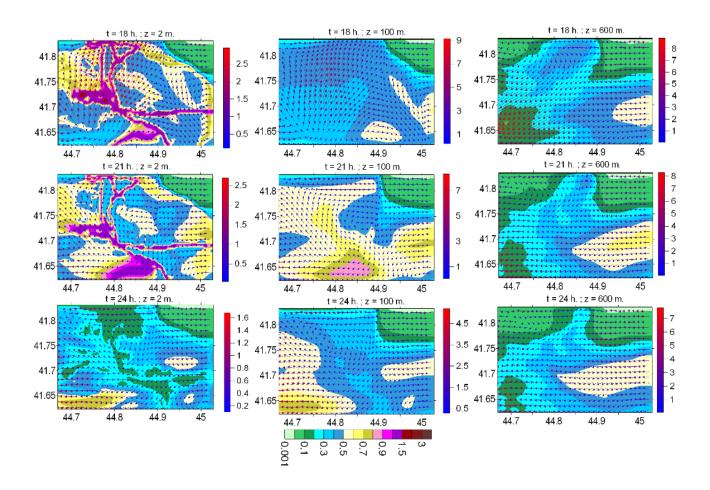


**Fig. 5.** Distribution of wind velocity (m/s) and dust concentration (MAC) in case of western background light air, when t = 9, 12 and 15 h at 2, 100 and 600 m height from the earth surface

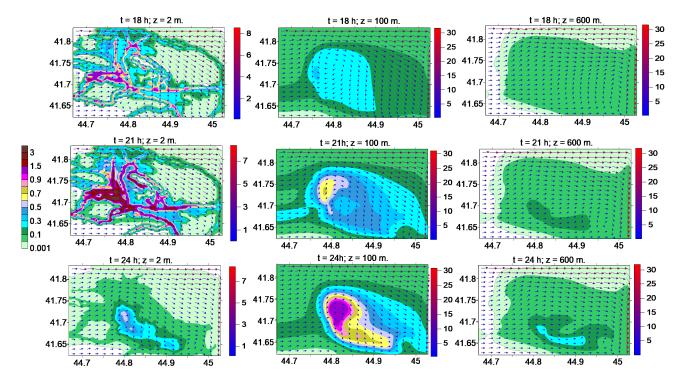
In case of eastern background wind, in the second half of the day, from 6 PM to 9 PM, despite the second "rush-hour" situation with motor transport traffic, a quasistationary distribution of pollution is settled at 2 m height from the earth surface (Fig. 6). Wherein, there is no significant change in values of concentration, however partial extension and deformation of pollution zones takes place. At that, the "excessive" dust emitted during "rush-hour" situation extends towards the upper part of surface layer of the atmosphere that causes a substantial increase of concentration at 100 and 600 m height when t = 21h. Maximum values of concentration in the city center, in the southern part of modeling area

are within a limit of 1.5-2.5 MAC. At 100 m height, the concentration is roughly equal to 1 MAC in the southern part of the city.

The dust propagation process in case of western background light air runs differently. The impact of "rush-hour" situation on atmosphere pollution becomes appreciable in the second part of a day. Calculations show that starting with 6 PM the concentration growth takes place not only in the central part of the city, but at the peripheral territories as well (Fig. 7). By 9 PM, the concentration at 2 m height from the earth surface reaches 2 MAC and afterwards it starts to reduce.



**Fig. 6.** Distribution of wind velocity (m/s) and dust concentration (MAC) in case of eastern background light air, when t = 18, 21 and 24 h at 2, 100 and 600 m height from the earth surface



**Fig. 7.** Distribution of wind velocity (m/s) and dust concentration (MAC) in case of western background light air, when t = 18, 21 and 24 h at 2, 100 and 600 m height from the earth surface

The processes of dust vertical transfer in the surface layer of the atmosphere are different, as well. In case of eastern background light air, the dust concentration change occurs synchronously at 2 and 100 m height, while during western background wind this process runs with 3-hour phase delay.

#### **CONCLUSION**

Change of kinetics of distribution of the dust generated by motor transport at the territory of Tbilisi is explored using numerical modeling for main two meteorological situations: in case of eastern and western background light airs. Daily change of dust spatial distribution is studied. Via modeling there are obtained concentration values, which are within a limit of magnitudes obtained via routine observations. Through analysis of the fields of wind velocity and dust concentration there is established that the spatial distribution of heavily polluted areas depends on the motor transport traffic intensity and city mains disposition, on one hand and on local circulation systems formed through diurnal temperature variation in the surface layer of the atmosphere.

There are explored those main differences and similar properties that are originated in case of background winds of both direction peculiar for the city. By means of modeling there is established that in case of eastern background light air the wind velocity field formed under the influence of terrain impedes the process of dust taking-out from the city and creates the area of high dust pollution level at the large territory of the city with 2-2.5 MAC maximum concentration. As for western background light air, the wind velocity field promotes the "selfpurification" process of city atmosphere and high level of dust pollution is registered during "rushhour" situation with motor transport traffic. At that, in a period of the second "rush-hour" situation the concentration significantly increases in the central part of the city and in the surroundings of connecting avenues.

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# Growing technology for soybeans with nanoherbicides

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#### ABSTRACT

Modern herbicide market in agriculture is about 2 billion tons and about 73 billion dollars industry with sophisticated multi-impact problems with food safety and human health, with increasing of weed resistance with every passing year at the topmost. Nanoherbicides under development in the current decade of our century could be a new strategy to address all the issues caused by the conventional non-nanoherbicides. From the beginning of 21 century group of Georgian scientists with farmers associations have begun development nanoherbicides (experimental name "Nanocooper 076", which is under registration) in soybean experimental pilot plots and farmer's fields, which will allow farmers to clear their soybean plantings from weeds without using toxic chemicals, like Glyphosate. As the potential use of nanostructured nanomaterials enables the use of nanoherbicides effectively and rules out the emergence of various weed-resistant population at an early stage of growing agricultural crops (first weeks after sowing), these very desirable nano technological methods and practices in general agriculture are reviewed by this article.

*Keywords:* Soya seed pilling, Nanocooper 076, Soil contamination, Friendly nanotechnologies, Nanoherbicides, Agriculture.

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#### Introduction

Recently definition of nanoherbicides in modern scientific dictionaries means like this – Nano herbicides also commonly known as weed killers, with chemical substances with various ingredients with nano metal composites, used to control unwanted plants, mainly weeds. Our nanoherbicide (Nanocooper 076) were synthesized and evaluated for herbicidal activity and cytotoxicity for the using in soybeans. The optimum formulation of nanoparticles was obtained using the cooper nano-technological composite. The basic properties - means particle size, stability time, morphology, and interaction between heavy metals (above 5 g/sm³) and herbicide were characterized using a particle size analyzer in

close labs. The nanoparticles were found to be in size range approximately 10-30 nm with low zeta potential value [1].

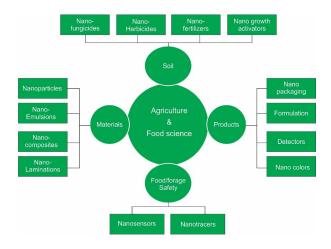
The genetically acquired capacity of the weed population to survive herbicide exposure under normal usage conditions could be stated as herbicide resistance. In a population of weeds exposed to herbicide, only a few individuals develop resistance, while the rest dies due to the herbicide action. This set resistant weed that survives eventually becomes a population of weeds with acquired resistance to a particular herbicide. The uncontrolled and repeated application of same herbicide will also select resistance plants. In some cases, multiple resistances can also appear due to sequential selection. Over the globe, nearly 250 herbicide-resistant weedy bio-

types have been identified in over 50 countries. This number constantly grows on an annual basis giving rise to new resistant weeds. Likely, some management practices also give a rise to the development of herbicide-resistant weeds [2,3].

The modern nanoherbicides has the potential to increase productivity- yield of field crops and guaranty food safety, while resolving the drawbacks of conventional pesticides and agrochemicals, which have negative environmental impacts [4,5].

Nanotechnology with his nanoherbicides offers exciting ways for averting the herbicide overuse and also a safe and effectual delivery. The usage of nanostructured systems in agriculture has increased tremendously in the current era for the controlled release of agrochemicals as well for plant nutrients (Fig. 1).

The nanostructured herbicide could substantially reduce the herbicide consumption rate 2-3 times and promise increased field crops productivity. This technology of exploiting nanomaterials guarantees to improve the current agricultural practices via the enrichment of field management methods. Nanoherbicides are one of the new-fangled strategies for combating the problems of conventional herbicides. These are being developed for addressing the issues in annual weed management and also for fatiguing the weed seed collection. The nanostructured formulation performs action through controlled release mechanism. The nanoherbicides comprise a wide range of entities such as polymeric and metallic nanoparticles. Nanoherbicides require a glance in order to place nanotechnology at the premier level [6].



**Fig. 1.** Applications of nanoherbicide in agriculture & food/forage

Modern nanotechnology has potential for efficient delivery of chemical and biological pesticides using nano-sized preparations or nanomaterials (in our case Nanocooper 076) based agrochemical formulations. The active ingredient is adsorbed, attached, encapsulated or entrapped unto or into the direct nano-matrix. Controlled release of the active ingredient is achieved due to the slow release characteristics of the Nanocooper 076, bonding of the other ingredients to the material and the soil and climate as well as whole of environmental conditions for future generations [7].

#### **Results and Discussion**

Influence of Nanocooper 076 efficiencies on soybean yield and food safety positive parameters, as well as main use for soybeans - weed control with using seed pilling technology during last 12 years field and lab research results showed that application of herbicide-loaded Nanocooper 076 particles could be used to reduce the use of herbicides with improved efficacy and ecological environmental protection and food safety. During 2007 to 2018 in labs and farmer's fields with very close cooperation of researchers from Georgian Agricultural University in Tbilisi and the University of Maryland in USA, we are collaborating on a project to research and develop a nanotechnology-based herbicide that would prevent weeds from germinating and starting period of time during first 5 leaves faze growing of soybeans in West Georgia region in 14 hectares [8,9].

Preliminary lab analysis proves that the benefits of such technologies – seed pilling + Nanocooper 076, based on formulations of improvement of efficacy due to higher surface area, higher solubility of herbicide and seed pills with mineral fertilizers, higher mobility and lower toxicity due to elimination of organic and mineral solvents. Our nanopesticide involve either very small particles of pesticidal active ingredients of cooper or other small engineered structure with useful active pesticide properties. Our nanoherbicide show that it can increase the dispersion and wettability of agricultural formulations and unwanted pesticide movement, it's working very next day [10,11].

Nanocooper 076 exhibit useful properties such as stiffness, thermal stability, solubility, permeability, crystallite stability and biodegradability needed for formulating nanopesticide. It can also offer large

specific surface area and hence increased affinity to the target, it's very useful for the application by small size drones or airplane and need law expenses – in our pilot plots for 14 ha only \$784.

The use of nanopesticid copper 076 may offer new ways to control of these biological agents. nanopesticides or fungal bio control agents are promising as they act by contact and do not need ingestion, can be easily mess produced, and are relatively specific. Microbial products such as enzymes, inhibitor, antibiotics and toxins are also promising as biopesticides against plant pests and pathogens. However, microbial products need stabilization and directed delivery mechanism towards identified targets, which need a very huge scientific research in collaboration with many biological and chemical specialists [12].

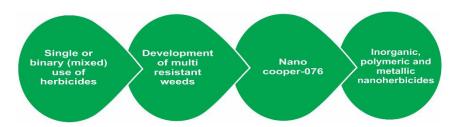


Fig. 2. Evolution of "Nano cooper 076"

Advancements in nanotechnology could be boon for mitigating the unsolvable herbicide resistance prevailing for centuries (Fig. 2). The high penetration efficiency of nanoherbicides helps in eliminating the weeds before resistance could develop [13].

Nanocooper 076 was formulated by exploiting the nano technological potential for effectual delivery of chemical pesticides with the help of nanosized preparations or nanomaterials-based herbicide formulations. Nanomaterials or nanostructures materials-based formulations could improve the efficacy of the herbicide in economically too, enhance the solubility and reduce the toxicity in comparison with well-known conventional herbicides on the base of Glyphosate (C<sub>3</sub>H<sub>8</sub>NO<sub>5</sub>P), which in the nearest future will be prohibited [14].

Weed control in soybeans with the use of nanoparticle-based herbicide Nanocooper 076 release systems could reduce the herbicide resistance potential, maintain the activity of the active ingredient and prolong their release over a longer period of time (2-3 years). The development of such specific herbicide molecule encapsulated with nanoparticle aims at specific receptors present at the root of the weed, especially in the spring before planting and during germination of soybeans. The developed Nanocooper 076 enters the root system of the weed and gets translocated to perform its action which in turn inhibits the glycolysis of the plant root system, while they are small and sick. The targeted action creates starvation of the plant and thus kills it. This Nanocooper 076 could also be used in rain-fed areas, where conventional herbicides get dissipated

through misty due to insufficient soil moisture (East Georgia). With the help of controlled release of Nanocooper 076 via encapsulation, approximately 90% weeds can be utterly destroyed with their formulated seeds.

Due to renewal of conventional herbicides with every passing year, the contaminant elements in soybeans increased by 0,3%. As a result, reducing the contamination of soils and correspondently soybeans by represented technology estimated by 0,04 per year. In constructed crop rotation with 4 fields of legumes, calculated on a century, using nanoherbisides significantly decrease contamination of soil: in wheat by 88 %, in alfalfa 3 years fields by 74%, in soybeans by 94%. The notion of proposed technology has to introduce efficient of nanoherbicedes in such rotations [15,16].

Additional successful new by point of view of food safety is that proposed new friendly technology used in our researches is the inoculation of soybean seed by the method of seed pilling [patent # 1180 GE], ensures considerable economy of micro and macro mineral fertilizers as well as Rhizobium bacteria treatment material. Technology protect the environment from its pollution, ecologically pure and safe production of legumes and later on the same land plots, high output of other crops, which are sown after those legumes and favorably use biological nitrogen fixed by legume crops. It's one of strong way for intensive accumulation of biological nitrogen in soil results in heightening of its fertility and growth of output of grain legumes by 4% and that of legume grasses by 17% (3 years stand). With

this technology is very effective using of nanoherbicide - Nanocooper 076, which practically do not contaminated soils by chemicals and adsorbed during calculate time of period (the century).

Table below presents the data on contamination, efficiency of the above-described technologies, clearly proving its profitableness by point of the view of soil contamination. Application of the technologies elaborated in grain legumes crop farm economies, irrespective of their small territories, proved that traditional technologies used in growing of these cultures cannot compete, even slightly, with scientific achievements, especially if we consider the indices, such as net income and environmental protection value with maximally of food safety.

Especially very well were matters in farm economies distributed in the arid zone of East Georgia, where we planted those crops using the technology of seed pilling with nanoherbicides.

**Table.** Contamination of soils conditionally for 100 years of different crops growing

| Crop Sequence (Rotation)           | Contamination of topsoil (0-30 cm) conditionally remaining (conventional herbicides / new technologies of seed pilling + Nanocooper 076 *), kg/ha |  |  |  |
|------------------------------------|---|--|--|--|
| Continuous Wheat                   | 689.56/84.31*   |  |  |  |
| Continuous Alfalfa (3 years stand) | 239.92/63.15*   |  |  |  |
| Continuous Soybean                 | 532.36/32.28*   |  |  |  |
|                                    |   |  |  |  |

<sup>\*</sup>Wheat, Alfalfa (3 years stand), Soybean (All grown by new technologies of seed pilling and "Nanocooper 076") during 5 years field rotation

In these regions, on the pilot plots of farmers' households, at about 4.2 metric t/ha soybeans (with irrigation), 2.6 t/ha lentils, and 4.1 t/ha faba bean was obtained on the small trials, while in West Georgia, in the humid zone (with drainage) soybeans on pilot plots reached 3.8 tons grain per ha without additional mineral fertilizers. The fact is to be emphasized that the advantage of the elaborated technologies (seed pilling with Nanocooper 076) were so apparent and reliable that they found great popularity among farmers and local governmental authorities during field days [17].

#### Conclusion

Field and lab scientific-research work show that from the point of view of soil rehabilitation and environmental protection, we carried out calculation of contamination of topsoil during 1 century, growing by traditional and new technologies (table). Using Nanocooper 076 and new friendly technologies of growing with seed pills have no alternative for

contamination of environment, as in the case of wheat, the difference is 363%, in the case of Alfalfa 235%, and 297% less in the case of soybeans. Joint scientific collective groups must be created in the nearest future, which will exchange the results of scientific-research achievements, will intensify the exchange training of farmers, take active part in advertising meetings of scientists and farmers, symposia and conferences in neighboring countries. All of results is open for any scientists and commercial farmers. Heightening of efficiency of the results of research work is a demand of the day and it must be supported by creation of necessary conditions for strengthening the economies of private farms not only in Georgia, but also in South Caucasus, EU countries, etc. This will be pane the way for successful implementation of the Food Security and Safety Programs of our country on the basis of this soil and environmental protection friendly technologies.

In Georgian agricultural practice for the destroying of typical for soybean weeds, for the growing of this cultural crop, farmers are using conventional herbicide when sprayed has a chance of getting affected to the foods crops too by this and there can be huge loss in the crop yield [18]. By using Nanocooper 076 which is cheap then very released and useful for soy herbicide Pivot on the base of Glyphosate by 32%, will try to mingle with the soil particle and try to destroy the entire weeds from their roots by not affecting soybean.

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#### Vibrocalibration of walnuts

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#### **ABSTRACT**

One of the directions for the development of agricultural product processing is the elaboration of small, energy-saving, high-productivity machinery and equipment. The article discusses a walnut calibration unit equipped with the direct current magnetic electromagnetic reciprocating vibration motor, with the ability to change the number of fractions and adjust the productivity; The trajectory of the walnut movement on the surface of the screen and the scheme of forces acting on it; Analysis of the optimal technological parameters obtained as a result of the study of the working modes of the walnut calibrator.

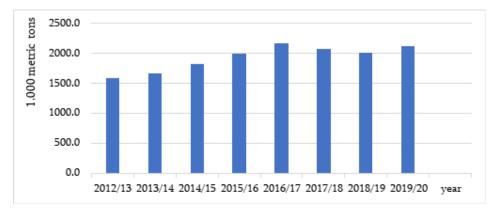
Keywords: Electromagnetic, Vibrating motor, Calibration, Walnut, Vibrating chute, Screen.

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#### Introduction

Walnut cultivation and processing is a highly profitable business. The world walnut market is characterized by high concentration of its production in a limited number of countries [1]. According to Food and Agriculture Organization (FAO), world's total volume of walnut production reached in 2010/2020 year 2,13 million metric tons; China

accounted for about 50% of the total walnut production, but consumes nearly all that it produces, the USA ranks second with above 30%. World exports, dominated by the USA, are expected to rise to 946,000 metric tons and European Union with 300,000 metric tons is the largest walnut import market in the world, representing almost half of all imports [2]. Fig. 1 shows the dynamics of walnut production worldwide in 2012-2020.



**Fig. 1**. Dynamics of walnut production worldwide, shelled, 1000 metric tons [3].

Walnut is considered to be the oldest culture in Georgia. The first information about its spread is found in Greek sources belongs to the VI-IV centuries BC. Walnuts have been present in the local traditional cuisine of Georgia (Satsivi, Churchkhela, Khartcho, Kuchmachi, Bazhe, Kaklis Muraba etc.). There are great prospects for its development – Georgia's soil and climate is favorable for walnut cultivation and the plant is cultivated in every region of the country on the elevation of 500...900 m from see level [4, 5] and the Autonomous Republic of Adjara is a leader in terms of production, followed by Regions of Shida Kartli, Kakheti, and Imereti [6]. Walnut production in Georgia in 2010-2019 is shown in Fig. 2 [7]. However, Georgia currently does not meet its own needs, demand on local market is more than 6 thousand metric tons of the inshell walnut production, so walnuts are imported from other countries in high market prices [8]. Georgia needs to build the capacity of its producers to become more competitive in domestic and export markets and one way to become competitive is to improve walnut production quality, its storage and processing, that is, to go as soon as possible the path that Georgia has passed with the development of its hazelnut production, having become today the second after Turkey exporter to the world market - mainly to the EU countries.

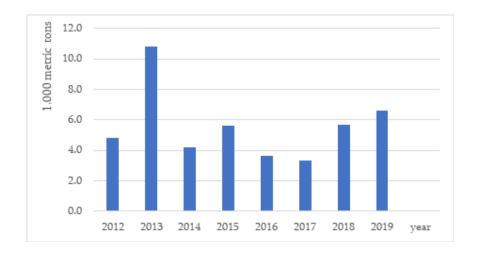


Fig. 2. Walnut production in Georgia in 2012-2019, 1000 metric tons

Recently, commercial development of walnut acreage is ramping, several hundred hectares of walnut orchards have been planted in Georgia and this process continues. Given the pace of orchard cultivation, we must assume that in a few years walnut imports will be reduced to a minimum and export opportunities will emerge, as result the country, which for a long time was a net importer of walnuts, began to dramatically increase the export of this product (Table 1).

| Table 1. Walnuts (HS 080231/in-shell and 080232/shelled) External Trade of Georgia, | 1000 US |
|---|---------|
| Dollars (8)   |         |

| Year          | 2012    | 2013   | 2014   | 2015   | 2016   | 2017    | 2018    | 2019    | 2020   |
|---------------|---------|--------|--------|--------|--------|---------|---------|---------|--------|
| Export        | 57.9    | 80.08  | 50.2   | 42.6   | 2.0    | 224.4   | 361.6   | 1361.9  | 1129.5 |
| Import        | 891.1   | 623.8  | 819.1  | 890.5  | 1830.9 | 4289.7  | 5525.2  | 5424.9  | 1877.2 |
| Trade Balance | - 833.2 | -543.8 | -840.9 | -847.9 | 1828.8 | -4065.3 | -5163.6 | -4063.0 | -747.7 |

To date, almost all varieties of walnuts grown in Georgia are rounded walnuts with thick shells, cultivated only in households and a significant part of the walnut processing is done by hand [4]. This is mainly due to the facts, that modern enterprises manufacture machines for processing walnuts with only thin shells, and when processing thickshelled walnuts, part of the kernel remains in the shell, which reduces the yield. As a result, varieties with thick-shelled nuts are processed only by hand and not in the factory. Sorting is predominantly also done manually to ensure quality and consistency of the product batches. Accordingly, walnut production in Georgia is still mostly a labor-intensive business, with the majority of walnuts harvested by hand or rudimentary nut picking devices in family farms used by the previous generation of growers. And as is well known, small farms with mostly manual labor are characterized by unstable quality of their product, which pushes them into the low-level segment among buyers.

There are a number of constructions for walnut crushing machines, the technical data's of which must be taken into account when deciding to use them. First of all, the principle of walnut cracking used in the machine and during which it is necessary to calibrate the nut is important. Moreover, the more thoroughly such a calibration is performed (the greater is the number of fraction sizes), the better the crack will be and the greater the output of the partitions, which in turn will affect the quality and price of the shelled walnuts. Thus, calibration occupies an important place in the walnut processing chain.

