

2019

ISSN 1512-1887

ANNALS OF AGRARIAN SCIENCE



Vol. 17 No. 4

Annals of Agrarian Science

Volume 17, Number 4, September 2019

Aims and Scope

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.

Annals of Agrarian Science

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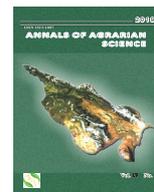
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Annals of Agrarian Science

Journal homepage: <http://journals-org.ge/index.php>



Laccases and their application in bioremediation of organic pollutants

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Received: 10 November 2018; accepted: 18 February 2019

ABSTRACT

Laccases are present abundantly in many white-rot fungi. They are blue multicopper oxidases, which catalyze the monoelectronic oxidation of a broad spectrum of substrates, such as ortho- and para-diphenols, polyphenols, aminophenols, aromatic, and aliphatic amines coupled with a full, four-electron reduction of O₂ to H₂O. Laccase-assisted reactions have a wide application potential. In particular, they decolorize and detoxify the industrial effluents and help in wastewater treatment. In the presence of specific mediators, they act on both phenolic and nonphenolic lignin-related compounds as well as highly recalcitrant environmental pollutants. Several techniques have been developed for the immobilization of laccase to preserve their enzymatic activity. In this review, we describe the fungal source of laccases and their application in environmental protection.

Keywords: Laccases, Enzymatic Biodegradation, Micropollutants, EDC, Immobilization, Enzymes.

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Introduction

Intensive industrial and agricultural activity over the past decades led to considerable contamination of soil and water with toxic organic pollutants having detrimental effects on the health of humans, animals, plants, and microbes. Wastewaters produced by industry and municipalities contain synthetic compounds including pharmaceutical and personal care products (PPCPs), endocrine-disrupting compounds (EDCs) are increasingly discharged through wastewater treatment plants (WWTPs) [1, 2]. This organic compounds, termed micropollutants, are present at such low concentration (pg/L to ng/L) in water that most established treatment processes seldom lead to their elimination and may pose serious risks to the environment and public health [3]. Various effective mechanical, chemical, physiochemical techniques are currently available to remove pollutants but they are complex, quite expensive, may generate toxic intermediates [4-6]. Bioremediation is seen as an attractive and suitable alternative due to its reputation as a low-cost, effective and environmentally friendly approach. Recent fundamen-

tal studies demonstrated the potential of white rot basidiomycetes (WRB) and their lignin-modifying enzymes (LME) application for degradation of various aromatic compounds and treatment of soils and effluents contaminated with organic pollutants [7-10]. Especially, multi-copper phenol-oxidizing laccases (EC 1.10.3.2) secreted by many fungal species associated with the degradation of lignocellulosic material appeared to be the most promising for their environmental applications. Due to a wide range of substrate specificity, these enzymes oxidize a broad range of compounds such as phenols, polyphenols, methoxy-substituted phenols, and amines while reducing oxygen to water; however, laccase activity can be extended to non-phenolic compounds by use of mediators like HBT (1-hydroxybenzotriazole) and ABTS (2,20-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid)), and syringaldehyde [11, 12].

Laccase producers

Laccases are common enzymes in nature and are found widely in plants and fungi as well as in some bacteria and insects [13]. The first bacterial laccase

was detected in the plant root-associated bacterium “*Azospirillum lipoferum*” [14], where it was shown to be involved in melanin formation [15]. An atypical laccase containing six putative copper-binding sites was discovered from *Marinomonas mediterranea*, but no functional role has been assigned to this enzyme [16]. *Bacillus subtilis* produces a thermostable CotA laccase which participates in pigment production in the endospore coat [17]. Laccases have also been found in *Streptomyces cyaneus* [18] and *Streptomyces lavendulae* [19]. Laccase producing *Bacillus* were isolated from soil and the enzyme was involved in phenol degradation [20, 21]. Although there are also some other reports about laccase activity in bacteria, it does not seem probable that laccases are common enzymes from certain prokaryotic groups [22].

Laccases of fungal origins have been the most intensively studied [23]. Fungal laccases are implicated in both intra- and extra-cellular physiological processes including morphogenesis, pigmentation, pathogenesis, and delignification [24-26]. Among fungi, ascomycetes, basidiomycetes, and deuteromycetes can produce laccases, but the WRB are the most efficient lignin degraders and laccase producers [24, 27]. WRB possess a strong biochemical and ecological capacity to degrade environmental organic chemicals either by chemical modification or by influencing chemical bioavailability. Ability of these fungi to form extended mycelia networks, the low specificity of their enzymes and their ability of using pollutants as a growth substrate make them well suited for bioremediation processes. In contrast to bacteria, fungi are able to extend the location of their biomass through hyphal growth in search of growth substrates [10, 28, 29]. Bacteria contain little energy, have a low bioavailability and cannot degrade substances having rare structural elements. Filamentous fungi are more advantageous where translocation of essential factors (nutrients, water, the pollutant itself) is required for the degradation of environmental chemicals by translocation resources between different parts of their mycelium. Fungi also release extracellular enzymes which allow for digestion of energy sources in their surroundings and further diffusion of these molecules through the substrate towards the fungus [30]. Owing to the broad substrate specificity of the LME and potent intracellular enzymes, in particular, cytochrome P450 monooxygenases, WRB are especially well-suited for PAH degradation compared to bacteria. Moreover, they can degrade high mo-

lecular-weight PAHs, whereas bacteria can degrade only smaller molecules. In addition, fungi are also known to produce large quantities of exudates that serve as auxiliary carbon sources for pollutant-degrading bacteria [31] delignification.

Laccases are secreted by WRB along with other ligninolytic enzymes including manganese peroxidase, lignin peroxidase, and versatile peroxidase, although the specific types of enzymes secreted may differ with the fungus [24, 32]. Many fungi contain several laccase-encoding genes, but their biological roles are mostly not well understood [33]. *Pleurotus ostreatus* and *Trametes versicolor* can be regarded as the model organisms in basic and applied laccase research. Other species of *Pleurotus* and *Trametes* genera as well as *Cerrena unicolor*, *Coriolopsis gallica*, *Ganoderma lucidum*, *Phlebia radiata*, *Pycnoporus cinnabarinus*, *Pycnoporus sanguineus* are well-known efficient laccase-producing basidiomycetes. Nevertheless, efforts are still being made with an expectation to screen naturally-occurring laccase overproducers with desired laccase yields and properties and elucidate factors regulating enzyme production [34-39].

Laccase production

The potential applications of LME in industrial and environmental technologies require huge amounts of these enzymes at low cost [27, 40]. Enhancing laccase yields is essential to lower production costs and promote industrial applications of the enzyme. Laccase production and yield are directly influenced by some parameters such as the substrate to be used, the fermentation technique, agitation, aeration, and cultivation time, among others. The production of laccase is linked straight to a complex regulation of nutrients that affects directly the expression of laccases, comprising the carbon and nitrogen sources, their respective concentration and the relationship between them, and also the concentrations and kind of inducers used [41]. However, no generalization can be made on the best nutrients or inducers and their optimal concentrations since basidiomycetes display a wide diversity in their responses to individual compounds [40, 42, 43]. Lignocellulosic wastes containing carbohydrates and inducers are often added resulting in benefits such as laccase production enhancement, lower production costs, and waste reuse [44, 45].

Literature describing laccase induction by xenobiotics, e.g., lignin breakdown products, dyestuffs

and organic pollutants evidences that the effects of organic compounds on laccase production depend on the compound structure, fungal strain, growth stage as well as the culture medium [42, 46]. Combinational induction of laccase production by metal ions and organic compounds can be either synergistic [46] or antagonistic [47].

Co-culture of laccase-producing strains with other microbes can be more advantageous and effective than chemical induction to enhance laccase production either through yield increase or induction of new isozymes. Microbial interactions with laccase inducing effects vary with the strain, but the structure of inducing metabolites and the inducing mechanism remain largely unknown [44, 48-52]. One proposed mechanism for laccase overproduction in the co-culture process is carbon source succession. Li et al. found that glycerol produced from glucose by the yeast *Candida* sp. is an efficient carbon source for *G. lucidum* upon glucose deprivation and crucial for laccase overproduction by prolonging laccase secretion time [52].

Molecular and catalytic properties of laccase

Laccases are copper-containing oxidoreductases (EC 1.10.3.2). More than 100 laccases have been isolated and characterized to various extents. Molecular structure of fungal laccases occurs often as isozymes with monomeric or dimeric protein structures. Both intracellular and extracellular isozymes may be produced from a single organism.

Laccases are glycoproteins with molecular weights 50–130 kDa. The carbohydrate portions of plant laccases constitute up to 45% of the molecule weight, whereas fungal laccases contain less carbohydrates (10–30%), that catalyse the monoelectronic oxidation of various substrates (e.g. phenols, and aromatic or aliphatic amines) to the corresponding radicals, using molecular oxygen as the final electron acceptor [53-56]. Laccases play important roles in several biometabolic steps including those involved in fungal pigmentation, plant lignification, lignin biodegradation, humus turnover and cuticle sclerotization, where in naturally occurring low-molecular-weight phenolic compounds and natural fibre polymers are utilized as substrates [56].

Laccases from different sources exhibit a wide range of redox potentials. The T1 site has a high redox potential reaching 780 to 800 mV for the *T. ver-*

sicolor and *Neurospora crassa* enzymes, whereas the plant *R. vernicifera* enzyme has a value of 420 mV [57, 58]. The redox potentials of T2 and T3 sites for the *R. vernicifera* laccase are, respectively, 390 and 460 mV at pH 7.5 [59]. The *T. versicolor* laccase T1 and T3 sites have been reported to be 785 and 782 mV, respectively [60]. In general, the T1 sites in fungal laccases are much higher than those of plant laccases and other blue copper oxidases, although significant differences in potentials also exist among fungal laccases [61]. Laccases exhibit low substrate specificity and can oxidize a range of compounds, such as diphenols, aryl diamines, and aminophenols. The K_m values are generally in the range of 1–10 mM, and the change in $\log(k_{cat}/K_m)$ increases proportionally to the redox potential difference between the T1 site acceptor and the substrate donor [62, 63].

Usually, fungal laccases have an optimal pH in the acid range, and the temperature profiles for the activity of these enzymes normally do not differ from those of other extracellular ligninolytic enzymes. Purified laccase was incubated in buffers of varying pH between pH 2.0 and 8.0 for up to 180 min to determine its pH stability. Similarly, some authors showed that the purified enzyme dissolved in 0.1 M sodium acetate, pH 5.0, was incubated at varying temperatures (40–80 °C) for up to 180min to determine its thermal stability. Laccase was found to be most stable at pH 5.0 and 30°C and 5(d)). Laccase activity decreased significantly (93%) after 180 min at pH 8.0 and it was fairly stable between pH 3.0 and 7.0 for up to 180 min. While the enzyme was relatively stable below 50°C, activity decreased significantly when the temperature was 70°C or higher. In fact, laccase activity was completely lost within an hour at 80°C and 5(d) [64].

Mediators of laccases

The biotechnological importance of laccases have increased after the discovery that the enzyme substrate range could be further extended in the presence of small readily oxidizable molecules called mediators [65]. Generally, the laccase reactivity decreases with the increase of the substrate size; therefore, the limited substrate accessibility is overcome through the use of appropriate laccase mediators. As the substrate due to its size cannot enter the laccase active site, the mediator acts as a carrier of electrons between the enzyme and the substrate thereby overcoming the steric hindrances that exist

between them [66]. In this case, during the initial reaction step, the mediator is oxidized to stable intermediates with high redox potential by laccase [66]. The ideal mediator should be non-toxic, economic, and efficient, must be a good laccase substrate and reduced forms that do not inhibit the enzymatic reaction [67]. Moreover, the redox mediator should be able to continuously maintain the cyclic redox conversion. Suitable mediators are needed to maximize the potential of laccase use as a bioremediation agent. The most commonly used mediators are the ABTS and the 1-hydroxybenzotriazole (HBT) [68]. ABTS [2,2-Azino-bis (3-ethylbenzthiazoline-6-sulphonic acid)] is the best organic redox mediator. Its use for oxidation of non-phenolic lignin structures gave impetus to search for new laccase mediators [69]. It is well known that cation radicals represent an intermediate oxidation step in the redox cycle of azines and, upon extended oxidation and abstraction of the second electron, the corresponding dictations can be obtained. These cation radical and dictation play role in the oxidation of the substrates, non-enzymatically. The redox potentials of $ABTS^+$ and $ABTS^{2+}$ were evaluated as 0.680 V and 1.09 V respectively [70].

1-hydroxybenzotriazole - HBT is another important organic redox mediator. HBT belongs to the N-heterocyclic compounds bearing N-OH groups mediators [69]. Consuming oxygen HBT is converted by the enzyme into the active intermediate, which is oxidized to a reactive radical (R-NO \cdot) and HBT redox potential has been estimated as 1.1-1.2 V [70]. HBT acts as a radical mediator rather than ionic, but a severe factor limiting catalytic productivity is again deoxygenation to benzotriazole. Indeed, for recalcitrant substrates in the presence of large amounts of enzyme and HBT, deoxygenation is the predominant process seen. When this occurs, rapid deactivation of the enzyme is also seen, suggesting some interaction of the oxidized mediator with the protein [71]. It has been proposed that the HBT radical forms a coupling intermediated product with lignin [72]. The intermediate product can subsequently degrade to release the reduced form of benzotriazole (BT), or form a stable complex that binds some of the HBT to lignin via covalent bonding.

2,2,6,6-tetramethyl-1-piperidinyloxyl (TMPO) is more efficient than ABTS, HBT, or the natural laccase mediator 3-hydroxyanthranilic acid. This mediator is present in the solution a stable N-oxyl radical, which can perform primary modification of some high potential substrates even without laccase [73]. Laccase oxidizes TMPO to produce the

oxo-ammonium ion, which reacts with the substrate. Proton removal yields the oxidized product and the reduced form of TMPO is converted to the oxidized form by laccase and then to oxo-ammonium ion. The laccase-TEMPO acts according to the ionic mechanism, whereas the HBT-mediated reactions favor the radical mechanism [73].

Despite all the associated advantages of using these mediators, there still have several drawbacks from a practical point of view: i) low catalytic efficiency, excess amounts of mediator (typically 30 mol% on substrate) are needed, some even need more than 1 equiv; ii) in some cases, laccase is inactivated by the mediator radicals, or the latter can be transformed into inactive compounds with no 10 more mediating capability (e.g. generation of benzotriazol from HBT by losing the hydroxyl group); iii) some of mediators can generate toxic derivatives; iv) long reaction time are usually required. In view of the aforementioned drawbacks of existing routes and the desire for aerobic oxidation of multi-functionalized 15 complex molecules, the development of new and highly efficient laccase-mediator system is particularly attractive [74].

Enzyme immobilization

Silicon dioxide offers a high versatility (particle size, surface area, porosity, surface chemistry, ease of synthesis, and scalability) and represents the support of choice for the enzymatic immobilization. Furthermore, silica materials are considered neutral and nontoxic and matches the natural environments.

Enzyme immobilization is one of the most promising techniques for highly efficient and economically competent biotechnological processes in the field of environmental monitoring, biotransformation, diagnostics, pharmaceutical and food industries. The immobilization of the biocatalysts onto a solid support can reduce the enzyme loss, limit the denaturation problems, and facilitate their possible reuse. Additionally, immobilization improves many properties of enzymes such as performance in organic solvents, pH tolerance, heat stability or the functional stability [13]. Laccase immobilization has been addressed as a way to intensify and stabilize biocatalytic activity [75].

The immobilization technique is based on the covalent attachment of oxidative enzymes on the nanoparticles. Different synthesis routes of nano-particulate materials exist or are under development. They comprise micro- and mini-emulsion-

based techniques, monophasic synthesis according to the method of Stoeber et al. [76] and flame spray pyrolysis (FSP). Due to the high specific surface of nontoxic nanostructured support material, fumed silica nanoparticles (fsNP) are expected to be of interest for the immobilization of laccases [77]. Furthermore, these systems are relatively cheap and are produced at industrial scale [78].

A covalent immobilization method based on glutaraldehyde and amino-functionalized SBA-15 supports has been successfully applied to covalently and stably immobilize laccase from *Trametes versicolor*. Aminopropyl-based laccase-SBA-15 biocatalysts displayed the best reusability properties, retaining higher activity after four repeated uses than the corresponding aminobutyl-based materials [79]. Alkyl moieties displayed higher enzyme loadings than phenyl moieties, being more adequate the larger n-butyl tethering residue likely due to its higher mobility.

Carbon materials have been long preferred as carriers for enzyme immobilization, mainly because they may provide a large surface area relative to the enzyme loading. In what concerns carbon nanotubes (CNTs), their exclusive structure and properties have been intensively studied in distinct applications ranging from energy storage, biotechnology and environmental remediation [80]. Additionally, CNT surface can be easily functionalized, tuning their properties towards specific applications and enhancing their efficiency either as supports or catalysts. Polysulfone (PSf) membranes are widely used in water applications due to their excellent heat resistance and chemical stability over a large range of pH [81].

Higher immobilization efficiency and recovered activity were obtained using CNTs oxidized with HNO_3 0.30 M (CNTox-0.30), which is related to the higher number of oxygen-containing groups available at the surface of the material. The laccase/CNTox-0.30-EN bioconjugate was successfully used for the treatment of a mixture of four phenolic compounds, the process showing similar efficiency as the analogous using the free enzyme, with the possibility of reusing the biocatalyst [82]. For improving stability of immobilized white-rot fungus to treat various effluents, some authors performed study where *Phanerochaete chrysosporium* cells and the combined cross-link enzyme aggregates (combi-CLEAs) prepared from *Trametes versicolor* were co-immobilized into the Ca-alginate gel particles in this paper [83]. The activity yields of obtained combi-CLEAs were 42.7% for lignin peroxidases (LiPs), 31.4% for manganese peroxidases

(MnPs) and 40.4% for laccase (Lac), respectively. And their specific activities were 30.2 U/g as combi-CLEAs-LiPs, 9.5 U/g as combi-CLEAs-MnPs and 28.4 U/g as combi-CLEAs-Lac. The combi-CLEAs improved the adaptability of the white-rot fungal particles to adverse environmental conditions.

The experimental results indicated that magnetic graphene oxide (MGO) nanomaterials were synthesized based on covalent binding of amino Fe_3O_4 nanoparticles onto the graphene oxide (GO), and the prepared MGO was successfully applied as support for the immobilization of laccase from *Trametes versicolor* (commercial enzyme from Sigma-Aldrich), it effectively improved the processing efficiency and expanded the industrial application of enzymes [84]. Compared with free laccase, the MGO-laccase exhibited better pH and thermal stabilities. The optimum pH and temperature were confirmed as pH 3.0 and 35°C. Moreover, the MGO-laccase exhibited sufficient magnetic response and satisfied reusability after being retained by magnetic separation. The MGO-laccase maintained 59.8% activity after ten uses. Compared with the free enzyme, the employment of MGO as enzyme immobilization support could efficiently enhance the availability and facilitate the application of laccase.

The operating stability of the immobilized enzyme is a very important parameter. Immobilized enzymes may be easily separated from the reaction solution and reused, which greatly decreases costs of the enzyme and increases its significance for practical application [85-87]. The immobilization of laccase from *Trametes versicolor* on the controlled porosity carrier (CPC) silica beads allowed laccase to remain stable and maintain more than 85% of its initial activity after 30 days. After immobilization, laccase degraded more than 90% of 2,4-dinitrophenol within 12 h of treatment [88].

Decrease in the activity of the immobilized laccase as a result of repeated usage may be expected due to the possibility of enzyme denaturation during the operation process. What is interesting, Wang et al. [86], observed the highest activity of commercial laccase immobilized on polyacrylonitrile or polyacrylonitrile/montmorillonite/graphene oxide (PAN/MMT/GO) at the second or third cycle. It is possible that during repeated usage, the membrane became slack and downy, contributing to more sites for the enzyme to reach the substrate.

Enzyme immobilization is the key step of biocatalytic membrane preparation which is promising in food, pharmaceutical, and water treatment

industries [89]. Polysulfone membranes containing functionalized CNTs demonstrated to be also an excellent support for the enzyme (re)-immobilization and their application in the degradation of 4-methoxyphenol. The PSf membrane with 0.1 wt% of CNTox-0.30-EN was the most efficient, presenting a comparable activity to CNTs solely and with a much lower CNT content [82].

Potential of laccases to solve ecological problems

The release of untreated industrial effluents into sewers that will empty into rivers represents a very big problem [90]. Sewage can contain many substances from industrial processes with carcinogenic, mutagenic, and teratogenic potential, including toxic effects to human beings and many fish and microorganism species. So the appropriate treatment of these wastewaters is important and can be realized by various methods, although each technique has some limitations [91]. Using of enzymatic processes in wastewater treatment is relatively new and has the advantage of decreasing the consumption of reagents and degradation of subproducts. Among the possible alternative methods can be the use of ligninolytic enzymes capable of degrading many toxic and persistent substances such as dyes, solvents, inks, pesticides, fertilizers, and others [92]. Moreover, enzyme treatments are eco-friendlier and energy efficient compared to chemical treatments [93, 94].

One of the applications of laccases (together with the peroxidases) in wastewater treatment is related to the textile industry, because of the necessity of eliminating liquid residues containing textile dyes that remain in the water after the dye industrial process. Synthetic dyes often have low biodegradability, several are toxic, carcinogenic and mutagenic and it is not uncommon that their degradation products show similar properties. Furthermore, the absorption and reflection of the sunlight by dyes in surface waters interferes with bacterial and plant growth, disturbing the ecological balance [95, 96]. Therefore, decolorization of wastewaters discharged in textile processing is a major problem. In recent years it was shown that many industrial dyes could be decolorized by fungal laccases frequently immobilized on different supports, such as glass-ceramic materials, imidazole-modified silica, montmorillonite, alginate-gelatin mixed gel, hydrophobic sol-gel and green coconut fiber [97-99]. Laccases physically entrapped or

immobilized by their covalent attachment to various supports are the most commonly used in decolorization processes [99]. The decolorization of waste waters by immobilized laccases is the result of enzymatic catalysis and support adsorption [98, 99]. It was shown that alginate and chitosan are biopolymers, which can be used as sorbents to remove dyes from the aquatic solutions. Although the entrapment of laccases in alginate mixed gels or hydrophobic Sol-gel decreases their activity and dye affinity, it improves their pH stability, thermostability and enables reusability of the enzymes [99]. Additionally, the immobilization of laccases on such insoluble supports limits their conformational changes that increases their operational stability and durability [99]. For example, the entrapment of laccase in alginate-gelatin, alginate-chitosan mixed gels or in hydrophobic Sol-gel matrix of trimethoxysilane and propyltetramethoxysilane led to its significant stability towards heat denaturation. However, under these conditions lower affinity of the enzyme to the substrates caused by diffusional limitations and decreased protein flexibility was observed [97-99].

In turn, Cristóvão et al. [98] immobilized a commercial laccase on green coconut fibers and verified the ability of the enzyme to decolorize different textile dyes. Decolorization is limited by the concentration of a mediator such as 1-hydroxybenzotriazole (HBT) and 2,2'-azinobis(3-ethylbenzthiazoline-6-sulfonate) (ABTS) [97,98]. The addition of ABTS during dye decolorization by laccase immobilized on green coconut fiber significantly increased enzyme activity [98]. The same effect was observed during decolorization carried out by laccase entrapped in the hydrophobic Sol-gel [97]. The desired properties of immobilized laccases such as improved characteristics, stability and reusability obtained after they are immobilized on different carriers show the suitability of these biocatalysts for continuous treatment of different industrial effluents.

Some authors demonstrated that immobilized laccase has potential applications in dyestuff treatment. Laccase encapsulation in Cu-alginate gels was active at different conditions of pH, temperature and ionic strength, and was able to decolorize the carcinogenic dyes Trypan Blue, Bromothymol Blue and Coomassie Brilliant blue R with yields close to 90% without mediator addition [100]. The Cu-alginate derivatives retained more than 70% of catalytic activity for at least 430 h of continuous

use. The decolorization of synthetic dyes by laccase from *Cotylidia pannosa* grown in submerged culture and used to investigate its ability to decolorize synthetic dyes showed that the fungus as well as the crude laccase preparation decolorized the synthetic dyes such as Congo red, bromophenol blue and coomassie brilliant blue R-250 to different extent. A decolorization efficiency of 94% by fungal biomass and 40% by crude laccase was observed for congo red [101].

A laccase (Lacps1) produced by *Pycnoporus sanguineus* RP15 grown in wheat bran and corn-cob under solid-state fermentation was purified and characterized and the potential of the pure Lacps1 and the crude culture extract for synthetic dye decolorization was evaluated. The data suggested good potential for treatment of industrial dye-containing effluents [96]. The maximum decolorization percentages of bromophenol blue (BPB), remazol brilliant blue R and reactive blue 4 (RB4), at 25 or 40 °C without redox mediators, reached 90%, 80% and 60%, respectively, using either pure and free Lacps1 or the crude extract.

The laccase from *Trametes* sp. MA-X01 is one of the promising candidates for the decolorization of textile wastewater containing various dyes, without redox mediators, the free enzyme had a good ability to degrade different kinds of dyes, including azo, heterocyclic and triphenylmethane dyes [102]. The characteristics of that laccase from the strain *Trametes* sp. MA-X01 were studied. The dye decolorization also researched to determine the degrading capability of laccase produced from *Pleurotus eryngii*, methyl orange, tartrazine, reactive red 2 and reactive black dyes were treated with this enzyme (SSB) [103]. The highest decolorization was performed with methyl orange as $43 \pm 2.8\%$ after 5 min of treatment among them.

Purified free laccase from *Coprinus comatus* showed 49.3% decolorizing activity against remazol brilliant blue R (RBBR) and 41.6% decolorizing activity against Poly R-478 after 12 h incubation [104]. MGO-laccase were utilized in the decolorization of dye solutions and the decolorization rate of crystal violet (CV), malachite green (MG), and brilliant green (BG) reached 94.7% of CV, 95.6% of MG, and 91.4% of BG respectively. The experimental results indicated the MGO-laccase nanomaterials had a good catalysis ability to decolorize dyes in aqueous solution [105]. The immobilized enzyme in the developed scalable bioreactor system showed high efficiency in degrading various synthetic dyes

under non-buffered conditions, in particular the indigoid dye Indigo Carmine [105].

Due to high efficiency and low-cost degradation of the pollutant, laccases are frequently used for the treatment of contaminated environments [85]. Given its environmental impacts, it is important to remove pesticides and herbicides compounds from wastewater [109]. Data showed that the laccase extracted from *Trametes versicolor* with an appropriate mediator promoted the degradations of five selected pesticides including atrazine, chlorothalonil, isoproturon, pyrimethanil and chlorpyrifos. Among them, the optimizing mediators were HBT for pesticide atrazine. Its degradation rate was up to 75.0% with HBT as a mediator. But with other mediators, the rates were found to be ranging from 21.7% to 38.9%. For isoproturon and pyrimethanil, the best mediators were found to be acetosyringone, ABTS, HBT and violuric acid. Syringaldehyde and vanillin were the best mediators for chlorpyrifos only. As the results shown, the mediators play important roles in the degradation of the pesticides by laccase catalysis except for chlorothalonil. The degradation of isoproturon and pyrimethanil reached above 60% in the presence of a proper mediator after 6 and 10 h incubation, respectively. However, it took 2 days for other selected pesticides to reach the similar degradation (60%).

Isoproturon belongs to type of phenylurea herbicide and produces a carcinogenic metabolates, has negatively affects to the environment [109]. The data demonstrated that the degradation rates of isoproturon and pyrimethanil were higher than other three pesticides. With violuric acid as a mediator, the decline rates of isoproturon and pyrimethanil were approximately 98% within 24h. The degradation of atrazine, chlorothalonil, and chlorpyrifos ranged from 70.4% to 91.6% (8 days) with an appropriate mediator. However, without any mediator, the degradation rate of chlorothalonil was 78.6%. Similarly, for other four pesticides, the rates were from 6.1% to 38.9% [108]. The kinetic parameters of reactions catalyzed by laccase immobilized on CMMC indicated less affinity for the substrate than that of the free enzyme. Nevertheless, the immobilized enzyme during 12 h utilized 78% and 84% of phenol and p-chlorophenol, respectively [85]. A similar high efficiency of degradation was observed by Wang et al. [86] and Xu et al. [87]. Laccase immobilized on magnetic Cu²⁺-chelated silica support removed pentachlorophenol with 82.9% efficiency [86],

whereas conjugation of the enzyme onto the surface of polyacrylonitrile electrospun fibrous membrane resulted in 87% efficiency of 2,4,6-trichlorophenol removal in four hours [87]. Using the aminobutyl-based laccase-SBA-15, 82%, 73%, and 55% conversion of naphthalene, phenanthrene and anthracene, respectively, were achieved after 48 h, very close to the values obtained with free laccase under the same reaction conditions [79].

Pharmaceutical compounds have shown toxic impact on aquatic ecosystems, specially related to endocrine disruption and reproductive disorders, and their bioaccumulation can also create toxic effects in humans [110]. These compounds as well as their by-products have been detected in both surface water and groundwater [110-113].

Researchers have used laccases in different forms for the degradation of a wide spectrum of contaminants particularly EOPs. The efficiency of the enzyme has been tested: (a) in a free form, (b) with co-substrates to promote the reaction and raise the redox potential as a “laccase-mediator system”(c) and in an immobilized. Most frequently types of micropollutants detected in water supplies are industrial pollutants [114] and pharmaceutical compounds such as antibiotics, analgesics and anti-inflammatory drugs [115] since WWTPs are not well-designed to remove harmful pollutants present at trace concentrations [116]. Diclofenac sodium (DFC), 5,7-Diiodo-8-hydroxyquinoline (DHQ), b-Naphthol (b-NP) and 2,4-dichlorophenol (2,4-DCP) belong to a non-steroidal anti-inflammatory drugs, prescribed as antipyretic analgesic [23] are classified as a harmful environmental pollutant because of their toxicity and biomagnification in the food chain [117]. Due to its extensive use, DCF has been found in several water supplies, even in groundwater at concentrations between 3.6 and 580 ng [118, 119]. Some authors studied the biocatalytic ability of free laccases from filtered culture supernatant of *Pycnoporus sanguineus* without mediators and under mild reaction conditions. This 100 U L⁻¹ laccase cocktail removed 50% Diclofenac, 97% b-Naphthol and 71% 2,4 Dichlorophenol within 8 h of reaction and 78% for 5,7-Diiodo-8-hydroxyquinoline within 3.5 h; at initial concentrations of 10 mg and at 25°C. Furthermore, this enzyme cocktail also removed in excess of 53% all tested compounds in a real groundwater sample from northwestern Mexico [120, 121].

The biotransformation of ritalinic acid by laccase was investigated for the first time. Free and immobilized enzymes were applied both with and

without redox mediators. The results showed that free and immobilized enzymes alone did not transform ritalinic acid. The effective transformation of ritalinic acid was observed only in TEMPO-mediated systems; however, laccase activity in these systems was reduced significantly. The most effective enzyme was the free laccase of *T. versicolor* 159. Immobilization onto nanoparticles caused an increase in transformation efficiency only for the laccase of *C. unicolor* 303 [122].

Magnetic cross-linked enzyme aggregates (M-CLEAs) were prepared for Cerrena laccase and used in antibiotic treatment. Of the seven antibiotics were examined in some study, Cerrena laccase M-CLEAs were most effective in degradation of tetracycline (TC) and oxytetracycline (OTC), followed by ampicillin, sulfamethoxazole and erythromycin. The redox mediator ABTS was not able to improve efficiencies of degradation of TC and OTC. Cerrena laccase at 40 U/mL eliminated 100 µg/mL TC at pH 6 and 25 °C in 48 h in the absence of a redox mediator, with over 80% degradation occurring within the first 12 h. Laccase treatment also significantly suppressed the antimicrobial activity of TC and OTC [23].

Conclusion

Because of their specific nature, laccases are receiving much attention from researchers around the globe. The interest in utilizing laccases for biotechnological applications has increased rapidly since the discovery of these enzymes potential application in emerging technologies including selective delignification in pulp bleaching, conversion of lignocellulosic materials into feed and biofuel, and treatment of environmental pollutants and toxicants generated in various industrial processes. Therefore, laccases have been widely studied for various applications, including the functionalization of lignocellulosic materials, wood fiber modification, and the remediation of soil and contaminated effluents as well as their use in biosensors. However, much more research is required to make use of laccases to protect environment and for other industrial applications.

Acknowledgment

The authors gratefully acknowledge the support of the Shota Rustaveli National Science Foundation of Georgia (SHRNSF), grant agreement #YS-2016-

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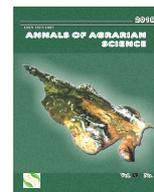
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Use of a Bacteriophage Cocktail to Reduce *Salmonella* Colonization in Experimentally Infected Chickens

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Received: 12 August 2019; accepted: 29 October 2019

ABSTRACT

Antimicrobial-resistant *Salmonella* is dangerous for animal health with possible transmission from animals to humans. The bacteriophages may be a safe, effective alternative of antibiotics for the treatment and prevention of *Salmonella* colonization in poultry. The main goal of this work is to study the efficacy of *Salmonella* phage cocktail in elimination, reduction and prevention of colonization in poultry of the chicken infectious model. Four groups of experimental animals (n=24) were enrolled in this experiment. Chickens of three groups were challenged orally with a single dose of salmonella (10^6 CFU/chicken). Group 1 - received orally 1 ml salmonella phage cocktail 10^9 PFU per chicken on the day before challenge, immediately after the bacterial challenge and 1 treatment per day the next 8 days. Group 2 – treated with the same dose of cocktail for the next 10 days post-challenge. Group 3 - treated with dialysis buffer for 10 days post-challenge (control group 1) and Group 4 - non-infected chickens (control). In each group, 4 chickens were euthanized on days 1, 3, 5, 7, 10 and 21. The cecum of chickens was checked for *Salmonella* quantification. The experiment results show that the use of phage cocktail before infection (group 1) significantly reduced colonization of *Salmonella* and show complete and irreversible elimination of the pathogen after 5 days post-challenge. The complete elimination of pathogen was reached on the 7th day of treatment in group 2.

Keywords: Bacteriophage cocktail, *Salmonella*, Antibiotic resistance, Elimination, Poultry, Infection.

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Introduction

Salmonella species induce bacterial illness and are also one of the leading causes of hospitalization among all the foodborne bacterial pathogens [1, 2].

CDC estimates *Salmonella* causes about 1.2 million illnesses, 23,000 hospitalizations, and 450 deaths in the United States every year. Among the illnesses acquired in the United States, CDC estimates that food is the source for about 1 million illnesses, 19,000 hospitalizations, and 380 deaths [3].

Salmonella can be found in different types of food, ranging from poultry, pig and bovine products and meat to vegetables, fish or other fishery

products. Most *Salmonella* associated infection occurs after ingestion of contaminated foods - mainly meat, poultry, eggs, milk, and vegetables.

Salmonella bacteria are a major problem in the poultry industry. This is largely the result of the entry of these bacteria into the human food chain through poultry. In chicks *Salmonella* colonizes the gastrointestinal tract but does not cause clinical disease. Infected chicks are able to shed pathogen into the environment for extended periods of time increasing the risk for environmental contamination, spread of the organism within the flock and contamination of the food supply [4,5].

Human *S. Enteritidis* cases are most commonly associated with the consumption of contaminated eggs and poultry meat. Reduction of the number of *S. Enteritidis*-contaminated eggs or egg contamination in flocks of laying hens is a main target for reduction of human salmonellosis. Food can become contaminated with pathogens at every stage from “field to fork.” Being free of pathogens or lightly contaminated at a farm, food might arrive heavily contaminated by the time it reaches the consumer. Controlling pathogens on the farm is a complex process, and for many years the main line of defense in disease prevention and treatment has been antibiotics, but important public health concern is the emergence of antibiotic resistant strains of *Salmonella* [6,7].

Bacteriophages (phages) are the natural enemies of bacteria and have proven to be a valuable natural weapon to fight against disease. Phages show great promise as alternatives to traditional antimicrobials in the control of pathogens. The extreme specificity of phages makes them ideal candidates for applications designed to increase food safety during production process (including the quick and specific identification of unwanted viable pathogens in food), and for decontaminating food surfaces and equipment surfaces in food-processing facilities [8-11].

The aim of this study was to test the efficacy of application of *Salmonella* phage cocktail for elimination, reduction and prevention of colonization in poultry of the chicken infectious model.

Methods

Bacteriophages: *Salmonella* phages Sal.phi13, Sal.phi18, and vB.Stm 21 were used in this experiment. Data about the cocktail's phages: phage morphology, host range, restriction analysis of genome, in vitro efficiency have already been documented [12].

Propagation of the phages in the liquid medium

To amplify the phages, 10 ml of overnight culture of host strains (10^9 CFU/ml) and 1 ml of corresponding phage were added to 500 ml of Luria-Bertani (LB) broth and incubated in the shaker at 37°C for 6 hrs. After that, 10 ml of chloroform was added to the cell-phage lysate to release any progeny phage, which remained associated with the host cells. Then, the suspension was incubated for additional 10 min in the shaker at 37°C. To remove bacterial debris, the suspension was centrifuged at

5,000 x g for 15 min; the supernatant was carefully separated and filtered through 0.22- μ m Millipore filters. The phage lysates were stored at 4°C.

Preparation of concentrated phage stocks (Bilayer agar method)

100 μ l of the host bacterium culture grown overnight and 1 ml of phage (10^3 PFU/ml) were mixed; then 3 ml of molten soft-agar (0.7%) was added to each tube and the mixture was gently vortexed and poured over LB agar plates (1.5% agar). The Petri dishes were incubated at 37°C. After 18-20 hours incubation, 3 ml of broth was spread over the agar and left for 15-20 min. Using spreading rod or spatula, the soft-agar with broth were scraped and transferred to a centrifuge tube, and centrifuged at 6000 g for 45 min. The supernatant was filtered through 0.22 μ m filter, transferred into the sterile vial and titrated.

Preparation of phage cocktail. The preparation was carried out individually for all three phages and the cocktail was mixed according to the adequate concentration in the proportion 1:1:1 of each phage.

Animals. One hundred twenty laying hens, aged 3 weeks, were obtained from two commercial grower salmonella-free poultry farms near Tbilisi. The hens were not vaccinated against *Salmonella* spp.

Bacterial inoculum. The *Salmonella* strain used in this experiment was a poultry isolate of *Salmonella* *tiphimurium*. A stock culture was prepared in sterile phosphate buffered saline (PBS) and was used for inoculation at a dose of 10^6 CFU/chicken.

