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G.O. Kantureyeva[1]*, A.A. Saparbekova[1], D.E. Kudassova[1], G.G. Rakhmonova[2], K.A. Urazbayeva[1]

[1]NC JSC M. Auezov South Kazakhstan University, Shymkent, Kazakhstan
[2]Academician A.S. Sadykov Institute of Bioorganic Chemistry of Uzbekistan Academy of Science, Tashkent, Uzbekistan
Corresponding author: gulzhan.kantureeva01@auezov.edu.kz

Study of acute toxicity and cumulative activity of polyphenolic powders derived from pomegranate peel and grape seed waste

Abstract. *This article presents studies to determine the acute toxicity and cumulative activity of polyphenolic powders derived from pomegranate peel and grape seed. When administered intragastrically to mice in doses from 500 to 2000 mg/kg, no acute toxicity was detected, no animal death was observed, and no deviations from the norm in other external parameters were observed. The study of acute toxicity of the preparations of polyphenolic powders from (PPPP) and polyphenolic powders derived from grape seed (PPGS) showed that they belong to the V class of chemical compounds (practically non-toxic) according to the OEC classification, the LD50 on mice was ->2000 mg/kg. The preparations belong to the group with average and weak cumulative properties. These polyphenolic powders with high biological activity were obtained from food waste in Turkestan region, such as pomegranate peel and grape seeds. In recent years, there has been an active search for plant sources for bioactive compounds, and among the widely consumed fruits, which are considered an excellent source of polyphenols, and in relation to the use of industrial waste, pomegranate and grape fruits, are the leaders. The appropriateness of their use for human consumption was confirmed by the low level of risks in terms of the absence of toxicity and pronounced cumulative actions in these preparations.*

Keywords: *biowaste, pomegranate peel, grape seed, polyphenolic powders, acute toxicity, cumulative activity.*

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Introduction

Pollution created by biowaste and byproducts from the agri-food chain is a growing concern in terms of animal and plant health and environmental sustainability [1]. In addition, the processing of by-products is a promising area that requires interdisciplinary research by food technologists, chemists, nutritionists and toxicologists, as well as biotechnologists [2].

However, only a small proportion of food waste is separated, recycled or used for animal feed. Especially since, for example, in Kazakhstan in 2021 the amount of food waste amounted to 35.9 thousand tons [3].

In recent years, there has been an active search for plant sources for bioactive compounds, and among the widely consumed fruits, which are considered an excellent source of polyphenols, and in relation to the use of industrial waste, pomegranate and grape fruits, are the leaders. All parts of the above mentioned fruits contain polyphenols, but, for example, 60-70% of extractable polyphenols are concentrated in grape seeds as flavonols, phenolic acids, catechins, proanthocyanidins and anthocyanins. Among them, catechins and proanthocyanidins are the main compounds, accounting for 77.6% of the total amount of certain phenolic compounds [4].

For example, pomegranate peel, which makes up 49 to 55% of the weight of the fruit, depending on its variety [5], is rich in phenolic compounds, including punicalagin (28,000-104,000 g/g), ellagic acid (1580-4514 g/g), catechin (115-613 g/g), gallic acid (10-73 g/g) and several other compounds [6] that are responsible for its biological activity [7].

However, knowing that the total production of pomegranate worldwide is three million tons, and its peel and seeds make up about 54% of the fruit, this leads to approximately 1.62 million tons of waste [8].

Grapes are considered the most common plant species worldwide, and *Vitis vinifera L.* is the main species used as a raw material for wine production. This industry is a sector with great potential worldwide. Global wine production in 2021 was 260 million tons. Wine production produces approximately 1.3-1.5 kg of residues per liter of wine produced [9].

Thus, a huge quantity of residues is being generated that is essential to find suitable methods to revalue it by optimizing the extraction of bioactive compounds from pomegranate and grape fruit residues and then converting them into value-added products.