Today, most walnut calibrators are rotary and have a sectional drum-shaped construction with

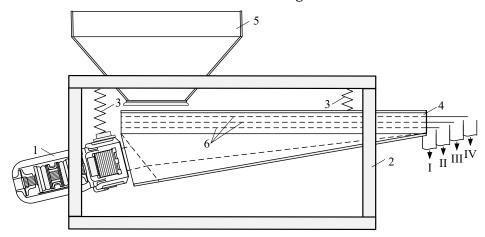
holes of different sizes in each section. The productivity of such a machine depends significantly on the number of fractions obtained. These types of calibrators are produced by the USA, France, Germany, Spain, China, Ukraine, Moldova and others. In these machines, the working frequency of the sorting drum is regulated by a frequency regulator, which increases the cost and complicates the operation process [9].

#### Purpose and methods

The aim of the work is to develop and research a vibrating walnut calibrator equipped with reciprocating electromagnetic vibrating motor; Analysis of the process of vibrational calibration of walnuts and determination of optimal technological parameters; Loose and solid matter sampling modes (methods) - oscillating with or without particle cutting from the working surface [10] - in the process of walnut calibration.

#### Research

An analysis of the calibration process and known constructions as a result of the study showed that the calibration of the nut can be achieved using vibration. Therefore, we have developed a nut calibration vibrating device (Fig. 3), which can be driven by our patented reciprocating electromagnetic vibrating motors 1 with direct current (DC) biasing [11-13], where it is possible to adjust the vibration of the working body by the use of bias current change.



**Fig. 3.** Walnut calibrator with reciprocating electromagnetic vibrating motor.

1 - vibrating motor; 2 - frame; 3 - suspension elastic system; 4 - vibrating chute; 5 - hopper;

6 - screens with openings of different sizes

The machine works as follows. The walnut from the hopper 5 falls on the vibrating chute 4, in which three screens 6 are placed in parallel under each other, with openings of different sizes. The walnuts will be divided into four fractions, of which I will be small-sized walnuts and impurities, which are necessarily found in raw materials for the first time, II-larger than fraction II, and so on. The number of fractions can be changed if necessary. The machine can be used to sort hazelnuts, chestnuts and others.

During the calibration process, the walnuts move on the screen in the direction of the outlet and are divided into two fractions: a) which fit and pass through the holes of the screen; b) which cannot fit and move along the surface of the screen towards the exit. Under the influence of vibration there is a transport of walnuts along the screen in vibrating chute, which is necessary for the continuity of the process and is characterized by the average speed (velocity) of the center of the whole bulked walnut mass.

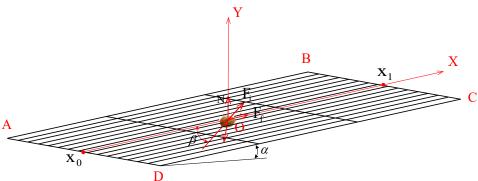
Consider one of the calibrators of a calibrator on the ABCD surface forming an  $\alpha$  angle with respect to the horizon, a walnut whose mass in m, kg is the process of displacement (Fig. 4).

In the *XOY* coordinate system, a walnut that fails to pass through the screen opening will have the following equilibrium condition [14]:

$$\begin{cases} m\ddot{x} = F_f - mg\sin\alpha + F_i\cos\beta = 0\\ m\ddot{y} = N - mg\cos\alpha + F_i\sin\beta = 0 \end{cases} \tag{1}$$

The force of gravity acts on the walnut is P = mg, N; g - free fall acceleration, m/sec<sup>-2</sup>; N - normal reaction, N;  $F_f = Nf_s$  - frictional force, N;  $f_s$  - sliding friction coefficient and  $F_i = mx_m\omega^2\sin\omega t$ , inertia, N;  $x_{m-1}$ 

amplitude of the screen oscillations, m;  $\omega$  - angular frequency of oscillation, sec<sup>-1</sup>; t - time, sec;  $\beta$  - angle of oscillation direction towards the surface of the *ABCD* screen, °.



**Fig. 4.** Scheme of action of forces on a walnut moving on the surface of the screen tilted at an angle to the horizon

From (1) is a normal reaction

$$N = m(g\cos\alpha - x_m\omega^2\sin\omega t\sin\beta) \qquad (2)$$

In order for the walnut to move from  $x_0$  to the point  $x_1$  on the screen surface (Fig. 4) it is necessary to fulfill the condition  $m\ddot{x} > 0$  or  $F_i \cos \beta > mg \sin \alpha - F_f$ , from which

$$x_m \omega^2 \sin \omega t \cos \beta > g \sin \alpha - \frac{N}{m} f_s$$
 (3)

Depending on the operating modes, the walnut may travel with or without disconnection from the vibrating working surface:

a) the travel disconnect from the surface we have when N < 0. Accordingly, the reset parameter - dependence on the amplitude of the transverse component of the inertial force from the amplitude of the transverse component of the gravity

force 
$$w_0^* = \frac{x_m \omega^2 \sin \beta}{g \cos \alpha} > 1$$
 (4)

b) without tear off the walnut from the surface, N > 0, namely when  $w_0^* < 1$ .

When the body slides on the vibrating surface by inertia, there is a certain aspiration to get the orientation defined by it, the direction of its movement [10]. This phenomenon can be used in our case as well, especially to determine the shape of the opening.

1. Self-sorting - sinking of the walnuts with small size and high density into the bottom layer (near the surface of the screen) and pushing out of walnuts of large size and low density into the upper layer; During the separation with screens, the self-sorting intensity is measured by the speed \$\mu\_{ss}\$ at which

the nuts are sinking from the upper layer to the holes on the surface of the screen, overcoming a distance  $h_{w}$  in the  $v_{ss} = \frac{h_W}{t_{ss}}$ 

2. Screening - the movement of a walnut screen smaller than the size of the openings through the openings.

Screening as the last stage of calibration comes from the bottom layer, so when the bulk mass is thicker then the average size of the walnut, it is necessary to sink the walnuts into the bottom layer that are subject to screening. If such walnuts are coming to the surface, for example due to their low density compared to the surrounding ones, for effective calibration it is necessary to roll the bulk mass or reduce the thickness of the walnut layer.

To ensure continuous flow of walnuts, the thickness of the layer of nuts at a distance x from the oopening of the screen

$$h_x = \frac{Q_W - Q_{WS}}{b v_x \rho_W}$$

where  $^{b}$  is the width of the vibrating chute, m;  $v_x$  - average speed across the walnut laver, m/sec;  $\rho_w$  - density of walnuts, kg/m<sup>3</sup>;  $Q_w$ - mass production of walnuts (initial material), kg/sec;  $Q_{ws}$  - number of nuts, passed the openings from  $x_0$  to x, kg/sec.

The screening of each walnut during its movement on the screen opening is a random event, the probability of which is subject to normal law when moving is without interruption

$$p_0 = 1 - \frac{1}{\sigma\sqrt{2\pi}} \int_0^v e^{-\frac{1}{2} \left(\frac{v - v_k}{\sigma}\right)^2} dv$$

where v is the speed of the center of mass of the nut relative to the grate, m/sec,  $\sigma = tg\alpha$ /  $f_s$  - the inclination parameter of the screen surface,  $v_k$  - the value of the critical speed, m/ sec, when  $p_0 = 1/2$ .

If the movement of the nuts comes off with interruption from the surface of the screen, the probability of screening is significantly reduced due to the reduced contact time of the nut with the screen. This happens not only by increasing the normal setting of the screen acceleration and violating the condition of movement without a smash, but also by "jumping up" after hitting the edge of the nut opening. The latter is observed when moving a thin layer. Increasing the layer

thickness to the optimum  $h^*$  magnitude increases the likelihood of screening, since the upper layers do not experience direct shocks and are resistant to cutting the lower layers from the screen. When  $h > h^*$  jumps do not exist for this reason, but the pressure of the upper layers increases the friction between the nuts of the lower layer, which complicates the screening process.

The specific throughput of the screen qw, kg/m<sup>2</sup>sec is proportional to the number of walnuts nw that pass through the openings in 1 sec and the probability of their screening  $p_w$ 

$$q_w = c_0 n_w p_w$$

where <sup>c</sup><sub>0</sub> is the coefficient of proportionality, which depends on the "live" area of the screen, the concentration of the outgoing nut in the lower layer, density and other properties.

Experimental research has shown that, given the physical and mechanical properties of the walnut, calibration with the reciprocating motion vibrating motor is relatively effective in moving the walnut take place without detachment from the surface. To do this, it is necessary to fulfill condition (4), respectively, to justify the following parameters and modes of the vibrator: the angle of inclination of the screen with respect to the horizon  $\alpha$ , oscillation frequency  $\omega$  and direction angle  $\beta$ , also relevant are the amplitude of the oscillations  $x_m$ , the shape and size of the screen opening. At this time Y = 0, and the friction force

$$F_f = \begin{cases} -f_s N, \ \dot{x} < 0 \\ f_s N, \ \dot{x} > 0 \end{cases} \tag{5}$$

and when the walnut, that cannot pass through the screen surface is in a stationary position (X = 0, Y = 0), then the force of dry friction is calculated from (1)  $F_{df} = -mg \sin \alpha + F_i \cos \beta$ , however, this state is maintained until the inequality  $-f_{fs}N < F_{df} < f_{fs}N$  is completed, where  $f_{fs}$ is the coefficient of friction of immobility, usually  $f_{fs} \ge f_s$ . The equation for the acceleration  $\ddot{x}$ , the speed  $\dot{x}$  and displacement xof the walnut on the screen surface is taken from (1), (2) and (5)

$$\ddot{x} = -g \frac{\sin(\alpha \pm \theta)}{\cos \theta} + x_m \omega^2 \frac{\cos(\beta + \theta)}{\cos \theta} \sin \omega t \tag{6}$$

$$\dot{x} = -gt \frac{\sin(\alpha \pm \theta)}{\cos \theta}; -x_m \omega \frac{\cos(\beta \mp \theta)}{\cos \theta} \cos \omega t \tag{7}$$

$$\ddot{x} = -g \frac{\sin(\alpha \pm \theta)}{\cos \theta} + x_m \omega^2 \frac{\cos(\beta \mp \theta)}{\cos \theta} \sin \omega t \qquad (6)$$

$$\dot{x} = -gt \frac{\sin(\alpha \pm \theta)}{\cos \theta} \div x_m \omega \frac{\cos(\beta \mp \theta)}{\cos \theta} \cos \omega t \qquad (7)$$

$$x = -\frac{gt^2}{2} \frac{\sin(\alpha \pm \theta)}{\cos \theta} - x_m \frac{\cos(\beta \mp \theta)}{\cos \theta} \sin \omega t \qquad (8)$$

where  $\theta = arctgf_s$  is the sliding friction angle; In the formulas (6) - (8), the upper mathematical signs (+ or -) correspond to sliding forward  $(\dot{x} > 0)$ , and the lower ones - to sliding back  $(\dot{x} < 0)$ .

Established modes of motion of a substance without detachment from the oscillating surface are discussed in [5]. For effective calibration of the walnut, taking into account the shape of the screen openings we have chosen, it is advisable to use the mode when the walnut slides in one direction from point  $x_0$  to point  $x_i$  (Fig. 4).

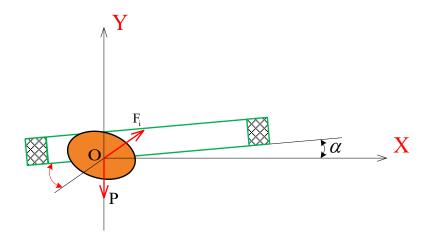


Fig. 5. Scheme of the action of forces on a walnut moving in the opening of the screen tilted at an angle  $\alpha$  to the horizon

In the XOY coordinate system, a walnut moving through an opening (Fig. 5) will have the following equation of motion:

$$\begin{cases}
m\ddot{x} = -F_i \cos \beta \\
m\ddot{y} = -mg + F_i \sin \beta
\end{cases}$$
(9)

from which, after appropriate little manipulation we obtain

$$\begin{cases} \ddot{x} = -x_m \omega^2 \sin \omega t \cos \beta \\ \ddot{y} = -g + x_m \omega^2 \sin \omega t \sin \beta \end{cases}$$
 (10)

$$\begin{cases} \dot{x} = x_m \cos \omega t \cos \beta \\ \dot{y} = -gt - x_m \omega \cos \omega t \sin \beta \end{cases}$$
 (11)

$$\begin{cases} \ddot{x} = -x_m \omega^2 \sin \omega t \cos \beta \\ \ddot{y} = -g + x_m \omega^2 \sin \omega t \sin \beta \\ \dot{x} = x_m \cos \omega t \cos \beta \\ \dot{y} = -gt - x_m \omega \cos \omega t \sin \beta \end{cases}$$
(10)  
$$\begin{cases} x = x_m \sin \omega t \cos \beta \\ y = -g \frac{t^2}{2} - x_m \cos \omega t \sin \beta \end{cases}$$
(12)

and from (11) - the movement speed of the walnut is

$$v_w = \sqrt{(x_m \cos \omega t \cos \beta)^2 + (-gt - x_m \omega \cos \omega t \sin \beta)^2}$$

which depends on the amplitude, frequency and angle of the oscillation direction. These values, as well as the distance to the next screen surface on which the walnut to be driven into the upper screen should fall, should be selected so that jumps are minimized.

The results of the obtained theoretical and experimental studies show, that if the width of vibrating chute of the calibrator is b = 0.6m, height h = 0.15 m, length l = 1.5 m and amplitude of oscillations  $X_m = 0...0,0015$  m with oscillation frequency  $\omega = 314 \text{ sec}^{-1}$ , the walnuts can be calibrated without disconnecting from the screen surface at the values of  $\beta$  and  $\alpha$ , given in the Table 2.

**Table 2.** Value limits of vibration angle  $\beta$  and chute angle  $\alpha$  in vibrating calibrator for calibration of walnuts without disconnecting from the screen surface for specific values of  $X_m$  and  $\omega$ 

| # | $X_m$ , m | $\omega$ , sec <sup>-1</sup> | $\beta^{0}$                            | $\alpha^{0}$                          | w <sub>0</sub> *        |
|---|-----------|------------------------------|--|---------------------------------------|-------------------------|
| 1 | 0,0005    | 314                          | $0^{\circ} \leq \beta \leq 11^{\circ}$ | $0^{\circ} \le \alpha \le 15^{\circ}$ | $0 \le w_0^* \le 0,992$ |
| 2 | 0,0010    | 314                          | $0^{\circ} \leq \beta \leq 5^{\circ}$  | $0^{\circ} \le \alpha \le 28^{\circ}$ | $0 \le w_0^* \le 0,991$ |
| 3 | 0.0015    | 314                          | $0^{\circ} \leq \beta \leq 3^{\circ}$  | $0^{\circ} \le \alpha \le 37^{\circ}$ | $0 \le w_0^* \le 0,987$ |

#### Conclusion

It is advisable to use a vibrating device for calibration of walnuts due to its design simplicity and high reliability of operation.

Analysis of the movement of the walnut on the surface of the vibrating screen and through the openings allowed us to determine the influence of the main factors on the design-kinetic parameters that ensure the calibration of the nuts.

For effective calibration of the walnuts, the mode should be used when the walnut slides in one direction from point  $x_0$  to point  $x_1$  (Fig. 4).

The productivity and quality of the vibrating calibrator depends on the speed of movement of the walnut on the screen surface, which in turn depends on the coefficient of friction, the angle of inclination of the screen towards the horizon, the frequency, amplitude and direction of vibration.

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# Checklist of carrion beetle (Coleoptera, Silphidae) from Sakartvelo (Georgia)

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#### ABSTRACT

A checklist of Carrion beetles of Sakartvelo (Georgian) contains 26 species, which are represented by nine genera (*Ablattaria* Reitter, *Aclypea* Reitter, *Dendroxena* Motschulsky, *Necrodes* Leach, *Nicrophorus* Fabricius, *Oiceoptoma* Leach, *Phosphuga* Leach, *Silpha* Linnaeus, *Thanatophilus* Leach) and two subfamilies Nicrophorinae and Silphinae. Distribution records are given by using available literature, web databases and collections. The pictures of few specimens are provided and species with doubtful records in Georgia are not included on the list but are given separately.

Keywords: Carrion beetles, Silphidae, Coleoptera, Distribution, Caucasus, Georgia.

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#### Introduction

Carrion beetles (Coleoptera, Silphidae) are a small family of beetles: fewer than 200 species have been described worldwide, however they have global distribution [1].

They play an important role in ecosystem as recycles of organic matter. While most of the carrion beetles are necrophagous, some of the members of Silphidae family are predaceous (*Ablattaria* Reitter, *Dendroxena* Motschulsky, *Phosphuga* Leach) or phytophagous (*Aclypea* Reitter). Larvae of most species feed on vertebrate carrions [1, 2].

Carrion beetles in Georgia (in Georgian: Sakartvelo) are poorly studied: one of the first list of Caucasian beetle was published in 1899 by Radde [3], later Zaitsev provided list of Carrion beetles for Caucasus region [4, 5], Kobakhidze indicated three species of Family Silphidae for Lagodekhi region [6], Jambazishvili provided carrion beetles from Akhaltsikhe and Akhalkalaki region [7], Qubaiová et al. provied lockations for Genus *Ablattaria* (Reitter, 1884), in their work [8], V. Pushkin provided data for Georgia for several species in his papers [2, 9]

Aim of this paper is to summarize all known records of carrion beetles in Sakartvelo (Georgia).

#### Material and Methods

Literature review was performed according published sources, which was provided by Institute of Entomology in Agricultural University of Georgia and National Scientific Library - Georgia and based on by using Google-Scholar and Researchgate web databases, using Keywords: "Carrion beetles", "Silphidae", "Distribution", "Caucasus", "Georgia" and combination of those keywords. Locations were looked in GBD (Georgian Biodiversity Database) [10], GBIF [11 - 18] (Global Biodiversity Information Facility) and collections listed in abbreviations were studied.

All species are listed alphabetically by subfamilies. Nomenclature and classification according to

Catalogue of Palaearctic Coleoptera [19]. For species with no exact locations, "Georgia, Exact location is unknown" was used in Distribution.

Pictures were taken by using a Canon EOS 550D camera with Canon EF 100mm f/2.8 Macro USM lens and Raynox DCR-250 Super Macro Snap-On Lens attached on it. Digital images were prepared using Zerene Stacker image stacking software and Adobe Photoshop CS6.

#### Abbreviation:

EG - East Georgia.

WG - West Georgia.

GCG - collection of Dr. Giorgi Chaladze, Sakart-velo (Georgia).

ZIISU - collection of Zoological Institute, Ilia State University, Sakartvelo (Georgia).

IEAUG - collection of Institute of Entomology, Agricultural University of Georgia.

CaBOLISU - collection of CaBOL (Caucasus Barcode of Life), Ilia State University, Sakartvelo (Georgia).

#### Results

Family Silphidae Subfamily *Nicrophorinae* Kirby, 1837 Genus *Nicrophorus* Fabricius, 1775

1 N. antennatus (Reitter, 1885)

Distribution: Georgia, Exect location is unknown [19].

2 N. germanicus (Linnaeus, 1758)

Distribution: Georgia, Exact location is unknown [2, 19].

3 N. humator (Gleditsch, 1767)

Distribution: EG: Borjomi [4], Borjom-Kharagauli National park (IEAUG), Tbilisi [4].

4 N. interruptus Stephens, 1830

(= *N. fossor* (Erichson, 1837)) [2]

Distribution: EG: Lagodekhi [6], Village Khevsha [10], WG: Bzyb ridge [2], Swaneti [3], Iprali [13].

5 N. investigator Zetterstedt, 1824

(= *N. funeror* (Reitter, 1884)) [23]

Distribution: EG: Lagodekhi [4], Bakuriani [4], Dighomi Village [10], WG: Swaneti [3, 23].

6 N. nigricornis Faldermann, 1835

Distribution: EG: Lagodekhi [4], Tskra-tskaro [4], WG: Swaneti [3, 4].

7 N. sepultor (Charpentier, 1825)

(= *N. confusus* Portevin, 1924) [20]

Distribution: EG: Tbatani [4], Tsalka [15], Chivtkilisa [22 as *N. confusus*], Akhalsheni (previously known as Kariaki) [22 as *N. confusus*], Tetri-tsqaro [22 as *N. confusus*], Akhaltsikhe region [22 as *N. confusus*], Akhalkalaki region [22 as *N. confusus*].

8 N. vespillo (Linnaeus, 1758)

Distribution: EG: Village Khevsha (GCG), Bakuriani forest (GCG), Bakuriani [4], Mountain likhi (GCG), Akhaltsikhe region [7], Akhalkalaki region [7], Ketrisi (ZIISU) WG: kintrishi Protected Areas (CaBOLISU), Chakvistavi [11], Kobuleti [12].

9 N. vespilloides Herbst, 1783

Distribution: WG: Bzyb range [2].

10 N. vestigator Herschel, 1807

Distribution: Georgia, Exect location is unknown [19].

#### Subfamily *Silphinae* Latreille, 1807 Genus *Ablattaria* Reitter, 1884

11 A. arenaria (Kraatz, 1876)

Distribution: WG: Sukhumi [2].

12 A. cribrata (Menetries, 1832)

Distribution: EG: Kaspi (GCG), Sioni [8], Lagodekhi [8], Didgori [10] Borjomi [3, 4], Borjom-Kharagauli, 2 km East Quabiskhevi [8], Chankaani, near Dedophliswyaro (CaBOLISU).

13 A. laevigata (Fabricius, 1775)

Distribution: EG: Bakuriani [4, 8], WG: Gagra [8], Kutaisi [8].

#### Genus Aclypea Reitter, 1884

14 *A. opaca* (Linnaeus, 1758)

Distribution: EG: Tsodoreti-Paravani [10].

15 A. undata (O.F Müller, 1776)

Distribution: WG: Avadkhara [9], EG: Samachablo (South Ossetia) [2], Surami [3], Kasbegi [3], Telavi [4], Tbilisi [17], Saghamo lake (CaBOLISU).

#### Genus Dendroxena Motschulsky, 1858

16 D. quadrimaculata (Scopoli, 1771)

(= *Xylodrepa quadrimaculata* (Schreber, 1759)) [3, 4]

Distribution: EG: Turtle lake, (Bulbulebis chala) [10].

#### Genus Necrodes Leach, 1815

17 N. littoralis (Linnaeus, 1758)

Distribution: EG: Likani [10], Samachablo (South Ossetia) [2], Akhaltsikhe region [7], Akhalkalaki region [7], WG: Avadhara, Akhalsheni, Sukhumi Region valley River Gumista [2].

#### Genus Oiceoptoma Leach, 1815

18 O. thoracicum (Linnaeus, 1758)

Distribution: EG: Samachablo (South Ossetia) [2], Bakuriani [2], Mt. Lomismta (IEAUG), Surami [3, 4], Kojori [4], WG: Avadkhara [2], Akhalsheni [2], Pskhu [4], Kodori [4], Shovi (ZIISU).

### Genus Phosphuga Leach, 1817

19 *P. atrata* (Linnaeus, 1758)

Distribution: EG: Didgori (GCG), Bakuriani [4], Telavi [4], Manglisi [4], National park of Borjom-Kharagauli [10], WG: Lezgara (Church Tekhish Jgrag) (CaBOLISU), Zeskho (ZIISU), Gagra [4].

