

GOCE DELCEV UNIVERSITY - STIP
FACULTY OF AGRICULTURE



JOURNAL OF AGRICULTURE AND PLANT SCIENCES

YEAR 2022

VOLUME 20, Number 2

Journal of Agriculture and Plant Sciences, JAPS Vol. 20, No. 2

ISSN 2545-4447 print

ISSN 2545-4455 on line

Vol. 20, No. 2, Year 2022

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**GOCE DELCEV UNIVERSITY - STIP
FACULTY OF AGRICULTURE**

doi.org/10.46763/JAPS201

Indexed in EBSCO database

ISSN 2545-4447 print

ISSN 2545-4455 on line

Vol. 20, No. 2, Year 2022



Journal of Agriculture and Plant Sciences, JAPS, Vol 20, No. 2

YEAR 2022

VOLUME XX, Number 2

Editorial Office

Faculty of Agriculture, Goce Delcev University - Stip,

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INTRODUCTION

The publication of the new issue of JAPS, No. 20., Vol. 2. coincided with the warning from the World Health Organization that a new subvariant of the corona virus "Kraken", which is more transmissible than any before, is spreading around the world. Just when it seemed that the corona virus pandemic would finally subside, the XBB.1.5 virus appeared, and experts named it Kraken, after the legendary sea monster, a giant octopus that dragged ships to the bottom of the sea and killed the sailors. It is a subvariant of the omicron strain of the corona virus, which is already affecting America, and Kraken has quickly spread to Europe, but the first information indicates that it causes a mild clinical presentation, as well as the omicron variant. The scientists discovered that the spike proteins have 14 new mutations, which means that it more easily infects people, who already have immunity from corona, either through vaccination or previous illness. This makes them more susceptible to infection, but not to a serious clinical presentation with fatal consequences.

It seems that the world, in all segments of its functioning, has adapted to a new post-pandemic way of functioning. In the past three years, science was also not safe from the consequences of the pandemic. Particularly in the agricultural production sector, where all activities were aimed to adapt to the new conditions and challenges. Researchers certainly have not stopped their research activities, which is confirmed with the publication of current issue of JAPS, No. 20., Vol. 2.

The Editorial Board of JAPS, in the conditions of the covid-19 pandemic, had continuously published all journal issues in order to share with the scientific and professional community the new research results in the field of agricultural production and plant sciences. We are honored and pleased to share with you five peer-reviewed scientific papers in JAPS issue No. 20., Vol. 2 and to invite and encourage our colleagues from Republic of North Macedonia, the region and wider to publish their research results in future issues of JAPS.

Editorial Board,

December, 2022

Editor in chief,

Prof. Liljana Koleva Gudeva, PhD



ASSESSMENT OF RADIOLOGICAL HAZARD FOR VARIOUS FOOD COMMONLY USED IN REPUBLIC OF NORTH MACEDONIA

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Abstract

Consuming food containing radionuclides is particularly dangerous. If anyone ingests or inhales a radioactive particle, it continues to irradiate the body as long as it remains radioactive and stays in the body. However, studies on the radioactivity of consumable foods assume importance as it is necessary to estimate the ingestion dose to the public. Due to all this, the focus of this research was on determining the activity concentrations of ^{226}Ra , ^{40}K and ^{232}Th . Forty-nine samples in three categories of vegetables, cereals (rice, wheat, corn), and milk, were collected from local markets (city of Skopje) in the Republic of North Macedonia and they were analysed by using high-purity germanium (HPGe) detector to assess natural and artificial radioactivity. The average activity concentrations of ^{226}Ra , ^{40}K and ^{232}Th of the tested samples were 2.85 ± 1.15 , 2.48 ± 0.85 , and $80.64 \pm 5.45 \text{ Bq kg}^{-1}$, respectively. No artificial radionuclide was found in any of these samples. The average value of the radium equivalent activity in all samples was 12.56 Bq kg^{-1} , which was less than the maximum permitted value of 370 Bq kg^{-1} . The values of the external hazard indices for vegetables, cereals and milk samples vary with an average value of 0.11, which is less than one in all samples indicating the non-harmfulness of the samples. The mean activity concentrations of ^{226}Ra , ^{40}K and ^{232}Th (Bq kg^{-1}) in the samples were used to calculate the absorbed dose rate whose mean value for all food samples was 6.16 Bq kg^{-1} . It was determined that the measured values are within the globally accepted values, i.e., they are quite lower than the data in literature. These data would be useful to establish a baseline for natural radioactivity concentrations in food products consumed in the Republic of North Macedonia.