At the same time, although the scientific community supports research on reuse of biological waste, the question of biological waste safety must necessarily remain open. Two aspects of biowaste call into question the safety of the products derived from them and their use for human consumption. The first is related to the chemical complexity of biological waste. The technology used to process this material can lead to the release of chemical compounds such as toxins, genotoxic compounds, and heavy metals into the final product, which are potentially dangerous to human health [10]. The second problem associated with the reuse of biological waste is the possible change in the absorption, distribution, metabolism, and excretion profile of the final product. Unexpected molecules can synergistically affect the biologically active fraction of the plant and change the overall pharmacokinetic parameters of the final product (bioavailability, bioavailability, bioactivity) as well as its overall toxicity [11]. Considering the requirements for drug safety in toxicological studies, it is also necessary to calculate the cumulation coefficient in order to determine whether the drug cumulates or it is addictive. The aim of our work was to define the acute toxicity and cumulative activity of polyphenolic powders obtained from waste products: pomegranate peel and grape seed.

Materials and methods

The object of the study was polyphenolic powders obtained from pomegranate peels and grape seeds according to the further described methodology. Aqueous-alcoholic extracts were obtained from dried samples by maceration with periodic stirring; in brief, 1000 g of pomegranate peel were mixed with 40% ethanol (1:4 by mass), stirred for 10 min, left to infuse for 24 hours at room temperature and finally filtered to extract aqueous-alcoholic extract. Then the extract was evaporated for efficient and gentle removal of solvents at a rotary evaporator, and a finely dispersed loose polyphenol powder (PP) from pomegranate peels was obtained by spray drying. When obtaining PP from grape seeds, the difference was the use of 70% ethanol during extraction.

Standardization of the preparation was carried out using chromatographic (thin layer chromatography, high performance liquid chromatography), spectroscopic and spectrophotometric methods. The high content of various polyphenolic compounds was revealed in both samples including flavonoids (rutin and catechin) 11.777 and 6.13 mg/100g respectively in PP from pomegranate peel; 26. 766 and 44.878, mg/100g respectively in PP from grape seed; phenolic acids (gallic acid) 118.217 mg/100g in PP from pomegranate peel (PPPP); 106.518, mg/100g, in PP from grape seed (PPGS).

Common methods were used to determine acute toxicity parameters. The investigation of acute toxicity of the preparations was performed on white male mice weighing 20±2.0 g, 5 animals in each group, 45 mice in total were used. All manipulations with them were performed in accordance with International Agreement for Humane Treatment of Animals (The European Communities Council Directives of November 24, 1986- 806/609/EEC). The animals were obtained from the cattery of Tashkent Zoo. All pharmacological studies were performed on healthy mature animals (mice) quarantined for at least 10-14 days in specialized vivarium (at the Academician Sadykov Institute of Bioorganic Chemistry - IBCh Uzbekistan AS). Preparations of PPPP and PPGS were administered to mice once intragastrically

in doses of 500, 1000, 1600 and 2000 mg/kg, control distilled water in the same volume.

The animals were observed every hour during the first 24 hours of the laboratory test, with survival during the analysis, overall status, potential breakdown and passing like indexes of the animals' functional condition. Then every day, during 2 weeks in the vivarium conditions, the animals of all groups were observed for their general state and activity, behavior features, number and deepness of breathing, condition of hair and epidermis, urination, body mass altering and other indicators. All tested mice were stayed under the equal circumstances and on the general ration with obtaining water and food for free [12]. When experiment was finished, the average lethal dose (LD_{50}) was calculated and the toxicity class was determined [13].

Study of cumulative properties of PPPP and PPGS preparations was implemented according to strategy proposed by R.R. Lim et al [14] on each group of 10 white male mice weighing 20±2 g. Scheme of the experiment included daily intragastric injection of PPPP and PPGS preparations, once through the probe. The first 4 days the dose was 200 mg/kg (0.1 LD_{50}), then every next 4 days the dose was increased and the injected dose was 300, 440, 680, 1000, 1500, and 2240 mg/kg. The injected doses were calculated based on the established LD_{50}=2000 mg/kg dose, when injected once to the animals. The maximum duration of the experiment was 28 days.