20 P. ruzickai Khachikov, 2011

Distribution: WG: Lake Ritsa [19, 21].

#### Genus Silpha Linnaeus, 1758

21 S. carinata Herbst, 1783

(= S. armeniaca Kolenati, 1846) [4]

Distribution: EG: Passanaur [4], Tsalka [4], Telavi [4], Lagodekhi [4], Mt. Lomismta (IEAUG), WG: Mt. Taginauri [2].

22 S. obscura Linnaeus 1758

Distribution: EG: Turtle lake (GCG), Khevsha

village (GCG), Tskra-tskaro pass (GCG), Passanauri [16], Surami [3], Tashiskari [10], David Gareja [10], Akhaltsikhe region [7], Akhalkalaki region [7], Zemo Tmogvi (CaBOLISU) WG: Adjara [3], Mt. Taginauri [2], Kintrishi (Khinu) (GCG), Swantei [3], Gagra[14], Near Shkhara (CaBOLISU).

23 S. tristis Illiger, 1798

Distribution: Georgia, Exect location is unknown [19].

#### Genus Thanatophilus Leach, 1815

24 T. rugosus (Linnaeus, 1758)

Distribution: EG: Samachablo (South Ossetia) [2], Teliani [4], Bakuriani [4], Mt. Lomismta [4], Mtskheta [3, 4], Tbilisi; Vakis park (GCG), Akhaltsikhe region [7], Akhalkalaki region [7], Rustavi (GCG), WG: Akhalsheni, Avadhara, Sukhumi Region valley River Gumista [2].

25 T. sinuatus (Fabricius, 1775)

Distribution: EG: Khevsha village (GCG), Lisi [4], Borjom [4], Bakuriani [4], Akhaltsikhe region [7], Akhalkalaki region [7], WG: Akhalsheni, Sukhumi Region valley River Gumista [2].

26 T. terminatus (Hummel, 1825)

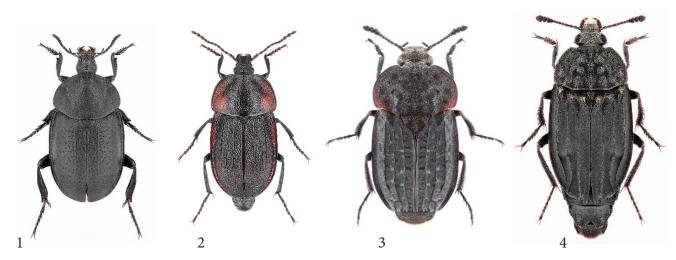
Distribution: EG: Lisi [4].

Doubtful species which was not included in the list

1 Thanatophilus dispar (Herbst, 1793)

Distribution: EG: Tskra-tskaro [4], Borjomi [18]

Comment: According to Pushkin [2], Notes for the Caucasus (Transcaucasia) is wrong. According to Zaitsev [5], this species must be crossed out from the list of Caucasians Species due to misidentification.



**Fig. 1.** Silphidae of Sakartvelo: 1. Ablattaria cribrata, male. 2. Phosphuga atrata, male. 3. Thanatophilus rugosus, male. 4. Thanatophilus sinuatus, female.



**Fig. 2**. Silphidae of Sakartvelo: 5. Necrodes littoralis, female. 6. Nicrophorus interruptus, male. 7. Nicrophorus investigator, female. 8. Nicrophorus vespillo, male.

#### Conclusion

In total, 26 species have been represented, from nine genera, the genus *Nicrophorus* Fabricius is the most diverse, with ten species occurred in Georgia. It is fair to say that the distribution of carrion beetles in Georgia is very diverse.

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# To study population polymorphism in animals (Population mechanisms of adaptation to the environment)

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#### ABSTRACT

As known, animals have many ways to adapt to the environment, one of which is based on intrapopulation heterogeneity, or polymorphism. It ensures an adequate transformation of the demographic structure of the population during any changes in the environment. In the paper, against the background of the general characteristics of animal populations, the population polymorphism of one of the representatives of the Acaroidea mites is described. Our findings are based on observations made at the end of the last century on the Mugan Plain, near the city of Salyan (Azerbaijan) and in Shirak, on the Eldar Valley, while studying the holes of the Microtus.

*Keywords:* Population, Polymorphism, Stacia, Homeostasis, Ontogeny, Homeomorphic (males).

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#### Introduction

Species are known to be unevenly distributed within their area, and usually their members form more or less separate groupings - populations. According to the modern view, population is the basic form of existence of a species, i.e. a group of individuals of one species, which occupies a certain area and has the ability to exist indefinitely for a long time. Its members copulate freely with each other and are usually spatially isolated from other populations of the same species.

The species exists in the form of populations. In any biocenosis it is represented by population. In this sense, population can be considered as part of biocenosis, and biocenosis - as a combination of different species of populations.

Any organism, especially if it reproduces sexually, is a member of this or that population. Its long existence without population is difficult, often impossible. Members of the population are interrelated

in many ways. Because of this, the population functions as a unified system [1].

No matter how homogeneous the species area is, not even two points can be found in it, with absolutely identical living conditions. This means that the action of natural selection is always different here. As populations are the result of long-term natural selection, each of them is more or less different from the others in a number of ways.

The population functions as a unified system, although this does not mean that it is homogeneous. Most populations are characterized by a complex structure. But intrapopulation units, unlike populations, do not spatially exclude each other and are more or less closely related. Polymorphysiologically different forms within a population.

Polymorphism is an adaptive feature; it significantly increases the vital capacity of the population and contributes to the existence of the species under changing conditions. The more pronounced the polymorphism, the wider the ecological plasticity of the population, the more easily it adapts to cyclical and sudden changes in the environment.

One of the manifestations of intrapopulation heterogeneity is the demographic structure of the population. It refers to the ratio of individuals of different ages and genders, which determines the birth rate, mortality rate and, ultimately, the change in population number over time. Examples of polymorphisms are castes - in public insects, seasonal races - in fish, races by location - in insects and rodents, etc. There are morphs that serve to transfer unfavorable environmental factors.

Deterioration of living conditions (drought, severe frosts, food shortages) is accompanied by a sharp decline in the number of animals, but this rarely affects the ability of the population to selfrecover. At the expense of individuals (morphs) resistant to extreme conditions, the optimal structure of the population is easily restored at the appropriate time [2, 3].

Any form of polymorphism should be considered as variants of genetic heterogeneity because they are genetically determined.

As A. Yablokov notes that "intrapopulation forms of animals (fast-growing, born in spring or early summer, rapidly reproducing, overwintering, etc.) provide high population polymorphism, thus accurately enhancing animal resilience to environmental variability" [4]. In the following years V. Bolshakov and L. Dobrinsky wrote: According to biologically important traits, any population is heterogeneous, which helps it to adapt to fluctuations in environmental conditions through genetic and ecological transformations [5].

The study of polymorphism is one of the priority areas of population ecology today. Undoubtedly, it has not only theoretical but also great practical significance, because it can be used to solve some issues of conservation biology - this topical field of science.

The study of animal population polymorphism began in the 60s of the last century. The object of study is mainly representatives of higher animals. In other animals this phenomenon is relatively poorly studied.

#### Main content

At the end of the last century we were able to detect a peculiar reveal of a polymorphism in one of the representatives of the type Arthropoda. The observations were made on the Mugan Plain (Azerbaijan), where the biology of Acaroids in extreme conditions has been studied for years. Dominant species was the Phizoglyphus echinopus F. et. R. The Microtus were being studied - the only place for these animals to exist in the mentioned region. The extraction and processing of material (content of holes) was carried out by the traditional method [6]. We used a field thermoeclectors.

The Phizoglyphus echinopus F. et. R. (family of the Acaridae) is a cosmopolitan species; Known as a pest of storage products, primarily bulbs, tubers, medicines, grains and its processing products. It is, at the same time, widespread in natural stacias. Inhabits in the holes of the warmblood animals, in the nests of ants, where it reproduces smoothly throughout the year.

The *Rh. echinopus* - is one of the most widespread species in the South Caucasus (Georgia, Armenia, Azerbaijan). It is found in all natural belts - from semi-deserts and fields to subalpine meadows, both in synanthropic conditions and in nature. Prefers high moisture substrates. The frequency of this species in Georgia is: 44% in synanthropic habitats and 56% in natural conditions. It is clearly characterized by high resistance to low temperatures [7-9].

The demographic structure of this species is noteworthy. Ontogenesis encompasses three stages. There are two types of males - the so-called. "Homeomorphic", which is identical in appearance to females, and "heteromorphic", which differs from females in a number of morphological features. Homeomorphic males are usually involved in breeding. The function of heteromorphic males is unknown until recently.

Over the years the demographic structure of the Rh. echinopus population during the summer months in the mentioned area was more or less stable. By average, it looked like this:

the share of females in the samples was 52-61%, the share of males - 18-24% (of which heteromorphic males did not exceed 3%),

the share of underage forms - 32-41%.

A remarkable fact of drastic structural change in the *Rh. echinopus* population was observed in July 1981. In the first decade of this month there was an unusual drought in

the area. The number of the *Rh. echinopus* population has decreased by almost 5.5 times compared to the summer months of other years. About 95% in the samples were heteromorphic males and females, in approximately equal numbers.

In mid-July, we again analyzed the holes of the Microtus. The result was similar: against the background of a small number of organisms, a large part of the population was still heteromorphic males and females.

In mid-September we were again given the opportunity to study the demographic structure of the Mugan population. This time a large part of the population was represented only in underage forms. The share of adult organisms did not exceed a few percent. There was noted the copulation process.

In the summer of 1990 we again discover the *Rh*. *Echinopus* population - this time on the Eldar Valley, in the holes of the public Microtus. The number of the population did not exceed three dozen (on the traditional sample), of which about 50% were heteromorphic males, more than 20% - females, and the rest of the population were homeomorphic males and underage forms.

Unfortunately, we were not able to detect a similar change in the Rh. echinopus population in other regions of the South Caucasus. Therefore we have to draw conclusions based only on the unit populations described here. We suppose that the only reason for such a drastic change in the Mugan population is a substantial change in the microclimate at its particular location. The multiple increase in the share of heteromorphic males suggests that they take on the function of painlessly endure unfavorable living conditions by the population. The same can be said for females found with heteromorphic males in populations. There is no doubt that the ability to endure unfavorable conditions painlessly is their inherited determinant feature, which ultimately serves to save the population.

We had similar occurrence in the case of the Rh. echinopus' Eldar population.

It should be noted that the heteromorphic males described here are not only typical for Rh. echinopus. It is noticed in other representatives of the Acaroidea superfamily, namely, in the genera: Acotyledon, Calo-

glyphus, Schwiebea, etc., although whether they perform the same function as in the species described here is difficult to say [10]. In some representatives of the superfamily Acaroidea, peculiar dimorphism is also observed in underage forms. In many species (genera Acarus, Histiogaster, Mycetoglyphus, etc.) between stages I and III of individual development, two types of different structures are involved, of which I - serves to locate the species, and the function of II is often unknown. Thus we can assume that polymorphism is a much more common phenomenon than is confirmed by established facts.

#### Conclusion

As we can see, *Rh. echinopus* heteromorphic male is an essential element of the population (morpha), which serves to save the population during adverse changes in the environment. But after the environment stabilizes, they intensively copulate with the remaining females, after which the population returns to its optimal size and structure.

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# Mineral qualities of Isabella grapevine (Vitis labrusca L.) leaf

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#### ABSTRACT

The goal of the present paper was to identify the content of heavy metals, also macro - and micro-nutrients, which play very important roles in human health in Isabella grapevine (Vitis labrusca L.) leaves as a raw material to produce a tonic drink rich in vitamins and active substrates. Samples of leaves and soils were analyzed by flame atomic absorption spectroscopy. The leaves of Isabella do not contain any heavy metals and are suitable for our purposes.

Keywords: Grape leaves, Nutrient content, Vitamins, Heavy metals, Soil samples, Analysis by FAASPS.

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#### Introduction

With the aim of keeping the population healthy and fit for work, usage of plant-based concentrates enriched with food vitamin complexes became very popular [1]. Taking this into the account, we studied the secondary product, such as grapevine leaf, which is enriched with vitamins, antioxidants, and is a dietary product. It is used in medicine and perfumery. Studies have shown that from the green grapevine biomass we can produce the following products: 1. grapevine poly vitamin and granular dragée; 2. liquid food concentrates; 3. dry, easily soluble food concentrates of the grapevine; 4. natural food dyes; 5. Green, yellow, red, and black granules vitaminized by the young offshoots of the grapevine; 6. protein and vitamin-enriched food for stockbreeding [2].

In Georgia, one of the widespread breeds of the grapevine is Isabela (Adesa). This is a natural hybrid with the qualities of cold resistance, phylloxera resistance, and fungi-related disease resistance,

therefore it doesn't require spraying with pesticides. Usage of the leaf of this grapevine and producing tonic, vitamins, and useful substances-enriched drinks will play a significant role in the food economy [3].

In the list of the most useful products, published by the dietologists and gastroenterologists of the European research association, Isabella takes fourth place in the terms of its biochemical content.

Our aim was to study the leaf of the Isabela grapevine for the production of tonic, non-alcoholic refreshing drinks.

As a result of the research, it was shown that the leaf of the Isabela grapevine contains metals. These observed metals don't include heavy metals such as lead, cadmium, and mercury.

#### Object and methods

As research material, the leaf of the Isabela grapevine was taken from the East Georgia (Ga-

chiani) and West Georgia (Chokhatauri) regions. Also, soil samples were taken from the place, where the raw material was collected. The soil samples were tested to study the presence of the significantly important micro and macroelements. Additionally, the seasonal variability (Spring, Autumn) of the grapevine leaf mineral composition was studied.

Reagents: high purity nitric acid, deionized water, especially pure air-like acetylene, metal composition standard models (standard solutions of the Perkin Elmer company)

Testing the presence of metals in samples was conducted with Buck Model 210VGP atomic absorption spectrophotometer [4-6]. Atomic absorption spectroscopy (AAS) is a physical analysis method. AAS is based upon the principle that free atoms in the ground state can absorb light of a certain wavelength. Absorption for each element is specific, no other elements absorb this wavelength. When the metals from the solution end up in flame, metal ions are transitioning to the atomic state and an aerosol is produced. The metals, which are to be determined, don't interfere with each other. From the spectral lightbulb of the metals, which are to be researched in the samples, the light crosses the flame, the atomic cloud will light up and atoms will absorb the part of the radiation. The relationship between the concentration of absorbed atoms and the beam of light is described by The Beer-Bouguer-Lambert law: A=Lg(1/T)=Lg(10/I), where T is the conductance of the flame, 10/1 is the intensity of the radiation which has been dropped and then passed. A is a quantity of absorbed light beams. Then the light beam will end up in the monochromator detector. In the detector will be measured the energy of the light beam which was absorbed by atoms in flame. The intensity of the light beam absorption depends

on the presence of a free, ground state number of atoms in flames.

#### **Experiment**

Since the Isabela grapevine is not a subject of pesticide usage, firstly we measured the heavy metal presence in the soil samples. Additionally, we measured the presence of micro and macroelements, necessary for viability.

The work was carried out in the Laboratory of Food Safety of Grocery department (Faculty of Agricultural Sciences and Biosystems Engineering GTU). During the work, the used samples were the spring leaves of the Isabela grapevine from Eastern (Gachiani) and western (Chokhatauri) parts of Georgia, the samples of autumn leaves, and the soil samples.

Various solutions containing different concentrations of metals were made on the deionized water to measure absorption index and to create the relevant mathematical model. For different metals, the corresponding lightbulb was chosen. Measuring the absorption wave was achieved by the manual atomic absorption spectrophotometer. The mode of the device is chosen by the recommendations of the enterprise which has produced the spectrophotometer. After turning on the spectrophotometer, the atomization method is chosen by the computer program WinLab32 (setting up the atomizer and adjustment is achieved automatically). The data which is presented in the following table is corresponding to the samples of: 1 (Chokhatauri, Spring leaf), 2 (Gachiani, Spring leaf), 3 (Chokhatauri, Autumn leaf), 4 (Gachiani, Autumn Leaf), 5 (Chokhatauri, soil), 6. (Gachiani, soil).

**Table 1.** The concentration of metal in the samples (mg/kg)

| Metals | M.A.C. | sample 1 | sample 2 | sample 3 | sample 4 | sample 5 | sample 6 |
|--------|--------|----------|----------|----------|----------|----------|----------|
| Cu     | 3,0    | 0,04     | 0,11     | 0,03     | 0,10     | 0,07     | 0,13     |
| Zn     | 23     | 4,1      | 4,78     | 4,23     | 4,69     | 4,25     | 4,79     |
| Pb     | 6,0    | 0,06     | 0,07     | 0,05     | 0,07     | 0,07     | 0,08     |

M.A.C. —maximum allowable concentrations - maximal concentration, when the substance does not affect (neither directly nor indirectly) the health of the population.

According to the results, there was almost no difference between the soil samples and leaf samples in terms of heavy metal presence. In the samples, there were practically no Cd, As, Hg, Ag, Ni, Cr, Sn present.

The samples were tested for the presence of several metals, significant for the development:

| <b>Table 2.</b> The presence of sev | eral significant metals (mg/kg) |
|-------------------------------------|---------------------------------|
|-------------------------------------|---------------------------------|

| Metals | sample 1 | sample 2 | sample 3 | sample 4 | sample 5 | sample 6 |
|--------|----------|----------|----------|----------|----------|----------|
| K      | 7,61     | 7,59     | 7,62     | 7,61     | 6,9      | 6,9      |
| Na     | 1,7      | 1,68     | 1,69     | 1,66     | 0,8      | 0,8      |
| Ca     | 5,69     | 4,99     | 5,66     | 5,0      | 4,09     | 4,09     |
| Mg     | 3,01     | 3,01     | 2,62     | 2,61     | 1,0      | 1,11     |
| Fe     | 2,37     | 2,87     | 2,4      | 2,75     | 2,0      | 2,0      |
| Mn     | 0.2      | 0.11     | 0.13     | 0.12     | 0.11     | 0.12     |

In the grapevine leaves most of the elements are entering the leaf as a salt, as an ion, or as a complex/organic substance. Therefore, they're crucial components (nutrients). Even though mineral substances don't have energetic value as proteins, lipids or polysaccharides do, they are essential substances for viability [7-11].

### Conclusion

- 1. The leaf of the Isabela grapevine doesn't contain heavy metals and it can be used for producing raw tonic drinks.
- 2. Usage of the leaves of this species and producing tonic, vitamin-enriched drinks will play a significant role in the food production economy.

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# Comparative study of lipase inhibitory activity of some Georgian wines obtained through Kakhetian and European winemaking techniques

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### ABSTRACT

The lipase inhibitory activity, total polyphenol content and antioxidant activity of some Georgian wines were studied. Fourteen commercially available samples of red and white wines were analysed, which differed by the production method (Kakhetian winemaking and Classic European winemaking methods). From the investigated wines, the highest total polyphenol content (TPC) was found in the wine made by Mukuzani Valley 2019 (3572.358  $\pm$  153.111 mg Gallic Acid equivalent (GAE) per liter. This wine had the highest antioxidant activity (AOA), too, 4729.199  $\pm$  88.162 mg ascorbic acid equivalent (AAE) L<sup>-1</sup>.

It was recognised that wines made with the Kakhetian fermentation method contain more total polyphenols than those made by the classical European method. The differences between the samples were statistically significant. White wines made with this Kakhetian method have comparable TPC to some European-style red wines. Red wines in general are characterised on average by higher anti-lipase and antioxidant activity than white wines, although white wine had the most increased anti-lipase activity among the investigated samples (82.63% mL<sup>-1</sup> of wine). Wines from Mukuzani microzone possess high anti lipase activity, which is ranged from 77.12 to 79.7 8% mL<sup>-1</sup> of wine. No correlation between TPC and lipase inhibitory activity among the red and white wine samples was found, nor between lipase inhibitory activity and the winemaking method.

*Keywords*: Georgian wine, Red Wine, White wine, HPLC, Kakhetian fermentation method, Pancreatic lipase.

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#### Introduction

Obesity, which has been considered a twenty-first-century disease and the "New World Syndrome", is a global public health concern [1, 2]. Many studies have shown a positive relationship between obesity and the intake of a lipid-rich diet [3, 4]. Efficient absorption of dietary fats is highly reliant on the action of pancreatic lipase (PL) [5]; thus, the inhibition of this enzyme has become an attractive approach to manage and treat obesity [6, 7]. Currently, the only available PL inhibitor at the market is Orlistat (Xenical) [6, 7], derived from the gram-positive bacterium [8] with adverse effects, such as fecal incontinence, stomach pain or discom-

fort, fatty/oily stools etc. [9-11]. Hence, the search for new substances with fewer undesirable effects and potent inhibitory activity against PL remains a hot topic in research [12,13].

A great deal of research showed that the class of polyphenols represents one of the essential sources to inhibit PL activity [14-16]. Grapes and their derivatives are considered one of the richest natural sources of phenolic compounds. The composition of phenolics is highly linked to wine quality properties, such as colour, flavour, and taste, as well to health-promoting properties, including antioxidants and cardioprotective properties among others [17, 18]. Furthermore, on account of health-promoting chemicals (e.g. phenolics), moderate wine con-

sumption is nowadays recognised as a risk-reducing factor in several human diseases, including type 2 diabetes [19, 20], several types of cancer [21, 22] and cardiovascular disease [21, 23]. However, the phenolic content in wine can be influenced by many factors such as grape cultivar, soil, climatic conditions, weather, winemaking procedure and conditions of maturation and storage [18, 24, 25].

In this manner, the country of Georgia is known to use the ancient method of winemaking (widely known as Kakhetian winemaking or Qvevri winemaking method), which differs from Western European techniques (referred to as the Classic winemaking, Industrial method). Kakhetian winemaking method involves placing crushed grapes and other parts, i.e., clusters (stem, skin, seeds) in a clay vessel called Qvevri, dug in the ground. Qvevri is then sealed, and the wine is left to mature. During fermentation, phenolic compounds are extracted from pomace, defining the composition and essence of Kakhethian wine [26]. This winemaking method has also been approved as an intangible cultural heritage convention by UNESCO [27]. Furthermore, Georgia is considered the "cradle of wine", as the earliest traces of winemaking have been found here [28]. Additionally, Georgia is home to over 500 varieties of indigenous grapes, many of which are unknown to the rest of the world [29].

Although a significant amount of research has been done about Georgian wines [30–32], to the best of our knowledge, only scarce information about their anti-lipase activity is available [33, 34]. Gulua et al. [33] studied chemical constituents, antioxidant and anti-lipase activity of some wines produced in Georgia. However, this study investigated the lipase inhibitory of six different wines, and it did not bring up any relationship between anti-lipase activity and technological methods.

In this matter, this study aims to investigate fourteen wines made from Georgian autogenous grapes (such as Rkatsiteli and Saperavi), obtained through Kakhetian and European winemaking methods and compare their PL inhibitory activity and polyphenol content and find a relationship, if any, between lipase inhibitory activity and winemaking method.