Experimental design

Chickens under the experiment (96 chicks) were randomly distributed into four groups, each group contained 24 chickens (n=24). Each group of birds was placed individually, provided feed and water and libitum. Chickens of three groups (group 1-3) were challenged orally at day two with a single dose of salmonella (10^6 CFU/chicken). Chickens of group 4 was left as a negative control and not inoculated with phage cocktail or *Salmonella*.

All chicks of group 1 were orally treated with the *Salmonella* phage cocktail 10^9 PFU per chicken before challenge, immediately after the bacterial challenge and the next 8 days 1 treatment per day. Chicks of group 2 – were treated with the same dose of cocktail the next 10 days after challenge, 1 treat-

ment per day. Chicks of group 3 – were treated with dialysis buffer the next 10 days after challenge, 1 treatment per day (positive control group).

Sampling

In each group, 4 chickens were euthanized on days 1, 3, 5, 7, 10 and 21 post challenge. The cecum of chickens was checked for *Salmonella* quantification. The cecum from each chicken was removed aseptically. Each sample was transferred to a sterile plastic bag and transported at approximately $4 \pm 2^\circ\text{C}$. Laboratory testing of all samples took place within 24 h after sampling.

Bacteriology

The homogenized cecum samples (0.25 g each) were diluted in 2.25 mL PBS (pH 7.0), decimal dilutions were prepared in PBS and spread onto XLT- plates to enumerate *Salmonella*. The plates were incubated at 41°C for 24 h, and the number *S. typhimurium* cells per g of cecal contents determined. The isolates were biochemically identified as *Salmonella* (methyl red, citrate utilization, triple sugar iron and catalase tests positive & negative to urease and indole tests).

Statistical Analyses

The mean and SEM for each cytokine/chemokine were calculated at each time and statistical analyses performed (Student's t-test). For all analyses, significance was considered if $P \leq 0.05$.

Results

For formulating phage cocktail 3 phages with wide, complementary, not-fully-overlapping host ranges were selected. *Salmonella* phages Sal.phi13, Sal.phi18, and vB.Stm 21 were mixed in the proportion 1:1: 1 (Fig 1).

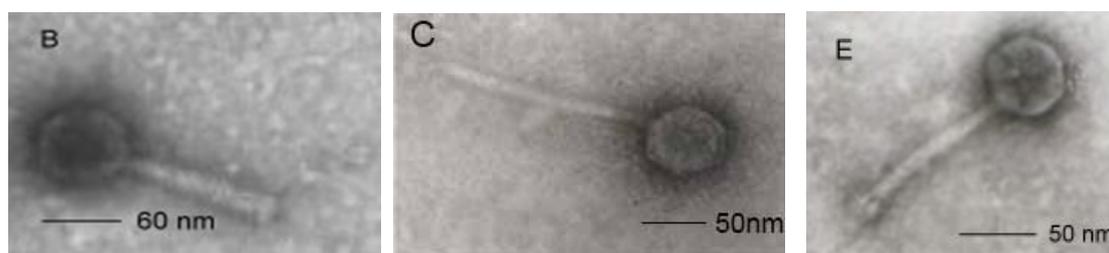


Fig. 1. B- *Sal.phi13*, C- *Sal.phi18*, and E- *vB_Stm 21* phages micrograph

We have studied the efficacy of this phage cocktail in the chicken model. 3 groups of chicken were infected by oral inoculation of the same dose *S. Typhimurium* (10^6 CFU/chicken). Chickens of groups 1 and 2 treated with the bacteriophage cocktail 10^9 PFU per chicken using different treatment schedules.

Significant reduction in *Salmonella* concentration (on day 5) and total elimination (on day 7) was reached when chickens were treated with bacteriophage cocktail 1 day after bacterial infection and then again next 9 days (group 2). On day 5 *Salmonella* concentration in cecum of chickens group 2 was decreased significantly compared to the control group ($1.6 \log_{10}$ vs $6.6 \log_{10}$) (Fig.2). From day 7 to the end of the experiment (day 21) all the chicks were cleared for *Salmonella*.

Treatment of chickens with the *Salmonella* phage cocktail before bacterial challenge immediately after the challenge and the next 8 days caused a significant delay in bacterial colonization of cecum ($1.6 \log_{10}$ Vs $3.0 \log_{10}$ on day 1 after challenge), and total elimination of *Salmonella* on day 5 (Fig 3). From day 7 to the end of the experiment all the chicks were cleared for *Salmonella*. In chickens of group 4 (negative control group) no pathogens were isolated during the whole course of experiment (Fig 4).

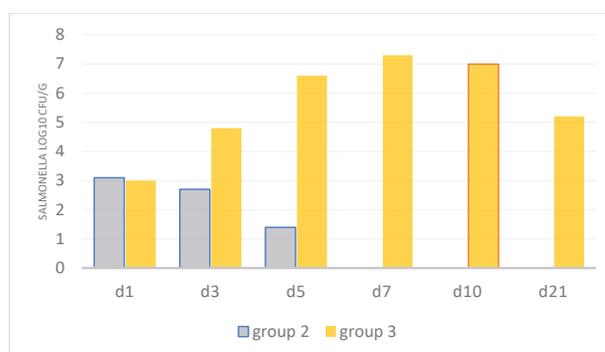


Fig. 2. Concentration of *S. Typhimurium* in the cecum of chickens post bacterial challenge. Group 2-chickens treated with salmonella phage cocktail 10 days post infection, Group 3 – untreated chickens.

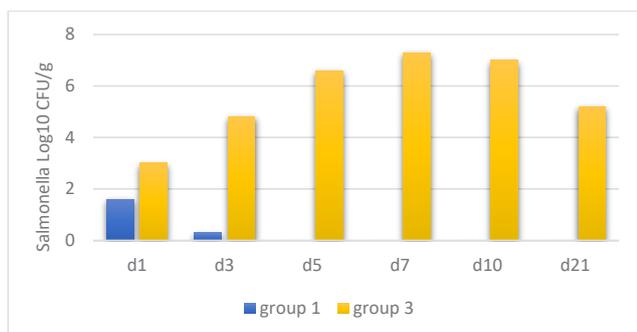


Fig. 3. Concentration of *S. Typhimurium* in the cecum of chickens post bacterial challenge.

Group 1-chickens treated with salmonella phage cocktail day before bacterial challenge, immediately after challenge and the next 8 days.
Group 3 – untreated chickens.

Discussion

According to the Center for Disease Control and Prevention (CDC) and the European Food Safety Authority (EFSA), *Salmonella* spp. are among the most important human pathogenic bacteria that frequently cause foodborne diseases worldwide. It was analyzed outbreaks reported to the United States' Foodborne Disease Outbreak Surveillance System from 1998 to 2012 in which the implicated food or ingredient could be assigned to one food category. Out of 1114 outbreaks, poultry was associated with 279 (25%), accounting for the highest number of outbreaks, illnesses and hospitalizations, and the second highest number of deaths. Out of the 149 poultry-associated outbreaks caused by a confirmed pathogen, *Salmonella enterica* (43%) was the most common pathogen [13, 14].

Antibiotic resistance is of great public health concern because the antibiotic-resistant bacteria associated with the animals may be pathogenic to human, easily transmitted to human via food chains, and widely disseminated in the environment via animal wastes. The routine employment of antibiotics, for prevention and growth promotion purposes in livestock farming, selects for antibiotic resistance among commensal and pathogenic bacteria. Owing to the fact that most of these antibiotics are not fully metabolized but released into the environment as waste products, antibiotic resistance has an ecological impact, since these waste products still have the potential to influence the bacteria population and promote antibiotic resistance [15].

Over the past 50 years, as poultry farms became larger and more concentrated, farmers began using



Fig. 4. Concentration of *S. Typhimurium* in the cecum of chickens.

Group 1-chickens treated with salmonella phage cocktail day before bacterial challenge, immediately after challenge and the next 8 days: Group 2-chickens treated with *Salmonella* phage cocktail 10 days post infection, Group 3 – infected but untreated chickens (positive control): group 4 (negative control, uninfected group).

antibiotics to prevent disease and speed growth in broiler chickens. The industry today remains dependent on their widespread use, and antibiotics are frequently given to birds that are not sick. When antibiotics are routinely given to entire flocks, resistant bacteria are likely to survive and proliferate. These resistant bacteria can even share resistance genes with other bacteria. Antibiotics misuse by poultry farmers has resulted in multidrug resistance and impeded efficiency of antibiotic treatments in the industry. The ability of pathogens to colonize in the gut increases after antibiotic administration because of a loss of resident microflora. (www.nrdc.org/sites/default/files/poultry-industry-antibiotic-stewardship-IB.pdf).

Salmonella spp. is one of the most common microbial contaminants in the poultry industry. Due to the common foodborne illness cases caused by *Salmonella*, prevention of *Salmonella* colonization in the gastrointestinal tract (GIT) of chickens is necessary. There is great potential for the use of phages as natural antibacterial remedy to control food pathogens at the pre- and postharvest stages of production [15-17].

Reduction of pathogen colonization in animals during primary production (*phage therapy*) is a strategy followed in primary production just before slaughter or during animal growth to reduce the probability of cross-contamination with the animal feces during food processing [18].

Previous studies have shown that phages significantly reduce the colonization level of *Salmonella* spp. in the avian gut. The results of our experiment

are in approximate agreement with other experimental studies [19-25]. Bardina et al. have investigated that frequent treatment of the chicken with bacteriophages, and especially prior to colonization of the intestinal tract by *Salmonella*, is required to achieve effective bacterial reduction over time. The best results, defined as a reduction of *Salmonella* concentration in the chicken cecum, were obtained when the bacteriophage cocktail was administered 1 day before or just after bacterial infection and then again on different days post-infection [26]. The study of Nabil N. et al. has shown that the effectiveness of bacteriophage treatments on *Salmonella* colonization in cecum of infected chicks was increased after five doses of phage treatment. At day 3 post-infection (dpi), cecal contents showed a marginal decrease in *Salmonella* loads with more reduction at 5 dpi. From 7 dpi to the end of the experiment at 15 dpi, all the chicks were cleared for *Salmonella* [27].

Our study has demonstrated that bacteriophages can be used for reduction and elimination of the cecal colonization of *Salmonella* in poultry. In this study, we have demonstrated that administration of salmonella phage cocktail to chickens before oral *Salmonella* infections followed by 9 consecutive phage treatments after the bacterial challenge as well as treatment with the same doses of phage cocktail (10 days post infection) were very effective. Further research should be done to investigate the possible use of our phage cocktail for prevention of salmonella colonization in farm level as our study showed that 1 day phage treatment before experimental infection decreased bacterial load from \log_{10} 3.1 (control group) to \log_{10} 1.6, no bacteria was detected in chickens of both groups on day 7 post-infection and all chickens were free of salmonella during 10 days after last administration of phage cocktail (21 day post-infection). This indicated that treatment with phage cocktail leads to complete elimination of *salmonella* contamination.

Conclusion

Our results demonstrate efficacy of used phage preparation in reducing *Salmonella* colonization in chickens and possibility of using this preparation as alternative to antibiotics for the reduction of *Salmonella* infection in poultry.

Acknowledgement

This work is funded by Shota Rustaveli National Science Foundation of (Project # FR17_293).

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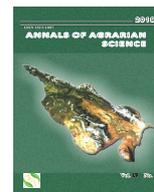
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Annals of Agrarian Science

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Dangerous regions of blizzard in georgia

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Received: 12 May 2019; accepted: 27 July 2019

ABSTRACT

Blizzards will do essential damage to the mountain and alpine regions of Georgia. Blizzards especially damages agriculture, is caused the uneven redistribution of snow and are created the bare sections of the pastures, which cause the freezing of winter cultures and the erosion of soil. In the regions, where winter pastures are arranged, blizzards frequently disrupts the normal mode of the feeding of animals and hampers wintering cattle. In this study the observational data of 85 meteorological stations and posts during the period of 1966-2017 are used. Most reliable are data during the period of 1966-1992. In 1993 the majority of alpine meteorological stations were closed; therefore period 1993-2017 was illuminated only by data of separate stations. The materials of the archive of the institute of hydrometeorology of Georgian technical university, data National Environmental Agency, literature data and climatic reference books also were used. Blizzards dangerous regions of Georgia are revealed and the corresponding map of division into districts is built. The dependence of a change in the blizzards on the height of locality is investigated.

Keywords: Mountain and alpine regions, Dangerous meteorological phenomena, Blizzard, Observational data, Agriculture, Meteorological station.

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Introduction

Blizzard is the transfer of snow by the wind of a sufficient force. Blizzard disrupts the uniformity of snow cover and causes the appearance of cornices, snowdrifts and other unstable forms of snow-accumulation [1]. Blizzards prevents the functioning of all forms of transport, is caused the destruction of the lines of communications and power transmissions, it creates emergency situations and frequently causes human victims.

Blizzards especially damages agriculture. High winds and friable structure of snow cover cause the uneven redistribution of snow and create the bare sections of the pastures, which cause the freezing of winter cultures and the erosion of soil. In the regions, where winter pastures are arranged, blizzards

frequently disrupts the normal mode of the feeding of animals and hampers wintering cattle [2].

The direction of blizzards dangerous winds is caused by the atmospheric processes, during which occurs the formation of blizzards. For the conditions of Georgia the winds with western and eastern components are such flows, although the relief introduces some correctives. The significant part of the blizzards is caused by the western processes, which cover almost entire territory of Georgia. In western Georgia they are accompanied by snow-falls, in the remaining regions surface blizzards can be formed. Wind speed upon the eastern intrusions is substantial less which respectively decreases snow-storm danger. During the eastern processes the blizzards covers entire eastern Georgia and significant part of the southern Georgian upland, in the

rare case it can be extended in the alpine regions of western Georgia.

The nature of blizzards is caused by the set of the conditions: a quantity of solid precipitation, speed and wind direction, the changeability of weather, the configuration of relief and the connected with it protection of relief. The intensity of blizzards to a considerable degree depends on speed of wind, stability of snow cover, dimensions of snowy particles, temperature and humidity of air [3].

Studies of blizzards in Georgia were begun in the 70-years of past century [4, 5], whereas in recent years with our participation were executed several articles on the base of contemporary data [2, 3, 6]. As a result were established laws governing allocation of frequencies, duration and intensity of blizzards for the territory of Georgia, the basic regions and periods of their propagation, meteorological regime of blizzard [3, 5].

Present article is the logical continuation of these studies and sets development and characteristic blizzards dangerous regions of Georgia as a goal in order to grant the data about the territorial distribution, the intensity, the repetition, the duration and the direction of snow-transfer to the interested persons from the state and the quotient of sectors.

Materials and methods

In this study observational data of 85 meteorological stations and posts during period 1966-2017 are used. Most reliable data are during period 1966-

1992. In 1993 the majority of alpine meteorological stations were closed; therefore period 1993-2017 was illuminated only by data of separate stations.

The materials of the archive of Institute of hydro-meteorology of Georgian technical university, data of the National Environmental Agency, literature data and climatic reference books were used also [4, 7, 8].

In a study the approved methods of climatological data processing, and also the methods of the mathematical statistics and probability theory are used.

Results and discussion

Table 1 depicts the average and maximum values of the number of blizzards days in different regions of the country with the indication of the height of the position of points.

It should be noted that in 90 years of past century the network of meteorological stations, which previously counted about 300 meteorological stations and posts, were practically destroyed. Thus, observations of different meteorological phenomena after this period, including of observation of blizzards, were conducted only at several stations, that, of course, it does not give the complete picture of the distribution of the number of days with the blizzards. However, the statistics from 1966 through 1992 is complete and can be examined in the form of united continuous network, which in the totality makes it possible for us to analyze the number of days with the blizzards in Georgia in the last 50 years.

Table 1 . Average annual and maximum number of days with the blizzards in the regions of Georgia (1966-1917)

Samegrelo-Zemo Svaneti				
Location	Average	Max	Region Center	Height above s.l., m
Местиа	2.1	16	Mestia	1500
Racha-Lechkhumi, Kvemo Svaneti				
Zeskho	12.3	27	Lentekhi	1800
Mamisoni Pass	146.3	235	Oni	2854
Мцхета-Мтиანети				
Kazbegi alpine	129	346	Stepantsminda	3665
Kazbegi	17.8	86	Stepantsminda	1750
Cross Pass	39	70	Stepantsminda	2380
Imereti				
Korbouli	13.7	33	Sachkhere	790

Mta-Sabueti	29.5	105	Kharagauli	1248
Kakheti				
Tsvi-Tura	8.3	68	Sagarejo	1990
Shida Kartli				
Ermani	28.9	64	Java	2220
Samtskhe-Javakheti				
Tskhratskaro	90.4	163	Borjomi	2462
Paravani	42.9	117	Ninotsminda	2073
Aragvi	17.3	74	Akhalkalaki	1669
Efremovka	24.9	90	Ninotsminda	2110
Guria				
Bakhmaro	27.46	135	Chokhatauri	1926
Kvemo Kartli				
Manglisi	7.2	36	Tetritskaro	1200
Abkhazeti				
Gagra Pass	29	73	Gagra	2432
Adjara				
Goderdzi Pass	78.8	162	Khulo	2025

It follows from Table, that the distribution of blizzards in the regions of Georgia has heterogeneous nature. From a quantitative point of view, taking into account the average and maximum num-

bers of days with blizzards, the regions of Georgia can be divided into 3 groups: less blizzards dangerous, blizzards dangerous and especially blizzards dangerous regions (Fig. 1).



Fig. 1. Blizzards dangerous regions of Georgia

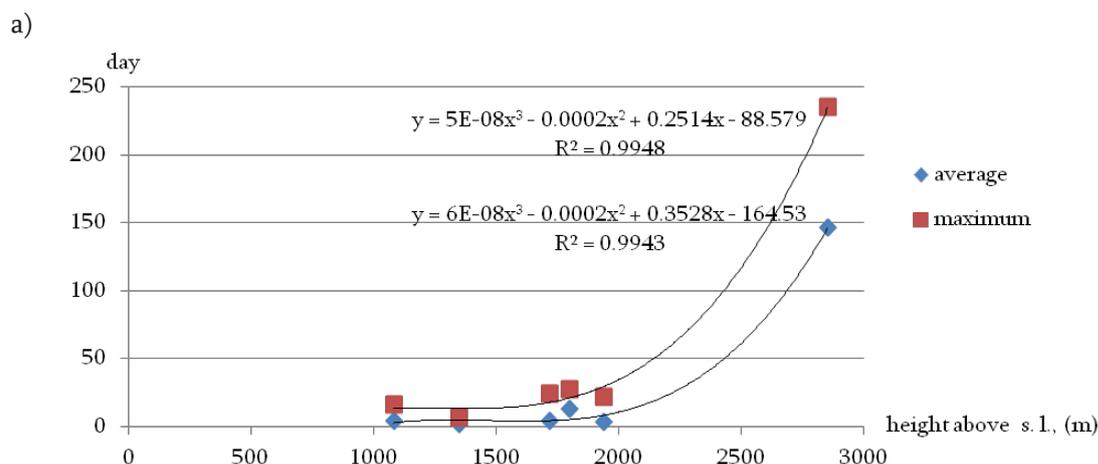
It is evident from Fig.1. that the Samegrelo-Zemo Svaneti region including alpine settlements of region, are less dangerous under the conditions of blizzards. The majority of blizzard is observed here, in the Mestia municipality, where their average annual number does not exceed 2 days, but the maximum of is equal to 16 days. Other regions are also less than blizzards dangerous. In particular in Kakheti on the Gombori ridge the maximum number of days with the blizzards composes 68 in the year, although on the remaining part of the territory of Kakheti the blizzard is rare phenomenon. In Shida Kartli is separated the Java municipality, where the average annual number of days with the blizzards composes 29, and maximum - 64. In Kvemo Kartli the blizzard is rare phenomenon, in Manglisi the average annual number of days with the blizzards composes only 7, and maximum - 36 days. Abkhazeti as a whole also is not characterized by blizzards activity, although the Gagra ridge can be considered as the separately blizzards dangerous region, where the average and maximum number of days with the blizzards they are 29 and 73 days respectively.

Blizzards dangerous regions include Imereti, Guria and Adjara. High mountain region of Imereti is characterized by blizzards activity, in particular the Sachkhere and Kharagauli regions. In Korbouli (Sachkhere municipality) at the height of 790 m the maximum number of days with the blizzards composes 33. In Mta-Sabuyeti (Kharagauli municipality) in the course of year is fixed 105 days with the blizzards. Frequent blizzards are noted also in the high mountain region of Guria. For example, into Bakhmaro (Chokhatauri municipality) the maximum number of days with the blizzards in the year reaches 135. Also, is separated blizzards activity in the mountain regions of Adjara. On the Goderdzi Pass (Khulo municipality) the average annual num-

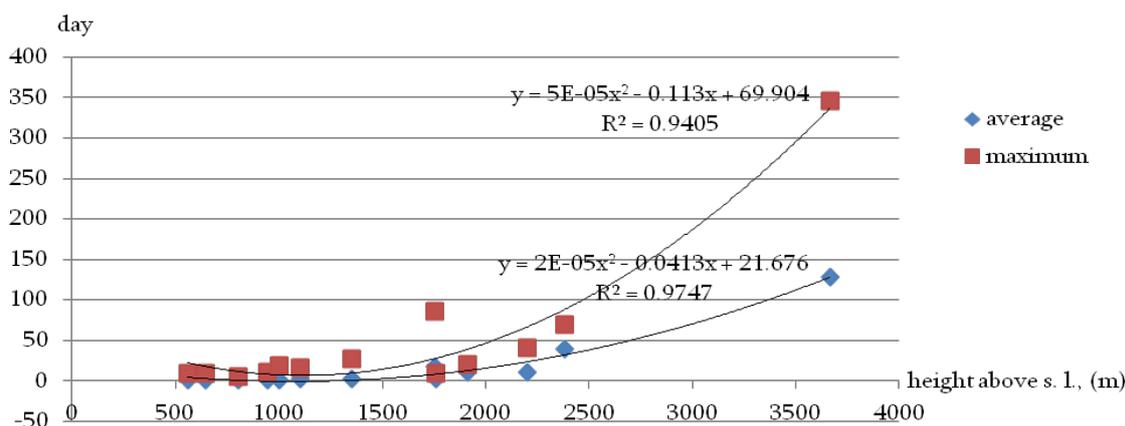
ber of days with the blizzards composes 80, and maximum reaches 162 days. On 11 November of 2015 on the Goderdzi Pass the lifesavers from the hearth of snow they took out 7 automobiles as a result of which they saved 30 people.

By especially blizzards dangerous regions on the territory of Georgia are Racha-Lechkhumi, Mtskheta-Mtianeti and Samskhe-Javakheti. Especially separated are Lentekhi and Oni municipalities in Racha-Lechkhumi region. On the Mamisoni Pass the average annual number of days with the blizzards composes 145, and the maximum number of days reaches 235. In Mtskheta-Mtianeti by blizzards activity is separated the Stepantsminda municipality, in particular Djvari Pass and Kazbegi alpine. In the latter of them the average annual number of days with the blizzards composes 145, and maximum - 340. Complex situation occurred on 4 January of 2016 years, when after closing of Georgian Military Road in the section of Kobi-Gudauri, about 150 machines remained with the blizzards and the 17 degree frost. The most blizzards dangerous region is Samtskhe-Javakheti. By blizzards dangerous activity it is especially separated the Javakheti upland, Borjomi, Ninotsminda and Akhalkalaki municipality. Here the average annual number of days with the blizzards exceeds 100 days. For example, into Tskhratskaro the average annual number of days with the blizzards composes 160. 5 and on 6 January 2015 years the lifesavers saved from the blizzards approximately 200 people, unfortunately one person passed away.

A significant effect on formation and development of blizzard has orography, in particular the height of locality. In Fig. 2 are represented the dependences of a change in the number of days with the blizzards on the height of locality in the separately blizzards dangerous regions, and also their approximating polynomials and coefficients of determination (R^2).



b)



c)

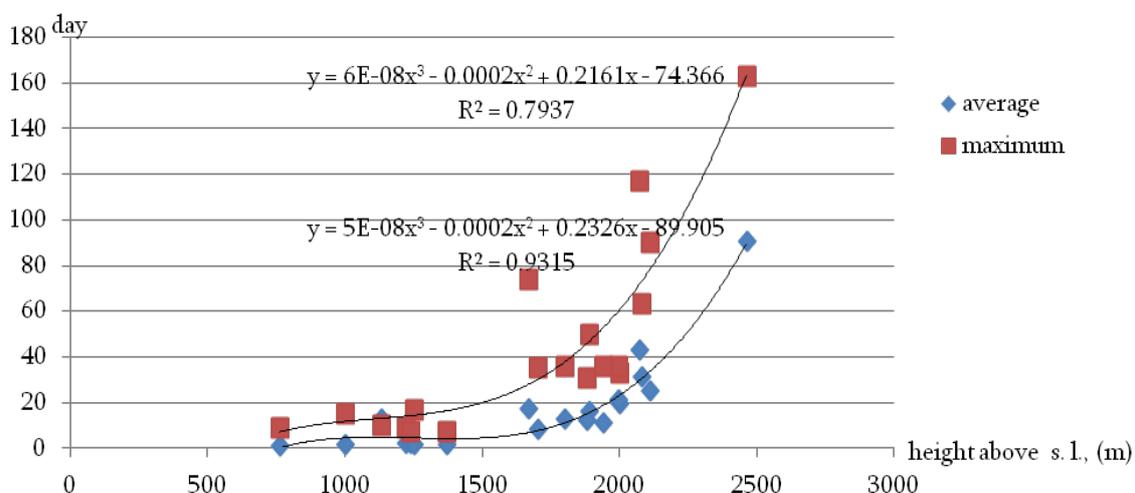


Fig. 2. Dependence of a change in the average and maximum number of days with the blizzards on the height of the locality: a) Racha-Lechkhumi, b) Mtskheta-Mtianeti, c) Samtskhe-Javakheti

It follows from Fig.2 that with an increase in altitude of locality increases the number of days with blizzards, also, in the alpine zone, at the heights more than 2000 m, in Kazbegi (3665 m) and on Mamisoni Pass (2850 m), blizzards are noted almost throughout year.

The represented polynomials make it possible to estimate the annual number of days with the blizzards in the dependence on the height of locality. y - annual number of days with blizzards, x - height above sea level. Equations completely satisfactorily describe a change of the number of days with the blizzards in the dependence on the height of locality. According to the coefficients of determination the contribution of the height of locality in a change in the number of days with the blizzards in Racha-Lechkhumi and Mtskheta-Mtianeti is especially high and composes 94-100%. Coefficient of determination is somewhat less in the region of Samtskhe-Javakheti, which in all probability,

is caused by the variety of relief and topographical-climatic conditions.

It should be noted that the number of days with blizzards depends not only on the height of locality, but also from other factors, including from the topographical-climatic conditions, the special features of micro relief and so forth [6]. The aforesaid data of table 1 confirm for example, in Korbouli at the height of 800 m the average annual number of days with the blizzards composes 14, and into Zeskho, at the height of 1800 m the number of days with the blizzards is less 12 days.

Conclusion and recommendations

The majorities regions of Georgia are not blizzards dangerous.

Blizzards are dangerous in Racha-Lechkhumi, Mtskheta-Mtianeti and Samtskhe-Javakheti. In these regions the alpine regions, where the blizzards

are noted throughout year, are separately blizzards dangerous.

As is known, mountain landscape occupies the significant part of the territory of Georgia; therefore the development of mountain regions has vitally important value for our country. For the development of different segments of the economy, including agrarian sector, in the mountain and alpine regions of Georgia, it is important to in proper time liquidate damage substituted by different dangerous meteorological phenomena, including by blizzards, to take all possible precautionary measures in order to minimize damage from the blizzards and to create safe medium in this respect. Also is important conducting effective measures for the purpose of control of blizzards for the uninterrupted functioning of the automobile, railroad and aviation transport junction of Georgia, which first of all provides for the installation of meteorological stations in the blizzards dangerous regions, in particular in the crossing sections, which will ensure observations of the blizzards and other dangerous meteorological phenomena in the continuous regime, thanks to which will become possible with the high accuracy in forecasting of the areas of blizzards and other phenomena, and conducting preventive measures.

Acknowledgment

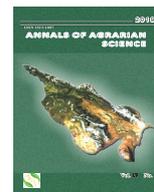
This work was supported by Shota Rustaveli National Science Foundation of Georgia (SRNSFG) [Grant number YS17-49].

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Studying residual pesticides in agricultural lands of the republic of armenia

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Received: 12 February 2019; accepted: 15 May 2019

ABSTRACT

In Armenia like other former Soviet states synthetic and particularly organochlorine pesticides were first introduced in the mid XX century and remained in wide use for almost 20 years, up to the 1970s. After the USSR disintegration numerous pesticide storage facilities throughout Armenia were left unmanaged and presently semi-collapsed and abandoned repositories are found in almost all marzes (provinces) of the country. However, no complex, in-depth researches of residual pesticides were ever done in the country, isolated studies had a limited character. The given research is a long-term one and is aimed at detection of residual pesticides in Armenia's agricultural lands and assessment of residual pesticide contamination in system 'agricultural soil-irrigation water-fruits and vegetables'. To assure and control the quality of field and lab works the CENS staff have developed respective SOPs (standard operating procedures) and QA/QC (quality assurance and quality control) plans. Chromatographic analyses were done by a gas chromatograph-mass spectrometer (GC-MS) Trace DSQ (Thermo Electron Corporation, USA). This paper considers a part of initial research results obtained for Armenia's 5 out of 10 marzes (provinces) in 2014 and 2015. Data generated from this research have indicated that in all the five studied marzes agricultural soils and those of abandoned pesticide storage sites are contaminated with residual DDT which exceeds national MAC (maximum acceptable concentration) by several tens of times. None of banned residual pesticides were detected in irrigation water and fruit and vegetable samples. Presently, the research is underway.

Keywords: Organochlorine pesticides, DDT (sum), Agricultural land, Pesticide contamination, Pesticide storage facility, Banned pesticides.

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Introduction

It is known that all organochlorine pesticides decay very slowly and hence remain in the environment for a long time. It is also known that due to good migration [1] and bioaccumulation properties pesticides travel into different environmental compartments vis. soils [2, 3], water [4, 5], vegetation [2, 3, 6] and penetrating into food chain ultimately affect human health [4, 3, 7]. In 2001 the Stockholm Convention [8] included those pesticides in the list of persistent organic pollutants to the environment, thus banning both their production and application in agriculture. In 2003 Armenia, too, joined the Convention.

The Republic of Armenia (latitude 38°50' - 41°18' N, longitude 43°27' - 46°37' E) is a landlocked high-land country located in the South Caucasus. A major portion of its total area (29 743 sq.km) belongs to agricultural lands (68.8 %) [9, 10]. In Armenia like other former Soviet states synthetic emphasizing organochlorine and organophosphate pesticides were first introduced in the mid XX century and remained in wide use for almost 20 years, up to the 1970s [11]. Since the 1990s, after the USSR disintegration numerous pesticide storage facilities throughout Armenia have remained unmanaged and presently semi-collapsed and abandoned repositories are found in almost all marzes (provinces) of the country [12]. The fact that pesticide storage fas

cilities are potential sources of environmental pollution is demonstrated particularly by recent researches [13, 14]. It should be stressed that though the issue is topical to Armenia, nonetheless little pesticide research has been done in the country since the 1990s. Moreover, the studies had a limited character and did not include in-depth and complex monitoring investigations particularly in system “agricultural soils-irrigation water-fruits and vegetables”. It should be mentioned that GAP (Good Agricultural Practice), has not been introduced in Armenia so far and that no data are available regarding pesticide contamination of agricultural crops. This particular research was implemented in the frames of a “Monitoring of residual pesticides in food produced in the Republic of Armenia, 2014-2018” Program designed to fill in the mentioned gaps and covering all the 10 marzes of Armenia. The research goal was detecting residual pesticides in Armenia’s agricultural lands and assessing pesticide contamination in system “agricultural soils-irrigation water-fruits and vegetables”.

Material and methods

The research covered 13 settlements located in 5 out of 10 marzes.

Selection of plots of agricultural land was done based on the size and productivity. The studied materials were samples of agricultural soils, irrigation waters, fruits and vegetables.

The research included implementation of both field and lab works in compliance with SOPs and QA/QC plans developed by the CENS staff consistent with national and international standards, methods and directives (Tab. 1, 2). Sampling was done in 2014 and 2015. Specimens of fruits (peach, plum, apple, fig, persimmon, pomegranate, pear, apricot, quince) and vegetables (potato, pepper, cabbage, beetroot) were gathered in the harvesting period from relatively large plots of agricultural land-orchards and fields; samples of soil were taken from the same orchards and fields; water was sampled from local irrigation canals. One should stress that when sampling soil, special attention was given to former pesticide storage sites.

Table 1. Standards, orders, guidance and directives used in SOPs and QA/QC plans

Kind of samples	Standards	Order	Guidance	Directives
Agricultural soils	ISO 10381-1:2002 ISO 10381-2:2002 ISO 103081-4:2003	Order № 01-Ն of the Minister of Health RA as of January 25, 2010 “Hygienic requirements to soil quality” № 2.1.7.003–10: About approval of sanitary regulations and norms”	US EPA. Field sampling guidance document #1205. Soil sampling.	-
Irrigation water	ISO 5667-1:2006 ISO 5667-2:1991	-	-	US EPA OSWER Directive #93240.0-05
Fruits and vegetables	ISO 874:1980	-	CAC/GL 41-1993 CAC/GL 33-1999	Commission Directive 2002/63/EC

Wholly, over the studied period 152 fruit and vegetable, 146 soils, 22 water samples were collected. Additionally, 6 soil samples were taken from some of former pesticide storage sites found in four out of five studied marzes.

Lab analyses were carried out at the Central Analytical Laboratory CENS accredited by ISO IEC 17025. Prior to extraction all the samples underwent pre-treatment. Extraction of the fruit, vegetable, agricultural soil and irrigation water samples was done to indicate the presence of 17 forbidden pesticides [15]: aldrin, DDT (sum), dieldrin, 1,2-dichloroethane, endrin, captafol, heptachlor, HCH (sum), methyl parathion, methamidophos, mirex, parathion, pentachlorophenol (PCP), toxaphene, chlordane, chlordecone, phosphamidon. All the reagents and solvents we used were of analytical grade and included twenty (20) different pesticide standards ordered from AccuStandard (New Haven, USA). Working standard solutions were made by dilution of the stock standards and mixtures of standards of different concentrations and were used mostly for the screening of the pesticide residues and metabolites. The volumetric glassware was teflon stoppered. Chromatographic analyses were done by a gas chromatograph-mass spectrometer (GC-MS) Trace DSQ (Thermo Electron Corporation, USA) (Tab. 2).

After thorough analyses the obtained data were collated with national MACs for soil (0.1 mg/kg) [16] and then compiled into a respective database.

Results and discussion

None of 17 banned pesticides (aldrin, DDT (sum), dieldrin, 1,2-dichloroethane, endrin, captafol, heptachlor, HCH (sum), methyl parathion, methamidophos, mirex, parathion, pentachlorophenol (PCP), toxaphene, chlordane, chlordecone, phosphamidon) were detected in sampled irrigation waters, fruits and vegetables. In 13.6% of agricultural soil samples we detected residual DDT only, the contents of which widely varied 0.0028 to 7.32 mg/kg (Tab.3).

DDT was not found in agricultural soil samples collected from settlements of Azatan (Shirak marz), Deghdzavan (Tavush marz), Meghri, Karchevan, Agarak (Syunik marz). The largest share of contaminated samples (54.5%) belongs to Armaviri mars, where DDT concentrations exceeded MAC (0.1 mg/kg) by 1.5-73.2 times (Tab.3). It is noteworthy that DDT and its metabolites were found out mainly in soil sampled from orchards except one sample collected from potato field in village of Akhurian. In this respect one should note that according to Harris M. et.al [17], pesticides remain in orchard soils for a longer time as compared to field soils. The impact of physical factors on orchard soils (soil treatment, direct sun rays, extensive moisture loss, erosion) is limited, this contributing to bioaccumulation properties of persistent pesticides.

Table 2. Standards and methods of analyses of agricultural soil, irrigation water, fruit and vegetable samples

Kind of samples	Standards	Methods
Agricultural soils	AST ISO 10382-2005 ISO 11465:1993 ISO 14507:2003 US EPA 3546	microwave extraction (MWE), GC-MS
Irrigation water	AST ISO 6468-2005 US EPA 608 US EPA 625	Liquid-Liquid Extraction (LLE), GC-MS
Fruits and vegetables	AST EN 12393-2-2011 AST EN 12393-3-2011 GOST R 30349-1996 GOST R 30710-2001	GC-MS

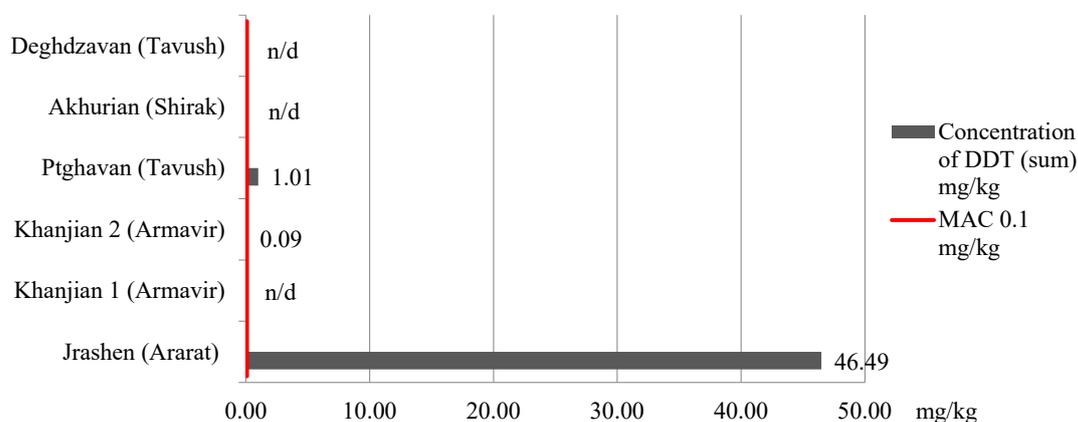
Table 2. Concentrations of DDT (sum) in agricultural soils of 5 studied marzes

n/n	Marzes	Studied settlements	The total amount of samples per settlement	The amount of contaminated samples	Share of contaminated samples, %	DDT range, mg/kg
1	Ararat	Qaghtsrashen	36	3	8.33 %	0.0028 - 4.165
		Narek	10	2	20 %	0.013 - 0.778
		Jrashen	10	4	40 %	0.44 - 5.36
2	Armavir	Khanjian	13	6	46.15 %	0.15 - 7.32
3	Shirak	Akhurian	12	1	8.3 %	0.424
		Azatan	14	n/d*	n/d	n/d
4	Tavush	Haghtanak	19	1	5.26 %	0.103
		Ptghavan	12	2	16.67 %	0.29 - 0.483
		Deghdzavan	9	n/d	n/d	n/d
5	Syunik	Syunik rural community	5	1	20 %	0.654
		Meghri	9	n/d	n/d	n/d
		Karchevan	2	n/d	n/d	n/d
		Agarak	1	n/d	n/d	n/d
TOTAL		13	152	20	13.6 %	n/d - 7.32

Note: *- not detected

DDT was detected only in three soil samples collected from pesticide storage facilities found in the four out of five studied marzes. Its concentrations varied rather widely: between 0.09 and 46.49 mg/kg. The highest concentration 46.49 mg/kg which exceeded national MAC by 469.9 times, was established in one soil sample taken from an abandoned pesticide storage facility in Ararati marz (Jrashen village). The lowest contents of DDT (sum) 0.09 mg/kg which did not overstep national MAC was

detected in one of two former pesticide storage sites in Armavir marz (Khanjian village). DDT detected in one soil sample from Ptghavan village (Tavush marz) exceeded national MAC by 10.1 times. No residual DDT was detected in soil samples collected from Akhurian and Deghdzavan villages located respectively in Shirak and Tavush marzes and one out of two former pesticide storage sites in Khanjian village (Armavir marz) (Fig.).