The cumulation coefficient (Cc), calculated as the ratio of the total dose received by the body during repeated (fractional) injection of the substance in an amount equal to the average lethal dose of LD_{50}, to the same value, but at a single injection, was calculated by the follow formula (1):

$$Cc = LD_{50;\,n} / LD_{50;1}, \quad (1)$$

where $LD_{50;n}$ is the cumulative mean lethal dose at n-fold injection,

$LD_{50;1}$ – average lethal dose at a single injection.

The qualitative estimation of Cc value was calculated by the L.I. Medvedja et al. scale modified by B.I. Lyublina [15], where Cc <1 corresponds to Supercumulation; Cc = 1-2.2 - Distinct cumulation; Cc = 2.2-5 - Average cumulation; Cc>5 - Weak cumulation.

Results

The total impact and acute toxicity of PP from pomegranate peel and PP from grape seed drugs were analyzed on mice by single inoculations into gastric. Every single dose of the preparation was analyzed on five mice. During fourteen days the experimental animals were monitored. PP from pomegranate peel and PP from grape seed preparations were administrated into the gastric at doses of 500, 1000, 1600, and 2000 mg/kg per mouse.

The results obtained are in the Table 1.

Table 1

Results of acute toxicity indicators during intragastric injection of PPPP and PPGS preparations into mice (n=5)

Groups	Animal species, The way of injection	Doses, mg/kg	Number of deaths /number of animals in groups	LD_{10} mg/kg	LD_{16} mg/kg	LD_{50} mg/kg	LD_{84} mg/kg
PPPP	Mice, i/g	500	0/5			>2000	
		1000	0/5				
		1600	0/5				
		2000	0/5				
PPGS	Mice, i/g	500	0/5			>2000	
		1000	0/5				
		1600	0/5				
		2000	0/5				
control	Mice, i/g	0,2 ml/mouse	0/5			-	

As shown in Table 1, acute toxicity was determined by assessing the survival rate of mice and the calculated average lethal dose. The results showed that no lethal effects were observed when the drugs were administered intragastrically (i/g), i.e., the doses causing possible lethal effects when administered i/g are higher than 2000 mg/kg.

The subchronic toxicity study of cumulation assessment criteria were as follows: animal condition before and after drug administration, behavioral reactions, condition of hair and visible mucous membranes, respiratory rate, heart rate, number of fallen and survived animals in groups. At the end of the experiment the cumulation coefficient was determined as the ratio of LD_{50} at multiple injections to LD_{50} at single injections. The observation period for animals in acute experiments in the presence of single death should be at least 14 days. The later the deaths occur, the more likely the substance under test has cumulative properties. It is believed that the greater the value of the cumulation coefficient, the more pronounced are the cumulative properties of the tested biologically active substances. The results of determining the cumulative activity of PPPP and PPGS preparations are shown in Tables 2 and 3.

Table 2

Subchronic toxicity study of cumulation by the PPPP preparation

Days of injection	Number of deaths/ number of animals in the group	Fraction of LD_{50} at a single injection	LD_{50} =2000 mg/kg
1-4	0/10	0.1	200
5-8	0/10	0.15	300
9-12	0/10	0.22	440
13-16	0/10	0.34	680
17-20	2/10	0.50	1000
21-24	4/10	0.75	1500
25-28	6/10	1.12	2240

Table 3

Subchronic toxicity study of cumulation by PPGS preparation

Days of injection	Number of deaths/ number of animals in the group	Fraction of LD_{50} at a single injection	LD_{50} =2000 mg/kg
1-4	0/10	0.1	200
5-8	0/10	0.15	300
9-12	0/10	0.22	440
13-16	0/10	0.34	680
17-20	2/10	0.50	1000
21-24	3/10	0.75	1500
25-28	4/10	1.12	2240

Discussion

Measuring the toxicity of plant extracts is important for determining biological indicators for *in vitro* studies because many chemicals and plants have different levels of toxicity. When injecting animals PP from pomegranate peel in doses of 500 and 1000 mg/kg, washing, narrowing of the eyes, and crowding were noticed, and behavior of the animals normalized in 1 to 3 hours. In the situation with the same preparation but in amount of 1600 and 2000 mg/kg, washing, lethargy, bunching, and eye narrowing were noticed. Animal behavior normalized in 3-4 hours. No mortality was observed in all doses (0/5).