Key words: *natural spectrometry, food, gamma spectrometry, radiation risk*

INTRODUCTION

Humans are constantly exposed to natural radioactivity at different levels depending on the natural radioactive elements present in each area; therefore, researchers are studying natural environmental radiation and radioactivity in order to conduct background checks and detect environmental radioactivity (Radhakrishna et al., 1993). Considering that radionuclides occur naturally in both rocks and soils, when these rocks decompose by natural processes, radionuclides are transported to the soil by rain and lowlands (Agbalagba & Onoja, 2011; Essiett et al., 2015). By means of migration, the present radionuclides can easily accumulate in the food chain through the soil - plants and the human

(Skwarzec & Falkowski, 1988). Due to this migration, radioactivity has always been present in all food products to some extent. In general, anthropogenic radionuclides in food products originate from the effects introduced into the biosphere. In fact, radionuclides can cause a number of health conditions and diseases as a result of their exposure, hence resulting in bio-accumulation and bio-toxicity (Ferdous et al., 2015). Therefore, the IAEA and international experts (Natural and induced radioactivity in food IAEA-TECDOC-1287, 2002) are developing guidelines for measuring and determining acceptable levels of natural radioactivity in food, with the ultimate goal of improving food

safety. Many studies have been conducted on the concentration of radioactivity in food products (Alsaffar et al., 2015) (Angeleska et al., 2021) where the natural radioactivity in food is usually within the range from 40 to 600 Bq kg⁻¹ of food (IAEA, 2002). This work aims to identify radionuclides in certain food samples that are

commonly used in households in the Republic of North Macedonia, by measuring the amount of specific concentrations of ²²⁶Ra, ²³²Th and ⁴⁰K. Based on these results, the potential radiological health risks associated with the consumption of these analysed types of food were assessed.

MATERIAL AND METHODS

- Sample collection and preparation

A total of 66 samples of locally widely consumed food products were collected from various markets in the Republic of North Macedonia. The main food selection was based on a survey questionnaire on the diet of 30 residents. The main food groups were selected, including wheat, rice, and milk. The samples were prepared fresh, i.e., they were homogenized and packaged in standard Marinelli cups (500 g) and sealed in order to achieve a radioactive balance between the parents and their daughter radionuclides. (IEEE, 1997; Walley El-Dine et al., 2001)

- Sample analysis

The analytical techniques for gamma-ray spectrum were used for determination of the natural radionuclides ²²⁶Ra, ²³²Th and ⁴⁰K. This measurement system includes a typical high-resolution gamma-spectrometer based on a high-purity germanium detector with energy resolution of 1.80 keV full width at half maximum for the gamma ray line of 1332 keV from ⁶⁰Co. According to the energy of the photopic line, the activities of ²³²Th were measured by taking the mean activity of the photopeaks of the daughter nuclides ²²⁸Ac (338.40, 911.07 and 968.90 keV) and ²¹²Pb (238.63 keV). The activities of ²²⁶Ra were calculated with ²²⁶Ra. keV, line energy of ²¹⁴Pb. The activities of ⁴⁰K were determined directly from its gamma emission at 1460.83 keV. (Righi et al., 2009) The counting time for each sample was 108 000 seconds.