**Fig.** Concentrations of DDT (sum) in soil samples collected from 6 former pesticide storage facilities

Conclusion

The research results for 2014-2015 support a conclusion that irrigation water and fruit and vegetable samples collected from five studied marzes selected for this research are not contaminated with banned residual pesticides. Agricultural soil samples are contaminated with residual DDT only. In some soil samples DDT concentrations exceed national MAC by several times. Residual DDT which exceeds national MAC by several tens of times is detected in three marzes: Ararati, Armaviri, Tavushi. Presently the research is underway.

Acknowledgment

The research results considered in this paper were obtained in the frames of a target Program “Monitoring of residual pesticides in food produced in the Republic of Armenia, 2014-2018” funded by the State Committee for Science to the Ministry of Education and Science RA.

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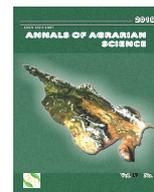
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Annals of Agrarian Science

Journal homepage: <http://journals-org.ge/index.php>



Water economy balance - the basis of water supply assessment

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Received: 15 March, 2019; accepted: 22 June, 2019

ABSTRACT

Georgia has reach water resources, however, their unequal distribution creates a gap between the amounts of available and consumed water. The main goal of the study is to identify most vulnerable regions as regards water resources on a basis of Water Economy Balance. Kvemo Kartli has been taken as the region under study. Its area is 6.5 thousands of square km (that is 9.3% of the country territory). There are 347 settlements – 7 cities, 6 towns and 334 villages. Its natural conditions are most favorable for agricultural purposes with 2-3 harvests per year that stipulate for high competitiveness of the region in comparison with other ones. Different branches of industry such as mining, metallurgy, chemical production of cement and construction materials, ceramics, glass, etc., along with power generation plants are the most active water consumers in the region. The main problem of nearly all municipalities in the region are irrigation schemes which are depreciated or in poor state today. The existing problems of water supply hinder development of agricultural branches considerably. The hydrography network of Kvemo Kartli is represented with trans-boundary river Mtkvari and its tributaries; 15 lakes are used for recreation, irrigation and fishing purposes; 6 reservoirs are used for fresh water supply, power and irrigation purposes. There are mineral, sulphur and thermal springs.

Keywords: Water resources, Management, Water economy balance, Deter-ration, Climate change, Hydrography network.

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Introduction

Georgia has reach water resources, however, their unequal distribution creates a gap between the amounts of available and consumed water. In east Georgia, which is considered to be the main water consumer part of Georgia, water consumption rates are four times lower compared to the West. In West Georgia, South and South-East parts are suffering from the most severe conditions. From main adverse factors influencing on water resources of Georgia are: the increasing pollution of hydrosphere and environment (atmosphere, lithosphere). The causes of qualitative deterioration of water resources are: irrigation, land-reclamation of salinized lands, run-off waters, malorganization of reservoirs' foundation pits, et cetera. The importance of the problem is proved by the fact that even purified, recycled waters need dilution 15 times with clean water to regain natural water quality [1]. Here,

river water flows are often lower than the environmental minimum during low water level periods. Considering the processes of climate change, we predict that freshwater availability issue will become more significant in the future. In large parts of eastern Georgia annual precipitation sum decreased at a rate of 1e3% per decade. Most precipitation decreasing decadal trend is observed in Kvemo Kartli region, south of Tbilisi, and is more than 5% [2]. Nowadays, in Georgia water resources are managed by an administrative principle. The administrative model cannot ensure efficient water management, considering the needs of water consumers' as well as environmental protection concerns. Therefore, there is a necessity for the shift to pound manageable water consumption [3, 4]. At the same time, Kvemo Kartli's administrative-territorial division model excellently illustrates the specificities of the region's natural and socio-economic conditions. The hydrography network of Kvemo Kartli is rep-

resented with trans-boundary river Mtkvari and its tributaries. Tbilisi – the capital of country is located next to Kvemo Kartli region. The nearby Azerbaijan and Armenia republics, Tbilisi international airport, high level of urbanization, transport and power corridors, etc., favor development of the region.

Georgia strives for deeper cooperation with Euro Union. Among the priority spheres of that cooperation one of the most important ones is sustainable development of water resources. Inculcation of controlled water consumption system will create better conditions for the rational and more efficient use of the budget expenditures that will have positive effect on economics and social sphere of the country.

Methods and Materials

Kvemo Kartli was choose as a study area. Agriculture is the central area of Kvemo Kartli's economics. 29% of the total population and 48% of the countryside population are employed/self-employed in agriculture. Compared to the other regions, there is an up-growing Natural Population Growth trend in Kvemo Kartli. Considering this, it is likely that in a longterm perspective, there will be a higher demand for social infrastructure, utilities, and employment. Hence, there will be a greater need for water resources.

The main goal of the study is to identify most vulnerable regions as regards water resources on a basis of Water Economy Balance. This is essential for long-term and short-term planning.

Kvemo Kartli is located in the Southeastern part of Georgia. It is surrounded by Samtskhe-Javakheti on the west; surroundings of Tbilisi, Shida Kartli and Mtskheta-Mtianeti on the north; Kakheti on the east; and the republics of Armenia and Azerbaijan on the south.

In the region, 45% of the population is ethnically Georgian; 45% - Azeri; 6% - Armenian; 4% - Abkhazian, Osetian, Russian, Greek, Ukrainian, Kurd. Self-governing units of the region are the Municipalities of Bolnisi, Gardabani, Dmanisi, Tetri Tsqaro, Marneuli and Tsalka, and the city Rustavi.

Climate is moderately humid in Kvemo Kartli. Towards east, humidity decreases. Winter is moderate cold, summer – hot. In the region, northern, north-western and south-eastern winds dominate. Average minimal temperature is not lower than 0,2°C. Average maximal temperature usually occurs in August and amounts to 23.9°C. The average sum of annual precipitate is 146 mm. The most precipitated month is May, the driest – January.

Compared to the other regions of Georgia, the volumes of local mineral and thermal waters are lower. Among them, the most significant rivers are the transborder river Mtkvari and its tributaries. Existing water resources are efficiently consumed in the region. Water Supply Data of Natural Water Objects (underground and surface) from 2011 year shows that water supply amounts to 452 million m³, from which 2% was used for drinking-agricultural means, 30% - for hydro energetic means, 55% for the industrial means, 11.7% for irrigation, 0.7% for agricultural water supply, and the rest - for pond economics and recreational means.

Climate conditions are convenient for producing agricultural products in the region. Harvesting is conceivable 2-3 times a year, which makes this region highly competitive (outstands the region) to the other regions of Georgia. There were no major changes in the structure of agricultural areas of the region during the past 5-7 years. However, there is an upward trend in the number of the farmers occupied in animal breeding and a decrease of the interest in seeding. The dysfunction of the irrigation system is a basic obstruction for land cultivation.

Irrigation system area amounts to 46 754 ha, which is 50% of the local arable land. Out of it, 20 274 ha (43%) requires rehabilitation. There are identical conditions in inner networks of irrigation systems since they are absorbed and broken. Water supply from water object surfaces are prevailing. At the time of stil and temperature inversion, cold air masses accumulate. The average minimum air temperature is not below 0,2⁰ C, the average maximum temperature in August reaches 23.9⁰ C. The average annual precipitation does not exceed 550 mm. The most precipitation is May, and dry – January.

Territorial water supplement is usually assessed using the structure of Quantitative Water Economy Balance (QWEB). The Water Economy Balance of a river basin, sub basin, or every part or economical region is the balance between water resources and water supply in certain time trims. Water economy balance (WEB) can be employed for multi-year goals as well as for certain periods (such as arid-climate year, vegetative season, etc.). As a rule, maximum of water-consumption indices are taken into account for developing water economy balance.

While developing Water Economy Balance, sequentially along the river (from the outfall to the estuary), Water Economy Units (WEU) are allocated. WEU is the part of the territory and its parameters are used to assess water intake limits of the water

object, Water Supply, and different water consumption parameters. For each water Economy Units, parameters of the structure of water Economy balance are developed. Based on them, status for each can be generated using the following classification:

- WEB can be secured by local flow;
- WEB includes local flows as well as flows that are transmitted from the upper side of the Unit's adjacent territories;
- WEB consists of local water, water transmitted from the upper side of the Unit's adjacent territories and returned water;
- Besides mentioned water resources, environmental minimum water also participates in WEB supplement.

In the mentioned classification, the first one is the most convenient for water supply, while the last one describes the most difficult conditions. Each WEB that includes returned or environmental minimum waters, face certain issues. Not to mention the last one, the water that is necessary for returned polluted water dilution might create severe problems related to water supply. Considering the aforementioned, Water Economy Balance also possesses a qualitative aspect.

The main goal of the study is to develop the Quantitative-Qualitative WEB for Kvemo Kartli using updated data and its analysis. The calculations are based on information from the 80s of the last century, which was extrapolated to 2011. It should be considered that during this time almost all fields of agriculture functioned normally in Georgia. The amount of the water necessary for dilution, was assessed using secondary data analysis (previous reports, projects, mentions, scientific works, etc.). In Kvemo Kartli saturation of Returned water in QQWEB was assessed using previous research findings of the surveys conducted in the region. Moreover, expert assessments were also considered by following scheme: Tsalka, Tetrtskaro (Basin of the River Khrami), Dmanisi, Bolnisi – 5 times more; Marneuli (Basin of the River Khrami) - 8-times more; Tetrtskaro (Basin of the River Alngeti), Marneuli (Basin of the River Algeti) - 10-times more.

Methodology for compiling Water Economy Balance has been carefully developed [5]. Its results are widely used in the regulation/planning of water relations. The general pattern QWMB is displayed by the following equation:

$$B_q = W_1 - W_i + W_r \geq G \quad (1)$$

In this formula, B_q represents Quantitative Water Economy Balance (flow given to neighbor/low

riverside Water Economy Section). W_1 represents a local flow, formed in the Water Economy Section. W_i refers to water in take, W_r - returned water, G – environmental minimum.

In general, conditions are tensed when water remaining in the river (transferred to the neighboring WEB) equals or is less than the environmental minimum. In this case, Water Economy Balance is considered to be in shortage.

Nowadays, the rule of computing environmental expenditure is not defined by Georgian law. Acknowledging largely admitted water expenditure computing methods, we presume that it would be advisable to use 25% of the river multi-year expenditure as the mean of the environmental expenditure in Kvemo Kartli, considering its natural resources.

In order to preserve water background quality, which is one of the main demands of EU Water Frame Directive, a minimum amount of the water is needed in the object which is a receiver of polluted water flows. This minimum amount is enough for water dilution to certain concentrations. Therefore, indices that show a quality of polluter water flow, is reduced to the quantitative indices as it considers the amount of the water needed for dilution.

To address freshwater-related issues caused by contemporary natural, demographical and economic conditions, it is necessary to modify Water Economy Balance. Precisely, it is important to involve the amount of the water needed for polluted water dilution. In other words, for complex assessment of territorial water supply, it is important to develop quantitative-qualitative water economy balance. According to the modified new approach (1), we should add to the display a new member that would count the amount of the water needed for the polluted water dilution.

Supplied returned water – $W_r = K_r * W_i$; Water needed for polluted water delution – $W_d = K_d * W_i$; K_d where K_r and K_d are return and delution amount coefficients. As a result, after simple transform, we can compute volume of the returned water at Water Economy Balance, needed for delution:

$$W_d = W_r * (K_d + 1) \quad (2)$$

Calculation of Quantitative-Qualitative Water Economy Balance is even more complex. This is due to the limited availability of observed data needed for computation. Because of that, pollution of received and returned waters are often ignored and they are taken as deemed and as appropriate to the established normative. Grin [5] and Lvovich [6] who are highly qualified and well know special-

ists in this area, also confirm that even clean flows should be diluted at least 5-10 times [5, 6]. At the same time, complex and costly activities are needed for consumed water dilution, that flows in in the water objects in order to be relevant for standards, rather than reducing its volume. In case high volume of water is needed for flown consumed water solution, it is recommended to reduce them.

After assessing the probity of compilers and existing data, as a rule, the number of compilers gets significantly reduced. Considering conditions of Kvemo Kartli, just 6-8 compilers can be enough for computing Quantitative-Qualitative WEB. Therefore, we develop a scheme of Quantitative-Qualitative WEB by sections:

$$\text{WEB } 1 - W_1 = W_0 + B_1 + A_1 + Q_1 - E_1 - D_1 - C_1 \pm \Delta V_1 \geq F_1 * (K_d + 1) \geq G_1,$$

$$\text{WEB } 2 - W_2 = W_1 + B_2 + A_2 + Q_2 - E_2 - D_2 - C_2 \pm \Delta V_2 \geq C_1 + C_2 * (K_d + 1) \geq G_2,$$

$$\text{WEB } 3 - W_3 = W_2 + B_3 + A_3 + Q_3 - E_3 - D_3 - C_3 \pm \Delta V_3 \geq C_1 + C_2 + C_3 * (K_d + 1) \geq G_3, \quad (3)$$

$$\text{WEB}_n - W_n = W_{n-1} + B_n + A_n + Q_n - E_n - D_n - C_n \pm \Delta V_n \geq \sum_{i=1}^n C_i * (K_d + 1) \geq G_n,$$

(3)

Were W_n is WEB in n cross,

B_n - Local flow;

Q_n - All types of returned water on the WEB,

A_n - Water flow in WEB, with territorial diversification systems of flows (between basins, inside basin),

$\pm \Delta V_n$ - Refilement/operation of pools, puddles and reservoirs on the WEB. During the computation period, it is defined as a corrective of available water resources that takes levels of incoming and outgoing parts of balance.

E_n - Water loss in the reservoirs due to water evaporation on the balance of WEU.

D_n - Withdrawal the parts of waterflow outside WEB

C_n - Sum of demand on water among WEB consumers

F_n - Water needed for returned water delution

K_d - Saturation coefficient of returned water delution

G_n - Environmental minimum

If the mentioned conditions are not fulfilled, WEB is considered to be under a shortage. The shortage can be addressed by:

- Transmitting of water release point to the cross of WEB which is better secured with water;
- Reducing water intake in crosses located

above WEB;

- Reducing water release in the river (water object);
- Building complex management water reservoirs and increasing water expenditure, considering it's beneficial volume during water release period.
- Concerning last activity, it is worth to mention that recent events (forest fires) perfectly illustrated the necessity for having small anti-fire reservoirs when the territory has a difficult relief. The reservoirs described above might combine anti-fire function as well.

We modified existing data related to water flows [7], which is a part of WEB return water, using contemporary, high-resolution digital elevation model of relief. We computed precise multi-year flows for ponds, sub basin, and municipalities.

Results

In order to better identify impacts of social-economic conditions, QQWEB was developed for municipalities and river basins (Table 1). Table 2 shows the vulnerability levels considering WEB and QQWEB. A vulnerability is defined as a difference between the Quantitative and Quantitative-Qualitative Water Economy Balances.

We assume that QQWEB structure analysis, expressed into different coefficients, is more feasible compared to absolute data (Table 2). The coefficients showing WEB balance are following: WEB coefficient, which is a ratio of WEB to an environmental minimum; Water Consumption Coefficient – a ratio of local flow to total water consumption; Potential Water Consumption Coefficient – a ratio of accessible flow to total water consumption.

The main part of Kvemo Kartli water resources is formed in the basin of the River Khrami, in the municipalities of Tsalka, Tetrtskaro, and Dmanisi. WEB structure in the basin of the River Khrami is satisfactory - withdrew and accessible flows are significantly higher than water consumption and environmental minimum. In each municipality, WEB coefficient is higher than 1 and variate between 2.2-3.7. In the municipalities of Tsalka, Tetrtskaro, and Dmanisi, WEB and its' Structure Coefficients are the most convenient. Only Marneli Municipality shows tensed condition.

Table 1. *Kvemo Kartli Quantitative-Qualitative Water Economy Balance, Million m³*

Municipality		Local Flow	Total waterintake	Returned Water	Total Flow	QWEB	Environmental Minimum	Consumed Water	Delution Water	QQWEB
River Khrami										
Tsalka		457	37.5	37.5	457	420	114	37.5	0	420
Tetrtskaro		142	34.5	34.5	599	532	150	29.8	23.5	508
Dmanisi		152	77.0	77.0	751	621	188	63.1	69.5	551
Bolnisi		87.1	65.4	65.4	838	652	210	55.9	47.5	604
Marneuli		103	240	240	941	523	235	232	59.2	463
River Algeti										
Tetrtskaro		111	61.5	61.5	111	49.5	27.8	61.5	0	49.5
Marneuli		16.7	48.8	48.8	128	27.7	31.9	38.5	103	-75.3

The flow in the River Algeti is mostly formed (>80%) in the territory of Tetrtskaro Mnicipality. In this part of the basin, WEB structure is satisfactory.

Table 1. *WEB Structure of Municipalities in Kvemo Kartli*

Municipality, River Basin	WEB Coefficient	Water Consumption Coefficient	Potential Water Consumption Coefficient
Tsalka - Khrami	3.7	12	12
Tetrtskaro – Khrami, Algeti	3.3	2.8	7.3
Dmanisi - Khrami	3.1	2.0	9.9
Bolnisi - Khrami	3.0	1.3	12
Marneuli – Khrami, Algeti	2.2	0.7	3.2

In Kvemo Kartli most tensed conditions are in the area where a minimum volume of local flow is 16.7 million m³. This is in the basin of the River Algeti in Marneuli Municipality. In this area, accessible water resources consist of the sum of flows and returned waters from Tetrtskaro Municipality. In the Municipality every WEB Structure Coefficients are minimal for the region. WEB coefficient lower than 1 exists only in this region, therefore,

here environmental minimum particulates in water consumption (Fig.).

Tsalka is the most well-ensured municipality of the region, while Bolnisi is the least ensured.

In the region, highest amount of water is needed in Marneuli Municipality, lowest – in Tsalka Municipality.

Nowadays, in the municipalities of Markenuli and Tetrtskaro, only 20%-30% of the total popu-

lation can be secured using local water resources. At the same time, the municipalities of Tsalka and Bolnisi have the capacity to supply 20 times more population (for city residents – 500 Liters per day; for village residents – 300 Liters per day).

Conclusion

Based on analysis of Kvemo Kartli Water Economy Balance, we can conclude the following:

Sum WEB of basins of the main rivers in Kvemo Kartli (Khrami, Algeti), is higher than the environmental minimum. Therefore, it is satisfactory.

WEB structure in the basin of the River Khrami is generally satisfactory – withdrawn and accessible flows are significantly higher than water consumption and environmental minimum. The same balance is shown considering delutin returned flows (8 times) and inveronmental flow (25% of norm). Tensed conditions are in Marneuli Municipality due to high volume of consumed water.

Areas of the River Algeti basin that are located in Tetrtskaro Municipality, WEB is satisfactory.

In terms of WEB, in Kvemo Kartli, most tensed conditions are in those areas of the River Algeti basin which is located in Marneuli Municipality. Here, we face a significant shortage in WEB.

Analysis of WEB by municipalities shows that in case of Marneuli Municipality there is a serious

problem which becomes even more visible when considering approximate spillway.

In parts the River Algeti basin, located in Marneuli Municipality, accessible water resources consist of the sum of withdrawn flows and returned water from Tetrtskaro Municipality.

Acknowledgments

The study was fulfilled with the support of the Shota Rustaveli National Scientific foundation (№216916).

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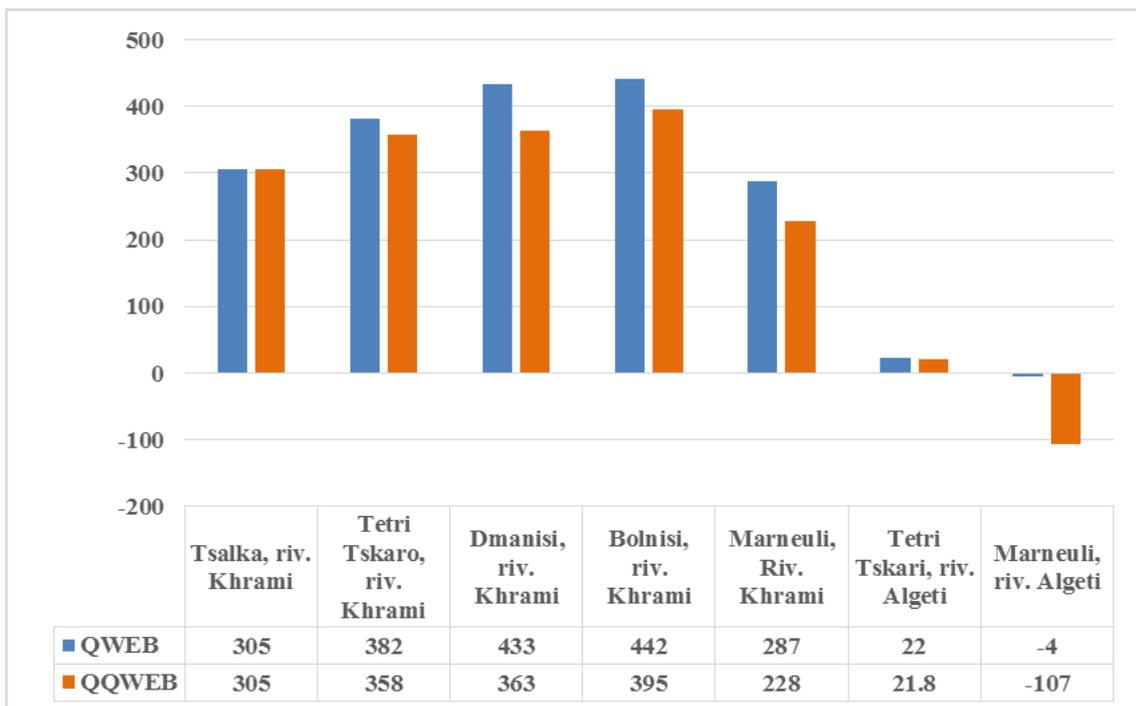


Fig. Vulnerability of the municipalities in terms of river basins, million m3 s

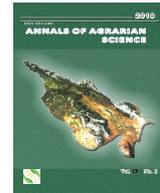
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Agroclimatic zoning of western regions of Georgia

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Received: 12 January 2019; accepted: 15 April 2019

ABSTRACT

Assessment and efficient use of agroclimatic resources is important for agroclimatic zoning in agriculture, as the growth and development of the agricultural crops and harvest mostly depend on the rational spatial distribution of the crops. When developing new areas, agrarian workers and farmers must consider the demands of crops for agroclimatic conditions what will improve their productivity. This will significantly increase their economic incomes and will contribute to the better provision of the population with the agricultural produce. Aiming at identifying the agroclimatic zones in the western regions of Georgia, based on the data of a 60-year-long meteorological observation, the agroclimatic resources of 6 regions (Samegrelo-Zemo Svaneti, Guria, Imereti, Racha-Lechkhumi-Zemo Svaneti, Apkhazeti) were evaluated. The thermal regime during the vegetation period of agricultural crops was evaluated by means of the sum of active temperatures ($>10^{\circ}\text{C}$), which changes by $\pm 300\text{-}400^{\circ}\text{C}$ annually and more. The parameters of atmospheric precipitations (in warm and cold periods), frosts (the first and the last one), periods without frost were determined according to vertical zoning in the agroclimatic zones identified within the region. In order to avoid the negative impact of frosts, it is recommended to use physical and biological methods against it. The perspective agricultural crops and soil types in the identified agroclimatic zones were described. Based on the agroclimatic zones identified in each region, the agricultural crops will be distributed in a rational manner and the prospects of their growth and development and high yield will be thoroughly identified.

Keywords: Agroclimatic zone, Active temperature, Atmospheric precipitation, Agroclimatic characteristics, Crops, Agroclimatic zone.

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Introduction

Provision of the population with agricultural products is one of the most important issues for the country. Therefore, profitable economy can be reached by employing the local agroclimatic resources (duration of sunshine, temperature, atmospheric precipitations, soil and air humidity, etc.) to the extent possible. These resources have an impact on harvest and economic income as a result. For Georgia, which is a land-poor country, the evaluation and efficient use of climatic and agroclimatic resources is of an utmost importance, and for the development of different branches of agriculture in the first place.

The agroclimatic resources must be evaluated on the basis of the climate properties (warmth, light,

atmospheric precipitations, etc.) associated with the harvest formation. In case of irrational use of the given agroclimatic resources, favorable growth or development of the agricultural crops or gaining guaranteed rich harvest will be impossible. The above-mentioned climatic parameters help select the relevant crops, expediently plan land cultivation, carry out various kinds of farming operations, etc. The average values of the following agroclimatic parameters are very important: temperature, precipitations, frosts, etc. They can be used to evaluate the agroclimatic resources of different regions in West Georgia [1-3] what allows the efficient use of agroclimatic properties, favorable growth and development of agricultural crops and their rational spatial distribution.

The regions differ with their agroclimatic properties. Therefore, it is necessary to differentiate between the agroclimatic zones of cereals, vine, tea, citrus, fruit, oil-bearing and ether-oil bearing, technical and other crops. Efficient use of the agroclimatic parameters of each zone will contribute to the efficient growth and development of crops and rich harvest forming the basis for profitable farming in an economic respect.

Study area

The territory of West Georgia, which, under the combined action of the Black Sea, solar radiation, high mountains of West Caucasioni and transformation of air masses, as well as orographic and complex mountainous relief, has several types of climate, in particular, humid subtropical climate up to 500-600 m above sea level, reaching Zemo Imereti where the humidity is a bit less. Moderate climate dominates 700 to 1300 m above sea level, and the cold continental climate dominates at 1400-2200 m above sea level. Above the latter altitude, there dominates very cold climate of eternal snow and glaciers. The agricultural conditions of the first three zones are favorable to grow relevant agricultural crops. There are 6 regions (Samegrelo-Zemo Svaneti, Guria, Imereti, Racha-Lechkhumi-Zemo Svaneti, Apkhazeti) with diversified agroclimatic resources in the study area.

Data and methodology

The work used: the database of the meteorological observations of the Institute of Hydrometeorology of Georgian Technical University; long-term observations data on the territories of the weather stations of 6 regions in West Georgia (Department of Hydrometeorology of the National Environmental Agency of Georgia); agroclimatic bulletins of the National Environmental Agency of Georgia (2008-2018); the agroclimatic resources of the region were evaluated by using the climatic and agroclimatic reference books: solar radiation, radiation balance and duration of sunshine (1968); air and soil temperatures (1967); atmospheric precipitations (1970); winds (1968), agroclimatic reference book of Georgia (1961); reference book of the agroclimatic resources of Georgia (1978); applied scien-

tific climatic reference book of Georgia (2004) and other relevant literary sources.

Besides, the classical study methods commonly used in agro-meteorology were used. The data of agro-meteorological observations were treated by using the method of mathematical statistics.

Results and discussion

Samegrelo-Zemo Svaneti region has a relief made up of plains, hills, mountains and high mountains (2000 m and more above sea level). The plain relief of Samegrelo (up to 100-150 m above sea level) is widely spread towards the Black Sea and created favorable geographical-economic conditions for social-economic development [4]. It extends north and north-eastwards with vertical zoning, reaches up the southern slope of main Caucasioni watershed ridge and covers the territory of Zemo Svaneti, above the altitude of 2000 m [5]. The given region is bordered by the Caucasioni mountain from the north, by Imereti region from the east, by Guria region from the south and by Apkhazeti region and the Black Sea from the west. Due to the influence of the Black Sea on the region, the plains and hilly (up to 500-600 m above sea level) relief of Samegrelo are located within the humid subtropical climate, while the average- and high-mountain locations of Zemo Svaneti are located within the moderate and continental climate characterized by moderately cold and cold climate, snowy winter and short summer, respectively [6]. The specific weight of the region in the production of annual and perennial plants is high. The following crops grow well on the plains and hills: tea, citruses, technical oil-bearing and essential-oil-bearing crops, vine (different varieties), kiwi (actinidia), nut, cherry laurel, fruit, etc. The following crops grow quite well in the mountainous and high-mountainous areas: cereals, vegetable, continental fruit, and there are vast areas of hay meadows and pastures (Table 1).

Table 1. *Agroclimatic zones characteristics of Samegrelo-Zemo Svaneti region*

Agroclimatic zones of Samegrelo-Zemo Svaneti region	Sum of active temperatures (>10°C)	Atmospheric precipitations (mm)		Frosts		Duration of periods without frost (day)
		Cold period	Warm period	First frost	Last frost	
I – zone	4400	630-850	800-1160	2.XII-7.XII	15.III-20.III	252-260
II – zone	4200-3700	860-990	1160-1220	23.XI-6.XII	21.III-29.III	236-257
III – zone	3700-2900	660-990	620-1220	1.XI-24.XI	30.III-11.IV	235-196
IV – zone	2800-2000	400-420	600-620	9.X-31.X	12.IV-25.IV	195-156
V – zone	1900-1100	870-1130	610-1130	16.IX-8.X	26.IV-8.V	155-116
VI – zone	1100-1000	390-490	650-790	7.IX-15.IX	7.V-22.V	115-75

The I agroclimatic zone covers the territory along the Black Sea coast, up to 200 m above sea level where Abasha, Zugdidi, Martvili, Senaki, partly Tsalenjikha, Khobi and Chkhorotsku municipalities are located. In the given area, along the Black Sea coast, there is a narrow strip of red-soil and podzolized soils. A bit far from this point, there are marsh-peats, and subtropical gleysols are found south, east and west of Khobi. Alluvial soils are spread around Abasha, Senaki and Khobi and partially, north-west of Chkhorotsku. North and east of Chkhorotsku, there are yellow soils spread and there are red podzolized soils spread around Zugdidi. Towards Mestia, there are brown acid and brown podzolized soils, and there are mountain-meadow turf soils north of Mestia [7, 8].

The factors in the given zone allow successful growing of cereals, vegetable-and-watermelons, citruses, some vine varieties (Tsolikauri, Ojaleshi, Tsitska), subtropical fruits, technical crops (tung, cherry laurel), kiwi (actinidia), nut, etc. Full ripening of oranges and grapefruit up to 200 m above sea level is limited due to the little necessary temperature sum in the vegetation period. In this zone, the production of citruses, besides warmth, depends on minimum winter temperatures, with the average indices of absolute air minimal temperature of -5°C to -6°C. These temperatures are not critical for citruses. However, they are more or less dangerous for lemon (unless it is protected against frosts). In

the given region, the areas with citruses may be expanded over the elevated slopes and hills, excepting plains and basins.

The II zone covers the territory north of the I zone, as well as Zugdidi, Chkhorotsku, Tsalenjikha and Martvili municipalities. It is located at 200-500 m above sea level. There are following soil types in the given zone: alluvial (south of Tsalenjikha), podzol (west of it), as well as red podzolized soils. The agroclimatic conditions in the given zone are favorable to grow only mandarin and lemon (provided the latter is protected against frosts), where the average absolute temperatures do not exceed -7°C, -8°C. The distribution area of the said crops in this zone incorporates the territories of piedmonts, up to 200-250 m above sea level and 300 m above sea level at some locations. The conditions in the given region are favorable to successfully grow: tea, vine, fruits, subtropical fruits, cereals, vegetable, etc.

The III zone covers the area adjacent to the north of the II zone, including Khaishi and Lebarde. It is located at 500-1000 m above sea level. The following soil types are spread there: rendzic leptosols, brown and brown acid soils. The given zone is favorable to grow cereals, vine (late variety, up to 800 m above sea level), fruits, nut, walnut, vegetable, tea (up to 600-650 m above sea level). Other perspective varieties are vine (early and late varieties) at 800-900 m above sea level.

The IV zone covers the area adjoining the III zone from the north, including Mestia municipality, at 1000-1500 m above sea level. The sum of active temperatures in this zone is relatively less. Therefore, the possibility to grow thermophytes in this zone is somewhat limited. Therefore, only early vine varieties can be grown at 1200-1300 m above sea level (on the southern slopes), as well as cereals (wheat, barley, oats, rye, early corn varieties), vegetable, potato, nut, fruits and berries.

The V zone covers the area adjoining the IV zone from the north, at 1500-2000 m above sea level. The following soils types are spread within this zone: mountain-meadow turf, brown acid and brown podzolized. In this zone, the following crops can be grown successfully: spring wheat, barley, oats, potato, vegetable and berries (black currant, currant chokeberry, sea-buckthorn), as well as juicy root crops for cattle. Besides, hay meadows and pastures can be developed successfully.

The VI zone covers a relatively small area adjoining the V zone from the north, which is very high, is located within the Alpine zone, at 2000-2500 m above sea level. The following soil types are spread in this zone: mountain-meadow turf and brown podzolized soils. The climate in the given zone is totally Alpine. Therefore, the crops needing less sums of active temperatures (1000-1100°C) can be grown there: early potato, oats, barley and

vegetable crops, as well as berries (blackcurrant, sea-buckthorn), root crops for cattle-breeding, and hay meadows and pastures can also be developed. The sum of said temperatures will be accumulated at 2100-2200 m altitudes above sea level.

Guria region covers partially hilly and high-mountainous relief and quite vast plain relief inclined towards the Black Sea, up to 100 m above sea level. The hilly locations are found up to approximately 400 m above sea level, the mountainous locations are found up to 1000 m above sea level, average-mountain relief is up to 1400 m above sea level and high-mountainous relief is at 2000 m above sea level. The given region is bordered by Ajara region from the south, partially by Samtskhe-Javakheti region from the south-east, by Imereti region from the north-east, Samegrelo-Zemo Svaneti region from the north and by the Black Sea from the west. The specific weight of Guria region in the development of agriculture of Georgia is very high, the following crops are produced in the region: cereals, tea, citruses, vine, subtropical, technical-oil-bearing and other crops. The agricultural crops are grown and produced in the region in terms of humid subtropical zone. Therefore, the leading branches in the region are: tea-growing and citrus-growing. Other important branches are: corn-growing, subtropical technical crops production, fruit-growing, etc. (Table 2).

Table 2. *Agroclimatic zones characteristics of Guria region*

Agroclimatic zones of Guria region	Sum of active temperatures (>10°C)	Atmospheric precipitations (mm)		Frosts		Duration of periods without frost (day)
		Cold period	Warm period	First frost	Last frost	
I – zone	4000	800-850	1000-1370	16.XII-25.XII	10.III-18.III	274-290
II – zone	3000-4000	850-900	1000-1200	11.XII-24.XII	11.III-21.III	265-288
III – zone	2000-3000	800	900	8.XI-26.XI	2.IV-18.IV	220-238
IV – zone	2000-1000	700	750-800	7.XI-20.XI	18.IV-27.V	202-113
V – zone	1000	650-700	700	20.IX-11.X	28.V-5.VI	96-113

The I zone covers the territory of Lanckhuti region, including the Black Sea coastal area and partially, the territory of Ozurgeti municipality. It is located at 10-200 m above sea level. The following soil types are spread in this zone: gleysols in the north, alluvial acid and marsh-peat soils in north-west, and red-soil and gleysols on the Sea coastal zone [7, 8]. The following crops can be grown successfully in the given zone: tea, citruses (mandarin, lemon), tung, kiwi (actinidia), feijoa, nut, vine (Tsolikauri, Chkhaveri, Izabela and some local varieties). As for the orange and grapefruit, full ripeness of their fruits, due to the little sum of active temperatures, is possible only 3 or 4 times in every ten years. The zone is also favorable to grow cereals, fruits and vegetable-and-watermelons.

The II zone covers the middle part of the region where the territories of Ozurgeti and Chokhatauri municipalities are located. The given zone is located at 200-300 m above sea level. The following types of soils are spread in the region: slightly alluvial and yellow podzolized north of Ozurgeti municipality, yellow soils and red soils east of Chokhatauri municipality and gleysols in the direction of the Sea coastal area. The given zone has quite favorable agroclimatic conditions in the west, on the piedmonts of Ozurgeti and Chokhatauri municipalities, at 200-300 m above sea level. Citruses (mandarin, lemon) can be grown successfully in the given zone, and vine, fruits, nut, cereals and other crops can be grown at higher altitudes. Tea can be produced at 500-600 m above sea level.

It should be noted that in the I and II zones, unless due protection is provided, lemon is expected to freeze for 3 or 4 times, orange and grapefruit - for 2 or 3 times in every ten years and mandarin - once in every 15 or more years.

The III zone spreads east and south-east of the II zone. It is located at 600-1000 m above sea level. The following soil types are spread in the east of the zone: yellow brown, red soils, brown acid soils and gleysols. The following varieties can be grown in the given zone: vine (average and early varieties), fruits, nut, berries, cereals, legumes and vegetable crops, as well as root crops for cattle-breeding and hay meadows and pastures can also be developed.

The IV spreads east and south-east of the III zone, from 1000 to 2000 m above sea level. The following types of soils are spread in the given zone: brown acid, yellow brown, red-soil and red podzolized soils. The sum of temperatures in the

given zone does not support the ripening of perennial thermophyte fruits but the given zone is favorable for berries, cereals (grain corn at 1000-1200 m above sea level), wheat, barley, oats, legumes and vegetable. The given zone is also favorable to grow juicy forage root crop for cattle-breeding and to develop for hay meadows and pastures.