To determine the acute toxicity of the second preparation, PP from grape seeds, intragastric administration was performed at doses of 500, 1000, 1600, and 2000 mg/kg per mouse. In the first hour after injection of the drug in all studied doses, animals showed washing, immobility, bunching, and some animals showed lethargy, especially with increasing dose of the drug. When injecting the PPGS drug in all doses used, no 0/5 death was recorded.

For other studied parameters throughout the study period (14 days): condition of hair and epidermis, tail state, quantity and texture of fecaloma, altering of body mass when injected with the drug in all doses used, no deviation from the norm was observed.

So, the investigation of the acute toxicity of PPPP and PPGS drugs showed that these drugs, when injectered intragastrically, correspond to an LD_{50} of more than - 2000 mg/kg. According to the modified classification of the Organization for Economic Cooperation and Development (OECD), this drug belongs to the V class of toxicity of substances (Practically non-toxic).

Further, throughout the duration of the study (14 days), the surviving animals were observed after administration of PPPP and PPGS preparations. Observation of experimental animals according to the studied indices did not reveal any deviations in the state of hair and skin, tail position, fecal mass consistency, diuresis, and change in body weight from the control group animals. Analyses of the acute toxicity of PPPP and PPGS preparations obtained from food waste of Turkestan region, were similar to those reported by other researches [16,17].

To identify the possibility of chronic poisoning of animals, the cumulative properties of experimental samples of the PPPP and PPGS preparations were studied in a subacute experiment on mice. According to table 2, the resulting total dose to the animal for 28 days is =12.72 $LD_{50;1}$. $LD_{50;n}$ - the total dose at which the beginning of a large number of deaths was recorded, was $LD_{50;n}$ = 25440 mg/kg, and this dose is higher than $LD_{50;n}$. On this basis we calculate coefficient Cc: Cc=6480 / 2000 = 3.24. According to the generally accepted hygienic classification (GOST 12.1.007-76), the drug belongs to hazard class 3 when administered into the stomach. The cumulation coefficient was 3.24, i.e. the drug

PPPP has average cumulative properties.

According to table 3, the cumulative dose received by the animal for 28 days is =12.72 $LD_{50;1}$. $LD_{50;n}$ - the cumulative dose at which the beginning of the deaths is fixed, was $LD_{50;n}$ = 2540 mg/kg, and this dose is higher than $LD_{50;n}$. On this basis we calculate Cc coefficient: Cc=9480 / 2000 = 4.74. The cumulation coefficient is - 4.74, which corresponds to the evaluation of the action of the substance PPGS as a drug with weak cumulative properties.

Conclusion

Thus, the study of acute toxicity of PPPP and PPGS preparations showed that they belong to the V class of chemical compounds (practically non-toxic) according to the OECD classification. Acute toxicity test showed that LD_{50} was on the level >2000 mg/kg and no significant impact on mice. The cumulation coefficient of PPPP is -3.24, which corresponds to the drug with average cumulative properties (according to the scale of L.I. Medvedya et al. modified by B.I. Lyublina). The cumulation coefficient of PPGS is 4.74, which corresponds to the drug with weak cumulative properties (according to the scale of L.I. Medvedja et al. in modification of B.I. Lyublina).

The results obtained justify the expediency of using the obtained polyphenolic powders from the residues of processed pomegranate and grape fruits with high content of bioactive compounds in the construction of value-added products.

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References

1. Castrica M., Rebucci R., Giromini C., Tretola M., Cattaneo D. & Baldi A. Total phenolic content and antioxidant capacity of agri-food waste and by-products // Italian Journal of Animal Science. – 2019. – Vol. 18(1). – P. 336-341. DOI: 10.1080/1828051X.2018.1529544.

2. Otles S., Despoudi S., Bucatari, C., Kartal C. Chapter 1- Food Waste Management, Valorization, and Sustainability in the Food Industry. In Food Waste Recovery. – San Diego, CA, USA: Academic Press, 2015. – P. 3-23.