#### Materials and methods

#### Chemicals

Ascorbic acid, Sodium Hydroxide, Folin-Ciocalteu reagent, Potassium dihydrogen Phosphate,

Olive Oil, Detergent Tween 80, Sodium carbonate, Ethyl acetate, glacial acetic acid and methanol were purchased from Sigma – Aldrich (Steinheim, Germany), TPTZ-2,4,6-Tris (2-pyridyl)-s-triazine (Sigma – Aldrich, Switzerland), hydrochloric acid, formic acid and phosphoric acid were provided by Merck (Darmstadt, Germany), lipase concentrate – HP was purchased from Integrative Therapeutics, LLC (USA). Orlistat® (trade name Xenical) by Roche (Italy) was purchased at the local pharmacy. All other reagents were commercially available at the local market and were of analytical grades.

### Wine Samples

Samples of commercial wines made from autochthonous and leading grape varieties grown in the region of Kakheti were provided from local producers or purchased from wine stores.

A total of 14 red and white wines of the varieties Saperavi (n = 9) and Rkatsiteli (n = 5) were assessed. The wine samples for the experiment were chosen at random. We did it from the consumer's point of view. We did that because, as consumers would do, promising in vivo potent lipase inhibitory activity can be the definitive factor behind consumer decision making. Detailed information about the tested wines is included in Table 1. The wines, packed in glass bottles, were stored at room temperature until being analysed.

To exclude the impact of alcohol on the lipase, de-alcoholised red and white wine samples were used in the lipase inhibition assay, too.

### Winemaking methods

The wines included in this study were either made based on the common "European methods" or by the Kakhetian method. Kakhetian method is one of the methods elaborated in Georgia. This style of wine is based on long period (up to 5 months) maceration and fermentation of must with the usage of 100% of grape pomace (skin, seeds, stems). Fermentation is carried in a clay vessel called "qvevri," buried underground. Qvevri is then sealed, and the wine is left to mature. During fermentation, phenolic compounds are extracted from pomace, defining the composition and essence of Kakhethian wine [26].

### Titratable acidity

Titratable acidity (TA) was determined by titration with 0.1 N sodium hydroxide using an automatic titrator (ZDJ-4A, NASA Scientific Instrument Co., Ltd, Anting Shanghai, China). The TA results were expressed as g of tartaric acid equivalents 1000 mL-1 of the sample [35].

### Total dry extract

For measurement of non-volatile dry matter, a 50 mL sample of wines were aliquoted into a porcelain dish. The dish was then placed onto a boiling water bath until the evaporation of water, alcohol, and other volatile compounds had occurred. The residual moisture was then evaporated from the samples by oven drying at 105°C for 16h. The total dry extract was determined gravimetrically as the residue remaining after drying.

# **Determination of total phenolic content** (TPC)

The total phenolic content (TPC) was determined spectrophotometrically (UV 1609, A&E Lab Co LTD, UK), according to the Folin-Ciocalteu method [36]. Briefly, the diluted samples of each wine (1 mL) were pipetted into separate disposable test tubes and mixed with 5 mL Folin-Ciocalteu phenol reagent (1:10 v/v distilled water). 8 min after, 4 ml of Sodium Carbonate solution (7.5% (w/v)) was added into each test tube. The mixtures were stirred well, and the tubes were allowed to stand for another 60 minutes at room temperature for colour development. Subsequently, their optical densities against the water were read at 765 nm, with a 10 mm path length cell. Diluted Gallic acid (10-50 µg mL-1) was used as a standard working solution. The calibration curve of absorbance vs concentration of a standard solution (equation y = 0.0123x + 0.0236, where Pearson's correlation coefficient:  $R^2 = 0.9918$ ) was used to determine TPC. Results were expressed as mg of gallic acid equivalents (GAE) per litre.

# Chromatographic determinationn of individual polyphenols

Individual polyphenols were separated and quantified by High-Performance Liquid Chromatography (HPLC) analysis performed on a Varian Prostar 500 series liquid chromatography (Varian Prostar 500, Walnut Creek, California, USA). Separation was achieved on a C18, 150 mm x 4.6 mm column (Waters Corporation, Milford, USA). Phenolic compounds were separated on an Acclaim® C18 (4.6 x 250 mm; 5µm) column (Dionex, USA), at 30°C using a temperature-controlled column compartment (TCC-3000). Data acquisition, peak integration, and calibrations were performed with Dionex Chromeleon software (Version 6.80 RS 10). Solvent A was 0.5% acetic acid, and solvent B was 100% methanol. Separation was achieved using the following gradient: isocratic 0% B and 100% A for 0 min; isocratic 40% B and 60% A over 40 min; 0% B and 100% A over 10 min; 0% B and 100% A over 10 min. The flow rate was 0.5 mL min<sup>-1</sup>, and eluent was monitored at 280 nm.

To prepare the sample for analysis, 4.0 mL of the wine sample was carefully deposited onto a C18 solid phase extraction cartridge (Agilent, Bond Elut, USA). Sugars and other polar substances were eluted using 2.0 mL of deionized water through the cartridge, whereas polyphenols were eluted using 2.0 mL of ethyl acetate, which was then evaporated under vacuum at 40-45 °C. Four mL of 50% ethanol was added to the dry extract. The extract was then filtered through 45  $\mu$ m filter paper (Whatman, Maidstone, UK) and 20  $\mu$ L was injected onto the HPLC system.

# Ferric reducing ability of plasma (FRAP) assay for total antioxidant activity

Ferric reducing ability of plasma (FRAP) assay has been applied for the evaluation of the total antioxidant activity (AOA), according to Benzie and Strain, 1996, with slight modifications [37]. The working FRAP reagent was prepared freshly by mixing acetate buffer (300 mM, pH 3.6), 2,4,6- tripyridyl-s-triazine (TPTZ) solution (10 mM, dissolved in 40 mM of HCl) and Ferric Chloride solution (20 mM) in the ratio 10:1:1. The FRAP reagent and ascorbic acid (1mM) were separately incubated for 15 min at 37 0C. 3 mL of working reagent was mixed with 100 microliters of the diluted sample. Ascorbic acid was used as a standard. The reduction was monitored at 593 nm, and the absorbance was recorded after 4 min. FRAP values of samples were compared to that of ascorbic acid and expressed as mg ascorbic acid equivalents (AAE) per 1 litre of wine.

### **Determination of Lipase inhibitory activity**

Titrimetric assay method was used to determine lipase activity as reported by Stoytcheva et al., 2012, with minor modifications [38]. Briefly, the initial reaction mixture consisted of 2.5 mL of deionised water, 1 mL 200 mM Tris HCl buffer (pH 7.2), 3 mL of olive oil, and 0.5 mL of detergent (Tween 80). To obtain a good result, the solution was vigorously mixed on a magnetic stirrer for 15 min. Subsequently, 110 mg of the lipase concentrate was then added to the emulsified mixture, which was then incubated at 37 °C for exactly 30 min. At the end of the incubation, 3 mL of 95% ethanol was added, and the final reaction mixture was titrated with 50 mM NaOH until the value of pH 9 at automatic titrator (ZDJ-4A, NASA Scientific Instrument Co., Ltd, Anting Shanghai, China) was achieved. Blank titration was carried out as above, but potent inhibitors were involved without lipase in test samples. One unit of lipase activity is defined as the amount of enzyme that hydrolyses 1.0 micro equivalent of fatty acid from a triglyceride in one hour at pH 7.2 at 37 °C. Lipase activity was calculated using the following equation:

Lipase Units = 
$$\frac{(A - B) (1000) (2) (DF)}{(1)}$$

where

A = volume of 50 mM NaOH consumed by the test sample in mL;

B = volume of 50 mM NaOH consumed by the blank sample in mL;

1000 = conversion factor from milli equivalents to micro equivalents;

2 = time conversion factor from 30 min to 1 hour;

DF = dilution factor

1 = Volume (in millilitre) of enzyme used

The percentage of inhibition was calculated in the presence and absence of inhibitors. Orlistat was used as a standard inhibitor. Lipase activity was measured in the presence of Orlistat (10mg), and the per cent of inhibition was calculated per 1 mg of Orlistat.

To measure the percentage of lipase inhibition, 1 mL of potent inhibitor (i.e. wine) was added separately to the initial mixture, the following procedures were identical to those described previously. The effect of inhibition of the sample was calculated as the percent of Orlistat inhibition value.

### Statistical analysis

The data represents the mean of a minimum of three replicates ± standard deviation (SD). Data were subjected to the one-way ANOVA and Tukey's HSD tests. One-way analysis of variance (ANOVA) was done to analyse the variation of the means between the experimental samples. Tukey's HSD test was used to differentiate between the mean values. All the analyses were done using XLSTAT (free trial version 2021, Addinsoft, Inc., Brooklyn, NY, USA)

### Results and discussion

Chemical constituents

The characteristics of wines studied herein are shown in Table 1. Wines included in this study were either made based on the common "European methods" i.e. Classic technology or the Kakhetian method, i.e. Qvevri technology. Investigated samples were dry wines, except for late harvest wines. Most of the red wine samples were Mukuzani (S4-S9), Appellation Controlled Origin (AOC) dry red wine, produced from Saperavi grapes grown in the Mukuzani micro-viticulture area Kakheti region.

Table 1. Wine characteristics

| Wine<br>Code | Name of the bottle                              | Producer                 | Vintage | Grape<br>variety | Туре         | Strength % | Making<br>method         |
|--------------|---|--------------------------|---------|------------------|--------------|------------|--------------------------|
| S1           | Glekhuri - Khasmi<br>Saperavi                   | Teliani Valley           | 2017    | Saperavi         | Dry<br>Red   | 13         | Kakhetian                |
| S2           | Matrobela                                       | Matrobela Wines          | 2018    | Saperavi         | Dry<br>Red   | 13.5       | European                 |
| S3           | Icewine - Saperavi<br>- Guramishvilis<br>Marani | Guramishvili's<br>Marani | 2017    | Saperavi         | Sweet<br>Red | 12         | late-harvest<br>European |

|      |                                    | 1  | 1    |            | 1              |      |                          |
|------|------------------------------------|--|------|------------|----------------|------|--------------------------|
| S4   | Mukuzani Valley -<br>Mukuzani      | Mukuzani Valley                            | 2016 | Saperavi   | Dry<br>Red     | 12.5 | European                 |
| S5   | Mukuzani Valley -<br>Mukuzani      | Mukuzani Valley                            | 2019 | Saperavi   | Dry<br>Red     | 12.5 | European                 |
| S6   | Rtvelisi - Mukuzani                | Rtvelisi - Mukuzani Rtvelisi 2018 Saperavi |      | Saperavi   | Dry<br>Red     | 13   | European                 |
| S7   | Zurab Tsereteli -<br>Mukuzani      | Tsereteli Wine and Spirits                 | 2015 | Saperavi   | Dry<br>Red     | 13   | European                 |
| S8   | Zhamurashvili's<br>wine - Mukuzani | Zhamurashvili's wine                       | 2018 | Saperavi   | Dry<br>Red     | 13   | Kakhetian                |
| S9   | Nekresi Estate -<br>Mukuzani       | Nekresi winery                             | 2016 | Saperavi   | Dry<br>Red     | 13   | Kakhetian                |
| RK 1 | Icewine Satrapezo                  | Telavi Wine Cellar                         | 2013 | Rkatsiteli | Sweet<br>White | 10.5 | Late harvest<br>European |
| RK 2 | Rkatsiteli Vine<br>Ponto           | The Spirit of<br>Georgia                   | 2016 | Rkatsiteli | Dry<br>White   | 12.5 | Kakhetian                |
| RK 3 | Mr Rkatsiteli from<br>Gurjaani     | Mr Wine                                    | 2018 | Rkatsiteli | Dry<br>White   | 13   | Kakhetian                |
| RK 4 | Rkatsiteli – Vaziani               | Vaziani company                            | 2016 | Rkatsiteli | Dry<br>White   | 12.5 | European                 |
| RK 5 | Rkatsiteli                         | Kindzmaruli's<br>Marani                    | 2018 | Rkatsiteli | Dry<br>White   | 13   | European                 |

The total acidity varied between 4.791 and 7.986 g L<sup>-1</sup> tartaric acid equivalent for white wines and between 5.25 - 8.794 g tartaric acid equivalent per liter for red wines. The established titratable acidity for Mukuzani wines provided by the legislation of Georgia is no less than 5 g L<sup>-1</sup> (source); all our Mukuzani samples met this requirement.

The highest total dry extract was presented in late-harvest wines,  $100.54 \pm 0.06$  g L<sup>-1</sup> in Saperavi and  $97.24 \pm 0.04$  in Rkatsiteli wine. This can be explained by using naturally dehydrated and completely frozen grape berries in winemaking. Icewine is a type of dessert wine produced from fully ripened grapes that have been frozen while still on the vine. Usually, grapes are left onto wine until the temperature drops below -9 degrees Celsius. This pre-harvest dehydration concentrates the soluble solids in grape berries. As a result, wine is rich in sugars, phenolic compounds and flavour (Moreno et al., 2008) [39].

Among the other samples, white wines made with the European method (Rk 4 and Rk5) had the lowest total dry extract,  $13.46 \pm 0.15$  and  $16.62 \pm 0.18$  g L<sup>-1</sup>, respectively.

S5 (Mukuzani Valley, 2019) and S2 (Matrobela wines) samples showed the highest total polyphenolic content, i.e.,  $3572.358 \pm 187.521$  and  $3482.927 \pm 136.204$  mg GAE L<sup>-1</sup>, respectively. The TPC among the rest of Saperavi grape wines varied between the range of 2415 and 2930 mg GAE L<sup>-1</sup>. Compared to

the other red wine samples, Zurab Tsereteli's Mukuzani contained a statistically significant amount of TPC,  $2415.176 \pm 19.163$  mg GAE L<sup>-1</sup>, which could be caused by the winemaking method or vintage, or both. The TPC of this sample was statistically significant to the white wine sample RK 2, fermented by the Kakhetian method. This is a good example of how the production method can increase the TPC in wine. Usually, consumers consider that white wines do not have polyphenols present in such large quantities as red wine; however, because the Kakhetian method differs from the European method, a white wine made with this method can have comparable TPC to some red wines.

White wines made by the Kakhetian method (RK2 and RK 3) possessed significantly higher TPC than white wines prepared by the common European method (RK1, RK4, RK5). These results are in good agreement with Shalashvili et al. [25], Khatchapuridze et al. [34].

### **Individual polyphenols**

Individual concentrations of 4 phenolic compounds (gallic acid, epicatechin, (+) catechin and caffeic acid, (-)) presented in each wine sample were quantified by HPLC-UV/Vis analysis (concentrations of compounds in all wine samples are shown in Table 2).

**Table 2.** Chemical composition of wines

|      |                   | 1                                |  | 1                                    |                  | 1                         |                                 |                                 |
|------|-------------------|----------------------------------|--|--------------------------------------|------------------|---------------------------|---------------------------------|---------------------------------|
| Code | TA g L-1          | Total<br>dry<br>extract<br>g L-1 | TPC mg<br>Gallic acid<br>equivalent<br>L-1 | acid mg /100mL Epicatechin mg /100mL |                  | Catechin<br>mg/<br>100m L | Caffeic<br>acid<br>mg/100<br>mL | AOA (mg<br>AAE) L <sup>-1</sup> |
| S1   | 6.588 ± 0.154     | 24.34 ± 0.02                     | 2734.959 ± 59.002 bc                       | 0.48 ± 0.02                          | $6.22 \pm 2.03$  | 2.62 ± 0.32               | 2.73±<br>0.19                   | 3397.031 ± 194.837 cd           |
| S2   | 5.776 ± 0.054     | 25.88 ± 0.07                     | 3482.927 ± 136.204 a                       | 0.88 ± 0.04                          | 1 271 + 0.16     |                           | ND                              | 4160.465 ± 126.339 b            |
| S3   | 5.254 ± 0.004     | 100.54<br>± 0.06                 | 1828.455 ± 28.455 e                        | 0.5 ± 0.02                           | $1.65 \pm 0.15$  | $0.5 \pm 0.05$            | 0.9 ±<br>0.07                   | 1921.397 ±<br>119.724 f         |
| S4   | 7.167 ± 0.0435    | 25.92 ± 0.02                     | 2930.081 ± 74.809 b                        | $5 \pm 0.19$                         | $11.5 \pm 0.31$  | 2 ± 0.2                   | 1.8 ± 0.21                      | 3371.412 ± 240.218 cd           |
| S5   | 6.984 ± 0.232     | 30.3 ± 0.01                      | 3572.358 ± 153.111 a                       | 5.26 ± 0.34                          | 0.9± 0.02        | 4.05 ± 0.07               | 10.83±<br>0.31                  | 4729.199 ± 88.162 a             |
| S6   | 7.119 ± 0.301     | 25.80 ± 0.12                     | 2810.840 ± 297.486 bc                      | 7.37 ± 0.14                          | $11.35 \pm 0.51$ | $2.3 \pm 0.12$            | 10.31±<br>0.28                  | 3145.968 ± 113.186 cd           |
| S7   | 7.15 ± 0.362      | 26.7 ± 0.09                      | 2415.176 ± 19.163 cd                       | $3.4 \pm 0.1$                        | $7.62 \pm 0.28$  | 3.67 ± 0.13               | 8.58 ± 0.3                      | 3012.751 ± 119.724 d            |
| S8   | 8.413 ± 0.381     | 29.38 ± 0.07                     | 2813.550 ± 49.823 bc                       | ND                                   | $4.4 \pm 0.13$   | $0.5 \pm 0.01$            | ND                              | 3299.678 ±<br>88.152 cd         |
| S9   | 6.625 ± 0.002     | 30.70 ± 0.01                     | 2965.312 ± 67.152 b                        | 2.3 ± 0.11                           | $9.19 \pm 0.32$  | 1.08 ± 0.02               | 1.37 ± 0.03                     | 3494.381 ± 94.199 c             |
| RK 1 | $7.455 \pm 0.032$ | 97.24 ± 0.04                     | 149.594 ± 8.13 g                           | 58.13 ± 0.45                         | $0.2 \pm 0.001$  | 1.78 ± 0.09               | $1 \pm 0.02$                    | 210.073 ± 28.984 g              |
| RK 2 | 7.932 ± 0.055     | 27.48 ± 0.01                     | 2515.477 ± 97.561 de                       | 4.02 ± 0.2                           | $11.38 \pm 0.43$ | 3.87 ± 0.22               | 11.17 ± 0.35                    | 2413.275 ± 43.476 e             |
| RK 3 | 4.961 ± 0.057     | 20.36 ± 0.02                     | 1572.358 ± 56.912 e                        | 13.6 ± 0.03                          | $8.39 \pm 0.26$  | 4.52 ± 0.15               | 8.03 ± 0.23                     | 1788.181 ± 69.123 f             |
| RK 4 | 4. 909 ± 0.118    | 13.46 ± 0.15                     | 489.577 ± 36.112 f                         | 0.4 ±<br>0.001                       | $0.4 \pm 0.001$  | $2.9 \pm 0.11$            | 6.78 ± 0.26                     | 179.330 ± 50.722 g              |
| RK 5 | 6.897 ± 0.226     | 16.62 ± 0.18                     | 190.244 ± 8.13 g                           | $1 \pm 0.01$                         | ND               | $2.2 \pm 0.04$            | 9.16 ± 0.32                     | 199.825 ± 43.476 g              |

Means  $\pm$  standard deviation (SD) in the same column with different alphabet letters indicate the significant difference at p < 0.05.

Due to the lack of corresponding standards, other individual polyphenols were not identified within this study. A reverse-phase HPLC separation profile of Mukuzani wine (Mukuzani Valley, 2016), is shown in Fig 1.

<sup>&</sup>quot;ND" individual polyphenol was not detectable in the sample.

S1- Glekhuri, Khasmi Saperavi; S2 - Matrobela, Saperavi; S3 S3 - Icewine, Guramishvilis Marani, Saperavi; S4 -Mukuzani Valley, Mukuzani (2016); S5 - Mukuzani Valley, Mukuzani (2019); S6 - Rtvelisi, Mukuzani; S7 - Zurab Tsereteli, Mukuzani; S8- Zhamurashvili's wine, Mukuzani; S9 - Nekresi Estate, Mukuzani; RK1 - Icewine Satrapezo, RK2 - Vine Ponto, Rkatsiteli; RK3 Mr Rkatsiteli from Gurjaani; RK4 - Vaziani, Rkatsiteli; RK5 Rkatsiteli;

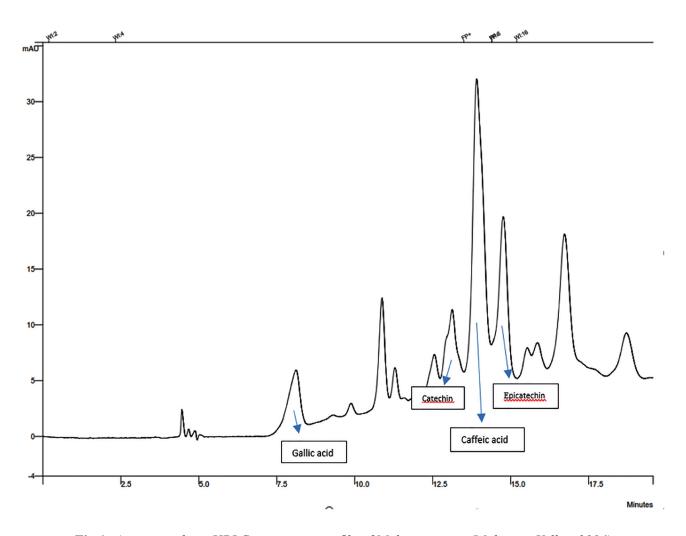


Fig 1. A reverse-phase HPLC separation profile of Mukuzani wine (Mukuzani Valley, 2016)

Highly significant differences in the four individual polyphenolic compounds quantified were observed in the wine samples analysed in this study. A high concentration of gallic acid could be found in Satrapezo Icewine, RK1 (58.13  $\pm 0.46$  mg / 100 mL), among the rest of Rkatsiteli samples, wines made with Kakhetian method (RK 2 and RK3) contained significantly higher amount of gallic acid (4.02  $\pm$  0.02 and 13.6  $\pm$  0.03 mg / 100 mL, respectively). Gallic acid was not detectable in Mukuzani Zhamurashvili's wine, S8 and in the rest of the Mukuzani samples, its content was varying between 2.19 - 7.52 mg /100 mL.

The highest caffeic acid content was presented in the following wines Rkatsiteli Vine Ponto 2016 (11.17 $\pm$ 0.16 mg / 100 mL) and Mukuzani Valley 2019 (10.83 $\pm$ 0.12 mg /100 mL), with no significant difference.

The concentration of catechin was varying between 0.5 - 4.52 mg/100 mL The highest content of epicatechin was found in wine S5 (Zhamurash-

vili Valley 2016) (11.5  $\pm 0.1$  mg /100 mL) RK2 (Rkatsiteli Vine Ponto samples) (11.38  $\pm 0.15$  mg /100 mL) and S6 (Rtvelisi) (11.35  $\pm 0.16$  mg /100 mL).