The V zone is relatively smaller and spreads south-east and south of the IV zone, at 2000-2200 m above sea level. The following types of soils are spread in the zone: brown podzolized, red-soil, red podzolized and brown yellow soils. The following crops can be grown in the given zone: berries (black currant, chokeberry, willow-leaved sea-buckthorn, etc.) and vegetable crops (fenugreek, fennel, cumin, garlic, onion, parsley, celery, radish, cauliflower and early cabbage, carrot, etc.), as well as forage root crops for cattle-breeding; besides, hay meadows and pastures can be developed.

Imereti is located in the center of Georgia, at 20-1500 m above sea level and higher. Following its relief conditions, it was named as Kvemo (Lower) Imereti and Zemo (Upper) Imereti. A part of Kvemo Imereti has a plain and low piedmont relief, while Zemo Imereti has a hilly relief spreading over quite large areas. In an agricultural respect, the territory of Zemo Imereti at 400-600 m, has a more complex relief. Therefore, grain-growing, vegetable growing, fruit-growing, vine-growing, etc. are better developed over the plains. Most of the area at 600-800 m is presented as dissected gorges. The said crops can be grown only over small number of land plots. The territory above 1000 m above sea level, due to the great inclination, is mostly presented by forests and hay meadows and pastures [9]. Imereti region is bordered by Mtskheta-Mtianeti region from the east, by Samtskhe-Javakheti region from the south, by Racha-Lechkhumi-Kvemo Svaneti region from the north and by Samegrelo-Zemo Svaneti region from the west. Agroclimatic resources of Imereti are favorable to grow many kinds of crops, particularly, cereals, vegetable, vine, fruits, etc. The specific weight of corn is high among the cereals and vegetables are also grown intensely. Another successful branch in the region is vine-growing. In addition, the region offers favorable conditions to develop such branches as cattle-breeding, poultry-raising, bee-raising and sericulture (Table 3).

Table 3. *Agroclimatic zones characteristics of Imereti region*

Agroclimatic zones of Imereti region	Sum of active temperatures (>10°C)	Atmospheric precipitations (mm)		Frosts		Duration of periods without frost (day)
		Cold period	Warm period	First frost	Last frost	
I – zone	>4000	640-830	600-1000	12.XI-30.XI	12.III-2.III	251-274
II – zone	3000-4000	450-950	450-1190	20.XI-29.XI	24.III-1.IV	194-249
III – zone	>2000	600-750	640-850	31.X-18.XI	2.IV-18.IV	185-231
IV – zone	>1000	600-700	900-950	17.X-27.X	23.IV-1.V	176-188
V – zone	<1000	800	1000	13.X	5.V	158

The I zone covers the plain (lowland) of Kvemo Imereti) and hilly locations of Zemo Imereti at 20-300 m above sea level. There are different soil types spread in the zone, e.g. west of the zone, around Samtredia and Khoni municipalities, there are alluvial calcareous soils, there are subtropical podzols south of Khoni, red-soils in the north and yellow soils in the north-east. South and west of Tskaltubo, there are subtropical podzols; there are yellow soils in the north and rendzic leptosols in the east; there are yellow-soils around Vani, and alluvial satiated soils in the north; there are yellow and limestone-calcareous soils around Bagdati and there are subtropical podzols in the north of it; north of Zestaponi, there are alluvial-calcareous soils, and there are rendzic leptosols in the south-west and east of it. East of the given zone, around Kharagauli, there are rendzic leptosols, and there are brown yellow-soils little eastwards [7, 8]. The agroclimatic resources in the given zone are favorable to grow many different kinds of agricultural crops, in particular, cereals, vine, tea, mandarin, lemon (provided the it is protected against the freeze), kiwi (actinidia), feijoa, subtropical persimmon, essential oil-bearing crops, nut, tung, continental fruits, vegetable-and-watermelons, etc. In some years, guaranteed harvest is possible to gain only if the soil is duly moisturized, mostly in the regions where the atmospheric precipitations are 700 mm or less.

The II zone borders the first zone from the north, east and south. It is located at 300-500 m above sea level. In the northern and southern parts of the given zone, there are yellow soils, and there

are rendzic leptosols in the east. South of Tkibuli, there are yellow-brown soils spread. The same soil type is spread east of Kharagauli; immediately east of the given zone, east of Sachkhere, there are yellow-brown soils, and there are of rendzic leptosols spread in the south of the zone. It is possible to grow cereals, vine (early and late variety) fruits, vegetable and other crops in this zone. In some years when the atmospheric precipitations are 700 mm and less, it is recommended to increase moisture in the soil to gain the guaranteed harvest.

The III zone covers the area adjoining the II zone from the north-east, east and south, at 500-1000 m above sea level. Red soils are spread in the north-west of the given zone; there are intensely washed-down soils north and east of Tkibuli; there are limestone-calcareous soils east of Sachkhere and yellow brown soils east of Kharagauli. It is possible to successfully grow cereals, vegetable, vine (early variety) and fruit in the zone.

The IV zone covers the territories adjoining the III zone from the north-east and south, at 1000-1200 m above sea level. The following soil types are spread in this zone: limestone-calcareous soils in north-east and yellow-soils in the south of the zone. The following agricultural crops can be grown within the given zone: cereals, potato, vegetable, some early fruits and berries, and it is possible to develop hay meadows and pastures for cattle-breeding.

The V zone spreads in the extreme north-western and extreme southern parts of the region, at 1200 m and higher. The soil types in this zone are the same as in the IV zone. The following crops are perspec-

tive to develop in the zone: vegetable, berries and juicy root crops for cattle-breeding, and besides, hay meadows and pastures can be intensely developed.

Racha-Lechkhumi-Kvemo Svaneti region has a complex hilly-mountainous relief (plains, mountains, mountain slopes, gorges). The region is located on the southern slopes of West Caucasioni where Caucasioni watershed ridges and glaciers surrounding the area provide the conditions unfavorable for the sustainable development of agriculture in the region. The area of the plain relief in the region below 500 m above sea level is very small (2,2% of the total area). Village Tvishi is located at the lowest hypsometric altitude (400 m) and Mtiskalta is located at the highest hypsometric altitude (1840 m) [10]. Racha-Lechkhumi-Kvemo Svaneti region is bordered by Caucasioni Mountain from the north, partially by Shida Kartli region from the east, by Imereti region from the south and by Samegrelo-Zemo Svaneti region from the west and partially, from the north. The said region occupies the area up to 800-1000 m above sea level, in the moderate climate zone, while above this altitude, it is located in the continental climate zone (severe climate zone). Following relatively severe climatic conditions and complex relief, the plots of agricultural lands are few in number limiting an intense development of versatile branch of agriculture. Despite this, the dominating agricultural crops in the region, depending on relevant conditions are: vine, fruits, cereals and legumes. The soil and climatic conditions favorable to grow vine are mostly observed in the lowland and over the mountain slopes up to the

altitude of 400-800 m. It is also promising to grow stone fruits, nuts and berries (Table 4).

The I zone covers the area of the region, in particular, western part of Oni region, central areas of Ambrolauri and Tsageri municipalities and southern part of Lentekhi municipality. The given zone is located at 400-800 m above sea level. Raw Humus Calcareous soils, rendzic leptosols and humus-acid soils are spread around Ambrolauri municipality. Soils of similar types are spread in Oni, Tsageri and Lentekhi municipalities [7, 8]. The agroclimatic conditions in the given zone, as compared to other zones, are most favorable to develop and grow many different agricultural crops. (Winter and spring) wheat, corn, barley, vine, fruit, kiwi (actinidia), vegetable and watermelons can be grown successfully in this zone. In the given zone, in the environs of Tsageri region, in village Tvishi (400-500 m above sea level), it is possible to grow dry subtropical crops (fig, pomegranate and subtropical persimmon). In some years, during intense winter frosts (-18°C, -19°C), the probability of preventing them from damage is low (%). The said crops freeze to the root collar once in every ten years, while 3-year-old plantings freeze by 3 or 4 times, 2-year-old plantings - by 5 times and 1-year-old plantings freeze by 7 times in every ten years. Clearly, growing them is associated with certain risks. However, these crops are perspective and it will be useful, if during the frosts (-15°C, -16°C or more), the relevant measures to protect them against the frosts are taken.

The II zone spreads over the territory adjoining the I zone from the east, north and west and it

Table 4. Agroclimatic zones characteristics of Racha-Lechkhumi - Kvemo Svaneti region

Agroclimatic zones of Racha-Lechkhumi - Kvemo Svaneti region	Sum of active temperatures (>10°C)	Atmospheric precipitations (mm)		Frosts		Duration of periods without frost (day)
		Cold period	Warm period	First frost	Last frost	
I – zone	3000-3600	420-550	630-750	31.X-13.XI	3-5.IV	199-222
II – zone	2000-3000	550-800	800-1300	9.X-29.X	17.IV-5.V	155-193
III – zone	2000-1000	500-550	700-800	23.IX-8.X	6.V-20.V	124-153
IV – zone	1000-600	550	800	15.IX-22.IX	21.V-27.V	110-123

also covers a small area in the south. It is located at 800-1400 m above sea level. The following types of soils are spread in the region: rendzic leptosols, mountain-meadow peat and brown acid soils. Cereals (winter and spring wheat), corn, vine (early), fruit, vegetable and other crops can be grown successfully in this zone.

The III zone covers the area adjoining the II zone from the east, north and west, at 1400-1800 m above sea level. The following types of soils are spread in the region: brown-podzolized, mountain-forest-meadow and mountain-meadow peat soils. Cereals (winter and spring wheat, barley and oats), potato, vegetable, berries and root crops for cattle can be grown successfully in this zone, and pastures and hayfields can be developed.

The IV zone covers the area adjoining the III zone from the north and partially from the east. It is located at 1800-2000 m or higher above sea level. There are mountain-meadow turf soils in the given zone. Some perspective vegetables and berries approved for growing in the high mountains giving rich harvest (fenugreek, coriander, fennel, anise, parsley, celery, potato, black currant, etc.) can be grown in the given zone. Besides, the zone offers favorable conditions to intensely grow juicy root crops for cattle-breeding and expand hay meadows and pastures.

In a geomorphological respect, **Ajara region** has quite a complex relief: lowlands, hills, deep gorges, and average and high mountains. The Black Sea coastal lowland is located at about 100 m above sea level, the hilly relief is found at 500 m above sea level. Average-mountainous relief is found at 1000 m above sea level, and high-mountainous relief is located at 2000 m or higher. The lowlands in the given areas occupy 13.6%, hills occupy 9.3%, and mountains and piedmonts occupy 77.1%. The region is bordered by Turkey from the south, by Samtskhe-Javakheti region from the east, by Guria region from the north and by the Black Sea from the west. Ajara region is located in the humid subtropical zone of West Georgia with its upper border reaching 500-600 m above sea level. Higher that altitude, the climate is relatively more moderate and continental. Therefore, various agricultural branches are developed in the given region: tea-growing, citrus-growing, corn-growing, and tobacco-growing tung growing, etc. as cultural crops. The agroclimatic conditions of the region support the development of the branch of subtropical and continental fruit-growing as well. In addition, the region offers favorable conditions for vine-growing (different varieties), potato-growing, vegetable-growing, etc. (Table 5).

Table 5. *Agroclimatic zones characteristics of Ajara region*

Agroclimatic zones Ajara region	Sum of active temperatures (>10°C)	Atmospheric precipitations (mm)		Frosts		Duration of periods without frost (day)
		Cold period	Warm period	First frost	Last frost	
I – zone	>4000	1150-1290	1360-1500	2.XII-7.I	4.III-15.III	246-304
II – zone	3000-4000	850-1100	800-1330	6.XII-1.I	13.III-26.III	255-273
III – zone	2000-3000	700-750	750-800	11.XI-1.XII	30.III-15.IV	202-247
IV – zone	2000-1000	700	800	14.X-7.XI	18.IV-8.V	158-202
VI – zone	<1000	650	700	20.IX-9.X	12.V-28.V	158-113

The I zone covers the low part of the Black Sea coastal zone, including the territories of Kobuleti and Khelvachauri, at 100-200 m above sea level. There are soils typical to the humid subtropics spread within the given zone: subtropical gleysols west of Khelvachauri and alluvial soils on the Sea coastal lowland [7, 8]. The given zone offers favorable conditions to grow tea, citruses (mandarin, lemon, orange, grapefruit), tung, kiwi (actinidia), feijoa, nut and vine (Tsolikauri, Izabela and some local varieties). Full ripening of orange and grapefruit is possible only 5 or 6 times in every 10 years. The zone offers favorable agroclimatic conditions to grow cereals, legumes, subtropical persimmon, fruits and other crops [11].

The II is relatively vast and covers the territories of Kobuleti, Kedi and partially Shuakhevi regions. The zone is located at 200-400 m above sea level. The following soil types are spread in the region: brown yellow-soils are spread in all parts of Kedi region and mountain-meadow turf and brown acid soils are spread in the east. The given zone has humid subtropical conditions offering favorable conditions to grow the crops specified for the I zone. Orange, grapefruit or mandarin late varieties cannot yield desirable commercial results due to the little sum of active temperatures in the zone. Lemon in the I and II given zones may freeze twice or 3 times, while mandarin may freeze once in every 20 years unless provided by relevant protection.

The III zone is located in the central part of the region, at 500-1000 m above sea level and covers the territories of Shuakhevi and Khulo regions. From all sides of Shuakhevi municipality, there is brown yellow-soil spread, while brown acid soils are spread in the north and south. It is promising to grow the following crops in the given zone: cereals, legumes, vegetable, vine (average and early varieties), fruits, nut, berries, etc. The given zone is also favorable to grow root crops for cattle-breeding and for developing hay meadows and pastures.

The IV zone spreads north, east and south of the region, over small areas. It is located at 1000-1500 m above sea level. There are brown yellow-soils and brown acid soils in the region [7, 8]. It is possible to grow wheat, barley, oats, grain corn (1000-1100 m above sea level), potato, vegetable, fruits and berries in the given zone. It is also possible to grow root crops for cattle and to develop hay meadows and pastures.

The V zone spreads in the extreme eastern part of the region and a small part of it spreads north-

east of it. It is located at 1500-2000 m above sea level. During the vegetation period, the zone obviously lacks warmth. The soil types are not diversified. Brown acid soils are spread in the west, and brown podzolized soils are spread in the north and east. The following crops are perspective to grow in the zone: vegetable (coriander, fennel, garlic, fenu-greek, beet, etc.) and berries (blackcurrant, chokeberry, sea-buckthorn cultural variety, etc.). The given zone is favorable to grow juicy root crops for cattle and for hay meadows and pastures.

The relief in **Apkhazeti** region is hilly, mountainous or high-mountainous (2000 m above sea level and more) and presents a low plain location along the Black Sea coastline, which spreads more extensively south-east (50 m above sea level) [12]. The region is bordered by Russia from the north-west, it is bordered by Caucasioni Ridge from the north, by Samegelo-Zemo Svaneti region from the east and the Black Sea from south-west. Apkhazeti is located in the humid, subtropical climatic zone of West Georgia. Therefore, the agroclimatic conditions of this region are favorable to develop intense subtropical farming (tea, citruses, oil-bearing and essential-oil-bearing, vine, continental fruit, etc.), (Table 6).

The I zone covers the plain zone of the Black Sea coast and hilly locations at 250-300 m above sea level. The soil types spread in the given zone are typical to the subtropical zones, in particular, subtropical podzols, red-podzolized, yellow-podzolized, and partially, alluvial soils. Yellow and red soils are spread in Gali municipality, as well as subtropical podzols and subtropical gley soils. Subtropical podzol, subtropical gley soils and brown podzolized soils are spread in Gulripshi region. There are alluvial-calcareous and yellow soils north of Sokhumi, while subtropical gleysols are spread north-east of Gudauta municipality. Subtropical podzol, yellow- and brown yellow-soils are spread south-east of Gagra [7, 8]. The agroclimatic conditions of the given zone are favorable to grow tea, citruses, vine, tung, kiwi (actinidia), feijoa, nut, subtropical persimmon, fruits, as well as cereals (corn, wheat) and vegetables and watermelons. In this zone, it is also possible to grow essential-oil-bearing crops and to gain two harvests of geranium. The first harvest is gained at the end of the III decade of July and the second harvest is gained from October 15 until the onset of frosts. In some years, it is also possible to gain two harvests of East Indies basil. Jessamine and essential-oil-bearing rose can also be grown. In the given zone, where the average absolute min-

Table 4. *Agroclimatic zones characteristics of Racha-Lechkhumi - Kvemo Svaneti region*

Agroclimatic zones Abkhazia region	Sum of active temperatures (>10°C)	Atmospheric precipitations (mm)		Frosts		Duration of periods without frost (day)
		Cold period	Warm period	First frost	Last frost	
I - zone	>4000	730-970	710-1010	13.XII-25.XII	10.III-20.III	269-291
II - zone	3000-4000	850-950	1100-1400	2.XII-6.XII	25.III-28.III	251-260
III - zone	2000-3000	900-1000	1000-1300	7.XI-26.XI	3.IV-18.IV	238-202
IV - zone	1000-2000	800-1300	900-1100	14.X-5.XI	22.IV-8.V	157-198
V - zone	<1000	850-1400	1150-1200	20.IX-7.X	14.V-28.V	144-113

imum temperature of -5°C is observed, it is not recommended to grow lemon unless it is protected against the frost, while in Gagra, Sikharuli, Bichvinta, Akhali Atoni, Eshera and Gulripshi (with the temperature of -4°C) lemon can be grown successfully, without any protection.

The II adjoins the I zone from the north and is located at 350-500 m above sea level. It spreads from Gali municipality to Psou River. The following types of soils are spread in the region: red-soil-podzolized, Raw Humus Calcareous, brown acid, rendzic leptosols, yellow-podzolized, subtropical podzol and yellow soils. In the given zone, it is possible to grow early varieties of mandarin. Production of oranges and grapefruit is limited due to the lack of the necessary sum of active temperatures. Lemon needs annual protection against frosts. Mandarin, orange and grapefruit may be damaged once or twice in every 10 or 15 years. The zone is favorable to grow and gain rich harvest tea, vine, cereals, fruits, vegetable and other crops.

The III zone covers the area adjoining the II zone from the north and is located in the center of the region, at 600-1000 m above sea level. The following types of soils are spread in the zone: mountain-meadow turf, brown acid and brown podzolized. The following crops can be grown successfully in the given zone: fruits, vine (early varieties), cereals, legumes and vegetable. In the piedmonts, it is possible to grow geranium and essential-oil-bearing rose. One harvest of them can be gained from the first decade of October.

The IV zone also includes the area adjoining the III zone from the north and is located at 1050-1500 m above sea level. The following soil types are spread in this zone: mountain-forest-meadow peat and mountain-meadow turf soils, as well as brown acid soils. The following agricultural crops can be grown in the given zone: cereals (wheat, oats, barley), early corn (1000-1100 m above sea level), potato, vegetable, continental fruits (apples, pear), stony fruits (plum, etc.) and berries (blackcurrant and redcurrant, etc.), as well as root crops for cattle-breeding and beside, hay meadows and pastures can be developed successfully.

The V zone spreads in the extreme northern area adjoining the IV zone and is located at 1600-2000 m above sea level. The following types of soils are spread in the zone: mountain-meadow turf, brown acid and podzolized soils. The following crops are perspective to grow in the given zone: vegetable and berries (black currant, sea-buckthorn, etc.), as well as root crops for cattle-breeding and besides, hay meadows and pastures can be developed.

Conclusion and recommendations

Due to the climatic, orographic and other conditions on the territory of West Georgia, the distribution of agroclimatic resources is different. Efficient use of agroclimatic resources is of a particular importance to make farming profitable. Successful

farming basically depends on the rational distribution of the agricultural crops in the specific area by considering their demands to agroclimatic conditions. Based on the data of 60-year-long meteorological observations, depending on the sum of active temperatures ($>10^{\circ}\text{C}$) and atmospheric precipitations (mm), for the 6 regions in West Georgia, the following agroclimatic zones were identified based on vertical zoning, in particular, 6 agroclimatic zones for Samegrelo-Zemo Svaneti, 5 agroclimatic zones for Guria, 5 agroclimatic zones for Imereti, 4 agroclimatic zones for Racha-Lechkhumi-Zemo Svaneti, 6 agroclimatic zones for Ajara and 5 agroclimatic zones for Apkhazeti. By considering the given zones, it is possible to develop branches of agriculture (grain-growing, tea-growing, citrus-growing, vine-growing, fruit-growing, vegetable-growing, potato-growing, cattle-breeding, etc.). For the agroclimatic zone identified in each region, the parameters of atmospheric precipitations (in the cold and warm periods), frosts (the first and the last one), period without frost, as well as relevant soil types were specified. The sums of active temperatures ($>10^{\circ}\text{C}$) used to evaluate the thermal regime in the vegetation period varies by $\pm 300\text{-}400^{\circ}\text{C}$ and more. The lack of the given sum results in a sharp reduction of the harvest, and vice versa.

It should be noted that in the agroclimatic zones, where the negative impact of frost is expected on the agricultural crops, it is necessary to apply physical and biological methods against the frost. Consideration of the recommended agroclimatic zones and climatic parameters will help the farmers select perspective agricultural crops to grow specific crops, plan their farming in an expedient manner and select the right type of agricultural production.

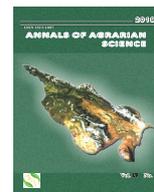
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Annals of Agrarian Science

Journal homepage: <http://journals-org.ge/index.php>



Vine culture in meskheti in old population census books

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Received: 05 June 2019; accepted: 02 October 2019

ABSTRACT

The article considers the areas of vine-growing in Meskheti covering not only the part of Meskheti on the territory of present-day Georgia, but also beyond it – the basins of the middle and upper reaches of the rivers Chorokhi and Mtkvari. The study used several population census books drafted for fiscal purposes during the Ottoman reign and giving the valuable social-economic data about the part of the territory of Georgia occupied by the Ottomans. The study demonstrated that vine grew only in few villages of Meskheti, though it must have been one of the leading cultures before the Ottoman domination. This is evidenced by the old Turkish population census books, which make it clear that vineyards were sparsely scattered on the territory of the whole Governorate and grew in the Nahiyesis and couple of villages much distanced from one another. Such a scattered location of vineyards is undisputable evidence of vine-growing being well developed in this region at some time in the past. Today, most of the villages where people commonly grow vine are on the territory of modern Georgia (Samtskhe-Javakheti, Ajara). However, today, vine-growing is not developed in most of them. It is typical that vine grew even further south, in the upper reaches of the Chorokhi River and in the basins of the rivers Oltisistskali, Banistskali and Bardusistskali (presently, in Turkey).

Keywords: Vine, Viticulture, Population census books, Georgia, Meskheti, Ottoman domination.

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Introduction

Historical-geographical and historical-cartographical studies have a long history in Georgia. However, there is still a series of questions not studied to date. Unfortunately, most such studies are focused on a single discipline - on history and do not highlight the questions belonging to other fields and also at the intersection of different sciences. Neither do they make use of modern technologies.

Historical cartography, as the discipline developed on the brink of cartography and geography, aims at compiling maps and atlases, developing the methods of their compilation and analyzing the peculiarities of the territorial allocation of the objects plotted on the maps. However, historical cartography in Georgia can be considered a less developed

discipline. One direction of historical cartography, which is creating a retrospective picture of the political situation, is more developed. It is many decades now, the historical maps with teaching, scientific or reference-and review purposes have been compiled in Georgia, but they are mostly historical-political. They depict the political situation of the past only – the territories of Georgia and its adjacent countries in different historical periods. We have no historical-physical-geographical, historical-demographic, historical-economic, historical-ethnographic, cultural-historical or other types of historical maps of the territories of Georgia and its adjacent territories, and consequently, there are no methods duly developed to compile them.

Historical cartography allows creating a retrospective picture of various political and social-eco-

conomic processes and identifying the types of rational nature use in the past. Such studies are important not only for providing a retrospective review, but also for identifying the traditional knowledge and experience bringing the economic benefit to the population in the past what can be used efficiently even today. Vine culture is one of them.

Historically, Meskheta covered the basins of the middle and upper reaches of the rivers Mtkvari, Chorokhi and its tributaries. Historically, more than one regions of Georgia were located here. Ivane Javakhishvili called them communities. These communities are: Artaani, Ajara, Erusheti, Tori, Klarjeti, Kola, Lazeti, Ligani, Oltisi, Palakatsio, Samtskhe, Tao, Potskhovi, Shavsheti, Javakheti and others. They cover quite a vast area, with only small part of it being a part of Georgia at present, and as per the modern administrative division of Georgia, it presents two regions only – Samtskhe-Javakheti and Ajara.

On the territory of modern Meskheta, vine grows only in very limited areas and vine-growing, as a branch, is developed in some areas only (only in Samtskhe, in the lower part of Akhaltsikhe basin slopes). However, the situation is absolutely different in a historical context. It is known that historically vine was quite widely spread on the territory of Meskheta. Besides, vine was widely grown not only on the territory of Meskheta being a part of modern Georgia, but also beyond the borders of the country. This fact was referred to in many scientific and scientific-popular literary sources. However, the exact area occupied by vineyards in the past is not still identified exactly.

Methods and initial data

An important original source to create a retrospective picture of vine-growing areas in the past is old Turkish population census books. In this respect, in our view, particularly important are some population census books drafted for fiscal purposes by the Ottoman authorities. They are:

- „The Grand Book of Gurjistan Governorate“, dated by 1574 [1];
- „The Grand Book of Gurjistan Governorate“, dated by 1595 [2];
- „Big and Brief Books of Ajara Liva (District)“ [3];
- „Great old census book of Tbilisi Governorate“ of 1728 [4].

These documents describe the period when

the Meskheta was a part of Ottoman Empire as a Governorates (Vilayets, Provinces) and give a real picture describe the situation at the moment of censuses quite accurately. Today, a certain part of this territory is included in Adjara, Samtskhe-Javakheti, while another part of it rests beyond the borders of Georgia (Artaani, Klarjeti, Kola, Lazeti, Potskhovi, Shavsheti, Tao, Tortumi, etc.). All of these documents are valuable for different specialists such are: historians, geographers, demographers, economists, linguists and etc.

The research was accomplished based on the interdisciplinary studies. The study was oriented on compiling thematic maps. For this purpose, the data in the old census books were ranked and classified; designing GIS database and relevant legend and compiling a series of thematic maps. The objects given on the map were identified by comparing them to other sources. This will be done by means of comparative, semantic, retrospective mapping and other methods of study. A geographical-cartographic analysis of the objects included in the old census books have been done.

The old cartographic sources, unlike modern maps, were compiled in a different metric system and their analysis needs absolutely different approaches. Virtually, no such practice has ever been used in Georgia. For this purpose, the methodology of GIS-registration and combination of the old with the modern cartographic basis was developed.

Main results

Peculiarities of territorial distribution of vine

The scientific literature gives much abundant data about the vine-growing areas on the territory of Meskheta, which is a part of present-day Georgia. Different sources [5-11] give different numbers of the vine cultivars. Taking into consideration numerous sources we have registered 59 vine cultivars in this region. Compared to other regions of Georgia this one is just poor in the quantity of cultivars but very significant for it's a original vine varieties By the areas and density of vineyards Meskheta is not a prominent region and it lags behind other regions as Samegrelo, Imereti, Kakheti and etc. [12]. It is clear that direct duplication of the present-day situation in the past cannot be right. However, by considering the natural conditions, the general state of affairs must be similar in the past and at present. In any case, the climatic conditions on the territory of pres-

ent-day Georgia supported vine-growing both, in the past and at present more than in the areas located further south in the country, where the mountainous relief and severe climate of the South Caucasus do not favor vine-growing.

In 1574, the Great Register of Gurjistan Governorate names vineyards in thirty out of more than 1000 Georgian villages what, at the first sight, must be the sign of poor distribution of vine in the area. However, if taking a closer look at the territorial distribution of the vineyards, we will see that they grew in the Nahiyesis, which were much distanced from one another (Artanuji, Didi Artaani, Tortomi, Oltisi) and in a couple of villages. Such a scattered nature must be undoubted evidence of vine-growing being well developed in this region in the past, while few sites with vine survived by 1574 must be viewed as relict remnants of the branch well developed in the past. What is the reason for making such a conclusion? Purportedly, the villages located almost in the same natural conditions had the same economic structure and specialization, i.e. if vine-growing was developed in one village, the same would be true for its neighboring villages. Otherwise, it is impossible to explain such a scattered nature of the vineyards. At the same time, we may suppose that vine was survived in some other villages, of which the Register says nothing. In this instance, we mean individual vine plantings (perhaps, high-growing

vineyards), which were not taxed due to very little harvest they gave. The reason for such a conclusion is the great many feral vines found on certain territories of Meskhети, presently within the borders of Georgia, where no vine grows at present.

A similar discourse will be fair for the Great Register of Gurjistan Governorate of 1595, according to which vine grew all over the given area, including Javakheti. It is noteworthy that in respect of the number and development of vineyards, out of 9 Livas of the Governorate, Akhalkalaki Liva ranked the fourth in Meskhети” [13]. Most of the villages where vine grew commonly are on the territory of present-day modern Georgia, though presently, vine-growing is not developed in most of them. Large areas with vineyards survived in Khertvisi and Akhaltsikhe Nahiyesis (Fig. 1). The vineyards in these areas have survived even to our days, what is hardly true with the territories near the southern border of Georgia, e.g. Akshehiri, Tmogvi or and Nialiskure Nahiyesis. Even A more interesting fact is that vine grew even further southwards, in the upper reaches of the Chorokhi River and in the basins of the rivers Oltisistskali, Banistskali and Bardusistskali (presently, in Turkey). Not only doesn't vine grow there, but the area does not offer any favorable climatic conditions for vine-growing.

In some instances, old Turkish Population census books allow identifying the number of vineyards in

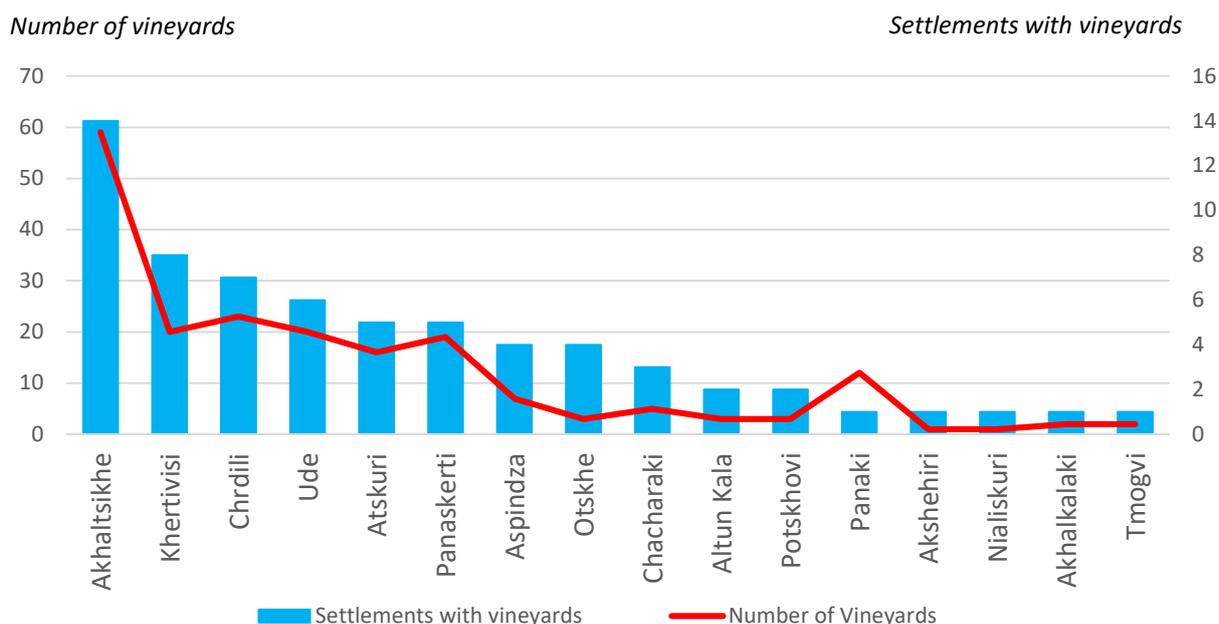


Fig. 1. Distribution of vineyards in Meskhети according to old Turkish Population census books

different villages and amount of taxes imposed to them. For instance, according to the Register of 1595, there were over 100 vineyards in the Governorate. Today, it is very difficult to determine the size of these plots; however, the taxes imposed to them give us a certain idea about the kind of soils and crop capacity of vine growing in such areas. So, we want to say that no fixed tax was imposed to each vineyard. The difference was seen among the villages as well and even on the territory of a single village. In particular, in village Nurmira (the same as Buzmareti), which was located in Panaskerti Nahiyesi of Didi Artaani Liva, one vineyard was evaluated to 40 akçe, while it was evaluated to 50 and 60 akçe at other locations. There were also the vineyards presumably giving very rich harvest. For instance, a half of one vineyard was evaluated to 48 akçe, i.e. 96 akçe for the whole plot. The factors used to determine the amount of tax can be assumed quite accurately – first of all, it was the crop capacity. This, first of all, depended on the grape variety and soil fertility.

The Register of Zemo Ajara also evidenced that vineyards were commonly grown there too, but not in very large areas. It is typical that at that time, vine grew on the territory of present-day Khulo Municipality. Such territories are villages Skhalta and Khula (the same as Khulo), where the tax for vineyards was 200 and 150 akçe, respectively. It should be noted that administratively, Zemo Ajara Liva of that time was divided into two units: Kvemo Ajara and Zemo Ajara Nahiyesis. Vine was not even mentioned in Kvemo Ajara, which was located more in the lower reaches of the Ajaristskali River, while several areas with vineyards were mentioned in Zemo Ajara Nahiyesi. It is without a doubt that vine was grown in other villages of Ajara located at lower hypsometric heights; however, the given Register gives data only about certain territories of present-day Ajara. The Sea coastal area and subtropical zone are not included in such territories, where vine-growing is one of the well-developed branches at present.

The Register of 1728 gives an absolutely different picture, with quite high vineyard taxes. This can be explained in two ways. First of all, the given Register was written two centuries later when the purchasing power of akçe could have been changed. On the other hand, the influence of the Ottoman economic policy was less in this area and consequently, the local people retained more vineyards. Some villages paid even 2000 akçe for vineyards

(e.g. villages of Petre Nahiyesi: Zanavi, Zemo Rveli, Kortaneti, Nua, etc.). It is also interesting that in some villages, vineyard tax ranked the second after the grain tax and made almost 25% of all economic taxes (Fig. 2).

One can cite other sources too evidencing the common nature of vine-yards in Meskheta, e.g. data by Vakhushti Bagrationi [14] and map compiled by Ivane Javakhishvili on their basis [9]. As it seems, the gorge of almost all upper, middle and lower reaches of the Chorokhi River (near the river) is occupied by vineyard-and-fruity area. This is hardly true with the River Mtkvari basin, where the vineyard-and-fruity areas were found within the borders of Kvemo Kartli and Shida Kartli lowlands, Borjomi Gorge and Akhaltsikhe Basin.

One of the historical records evidences that the vineyards grew in Akhalkalaki uyezd (district): “As Balas states, in 1895, 10 vineyards with the area of 3 desiatina (3,27 ha) were registered in Akhaltsikhe uyezd and there were 3 vineyards registered in Akhalkalaki uyezd” [6].

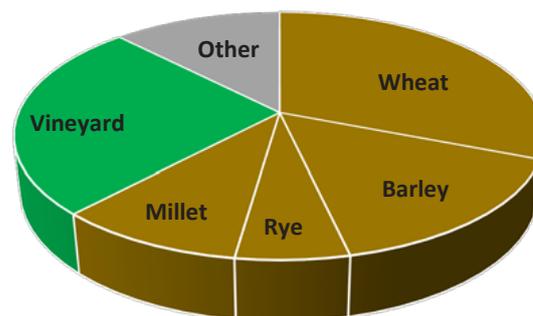


Fig. 2. Taxes in village Nua (as per Tbilisi Register of 1728)

In Meskheta, including the territory of historical Georgia, vine also grew over quite large areas in the Mtkvari River gorge – from Khretvisi to Artaani [15].

According to the villages incorporated in the old Turkish Population census books, it became possible to determine the hypsometric distribution of vineyards. By fixing the absolute heights of these villages, it was identified that vine in Meskheta grew at absolutely different heights within a quite large vertical range. For instance, in Ajara vine grows within the whole altitudinal range where vine commonly grows across Georgia what is hardly true with other territories of Meskheta, Samtskhe in particular, where vine grows only at certain hypsometric steps.

There is another noteworthy fact – in present-day Georgia, vine grows at 1200 (1340) m above sea

level, while as the Great Register of Gurjistan Governorate states, it grew even higher, in particular, the vineyards in the gorge of the Olaverdi River (the Paravani basin) reached 1900-1940 m above sea level [12]. If observing modern climatic conditions, we will be sure that in such areas, growing even early vine varieties is virtually impossible. The average temperature of the coldest month of the year is $-8-10^{\circ}\text{C}$ and the absolute minimum is -38°C . Therefore, we can suppose that people used to bury vine in winter. However, this technique allowed growing vine only at some places and not developing the vine-growing as a branch.

Evidence of developed vine-growing in Meskheti

The facts of widely spread vineyards and well-developed branch of vine-growing in Meskheti are strongly evidenced, and as mentioned above, the old Turkish Population census books are the primary evidence. However, due to few numbers of villages charged with vineyard taxes (and due to little taxes as well), we considered it necessary to use other evidences as well. Archeological remnants are one of the undoubted evidences. Clay amphorae buried in earth, wine presses, wine bowls and prints of vine leaves and grape pips were found at different locations: Bagebi, Gugata, Vardzia, Idumala, Ikhtila, Minadze, Oshora, Okhera, Ude, Sadzeli, Toloshi, Tsira, Tsnisi, Chobiskhevi, etc. [5-7, 13].

They find old vine roots in Meskheti even today. What is most important, Meskheti is outstanding for a great number of cultural indigenous vine varieties what is an undisputed evidence of a great tradition of centuries-long growing and cultivating of this culture. Unfortunately, this is hardly true with the part of Meskheti on the modern territory of Turkey. Archeological data are also quite scarce. Therefore, the remnants found on the territory of historical Meskheti are of a particular interest. For instance, vine jars or different shapes and capacities are found buried in ground in almost all villages of Ligani gorge (an area in the lower reaches of the Chorokhi River, south of Machakhela). As per the oral traditions, they were made locally as well, but were mostly delivered from Borchkhi. Even wine and grappa were found in many of them [15].