3. Как эффективно работать с отходами и развить отрасль переработки пищевых отходов в Казахстане. [Electronic resource] – URL: https://the-steppe.com/lyudi/kak-effektivno-rabotat-s-othodami-i-razvit-otrasl-pererabotki-pishchevyhothodov-v-kazahstane (Accessed: 30.10.2022).

4. Silvan J.M., Mingo E., Hidalgo M., Pascual-Teresa S. de, Carrascosa A.V. and Martinez-Rodriguez A.J. Antibacterial Activity of a Grape Seed Extract and Its Fractions against *Campylobacter spp* // Food Control. – 2013. – Vol. 29. – P. 25-31. DOI: http://dx.doi.org/10.1016/j.foodcont.2012.05.063.

5. Mo Y., Ma J., Gao W., Zhang L., Li J., Zang J. Pomegranate Peel as a Source of Bioactive Compounds: A Mini Review on Their Physiological Functions // Frontiers in Nutrition. – 2022. – Vol. 9. – P. 887113. DOI: 10.3389/fnut.2022.887113.

6. Man G., Xu L., Wang Y., Liao X. and Xu Z. Profiling Phenolic Composition in Pomegranate Peel From Nine Selected Cultivars Using UHPLC-QTOF-MS and UPLC-QQQ-MS // Frontiers in Nutrition. – 2022. – Vol. 8. – P. 807447. DOI: 10.3389/fnut.2021.807447.

7. Magangana T.P., Makunga N.P., Fawole O.A., Opara U.L. Processing factors affecting the phytochemical and nutritional properties of pomegranate (*Punica granatum L.*) peel waste: A review // Molecules. – 2020. – Vol. 25(20). – P. 4690. DOI: https://doi.org/10.3390/molecules25204690.

8. Cano-Lamadrid M., Martínez-Zamora L., Castillejo N., Artés-Hernández F. From Pomegranate Byproducts Waste to Worth: A Review of Extraction Techniques and Potential Applications for Their Revalorization // Foods. – 2022. – Vol. 11. – P. 2596. DOI: https://doi.org/10.3390/foods11172596.

9. State of the world. Vine and wine sector 2021. [Electronic resource] – URL: eng-state-of-the-world-vine-and-wine-sector-april-2022-v6.pdf (Accessed: 25.10.2022).

10. Kirkland D., Kasper P., Martus H.-J., Müller L., van Benthem J., Madia F., Corvi R. Updated Recommended Lists of Genotoxic and Non-Genotoxic Chemicals for Assessment of the Performance of New or Improved Genotoxicity Tests // Mutation Research -Genetic Toxicology and Environmental Mutagenesis. – 2016. – Vol. 795. – P. 7-30.

11. Musto G., Laurenzi V., Annunziata G., Novellino E., Stornaiuolo M. Genotoxic Assessment of Nutraceuticals Obtained from Agricultural Biowaste: Where DoWe "AMES"? // Antioxidants. – 2022. – Vol. 11. – P. 1197. DOI: https://doi.org/10.3390/antiox11061197.

12. Руководство по проведению доклинических исследований лекарственных средств. Ч. 1. Методические рекомендации по изучению общетоксического действия лекарственных средств. Изучение острой токсичности. – Москва: Гриф и К, 2012. – С. 15-17.

13. OECD, Test No. 420:2001 «Острая токсичность при внутрижелудочиом поступлении. Метод фиксированной дозы» («Acute Oral Toxicity — Fixed Dose Procedure», IDT).

14. Lim R.R. et al. A method for the evaluation of cumulation and tolerance by the determination of acute and subchronic median effective doses // Arch. Intern.Pharm. Ther. – 1961. – Vol. 130. – P. 336-352.

15. Основы общей промышленной токсикологии (руководство). – Ленинград: Медицина, 1976. – 304 с.

16. Vale E.P., Rego L.R. do, Pureza D.D.N, Silva P.G. Cytogenetic and toxicological effects of Punica granatum Linnaeus fruit peel hydroethanolic extract in mice // South Afr. J. Bot. – 2020. – Vol. 130. – P. 465-470. DOI: https://doi.org/10.1016/j.sajb.2020.01.041.