Samples S9, RK3, S7 and S1, contained intermediate levels of epicatechin, i.e.,  $9,19 \pm 0.1$ ;  $8.39 \pm 0.1$ ;  $7.62 \pm 0.1$ ;  $6.22 \pm 0.1$  mg /100 mL, respectively. Epicatechin was not detectable in the RK 5 sample. In general, higher content of catechin and epicatechin was found in white wines made by the Kakhetian method than those prepared by the European method. These are the main flavonoids found in the skin and seed of grapes [32,33]. Probably due to this reason, wines made with the Kakhetian method resulted in richer wines. Samples S4 and S5 are made from the same producer, by the same technology and are different by vintage. As seen, the content of gallic acid, caffeic acid and catechin is decreased by vintage, whereas the content of epicatechin is significantly higher.

### **Antioxidant activity**

White wines were significantly less effective in antioxidant assays than red wines. However, wines fermented using the Kakhetian method exhibited higher antioxidant activity and were comparable to some red wines made by the European method.

The wine S5 exhibited the highest AOA, 4729.199  $\pm$  88.162 mg AAE L<sup>-1</sup>. Wine S2 showed the second-highest antioxidant activity, 4160.465  $\pm$  126.339 mg AAE L<sup>-1</sup>. These two wines were statistically similar regarding TPC. No statistically significant difference between the samples S9, S1, S4, S8 and S6 was detected. The observed antioxidant activity was 3494.381  $\pm$  94.199, 3397.031  $\pm$  194.837. 3371.412  $\pm$  240.218; 3299.68  $\pm$  88.152; 3145.968  $\pm$  113.186 mg AAE L<sup>-1</sup>, respectively. The sample S7 showed no statistically significant difference compared

to the sample S6, which was  $3012.751 \pm mg$  AAE L<sup>-1</sup>. The antioxidant activity of white wine samples, RK2 ( $2413.275 \pm 53.247 mg$  AAE L<sup>-1</sup>), was higher than red wine sample S3 ( $1921.397 \pm 146.631 mg$  AAE L<sup>-1</sup>). Moreover, the AOA between these samples was statistically significant. However, the AOA of this late-harvested Saperavi wine was statistically similar to that of RK 3 ( $1788.181 \pm 84.658 mg$  AAE L<sup>-1</sup>). As stated earlier, wines fermented by the European method, RK1, RK5 and RK4 showed the lowest antioxidant activity; results were statistically similar and varying between 179 and 211 mg AAE L<sup>-1</sup>.

We could demonstrate a high correlation ( $R^2$ = 0.9731) between the total polyphenol content and the antioxidant activity of wines (Fig 2). A significantly positive correlation was also reported other researches Han et al. [40], Paixão et al. [41].

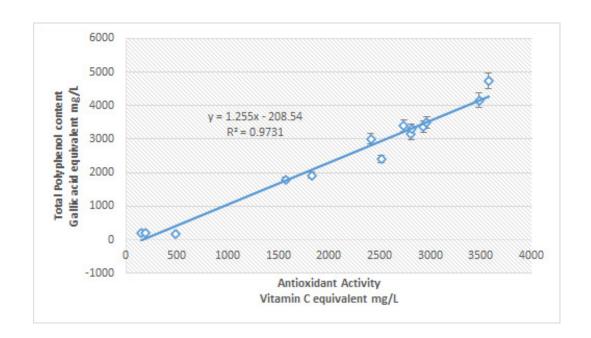


Fig. 2. Correlation between the total phenolic content and antioxidant activity

### Lipase inhibition

Lipase activity and its inhibition by wines are shown in the Table 3. The highest level of lipase inhibition among the given samples was exhibited by the sample RK 3, 82.63% mL<sup>-1</sup> of wine. This white wine sample was made by Qvevri winemaking method. It is notewor-

thy, that wines from Mukuzani microzone showed high anti lipase activity; their lipase inhibitory activity ranged from 77.12 to 79.7 8% mL<sup>-1</sup> of wine. No significant difference was observed among these samples. Orlistat (10mg) itself showed 75.84% inhibition of lipase activity.

**Table 3.** *Lipase activity in the presence of various inhibitors* 

| Inhibitor | Lipase activity               | Inhibition %                | Inhibition % based on 1 mg inhibitor | Effect of 1 mg inhibitor as<br>the percent of 1 mg Orlistat<br>inhibition value |
|-----------|-------------------------------|-----------------------------|--------------------------------------|---|
| Orlistat  | $487.67 \pm 21.142$           | 75.84                       | 7.58                                 | 100   |
| S1        | 422.84 ± 17.26 fg             | $79.05 \pm 0.86 \text{ ab}$ | $3.25 \pm 0.04 \text{ b}$            | $42.82 \pm 0.46 \ b$  |
| S2        | 562.71 ± 22.97 cd             | $72.13 \pm 1.14$ de         | $2.787 \pm 0.04 \ def$               | $36.75 \pm 0.58 \text{ def}$  |
| S3        | $504.63 \pm 20.60 \text{ de}$ | $75 \pm 1.02 \text{ cd}$    | $0.75 \pm 0.01$                      | $9.84 \pm 0.13 \text{ g}$   |
| S4        | $461.81 \pm 18.85 \text{ ef}$ | $77.12 \pm 0.90 \text{ bc}$ | $2.98 \pm 0.04 \text{ cd}$           | $39.23 \pm 0.48 \text{ cd}$   |
| S5        | 422.21 ± 17.24 fg             | $79.09 \pm 0.85$ ab         | $2.61 \pm 0.03$                      | $34.41 \pm 0.37 \text{ ef}$   |
| S6        | 409.41 ± 16.71 fg             | $79.72 \pm 0.83$ ab         | $3.09 \pm 0.03 \ bc$                 | $40.74 \pm 0.42 \text{ bc}$   |
| S7        | $449.64 \pm 18.36$ ef         | $77.73 \pm 0.91$ bc         | $2.91 \pm 0.03$ cd                   | $38.38 \pm 0.45 \text{ cd}$   |
| S8        | $444.72 \pm 18.16$ ef         | $77.97 \pm 0.90 \text{ bc}$ | $2.65 \pm 0.03 \text{ ef}$           | $34.99 \pm 0.40 \text{ ef}$   |
| S9        | 408.14 ± 16.66 fg             | $79.78 \pm 0.83 \text{ ab}$ | $2.60 \pm 0.03 \text{ f}$            | $34.27 \pm 0.35 \text{ f}$  |
| RK 1      | 593.74 ± 24.24 c              | $70.59 \pm 0.71$ e          | $0.73 \pm 0.01 \text{ g}$            | $9.57 \pm 0.16 \text{ g}$   |
| RK 2      | $469.50 \pm 19.17 \text{ ef}$ | $76.74 \pm 0.95 \text{ bc}$ | $2.793 \pm 0.03 \text{ de}$          | $36.82 \pm 0.de$  |
| RK 3      | $350.57 \pm 14.31 \text{ g}$  | $82.63 \pm 0.71$ a          | $4.06 \pm 0.03$ a                    | $53.51 \pm 0.46a$   |
| RK 4      | 910.25 ± 37.16 a              | $54.91 \pm 1.84 \text{ g}$  | $4.08 \pm 0.14$ a                    | $53.79 \pm 1.80a$   |
| RK 5      | $708.39 \pm 28.92 \text{ b}$  | 64.91 ± 1.43 f              | $3.91 \pm 0.09$ a                    | $51.49 \pm 1.14a$   |

Means  $\pm$  standard deviation (SD) in the same column with different alphabet letters indicate the significant difference at p < 0.05.

S1- Glekhuri, Khasmi Saperavi; S2 - Matrobela, Saperavi; S3 S3 - Icewine, Guramishvilis Marani, Saperavi; S4 - Mukuzani Valley, Mukuzani (2016); S5 - Mukuzani Valley, Mukuzani (2019); S6 - Rtvelisi, Mukuzani; S7 - Zurab Tsereteli, Mukuzani; S8- Zhamurashvili's wine, Mukuzani; S9 - Nekresi Estate, Mukuzani; RK1 - Icewine Satrapezo, RK2 - Vine Ponto, Rkatsiteli; RK3 Mr Rkatsiteli from Gurjaani; RK4 - Vaziani, Rkatsiteli; RK5 Rkatsiteli;

The lowest lipase inhibitory activity was shown by the sample RK4 54.91% per mL wine. RK4 belongs to the class of dry white wines made with classic European technology. This sample also contained a low amount of dry extract, TPC and AOA compared to the other samples. The second sample with a low lipase inhibitory activity was also a white wine sample, RK5 (64.91%). The amount of TPC in it was the lowest compared to other samples - 190.24 mg GAE L-1. At an average rate, white wines made by the classical technology possessed lower anti-lipase activity (69.96% per ml of wine) compared to red wines (77.51% per ml of wine). However, white wines made with Qvevri technology (RK1 and RK 2) showed higher lipase inhibitory activity than Saperavi samples (S2, S3), made with the European

winemaking method. No significant difference was observed among the other Saperavi samples, which differed by the winemaking method.

No significant correlation was found between polyphenol content and anti-lipase activity in wine samples, nor between lipase inhibitory activity and winemaking method. In a previously published study, the R<sup>2</sup> correlation value between lipase inhibitory activity and TPC was also found to be low [34].

Some differences in the results were observed when calculating lipase inhibitory activity per mg dry extract. The highest level of lipase inhibition was shown by the white wine samples (RK 4, RK 3 and RK 5) and no statistically significant differences existed between the samples. A significant relationship was found between late harvest wines (S3 and RK 1), and they exhibited the lowest percentage of inhibition (0.75% and 0.73%) due to the high content of the dry extract. The anti-lipase activity of Orlistat® itself calculated per mg was equal to 7.58 %.

From obtained results, one can calculate the optimal dose required to achieve the same results as the intake of 120mg Orlistat gives. However, further studies are needed to define the mechanism of action and kinetics of inhibition in the wine samples.

### Conclusion

In white wines produced by the Kakhetian method, antioxidant activity and TPC is significantly higher than in the white wines prepared by the common European method. No such effect was found in the samples of red wines. Probably because most of the studied samples of red wines belong to the Appellation of Controlled Origin and the Saperavi grapes grown in the Mukuzani micro-viticulture area are rich with polyphenols.

No significant correlation was found between polyphenol content and anti-lipase activity in wine samples, nor between lipase inhibitory activity and winemaking method.

Overall, it can be concluded that most of the wine samples we have examined, are characterised by noticeable or significant high anti-lipase activity. Regarding the results, we may conclude that the wines on the Georgian market made from local cultivars, i.e. Saperavi (red) and Rkatsiteli (white), are characterised by high anti-lipase and antioxidant activity and high polyphenol content.

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### Determining the likelihood of emerging illegal dumpsite on the example of Georgia

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### ABSTRACT

On the example of the Mtskheta-Tianeti highland rural region of Georgia, the probability of illegal landfills is studied using the math-ematical equation developed within the research. Statistical and experimental data were collected and processed to calculate the likelihood of illegal landfills as a result of surveys and visual inspections of municipal services and residents. As a result of airbrushing and visual field work, the results of determining the probability of illegal landfills in the study area obtained by theoretical calculations were verified. It is established that the theoretically obtained results, which depend on the reliability of the data available in the municipality, are ingood agreement with the real picture. It is also estimated that, on average, the probability of illegal landfills in the study highland rural areas of five municipalities in the Mtskheta-Mtianetiregion varies from 0.1 to 1.6 units per village.

*Keywords:* Illegal Dumpsite, Likelihood of Emerging, Identification, Equation, Highland rural settlements, Mtskheta-Mtianeti region.

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### Introduction

The uncontrolled occurrence of illegal dumpsites in populated areas is a global problem. It is especially relevant for Georgia as local governments are unable to provide appropriate services in the regions. This problem becomes even more urgent for the high mountainous rural regions.

Georgia's highmountainous regions are characterized by significant structural weaknesses compared to the lowlands and urban areas - weak economic diversification, population migration, extreme poverty, weak infrastructure, lack of health care facilities, limited access to public services, and more. 66% of the territory of Georgia consists of mountainous areas, where 6.5% of the country's population lives permanently [1,2]. The Mtskheta-Mtianeti region is particularly interesting in this regard, as 75% of the villages in the region (≈450)

units) belong to high mountainous villages, and the population living in these villages is 22% of the total population of the region [3].

Mtskheta-Mtianeti region is located in eastern Georgia and includes the following historical-geographical provinces: Khevi, Ertso-Tianeti, Pshavi, Khevsureti, Mtiuleti, Gudamakari and partly Ksniskhevi. It is bordered on the north by the republics of the Russian Federation: North Ossetia, Ingushetia and Chechnya, on the east by the Kakheti region, on the south by Kvemo Kartli, and on the west by Shida Kartli. Mtskheta-Mtianeti region is a heterogeneous and, therefore, quite specific terrain, of which more than 4/5 belongs to the high mountainous category. The administrative center of the region is Mtskheta, and the region consists of the following administrative units: Akhalgori Municipality, Dusheti Municipality, Tianeti Municipality, Mtskheta Municipality, Kazbegi Municipality. The total area of the Mtskheta-Mtianeti region is 5.8 thousand sq. km. (8.3% of the total territory of Georgia), and the population in recent years is 94 3701 people (2.53% of the population of Georgia); Population density - 1 sq. Km. 16.3 men per km [4].

Today, 40% of the region's population lives in the territory of Mtskheta Municipality, and the lowest, 5% - in Kazbegi Municipality (Table 1) [5]. As mentioned, the share of rural population in the total population of the region is 75%. The region is characterized by an abundance of small villages. There is only one village in the whole region with 5,000 inhabitants (Mukhrani village of Mtskheta municipality, which does not belong to the category of high mountainous villages) [3].

### Objectives and methods

The aim of the study was to develop and use an integrated methodology for determining the probability of illegal landfills in the border, highland rural regions of Georgia.

The research methodology is an innovative approach that involves determining the likelihood of illegal landfills in a geographically complex area of the country (particularly highland villages) based on statistics provided by municipalities. The results obtained using the developed theoretical methodology were verified by visualization (using a remote-controlled unmanned aerial vehicle-drone and with the help of expedition work) and a population survey. Based on the collection of statistical data on the study areas, relevant municipalities and population, the probability of illegal landfills was determined using a mathematical formula developed by us(1). This formula allows to determine the approximate amount of waste left without service in the study area and, consequently, to determine the likelihood of the existence / occurrence of an illegal landfill / landfills. In order to collect data, questionnaires were compiled and used for the representation of both the population and the municipality representatives, which were filled out on the basis of the

on-site (expedition) survey. In particular, the questionnaire was filled with the following data - population, availability of specific service / frequency / efficiency, number of containers (by size), amount of waste per capita, presence of waste recycling, etc. (Table 2).

D= $(AC-(Ln(L)+Sn(S))f(q/h))/AC \cdot 100\%$  (1) where.

D- Percentage generating possible illegal dumpsite;

A- amount of waste per capita;

C- population;

L- Large container capacity, kg;

n (L) - number of large containers, pieces;

S- small container capacity, kg;

n (S) - number of small containers, pieces;

f- collection frequency (frequency of waste truck service per week), coefficient from 0 to 1;

q- collection efficiency (ratio between the result achieved and the resources used, which implies the actual state of waste collection), coefficient from 0 to 1;

h - the relative rate of container overload, a ratio of 1 to 10, which shows how full the container is at the time of its emptying, and which depends on the population and the number of containers).

Parameter A: From 2015 to 2017, using the gravimetric method, we examined the amount of solid household waste (SHW) per capita and its morphological composition, which was supported by the Shota Rustaveli National Science Foundation (SRNSFG) [6] project "Development of a methodology for determination of amounts and morphological composition of municipal solid waste and creation of a data base" #FR/88/9-220/14 (Table 1) [7-10]. Accordingly, the average quantity of municipal solid waste (MSW) per capita living in cities in Georgia is 250 kg / person / year, and 75-80 kg / person / year per capita living in the region. This sharp difference, as already mentioned above, is due to the difference in living conditions and peculiar standards.

**Table 1**. Morphological Composition of Municipal Solid Waste in Georgia

| # | Regions                   | Glass | Paper | Metal | Plastic | Nappies | Rubber /<br>Leather/<br>Textile | Wood/<br>green<br>waste | Fine<br>Fraction | Hazardous | Food | Total |
|---|---------------------------|-------|-------|-------|---------|---------|---------------------------------|-------------------------|------------------|-----------|------|-------|
| 1 | Tbilisi                   | 3.5   | 13.2  | 1.7   | 15.2    | 8.0     | 2.2                             | 0.7                     | 3.7              | 0.8       | 51.1 | 100.0 |
| 2 | Ajara                     | 2.8   | 15.5  | 2.5   | 15.4    | 7.2     | 1.7                             | 0.2                     | 9.8              | 0.8       | 44.1 | 100.0 |
| 3 | Guria                     | 2.2   | 11.6  | 3.5   | 15.5    | 5.7     | 0.4                             | 0.5                     | 17.7             | 0.8       | 42.2 | 100.0 |
| 4 | Samegrelo-Zemo<br>Svaneti | 2.6   | 9.9   | 1.9   | 12.1    | 7.0     | 1.2                             | 1.5                     | 19.5             | 0.8       | 43.8 | 100.0 |

| 5  | Racha-Lechkhumi    | 2.4 | 13.5 | 1.5 | 13.2 | 9.0 | 1.3 | 0.2 | 16.5 | 0.4 | 42.0 | 100.0 |
|----|--------------------|-----|------|-----|------|-----|-----|-----|------|-----|------|-------|
| 6  | Imereti            | 0.5 | 9.2  | 0.3 | 1.9  | 9.1 | 1.0 | 0.1 | 29.2 | 0.6 | 48.2 | 100.0 |
| 7  | Mcxeta-Mtianeti    | 1.5 | 9.9  | 3.8 | 15.5 | 5.9 | 1.8 | 0.7 | 16.0 | 1.0 | 43.9 | 100.0 |
| 8  | Shida Kartli       | 3.1 | 11.3 | 2.8 | 15.6 | 5.2 | 0.5 | 0.7 | 16.7 | 0.3 | 43.7 | 100.0 |
| 9  | Kvemo Kartli       | 0.7 | 11.3 | 3.0 | 13.8 | 7.9 | 1.6 | 0.4 | 19.2 | 0,7 | 41.4 | 100.0 |
| 10 | Samtskhe-Javakheti | 2.8 | 12.0 | 3.2 | 13.8 | 6.8 | 0.8 | 0.8 | 10.8 | 0.4 | 48.7 | 100.0 |
| 11 | Kakheti            | 2.8 | 11.2 | 2.9 | 11.5 | 5.0 | 1.0 | 0.3 | 22.6 | 0.5 | 42.3 | 100.0 |
| 12 | Average            | 2.4 | 11.9 | 2.7 | 14.2 | 6.8 | 1.3 | 0.6 | 15.3 | 0.8 | 44.1 | 100.0 |

The parameters C, L, S, n (L), n (S) provided and subsequently determined by the specialists of the municipal service and as a result of a population survey.

Table 2 presents an example of a questionnaire compiled for municipalities to determine the above parameters.

 Table 2. Questionnaire for Municipal Service

| #  | 1   | 2                |
|----|---|------------------|
| 1  | Municipality  | Name             |
| 2  | Population  | Person           |
| 3  | Population which have not served of waste disposal service    | Person/household |
| 4  | Number of villages  | Units            |
| 5  | Number of high mountain villages                              | Units            |
| 6  | The least populated high mountain villages                    | Units and Name   |
| 7  | High mountain villages with the densest settlements           | Units and Name   |
| 8  | High mountain villages where there is no road infrastructure  | Units and Name   |
| 9  | Total number and type of waste containers (volume)            | Units and Name   |
| 10 | High mountain villages where there are not waste containers   | Units and Name   |
| 11 | High mountain villages where there is no waste truck          | Units and Name   |
| 12 | Total number of waste trucks                                  | Units and Name   |
| 13 | High mountain villages where illegal dumpsites are registered | Units and Name   |

Table 3 shows the gradations of the parameters f, q and h in Formula 1, which are selected during the calculations by visual evaluation at the study site.

**Table 3.** Ratios Classification of Waste Collection Frequency and Waste Collection Efficiency

| f   | Waste Collection Frequency | q   | Waste Collection Efficiency | h    | Relative rate of container overload |
|-----|----------------------------|-----|-----------------------------|------|-------------------------------------|
| 0   | Not                        | 0   | Not                         | 0    | There is no container               |
| 0.2 | Once a week                | 0.2 | Very bed                    | 1    | Too overloaded                      |
| 0.4 | Twice a week               | 0.4 | Bed                         | 2    | Overloaded                          |
| 0.6 | Every second day           | 0.6 | Satisfactorily              | 4    | Full                                |
| 0.8 | Every day                  | 0.8 | Good                        | 6    | Incompletely filled                 |
| 1   | Twice a day                | 1   | Very good                   | 8-10 | Less than half / insignificant      |

Additional information about the survey region was obtained with the help of the online resource of the National Statistics Office of Georgia [11], (Table 4).

Currently, about 1/6 of Mtskheta Mtianeti - the entire territory of Akhalgori Municipality is occupied by the Russian Federation and information about it is not officially found (Map 1). A total of 601 villages are registered in the regions controlled by Georgia, of which 448 are mountainous villages, the total population of the municipality is 102,044

people (in the case of Akhalgori municipality according to the Geostat data of 2008), territorial area: 6 785 km2. The share of the population using the municipal cleaning service is 65%, which is served by three official dumpsites.

The average result of the calculation by our proposed formula for the individual regions studied is given in Table 4, which shows that the average likrlihood of illegal dumpsites in the high mountainous rural areas for the study region is 52%. In addition, significant differences were identified be-

tween municipalities in terms of waste management (Table 4). Table 4 shows that the situation is more difficult in the mountainous villages of Dusheti and Mtskheta municipalities, where the result is more

than 50%, while the situation is better in Kazbegi municipality, which is mainly explained by the influence of tourism policy and responsible local government work.