Centuries-long vine-growing tradition in Meskheti is also evidenced by the fact that the local people were much familiar with vine culture and used to successfully select various vine species for concrete natural local conditions. Traditionally,

farmers used the slope inclination and exposition and peculiarities of the soils efficiently. In mountainous areas, they often grew vine over terraces. Usually, they did not plant any fruit trees near the vine. However, in order to prevent land slide, at the ends of the terraces, they planted perennial crops, such as walnut, oak, chestnut and other big trees. They used to deliver fertile soil from the river floodplains and manure to the terraces. Over the terraces in Meskheti, they grow low-growing vine. The vines were planted at the bottom of the terrace wall so that its sprouts would spread over the wall and grapes would also ripen there. Thus, vine did not need any support.

In Meskheti, people used to make terraces in two ways: terraces without walls (lari)- arranged over the slopes with $8-10^{\circ}$ inclination surfaces and walled terraces (darija) – arranged over the slopes with greater inclinations. Due to the little inclination of the relief the lari did not need any walls. The situation was quite different in the case of darija. It was arranged on steep slopes along river gorges in order to use the river water for the soil irrigation. For instance, the darijas in Khertvisi were irrigated from the River Paravani. The water flew along several hundred meters through a special channel made of stone to the vineyards and here it had switchers at the head of every terrace to run the water to it. The channel was constructed in such a way that the water flew from the highest terraces down to the lower ones [12]. Vine was mostly grown on the terraces with stone walls, together with fruit trees. As for such terraces, they were provided in the gorges of the Rivers Chorokhi, Mtkvari and their tributaries.

For centuries, people followed this tradition and passed it across generations. However, the situation changed later: the terraces turned desolate in the Soviet years, as they were inaccessible for heavy techniques, while in Turkey, they were destroyed due to the decreased scales or ultimate demolition of the branch of vine-growing. Old artificial terraces have survived at some places to date. However, their trace has disappeared at many locations – they were either washed down by torrents, or covered with forests. Therefore, the terraces on the territory of historical Meskheti are of a particular interest. For instance, S. Timofeev talks about terraces in Artvini environs, which the people made over the slopes due to insufficient land or mountainous relief and grew magnificent vine varieties [16].

Vine being one of the major agricultural crops and a religious symbol not only in Meskheti, but

also all over Georgia, is evidenced by ancient Georgian ornaments with vine leaves and grape decorations, ornaments and frescoes used as the decorations of many Christian cult buildings. The Zarzma, Vardzia, Oshki, Bana, etc. are some vivid examples.

In Meskheti, like all over Georgia, customs and traditions associated with vine culture have survived. People used wine as a ritual drink for many different celebrations.

Rich ampelographic Georgian language, including Meskhi terms, is the primary evidence of well-developed vine-growing and farmers' favorable scrutiny in this region. The vocabulary of Meskheti is primarily associated with vine, e.g. with individual parts of vine, shape and size of vine grains, vine growing peculiarities, terms of the first waking and further growing of vine, diseases, etc. There are also very many terms describing the rows of vineyards and spaces between them, vine stakes, etc. New sprouts of vine branches were called "Chavli" in Ajara, while they called it "Deda-Vasi" (Mother Vine) in the rest of Meskheti territory; for wild vine they used terms "Useless vine" and "Bad-luck vine", respectively [17]. Even at different places on the territory of Meskheti, people used different terms to denote the same objects. For instance, in Ajara they called wild vine not only useless vine, but also forest vine, wild vineyard, wild grape, Krikina, etc.

Vine varieties in Meskheti

Growing vine depends on two important circumstances: 1) natural factors (mostly, orographic, climatic and soil conditions), and 2) anthropogenic factors (totally determined by a man). An important and leading role of the natural factors is undoubted and a number of works have been dedicated to this topic. Much fewer are the works considering anthropogenic factors. Out of anthropogenic factors, it is important to identify how efficiently the whole complex of optimal natural conditions necessary to grow vine is chosen: the orographic, climatic and edaphic conditions; vine-growing tradition (distance between the plantings, distance between the rows, etc.); seasonality of vine-growing and caring, wine-making technique, etc.

Selecting optimal environmental conditions for vine is immediately associated with centuries-long farmers' knowledge and experience and folk traditions. Besides, anthropogenic factors often differ across the regions, even within the limits of the

same country. This is particularly true with Georgia, the country with extremely diversified natural conditions and culture, diverse botanic and ampelographic features and properties of vine varieties, as well as rich and diversified experience of vine-growing. However, quite often, centuries-long experience gets forgotten: the techniques to choose the right location or village to grow concrete vine varieties to make best wine are often forgotten, with some exceptions. This is why it is important to analyze various sources and discover the forgotten traditions of vine-growing and wine-making and introduce them to practice.

As for the old Turkish Population census books, they say nothing about vine varieties. They only mention vineyard plots. Neither do other historical documents give any valuable data about this topic. Consequently, based on various sources of a later period and by considering the natural conditions, we may talk about the vine varieties spread in the given period. Besides, it should be noted that many varieties have had their names changed from Georgian to the Ottoman ones. Therefore, a full picture is impossible to restore. For example, the name "Tskhenis-Dzudzu" was replaced by "At-Memesim" and "Khanum-Barmaghma" (Lady's finger in Turkish), "Saliklevi" was replaced by "Sirklev", "Meliskuda" was replaced by "Melviskvit", "Kharistvala" was replaced by "Oqvim-Gozim", etc. [18].

As per D. Bakradze, they mostly grew high-growing vine in Ligani gorge, like in neighboring Ajara and Shavshet-Klarjeti. All grape varieties grew there and they were set growing on high trees [19].

N. Marri also talks about grape varieties in Shavshet-Klarjeti [20]. He names 20 grape varieties and gives their brief ampelographic description. At some points in his work, he even names their locations.

More thorough data are given in the works by S. Timofeev, Er. Nakashidze and St. Menteshashvili. Based on them, Iv. Javakhishvili listed the following grape varieties: white/pink varieties (Khopaturi, Klarjuli, Mtsvane, Tsvite, Chichibe, Bagis Kurdzeni (Garden Grapes), Tskhushi, Brola, Butkoi, Burdzgla, Tetri Kurdzeni (White Grapes), Llikana Kurdzeni) and black/red varieties (Kharistvali, Tskhenis-Dzudzu, Chkhaveri, Mekrenechkhvi, Jineshi, Chodi, Satsuri, Matanauri, Makhaturi, Oriona, Mtevandidi, Shavi Kurdzeni (Black Grapes), Shavshuri, Povnili) [18]. As per the records by S. Timofeev and N. Marri, white grapes: Khevarduli, Tskhenis-Dzudzu, Meliskuda, Mtsvanura,

Soreki, Orjokhuli, Gorgouli, Tskaltetuli, Alichelebi, Oqviz-Gozi (Turkish for “Kharistvali”), Stambul-Vazi, Turvanda (Persian/Turkish for “Adreuli” (“Early Varieties”) and black/red grape varieties: Butko, Tskhenis-Dzudzu, Shishveli, Jghe, Shavropi, Saperavi, Saliklevi, Orjokhuli, Khalturi, Akhalaki, Jineshi, Pundukh-Vizvim, Dervish-Ali commonly grew in Shavshet-Klarjeti [18].

It is known that some names of grape varieties indicate their origin. Such names are many on the territory of Georgia. For instance, Ivane Javakhishvili names some grape varieties in Meskheta, four in each region: In Ajara (Matanauri, Klarjuli, Keduri, Khopaturi), in Shavshet-Klarjeti (Shavshura, Klarjuli, Orjokhuli and Tskaltetuli), in Chaneti (Kapis-

toni or Kapito, Opoura, Atinuri and Kvapaturi or Khopaturi) [18]. As for Ligani Gorge, such varieties as Livanuri, Shavshuri, Orjokhuli, Singoturi, Istanbulai (Chaushi) grew widely there [21].

It is noteworthy that Ivane Javakhishvili named Chaneti as the center of creative vine-growing in west Georgia after Egrisi and Argveti, whose contribution is outstanding if considering that grape variety Kabistoni or Kapito growing all over Georgia must have been spread from Chaneti to the rest of the country [18].

Based on the above-listed sources, we may contemplate about the vine varieties spread on the territory of historical Meskheta (Table 1).

Table 1. Major vine varieties spread on the territory of historical Meskheta

Vine varieties	Black/red				Vine varieties	White/pink			
	Ajara	Shavshet-Klarjeti	Chaneti	Samtskhe		Ajara	Shavshet-Klarjeti	Chaneti	Samtskhe
Akhalaki		+			Alichelebi		+		
Butko	+	+			Bagis Kurdzeni	+			
“Tamaris Vazi” / “Tamaris Tsremlebi”				+	Bezhana / Chreli / „Mosaika“				+
Matanauri	+				Butko	+			
Makhaturi	+				Brola	+			
Mekrenchkhi	+				Burdzгла	+			
Mtevandidi	+				Gorgouli		+		
Orona	+				Tavdakiduli				+
Orjokhuli		+			Tavtseskhlа				+
Povnili	+				Tita Meskhuri				+
Saliklevi		+			Klarjuli	+			
Saperavi		+			Kolosha		+		
Satsuri	+				Meliskuda		+		
Shavi Kurdzeni	+				Mtsvane/Meskhuri	+			+
Shavropi		+			Mtsvanura		+		
Shavshuri	+				Orjokhuli		+		
Shishveli		+			Oquz-Gozi		+		
Chkhaveri	+				Roketula				+
Tskhenis-Dzudzu	+	+		+	Samariobo				+
Chodi	+				Saparuli				+
Khalturi		+			Soreki		+		
Kharistvala	+	+	+		Stambul-Vizvim		+		
Jineshi	+	+			Chichikhe	+			
Jghe		+			Tskhenis-Dzudzu				+
					Chkhushi	+			
					Tsvite	+			
					Tskaltetuli		+		+
					Kharistvala Meskhuri				+
					Khevarduli	+			
					Khopaturi		+		

Reasons for decreased vineyard areas

A number of scientific literary sources state about the decreased vineyard areas in Meskhети. An accent is made on the raging of foreign invaders and Islamization of the local people for centuries. “Due to the Islamization of local people, the branch of vine-growing, which was well developed at one time, was destroyed. As for wine-making, it was ultimately suppressed because of the ban of wine drinking, and almost all famous local grape varieties became extinct” [11, 22, 23]. This fact is referred to in a number of historical documents and other scientific works [6, 18, 10, 23, 13].

We support the fact that the conquest of this territory of Georgia by the Ottomans must have resulted in the diminution or total extinction vine-growing, the branch well-developed before. As it is known from the history, vine was invaluable material and spiritual wealth of a Georgian man and therefore, foreign invaders always tried to destroy it. The situation was aggravated by the fact that the religion forbade the Ottomans to drink wine and this was one more cause for them to destroy vineyards.

However, not only political processes must have been the cause of vine destruction. We think that the natural conditions also played an important role in reducing the vineyard areas. In particular, we talk about the climate cooling in the XIII-XVIII centuries. This stage glaciation reached its peak at the beginning of the XIX century [24]. Besides, the palynological studies [25] demonstrated that the period following the XVI century was characterized by climate cooling followed by short (40-year-long), but severe climatic conditions at the turn of the XVII century resulting in the extinction of vine-growing in South Georgia. This makes us think that diminution and extinction of vine-growing and wine-making in this region of Georgia was resulted not only by the political situation. Rather, it was a natural process as well. However, it is clear that the political situation also played a certain role in this respect [12].

Conclusion

Despite the fact that according to old Turkish Population census books, vineyards grew only in few villages of Meskhети (particularly in historical Meskhети left beyond the territory of present-day Georgia), it may be said for sure that vine must have been one of the leading cultures there. The reason for such a statement is the scattered nature of the

vineyards all across the territory of the Governorate, survived only as fragments of the vineyard areas, which were so vast sometime in the past.

As per the old Turkish Population census books, it becomes clear that the vineyards grew in the Nahiyesis and villages much distanced from one another. Such a scattered nature in a couple of villages in the Mtkvari and Chorokhi River basins must be considered as undisputable evidence of vine-growing being a leading branch in the past in the given area. Such scattered distribution of the vineyards cannot be explained otherwise. At the same time, we may suppose that vine would have been survived in some of those villages of which the Register says nothing. We mean small number of vine plantings, which were not taxed due to little harvest.

Most of villages where vine grew widely, are today a part of present-day Georgia, though in most of them, vine-growing is not developed at all. It is interesting that vine grew even further south, in the upper reaches of the Chorokhi River and in the basins of the rivers Oltisistskali, Banistskali and Bardusistskali (presently, in Turkey). Not only doesn't vine grow there, but the area does not offer any favorable climatic conditions for vine-growing.

Acknowledge

The authors cordially thank the Shota Rustaveli Science Foundation for support, under whose financial aid it became possible to accomplish the present work within the scope of project “Mapping and analyzing Georgian cultural heritage of south-west Georgia according to old Turkish Population census books”, Grant #HE-18-448, 26.07.2018).

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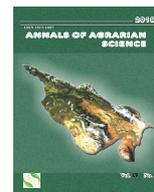
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Annals of Agrarian Science

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The determination of heavy metals in medical plants in georgia

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Received: 25 March 2019; accepted: 29 May 2019

ABSTRACT

The aim of this investigation is the determination of heavy metals (lead, cadmium, cooper and zinc) in the medicinal plants grown at the various part of Georgia. For the purposes to use of medicinal plants it is important to determine in them the content of the toxic microelements, especially since the ecological state in the world is changing for the worse. Accumulation of heavy metals in the various parts of plants is particularly important, because extreme demands are placed not only on the content of effective ingredients in these plants but also on their harmless. The following parts of the plants: 1. Rhizoma Potentillae, 2. Fructus Foeniculi, 3. Herba Hyperici, 4. Flores Chamomillae, 5. Folia Plantaginis Majoris, 6. Rhizomata et radices were investigated by differential-pulls polarographic method. The results of the study showed that medicinal herbs collected in Georgia have high quality, since one side the content of the toxic element – Pb in them is much lower than acceptable by the international standard and the concentration of Cd is equal to zero. On the other side, concentration of essential microelements – Cu and Zn are normal.

Keywords: Heavy metals, Medicinal grass, Polarographic method, Rhizoma Potentillae, Fructus Foeniculi, Herba Hyperici.

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Introduction

Recent years, interest to medicinal plants has been increased and the potential of their use is in progress. It is well known, that they can be used for various chronic and acute diseases, since they have high biological activity and less toxicity. Metabolic processes occurring during the ontogenesis period of plants form very important and valuable compounds, such as essential oils, alkaloids, glycosides, tannins, vitamins or other biological active substances that have a mild and long-lasting effect on the human body. For the purposes of use of medicinal plants it is important to establish in them the content of the toxic microelements, especially since the ecological state in the world is changing for the worse. Proceeding from the fact that medicinal plants are used for a long time, a great deal of determination has in them the content of heavy metals as they can accumulate in the human body.

Georgia occupies an interesting geo-botanical position as a part of Caucasia – the region which links Europe with Asia. The country is characterized by rather contrasting natural conditions, which account for the extremely high degree of divergence of plant communities within this comparatively small area. The rich and unique phylogenetic fund of Georgia represents a natural-historical treasure and requires the permanent conservation and rehabilitation, as it progressively exterminates or changes under the influence of various natural disasters. There is spread the unique medicinal, aromatic, melliferous, spicy and poisonous plants in Georgia, among them there are some varieties of plants which are not to be found anywhere in the world [1]. Due to their current state, most of these plants are on the verge of extinction. The erosive processes of genetic resources and uncontrolled export are going on. To all this is added the development of

industrialization, urbanization and construction. Heavy metals have generated several serious environmental problems that they are easy to transfer into soil and water, which will pose extreme toxicity to plants and aquatic organisms. The heavy metals contents of herbal plants are variable due to the factors like differences between the plants species, geographical area and exposure to different pollution sources. It is important with medicinal plants because extreme demands are placed not only on the content of effective ingredients in these plants but also on their harmlessness, including the content of heavy metals, namely Cd and Pb.

In previous years, we investigated the content of heavy metals in the medicinal herbs (*Melissa Officinalis* L., *Salvia Officinalis* L., Flaxseeds and its oil) grown on the own area of TSMU I. Kutateladze Institute of Pharmacochemistry [2-4]. The content of all four metals (Cd, Pb, Zn, Cu) was normal in above mentioned plants.

Current study belongs to study of medicinal plants growing on the outskirts of large cities (Tbilisi and Gori) of Georgia. Namely, the first - village Kiketi, which is located 13 km from Tbilisi on the south-east slope of the Trialeti ring; altitude is 1200 m above sea level, with a mild, temperate climate. The second - village Ateni, which is located 10 km from Gori on the northern slope of the Trialeti ring; altitude is 730 m above sea level, with a moderately, humid climate.

It is well known, that the plants tissue contents almost all the chemical elements. However, 12 of these elements (C, H, O, N, P, K, S, Ca, Mg, Fe, Zn, Cu) are essential nutrients for all plants [5-7]. They are required for body structure, fluid balance, protein structures and to produce hormones. They are a key for the health of every body system and function, but if their concentration does not meet the generally accepted norm, then they become harmful. The heavy metals As, Pb, Cd and Hg have not any function in the living organism, only are very harmful to plant, animal and human bodies. The main objective of our study was set to determine the quantity of the heavy metals (Cd, Pb, Cu, Zn) in plants spread in Georgia. The parts of following plants: 1. *Rhizoma Potentillae*, 2. *Fructus Foeniculi*, 3. *Herba Hyperici*, 4. *Flores Chamomillae*, 5. *Folia Plantaginis Majoris*, 6. *Rhizomata et radices Inulae* have been investigated by us. The listed medicinal plants are used all over the world and in particular in Georgia from the past centuries. These plants have many medicinal properties such as:

1. *Rhizoma Potentillae* and its rhizome extracts have been known for a long time in traditional medicine as a remedy for the treatment of inflammations, wounds, and gastrointestinal disorders. There are 30 species in Georgia and two of them are Georgia's endemic.
2. *Fructus Foeniculi* is very popular in Georgia. Sweet fennel fruit medicines are available in various forms to be taken by mouth. Preparations made from sweet fennel fruit can also be found in combination with other herbal substances. This combination is the essential herbs for hernia of cold type with abdominal pain because of its actions of warming kidney and liver.
3. *Hypericum herb* is used as part of many pharmaceuticals and its medicinal properties have such as: anti-inflammatory, antiseptic, spasmolytic action, stimulates skin regeneration.
4. *Flores chamomile* (*Matricaria recutita*, also known as *Matricaria chamomilla* or *Chamomilla recutita*) is one of the best-known medicinal herbs in the world. It has anti-inflammatory, healing, anti-allergic effects. The extract is used in a wide range of cosmetics.
5. *Folia Plantaginis* in Georgia there are 11 types. Extract of its leaf is characterized by anti-inflammatory, bactericidal, sedative, anti-depressive action. Used for respiratory tract infections, nervous disorders and insomnia, wounds and ulcers, gastritis, colitis.
6. *Rhizomata et radices Inulae* show a wide range of pharmacological action: anti-inflammatory, antimicrobial, blood-stopping, wound healing. decreased inflammatory process activity in the stomach, decreases the stomach acidity, strengthens the mucous substance.

Objective and Method

The following parts of the plants: 1. *Rhizoma Potentillae*, 2. *Fructus Foeniculi*, 3. *Herba Hyperici*, 4. *Flores Chamomillae*, 5. *Folia Plantaginis Majoris*, 6. *Rhizomata et radices* were investigated by differential-pulls polarographic method with a dropping mercury electrode ($t=3.5$ s, $m=2.6$ mg/s) by a three-electrode cell. The analytical procedure included: a careful washing technique by acetone and rinsing many times with redistilled water, drying at the 100°C for an hour, weighing and burning at the 450°C for 5 h in the quartz vessel. The received ash was treated by 1 N HNO₃ and evaporated. Af-

ter this 1 N HCl was added and evaporated again. The ash obtained in result of the mineralization of a plant was dissolved in 10 ml of 0.1 N HCl. After this, the solution is placed into the thermostatic cell ($t=25^{\circ}\text{C}$) and during in 10 minutes is blazing by inert gas. The value of potential was taken towards the saturated calomel electrode potential.

Results and analysis

The results of polarographic analysis of heavy metals content in plants are represented in Fig. (given as an example for *Rhizoma Potentillae*), which shows that each of investigated microelement gives a sharp peak at a certain very specific value of potential, namely, the value corresponding to the half-wave potential of the given microelement for Cu, Pb, Zn $E_{1/2} = -0.2\text{V}; -0.5\text{V}; -1.05\text{V}$ consequently. The peak correspondent of cadmium is absent, because concentration of this metal in the investigated plants from all above mention villages equals zero.

In Table are listed the concentration of 4 metals in all investigated by us in medicinal plants.

On the basis of our investigation it was established, that the toxic metal content in the medicinal plants from the villages near the cities (Tbilisi and Gori) of Georgia does not exceed the limits allowed by World Health Organization [8]. The maximum permissible levels in raw materials of medicinal plants for Cd and Pb are amount of 0.3 mg/kg and 10 mg/kg, respectively. As for Zn and Cu the WHO limits for these metals have not yet been established. It should be noted that the content of copper and zinc varies depending on the plant itself, with which

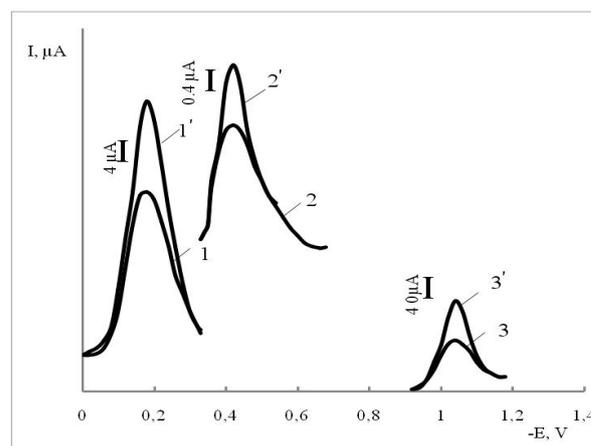


Fig. Polarograms for heavy metals in 1 g sample of the plant in the supporting electrolyte of 0.1 M HCl: 1 - Cu(II), 2 - Pb(II), 3 - Zn(II); 1,2,3- corresponding standard solutions.

their various healing properties are apparently related. Zinc is a component of many metal-enzymes, especially some enzymes which play a central role in nucleic acid Metabolism [9]. Zinc is also a membrane stabilizer and a stimulator of the immune response [10]. Manifestations of acute zinc poisoning include nausea, vomiting, diarrheal, fever and lethargy. The estimated safe and adequate daily intake of zinc is between 10.0 and 20.0 $\mu\text{g}/\text{day}$. As well as zinc, copper is essential to the human body since it forms a component of many enzyme systems, such as cytochrome, oxidise and ceruloplasmin, an iron-oxidizing enzyme in blood. The observation of anaemia in copper deficiency may probably be related to its role in facilitating iron absorption and in the incorporation of iron in haemoglobin [11].

Table. Results of research of the medicinal plants content of ions

#	Name of the plant	Content, mg/kg			
		Cu	Pb	Cd	Zn
1	HerbaHyperici	31.80	2.63	0.00	96.70
2	Flores Chamomillae	3.01	2.04	0.00	99.98
3	Rhizomata et radices Inulae	29.40	1.60	0.00	65.90
4	FructusFoeniculi	20.10	1.98	0.00	97.70
4	FructusFoeniculi	20.10	1.98	0.00	97.70
6	RhizomaPotentillae	1.50	2.40	0.00	82.90

The maximum permissible level of copper is 12.0 µg/ day. Therefore, the magnitudes of the content of zinc and copper in medicinal plants correspond to the rate necessary for medicinal properties.

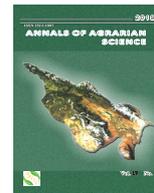
The absence of Cd and the low value of Pb in all of by us studying medicinal herbs are logically associated with relatively high levels of Cu and Zn. The absorption and distribution of Cd is usually influenced by low intake of Zn and Cu and contrary. Cadmium has a negative effect on enzymatic systems of cells with its ability to substitute for other metal ions (mainly Zn²⁺, Cu²⁺ and Ca²⁺) in metal-enzymes and has a strong affinity for biological structures containing sulfhydryl-groups.

Conclusion

We determined the content of Pb, Cd, Zn and Cu in various parts of plants located proximity to the vehicle big towns (Tbilisi, Gori). The investigation was performed through differential-pulse polarographic method. On the basis of our investigation it was established that the toxic metal content in the medicinal plants content of lead, cadmium, copper and zinc does not exceed the limits allowed by World Health Organization standards. It should be noted that the content of copper and zinc varies depending on the plant itself, with which their various healing properties are apparently related. The results of the study showed that medicinal herbs collected in Georgia are of high quality, since one side the content of the toxic element – Pb in them is much lower than acceptable by the international standard and the concentration of Cd is equal to zero. On the other side, concentration of essential microelements – Cu and Zn are normal.

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Theory of soil compaction by running bodies of mountain tandem wheeled self-propelled chassis

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Received: 10 February 2019; accepted: 19 April 2019

ABSTRACT

The theory of soil compaction by running bodies an mountain tandem-wheeled self-propelled chassis is developed. Proceeding from the soil rheological properties and parameters of adaptive mountain tandem-wheeled self-propelled chassis in the deformation distribution zone is defined functional dependency between soil average density and running parts of tandem-wheeled self-propelled chassis. Is developed the technique of calculation of all physical values that are stipulated due standard for assessment of running system's parameters impact on ground. Due these parameters is stated the comparison of experimental and serial self-propelled chassis.

Keywords: Rheological models, Deformation depth, Equalizing beam suspension, Soil compaction, Chassis, Machine-tractor.

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Introduction

Density represents one of the most important characteristics of the soil. For different crops value of optimal density is in a rather narrow range (for grain - 1.1 ... 1.3 g/cm³ for the row - 1.1 ... 1.5 g/cm³). Due to the impact of mobile members of machine-tractor units on the soil increases its density in the plow and subsurface horizons that leads to serious changes in the soil structure and characteristics, deterioration of root system's habitation and, ultimately, in lower crop yields [1].

Nowadays in the field of agricultural production mechanization occurs a gradual transition to an intensive and high technology machine crop technologies that cause the complication of machinery, extension of their functionality, increase in capacity, and as a result, increasing of weight, number of passes and speed of movement on the fields. In the process of soil preparation, planting, plant care and harvesting various machines are passing through the field from 5 to 15 times, resulting in the total area of vehicles propulsion tracks of 2 times exceed the size of field, the 10 ... 12% of field area are undergoing propeller impact of 6 up to 20 times, 65 ...

80% - from 1 to 6 times and only 10 ... 15% of the area is not compacted by machines [1].

It was defined that as a result of machine-tractor unit's passage on the field in the soil are created large-sized compaction zones, concentrating around the tracks of running bodies and extending to a distance of 0.8 ... 1.0 m on either side of the tracks of caterpillars or wheels. On depth these zones are extended to the whole topsoil and reach 0.6 m [1].

As a result of multiple impacts of propulsion, as well as tillage tools occurs an accumulation of compaction deformations not only in the plow, but also in the subsurface layer of soil that is harmful to fertility. The resulting plow sole prevents the water penetration into the soil, leads to water erosion, or to its rapid drying and wind erosion during drought. Plow sole violates the capillary flow of moisture from the deeper layers to the surface and prevents the development of root system. The destruction of plow sole is using chisel plows or chisels that significantly improve the soil fertility, but raise the price of technological process.

Recently the reducing of compaction effect impact on soil by agricultural machinery propellers occurs in three ways [1].

Technological way consists in improving the crops cultivation technology, including reducing the number of passes, a rational of routing machines movement, application combined and wide-cut units, minimum tillage, arrangement of permanent travel lanes for vehicles, application of overload technology in the interaction with vehicles, etc.

Agronomical way consists in increasing the soil capacity to resist compaction and shear loadings due organic fertilizers application, mulching, and keeping the all quality indicators for soil cultivation, the introduction of additional decompression operations.

Design way that consist in the improvement of tractors, agricultural machinery and their propulsion in the direction of eliminating or reducing harmful effects on the soil.

A certain reducing of on the soil would be achieved by increasing of tires size, but simultaneously are compacted the deeper layers of soil. The perspective would be the application of multi-axis tractors in order to reduce pressure on soil, and consequently, it compaction [2]. This would be achieved without increasing the number of tractor axles, by setting on the rear drive axle of tandem wheels suspension equalizer. This self-propelled chassis can be given also adaptive properties to expand their application scopes in small farms [3].

Basic part

The schematic diagram of the adaptive self-propelled chassis is shown in Fig. A.

With regard to running bodies of this energy means theory of soil compaction requires significant improvements with application of rheological models of soil deformation.

For the averaged estimation assessment of the soil density on the tractor trail with an area F we can write down:

$$\rho = \frac{M}{F(H^* - h)} = \frac{M}{FH^* \left(1 - \frac{h}{H^*}\right)} = \frac{\rho_0}{1 - \varepsilon}, \quad (1)$$

where M - is the mass of the soil compacting layer, kg;
 H* - is the depth of deformation distribution, m;
 h - is the depth of gauge, m;
 ρ0 - is the soil initial density, kg/m3;
 ε - is the relative deformation.

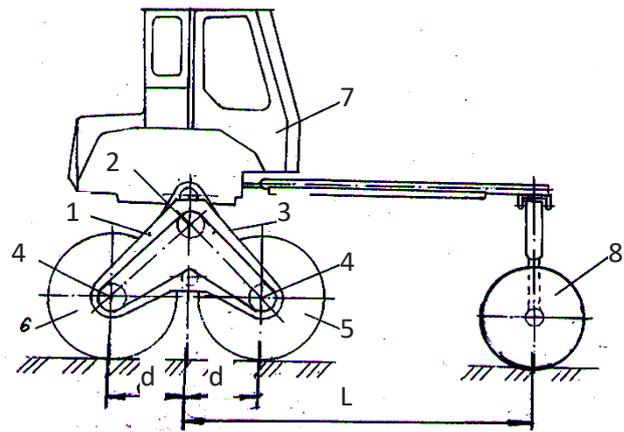


Fig. Schematic diagram of the adaptive self-propelled chassis.

1-balancer gear; 2-drive axis; 3-chain transmission; 4- drive wheels axis; 5-front drive tandem wheel; 6-rear drive the tandem wheels; 7-propelled chassis, 8-front wheels.

With the application of Voigt rheological model we obtained a design formula of gauge relative deformation after the passage of the three wheels of adaptive self-propelled chassis in the following form [4].

$$\varepsilon = \frac{G_g}{3F_0H} \left(1 - e^{-\frac{\ell}{\tau v}}\right) \left(1 + e^{-\frac{2d}{\tau v}} + e^{-\frac{L+d}{\tau v}}\right), \quad (2)$$

and with the application of general law of linear deformation equation

$$\varepsilon = \frac{G_g}{3F_0} \left[\frac{1}{E} + \left(\frac{1}{H} - \frac{1}{E}\right) \left(1 - e^{-\frac{\ell}{\tau v}}\right) \left(1 + e^{-\frac{2d}{\tau v}} + e^{-\frac{L+d}{\tau v}}\right) \right], \quad (3)$$

where G_g - is the total weight of self-propelled chassis, N;

S - is the instantaneous modulus of elasticity, N/m²;

H - is the long-term modulus of elasticity, N/m²;

$\tau = \frac{\mu}{H}$ - is the deformation delay time or time, s;

μ - is the coefficient of viscosity, N.s/m2;

ℓ - is the length of propeller bearing surface, m;

F0 - is the average area of wheels contact with the ground, m2.

At $\frac{1}{E} \approx 0$ the design formulas (2) and (3) coincide. For example, substituting in (1) the expression (2) we obtain the averaged soil density on the tractor trail in the following form:

$$\rho = \frac{\rho_0}{1 - \frac{G_g}{3F_0H} \left(1 - e^{-\frac{\ell}{\tau v}}\right) \left(1 + e^{-\frac{2d}{\tau v}} + e^{-\frac{L+d}{\tau v}}\right)} \cdot \quad (4)$$

In terms of herbage conservation, the criterion for the compaction effects on soil is not the averaged density but the soil density in the propeller's trail [5]:

$$\rho_t = \rho_0 + \alpha U, \tag{5}$$

$$\alpha = \frac{\rho_0(1-\nu^2)}{E_0 H^*}, \quad U = \omega b^* q_{\max} (1 + \ell_g n). \tag{6}$$

where H^* – is the deformation distribution depth, m;
 ν – is the coefficient of lateral expansion (Poisson ratio);
 E_0 – is the module of total deformation, N/m²;
 ω – is the coefficient depending on the size and shape of the propeller bearing surface (For wheeled propeller $\omega = 1.25$ [1]);
 b^* - is the width of single propeller trail, m;
 q_{\max} – is the maximum pressure on soil, N/m².
 For the wheel of system:

$$q_a = \frac{M \cdot g}{\sum_{i=1}^n F_i}, \tag{7}$$

where n - is the number of wheels on one track;
 M – is the mass of tractor.

It was defined that allowable propeller impact without crop yields decreasing is in the range $U < 75$ kN/m that is equivalent to the soil moisture at 25 ... 30% $q_{\max} \leq 0.075$ MPa, and at 8 ... 12% - $q_{\max} \leq 0.15$ MPa [1].

In the design formulae (5) and (6) the unknown quantity is the depth of deformation distribution H^* . As it is known, the relative deformation $\varepsilon = \frac{h}{H^*}$, i.e. $H^* = \frac{h}{\varepsilon}$. With consideration of the unit movement speed, soil rheological properties, as well as wheel geometrical and loading parameters, is obtained the expression for determining the depth of gauge at one wheel [6]:

$$h = \sqrt[3]{\frac{G^2}{b^2 K^2 D}} \cdot \sqrt[3]{\left[\frac{1 + (\tau \omega_0)^2}{1 + (2\tau \omega_0)^2} \right]^2}, \tag{8}$$

where G – is the load on wheel, N;
 D – is the diameter of wheel, m;
 b – is the width of wheel, m;
 K – is the coefficient of volume collapse, N/m³;
 τ – is the soil relaxation time, sec;
 ω_0 – is the angular velocity of wheel, 1/sec.

At increasing of re-loading to a value that precedes it, the wheels subsidence occurs, as at the re-deformation, by the same load. The further growth of re-load causes deformation of the deeper layers of soil, at this occurs the subsidence, as in the first loading or it slightly decreases because of the upper layers compaction. The impact reducing would be estimated by the factor K_L [2]:

$$K_L = 1 - \left(\frac{q_i}{q_{i+1}} \right)^{2n'} \tag{9}$$

where q_i and q_{i+1} – are the pressure values at previous and subsequent loadings, N/m²;
 n' – is the trial coefficient.

Then the dependence of subsidence on the various values of wheels pressures for consecutive arranged three-wheel self-propelled chassis would be written down as:

$$h = \sqrt[3]{\left[\frac{1 + (\tau \omega_0)^2}{1 + (2\tau \omega_0)^2} \right]^2} \cdot \frac{\left[\sqrt[3]{G_1^2 + \sqrt[3]{G_2^2} \left(1 - \frac{q_1}{q_2}\right)^{2n'}} + \sqrt[3]{G_3} \left(1 - \frac{q_2}{q_3}\right)^{2n'} \right]}{\sqrt[3]{b^2 K^2 D}}. \tag{10}$$

At loading decreasing from the passage to the passage the soil gauge depth is determined by the pressure developed during the first pass. At the same load on all wheels magnification factor of wheel is estimated by [1]:

$$K_0 = 1 + \ell_g n. \tag{11}$$

Then the dependence of subsidence on pressure of consecutive arranged three-wheel self-propelled chassis will be as:

$$h = \sqrt[3]{\frac{G^2}{b^2 K^2 D}} \cdot \sqrt[3]{\left[\frac{1 + (\tau \omega_0)^2}{1 + (2\tau \omega_0)^2} \right]^2} (1 + \ell_n 3),$$

or

$$h = 1,477 \sqrt[3]{\frac{G^2}{b^2 K^2 D}} \cdot \sqrt[3]{\left[\frac{1 + (\tau \omega_0)^2}{1 + (2\tau \omega_0)^2} \right]^2}. \tag{12}$$

At load decreasing on the passage to the passage the soil gauge depth is determined by the pressure developed during at the first pass, i.e. by expression (8).

We obtain that for a self-propelled chassis with equalizing beam suspension of two driving wheels the depth of deformation distribution in Voight rhe-

ological model is defined by:

$$H^* = \frac{h}{\varepsilon} = \frac{1,477 \sqrt[3]{\frac{G^2}{b^2 K^2 D}} \cdot \sqrt[3]{\left[\frac{1+(\tau\omega_0)^2}{1+(2\tau\omega_0)^2}\right]^2}}{\frac{G_g}{3F_0 H} \left(1 - e^{-\frac{\ell}{\tau v}}\right) \left(1 + e^{-\frac{2d}{\tau v}} + e^{-\frac{L+d}{\tau v}}\right)} \quad (13)$$

Similarly, when using the rheological model of the general law of linear deformation, we obtain:

$$H^* = \frac{1,477 \sqrt[3]{\frac{G^2}{b^2 K^2 D}} \cdot \sqrt[3]{\left[\frac{1+(\tau\omega_0)^2}{1+(2\tau\omega_0)^2}\right]^2}}{\frac{G_g}{3F_0} \left[\frac{1}{E} + \left(\frac{1}{H} - \frac{1}{E}\right) \left(1 - e^{-\frac{\ell}{\tau v}}\right) \left(1 + e^{-\frac{2d}{\tau v}} + e^{-\frac{L+d}{\tau v}}\right)\right]} \quad (14)$$

At $\frac{1}{E} \approx 0$ formulae (13) and (14) are coincide.