17. Chedea V.S., Macovei S.O.,Bocsan I.C. Grape Pomace Polyphenols as a Source of Compounds for Management of Oxidative Stress and Inflammation-A Possible Alternative for Non-Steroidal Anti-Inflammatory Drugs? // Molecules. – 2022. – Vol. 27. – P. 6826. DOI: https://doi.org/ 10.3390/molecules27206826.

Г.О. Кантуреева[1], А.А. Сапарбекова[1], Д.Е. Кудасова[1], Г.Г. Рахмонова[2], К.А. Уразбаева[1]

[1]НАО М. Әуезов атындағы Оңтүстік Қазақстан университеті, Шымкент, Қазақстан

[2]Академик А.С. Садықов атындағы Биоорганикалық химия институты, Өзбекстан Республикасы Ғылым академиясы, Ташкент, Өзбекстан

Өндіріс қалдықтарынан алынған полифенол ұнтақтарының жедел уыттылығын және жинақтаушы белсенділігін зерттеу: анар қабығы мен жүзім тұқымы

Андатпа. Бұл мақалада анар қабығы мен жүзім тұқымдарынан алынған полифенол ұнтақтарының өткір уыттылығы мен кумулятивтік белсенділігін анықтау бойынша зерттеулер ұсынылған. Тышқандарға 500-ден 2000 мг/кг дейінгі дозада асқазанішілік енгізу кезінде өткір уыттылық анықталған жоқ, жануарлардың өлімі болған жоқ, басқа сыртқы көрсеткіштер бойынша нормадан ауытқулар байқалмады. Анар қабығының полифенолды ұнтақтары (ППКГ) және жүзім тұқымының полифенолды ұнтақтары (ППВК) препараттарының өткір уыттылығын зерттеу OECD, LD_{50} жіктемесі бойынша тышқандардағы химиялық қосылыстардың V класына (іс жүзінде уытты емес) жататынын көрсетті -2000 мг/кг құрады. Препараттар орташа және әлсіз

кумулятивті қасиеттері бар топқа жатады. Бұл биологиялық белсенділігі жоғары полифенолды ұнтақтар Түркістан облысының тамақ өндірісінің қалдықтарынан алынды. Соңғы жылдары биоактивті қосылыстардың өсімдік көздерін іздеу белсенді түрде жүргізілуде және полифенолдардың тамаша көзі болып саналатын кеңінен тұтынылатын жемістердің ішінде, сондай-ақ өндірістік қалдықтарды пайдалану тұрғысынан анар мен жүзім жемістері жетекші орында. Оларды адам тұтынуы үшін қолданудың орындылығы уыттылықтың болмауы және осы препараттарда айқын кумулятивті әрекеттер тұрғысынан қауіптің төмен деңгейімен расталды.

Түйін сөздер: биоқалдықтар, анар қабығы, жүзім тұқымы, полифенол ұнтақтары, өткір уыттылық, кумулятивтік белсенділік.

Г.О. Кантуреева[1], А.А. Сапарбекова[1], Д.Е. Кудасова[1], Г.Г. Рахмонова[2], К.А. Уразбаева[1]
[1]НАО «Южно-Казахстанский университет им. М. Ауэзова», Шымкент, Казахстан
[2]Институт биоорганической химии им. академика А.С. Садыкова АН РУз., Ташкент, Узбекистан

Изучение острой токсичности и кумулятивной активности полифенольных порошков, полученных из отходов производства: кожуры граната и виноградных косточек

Аннотация. Данная статья представляет исследования по определению острой токсичности и кумулятивной активности полифенольных порошков, полученных из кожуры граната и виноградных косточек. При внутрижелудочном введении мышам в дозах от 500 до 2000 мг/кг острой токсичности не выявлено, гибель животных отсутствовала, не было отклонений от нормативов по другим внешним показателям. Изучение острой токсичности препаратов полифенольных порошков из кожуры граната (ППКГ) и полифенольных порошков из виноградных косточек (ППВК) показало, что они относятся к V классу химических соединений (практически нетоксично) по классификации OECD, LD_{50} на мышах составила –>2000 мг/кг. Препараты относятся к группе со средними и слабыми кумулятивными свойствами. Данные полифенольные порошки с высокой биологической активностью были получены из отходов пищевых производств Туркестанской области. В последние годы ведется активный поиск растительных источников биоактивных соединений, и среди широко потребляемых фруктов, которые считаются отличным источником полифенолов, а также с точки зрения использования промышленных отходов, лидируют гранат и плоды винограда. Целесообразность их использования для потребления человеком была подтверждена низким уровнем рисков с точки зрения отсутствия токсичности и выраженных кумулятивных действий в данных препаратах.