- The activity concentration

For all samples, the corresponding specific activities according to the methods of Shanthi were determined (Shanthi et al., 2009) as shown in Equation 1.

$$A = \frac{\frac{N}{t} - \frac{N_0}{t_0}}{\varepsilon \cdot \gamma \cdot m} \text{ (Bq} \cdot \text{kg}^{-1}) \text{ (1)}$$

Where, N is clean surface of peak accumulated from a specific radionuclide in analysis of a specific sample (number of readings), N₀ is clean surface of peak accumulated from the spot of a specific radionuclide without an analysis of sample (number of readings), t is live time of accumulation of the sample spectrum (s), t₀ is live time of accumulation of the phone spectrum (s), ε is detector efficiency for a given energy (for a specific peak), γ is intensity of gamma transition in radioactive decay for a respective radionuclide (%), and m is sample mass (kg).

- Radiological hazard assessment

- Assessment of Radium Equivalent

Gamma-ray radiation hazards caused by specific radionuclides of ²²⁶Ra, ²³²Th, and ⁴⁰K were evaluated using different indices, among which, the radium equivalent activity is the most widely used radiation hazard index (Beretka et al., 1985) It is determined by the weighted sum of activities of the three radionuclides, based on the assumption that 370 Bq, 259 Bq, and 481 Bq produce the same gamma-ray dose rate as given by (Kessaratikoon & Awaekuchi, 2008)

$$Ra_{eq} \text{ (Bq/kg)} = A_{Ra} + 1.43A_{Th} + 0.07A_K \text{ (2)}$$

where Ra, Th, and K are the activity concentrations of ²²⁶Ra, ²³²Th, and ⁴⁰K (in Bq kg⁻¹), respectively.

To keep the annual radiation dose below 1.5 mGy y⁻¹, the maximum value must be less than 370 Bq kg⁻¹.

- Absorbed Dose Rate in Air

The absorbed dose rates (nGy h⁻¹) in outdoor air from global gamma radiation at 1 m above the ground were analysed by the next equation (Özmen et al., 2014; Curtin et al., 2008)

$$D \text{ (nGy / h)} = 0,462 A_{Ra} + 0,604 A_{Th} + 0,0417 A_K \quad (3)$$

- External hazard index

The internal and external hazard indexes are calculated by the following expressions (Rafique et al., 2013)

$$H_{eks} = A_{Ra}/370 + A_{Th}/259 + A_K/4810 \leq 1 \quad (4)$$

A_{Ra} , A_{Th} , A_K specific activities (Bq/kg), ^{226}Ra , ^{232}Th and ^{40}K , respectively

- Ingestion effective dose

The Ingestion effective dose due to the intake of ^{238}U , ^{232}Th and ^{40}K in foods can be evaluated using the following expression ICRP (ICRP, 2006).

$$HT_r = \sum_i U_i \times C_{i,r} \times g_{T,r} \quad (5)$$

where, i denotes a food group, the coefficients U_i and $C_{i,r}$ denote the consumption rate (kg/y) and activity concentration of the radionuclide r of interest (Bq kg⁻¹), respectively, and $g_{T,r}$ is the dose conversion coefficient for ingestion of radionuclide r (Sv Bq⁻¹) in tissue T . For adult members of the public, the recommended dose conversion coefficient $g_{T,r}$ for ^{40}K , ^{226}Ra (^{238}U), and ^{232}Th , are 6.2×10^{-9} , 2.8×10^{-7} and 2.2×10^{-7} Sv Bq⁻¹ respectively IAEA (IAEA, 1996). In fact, the aggregate quantities for Average annual intake for adults in the Republic of North Macedonia by groups are: vegetables 44.5 kg, cereals 75.7 kg, legumes 8 kg and milk 35l kg.

- Annual effective dose equivalent (AEDE)

The annual effective dose equivalent received was computed from absorbed dose rate by applying a dose conversion factor of 0.7 Sv Gy⁻¹ and the occupancy of 0.8 (19/24 h) recommended by UNSCEAR. Therefore, the annual effective dose equivalent (μSv y⁻¹) was calculated using the equation 6 (UNSCEAR, 2000) AEDE (μSv y⁻¹) = absorbed dose (nGy h⁻¹) × 8760 h × 0.7 Sv Gy⁻¹ × 0.8 × 10⁻³ (6)

RESULTS AND DISCUSSION

Table 1. Mean values of specific activities (A) of values of ^{226}Ra , ^{232}Th and ^{40}K in foods.