The depth of deformation distribution for a commercial self-propelled chassis with application of Voigt model and results of the study [3] would be:

$$H_s = \frac{1,301 \sqrt[3]{\frac{G^2}{b^2 K^2 D}} \cdot \sqrt[3]{\left[\frac{1+(\tau\omega_0)^2}{1+(2\tau\omega_0)^2}\right]^2}}{\frac{G_g}{2F_0 H} \left(1 - e^{-\frac{\ell}{\tau v}}\right) \left(1 + e^{-\frac{L}{\tau v}}\right)} \quad (15)$$

At the same load on the wheel, the depth of deformation distribution for mountain self-propelled chassis would be increased on the value of:

$$K = \frac{H^*}{H_s} = 1,704 \frac{1 + e^{-\frac{L}{\tau v}}}{1 + e^{-\frac{2d}{\tau v}} + e^{-\frac{L+d}{\tau v}}} \quad (16)$$

For example, let's assume that: $\tau=1\text{m}$; $L=2.55\text{m}$; $d=0.546\text{m}$; $\ell=0.29\text{ m}$; $v=5\text{ km/h} = 1.39\text{ m/sec}$; $G_g=23100\text{ N}$; $G_a=3850\text{ N}$; $b=0.5\text{ m}$; $D=0.71\text{ m}$; $K=2500\text{ kN/m}^3$; $H \approx E=2 \cdot 10^5\text{ N/m}^2$; $\omega=1.25$; $q_{\text{max}}=1.5q_a=161\text{ kN/m}^2$. We obtain: $K = 1.28$, $h \approx 0.05\text{ m}$ (formula 8); $\varepsilon \approx 0.05$ (formula 2), $H^* = \frac{h}{\varepsilon} \approx 1\text{ m}$; $\omega=29.756\text{ kN/m}$ (formula 6); $\rho/\rho_0=1.053$ (formula 5).

Due to the increasing in deformation distribution depth H^* the value of α in the formula (6) will be decreased. Also would be reduced the maximum pressure q_{max} on the wheels because increased number of wheels. As a result, significantly would be reduced the soil density in the propeller trail ρ_i . According to the formula (5), by adjusting the of self-propelled chassis parameters the soil density in the propeller

trail ρ_i and propeller impact without crop yields reducing would be improved to the optimum value in terms of grass cover keeping.

Conclusion

The theory of soil compaction by running bodies of mountain tandem-wheeled self-propelled chassis is developed. Are defined functional dependency between soil average density in the soil deformation distribution zone and soil density in the propeller trail on rheological properties of soil and parameters of adaptive self-propelled chassis. The method of all physical values calculation that are needed to evaluate the assessment of impact indicators of running system on ground by standards is developed. According to these indicators is given the comparison of experimental and commercial self-propelled chassis.

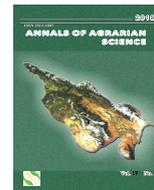
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The studies of technological and physical-chemical properties of rabbit meat

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Received: 03 May 2019; accepted: 24 July 2019

ABSTRACT

Each year worldwide, there is an increase in the consumption of rabbit meat, which is due to its high nutritional value and recognized dietary properties. We have studied the technological and physical-chemical properties of the rabbit meat of the most common in Georgia Californian and New Zealand white rabbit breeds. The modern, standard, generally accepted methods of research were used in this work. There have been determined the share of products obtained as a result of the slaughter of rabbit breeds under study, the yield capacity of anatomical parts obtained as a result of disintegration of the slaughtered carcass, their morphological composition and the meatiness index, on the basis of which, it is possible to develop technologies for the rational use of different parts of the processed rabbit meat. The studies of physical-chemical properties showed that pH value determining the stability of meat relative to decomposing microflora and the amino-ammoniac nitrogen content defining the goodness of meat did not exceed the norms and standards, the test for primary products of decay of proteins in the broth was negative, and the reaction of peroxidase was positive, indicating that the meat was produced by healthy animal. The studies of the contents of lead, cadmium, arsenic and mercury in different rabbit breeds do not exceed acceptable standards that indicate their safety, as well as their sanitary and hygienic reliability.

Keywords: Rabbit meat, Tissue composition, Physical-chemical characteristics, Toxic substances, Safety, Standard.

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Introduction

Meeting people's needs for high quality and safe foods is a major challenge of our day.

The meat is a nutritious food that must be included in the human diet. It contains a number of essential biologically active substances needed to regulate growth, regeneration, plastic and energy processes, as well as to restore protein resources of the organism.

In today's conditions, there is a deficit of meat in Georgia, as well as the significant increase in meat prices, which in turn leads to large quantities of imported meat products. According to data provided by Geostat, in 2017, Georgia imported 124 thousand tons of frozen meat [1]. In order to solve this very important problem, we need to find new types

of local high quality meat.

Rabbit-breeding is a highly promising branch of livestock farming. Rabbit is distinguished by early maturity, high fecundity and high meat productivity. Its breeding does not require too much spending on major construction, energy and human resources. Consequently, costs of production for rabbit meat is also lower in comparison with other types of meat. On average, one doe produces 100 kg of meat and raw materials for producing 55 pieces of jackets [2-8].

Each year worldwide, there is an increase in the consumption of rabbit meat. The world leading countries manufacturers of rabbit meat are China, Italy, Spain and France. Its consumption per capita in Italy, for example, is 4,5 kg per year, while in Europe the average indicator is 2 kg [2,9-12].

In Georgia, the formerly well-developed rabbit-breeding branch, requires now the rejuvenation.

Rabbit meat is an ecologically clean product. American scientists have established that until the age of six months, rabbit does not accept at all strontium-90, as well as other products of atomic disintegration of pesticides and herbicides. Rabbit meat pertains to the “white meat”, has a finely-fibred and gentle consistency, well-digested by the organism. Compared with beef, pork and poultry meat, morphological characteristics, technological properties, nutritional and biological value of rabbit meat of rabbit meat are higher: they distinct from them by significant yield of muscle tissue, and smaller quantity of connective tissue, the relatively larger amounts of easily digested complete and quite small amounts of fat and cholesterol. Rabbit meat is also a source of vitamins and minerals [13-25].

The above mentioned properties of rabbit meat indicate expediency of its use, as high-quality meat, in production of healthy foods, dietary and clinic nutrition. [9,26-29].

The aim of the work is to study the technological and physical-chemical properties of the rabbit meat.

Objectives and methods

The studies were carried out in the laboratories of the Department of Food Technologies of Akaki Tsereteli State University. The following rabbit breeds were chosen as the objects of study: the Californian rabbit and the New Zealand white rabbit, their age was 130 days. The rabbit meat slaughtered according to generally accepted methodology was stored at a temperature of +4 0C for 24 hours.

We assessed the quality of rabbit meat on the basis of technological and physical-chemical indicators. To this end, we used generally accepted methods of the analysis of meat products described in the appropriate standards, normative documents and special literature.

The share of products obtained as a result of the slaughter of different rabbit breeds, the yield capacity of anatomical parts remained after technological disintegration of the slaughtered carcass and its morphological composition were determined by calculating after removing non-food products from slaughtered carcass, disintegration of carcass into several parts and after they are weighted.

The index of meatiness of separate anatomical parts was determined by quantitative ratio of muscle and bone tissues.

We studied the physical-chemical properties of rabbit meat by State Standard [30] according to the following indicators: pH medium, amount of volatile fatty acids (mg/KOH), test for primary products of decay of proteins in the broth (reaction with CuSO₄), amount of amino-ammoniac nitrogen (%), the reaction of peroxidase.

Stability of meat relative to decomposing microflora – pH medium was determined by potentiometric method, on a pH meter Mi150 according to the instruction [31]. Meat freshness was studied by the content of volatile fatty acids and test for primary products of decay of proteins in the broth (reaction with a 5%-solution of CuSO₄); goodness of meat – by amount of amino-ammoniac nitrogen; and meat healthiness – by the reaction of peroxidase.

The heavy metal content was determined on an atomic adsorption spectrophotometer SHIMADZU AA-6200, the lead content was determined by State Standard GOST 26932-86 [32], the cadmium content - by State Standard GOST 26933-86 [33], the mercury content - by State Standard GOST 26927-86 [34], and the arsenic content - by State Standard GOST 26930-86 [35].

Statistical processing of the results obtained and the assessment of the reliability of the data were made by the mathematical statistics methods using the Windows' IBM SPSS Statistics IBM SPSS Statistics software (version 20.0). To describe the variation series, we measured the arithmetic mean and the average standard error.

Graphical interpretation of the results was made by using Microsoft Excel.

Tables and graphs illustrate the data of the experiments performed, each value is a mean of at least five determinations.

Results and analysis

At the first stage of the work, we identified the share of products obtained as a result of the slaughter of Californian and New Zealand white rabbit breeds (Table 1).

Table 1. *The share of products obtained as a result of slaughtering different rabbit breeds*

Name of part	Yield capacity, % of live weight	
	Breeds	
	Californian	New Zealand white
Live weight, kg	3,00±21,5	3,260±21,5
Slaughtered carcass	64%	65%
Head	4,3%	4,2%
Skin	8,3%	8,0%
Ears, feet, tail.	2,9%	2,5%
Blood	2,0	2,0%
Liver	2,5%	2,5%
Heart, lungs.	0,8%	0,8%
Intestines without the contents.	5,4%	5,3%
Intestines fat	0,5%	0,5%
Other waste	9,3%	9,2%
Total	100	100

Table 1 demonstrates that both rabbit breeds are distinguished by high yield capacity, while the yield capacity of the New Zealand white rabbit breed's slaughtered carcass is greater than the yield capacity of the Californian rabbit breed's slaughtered carcass. Compared to other parts, the yield capacity jacket material - leather is high (8,3%). The total number of sub-products amounted to 16,4 - 15.8%. Still, it should be mentioned that the processing of rabbit meat is near zero-waste, because sub-products obtained as a result slaughtering (except the liver, which is deemed a valuable nutrient), blood and the intestinal contents are used for producing protein supplement (in the form of flour) for the formula feed.

The results obtained are in compliance with the similar data available in the literature on rabbit breeds under study. At the same time, according to a

number of authors, the yield capacity of some of the parts obtained from the slaughtered carcass of other rabbit breeds (white Goliath, silver, chinchilla) differ from those that we obtained. In particular, the yield capacity of head among these breeds is 6,0-6,8%, skin's yield capacity is 13,3-13,4%, blood's yield capacity - 2,0-2,1%, liver's yield capacity - 3, 2-3.5%, intestine's yield capacity (without the contents) - 6,2-6,3%, other waste's yield capacity - 9,8-10,8%. There is also a significant difference in yield capacities of the slaughtered carcass, which according to Vasilenko vary between 52,9 and 55.9% [4]. This can be explained by the breed characteristics of rabbit.

For further studies, the slaughtered carcass of rabbit breeds under study was divided into four anatomical parts: coxofemoral, lumbosacral, scapulo-humeral, and cervicothoracic. Their yield capacities are given in Table 2.

Table 2. *Yield capacities of anatomical parts of the rabbit's slaughtered carcass*

Indicator	Rabbit breeds	
	Californian	New Zealand white
Carcass mass, g	1920±21,5	2119±20,8
Coxofemoral part, g	679,7± 5,6	745,9±4,8
Yield capacity of coxofemoral part, %	35,4	35,2
Lumbosacral part, g	485,8±2,3	542,5±2,7
Yield capacity of lumbosacral part, %	25,3	25,6
Scapulohumeral part, g	472,3±3,7	506,4±3,6
Yield capacity of scapulohumeral part, %	24,6	23,9
Cervicothoracic part, g	282,2±3,4	324,2±3,9
Yield capacity of cervicothoracic part, %	14,7	15,3

An analysis of the data contained in Table shows that with yield capacity of anatomical parts, the rabbit breeds under study do not differ significantly from each other.

In both studied rabbit breeds, coxofemoral part is distinguished by highest yield capacity, yield capacities of cervicothoracic part is lower by 58.5

and 56.5% (by breeds), 30.5 and 32.1% lower is yield capacities of scapulo humeral part and 28.5 and 27.3% lower is yield capacities of lumbosacral part.

The results of studying the morphological composition of individual parts of these rabbit breeds are shown in Fig. 1 and 2.

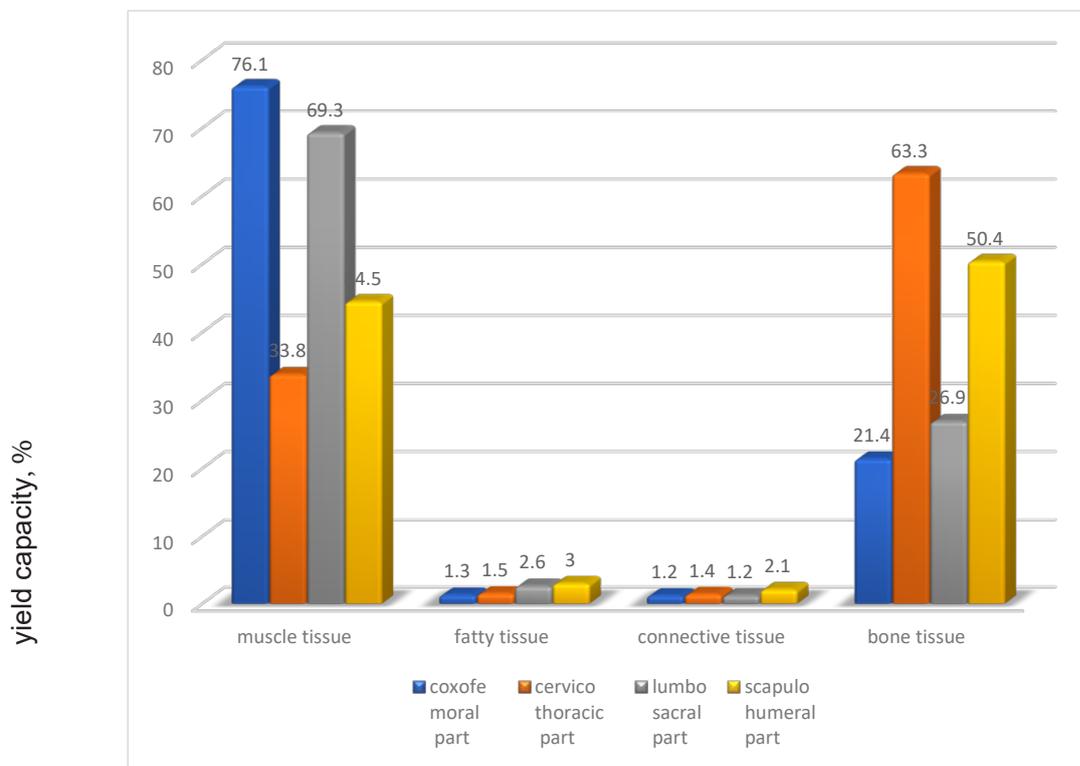


Fig. 1. Morphological composition of the Californian rabbit's slaughtered carcass

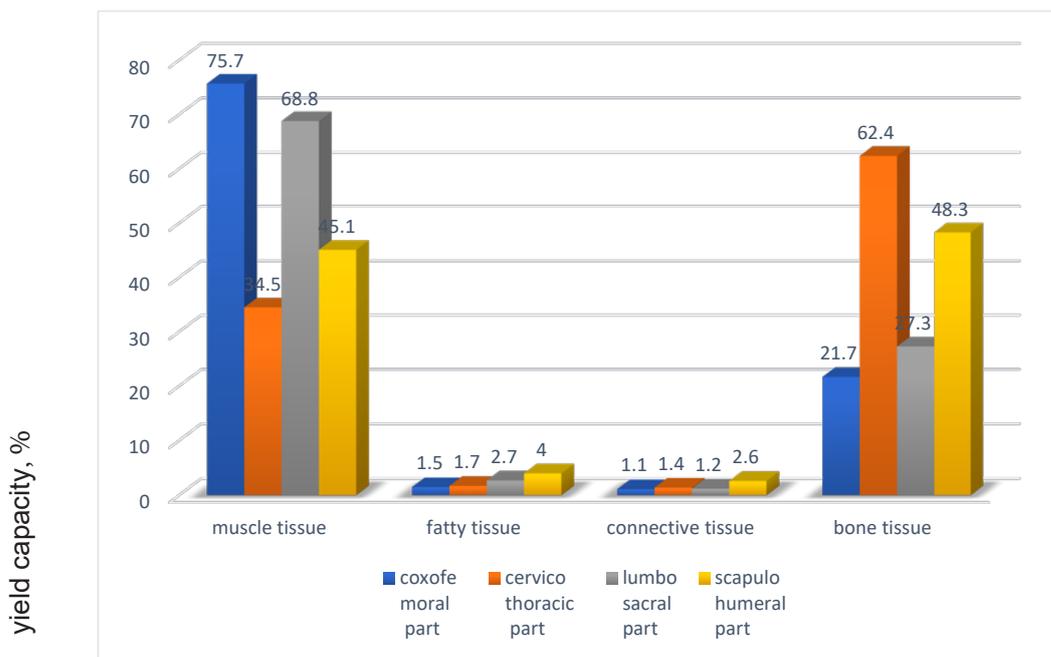


Fig. 2. Morphological composition of the New Zealand white rabbit's slaughtered carcass

Fig. 1 and 2 show that coxofemoral part contains the highest amount of muscle tissue (76.1% and 75.7%, by breeds), the lowest amount of bone tissue (21.4% and 21.7%) and the minimum amount of fatty (1.3% and 1.5%) and connective tissues (1.2% and 1.1%). The highest fatty tissue content is in scapulohumeral part. The percentage of muscle tissue in cervicothoracic part is 42.3 and 41.2 lower than in coxofemoral part, and 31.6 and 30.6% lower than coxofemoral part. The highest bone tissue mass content is observed in cervicothoracic part, and its content is 12.5 and 14.1% lower in scapulohumeral part, and the lowest content is observed in coxofemoral part (21.4% and 21.7%, accordingly).

It is noteworthy that meat muscle tissue differs from other tissues in a significantly higher nutritional value. Its content in coxofemoral part of the rabbit's slaughtered carcass, and the data we obtained earlier in the whole slaughtered carcass on muscle tissue (75.45% and 76.75%) [19], as well as similar data available in the literature [4,5,15,22, 36] demonstrate that the muscle tissue content in the rabbit's slaughtered carcass considerably exceeds its content in the slaughtered carcasses of other animals. In particular, the muscle tissue content in the slaughtered carcass of cattle is 57-62%, in the sheep's slaughtered carcass- 50-60%, in the pig's slaughtered carcass - 40-52%, and in broiler poultry - 51-53% [5]. Thus, the rabbit meat is more complete, soft and gentle.

An analysis of Figures 1 and 2 also shows that the meat of the front part of the rabbit's slaughtered carcass contains more quantities of connective tissue (4.5-6.6%) than its back part (2.3-2.4%). This indicates that the meat of the front part of the slaughtered carcass is more hard than the back part's meat.

Based on the obtained data, it is possible to choose those technologies for the rational use of anatomical parts of the different rabbit breeds, which contribute to maintaining their useful properties as long as possible.

It is believed that the most objective quality indicator of meat and its individual anatomical parts is the "meatiness index", which implies the quantitative ratio of muscle and bone tissues in the slaughtered carcass.

Fig. 3 illustrates the meatiness index of anatomical parts of meat of rabbit breeds under study.

As shown in Fig. 3, anatomical parts of the rabbit meat, according to the meatiness index, can be arranged in the following sequence: coxofemoral part, lumbosacral part, scapulohumeral part and cervicothoracic part.

When studying the technological and physico-chemical properties of meat, we determined their significant indicators, such as pH medium, reaction of peroxidase, test for primary products of decay of proteins in the broth, amount of volatile fatty acids and amount of amino-ammoniac nitrogen (Table 3).

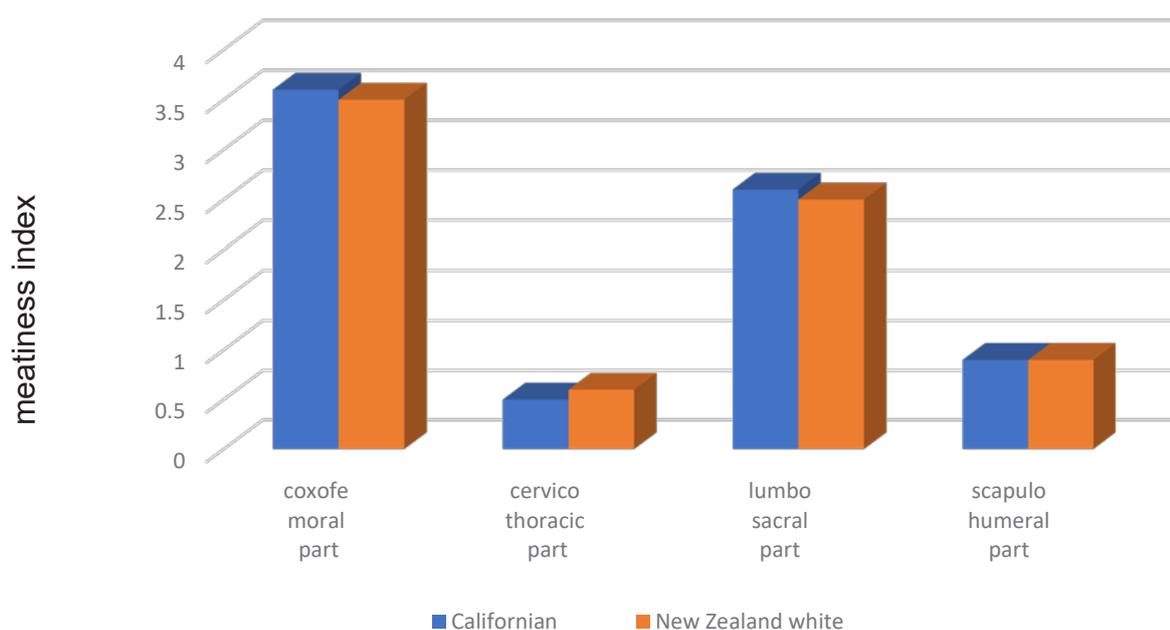


Fig. 3. The meatiness index of anatomical parts of meat of different rabbit breeds

Table 3. Physical-chemical indicators of rabbit meat

Indicators	Acceptable norm in frozen meat	Rabbit breeds	
		Californian	New Zealand white
pH	5,7- 6,2	6,03±0,31	5,87±0,29.
Reaction of peroxidase	Positive*	Positive	Positive
Test for primary products of decay of proteins in the broth (reaction with a 5%-solution of CuSO ₄)	Negative*	Negative	Negative
Amount of volatile fatty acids (mg/KOH)	≤2,25*	2,09±0,12	2,14±0,10
Amount of amino-ammoniac nitrogen (%)	1,4-2,0	1,65±0,08	1,52±0,03

An analysis of Table indicates that pH value determining the stability of meat relative to decomposing microflora in experimental samples does not exceed the norms and standards. In addition, the amount of fatty acids that determine the freshness of meat and the amino-ammoniac nitrogen content defining the goodness of meat are also within the limits of the standards, the test for primary products of decay of proteins in the broth was negative, but the reaction of peroxidase was positive, indicating that the meat was produced by healthy animal, since the enzyme peroxidase was active in all samples of meat. The extracted meat, after adding the Nessler's reagent, remained transparent in all cases.

Thus, the physical-chemical indicators of the rabbit breeds meat under study meet the quality requirements of meat.

In today's difficult environmental conditions, food safety issue is of high relevance. Therefore, we identified the content of the toxic substances in the meat of rabbit breeds under study (Table 4).

An analysis of Table shows that the contents of lead, cadmium, arsenic and mercury in different rabbit breeds do not exceed acceptable standards that indicate their safety, as well as their sanitary and hygienic reliability.

Conclusion

1. With a view to enhancing the use of products produced from rabbit meat, there have been studied the share of products obtained as a result of its slaughtering, yield capacity of anatomical parts and morphological composition of the rabbit's slaughtered carcass.
2. It has been established that the highest amounts of muscle tissue (76.1% and 75.7%) are contained in coxofemoral part and lumbosacral parts (68,8-69,3) of breeds, and connective tissue – in cervicothoracic scapulohumeral parts. Based on the obtained data, it is possible to choose those technologies for the rational use of anatomical parts of the different rabbit breeds, which contribute to maintaining their useful properties.
3. Anatomical parts of the rabbit meat, according to the meatiness index, can be arranged in the following sequence: coxofemoral part (3,5-3,6), lumbosacral part (2,5-2,6), scapulohumeral part (0,9) and cervicothoracic part (0,5-0,6).
4. It has been established that the physical-chemical indicators of the rabbit breeds meat under study

Table 4. The content of toxic substances in rabbit meat

Indicators	Acceptable norm*, mg/kg	Rabbit breeds	
		Californian	New Zealand white
Lead	Up to 0,5	0,038±0,01	0,12-0,14 მგ/კგ.
Cadmium	Up to 0,05	не обн. <0,01	0,007-0,019 მგ/კგ.
Arsenic	Up to 0,1	не обн. <0,0025	
Mercury	Up to 0,03	0,011±0,005	

* SanPiN 2.3.2.1078-01 p. 59

meet the quality requirements of meat.

5. The content of toxic substances in the rabbit meat indicates their safety, as well as their sanitary and hygienic reliability.
6. The data on the morphological and technological characteristics and physical-chemical properties of the slaughtered carcass should be taken into account when developing technologies for producing from them dietetic therapeutic healthy foods.

Acknowledgments

This work was supported by Shota Rustaveli National Science Foundation of Georgia (SRNSFG) [grant FR 17 – 353 “Development of technologies for Producing and Storage of Healthy foods by Using Rabbit Meat”].

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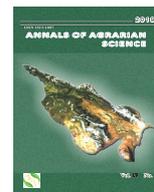
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Relationship of rainfall rate and radar reflective in east georgia

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Received: 15 March, accepted: 02 June 2019

ABSTRACT

Because the Country territory is prone to flash floods and mudflows, Quantitative Precipitation Estimation (QPE) and Quantitative Precipitation Forecast (QPF) on any leading time are very important for Georgia. Weather ground radar is the main tool for nowcasting and very short-range forecast. Two such radars operate in eastern Georgia. In this study, we presented calibration results one of them, which located in the Kakheti region. Precipitation estimation was compared to measured rainfall on 10 automated rain gauges within the radar coverage area. The calibration time step is an hour with the analysis period of 2017-18. We divided data into two subsets. The first set of 300 pairs with rainfall intensity from Georgian NHMS rain gauges network was used to calibrate the Z-R and to obtain parameters by minimizing RMSE and mean bias. The second set of 450 members was used for validation. The corresponding Z-R relationships for this region is $Z = 148R^{1.5}$. The minimization of mean root square errors between the rain gauge and the radar-derived measurements of rainfall shows significant improvement of radar estimated precipitation after the calibration of Z-R dependence.

Keywords: Weather radar, Reflectivity, Calibration, Correction, Rainfall rate, Nowcasting.

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1. Introduction

Georgia's orography and its interaction with airflows is the basic spotting factors of synoptic processes spread in the country. Peculiarities of local weather phenomena, which don't depend on the season are often characterized by diversity and extreme feature. The nowcasting (NWC) and very short-range forecast (VSRF) of the synoptic processes such as convective storms with attendant phenomena, fog and low clouds, locally forced precipitation events, wintertime weather (snow, ice, glazed frost, avalanches) have great importance for Georgia.

NWP model's predictability strength within the NWC range (0–6 h) is still relatively low. Valid NWC and very short range forecast require a high density of weather information, on the surface as well as radar and satellite observation and high-resolution local area model (LAM) output. NWC tech-

niques commonly use an extrapolation (with the heuristic rules) of the observations to make the forecast. The ground weather radar is the main tool for the nowcasting, especially for rainfall rate. Radars have the advantage in coverage of a large area for the real-time precipitation measurement but don't have a good enough accuracy for hydrological applications. Also, the advantages of weather radar are the ability to detect the rainfall and clouds as well as their structures in real-time [1-5]. Climate applications can use these data sets as well.

The very first weather radar in Georgia was "MRL" in the 1960 year, which was mainly used for anti-hail and air navigation system. There were running 10 "MRL" before the Soviet Union's collapse but they were stopped after it. National Environmental Agency hasn't its weather Radar today, but the three other organizations officially share their weather radar data to it. Scientific technical center "Delta" recovered the anti-hail system to

protect vineyards from hail in Kakheti region, using dual-polarization c-band “Selex ES” weather radar near the village Nukriani. The Georgian Air Navigation Service uses x-band dual-polarization radar from “EEC” installed near Tbilisi International Airport. Also Trabzon and Erzurum weather radars from Turkish State Meteorological Service which provide only composited pictures. These radars partially cover the country’s territory [6-9].

Weather Radar estimates volume averaged rainfall rate R , based on reflectivity factor Z measured in Decibels. Z depends on the size of the drops (to the sixth power) and their spatial distribution. R depends on the raindrop size distribution, the size of the drops (to the third power), and the fall velocity for a given drop diameter [10-16]. Marshall et al. (1947) reported a good correlation between reflectivity (Z) and rainfall rate (R) first time. Marshall-Palmer (1948) contained the results of research relating the size distribution of raindrops as a function of rainfall rate. From this relationship, the famous Marshall and Palmer relationship of $Z = 200 R^{1.6}$ was derived (where 200 and 1.6 are empirical constants [17-21]). Because of these different relationships, the Z - R relationship is not unique, as there can be different rainfall rates for a particular reflectivity, and vice-versa.

Rain gauges are used to validate and calibrate weather radar precipitation data, as rain gauges are recognized to be the most reliable source of rainfall data. Several disadvantages should be taken into account during the comparison: 1. rain gauges give almost approximate values of point rainfall rate when it compared with gridded precipitation field from radar; 2. The values of the cloud may look stronger than observed precipitation on the ground, especially in spring and summertime, when there are convective clouds, with strong wind gusts and evaporation goes up and holds rain particles in the air and could not fall; 3. The radar beam is usually not uniformly fell with precipitation; 4. Even if the rain measurements are below the cloud base the readings affect by drifting of low-level winds, and also the time delay between radar and rain gauges causes a further discrepancy between the two measurements [22-25].

New techniques and methodologies have been made to improve the accuracy of radar estimations and measurements over time. The international scientific literature on merging rain gauge measurements and radar estimations is vast and, a few useful reviews have been published recently [26-29].

In this paper, we present the relationship between reflectivity factor (Z) empirically derived from C-band weather radar located in Kakheti and precipitation amounts (G) from 10 rain gauges within the radar coverage area, as well as the validation results reflected in the several statistical parameters.

2. Materials and methods

2.1. Weather radar data

We used archived reflectivity data of the C-band radar for selected rainy episodes in the 2017-18 period. The radar location and its coverage area are presented in Figure 1. and the characteristics of the radar in Table 1. The radar performs Volume and Azimuth scan, moving circularly, changing elevation 17 times during 3 minutes.

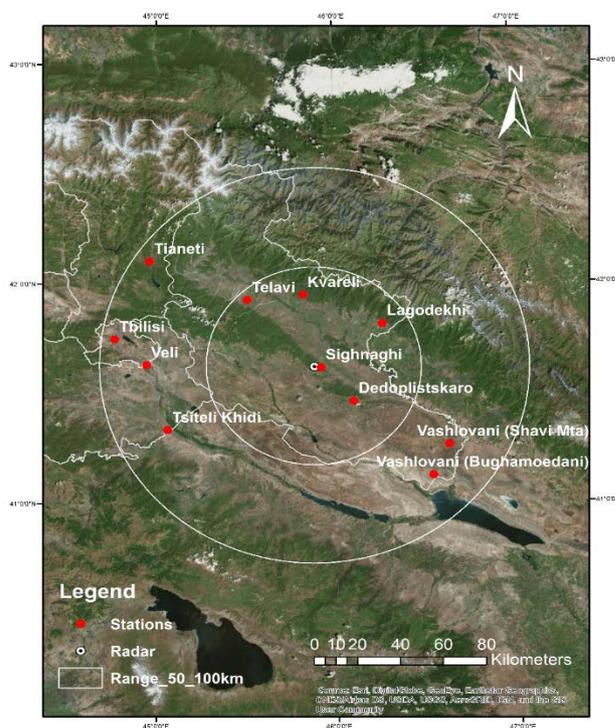


Fig. 1. Locations of Radar and Rain gauges

With its short wavelength (5.4 CM) and high frequency (4-8 GHz), C-band radar can easily identify intensity of clouds and show in decibels, from weak to extreme and dual-polarization is an advantage to classify type of precipitation. This is the general purpose of this system, but to predict quantity of precipitation per hour, products with its algorithms inside weather radar are more or less useful, these data needs to be more accurate and close to the real ground rainfall, measured by rain gauge and needs to find correct way of calculation [30-31].

The 6 minute interval of volume scan data with 10 elevation angles is used. These volume data are one of the radar collections that record the reflectivity and their properties of the precipitation in the sky.

Weather radar systems. The quality control of reflectivity was performed as ground clutter and noisy signal were removed from the measured reflectivity data. The reflectivity overlapped to the other adjacent radars should be checked and it reveals almost similar in both reflectivity shape and dbz. Hence, there is only reliable radar data were used in the analysis.

Reflectivity data that were greater than 53 dbz were limited to mitigate contamination from hail [32-34]. Additionally, the reflectivity less than 15 dbz cases were also excluded from the analysis in order to avoid the effect of noise in the measured radar reflectivity.

2.1. Rainfall data collection

Rain gauges are observational instruments capable of accurately and directly measuring near-surface rainfall. However, these instruments do not provide rainfall information at high spatial resolutions, and observational errors may be introduced into their measurements by strong ground winds or calibration errors. Particularly, in the case of tipping-bucket rain gauges (widely used for automatic rainfall observations globally due to easy digitization of the signals), rainfall data may vary significantly depending on whether the center or the edge of the rain cell passes over the rain gauge.

Hourly rainfall data from 10 automatic rain gauge stations located in eastern part of Georgia and fall in radar coverage area was collected from

GNHS. The location of study areas and these automatic rain gauges is illustrated in Figure 2. The quality control of gauge Rainfall data series was performed by comparing rainfall data of the considered rain gauge with rainfall data of the nearby stations. Rainfall data of the considered rain gauge should be consistent with rainfall data of the adjacent stations. All of the automatic rain gauges are of tipping bucket type with 0.245 mm accuracy. The hourly rainfall intensity of each rain gauge station from the selected rainfall event was used to calibration using average hourly reflectivity values in the gridded radar data.

2.1. Method

As it was described above, Reflectivity factor measured by radar for 2017-18 period was taken, values less than 5 and more 53 DBZ was removed and converted into Rainfall rate using default $Z-R$ relationship ($Z=200R^{1.6}$) formula. An average hourly reflectivity was obtained from 6 min radar readings, to make dataset in the same temporal interval (hourly) as corresponding amounts from 10 rain gauges in the coincident pixels, at 1 km resolution. Totally, 750 couple of rainguge – radar data was fitted (from 87 events). Mean bias, correlation and RMSE were calculated.

For calibration precipitation derived from radar, the ‘a’ and ‘b’ parameters from the $Z-R$ relationship $Z=aR^b$ will be corrected by minimizing the Root Mean Square Error (RMSE) [35-37] and mean bias (ratio G/Z) between radar and corresponding rain gauge rainfall.

The constant of ‘b’ parameter equal to 1.5 was taken, due to several studies explaining that variation of ‘b’ parameter did not affect the RMSE between radar and rain gauge rainfall much [38-42].

Table 1. Radar Characteristics

Detail of radar	Characteristics
Type of radar	Doppler, dual polarization METEOR 750C
Radar band	C band
Radar frequency (MHz)	3000
Wave length (cm)	5.4
Beam with (degree)	0.95°
Pulse length (μs)	2
Resolution of recorded data in Cartesian coordinates (m)	400X400
Gate width (m)	250
Maximum transmission power (kW)	250
Maximum Range (km)	500
Sequence of elevation angles (10 angles)	0.5°,1.7°,3.2°,4.9°,7.1°,10.2°,14.6°,21.5°,33.2°, 60°

2.1. Results and Discussion

87 rainy episodes have been selected, with different rain intensity released within one or several days, due to different synoptic processes. It worth to be mentioned, that radar covers the part of the country with small yearly precipitation sums, but characterizing with intensive hourly and daily rainfall rate, especially in spring and summer, with high probability of hail. As we rejected DBZ values above 53, the events with hail are not considered in our study.

We selected 750 radar-rainfall matched pares of hourly (mm/h) for above-mentioned 10 raingauges satisfying our criteria. Dataset was divided into 2 parts 300 from them was used for minimalizing RMSE during calibration and rest of them for verification. Distribution of the full set of rainfall data is following: 80% of them are below 6 mm/h, approximately 15% is in the range of 7-25 mm/h and only 1% is above 30 mm/h. The highest observed value

was 36.6 mm/h (Veli 18.06.2018 18:00) Radar estimated precipitation by default $Z-R$ relationship ($Z = 200R^{1.6}$) was 41 DBZ. In the table 2. Correlation coefficients and RMSE are given for the individual raingauges for full dataset, when radar estimated precipitation is calculated for $a=200$ and $b=1.6$ default values.

By minimizing RMSE from 7.521 to 6.163 in train data sample $a=148$ was derived empirically, while bias (G/Z) was reduced by 0.66 and correlation coefficient increases by 0.1. In the table.B.3. all statistical parameters, for calibration and verification data with default “a”, “b” constants, and empirically derived ones are presented.

The verification results prove that the radar rainfall estimated with the new coefficients are in much better agreement with the rain gauges data.

As it is presented on the Fig. 2. In calibration dataset linear regression’s slope is 0.985, and in verification data 1.017.

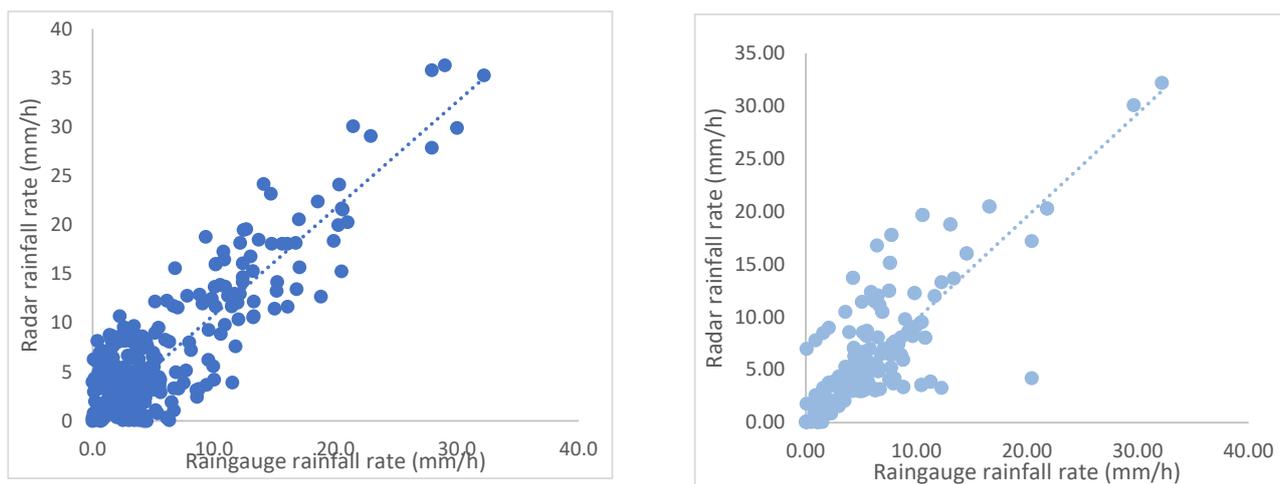


Fig. 2. Scatter plot between measured and estimated rainfall rate for $Z=148R^{1.5}$ for (left) calibration and (right) validation dataset.