Ключевые слова*:* биоотходы, кожура граната, виноградные косточки, полифенольные порошки, острая токсичность, кумулятивная активность.

References

1. Castrica M., Rebucci R., Giromini C., Tretola M., Cattaneo D. & Baldi A. Total phenolic content and antioxidant capacity of agri-food waste and by-products, Italian Journal of Animal Science, 18(1), 336-341 (2019). DOI: 10.1080/1828051X.2018.1529544.

2. Otles S., Despoudi S., Bucatari, C., Kartal C. Chapter 1- Food Waste Management, Valorization, and Sustainability in the Food Industry. In Food Waste Recovery, San Diego, CA, USA: Academic Press, 3-23 (2015).

3. Kak effektivno rabotat' s othodami i razvit' otrasl' pererabotki pishchevyh othodov v Kazahstane [How to effectively manage waste and develop the food waste processing industry in

Kazakhstan]. [Electronic resource] – Available at: https://the-steppe.com/lyudi/kak-effektivno-rabotat-s-othodami-i-razvit-otrasl-pererabotki-pishchevyhothodov-v-kazahstane (Accessed: 30.10.2022). [in Russian]

4. Silvan J.M., Mingo E., Hidalgo M., Pascual-Teresa S. de, Carrascosa A.V. and Martinez-Rodriguez A.J. Antibacterial Activity of a Grape Seed Extract and Its Fractions against *Campylobacter spp*, Food Control, 29, 25-31 (2013). DOI: http://dx.doi.org/10.1016/j.foodcont.2012.05.063.

5. Mo Y., Ma J., Gao W., Zhang L., Li J., Zang J. Pomegranate Peel as a Source of Bioactive Compounds: A Mini Review on Their Physiological Functions, Frontiers in Nutrition, 9, 887113 (2022). DOI: 10.3389/fnut.2022.887113.

6. Man G., Xu L., Wang Y., Liao X. and Xu Z. Profiling Phenolic Composition in Pomegranate Peel From Nine Selected Cultivars Using UHPLC-QTOF-MS and UPLC-QQQ-MS, Frontiers in Nutrition, 8, 807447 (2022). DOI: 10.3389/fnut.2021.807447.

7. Magangana T.P., Makunga N.P., Fawole O.A., Opara U.L. Processing factors affecting the phytochemical and nutritional properties of pomegranate (*Punica granatum L.*) peel waste: A review, Molecules, 25(20), 4690 (2020). DOI: https://doi.org/10.3390/molecules25204690.

8. Cano-Lamadrid M., Martínez-Zamora L., Castillejo N., Artés-Hernández F. From Pomegranate Byproducts Waste to Worth: A Review of Extraction Techniques and Potential Applications for Their Revalorization, Foods, 11, 2596 (2022). DOI: https://doi.org/10.3390/foods11172596.

9. State of the world. Vine and wine sector 2021. [Electronic resource] – Available at: eng-state-of-the-world-vine-and-wine-sector-april-2022-v6.pdf (Accessed: 25.10.2022).

10. Kirkland D., Kasper P., Martus H.-J., Müller L., van Benthem J., Madia F., Corvi R. Updated Recommended Lists of Genotoxic and Non-Genotoxic Chemicals for Assessment of the Performance of New or Improved Genotoxicity Tests, Mutation Research -Genetic Toxicology and Environmental Mutagenesis, 795, 7-30 (2016).

11. Musto G., Laurenzi V., Annunziata G., Novellino E., Stornaiuolo M. Genotoxic Assessment of Nutraceuticals Obtained from Agricultural Biowaste: Where DoWe "AMES"? Antioxidants, 11, 1197 (2022). DOI: https://doi.org/10.3390/antiox11061197.