### Results and analysis

**Table 4.** Calculation of Likelihood of Emerging Illegal Dumpsites in High Mountainous Rural Areas of Some Border Region of Georgia

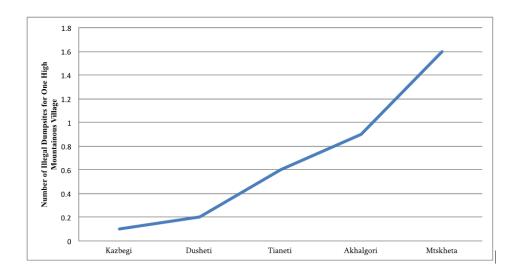
|  |                   |   |                           |         |         | Index   | of Regions |           |        |
|--|-------------------|---|---------------------------|---------|---------|---------|------------|-----------|--------|
| Parameters   | Measurement       |   | Symbol                    | Dusheti | Tianeti | Kazbegi | Mtskheta   | Akhalgori | Total  |
| Quantity of MSW  | kg/Person/<br>day |   | A                         | 0.3     | 0.3     | 0.3     | 0.3        | 0.3       | 0.3    |
| Quantity of village  | Units             |   | -                         | 289     | 87      | 47      | 62         | 116       | 601    |
| Quantity of high mountain village  | Units             |   | -                         | 242     | 87      | 47      | 19         | 53        | 448    |
| Population   | Person            |   | С                         | 9100    | 7700    | 2469    | 1 632      | 1500      | 22 401 |
| Residents of<br>high mountain<br>village who are<br>not provided with<br>a waste disposal<br>service | Person            |   | -                         | 6777    | 664     | 0       | 1632       | 1500      | 10573  |
| Value of big containers  | kg                |   | L                         | 220     | 220     | 220     | 220        | 220       | 220    |
| Quantity of big containers   | Units             |   | n(L)                      | 90      | 60      | 40      | 0          | 0         | 190    |
| Value of small containers  | kg                |   | S                         | 20      | 20      | 20      | 20         | 20        | 20     |
| Quantity of small containers   | Units             |   | n(S)                      | 0       | 0       | 0       | 0          | 0         | 0      |
| Waste Collection<br>Frequency  | 0-1               |   | f                         | 0.2     | 0.2     | 0.6     | 0          | 0.2       | 0.4    |
| Waste Collection<br>Efficiency   | 0-1               |   | q                         | 0.3     | 0.5     | 0.8     | 0          | 0.2       | 0.4    |
| Relative indicator of container load   | 1-10              |   | h                         | 1       | 1       | 6       | 0          | 1         | 1.8    |
| Total quantity of emerging waste   | kg/day            | В | AC                        | 2730    | 2310    | 740.7   | 489.6      | 450       | 6720.3 |
| Total quantity of disposal waste   | kg/day            | Е | L*n(L)+S*n<br>(S)*f*(q/h) | 1188    | 1320    | 704     | 0          | 0         | 3212   |
| Total quantity of remaining waste  | kg/day            | Н | В-Е                       | 1542    | 990     | 36.7    | 489.6      | 450       | 3508.3 |
| Likelihood of<br>Emerging Illegal<br>Dumpsites   | %                 | D | H/B*100                   | 56.48   | 42.85   | 4.95    | 100        | 100       | 52.20  |

By entering some of the values taken from Table 4 into Equation 1, we obtain the corresponding results for the likelihood of emerging an illegal dumpsite, according to the districts we have selected. The results show which of them is more unsatisfactory or better, and also where and what type of work needs to be done to improve the current situation.

The results can be presented as follows - for example: in the example of Kazbegi in 47 high mountainous villages (MMS) we got the likelihood of

only 5 illegal dumpsites, if we recalculate these values for one village (or how many dumpsites can be emerging in one village), we get that in one village Generate only 0.1 dumpsites; According to the relevant data - the likelihood of creating a dumpsite in one of the high mountainous villages of Dusheti district is 0.2 dumpsites; 0.6 dumpsites in one village of Tianeti, 0.9 dumpsites in Akhalgori, 1.6 dumpsites in Mtskheta.

The obtained results are well presented in Fig.



**Fig.** Likelihood of Quantitative Emerging of Illegal Dumpsites for One High Mountainous Village (Example of Mtskheta-Mtianeti Region) The results obtained are well presented in Graph 1.

### Conclusion

- 1. It is established that the results obtained using Equation-1 (with their values) are descriptive and fairly close to the real picture observed by the fieldwork team in the field of the project.
- 2. The number of illegal landfills that may arise in each municipality has been reduced to one village. Accordingly, it was determined how many illegal landfills are likely to occur in a single village in a particular district. The results show that this figure ranges from 0.1 to 1.6 units.
- 3. Based on the data obtained from the study, the most satisfactory situation is in Kazbegi region, while the situation is relatively poor in the villages of Akhalgori and Mtskheta regions, where the local government pays less attention to the population living in these

mountainous villages. In particular, they are not provided with all the necessary services needed to solve these existing problems

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# Formation of yield and quality of vegetable crops when using a biological preparation in the conditions of dark chestnut soils of the south-east Kazakhstan

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### ABSTRACT

The study of the biological product "BioEcoGum" on vegetable crops (salad, cabbage, broccoli, beans). It has been established that the varietal reaction during treatment with a biopreparation is unusual. The selective feature of vegetable crops is noted. Changes in the agrophysical properties of soil under the influence of preparations of biological origin during the cultivation of vegetables were revealed. The analysis of vegetable crops treated with a biological product in terms of yield, growth rates of plants in comparison with the control was carried out. The use of a biological product in the Almaty region of the southeast of Kazakhstan is promising in order to increase the adaptability and productivity of plants. Studies have found that having a positive effect on the growth and development of plants, improving the physiological processes in the plant organism, increasing the immunity and stress resistance of lettuce, broccoli cabbage, beans, biological product contributed to an increase in yields and product quality.

Keywords: Biological product, Lettuce, Broccoli, Beans, Plant productivity, Product quality.

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### **INTRODUCTION**

The use of a biological product is explained by the exacerbation of a whole complex of negative phenomena in agricultural production. As a result of the use of chemical plant protection products, the most important of which are various forms of resistance of phytophages to the main groups of widely used pesticides, and as a consequence, the continuing increase in pesticide pressure, disruption of agrobiocenotic relations and a general deterioration of the ecological situation.

The solution to the problem is possible when biological methods are included in the plant protection system, characterized by various

methods of impact on harmful populations - from the use of entomophages to the use of biological products. The cultivation of vegetables and the use of the crop for the preparation of baby and dietary food makes it necessary to use environmentally friendly preparations in protective measures. The first developments in the use of a biological product against pests of vegetable crops were carried out in 2020 on the territory of the experimental site of the UPH "Agrouniversitet". The use of biologics is already recognized in many countries around the world. Increasing yields and improved soil conditions are the best evidence for the quality of humic fertilizers. Refusal from the use of chemical (synthesis) pesticides, and the transition to new organic fertilizers will help preserve soil fertility and grow high-quality and healthy products without threatening the environment and human health. The main way to increase the productivity of agricultural crops is to apply high-quality fertilizers. Excessive use of mineral fertilizers and chemical means of protection can have a negative impact on soil fertility, the ecological state of the environment, and the quality of cultivated products. The most important role in increasing soil fertility and improving the quality of crop products belongs to fertilizers and preparations of a biogenic nature, the development and implementation of which are engaged in all over the world. Vegetables occupy the most important place in the human diet, as they are a source of vitamins, carbohydrates, organic acids, trace elements necessary to meet human physiological norms. Vegetables are the main source of vitamins, mineral salts, organic acids, aromatic and other substances, without the use of which the normal physiological activity of the human body is inconceivable. The great Russian physiologist I.P. Pavlov wrote about them: "A person can extend his life by at least a third if he eats fresh vegetables every day" [1].

Vegetable growing is an important branch of agriculture, designed to provide the population and the processing industry of the republic with full-fledged, balanced food products and high-quality raw materials all year round. According to the normative data of the Kazakh Academy of Nutrition, 120 kg of vegetables, 100 kg of potatoes and 26 kg of melons should fall on 1 inhabitant of the republic per year [2].

In the southeastern zone of Kazakhstan, with an abundance of solar heat and the availability of irrigation water, vegetable products allow you to get a good harvest and can be highly profitable. With a high potential, the productivity of the current state of vegetable growing in Kazakhstan remains low. It is known that the main factor in increasing the productivity of vegetable crops was and remains the assortment, which is being improved both through the introduction of varieties based on soil-climatic analogues, and by breeding and introducing genotypes created by combinative selection methods on a genetic basis [3].

Today in our country the area of vegetable crops planting is about 150 thousand hectares, new territories are being developed, serious work is being done by scientific workers of the research institute. About 65% of the sown area of vegetable crops is concentrated in the southeast of the country [4].

When creating highly productive crops of vegetables, it is especially important to switch the industry to highly efficient, low-cost, energy-saving technologies that ensure the maximum use of environmental resources, affecting the good survival rate of seedlings and high yields of vegetables.

The development and implementation of such biological products in the post-Soviet space is carried out by various SRI independently or with the participation of commercial organizations. A biological product that has a complex protective and stimulating effect on plants that increase productivity and product quality, was developed by the U.U.Uspanov Kazakh Research Institute of Soil Science and Agrochemistry. Natural preparation of biological origin: increases the immunity of plants to diseases (late blight, alternaria, cercospora, white rot, etc.) and extreme environmental conditions (soil salinity, alkali-forming factors, pesticides, drought, low soil and water temperatures, frosts, etc.), limiting productivity of agrocenoses;

- promotes enhanced growth of roots and aboveground parts of plants;
- accelerates the ripening of vegetables and potatoes;
- increases the yield of vegetable crops by 15-30%;

improves the biological and nutritional value of the manufactured products, as well as extends the shelf life.

The effectiveness of bioorganic fertilizers is also manifested in cereals and legumes. The use of humic biological products based on vermicompost together with microelements and growth stimulants helps to increase the yield of winter wheat, corn and soybeans, improves soil fertility, reduces the cost of their cultivation, and also allows you to get environmentally friendly products [5,6].

However, in the southeastern zone of Kazakhstan, which is characterized by its soil and climatic characteristics, there are practically no such works. Until recently, there were almost no recommendations in the region on a scientifically grounded system for introducing biological products into vegetable crop rotations, taking into account not only their effect, but also the aftereffect on subsequent crops in the crop rotation, and especially if we take into account the healing properties of vegetables and their effect on the general condition of the body.

Thus, the use of the domestic biological product "BioEcoGum" based on biohumus prompted to look for their own approaches to the problem and ways to solve it, taking into account the already accumulated domestic and foreign experience. The strength of scientific work is a multidimensional research approach. This is at the same time control over the reclamation of contaminated soils using a natural preparation of biological origin, the quality of cultivated vegetable products and the preservation and restoration of soil fertility. The biological product increases the defense mechanism of plants against the action of unfavorable factors, does not pose a threat to disturb the ecological balance in the biosphere, and plays an essential role in the anti-resistance strategy. The use of the biological preparation "BioEcoGum" is becoming more and more economically profitable and environmentally expedient.

### MATERIALS AND METHODS

The research was carried out in the UPH "Agrouniversity" of the Almaty region. The object of the study is dark chestnut soils, vegetables (lettuce, broccoli, beans). Dark chestnut soils are formed in the foothill desert-steppe zone at an altitude of 700-800 m above sea level under ephemeroid-fescue-wormwood vegetation (fescue, bluegrass, wormwood, sedge, sage). Mechanical composition - medium and heavy loams. Dark chestnut soils have a humus horizon on average 50 cm thick. A characteristic feature of these soils is their carbonate content. Groundwater is deep and has no effect on the soil-forming process [7,8].

The climate of the region is sharply continental with little precipitation in the spring, including the

dry month. The average air temperature in Talgar is 100C, the average annual precipitation is 594 mm. The warmest month of the year is July (22.10C), the coldest January is -120C.

The research was carried out by the method of laboratory and laboratory-field experience. The planning of experiments, the establishment and conduct of experiments was carried out according to the Dospekhov method [9], the Belikov and Bondarenkov method [10] and the method of the State variety testing of agricultural crops [11].

Phenological observations were carried out according to the form adopted by the state variety testing. From the moment of sowing the seeds to the end of fruiting, the timing of the onset and passage of phenophases is marked - the phase of the appearance of single and mass shoots, the beginning of the formation of the food organ, the first and last collection. The use of natural preparations of biological origin will be carried out according to the generally accepted method of S. Litvinov [12].

Humidity - by weight method, total humus according to I.V. Tyurin by wet combustion of humic carbon and its oxidation with dichromate; specific gravity - by pycnometric method; bulk density using Kachin borax; total porosity - by calculation method; gross forms of nitrogen, phosphorus from one sample in Gizburg and Shcheglov; determination of nitrogen according to Kjeldahl; calorimetric determination of phosphorus, potassium on a flame photometer; nitrate nitrogen according to Grandval-Lyazh, mobile phosphorus according to the method of Machigin B.A. [13]. Phenological observations of vegetable crops according to A. Druzhkin, G. Nikitenko [14,15]. Analyzes of soils and plants were carried out in certified laboratories of the U.U. Uspanov Kazakh Scientific Research Institute of Soil Science and Agrochemistry and KazNU them. I. al-Farabi (Center for Physicochemical Methods of Research and Analysis.

Vegetables were used for the experiment (lettuce, broccoli, beans). The size of the registration plots is 4–6 m2 with 3-fold repetition. The experiments were carried out in vegetable crop rotations on agrotechnical backgrounds recommended for the Almaty region [16]. Fertilizing of vegetable crops will be carried out by spraying, i.e. foliar method, the drug will also be applied by the root method, watering vegetables, at the rate of 2.5 liters of solution per plant.

The regulations for the use of biological products on tomatoes are presented in Table 1.

**Table 1**. Regulations for the use of the drug "Bio EcoGum"

| Culture  | Consumption rate       | Treatment method   | Period and number of treatments  |
|----------|------------------------|--|--|
| Salad    | 25 ml / 4-5 l of water | <ul><li>Soaking seeds for 3 hours.</li><li>Watering the soil before sowing seeds</li></ul> | During the growing season of plants 1-2 times with an interval of 7-14 days. |
| Broccoli | 35 ml / 4-5 l of water | <ul><li>Soaking seeds for 3 hours.</li><li>Watering the soil before sowing seeds</li></ul> | The total number of treatments during the growing season of plants: 2 - 4    |
| Beans    | 70 ml / 5 l of water   | - Soaking seeds for 3 hours Watering the soil before sowing seeds                          | No more than two times   |

Liquid humic biological product "BioGumEco", has the composition: humic substances 20%, macroelements, g / l: N - 5,  $P_2O_5$  - 10,  $K_2O$  - 10, Ca - 7, Mg - 2, trace elements (g / l): Mn - 30, Mo - 30, Zn - 25, Se - 3 [6]. The biological product was introduced into the soil at a dose of 2.5 ml / kg of soil, before sowing, the seeds were treated with the biological product ".

### RESULTS AND DISCUSSION

In the experiments, we used varieties of vegetable crops approved for use in the Almaty region - lettuce, beans, broccoli. In the southeast of Kazakhstan, as well as in Kazakhstan as a whole, the production of salad products

represents an insignificant part of the vegetable assortment. However, in recent years, interest in the culture of lettuce has increased on the part of the population and greenhouse complexes, but the need is far from being fully satisfied. The salad is distinguished by its early maturity, cold resistance and is of great dietary importance. The research was carried out on four types of lettuce: Ritzia, Poalli, Miracle of the four seasons, Large cabbage. The short growing season of this crop allows you to get several harvests even in open field conditions. Research has been carried out on the influence of the biological product "BioEcoGum" on the salad plant, both on the weight of food organs and on the yield (Table 2).

**Table 2**. Influence of the biological product "BioEcoGum" on the mass of food organs, yield and characteristics of lettuce seeds

| Option      | Variety                         | Mass, g |                       |                       |               | Seed          |                         |
|-------------|---------------------------------|---------|-----------------------|-----------------------|---------------|---------------|-------------------------|
|             |                                 | plants  | Incl. head of cabbage | Productivity,<br>c/ha | Seed color    | length,<br>mm | Weight of 1000 seeds, g |
| The control | Ritzia                          | 400     | _                     | 296,9                 | grayish white | 3,8           | 1,14                    |
|             | Poally                          | 240     | _                     | 175,7                 | grayish white | 3,7           | 1,18                    |
|             | The miracle of the four seasons | 463     | 254                   | 326,5                 | black         | 3,3           | 1,32                    |
|             | Large cabbage                   | 502     | 294                   | 352,1                 | grayish white | 3,6           | 1,29                    |
| "BioEcoGum" | Ritzia                          | 420     | _                     | 304,5                 | grayish white | 4,4           | 1,21                    |
|             | Poally                          | 270     | _                     | 195,8                 | grayish white | 4,4           | 1,20                    |
|             | The miracle of the four seasons | 483     | 284                   | 350,1                 | black         | 4,2           | 1,25                    |
|             | Large cabbage                   | 520     | 300                   | 377,0                 | grayish white | 4,7           | 1,24                    |

Our calculation of the total mass of lettuce showed significant differences in the development of the food organs of the tested varieties.

Plants of the Ritzia variety - 420 g (Figure 1), had the largest mass in leafy lettuce, the Poalli variety - 251 g.

Studies have shown that the high yield was noted in the Ritsia variety - 304.5 c/ha, the low was in the Poalli variety - 195.8 c/ha. Seeds of leafy lettuce varieties are grayish-white. The seed shape of these varieties is lanceolate.



Fig. 1. Salad of the Ritzia variety

Measurement of the length of the seed showed that the longest (4.4 mm) were the seeds in the varieties Ritsia and Poalli, short (3.4 mm). The weight of 1000 seeds (1.20–1.25 g) in leaf and head varieties was within the experimental error. Head varieties differ in the weight of food organs and yield. The Large cabbage variety had a large plant mass - 520 g, the Chudo variety of the four

seasons had a smaller mass - 483 g.A high yield was obtained for the head lettuce of the Large cabbage variety - 377.0 centner /ha, the low one was for the Chudo variety of four seasons - 350.1 centner /ha. The variety Chudo of the four seasons had a black color of seeds, and the variety Large cabbage had a grayish-white color. The longest were the seeds of lettuce variety Large. The shape of the seeds of all varieties is lanceolate.In terms of economic indicators, among the headed varieties, the Krupnokochanny variety stood out. The data obtained allow us to draw the following conclusions: - the soil and climatic conditions of the Almaty region are favorable for growing lettuce. To increase productivity and economic efficiency, lettuce should be grown from leafy varieties Ritzia and Poalli, and from cabbage varieties -Large cabbage. The soil and climatic conditions of the southeast of Kazakhstan correspond to the biological characteristics of common beans, which are widely cultivated as a vegetable garden. Beans are distinguished by a high content of protein in seeds of complete amino acid composition. It is of great economic value in human nutrition. In addition, the seeds of leguminous crops contain a lot of carbohydrates (24-50%), vitamins (A, B6, B2, C). In recent years, the demand of the population of many countries, including Kazakhstan, for food products from beans has been rapidly growing: seeds in dry and canned form, freshly frozen, as well as canned beans. In terms of acreage, beans ranks 12th in the world among leguminous crops after soybeans. We carried out studies on the effect of the biological product on the yield of vegetable beans (Table 3).

Table 3. Influence of the biological product "BioEcoGum" yield and characteristics of beans seeds

| Option      | Variety                 | Productivity,<br>c / ha | Pods per plant, pcs | Seeds in a pod, pcs | Weight of 1000 seeds, g |
|-------------|-------------------------|-------------------------|---------------------|---------------------|-------------------------|
| The control | Sachs without fiber 615 | 23,2                    | 9,8                 | 4,5                 | 336                     |
|             | Rand                    | 30,0                    | 11,2                | 4,6                 | 394                     |
|             | Laura                   | 22,2                    | 13,7                | 4,9                 | 219                     |
| "BioEcoGum" | Sachs without fiber 615 | 25,0                    | 10,5                | 5,8                 | 344                     |
|             | Rand                    | 34,1                    | 13,3                | 5,2                 | 406                     |
|             | Laura                   | 25,4                    | 15,9                | 5,5                 | 242                     |

The analysis of yield data showed that when processing with a biological product, the largest bean yield was obtained for the Rant variety - 34.1 c / ha, and the smallest was for the Saksa variety without fiber 615 - 25.0 c / ha.

Counting the number of pods on plants of the studied bean varieties made it possible to establish that the maximum number of pods was formed in the Laura variety (15.9 pcs.), The minimum was in the Saksa variety without fiber 615 - 10.5 pcs.

The Rant variety had the largest leaf surface area - 793 cm<sup>2</sup>; it was smaller in the Laura variety - 701 cm<sup>2</sup>.

Counting the number of pods on plants made it possible to establish that it was the highest in the Saksa variety without fiber 615 (5.8 pcs), and the minimum in the Rant variety (5.2 pcs).

The absolute weight of seeds in plants of the studied bean varieties also differed significantly.

The highest it was in the Rant variety - 406 g, the minimum in the Laura variety (242 g).

It was found that the maximum amount of beans

was formed in the Laura variety (15.9 pieces), the minimum was in the Saksa variety without fiber 615 - 10.5 pieces. (Figure 2).



**Fig. 2.** Beans of the Saksa variety without fiber 615

We have conducted research on the effect of a biological product on the development and productivity of broccoli cabbage plants (Table 4).

**Table 4.** Influence of the biological product "BioEcoGum" on the biometrics and productivity of broccoli cabbage plants

| Option      | Variety                    | Plant height, cm | Leaf blade diameter of a<br>large leaf, cm | The length of the petiole of<br>a large leaf, cm | Head diameter, cm | Head branches of the first<br>order, pcs | Plant leaf area, cm² | Productivity, c / ha |
|-------------|----------------------------|------------------|--|--|-------------------|--|----------------------|----------------------|
| The control | Caesar                     | 43,2             | 19,7                                       | 8,5  | 11,6              | 12,2                                     | 5479                 | 240,5                |
|             | Gnome                      | 44,5             | 20,4                                       | 9,7  | 11,9              | 12,8                                     | 6576                 | 278,3                |
|             | Christmas<br>from Calabria | 39,9             | 19,5                                       | 12,3   | 12,5              | 10,9                                     | 4674                 | 248,7                |
|             | Tone                       | 46,8             | 20,8                                       | 12,5   | 10,2              | 8,1                                      | 6496                 | 252,9                |
| "BioEcoGum" | Caesar                     | 45,6             | 22,8                                       | 10,4   | 13,9              | 13,1                                     | 5537                 | 270,8                |
|             | Gnome                      | 46,9             | 21,2                                       | 11,3   | 12,4              | 12,0                                     | 6676                 | 298,3                |
|             | Christmas<br>from Calabria | 40,9             | 20,1                                       | 14,1   | 14,0              | 11,1                                     | 4772                 | 268,4                |
|             | Tone                       | 48,9             | 21,6                                       | 13,3   | 11,2              | 9,8                                      | 6596                 | 272,7                |

Studies of yield data showed that treatment with a biological product contributed to the formation of the highest yield in the Gnome variety - 298.3 c / ha (Figure 3), while in the control it was 278.3 c / ha. The smallest yield was noted for the variety Christmas from Calabria –268.4 c / ha, compared to the control 248.7 c / ha.



Fig.3. Cabbage variety Gnome

The influence of the biological product also influenced the variety study of broccoli cabbage. It was noted that the smallest diameter of the flower head was in the Tonus cultivar - 11.2 cm, the largest in the cultivar Christmas from Calabria - 14.0 cm.

The largest leaf area was in the broccoli cabbage variety Gnome - 6676 cm<sup>2</sup>, the smallest - in the variety Christmas from Calabria - 4772 cm<sup>2</sup>.

The conducted studies allow us to recommend the production of methods of using the biological product "BioEcoGum" when growing vegetables, providing maximum economic and economic efficiency, taking into account environmental requirements.

### **CONCLUSION**

As a result of a comparative study of varieties of vegetable crops carried out in the UPH "Agrouniversity", Almaty region, the following conclusions can be drawn:

The use of the biological product "BioEcoGum" has a significant effect on the onset of phenological phases and the duration of interphase periods in vegetable crops - lettuce, broccoli, beans. A high yield was obtained from the head lettuce variety Large cabbage - 377.0 c / ha, low was the variety Miracle of the four seasons - 350.1 c / ha.

The variety had a large mass of the plant Large cabbage - 520 g, the smaller mass was in the Miracle variety of the four seasons - 483 g.

It has been established that in order to increase the productivity and economic efficiency of the salad, it is necessary to grow the Ritsia and Poalli varieties, and from the cabbage - Large cabbage.

The influence of the biological product on the bean yield was the highest for the Rant variety  $34.1\ c$  / ha, the smallest was for the Saksa variety without fiber 615 -  $25.0\ c$  / ha.