Sampling sites	A±SD (Bq kg ⁻¹) ^{226}Ra	A±SD (Bq kg ⁻¹) ^{232}Th	A±SD (Bq kg ⁻¹) ^{40}K
Potatoes (n=5)	0.41±0.15	1.96±0.05	92.15±5.50
Tomatoes (n=5)	0.71±0.05	1.26±0.02	102.50±6.50
Beans (n=7)	0.24±0.01	0.95±0.02	69.70±3.00
Apple(n=7)	0.65±0.20	1.41±0.50	25.35±3.50
Rice (n=7)	6.02±2.20	3.90±1.50	67.55±2.55
Wheat (n=7)	0.62±0.05	0.25±0.03	155.54±5.30
Corn (n=7)	1.92±1.00	2.19±1.00	37.02±5.90
Pea (n=7)	12.56±6.09	9.47±4.20	78.85±7.50
Red Lentil (n=7)	3.58±1.52	2.38±1.01	142.87±10.25
Milk (n=7)	1.83±0.31	1.04±0.15	31.87±4.53
Average	2.85±1.15	2.48±0.84	83.52±5.46

The specific activity due to ^{226}Ra , ^{232}Th and ^{40}K in different types of food was measured as presented in Table 1. The specific activity of ^{226}Ra is found within the range from 0.24 ± 0.01 Bq kg⁻¹ to 12.56 ± 6.09 Bq kg⁻¹, the one of ^{232}Th from 0.25 ± 0.03 Bq kg⁻¹ to 9.47 ± 4.20 Bq kg⁻¹ and the specific activity of ^{40}K was within the range from 25.35 ± 3.50 Bq kg⁻¹ to 142.87 ± 10.25 Bq

kg⁻¹. There is a variation in the specific activity of radionuclides in different food samples where it can be seen that in wheat, the concentration value of ^{40}K is higher than in other food products. The highest concentration level of the ^{226}Ra and ^{232}Th radionuclides were in pea and red lentils, respectively. The activity concentration of ^{226}Ra in cereal crops (wheat and rice) was higher than

the UNSCEAR reference values of 0.08 Bq/kg (UNSCEAR, 2000). The source of contamination of ^{226}Ra , ^{232}Th and ^{40}K of rice and wheat is considered to be due to the absorption of soil by the plant roots and irrigation water through

the root and also the irrigation period of wheat and rice is longer. The obtained average activity concentration in milk for ^{40}K was 31.87 Bq kg^{-1} , which is lower when compared to other results (ICRP,1996).

Table 2. Obtained values from the absorbed dose rate in the air (D), the radiation risk index (H_{eks}), the radium equivalent (Ra_{eq}), and AEDE.

Sample	D (nGy h ⁻¹)	Ra_{eq} (Bq kg ⁻¹)	Hex	AEDE (μSv y ⁻¹)
Potatoes	5.21	9.66	0.02	25.55
Tomatoes	5.35	9.68	0.02	26.24
Beans	3.58	6.46	0.01	17.56
Apple	2.20	4.43	0.01	10.79
Rice	7.94	16.31	0.04	38.95
Wheat	6.91	11.85	0.03	33.89
Corn	3.74	7.64	0.02	18.34
Pea	14.79	31.61	0.08	72.55
Red Lentil	9.03	16.89	0.04	44.29
Milk	2.78	5.54	0.01	13.63
Average	6.15	12.07	0.03	30.17

In this study, the values of the absorbed dose rate D (nGy h⁻¹) did not exceed the safety limits, emphasizing the negligible radiation hazard arising from naturally occurring terrestrial radionuclides. The radium equivalent activity values in all food samples vary with a mean value of 12.07 Bq kg^{-1} which is far below the internationally accepted value of

370 Bq kg^{-1} . On the other hand, the values of the hazard index varied from 0.01 to 0.08 with an average value of 0.03. The annual effective dose equivalent has been calculated from $10.79 \mu\text{Sv y}^{-1}$ to $44.29 \mu\text{Sv y}^{-1}$ with an average value of $30.17 \mu\text{Sv y}^{-1}$ respectively which is less than the recommended value of the IAEA, which is $1000 \mu\text{Sv y}^{-1}$.