12. Rukovodstvo po provedeniyu doklinicheskih issledovanij lekarstvennyh sredstv. CH. 1. Metodicheskie rekomendacii po izucheniyu obshchetoksicheskogo dejstviya lekarstvennyh sredstv. Izuchenie ostroj toksichnosti, Moskva: Grif i K [Guidelines for conducting preclinical studies of drugs. Part 1. Guidelines for the study of the general toxic effect of drugs. Acute toxicity study, Moscow: Grif and K] 15-17 (2012). [in Russian]

13. OECD, Test No. 420:2001 «Ostraya toksichnost' pri vnutrizheludochiom postuplenii. Metod fiksirovannoj dozy» («Acute Oral Toxicity - Fixed Dose Procedure», IDT) [OECD Test No. 420:2001 "Acute intragastric toxicity. Fixed Dose Method" ("Acute Oral Toxicity - Fixed Dose Procedure", IDT)]. [in Russian]

14. Lim R.R. et al. A method for the evaluation of cumulation and tolerance by the determination of acute and subchronic median effective doses, Arch. Intern.Pharm. Ther., 130, 336-352 (1961).

15. Osnovy obshchej promyshlennoj toksikologii (rukovodstvo) [Fundamentals of general industrial toxicology (manual)] (Leningrad: Medicina, 1976, 304 s.) [Leningrad: Medicine, 1976, 304 p.]. [in Russian]

16. Vale E.P., Rego L.R. do, Pureza D.D.N, Silva P.G. Cytogenetic and toxicological effects of Punica granatum Linnaeus fruit peel hydroethanolic extract in mice, South Afr. J. Bot., 130, 465-470 (2020). DOI: https://doi.org/10.1016/j.sajb.2020.01.041.

17. Chedea V.S., Macovei S.O.,Bocsan I.C. Grape Pomace Polyphenols as a Source of Compounds for Management of Oxidative Stress and Inflammation-A Possible Alternative for Non-Steroidal Anti-Inflammatory Drugs? Molecules, 27, 6826 (2022). DOI: https://doi.org/ 10.3390/molecules27206826.

Information about authors:

Kantureyeva G.O. – doctoral student of the Department of Biotechnology, M. Auezov SKU, 5 Tauke-Khan Ave., Shymkent, Kazakhstan.

Saparbekova A.A. – Professor of the Department of Biotechnology, M. Auezov SKU, 5 Tauke-Khan Ave., Shymkent, Kazakhstan.

Kudasova D.E. – teacher of the Department of Biotechnology, M. Auezov SKU, 5 Tauke-Khan Ave., Shymkent, Kazakhstan.

Rakhmonova G.G. – Junior Research Fellow, Institute of Bioorganic Chemistry named after Academician A.S. Sadykov of the Academy of Sciences of the Republic of Uzbekistan, Tashkent, Uzbekistan.

Urazbayeva K.A. – Professor of the Department of Food Engineering, M. Auezov SKU, 5 Tauke-Khan Ave., Shymkent, Kazakhstan.

Кантуреева Г.О. – «Биотехнология» кафедрасының докторанты, М. Әуезов атындағы ОҚМУ, Тәуке-хан даңғылы, 5, Шымкент, Қазақстан.

Сапарбекова А.А. – «Биотехнология» кафедрасының профессоры, М. Әуезов атындағы ОҚМУ, Тәуке-хан даңғылы, 5, Шымкент, Қазақстан.

Кудасова Д.Е. – «Биотехнология» кафедрасының оқытушысы, М. Әуезов атындағы ОҚУ, Тәуке-хан даңғылы, 5, Шымкент, Қазақстан.

Рахмонова Г.Г. – кіші ғылыми қызметкер, академик А.С. Садықов атындағы Өзбекстан Республикасы ҒА Биоорганикалық химия институты, Ташкент, Өзбекстан.

Уразбаева Қ.А. – Тамақ инженерия кафедрасының профессоры, М. Әуезов атындағы ОҚУ, Тәуке-хан даңғылы, 5, Шымкент, Қазақстан.