It was found that the maximum amount of beans was formed in the Laura variety (15.9 pieces), the minimum was in the Saksa variety without fiber 615 - 10.5 pieces.

The influence of the biological product on the vegetable culture of broccoli showed that the smallest diameter of the flower head was in the Tonus variety - 11.2 cm, the largest in the variety Christmas from Calabria - 14.0 cm.

The largest leaf area was in the broccoli cabbage variety Gnome - 6676 cm2, the smallest - in the variety Christmas from Calabria - 4772 cm2.

The greatest weight of the head of cabbage was in the plants of the Gnome variety (628 g), the highest yield was obtained here (298.3 centners / ha); The smallest yield was given by the variety Rozhdestvensky from Calabria - 268.4 kg / ha.

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# The checklist of semi-woody plants of Georgia (South Caucasus) and their regional assessment according to IUCN categories and criteria

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### ABSTRACT

A checklist of semi-woody plants of Georgia is presented for the first time. The cheklist includes 72 species, 13 subspecies and 1 variation of vascular plants, which belong to 32 genera and 13 families. Each taxon (species, subspecies and variation) was assessed according to IUCN criteria and assigned an appropriate regional category. Regionally (Republic of Georgia) 1 species is critically endangered (CR), 4 taxa - endangered (EN), 4 - vulnerable (VU), and 12 are near threatened (NT). Due to lack of data, 12 taxa could not be assessed and fell under the category of data definicient. 56 taxa are not endangered at present and have been assigned the category least concern. References to taxsonomy, key synonyms, assessment argumentation and references used in species assessment are given for each taxon.

**Keywords:** Semi-woody plants, Taxon, Cheklist, IUCN categories and criteria, References to taxsonomy, Assessment argumentation.

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### Introduction

Semi-woody plants (semishrubs, undershrubs and dwarf semishrubs) are common in almost all ecosystems and are one of the most important components of vegetation. Their role in vegetation cover of extreme and close to extreme ecosystems is particularly high. Desert vegetation is one of the most prominent in this respect, where semi-shrubs and undershrubs are the dominant forming species for vegetation. They are also widespread in other types of vegetation of arid and semi-arid ecosystems (phryganoid vegetation, tragacanthic shrubberies, steppes, xerophytic forests, vegetation of various rocky and scree ecotopes, etc.). They are also common in high-mountain vegetation. Their participation in forest ecosystems is relatively rare.

As mentioned, semi-woody plants are most

common in extreme and close to extreme environments. Moreover, they are under strong anthropogenic pressure (mostly grazing). Therefore, their recording, condition and assessment according to international standarts is an urgent issue. Despite the above, the composition of semi-woody plants common in Georgia is still unknown and no checklist exists for them either.

### **Objectives and Methods**

The aim of our research was to compile a checklist of semi-woody plants of Georgia and to make their regional assessments according to IUCN categories and criteria, which will promote the protection and conservation for this group of plants.

In addition to our own research, the second edi-

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tion of "Flora of Georgia" [1-3] and the "Nomeno clatural checklist of flora of Georgia" [4] laid the foundation for the creation of the checklist. Names and authors of taxa are checked with the international databases: The Plant List (2021), Euro + Med (2006+), IPNI (2021), GBIF.org (2021), Tropicos. org (2021) [5-9]. As a result, some of the "narrow" species included in the "Flora of Georgia" [1-3] and the "Nomenclatural checklist of flora of Georgia" [4] could not be found in this checklist. These species are given as synonyms.

Taxa assessment and categorization were carried out in accordance with IUCN Red List [10] criteria. The assessment of taxa is based on our own research, literary data and materials preserved in various herbariums of Georgia (TBI, TGI, BATU). Some taxa are assessed based on the data from the "Red list of the endemic plants of the Caucasus" [11].

### **Results and Analysis**

Semi-woody plants of Georgia are presented by 72 species, 13 subspecies and 1 variation. They belong to 32 genera and 13 families of vascular plants.

According to the IUCN Red List assessments, regionally (Georgia) 1 taxon is critically endangered (CR), 5 are endangered (EN), 4 are vulnerable (VU), and 12 are close to vulnerable. (NT). Taxa are distributed according to IUCN categories as follows:

Critically Endangered (CR) - Anabasis aphylla L.;

**Endangered (EN)** - Genista abchasica Sachokia, Helianthemum georgicum Juz. & Pozdeeva, Scutellaria orientalis subsp. karatschaica, Thymus karjaginii Grossh., Ziziphora woronowii Maleev;

**Vulnerable (VU)** - Camphorosma monspeliaca L., Genista mingrelica Albov, Genista suanica Schischk. ex Grossh., Thymus ladjanuricus Kem.-Nath.;

Near Threatend (NT) - Genista flagellaris Somm. et Levier, Genista humifusa L., Genista
kolakowskyi Sachokia, Onobrychis cornuta (L.)
Desv., Reaumuria kuznetzovii Sosn. et Manden.,
Salvia garedjii Troitzky, Satureja bzybica Woronow, Scutellaria leptostegia Juz., Scutellaria ossethica Charadze, Scutellaria raddeana Juz., Teucrium chamaedrys subsp. trapezunticum Rech. f.,
Thymus coriifolius Ronniger.

Due to data deficiency, 12 taxa could not be evaluated and were assigned the category Data

Definicient (DD). They are: Atriplex cana C.A. Mey., Noaea mucronata subsp. leptoclada (Woronow) Assadi, Helianthemum buschii (Palib.) Juz. & Pozdeeva, Helianthemum orientale (Grosser) Juz. et Pozdeeva, Andrachne telephioides L., Genista monspessulana (L.) L. A. S. Johnson, Genista transcaucasica Schischk., Scutellaria oreophila Grossh., Scutellaria pontica K. Koch, Thymus helendzhicus Klokov & Des.-Shost., Thymus karamarianicus Klokov et Des.-Shost., Linum tauricum Willd..

Although most of the semishrubs, undershrubs and dwarf semishrubs are under anthropogenic pressure and a significant proportion of them grow in close to extreme environments, most of them (56 taxa) are not endangered and have been assessed as Least Concern (LC).

Below is a checklist of semi-woody plants of Georgia. Taxa are sorted by families. For each taxon (species, subspecies and variation) the reference to taxonomy, the IUCN Red List category and criteria, the assessment argumentation and the references used in species assessment are given.

### AMARANTHACEAE Anabasis aphylla L.

References to taxsonomy: [4-8];

<u>IUCN red list category and criteria:</u> CR B1ab (iii) + B2ab (iii);

Assessment argumentation: extent of occurrence – 5 km², area of occupancy – 4 km², number of area fragments - 1, decline in habitat quality - habitat degradation;

<u>References used in species assessment:</u> [12, 13, 14]; the herbarium specimens preserved in the National Herbarium of Georgia (TBI); The information provided verbally by N. Lachashvili.

Atriplex cana C.A. Mey. (Atriplex cana Ledeb.) References to taxsonomy: [4-8];

*IUCN red list category and criteria:* DD;

<u>Assessment argumentation:</u> data deficient (unknown extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, continuing decline in quality of habitat etc.);

<u>References used in species assessment:</u> [5, 6, 18]; the herbarium specimens preserved in the National Herbarium of Georgia (TBI).

Bassia prostrata (L.) Beck [Kochia prostrata (L.) Schrad.]

References to taxsonomy: [5-7];

### *IUCN red list category and criteria:* LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

<u>References used in species assessment:</u> [12, 13, 15].

### Camphorosma monspeliaca L.

*References to taxsonomy:* [4-8];

<u>IUCN red list category and criteria:</u> VU B1 ab (iii);

<u>Assessment argumentation:</u> extent of occurrence - 10100 km<sup>2</sup>, area of occupancy - <2000 km<sup>2</sup>, area - fragmented, number of area fragments - 5, decline in habitat quality - habitat degradation;

<u>References used in species assessment:</u> [12, 13, 15, 16]; The information provided verbally by N. Lachashvili.

Halothamnus glaucus (M. Bieb.) Botsch. [Salsola glauca M. Bieb.; Aellenia glauca (M. Bieb.) Aellen]

*References to taxsonomy:* [5-8, 12, 13];

IUCN red list category and criteria: LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

<u>References used in species assessment:</u> [12, 13, 15].

### Krascheninnikovia ceratoides (L.) Gueldenst.

(Ceratoides papposa Botsch. & Ikonn)

*References to taxsonomy:* [5-8];

IUCN red list category and criteria: LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

<u>References used in species assessment:</u> [15]; the herbarium specimens preserved in the National Herbarium of Georgia (TBI).

Noaea mucronata (Forssk.) Aschers. & Schweinf.

Noaea mucronata subsp. leptoclada (Woronow) Assadi [Noaea leptoclada (Woronow) Iljin; Noaea mucronata subsp. tournefortii (Spach) Aellen]

References to taxsonomy: [8];

*IUCN red list category and criteria*: DD;

Assessment argumentation: data deficient (un-

known extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, continuing decline in quality of habitat etc.);

<u>References used in species assessment:</u> [15, 16]; the herbarium specimens preserved in the National Herbarium of Georgia (TBI).

## Noaea mucronata (Forssk.) Asch. & Schweinf. subsp. mucronata

References to taxsonomy: [4-8];

*IUCN red list category and criteria:* LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

<u>References used in species assessment:</u> [12, 13, 15, 16].

Salsola dendroides Pall. [Caroxylon dendroides (Pall.) Tzvelev]

*References to taxsonomy:* [4-8];

*IUCN red list category and criteria:* LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

<u>References used in species assessment:</u> [12, 13, 15].

Salsola ericoides M. Bieb. [Caroxylon ericoides (M.Bieb.) Akhani & Roalson]

References to taxsonomy: [4-8];

*IUCN red list category and criteria:* LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

<u>References used in species assessment:</u> [12, 13, 15].

Salsola nodulosa (Moq.) Iljin (Caroxylon nodulosum Moq.)

*References to taxsonomy:* [4, 5];

IUCN red list category and criteria: LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

*References used in species assessment:* [12, 13, 15].

### **ASTERACEAE**

Artemisia alpina Pall. ex Willd. (Artemisia caucasica Willd.)

*References to taxsonomy:* [5-8];

*IUCN red list category and criteria:* LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

References used in species assessment: [17].

### Artemisia fragrans Willd.

References to taxsonomy: [4-8];

*IUCN red list category and criteria:* LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

<u>References used in species assessment:</u> [12, 13, 17].

### Artemisia incana (L.) Druce

References to taxsonomy: [4-8];

*IUCN red list category and criteria:* LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

<u>References used in species assessment:</u> [16, 17, 18].

Artemisia marschalliana Spreng. (Artemisia campestris subsp. inodora Nyman)

*References to taxsonomy:* [4, 7, 8];

*IUCN red list category and criteria:* LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

*References used in species assessment:* [16, 17, 19].

### Artemisia splendens Willd.

References to taxsonomy: [4-8];

*IUCN red list category and criteria:* LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

<u>References used in species assessment:</u> [17, 19]. **Artemisia szowitziana (Besser) Grossh.**  References to taxsonomy: [4-8, 13];

*IUCN red list category and criteria:* LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

<u>References used in species assessment:</u> [13]; The information provided verbally by N. Lachashvili.

### **CAPPARACEAE**

*Capparis spinosa* L. [*Capparis herbacea* Willd; *Capparis spinosa* var. *herbacea* (Willd.) Fici]

*References to taxsonomy:* [4-8];

*IUCN red list category and criteria:* LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

<u>References used in species assessment:</u> [12, 13, 20].

### **CARYOPHYLLACEAE**

### Cerastium argenteum M. Bieb.

*References to taxsonomy:* [4-8];

*IUCN red list category and criteria:* LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

References used in species assessment: [16, 21].

### Dianthus orientalis Adms

*References to taxsonomy:* [4-8];

*IUCN red list category and criteria:* LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

<u>References used in species assessment:</u> [12, 16, 22].

### **CISTACEAE**

Helianthemum buschii (Palib.) Juz. & Pozdeeva

*References to taxsonomy:* [4-8];

*IUCN red list category and criteria:* DD;

<u>Assessment argumentation:</u> data deficient (unknown extent of occurrence, area of occupancy, the number of mature individuals and the tendency of

their decline in numbers, continuing decline in quality of habitat etc.);

References used in species assessment: [23, 24].

### Helianthemum georgicum Juz. et Pozdeeva

*References to taxsonomy:* [4-8];

IUCN red list category and criteria: EN Blab (iii, v) + 2ab (iii, v);

Assessment argumentation: extent of occurrence - <5000 km<sup>2</sup>, area of occupancy - <500 km<sup>2</sup>,

*References used in species assessment:* [16, 24]; the herbarium specimens preserved in the National Herbarium of Georgia (TBI); The IUCN category was given according to Solomon & al., 2013 [11].

Helianthemum nummularium (L.) Mill.

Helianthemum nummularium subsp. grandiflorum (Scop.) Schinz & Thell. [Helianthemum grandiflorum (Scop.) DC.]

*References to taxsonomy:* [5, 6, 8];

IUCN red list category and criteria: LC;

Assessment argumentation: at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

References used in species assessment: [16, 19, 23-25].

### Helianthemum nummularium (L.) Mill. subsp. nummularium

References to taxsonomy: [4-8];

*IUCN red list category and criteria:* LC;

Assessment argumentation: at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

References used in species assessment: [19, 24, 25];

### Helianthemum orientale (Grosser) Juz. et Pozdeeva

*References to taxsonomy:* [4-8];

*IUCN red list category and criteria:* DD;

Assessment argumentation: data deficient (unknown extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, continuing decline in quality of habitat etc.);

References used in species assessment: [12, 16, 24]; the herbarium specimens preserved in the National Herbarium of Georgia (TBI).

### **EUPHORBIACEAE**

Andrachne telephioides L. (Andrachne rotundi-

folia C.A. Mey. ex Eichw.)

References to taxsonomy: [4-8];

*IUCN red list category and criteria:* DD;

Assessment argumentation: data deficient (unknown extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, continuing decline in quality of habitat etc.);

References used in species assessment: [12, 26]; the herbarium specimens preserved in the National Herbarium of Georgia (TBI).

### **FABACEAE**

### Astragalus xiphidium Bunge

References to taxsonomy: [4-8];

IUCN red list category and criteria: LC;

Assessment argumentation: at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

References used in species assessment: [12, 27]; Herbarium specimens preserved in the National Herbarium of Georgia (TBI).

### Genista abchasica Sachokia

References to taxsonomy: [4-8];

IUCN red list category and criteria: EN Blab (iii) + 2ab (iii);

Assessment argumentation: extent of occurrence - <5000 km<sup>2</sup>, area of occupancy - <500 km<sup>2</sup>;

References used in species assessment: [28, 29]; The IUCN category was given according to Solomon & al., 2013 [11].

### Genista flagellaris Somm. et Levier

*References to taxsonomy:* [4-8];

*IUCN red list category and criteria*: NT;

Assessment argumentation: the species assessment data are approximate to VU category parameters and/or likely to be close to it in the future;

References used in species assessment: [28].

Genista humifusa L. (Genista sachokiana A. I. Kuth.)

*References to taxsonomy:* [4-8];

*IUCN red list category and criteria*: NT;

Assessment argumentation: the species assessment data are approximate to VU category parameters and/or likely to be close to it in the future;

References used in species assessment: [25, 28, 29].

### Genista kolakowskyi Sachokia

References to taxsonomy: [4-8];

*IUCN red list category and criteria:* NT;

<u>Assessment argumentation:</u> the species assessment data are approximate to VU category parameters and/or likely to be close to it in the future;

References used in species assessment: [28, 29].

### Genista mingrelica Albov

*References to taxsonomy:* [4-8];

IUCN red list category and criteria: VU D2;

<u>Assessment argumentation:</u> area of occupancy - <20 km<sup>2</sup>;

<u>References used in species assessment:</u> [28]; The IUCN category was given according to Solomon & al., 2013 [11].

### Genista monspessulana (L.) L. A. S. Johnson [Teline monspessulana (L.) K. Koch]

References to taxsonomy: [5-8];

*IUCN red list category and criteria:* DD;

<u>Assessment argumentation:</u> data deficient (unknown extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, continuing decline in quality of habitat etc.);

References used in species assessment: [28].

Genista tinctoria L. (Genista patula M. Bieb.)

*References to taxsonomy:* [5-8];

*IUCN red list category and criteria*: LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

References used in species assessment: [28].

Genista suanica Schischk. ex Grossh. (Genista adzharica Popov)

*References to taxsonomy:* [4-8];

<u>IUCN red list category and criteria:</u> VU B2ab (iv);

<u>Assessment argumentation:</u> extent of occurrence - <20000 km<sup>2</sup>, area of occupancy - <2000 km<sup>2</sup>;

<u>References used in species assessment:</u> [25, 28, 29, 30]; The IUCN category was given according to Solomon & al., 2013 [11].

### Genista transcaucasica Schischk.

*References to taxsonomy:* [4-6, 8, 11];

*IUCN red list category and criteria:* DD;

<u>Assessment argumentation:</u> data deficient (unknown extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, continuing decline in quality of habitat etc.);

References used in species assessment: [28].

Onobrychis cornuta (L.) Desv.

References to taxsonomy: [4-8];

*IUCN red list category and criteria:* NT;

<u>Assessment argumentation:</u> the species assessment data are approximate to VU category parameters and/or likely to be close to it in the future;

<u>References used in species assessment:</u> [16, 31]; the herbarium specimens preserved in the National Herbarium of Georgia (TBI).

Prosopis farcta (Banks & Sol.) J. F. Macbr. (Lagonychium farctum (Banks & Sol.) Bobrov)

*References to taxsonomy:* [5-8];

IUCN red list category and criteria: LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

<u>References used in species assessment:</u> [12, 13, 32]; the herbarium specimens preserved in the National Herbarium of Georgia (TBI).

### **HYPERICACEAE**

### Hypericum androsaemum L.

References to taxsonomy: [4-8];

IUCN red list category and criteria: LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

<u>References used in species assessment:</u> [25, 33, 34]; the herbarium specimens preserved in the National Herbarium of Georgia (TBI).

### **LAMIACEAE**

### Salvia garedjii Troitzky

References to taxsonomy: [4-8];

*IUCN red list category and criteria:* NT;

<u>Assessment argumentation:</u> the species assessment data are approximate to VU category parameters and/or likely to be close to it in the future;

*References used in species assessment:* [12, 35, 36].

### Satureja bzybica Woronow

*References to taxsonomy:* [4-8];

IUCN red list category and criteria: NT;

<u>Assessment argumentation:</u> the species assessment data are approximate to VU category parameters and/or likely to be close to it in the future;

References used in species assessment: [33, 37].

### Scutellaria helenae Albov

*References to taxsonomy:* [4-8];

IUCN red list category and criteria: LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

<u>References used in species assessment:</u> [25, 33, 38].

### Scutellaria leptostegia Juz.

References to taxsonomy: [4-8];

*IUCN red list category and criteria:* NT;

<u>Assessment argumentation:</u> the species assessment data are approximate to VU category parameters and/or likely to be close to it in the future;

References used in species assessment: [19, 39].

### Scutellaria oreophila Grossh.

References to taxsonomy: [4-8];

*IUCN red list category and criteria:* DD;

<u>Assessment argumentation:</u> data deficient (unknown extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, continuing decline in quality of habitat etc.);

References used in species assessment: [19, 39].

Scutellaria orientalis L.

*Scutellaria orientalis* subsp. *karatschaica* (Kharadze) Menitsky [Scutellaria karatschaica Kharadze]

*References to taxsonomy:* [5-8];

<u>IUCN red list category and criteria:</u> EN B1ab (iii) + 2ab (iii);

<u>Assessment argumentation:</u> extent of occurrence - <5000 km<sup>2</sup>, area of occupancy - <500 km<sup>2</sup>;

<u>References used in species assessment:</u> [39]; The IUCN category was given according to Solomon & al., 2013 [11].

### Scutellaria orientalis L. subsp. orientalis

References to taxsonomy: [4-8];

*IUCN red list category and criteria:* LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

References used in species assessment: [12, 39].

### Scutellaria ossethica Charadze

*References to taxsonomy:* [4-8];

*IUCN red list category and criteria*: NT;

<u>Assessment argumentation:</u> the species assessment data are approximate to VU category parameters and/or likely to be close to it in the future;

References used in species assessment: [25, 39].

Scutellaria pontica K. Koch

*References to taxsonomy:* [4-8];

*IUCN red list category and criteria:* DD;

<u>Assessment argumentation:</u> data deficient (unknown extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, continuing decline in quality of habitat etc.);

References used in species assessment: [39].

### Scutellaria raddeana Juz.

*References to taxsonomy:* [4-8];

*IUCN red list category and criteria:* NT;

<u>Assessment argumentation:</u> the species assessment data are approximate to VU category parameters and/or likely to be close to it in the future;

References used in species assessment: [19, 39].

Scutellaria sosnowskyi Takht. [Scutellaria orientali subsp. sosnowskyi (Takht.) Fed.]

References to taxsonomy: [4, 5, 7, 8];

*IUCN red list category and criteria:* LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

References used in species assessment: [16, 39].

### Stachys fruticulosa M.Bieb.

*References to taxsonomy:* [4-8];

*IUCN red list category and criteria:* LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

<u>References used in species assessment:</u> [12, 13, 40].

### Teucrium chamaedrys L.

Teucrium chamaedrys subsp. nuchense (K.

Koch) Rech. f. (Teucrium nuchense K. Koch)

*References to taxsonomy:* [5-7];

*IUCN red list category and criteria:* LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

<u>References used in species assessment:</u> [12, 13, 16, 19, 33, 41].

Teucrium chamaedrys subsp. trapezunticum Rech. f. [Teucrium trapezunticum (Rech. f.) Juz.]

*References to taxsonomy:* [5-7];

IUCN red list category and criteria: NT;

Assessment argumentation: the species assess-

ment data are approximate to VU category parameters and/or likely to be close to it in the future;

References used in species assessment: [41];

Teucrium chamaedrys var. multinodum Bordz. [Teucrium multinodum (Bordz.) Juz.]

References to taxsonomy: [5-8];

*IUCN red list category and criteria:* LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

<u>References used in species assessment:</u> [41].

### Teucrium polium L.

*References to taxsonomy:* [4-8];

*IUCN red list category and criteria:* LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

<u>References used in species assessment:</u> [12, 13, 16, 41].

### Thymus collinus M. Bieb.

References to taxsonomy: [4-8];

*IUCN red list category and criteria:* LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

<u>References used in species assessment:</u> [16, 19, 25, 42].

Thymus coriifolius Ronniger (Thymus sosnowskyi Grossh.)

*References to taxsonomy:* [4-8];

*IUCN red list category and criteria:* NT;

<u>Assessment argumentation:</u> the species assessment data are approximate to VU category parameters and/or likely to be close to it in the future;

References used in species assessment: [42].

Thymus helendzhicus Klokov & Des.-Shost

*References to taxsonomy:* [5-8, 11];

*IUCN red list category and criteria:* DD;

Assessment argumentation: data required for species assessment are not available (Literary data on the area and the number of species in Georgia are not available; no herbarium specimens of this species collected in Georgia are stored in the herbariums of Georgia (TBI, TGI, BATU);

<u>References used in species assessment:</u> the herbarium material of this species is not available in

the herbariums of Georgia (TBI, TGI, BATU).