Table 2. Correlation between hourly precipitation measured by raingauges and estimated by Radar using default $Z-R$ relationship ($Z = 200R^{1.6}$).

Rain gauges	Distance from radar (Km)	X Coordinate	Y Coordinate	RMSE	correlation
Bugha Moedani	78.7	629157.16	4553858.75	7.824	0.84
Dedoplistskaro	25.5	592031.41	4591078.94	7.783	0.96
Veli	78.6	495704.68	4608946.52	7.872	0.80
Tbilisi	93.7	480824.98	4622050.1	8.411	0.81
Tianeti	93	496996.82	4661412.79	7.974	0.70
Lagodekhi	38.7	605184.72	4630392.67	8.885	0.78
Sighnaghi	4.3	576778.71	4607994.60	8.329	0.68
Kvareli	36.5	568293.40	4644663.02	8.977	0.56
Vashlovani (Shavi Mta)	75.2	636590.77	4569408.07	7.541	0.83
Tsiteli Khidi	75.8	505473.1491	4576150.437	9.084	0.47

Table 3. RMSEs, Bias and correlations obtained from Z-R relationships using empirically derived and default parameters.

	Z=148R ^{1.5}			Z=200R ^{1.6}		
	RMSE	Bias (G/Z)	correl	RMSE	Bias (G/Z)	correl
calibration	6.163	0.985	0.95	7.521	2.01	0.857
verification	5.979	1.017	0.91	6.984	2.12	0.843337

1. Conclusion

The Z-R relationships were derived from statistical measurements of precipitation during the 2017-18 period in eastern Georgia, varying by parameter “a”, while keeping parameter “b” fixed at 1.5. 750 Z-R pairs were obtained from 87 precipitation cases, and then dataset was divided into two groups. The first set of 300 pairs with rainfall intensity from the Georgia’s NHMS raingauges network was used to calibrate the Z-R and the obtained parameters. The second set of 450 events was recognized as validation data. The relationship between point rainfall from the rain network and radar rainfall at a certain height of 1.5 km was calibrated by minimizing RMSE. The corresponding Z-R relationships for this region is $Z = 148R^{1.5}$. The result shows significant improvement with improved Z-R dependence in obtaining minimized errors from more accurate estimated rainfall. These results will provide to assess for evaluating of rainfall estimation for the sectors where accurate rainfall information is essential.

Acknowledgment

The research was funded by the Shota Rustaveli National Science Foundation (SRNSF) of Georgia (grant FR17_529).

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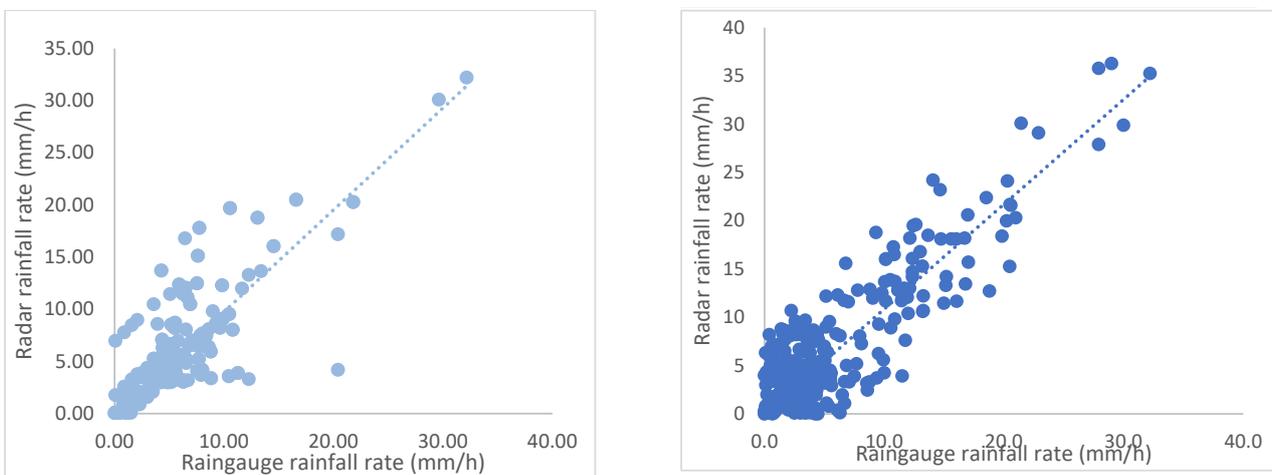


Fig. 2. Scatter plot between measured and estimated rainfall rate for $Z=148R^{1.5}$ for (left) calibration and (right) validation dataset.

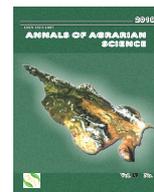
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Background error covariance in numeral weather production

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Received: 29 March 2019; accepted: 15 May 2019

A B S T R A C T

Improving of weather forecast quality is a continuous work, as it is an invaluable for society and environment. WRF model have been tuned and tested over Georgia's territory for years. Nowadays as local meteorological network became denser and many remote observational sources are available data assimilation with variational methods is current challenge. First time in Georgia the process of data assimilation in Numerical weather prediction is developing, the way for forecast initial conditions' correction. Assessment of the forecast error is one of the first and most important steps in data assimilation. This work presents how forecast error statistics appear in the data assimilation problem through the background error covariance matrix – B, where the variances and correlations associated with model forecasts are estimated. Statistics of B are usually determined for a limited set of variables, called control variables that minimize the error covariance between variables. Results of generation and tuning of background error covariance matrix for five control variables using WRF model over Georgia with desired domain configuration are discussed and presented. The mathematical and physical properties of the covariances are also reviewed.

Keywords: Weather forecast, Numerical Weather Prediction, Variational assimilation, Background errors statistics, Analysis increment, Distribution function.

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Introduction

Weather forecast accuracy very demanding on initial conditions, as small changes in initial conditions can lead to large changes in prediction. Variational data assimilation (VAR) is the method to estimate the initial state of the atmosphere for weather prediction to improve forecast quality [1, 2]. VAR usually combining measurements and models takes a forecast (also known as the first guess, or background information) and applies a correction to the forecast based on a set of observed data and estimated errors that are present in both the observations and the forecast itself. The forecast error is represented as background error covariance matrix (B). The specification of background error statistics is a key component of data assimilation since it affects the impact observations will have on the analyses.

In the variational data assimilation approach, applied in geophysical sciences, the dimensions of the background error covariance matrix (B) are usually too large to be explicitly determined and B needs to be modeled. Statistics of the background error covariance matrix B are usually determined for a limited set of variables, called control variables that minimize the error covariance between variables. Then, several parameters need to be diagnosed to drive the series of operators that model B. There are now many leading centres around the world- European Centre for Medium-Range Weather Forecast (ECMWF), the National Centers for Environmental Prediction (NCEP), or the UK Met office etc. that use VAR for weather forecasting, and there are often differences in the way that forecast error statistics are measured, described and used by each [3-5]. In this paper, we present background covariance

matrix' properties generated for WRF-ARW model with GEN_BE code over South Caucasus domain and testing results within the two assimilation systems GSI and WRFDA. Originally, the GEN_BE code was developed [6] as a component of a three dimensional variational data assimilation (3DVAR) method to estimate the background error of MM5 for a limited-area system. Since this initial version, various branches of code have been developed at NCAR and at the UK Met Office to address specific needs using different models such as (WRF) and the Unified Model (UM) on different data assimilation platforms such as the Weather Research Forecast Data Assimilation system (WRFDA) and the Grid point Statistical Interpolation system [7, 8]. The first section of this document describes the role of the background error covariance matrix B, difficulties and opportunities of its estimation. The second one presents general structure of GEN_BE code version 2.0. with some technical details in our application and provides results of pseudo observation case in two different systems of data assimilation (WRFDA and GSI) using different B matrix involving the same set of five control variables (CV5). All the results presented in these papers have been obtained from WRF model output for 9.2 km resolution domain configured and tuned over South Caucasus domain (Fig.1).

Methods and materials

2.1 Background error covariance matrix and initial state of atmosphere

The objective of VAR is a cost function $J(\delta x, x^g)$ minimization. This objective function is a combination of forecast and observation deviations from the desired analysis, weighted by forecast and observation-error covariance matrices.

$$J(\delta x, x^g) = 1/2(\delta x^b - \delta x)^T B (\delta x^b - \delta x) + 1/2[y_o - H(x^g + \delta x)]^T R^{-1} [y_o - H(x^g + \delta x)] \quad (\text{Eq.A.1})$$

Where x is the state vector composed of the model variables (e.g. winds, pressure, temperature, humidity, etc.) to analyses, at every grid point Dist of the 3-dimensional (3-D) model computational grid [6].

δx is difference between the analysis x^a and reference state or the 'first guess' x^g , i.e.

$$x^a = x^g + \delta x \quad (\text{Eq.A.2})$$

y_o is the vector of observations and H called the observation operator, is a mapper from the gridded

model variables to the irregularly distributed observation locations. R is the observational error covariance matrix. B is the background error covariance matrix. The background error covariance matrix describes the probability distribution function (PDF) of forecast errors. Theoretically exact knowledge of R and B would require the knowledge of the true state of the atmosphere at all times and everywhere on the model computational grid, what is not possible. Therefore, both matrices have to be estimated in practice. Dimension of the B matrix is the square of the 3-D model grid multiplied by the number of analyzed variables. For typical geophysical applications as in meteorology, the size of the B matrix, comprised of nearly $10^7 \times 10^7$ entries, is too large to be calculate explicitly nor be stored in present computer memories. As a result, the B matrix needs to be parameterized [9, 10].

2.2 Background errors covariance matrix modeling.

The cost function as defined in Eq. (A.1) is usually minimized after applying the change of a variable:

$$\delta x = B^{1/2} u \quad (\text{Eq.A.3})$$

$B^{1/2}$ is the square root of the background error covariance matrix. The variable u is called the control variable and the cost function becomes:

$$J(u) = 1/2 u^T U u + 1/2 (d - H B^{1/2} u)^T R^{-1} (d - H B^{1/2} u) \quad (\text{Eq.A.4})$$

Where d is the innovation vector defined as $d = (y_o - H(x^b))$ and it represents the difference between observations and their modeled values using a non-linear observation operator.

The square root of the B matrix as defined in Eq. (A3) is decomposed to a series of sub-matrices, each corresponding to an elemental transform that can be individually modeled:

$$U = S U_p U_v U_h \quad (\text{Eq.A.5})$$

Where, S diagonal matrix and composed of the standard deviations of the background errors.

U_p matrix - Physical Transform - defines the cross-correlations between different analysis Variables via statistical balance (linear).

U_h - Horizontal Transform - defines the horizontal auto-correlations for the control variables. It is modeled through successive applications of recursive filters [11],

The matrix U_v defines the vertical auto-correlations for each of the control variables [12].

If the EOF (Empirical Orthogonal Function) decomposition is used, the eigenvectors model the vertical transform (U_v) and the associated eigenvalues represent the variance. The length scale is estimated in the EOF space and represents the horizontal transform (U_h). In the data assimilation process, the eigenvalues weight the analysis increment and the recursive filter first spreads out the information in the EOF space according to length scale value. Then, the transformation from EOF mode to physical space spreads out the information vertically.

Calculations and results

For this study WRF-ARW model over the 9.2 km domain (Fig. B.1) with 151 x 100 x 36 grid cells have been used.

Background error covariance matrix **B** was generated using GEN_BE code version 2.0 in WRFDA. The code comprises from 5 stages, having separate input output infrastructure and managed via name list file, where control variables and all parameters to model B are defined by user.

Since the background error covariance matrix is a statistical entity, samples of model forecasts are required to estimate the associated variances and correlations of desired variables. The input data for gen_be are WRF forecasts, which are used to generate model perturbations, used as a proxy for estimates of forecast error.

NMC (named for the National Meteorological Center) method [13] was used to represent a sample of model background errors, where differences between two forecasts valid at the same time but initiated at different dates (time lagged forecast, e.g. 24-minus 12 h forecasts) was taken. This is done for many different dates to build up a large sample size for calculating statistics. Climatological estimates of background error may then be obtained by averaging these forecast differences over a period of time (e.g. one month).

For this run, February 2018 12 and 24-hour WRF-ARW forecasts, initialized both at 00 and at 12 UTC was used. Thus in all 60 pairs of perturbations are utilized to generate WRF-ARW Background Error.

On the initial stage analyses control variables stream function (ψ) and unbalanced velocity potential (χ_u) are calculated from u and v wind, then

differences for following 5 control variables: stream function (ψ), unbalanced velocity potential (χ_u), Temperature (T), Relative Humidity (q), Surface Pressure (ps) have been created. On the next stage statistics are calculated, such as mean from differences, created on the initial stage, then performs perturbation for each control variable and computes covariance of the respective fields [14].

On the stage 3 regression coefficient & balanced part of χ , T and p_s variables computed. The estimation error for one analysis variable may affect the value of another if they are correlated. The simplest way to model them is to use linear regression. Firstly, the regression coefficients between variables calculated, then, linear regressions are performed to derive uncorrelated control variables and then remove the balanced part for each other variable. This part achieves the Up transform: it models correlations between variables and allows transforming the matrix as a diagonal bloc in the control (uncorrelated) space. Computes unbalanced parts for the same variables:

$\chi_u' = \chi' - \chi_b$; $T_u' = T' - T_b$; $p_{s,u}' = p_s' - p_{s,b}$ is the preliminary step before estimating the vertical and horizontal auto-correlation parameters for each control variable.

Stage 4 Removes mean for χ_u' , T_u' & $p_{s,u}'$ and computes eigenvectors and eigen values for vertical error covariance matrix of ψ , T_u' , χ_u' and q fields, variance of $p_{s,u}'$ and eigen decomposition of ψ' , χ_u' , T_u' and q fields.

On the last stage “lengthscale (s)” calculated for each variable and each eigen mode.

Bellow on the fig. B. 2 some properties of B matrix displayed. Namely Fig.B.2. a) (left panel) represents the first five eigenvectors of psi –Stream function, chi_u, -unbalanced part of velocity potential, t_u, - unbalanced part temperature and rh-relative humidity variables. The eigenvectors are the results of EOF decomposition of the vertical auto covariance matrix and define vertical transform. On the Fig.B.2. b) horizontal length scales are shown for the same 4 variables.

The stream function and the potential velocity have the largest length scale value reaching 160 km and 120 km correspondingly. While, the unbalanced temperature length scale has a strong variation for the three first EOF passing approximately from 5 to 15 vertical modes and from there decreases from 40km to reach 10 km for the last EOF mode.

As the domain specific forecast error statistics computed, for diagnose and visualize B matrix prop-

erties is a good chose to run a single observation test, where only one (pseudo) observation is assimilated from a specific time and location within the analysis domain. In this case in analysis equation:

$$x^a = x^b + BH^T(HBH^T + R)^{-1}[y^o - H(x^b)] \quad (\text{Eq.A.6})$$

it's assumed that for any control variable $[y^o - H(x^b)] = 1.0$; $R = I$. Thus, $x^a - x^b = B^*$ constant delta vector and only B matrix is corresponding on spread of increments in the point across the domain horizontally and vertically. In addition, how it affects the other variables.

We design our single observation experiment in this way: temperature was increased with 1 Kelvin in the center of the domain on the 500-hpa height. Two variational data assimilation systems WRFDA with WRF-ARW domain specific background errors and GSI with NAM regional background errors have been used. For GSI we performed two runs with B_{nam} matrix. One of them was without tuning (lengthscale and variance options were set to 1) and with tuning (hzscl= 0.373, 0.746, 1.50). Background Forecast files have been similarly defined in all cases.

Fig. 3 shows analyses innovation for T, U and V variables for above-mentioned three runs. Fig.B.3. a) – corresponds to the results from GSI with regional B_{nam} (without tuning), Fig.B.3. b) – GSI with regional B_{nam} (with tuning), Fig.B.3. c) - WRFDA with our B. Each part of Fig.B.3 (a,b,c) shows two panels together left side - horizontal (XY at 11th sigma level) cross-sections of above mentioned three variables and right part - vertical cross-sections (XZ).

The first row on all figures show how temperature increment in the domain center spreaded horizontally and vertically. From figure 3a to 3b area where increment affects surrounded area reduces due to tuning length scale and variance parameters. On the figure 3c the affected area more concentrated in the center and more reduced.

Thus, the temperature Perturbation area produced from GSI recursive filter is larger than from WRFDA produced one with EOF mode. On the vertical cross-sections XZ, the temperature innovation has a larger impact on the vertical using our B than B_{nam} . These differences come from the dataset used to model these B matrices, the statistics in B_{nam} are more climatological as they are averaged over time and they are interpolated on the mesh grid of our domain during the data assimilation process. While our B constructed from 2-month data set.

The second and third rows of figure 3 show how wind U and V components response on temperature perturbation. Horizontal and vertical cross-sections of this parameters show similar features.

To validate B matrix within both assimilation systems the single observation tests' result are realistic and very close to each other with expected differences.

Conclusions

WRF model is the main tool for weather forecast in Georgia. The model have been tuned and tested over Georgia's territory for years. Nowadays as local meteorological network became denser and many remote observational sources are available to assimilate with variational methods is current challenge. We are working with two variational assimilation platforms suitable for this model namely WRFDA and GSI.

To estimate model forecast error in variational assimilation system, background error covariance matrix B, was successfully modeled and validated for Georgia's territory. To model B matrix GEN_BE v2.0 code has been used where model univariate or multivariate covariance errors from five control variables was taken as an input. This code gathers some methods and options that can be easily applied to different model inputs and used on different data assimilation platforms.

Different stages and transforms that lead to the modeling of the background error covariance matrix B and testing results by performing single observation tests was described and shown in this paper. B matrix modeled for our domain was tested on WRFDA platform using the EOF decomposition and was compared with the similarly designed test results on GSI platform using the recursive filters to model the vertical transform. The test shows similar results with comprehensive differences for the set of five control variables.

Appendix B: Figures

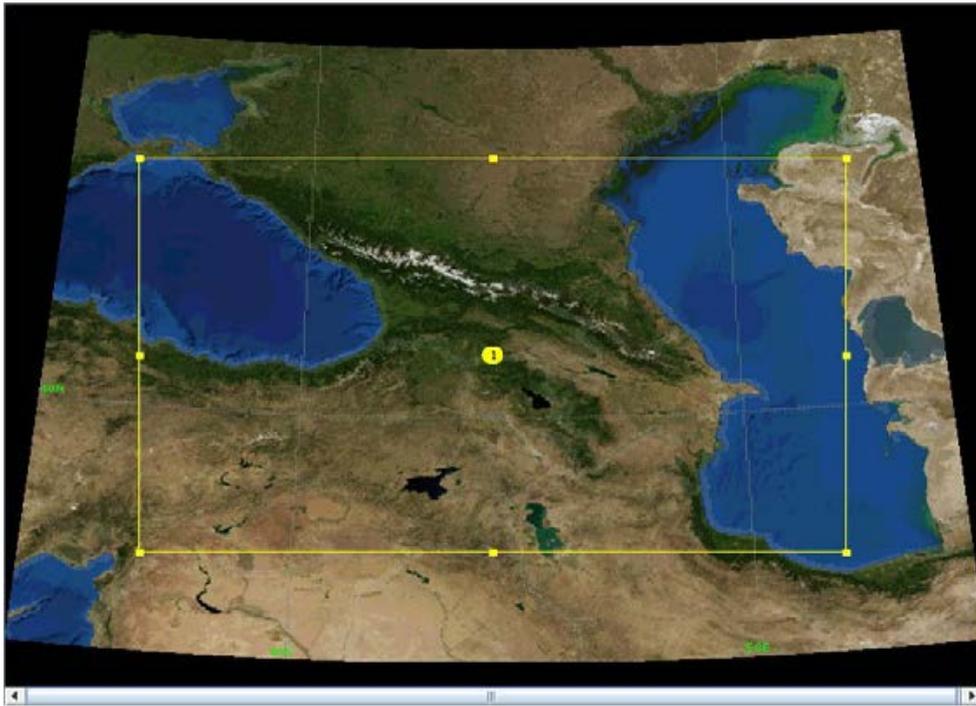


Fig. B.1. Extension of the WRF-ARW computational domain

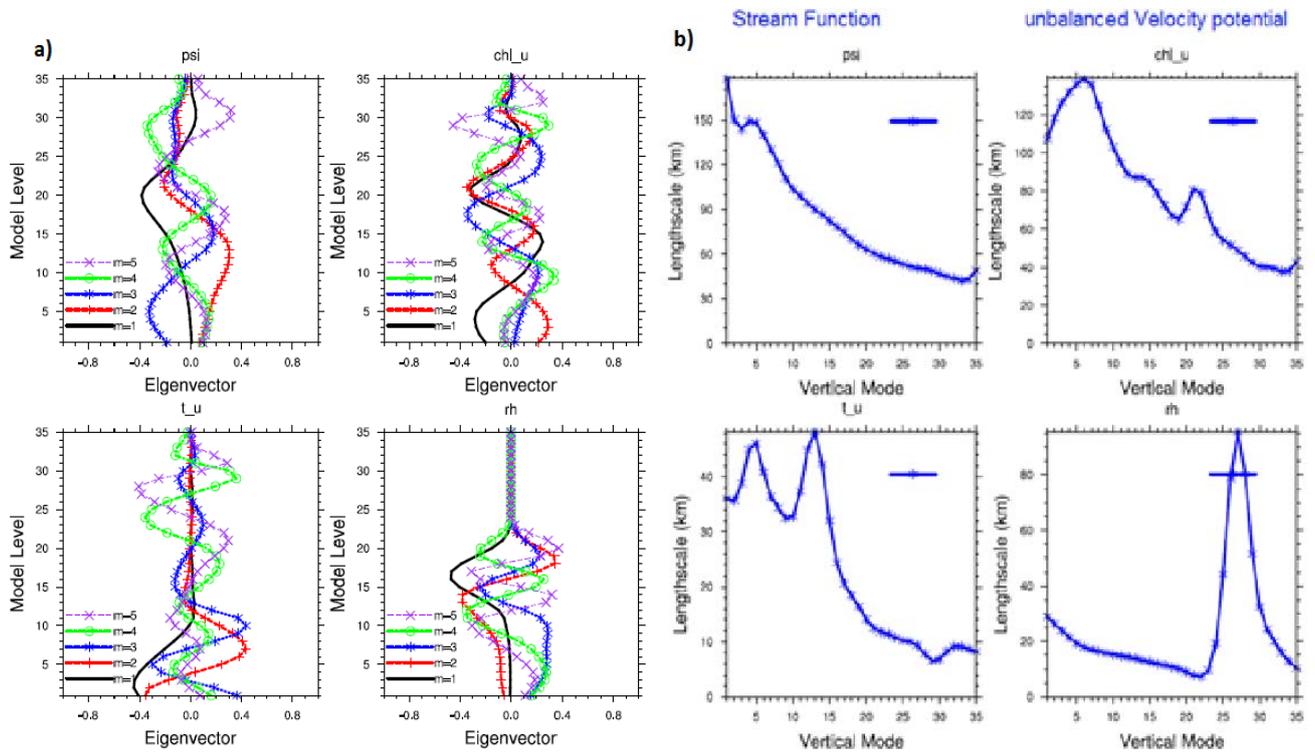


Fig. 2. a). Five eigenvectors of ψ – Stream function, chl_u , -unbalanced part of velocity potential, t_u , - unbalanced part temperature and rh -relative humidity variables; b) length scale factor for the same variables.

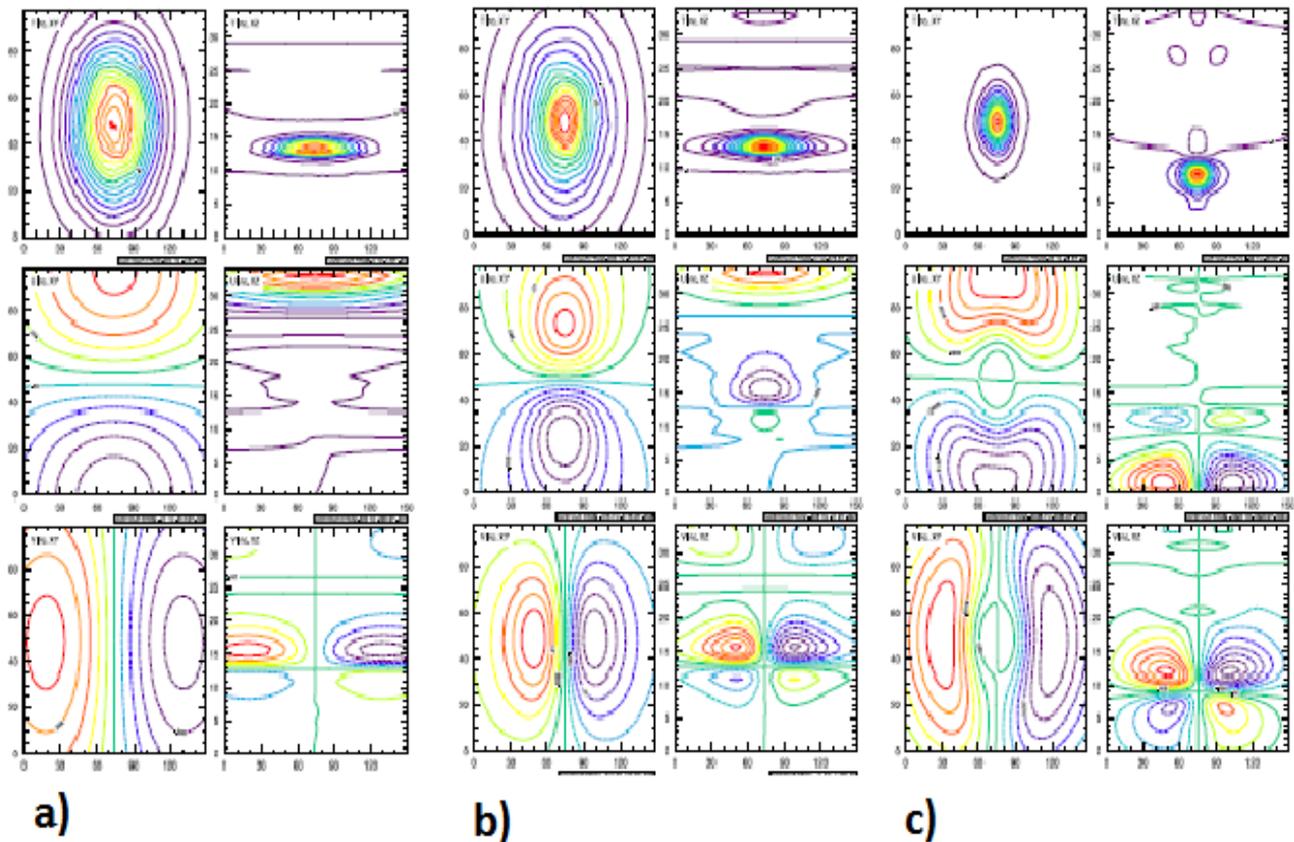


Fig. 2. a). Analyses innovation for T , U and V variables. 3a – results from GSI with regional B_{nam} (without tuning), 3b – GSI with regional B_{nam} (with tuning), 3c – WRFDA with our B . Each part of figure 3 (a,b,c) shows two panels together left side – horizontal (XY at 11th sigma level) cross-sections of above mentioned three variables and right part – vertical cross-sections (XZ).

Acknowledgment

The authors would like to thank to the Shota Rustaveli National Science Foundation (SRNSF) for the financial support (grant FR17_529).

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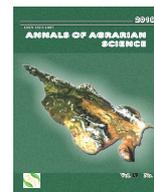
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Annals of Agrarian Science

Journal homepage: <http://journals-org.ge/index.php>



A liquid bio-flavanoid concentrate “Red aladasturi”

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Received: 23 February 2019; accepted: 06 May 2019

ABSTRACT

This paper dwells on the uvological characteristics of “Aladasturi” colored grape variety raw materials growing in the viticulture and winemaking zone of Imereti (Georgia), as well as biologically active compounds and antioxidant activity of hydrophilic extracts and liquid concentrates of its solid matters (stone and skin). The amount of phenolic compounds was determined using Folin Chocalteu reagent. The flavonoid, catechin and anthocyanin contents were determined by spectral methods, and antioxidant activity was determined by DPPH method. Research resulted in the establishment of the dynamics of changes in the biologically active compound content in test samples of “Aladasturi” variety growing in certain zones of Imereti region according to its growing areas. „The bio-flavanoid concentrate “Red Aladasturi” containing 64-75% of solid matters, which was thickened by vacuum-sublimation method constitutes a composition of the following components: 33% grape-stone ethanol extract, 33% grape-stone super-fluid extract and 33% grape-skin hydrophilic extracts. It has been established that the bio-flavanoid liquid concentrate “Red Aladasturi” is strong antioxidant (55.31-57.45%), and one tablespoon or 8-9 ml of it contains 52-57 mg of flavanoids, which is 109-112% of a full day of rations per person per day.

Keywords: “Aladasturi”, Colored grapes, Phenolic compounds, Anthocyanins, Antioxidant activity, Flavanoids.

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Introduction

Flavanoids are the largest group of phenolic compounds, and owing to their high biological activity, they are often referred to as bioflavanoids. Deficiency of flavonoids in the human body manifests with the following symptoms: the general weakness and chronic fatigue, nasal hemorrhage, the reduction in immunity, recurrent colds and infections, the formation of hematomas and vesication, the reduction in vascular conductance and elasticity, pains in the upper and lower extremities during the movement, and so on [1-3].

There is an extensive literature on high antioxidant activity of bioflavpnoid-rich colored grape seed and skin hydrophilic extract and red and white wine produced from it, as well as on inactivation of free radcals [4, 5].

In 2011, the VITAL (Fred Hutchinson Cancer Research Center, Seattle, Washington) published

studies on prostate cancer, with 35 239 males aged 50-76 voluntarily taking part in the study. Patients who received systemic hydrophilic extracts of grape juice were found to be 41% less likely to have prostate cancer than those receiving other drugs such as chondroitin, coenzyme Q10, fish oil, ginseng, ginkgo biloba, garlic, and glucosamine and palmetto [6].

In 2011, the VITAL (Fred Hutchinson Cancer Research Center, Seattle, Washington) published a study on prostate cancer, and 35 239 men aged 50-76 volunteered for this study. It was found that patients regularly consuming grape-seed hydrophilic extracts were 41% less likely to suffer from prostate cancer than patients taking other drugs such as chondroitin, coenzyme Q10, fish oil, ginseng, ginkgo biloba, garlic, and glucosamine and palmetto [6].

The aim of the study was to investigate a polyphenolic complex and antioxidant activity of secondary resources remained after the initial processing of “Aladasturi” grape variety growing in Imereti

and different micro-zones, as well as to explore the possibilities of using them for the production of drastic, antioxidant polyphenolic concentrates, because the solid parts of colored grapes, with the content of biologically active compounds are the best raw materials for the production of therapeutic extracts and concentrates to treat various pathologies [7-9].

“Aladasturi” is a Georgian, aboriginal, red, late-ripening, industrial variety, mostly common in the viticulture and winemaking zones of Imereti and Guria. Grapes ripen in late October and early November, and in full maturity, sugar content reaches 19.5-24.5%, and titrable acidity varies in the range of 8-9.3 g /dm³ [10].

It has been established that grape raw materials grown in different micro-zones differ in their sensory characteristics, uvological and chemical composition, as well as in antioxidant, antiradical and antimicrobial properties [11,12].

Secondary resources accrued from the processing of colored grapes (in the form of skin and stone), by the contents of biologically active compounds have barely analogs in the autotrophic organisms, and they are not of less value products than wine itself. Only 9-12% of the total amount of phenolic compounds is contained in grape juice and pulp, accounting for 75-81% of the total mass of raceme, while the remaining 88-91% of phenolic compounds is mostly localized in the skin and stone, the mass of which is only 18-25% of raceme. This clearly shows how rich the biologically active compounds are in the solid parts of colored grapes, as well as how big is their role in the production of powerful antioxidant polyphenolic concentrates.

Accordingly, research in this field is of high relevance.

Materials and methods

Object of study

Research covered the raw materials of “Aladasturi” grape variety from different vineyards of the Imereti viticulture and winemaking zone, particularly: sample No. 1 - Lifnari vineyards (Rokhi Village, Baghdati district, 120-160 m above sea level), sample No. 2 - Svirivineyards (Svirivillage, Zestafoni district, 230-250 m above sea level) and sample No. 3 - Bagineti vineyards (Bagineti Village, Vani district, 580-600 m above sea level).

Research also covered hydrophilic extracts of grape skin and stone thickened by the vacuum of

“Aladasturi” colored grapes raw materials, as well as the concentrates produced from their composition.

Research Methods

For research, there were used gravimetric, extractive, spectral and chromatographic methods [13-22].

In test samples, we determined: the moisture and solid matter contents by heat-gravitational (GOST 28561- 90) and refractometric methods.

Quantitative analysis of total phenols was performed spectrophotometrically, by Folin-Ciocalteu reagent. In particular, we extracted the crushed test samples with 75-81% ethyl alcohol at the temperature of 72-75 °C and under conditions of periodic stirring for 6-7 hours. 1 ml of extract obtained, we placed into a 25 ml flask and added 0.5 ml of H₂O, 1 ml of Folin-Ciocalteu reagent, and settled for 8 minutes at room temperature, then we added 10 ml of 7% Na₂CO₃, filled the flask with H₂O, and settled it for 2 hours at room temperature.

The determination was carried out at 750 nm. As a control, we took 1 ml of the appropriate extracting agent and went through the same process. Calculation of the data obtained from the determination was carried out on the calibration curve of gallic acid.

The total phenol content shall be calculated in accordance with the formula:

$$X = (D K V F)1000 / m,$$

where X - the total phenol content, mg/kg;

D – optical density;

K – gallic acid conversion factor;

F – solubility;

V – the total volume of extract, ml;

m – raw materials mass taken for extraction, g.

Antioxidant activity in test samples was determined by one of the most common methods - DPPH method. DPPH is a rapid, simple and accurate test method for determining antioxidant activity.

DPPH - (C₁₈H₁₂N₅O₆ M=394,33) is a stable free radical with maximum absorption at 515 - 517 nm, and purple-violet coloration of its methanol extracts changes to bright yellow as a result of the recovery. The reaction occurs in accordance with the following pattern:



where AH is antioxidant and R is a free radical.

Quantification of **total flavonoids** was carried out with AlCl₃ reagent by spectral method - test sample was extracted with 80% ethyl alcohol at the

temperature of 70 - 75 °C. 1 ml of extract obtained from the total volume was placed into a 10 ml flask, then we added 5 ml of H₂O, 0.3 ml of 5% NaNO₂ was settled for 5 minutes, and then we added 0.3 ml of 10% AlCl₃ and settled for 6 minutes, then we added 2 ml of 1N NaOH- R and the determination was performed at 510 nm. As a control, we took 1 ml of the appropriate extracting agent and then went through the same process.

Calculation of the data obtained from the determination was carried out on the rutin calibration curve. The total flavonoid content shall be calculated in accordance with the formula:

$$X = (D K V F) \cdot 1000 / m$$

where X - the total flavonoid content, mg/kg;

D – optical density;

K – rutin conversion factor;

F – solubility;

V – the total volume of extract, ml;

m - raw materials mass taken for extraction, g.

The course of the pH-differential method for quantification of **monomeric anthocyanins** was as follows: we take test sample from 1 to 5 grams and carry out extraction with 45% ethyl alcohol. The volume of extract was reduced to 50 or 100 ml according to the extraction quality. From the total volume of extract, we take in two test-tubes 1 ml of extract in each, and add 4 ml of buffer solution in each. In one test-tube, we add 0,025 M of potassium chloride, and in the other test-tube, we add 0,4 M of sodium acetate, and 20 minutes later, we determine the optical density of the test solutions at 520 nm and 700 nm.

Quantification of leucoanthocyanins and catechins by spectral method - extraction of test sample was carried out with 80% ethyl alcohol at the

temperatures of 70 - 75 °C. 1 ml taken from the total volume of extract was added with 3 ml of vanillin reagent and, 3 minutes later, we determine the optical density of red test sample at 500 nm. As a control, we shall take 1 ml or 3 ml of vanillin reagent. Calculation of the data obtained from the determination was carried out on the (+)catechin calibration curve. The catechin content shall be calculated in accordance with the formula:

$$X = (D K V F) \cdot 1000 / m$$

where X - the catechin content, mg/kg;

D - optical density;

K – 35,0 ((+) catechin conversion factor;

F – solubility;

V – the total volume of extract, ml;

m - raw materials mass taken for extraction, g.

Results and discussion

“Aladasturi” is a late-ripening colored grape variety with a very special aroma that reaches full maturity in the second half of November, and the range of aromatic compounds in it, increases in proportion with the increase in the sugar content. The area of our concern was represented by polyphenolic compounds, and we were less interested in the sugar and aroma compound contents. Accordingly, the grape raw materials were taken during the period of their technical maturity, while phenolic compounds were present in grapes to the extent possible.

Grape samples were taken on 16 October 2018. The analysis of the uvological characteristics of individual samples of grape raw materials is given in Table 1.

Table 1. Uvological characteristics of individual samples of grape raw materials

Characteristics		October 16, 2018		
		Sample N1	Sample N2	Sample N3
Parts of the cluster of grapes, %	Juice and flesh	78,60	79,67	79,83
	Grape stalk	4,71	4,74	4,69
	Grape skin	11,87	10,85	10,82
	Grape stone	4,48	4,44	4,39
Number of seeds in the grain		1-4		
Solid remains (Grape stalk +Grape skin +Grape stone)		21,06	20,03	19,90
Structural indicator		3,74	3,98	4,02

The study of the urological characteristics of selected samples showed that structural indicators of all three samples of grapes (the ratio of flesh and juice to solid waste), at both stages of the grape harvest, were almost similar (relatively smaller for sample No. 1, and relatively larger for sample No. 3), indicating small differences in the quantitative phenolic complex contents in these samples [22].