*Thymus karamarianicus* Klokov et Des.-Shost.

*References to taxsonomy:* [4-8, 11];

*IUCN red list category and criteria:* DD;

Assessment argumentation: data required for species assessment are not available (Literary data on the area and the number of species in Georgia are not available; no herbarium specimens of this species collected in Georgia are stored in the herbariums of Georgia (TBI, TGI, BATU);

<u>References used in species assessment:</u> the herbarium material of the species is not available in the herbariums of Georgia (TBI, TGI, BATU).

### Thymus karjaginii Grossh.

*References to taxsonomy:* [4-8];

<u>IUCN red list category and criteria:</u> EN B1ab (iii, v) + 2ab (iii, v);

<u>Assessment argumentation:</u> extent of occurrence - <5000 km<sup>2</sup>, area of occupancy - <500 km<sup>2</sup>, number of area fragments - 1, decline in habitat quality - habitat degradation;

References used in species assessment: [12, 42].

*Thymus ladjanuricus* Kem.-Nath.

*References to taxsonomy:* [4-8];

IUCN red list category and criteria: VU D2;

<u>Assessment argumentation:</u> area of occupancy - <20 km<sup>2</sup>;

<u>References used in species assessment:</u> [25, 42]; The IUCN category was given according to Solomon & al., 2013 [11].

*Thymus nummularius* M. Bieb. (*Thymus pseudopulegioides* Klokov & Des.-Shost.)

*References to taxsonomy:* [4-8, 43];

*IUCN red list category and criteria:* LC;

Assessment argumentation: at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

*References used in species assessment:* [19, 25, 42].

### Thymus praecox Opyz

Thymus praecox subsp. caucasicus (Willd. ex Ronniger) Jalas (Thymus caucasicus Willd. ex Ronniger)

*References to taxsonomy:* [5-8, 43];

*IUCN red list category and criteria:* LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

*References used in species assessment:* [25, 33, 42].

*Thymus praecox* subsp. *grossheimii* (Ronniger) Jalas (*Thymus grossheimii* Ronniger)

References to taxsonomy: [5-8, 43].

*IUCN red list category and criteria:* LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

*References used in species assessment:* [16, 42].

*Thymus pulegioides* subsp. *pannonicus* (All.) Kerguélen (*Thymus marschallianus* Willd.)

References to taxsonomy: [5-8];

*IUCN red list category and criteria*: LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

<u>References used in species assessment:</u> [16, 42]. <u>Thymus sipyleus</u> Boiss. (Thymus rariflorus K. Koch)

References to taxsonomy: [5-8];

*IUCN red list category and criteria:* LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

<u>References used in species assessment:</u> [12, 16, 42].

### Thymus tiflisiensis Klokov et Des.-Shost.

*References to taxsonomy:* [4-8];

*IUCN red list category and criteria*: LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

*References used in species assessment:* [12, 42].

### Thymus transcaucasicus Ronniger

References to taxsonomy: [4-8];

*IUCN red list category and criteria:* LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

<u>References used in species assessment:</u> [16, 19, 42].

Ziziphora clinopodioides Lam. subsp. clinopodioides (Ziziphora borzhomica Juz. ex Grossh.; Ziziphora dzhavakhishvilii Juz.; Ziziphora serpyllacea M. Bieb.)

*References to taxsonomy:* [5-8];

*IUCN red list category and criteria:* LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

*References used in species assessment:* [12, 16, 44].

### Ziziphora puschkinii Adams

*References to taxsonomy:* [4-8];

IUCN red list category and criteria: LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

References used in species assessment: [19, 44].

### Ziziphora woronowii Maleev

*References to taxsonomy:* [4-8];

<u>IUCN red list category and criteria:</u> EN B1ab (iii, iv) + 2ab (iii, iv);

<u>Assessment argumentation:</u> extent of occurrence - <5000 km<sup>2</sup>, area of occupancy - <500 km<sup>2</sup>;

<u>References used in species assessment:</u> [33, 44]. The IUCN category was given according to Solomon & al., 2013 [11].

### LINACEAE

Linum mucronatum Bertol.

Linum macronatum subsp. armenum (Bordz.)

**P. H. Davis** (*Linum orientale* subsp. *armenum* Bordz.)

*References to taxsonomy:* [6, 8, 43];

*IUCN red list category and criteria:* LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

<u>References used in species assessment:</u> [12, 13, 45]; the herbarium specimens preserved in the National Herbarium of Georgia (TBI).

### Linum tauricum Willd.

*References to taxsonomy:* [4-8];

*IUCN red list category and criteria:* DD;

<u>Assessment argumentation:</u> data deficient (unknown extent of occurrence, area of occupancy, the

number of mature individuals and the tendency of their decline in numbers, continuing decline in quality of habitat etc.);

References used in species assessment: [29, 45].

#### **ONAGRACEAE**

*Epilobium colchicum* **Albov** [*Chamerion colchicum* (Albov) Holub]

*References to taxsonomy:* [5-8];

*IUCN red list category and criteria*: LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

*References used in species assessment:* [19, 25, 46].

*Epilobium dodonaei* Vill. [*Chamerion dodonaei* (Vill.) Holub]

References to taxsonomy: [5-8];

IUCN red list category and criteria: LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

*References used in species assessment:* [19, 25, 29, 46].

# **SOLANACEAE**

# Solanum persicum Willd. ex Roem. & Schult.

*References to taxsonomy:* [4-6, 8, 43];

*IUCN red list category and criteria:* LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

<u>References used in species assessment:</u> [16, 19, 29, 47].

#### **TAMARICACEAE**

# Reaumuria alternifolia (Labill.) Britten

<u>References to taxsonomy:</u> Davlianidze & al., 2018; The Plant List (2021); Euro+Med (2006+); IPNI (2021); GBIF.org (2021) [4-8];

IUCN red list category and criteria: LC;

<u>Assessment argumentation:</u> at the current stage, neither the existing nor the expected threats to the species (extent of occurrence, area of occupancy, the number of mature individuals and the tendency of their decline in numbers, etc.) are noticeable;

<u>References used in species assessment:</u> [12, 13, 48].

#### Reaumuria kuznetzovii Sosn. & Manden.

References to taxsonomy: [4, 7, 8, 11]; IUCN red list category and criteria: NT; Assessment argumentation: Khintibidze, 1983; References used in species assessment: [48]; The

IUCN category was given according to Solomon & al., 2013 [11].

#### Conclusion

Semi-woody plants of Georgia are presented by 72 species, 13 subspecies and 1 variation. They belong to 32 genera and 13 families of vascular plants.

Regionally (Georgia) 1 taxon (Anabasis aphylla L.) is critically endangered, 5 – endangered (Genista abchasica Sachokia, Helianthemum georgicum Juz. & Pozdeeva, Scutellaria orientalis subsp. karatschaica, Thymus karjaginii Grossh., Ziziphora woronowii Maleev), 4 - vulnerable (Camphorosma monspeliaca L., Genista mingrelica Albov, Genista suanica Schischk. ex Grossh., Thymus ladjanuricus Kem.-Nath.). 12 taxa are close to vulnerable. As of today, 56 taxa are not in danger (Least Concern). Due to data deficiency, 12 taxa were assigned the category Data Definicient (DD).

# Acknowledgements

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# Tulipa biflora Pall. (Liliaceae) - a new species for the flora of Georgia (South Caucasus)

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#### ABSTRACT

Tulipa biflora Pall. is reported from Georgia for the first time. The species was found on Iaghluja Ridge in the vicinity of Rustavi (Kvemo Kartli region). The area belongs to the semi-arid climate zone. *T. biflora* grow on a ridge of marine and continental molasses on grey-cinnamonic soils with clay and small stones in phryganoid vegetation habitat, particularly in the plant community of thorny goat's-wheat (Atraphaxietum spinosae). The total area of occupancy of the newly discovered populations of *T. biflora* is approximately 350 m²; they are under heavy anthropogenic pressure. The populations consist of approximately 350-400 individuals. The article contains coordinates and a map of the new location of *T. biflora*, as well as a description of the habitat with reference to all the key features. The herbarium specimens of *T. biflora* are stored at the National Herbarium of Georgia (TBI). The photo documentation is included in this article.

Keywords: Tulipa biflora, Distribution area, Iaghluja Ridge, Rustavi environs, Habitat, Populations.

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# Introduction

Tulipa biflora Pall. (Fig. 1) belongs to the family Liliaceae. It is an ephemeroid bulbous plant (Fig. 2), mostly developing two gray-green leaves. It produces 1-3 or more flowers. Perianth is white, tinged on the outside with greenish and pale brownish-crimson, and with yellow at the base inside (Fig. 3a, 3b). According to the Raunkiaer classification of plant life forms [1], it is a geophyte.



Fig. 1. Tulipa biflora Pall. growing on Iaghluja Ridge (photo by N. Lachashvili).



**Fig. 2.** Bulb of *Tulipa biflora* Pall. (Photo by N. Lachashvili)



**Fig. 3a.** Flower of *Tulipa biflora* Pall. - front-view (Photo by N. Lachashvili)



**Fig. 3b.** Flower of *Tulipa biflora* Pall. - view from above (Photo by N. Lachashvili)

The distribution area of *Tulipa biflora* includes South-East Europe, South-West Asia, Middle Asia, the western part of Central Asia (North-West China - North and West Xinjiang) and Caucasus. It is also found in North-East Africa (Egypt) [2-5]. Considering its global distribution area and center of gravity of spread, in our opinion, it belongs to the Eurasian Steppe-Southwest Asian chorotype.

*T. biflora* grows on dry slopes and plains, scree and stony ecotopes, clay and saline soils, etc. It is distributed in plant communities of various arid and semi-arid ecosystems (desert and semi-desert, steppe, arid open woodland, hemixerophilous and xerophilous shrubberies, etc.).

# **Objectives and Methods**

The aim of our study was to determine the area and habitat of the identified populations of *Tulipa biflora*, to assess population abundance and condition of specimens and habitat of *T. biflora*, to identify current and potential threats and make a preliminary regional (Georgia) assessment according to IUCN categories and criteria [6].

The target species was recorded and monitored during both flowering (April) and fruiting (May) phases. Therefore, the phytosociological surveys of the vegetation were conducted twice. This allowed to identify vegetation structure of the *Tulipa biflora* habitat and the rhythm of the plant community development. Phytosociological surveys were carried out using traditional methods [7-13].

Taxa are cited according to The plant list (2021) [14]. *T. biflora* was assessed using IUCN Red List categories and criteria.

Climatic data follows Kordzakhia 2018 [15] and Kartvelishvili [16], soils - T. Urushadze [17, 18] and that of geology - Gamkrelidze [19].

Coordinates and altitude above sea level were recorded using Global Positioning System (GPS).

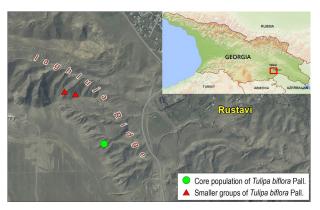
# **Results and Analysis**

*Tulipa biflora* was found in the eastern part of Iaghluja Ridge (Fig. 4) in the vicinity of Rustavi, at the altitude of 500-550 m asl. In adv dition to the core population at N41.541083,

E44.972217 smaller groups comprising several individuals are scattered within approximately 500 m long zone along the eastern slopes of the ridge (Fig. 5).



**Fig. 4.** Eastern part of Iaghluja Ridge – distribution area of *Tulipa biflora* Pall. (Photo by N. Lachashvili)



**Fig. 5.** Map of distribution of *Tulipa biflora* Pall. on Iaghluja Ridge

**Terrain.** The area is characterized by the alternation of slopes and ravines of different exposures and inclinations. The surface of the slopes is slightly hillocky and traversed by trails.

Climate. The study site is situated in the semi-arid climate zone, particularly in moder-

ately warm steppe climate with dry summers and two minimum of precipitation per year. The average annual temperature is up to 13°C. The average annual precipitation is in the range of 400-440 mm. Evaporability exceeds 1000 mm. Humidity ratio is lower than 0.6.

**Soil.** Loamy and skeletal (with small stones) dry gray-cinnamonic soils are developed.

Habitat. Tulipa biflora grows in phryganoid vegetation habitat, particularly in the plant community of thorny goat's-wheat (Atraphaxietum spinosae) (Fig. 6). This plant community is developed on the slope of the macro north-eastern exposure. The micro exposures of the slopes are north-east, southeast and north-west. The slope inclination is  $30^{\circ}-35^{\circ}$ .



**Fig. 6.** Habitat of *Tulipa biflora* Pall. on Iaghluja Ridge (Photo by K. Kereselidze)

The data summary of phytosociological surveys, conducted in April and May, are given below (Table). The field data collection was carried out in the core part of population supporting highest number of specimens.

Table. Key characteristics of Tulipa biflora habitat

Abbreviations: M – meter, N- north, E – east, S – south, W – west, H – hemicryptophyte, G – geophyte, Th – therophyte, Spec. – Specimen

| Date of description                   | 05.04. 2021  | 11.05. 2021 |  |  |  |
|---------------------------------------|--|-------------|--|--|--|
| Physical-geographical characteristics |  |             |  |  |  |
| Location                              | East Georgia, Kvemo Kartli, vicinity of Rustavi, eastern part of Iaghluja<br>Ridge |             |  |  |  |
| Coordinates                           | N41.541083, E44.972217   |             |  |  |  |
| Elevation (m)                         | 500-550  |             |  |  |  |

| Relief   | Slightly hillocky and traversed by trails     |                     |                     |                   |  |  |  |
|--|---|---------------------|---------------------|-------------------|--|--|--|
| Exposition macro   | N-E   | una naversea by tre |                     |                   |  |  |  |
| Exposition micro   | N-E, S-E, N-W                                 |                     |                     |                   |  |  |  |
| Inclination  | 30°-35°                                       |                     |                     |                   |  |  |  |
| Soil   | Grey cinnamonic, clay, small stony            |                     |                     |                   |  |  |  |
| Exploitation   | Pasture                                       |                     |                     |                   |  |  |  |
| Geobotanical characteristics of the vegetation             |   |                     |                     |                   |  |  |  |
| Overall projection coverage (%)                            | 40  |                     | 58-60               |                   |  |  |  |
| Distribution   | Even  |                     | Even                |                   |  |  |  |
| Layer I Layer  | Two vegetation layers Three vegetation layers |                     |                     |                   |  |  |  |
| Average height (cm)  | 30 65-70                                      |                     |                     |                   |  |  |  |
| Projection coverage (%)                                    | 20-25   |                     |                     | 25                |  |  |  |
| Distribution   | More or less eve                              |                     |                     | More or less even |  |  |  |
| II Layer   | 1,1010 01 1000 010                            |                     | 111010 01 1000 0101 |                   |  |  |  |
| Average height (cm)  | 5   |                     | 50                  |                   |  |  |  |
| Projection coverage (%)                                    | 25  |                     |                     | 25                |  |  |  |
| Distribution   | More or less eve                              | n                   | Even                |                   |  |  |  |
| III Layer  | 141010 01 1035 046                            |                     | LVOII               |                   |  |  |  |
| Average height (cm)  | -   |                     | (5)10-25(30)        |                   |  |  |  |
| Projective coverage (%)                                    | -   |                     |                     | 18-20             |  |  |  |
| Distribution   | -   | -                   |                     | More or less even |  |  |  |
| Floristic composition                                      | 1   |                     |                     |                   |  |  |  |
| Species  | Layer   | Coverage (%)        | Layer               | Coverage (%)      |  |  |  |
| Shrubs   |   |                     |                     |                   |  |  |  |
| Atraphaxis spinosa L.                                      | I   | 25                  | II                  | 25                |  |  |  |
| Caragana grandiflora (M.Bieb.) DC.                         | I   | +                   | II                  | +                 |  |  |  |
| Semishrubs and undershrubs                                 |   |                     |                     |                   |  |  |  |
| Artemisia fragrans Willd.                                  | II  | 4-5                 | III                 | 10                |  |  |  |
| Bassia prostrata (L.) Beck [Kochia prostrata (L.) Schrad.] | I   | + (1 spec.)         | II                  | + (1 spec.)       |  |  |  |
| Scutellaria orientalis L.                                  | II  | +                   | III                 | +                 |  |  |  |
| Teucrium polium L.   | II  | +                   | III                 | +                 |  |  |  |
| Thymus coriifolius Ronniger                                | -   |                     | III                 | +                 |  |  |  |
| Perennial herbs (H)  |   |                     |                     |                   |  |  |  |
| Prangos ferulacea (L.) Lindl.                              | II  | 6-7                 | I                   | 25                |  |  |  |
| Malabaila dasyantha Fisch. &                               | 11  |                     | 1                   |                   |  |  |  |
| C.A.Mey ex K.Koch  | II  | +                   | I                   | +                 |  |  |  |
| Astragalus bungeanus Boiss.                                | -   | -                   | III                 | +                 |  |  |  |
| Scorzonera biebersteinii Lipsch.                           | -   | -                   | III                 | +                 |  |  |  |
| Medicago sp.   | -   | -                   | III                 | +                 |  |  |  |
| Perennial herbs (G)  |   |                     |                     |                   |  |  |  |
| Allium sp.   | II  | +                   | III                 | +                 |  |  |  |
| Gagea commutata K.Koch                                     | II  | 1                   | III                 | +                 |  |  |  |
| Poa bulbosa L.   | II  | +                   | III                 | +                 |  |  |  |
| Podospermum canum C.A.Mey.                                 | -   | -                   | III                 | +                 |  |  |  |
| Tragopogon tuberosus K.Koch                                | -   | -                   | III                 | +                 |  |  |  |
| Tulipa biflora Pall.                                       | II  | 3-4                 | III                 | 3                 |  |  |  |
| Biennials (H)  |   |                     |                     |                   |  |  |  |

| Nonea lutea (Desr.) DC.  | II | 1 | III | 1 |
|--|----|---|-----|---|
| Silene cyri Schischk.  | -  | - | I   | + |
| Annuals (Th)   |    |   |     |   |
| Aegilops tauschii Coss.  | -  | - | III | + |
| Carduus pycnocephalus subsp.<br>cinereus (M.Bieb.) P.H.Davis                       | -  | - | III | + |
| Carthamus lanatus L.   | -  | - | III | + |
| Caucalis platycarpos L.  | -  | - | III | + |
| Cerastium pumilum var. glutinosum<br>(Čelak) E.Rico (Cerastium<br>glutinosum Fr.)  | -  | - | III | + |
| Echinaria capitata (L.) Desf.  | -  | - | III | + |
| Erodium cicutarium (L.) L'Hér.   | II | 1 | III | 1 |
| Filago pyramidata L.   | -  | - | III | + |
| Holosteum marginatum C.A.Mey.  | II | + | III | + |
| Lamium amplexicaule L.   | II | + | III | + |
| Linum sp.  | -  | - | III | + |
| Lolium rigidum Gaudin  | -  | - | III | + |
| Medicago minima (L.) L. [Medicago minima (L.) Bartal.]                             | -  | - | III | + |
| Alyssum linifolius Stephan ex Willd. [Meniocus linifolius (Stephan ex Willd.) DC.] | II | + | III | + |
| Papaver arenarium M. Bieb.   | -  | - | III | + |
| Crepis sancta (L.) Bornm.  | -  | - | III | + |
| Rapistrum rugosum (L.) All.  | II | + | I   | + |
| Sideritis montana L.   | -  | - | III | + |
| Thlaspi perfoliatum L.   | II | + | III | + |
| Veronica hederifolia L.  | II | + | III | + |
| Viola kitaibeliana Schult.   | II | + | III | + |

The floristic nucleus of the habitat consists of characteristic species of arid and semiarid ecosystems. They are xerophilous and hemixerophilous plants, growing on the dry loamy and skeleton soils. Almost all plant life forms were present, from which shrubs and perennial herbs (hemicryptophytes) are distinguished by their coenotic role. However, annual weeds and widespread plants were also recorded (Rapistrum rugosum, Carthamus lanatus, Aegilops tauschii, Thlaspi perfoliatum, Carduus pycnocephalus subsp. cinereus, Erodium cicutarium, Papaver arenarium, Filago pyramidata, etc.), which should be considered as a result of grazing. Nevertheless, the structure and condition of the vegetation is satisfactory and corresponds to the physical-geographical parameters and the structure of phryganoid vegetation.

**Data on** *Tulipa biflora*. The area of occupancy of *Tulipa biflora* populations is ca. 350 m<sup>2</sup>. The core population is comprised of unevenly distributed approximately 350-400 individuals (the average density is 2 individuals per 1 m<sup>2</sup>), while other smaller populations support 3-5 individuals each.

Observations of the target species revealed its main phenophases:

Flowering: End of April – May; Fructification: End of April – May; Seed maturity: The second half of May.

The developmental rhythm of *Tulipa biflora* in different parts of the global area are largely the same. However, in some parts of the area, such as the Negev Desert (Israel), it blooms relatively early (early March) [20]. The main phenophases of *Tu*-

*lipa biflora*, growing on Iaghluja Ridge, coincide with its phenophases in different areas.

Threats, concerns and ecological status. The area is used for grazing and is under anthropogenic impact. During the field data collection, cattle were grazing near the area. Given that Iaghluja Ridge is a winter pasture, it is assumed that sheep flocks over-winter here from autumn to early spring. Due to heavy grazing pressure most of the individuals of *T. biflora* showed gnawed leaves (Fig. 7) and a considerable part of them were small and undersized indicating stress-induced flowering (A. Schröter, unpublished raw data).



**Fig. 7.** Damaged leaves of *Tulipa biflora* Pall. (Photo by N. Lachashvili)

Most individuals of *Tulipa biflora* produce flowers and fruits (Fig. 8, 9) and fully complete the developmental life cycle.



**Fig. 8.** *Tulipa biflora* Pall. in fruits (Photo by N. Lachashvili)



**Fig. 9.** Mature fruits of *Tulipa biflora* Pall. with seeds (Photo by K. Kereselidze)

The preliminary regional assessment of *Tulipa biflora* using the IUCN categories and criteria. According to the IUCN categories and criteria [10], based on the decline of distribution area and habitat quality (grazing), the target species in Georgia was preliminarily assessed as critically endangered [CR B1 ab (iii) + 2ab (iii)].

# **Conclusions**

- 1. The populations of *Tulipa biflora*, a species new for the flora of Georgia are located in East Georgia, in the eastern part of Iaghluja Ridge, in the vicinity of Rustavi, at an altitude of 500-550 m asl.
- 2. Physical-geographic characteristics of the area (climate, terrain, soil, etc.) and habitat both largely coincide with the relevant data typical for *Tulipa biflora*.
- 3. The area of occupancy of T. biflora populations is about 350 m<sup>2</sup>.
- 4. The populations of *T. biflora* are composed of approximately 350-400 individuals;
- 5. Most individuals of *T. biflora* produce flowers, bear fruits and fully complete the developmental life cycle.
- 6. The populations of *T. biflora* are under permanent anthropogenic pressure (grazing).
- 7. In accordance with the IUCN regional assessment, *T. biflora* was preliminarily assessed as critically endangered [CR B1ab(iii) + 2ab(iii)].

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