We processed samples of grapes raw materials according to the following pattern:

- Identifying qualitative indicators of grapes raw materials;
- Passing grapes raw materials through the DMCSI-type grape clustercomb divider;
- Pressing-out the comb-less must in a basket press and separation of juice;
- Vacuum sublimation drying of juice-less sweet pomace with an initial moisture content of 45-65% to a final moisture content of 9-10%;
- Separation of the “Aladasturi” variety’s skin and stone dried to the moisture content of 9-10%, using tea sorting machine designed by G. Lominadze;
- Crushing separately the skin and stone in a micro-mill (TP2 Hammer Mill) until the fraction of 50-100 μm .

The crushed grape-stone was extracted by two different methods.

The first method (Grape-stone I - extract): - As an extracting agent for extraction of the grape-stone micropowder, we have selected a complex hydrophilic solvent – ethanol containing 40% volumetric alcohol, which was diluted with mineral drinking water “Borjomi”, whose pH = 3.6-6.3 Borjomi and mineralization is in the range of 7-14 g/dm³. This mineral water contains sodium (potassium) hydrogen carbonate and boric acid. Preliminary experiments have demonstrated that the extracting agent of ethanol diluted with mineral water can successfully replace the extracting agent diluted with water of ethanol containing 40% volumetric alcohol, which is oxidized by hydrochloric acid.

- We have determined experimentally the mass ratio of the extracting agent and the grape-stone microdispersed powder, which is 5 l/kg
- We have also determined experimentally the extraction parameters: temperature - 54-57 0C, duration - 180-210 minutes., pulsation - 4 sec-1 and the pulsation amplitude - 2-3 mm.
- Grape-stone ethanol extract at the initial stage, at the temperature of 4-5 0C, is subject to sedimentation for 7-9 hours, removal from sediment and filtration with a wine filter with plates.

The second method (Grape-stone II - Extract): - Extraction of a bioflavanoid complex from the grape-stone micro-powder was carried out using a supercritical super-fluid extractor (SFE - 100-2-C10) produced by Water Corporation, where the extracting agent was present together with CO₂ ethyl alcohol. For maximal extraction of the bioflavanoid complex, we have determined experimentally the optimal fluid extraction parameters: pressure - 95 bar, CO₂ delivery rate - 6.5 kg/h. In addition, the extraction quality was also affected by 72% ethanol as co-solvent, whose ratio to CO₂ was 21-22%.

- Grape-stone fluid extract at the initial stage, at the temperature of 4-5 0C, is subject to sedimentation for 7-9 hours, removal from sediment and filtration with a wine filter with plates. The data of the studies of biologically active compounds of the grape-stone superfluid extract are shown in Table 2 .
- We have blended the grape-stone extracts obtained by both methods at a ratio of 1:1. The filtered extract contained 5,2-6,3% of solid matters, and it was concentrated using a vacuum-rotary evaporator at the temperature of 54-57 0C to the solid matter content of 63%;

The composition of the concentrated grape-stone hydrophilic extracts was pumped over into the enameled collecting tank, from which test samples have been taken for the analysis on the biologically active compound content and antioxidant activity (see Table 3).

From the crushed grape skin, we obtained a hydrophilic liquid extract rich in bioflavonoids in accordance with the following technological scheme (grape skin extract):

- To effectively carry out extraction of anthocyanins from the grape skin, we processed the grape skin micropowder in advance to 0.4% with potassium metabisulphate.
- As an extracting agent, we selected 36% -45% volumetric ethanol processed by 2% citric acid. The optimal ratio of microdispersed raw materials and the extracting agent we determined experimentally at 3 l/kg.
- We determined experimentally the extraction optimal parameters: temperature - 54-57 0C; duration - 180-210 minutes; the extraction mass pulsation - 4 minutes; the amplitude - 5 mm.
- Prior to sedimentation and filtration, the obtained grape skin extract was processed by potassium bicarbonate (KHCO₃ - Potassium bicarbonate) for correcting 0.7-0.9 g/dm³ excessive acidity.

Table 2. *Biologically active compounds of the grape-stone fluid extract*

B.A.C., mg / 100 g. on dry weight basis	Stages of superfluid extraction								Total
	1	2	3	4	5	6	7	8	
Sample N1									
Phenolic compounds	131,6	977,66	782,9	395,9	344,6	114,1	137,5	95,1	2979,3
Flavonoids	290,8	505,6	421,9	310,3	243,6	144,6	219,4	89,0	2225,2
Flavan-3-ols	120,6	293,7	414,4	284,9	192,5	104,2	100,4	84,5	1594,2
Leukoanthocyanins	-	123,4	253,0	148,37	-	-	-	-	524,7
Sample N2									
Phenolic compounds	123,8	943,0	762,1	382,7	332,4	184,9	129,5	87,9	2946,3
Flavonoids	289,6	500,2	418,1	308,7	243,4	146,4	219,6	91,8	2217,8
Flavan-3-ols	118,0	287,6	406,0	279,0	188,4	101,8	97,2	82,6	1560,6
Leukoanthocyanins	-	130,6	257,7	153,2	-	-	-	-	541,5
Sample N3									
Phenolic compounds	132,4	953,3	764,2	388,7	343,9	201,9	142,0	99,8	3026,1
Flavonoids	292,9	501,1	421,8	310,5	247,7	149,9	219,3	98,5	2242,7
Flavan-3-ols	119,7	288,6	403,8	271,1	190,4	105,6	101,1	86,6	1566,9
Leukoanthocyanins	-	114,3	249,1	147,9	-	-	-	-	511,3

Table 3. *Biologically active compounds and antioxidant activity of grape-stone extracts with 61-63% of solid matter content*

Composition of hydrophilic extracts	Biologically active compounds , mg / 100 g. on dry weight basis				AOA, % (F=100)
	Phenolic compounds	Flavonoids	Flavan-3-ols	Leukoanthocyanins	
Sample N 1	3043,76	2293,94	1643,9	567,2	51,5
Sample N 2	3014,78	2276,10	1597,7	585,9	50,6
Sample N 3	3181,23	2308,65	1603,8	549,8	52,3

Table 4. Biologically active compounds and antioxidant activity of grape-skin hydrophilic extracts

Grape skin hydrophilic extract	Biologically active compounds , mg / 100 g. on dry weight basis				AOA, % (F=100)
	Phenolic compounds	Flavonoids	Flavan-3-ols	Leukoanthocyanins	
Sample N 1	3178,5	646,9	1295,9	2106,1	46,6
Sample N 2	3098,8	396,0	1484,5	1302,4	45,3
Sample N 3	3265,3	520,6	1667,8	1954,8	47,1

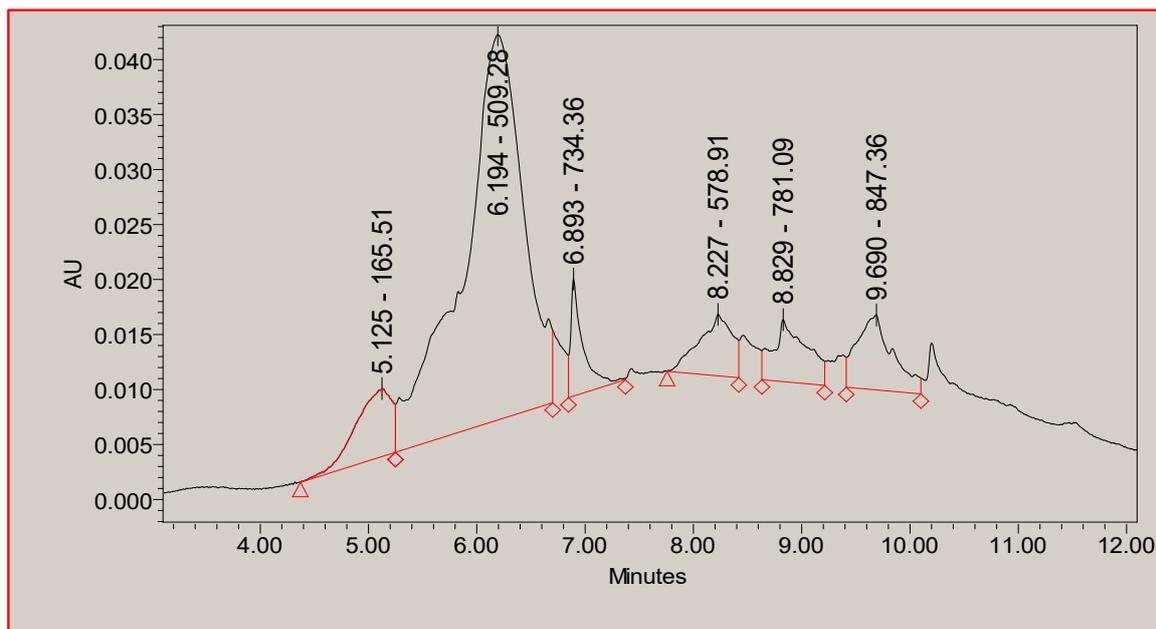


Fig. 1. Chromatogram of anthocyanins containing extract (61-63%) of Aladasturi peel (sample N1), m/z 509.28 [M-2H+H₂O] Malvidin-3-O-Glucoside

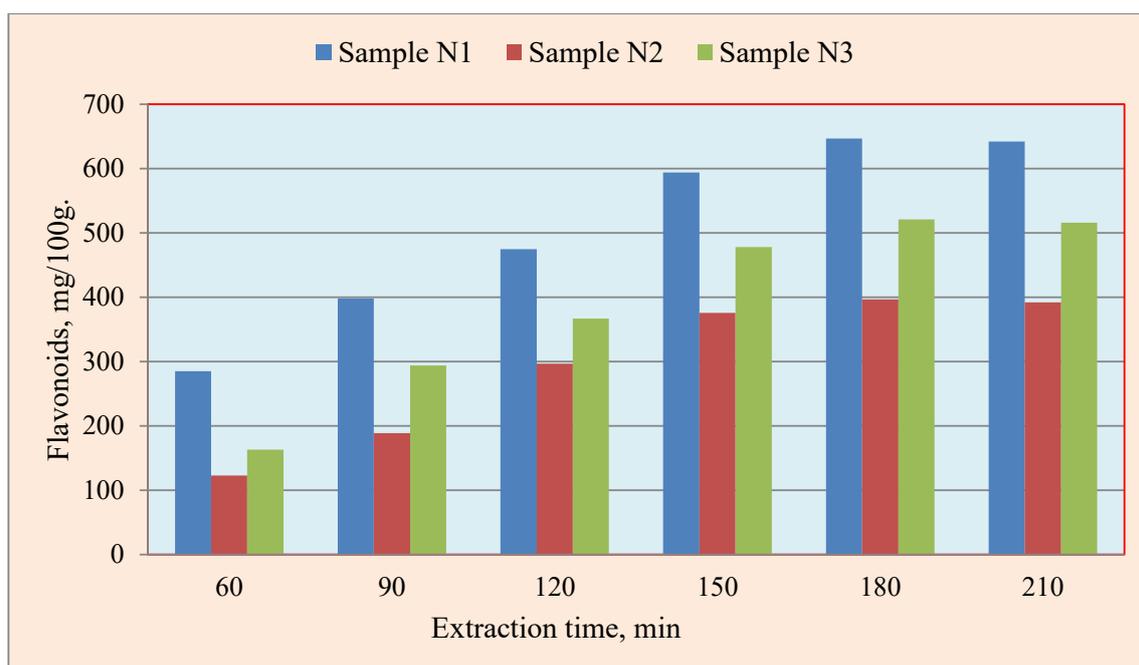


Fig. 2. Diagram of the extraction of flavonoids from grape skin

- The obtained extract, at the temperature of 4-5 0C, is subject to sedimentation for 7-9 hours, removal from sediment and filtration with a wine filter with plates.
- The composition of the filtered grape skin extracts contained 4,5-5,2 % of solid matters, and it was concentrated using a vacuum-rotary evaporator at the temperature of 54-57 0C to the solid matter content of 61-63%, and then we assessed biologically active compounds and antioxidant activity (Table 4). Figure 1 illustrates the chromatogram of anthocyanins of extract containing 61-63% of solid matters of the micro-dispersed skin of Lifnari's "Aladasturi" variety, and Figure 2 illustrates diagram of the extraction of flavonoids from grape skin.

We have blended the obtained grape-stone ethanol and fluid extracts containing 61-63% of solid matters at an equal ratio (1:1:1) and assessed biologically active compounds and antioxidant activity in this composition (Table 5).

The second stage of concentration was implemented by method of vacuum-sublimation or lyophilization to 74-75% of the solid matter content and pumped over into the enameled collecting tank, from which test samples have been taken for the analysis. The results of the assessment of biologically active compounds and antioxidant activity of bio-flavonoid liquid concentrate "Red Aladasturi"

are shown in Table 6.

The studies have shown that the bio-flavonoid concentrates containing 74-75% solid matters of of „Red Aladasturi” obtained from different samples of colored grapes are slightly different from each other in the biologically active compound contents, but all three samples produce the bio-flavonoid concentrates with high antioxidant activity.

Conclusion

It has been studied that the grape-stone and skin hydrophilic extracts of "Aladastur" colored grape variety's raw materials taken in the separate viticulture and winemaking micro-zones of Imereti and the liquid bio-flavonoid concentrates are characterized by high antioxidant activity (N1-56.6%; N2-55.45% and N3-55.45%).

The bio-flavanoid liquid concentrates obtained from sample No. 1 are characterized by a high anthocyanin content, while the concentrates obtained from sample No. 2, are characterized by a high leucoanthocyanin content, and the bio-flavanoid liquid concentrates obtained from sample No. 3 are characterized by the content and antioxidant activity of phenolic compounds and flavan-3-ols.

Anthocyanins in samples of "Aladasturi" variety are localized in the grape skin.

Table 5. *Biologically active compounds of grape-stone and skin ethanol and fluid extracts with 61-63% of solid matter content*

Sample number	Biologically active compounds , mg / 100 g. on dry weight basis					AOA, % (F=100)
	Phenolic compounds	Flavonoids	Flavan-3-ols	Anthocyanins	Leukoanthocyanins	
Sam. N 1	3089,8	1746,3	1529,1	2131,9	572,5	51,4
Sam. N 2	3044,7	1651,8	1562,6	1332,7	591,0	50,3
Sam. N 3	3210,6	1714,2	1625,9	2011,8	554,9	52,2

Table 6. *Biologically active compounds and antioxidant activity of "Red Aladasturi"*

Biologically active compounds , mg / 100 g. On dry weight	Sample N 1	Sample N 2	Sample N 3
Phenolic compounds	3401,8	3351,1	3533,3
Flavonoids	1921,2	1808,4	1886,7
Flavan-3-ols	1682,2	1719,0	1788,9
Anthocyanins	2348,3	1467,9	2213,2
Leukoanthocyanins	578,9	597,1	560,6
Dry matter, %	74-75	74-75	74-75
AOA, (F=100), In, %	56,6	55,31	57,45

Acknowledgment

This study was supported by Shota Rustaveli National Science Foundation (SRNSF) [N216752, Developing Innovative Technologies of Drastic Antioxidant Polyphenol Concentrates].

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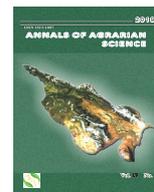
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Occurrence of stilbenoids in grapevine under Crown gall infection

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Received: 22 May 2018; accepted: 19 April 2019

ABSTRACT

It is established occurrence of phytoalexin stilbenoids in white and red grape varieties infected with crown gall disease (*Agrobacterium tumefaciens*) in Georgia. The infected vine varieties – Rkatsiteli, Saperavi, Cabernet Sauvignon, Tsitska and Tsolikouri were identified in East and West regions of Georgia. Healthy vine varieties were taken from the same vineyards for studying. Stilbenoid-containing fractions were isolated from the infected and healthy vine trunks and was identified their stilbenoids profile. It is studied the variation of the physiological concentration of stress- metabolite stilbenoids – trans-resveratrol and trans- ϵ -viniferin in terms of crown gall disease. It is identified, that occurrence of the physiological concentration of stilbenoids-trans-resveratrol and trans- ϵ -viniferin in vine infected with crown gall disease depends on variety factor. The obtained results are important data to identify the correlation of the immunity of the grape varieties to the phytoalexin stilbenoids.

Keywords: Vine, Phytoalexin, Stilbenoids, Trunk, Crown gall, Grape varieties.

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Introduction

Vine and grape stilbenoids are one of the groups of a wide class of phenol compounds, which incorporates cis- and trans-isomers of monomer resveratrol and their derivatives, as dimers, trimers, tetramers and glycosides [1-9]. Stilbenoids have diversified high biological activity and these compounds are very important for plants, as phytoalexins. Stilbenoids act against different vine diseases caused by biotic factors. The following stilbenoids were identified in the extract of vine (*Vitis vinifera*) trunk, roots and annual shoots: Ampelopsin A, (E)-piceatannol, Pallidol, E-resveratrol, hopeaphenol, isohopeaphenol, (E)- ϵ -viniferin, (E)-miyabenol C, (E)- ω -viniferin, r- and r2-viniferin. It was established that the extract inhibits the growth of sporulation of fungus *Plasmopara viticola* by 50%, while the most active inhibitor of it turned out to be r2-viniferin [10]. Under the influence of *Botrytis cinerea* on the mixture of Pterostilben and Resveratrol 7 new stilbens were formed, while 5 new

stilbens were formed from Pterostilben under the same terms. The anti-fungus effect of these stilbenoids was fixed against *Plasmopara viticola* [11]. At three stages of the grape (*Vitis vinifera*) grain development, the grains were infected on purpose with *Botrytis cinerea* spores „in vitro“. In the infected grain, stilbenoids: Pterostilben, (E)- ϵ -viniferin and trans-resveratrol were fixed. Dominating among them was (E)- ϵ -viniferin. [12] The grains of *Vitis Vinifera* L. cv. Barbera in the ripening period were infected with conidial suspension of *Aspergillus janicus*, *A.ochraceus*, *A. fumigatus* and *A.carbonarius*. The process of formation of ochratoxin A and stilbenoids was supervised. It was found out that all experimental fungi except *A. Fumigatus* significantly increase the concentration of trans-resveratrol and at the same time, trans-Piceid stays unchanged. In the grape grain damaged by *A.ochraceus*, the concentration of piceatannol increased significantly. A large amount of *A.carbonarius* was synthesized in the grain infected with *A.carbonarius* isolate and

the anti-fungicidal activity occurred with the following concentrations: 300 mkg/gr and 20 mkg/gr, what was sufficient for the total inhibition of fungus *A.carbonariuces* [13]. Besides above mentioned biological activity stilbenoids have many other functional purposes[14-22]. The vine and grape impacts some factor[23-26]. The vine varieties of Georgia are rich in biologically active stilbenoids. trans-resveratrol, trans- ϵ -viniferin, 2 tetrameric stilbens, including hopeaphenol as one of them, were isolated and identified from an annual shoot of Rkatsiteli variety. These stilbenoids and new stilbenoids identified by us were identified in the Georgian red-grape vintage varieties and their wines [27-31]. The study of stilbenoids in Georgian vintage varieties as that of phytoalexins, qualitative and quantitative analyses of their physiological concentrations and stress-metabolites and their impact on the microorganisms causing bacterial and fungus diseases is an urgent issue of the research. Consequently, our goal was to identify the vine varieties infected with crown gall disease, identify and determine their stress-metabolite stilbenoids and compare them with healthy vine stilbenoid profile.

Objects and Methods

We used healthy trunks of white- and red-grape vine varieties and those infected with crown gall disease from the same vineyard as study objects. Samples were taken in period February – march in 2018 year. Rkatsiteli from Gurjaani region (Alluvial soil; vineyard 15-16 old); Saperavi from Kvareli region(Alluvial soil; vineyard 15-16 old); Cabernet Sauvignon from Akhmeta region(Cinamonic soil; vineyard 15-16 old); Tsitska and Tsolikouri from Zestaponi region(Yellow- brown Forest soil; vineyard 10-11 old) (Fig.1). For analyzing we used vertical parts of the trunks. We isolated stilbenoid-containing fractions from the healthy and infected vine trunks as a result of treatment according to the chart (Fig. 2).

Stilbenoids were determined by the method of high-performance liquid chromatography (HPLC) [9]. For this purpose, we used the Varion chromatograph SupelcosilPM LC18 Column, 250*4,6mm, eluents: A. 0,025% trifluoroacetic acid, B.Acetonitrile: A80/20. Gradient mode: 0-35 min, 20-50% B, 48-53min, 200% B. Flow rate of the eluent- 1 ml/min; wavelength-306 and 285nm. Analyzed samples: isolated stilbenoid-containing fractions were filtered using a membrane filter (0,45 μ) be-

fore the chromatographic procedure. The chromat-mass-spectral investigations were carried out under the above-mentioned conditions; mass-spectra were detected by obtaining of positive ions.

Results and Discussion

Trans-resveratrol and trans- ϵ -viniferin were identified as dominants in the stilbenoid profile trunks of healthy study vine varieties(fig.3). Therefore, in the present article we considered it purposeful to describe the quantitative variability of the said stilbenoids following the infection with crown gall disease. The physiological concentrations of trans-resveratrol and ϵ -viniferin in different varieties of healthy vine trunks vary a lot. The highest concentrations of trans-resveratrol of 7,8 g/kg were fixed in the vine trunk of Rkatsiteli variety, while the lowest concentration was fixed in the vine trunks of Tsitska and Tsolikouri of 0,83 g/kg and 0,81 g/kg, respectively. At the same time, it should be noted that the concentration of trans- ϵ -viniferin in the vine trunk of Tsitska and Tsolikouri much exceeds the concentration of trans-resveratrol. At the same time, the vine trunks of Rkatsiteli, Saperavi and Cabernet Sauvignon show much higher concentration of trans-resveratrol than the concentration of trans- ϵ viniferin. Different results were obtained with the concentration changes of trans-resveratrol and trans- ϵ -viniferin, as stress-metabolites in the vine infected with crown gall disease. In particular, in Saperavi, the concentration of trans-resveratrol decreased from 2,6 g/kg to 0,85g/kg,while at the same time, the concentration of trans- ϵ -viniferin increased from 0,81g/kg to 2,1 g/kg; in Cabernet Sauvignon infected with crown gall disease, the concentration of trans-resveratrol decreased from 4,8 g/kg to 3,2gr/kg; at the same time, the concentration of trans- ϵ -viniferin also decreased from 2,2 g/kg to 1,5 gr/kg; infected Rkatsiteli variety with vine crown gall disease resulted in the reduction of the concentration of trans-resveratrol from 7,8 g/kg to 5,7g/kg and a quantitative increase in the amount of trans- ϵ -viniferin from 0,26 g/kg to 1,2 g/kg; in Tsitska infected with crown gall disease, the concentrations of both, trans-resveratrol and trans- ϵ viniferin increased. In the trunk of Tsolikouri infected with crown gall disease, the concentration of the given stilbenoids showed a less quantitative reduction (Fig. 4,5).



Fig. 1. The trunk of Vine (Tsitska) infected by crown gall disease.

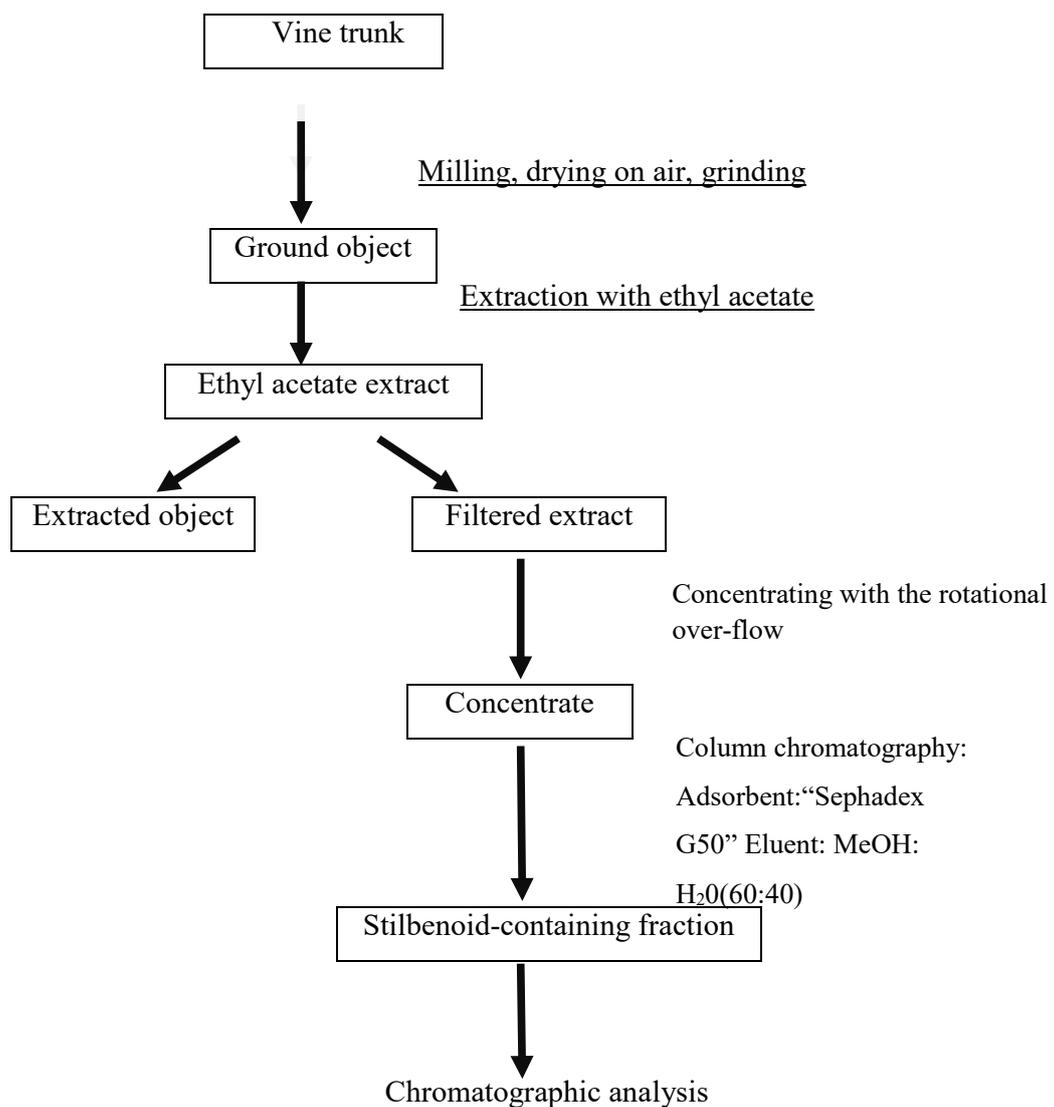


Fig. 2. Chart of isolating a stilbenoid-containing fraction from vine trunk

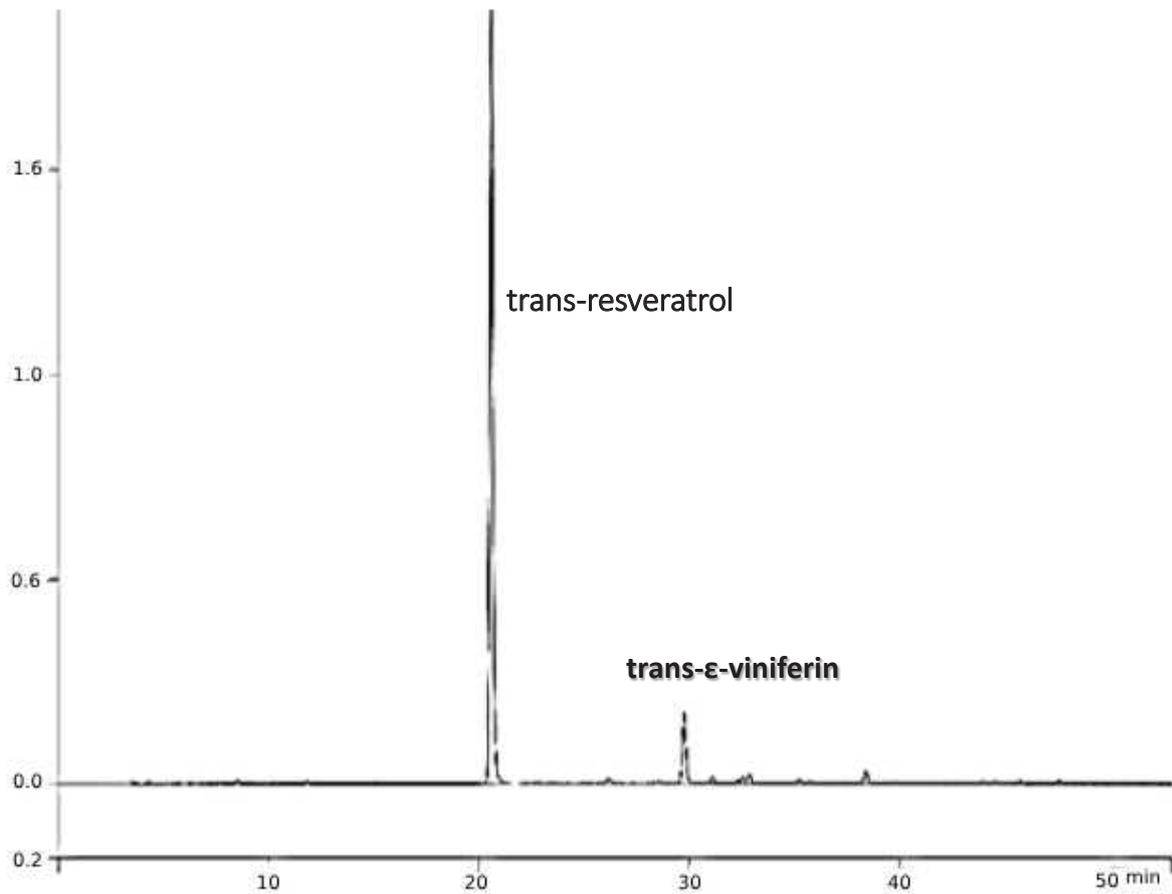


Fig. 3. HPLC of stilbenoid-containing fraction of Vine trunk of Healthy Rkatsiteli

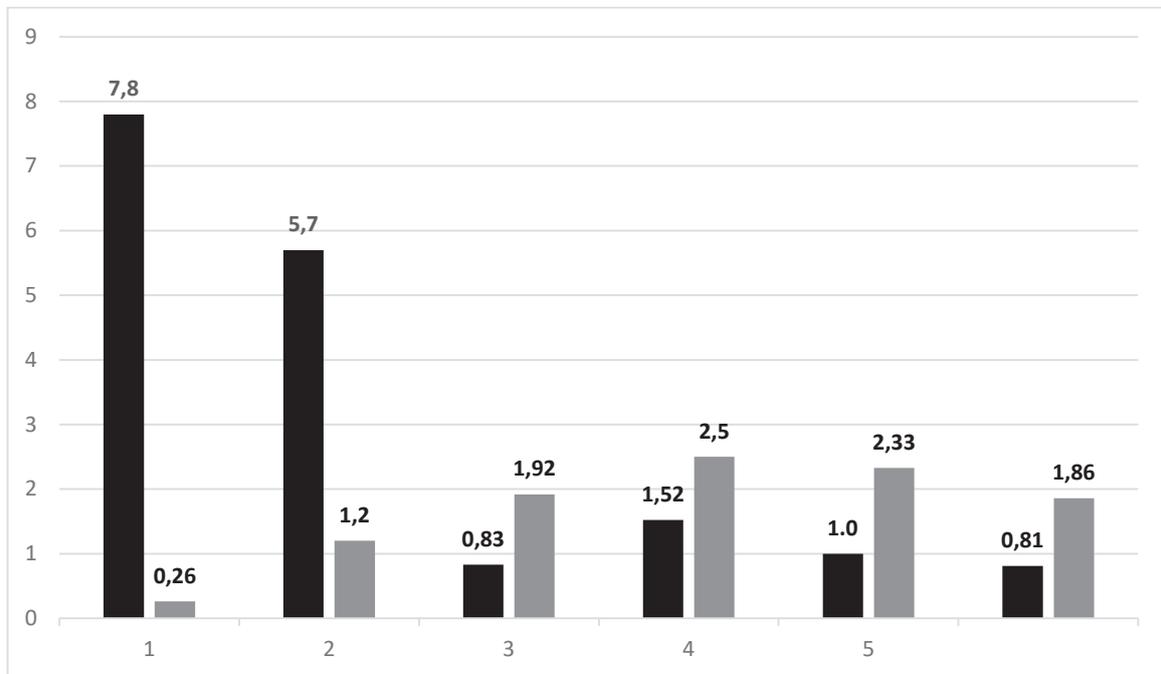


Fig. 4. Quantitative variation of trans-resveratrol and ε-viniferin (g/kg) in white-grape vine varieties infected with crown gall disease. 1. Healthy Rkatsiteli, 2. Infected Rkatsiteli, 3. Healthy Tsitska, 4. Infected Tsitska, 5. Healthy Tsojikouri, 6. Infected Tsojikouri .

■ -trans – Resveratrol, ■ - trans – ε-Viniferin

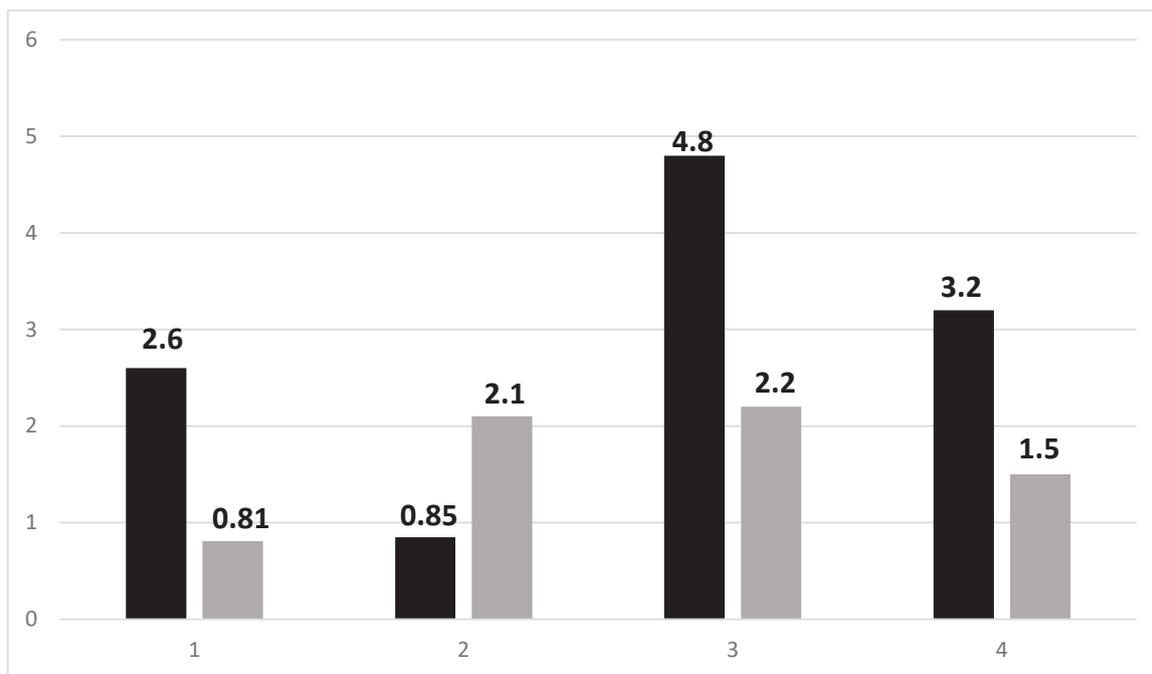
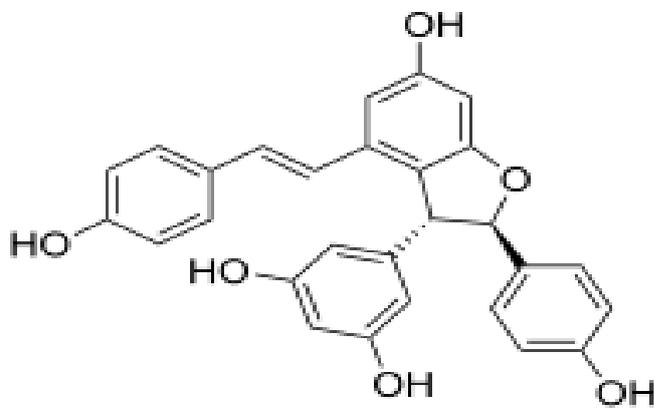
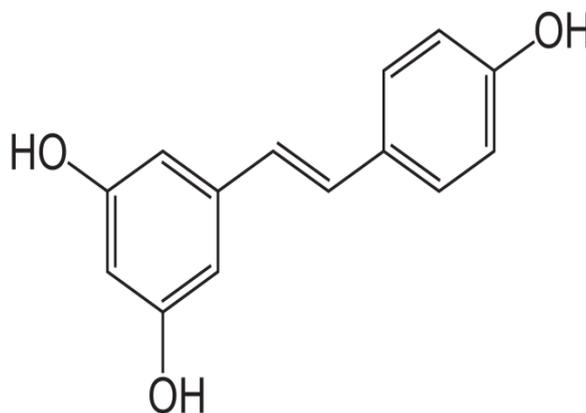


Fig. 5. Quantitative variation of trans-resveratrol and ε-viniferin (g/kg) in red-grape vine. 1. Healthy Saperavi, 2. Infected Saperavi, 3. Healthy Cabernet Sauvignon, 4. Infected Cabernet Sauvignon.

■ - trans – Resveratrol, ■ - trans – ε-Viniferin



Trans-ε-viniferin



Trans-resveratrol

Conclusion

Thus, as the accomplished study revealed the white- and red-grape vine varieties spread in Georgia: Rkatsiteli, Saperavi, Cabernet Sauvignon, Tsitska and Tsoolikouri are characterized by the relevant phytoalexin-stilbenoid profile. The relevant vine varieties infected with crown gall disease differ with the qualitative and quantitative contents of stilbenoids. The variation of the physiological concentrations of stress-metabolite stilbenoids: trans-resveratrol and ϵ -viniferin in the vine infected with crown gall disease takes place under the influence of the generic factor. In particular, in Saperavi and Rkatsiteli vine varieties, the concentration of trans-resveratrol decreased and the concentration of ϵ -viniferin increased; in Tsitska infected with crown gall disease, the concentration of both phytoalexins increased; in Tsoolikouri and Cabernet Sauvignon vine varieties, the amount of trans-resveratrol and ϵ -viniferin decreased. One of the reasons for such a quantitative reduction is the formation of other stress-metabolites of the derivatives of these compounds what helps the vine to fight against *Agrobacterium tumefaciens*. The obtained results are important data to establish the correlation of the vine varieties immunity with phytoalexins stilbenoids.

Acknowledgment

This work was supported by Shota Rustaveli National Science Foundation of Georgia (SRNSFG) [grant number FR 17_486].

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