Table 3. Estimated radiation hazard indices and ingestion effective dose in food at the markets in North Macedonia.

Sample	^{226}Ra	^{232}Th	^{40}K	Sum
Potatoes	0.0051	0.019	0.025	0.044
Tomatoes	0.0088	0.012	0.028	0.048
Beans	0.0005	0.001	0.003	0.004
Apple	0.008	0.013	0.006	0.027
Rice	0.013	0.006	0.031	0.050
Wheat	0.013	0.004	0.070	0.087
Corn	0.040	0.036	0.017	0.093
Pea	0.028	0.016	0.003	0.047
Red Lentil	0.008	0.004	0.007	0.019
Milk	0.001	0.008	0.006	0.015

Table 3 shows the results of the effective dose of ingestion in mSv y⁻¹ for an adult person due to specific activity of ^{226}Ra , ^{232}Th and ^{40}K in food samples calculated by using the equation (4). The sum of the effective ingestion doses varies from 0.004 mSv y^{-1} (in the beans sample) to 0.093 mSv y^{-1} (in the corn sample). The

average effective ingestion dose due to ^{40}K was higher than due to ^{232}Th and ^{40}K as a result of the increased dose conversion ratio for radionuclide ingestion. This indicates that the effective ingestion dose in all food samples is lower than the permitted limits of 1 mSv y^{-1} recommended by the ICRP.

CONCLUDING REMARKS

The activity of the ^{226}Ra , ^{232}Th and ^{40}K radionuclides was determined for the most available food products consumed in the Republic of North Macedonia. It was determined that the measured values are within the world range as published in this literature. The mean activity values of the ^{226}Ra , ^{232}Th and ^{40}K radionuclides in 66 food products were identified as $2.85 \pm 1.15 \text{ Bq kg}^{-1}$, $2.48 \pm 0.84 \text{ Bq kg}^{-1}$ and $83.52 \pm 5.46 \text{ Bq kg}^{-1}$, respectively. All calculated values for radiological hazard assessment are lower than the global average values. The dose of different

components reduces in the following order corn>wheat>rice>vegetables>milk. It should be noted that the geology of the region and the food processing methods have a major impact on the ^{40}K concentration. This study established a map of basic information about the future studies on radiation levels and the distribution of radionuclides in food products in the Republic of North Macedonia. The results of the study will also be used as a reference for future assessment.

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ОЦЕНКА НА РАДИОАКТИВНАТА ОПАСНОСТ ОД РАЗЛИЧНА ХРАНА КОЈА Е НАЈЧЕСТО КОРИСТЕНА ВО РЕПУБЛИКА СЕВЕРНА МАКЕДОНИЈА

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Резиме

Консумирањето храна што содржи радионуклиди е особено опасно. Ако некој внесе или вдиши радиоактивна честичка, таа продолжува да го зрачи телото сè додека останува радиоактивна и останува во телото. Сепак, студиите за радиоактивноста на потрошната храна имаат значење бидејќи е неопходно да се процени дозата за голтање на јавноста. Поради сето ова, фокусот на ова истражување беше на одредување на концентрациите на активност на ^{226}Ra , ^{40}K и ^{232}Th . Четириесет и девет примероци во три категории зеленчук, житарки (ориз, пченица, пченка) и млеко беа собрани од локалните пазари (Град Скопје) во Република Северна Македонија и беа анализирани со употреба на германиум со висока чистота (HPGe) детектор за проценка на природна и вештачка радиоактивност. Просечните концентрации на активност на ^{226}Ra , ^{40}K и ^{232}Th од тестираните примероци беа $2,85 \pm 1,15$, $2,48 \pm 0,85$ и $80,64 \pm 5,45 \text{ Bq kg}^{-1}$, соодветно. Во ниту еден од овие примероци не беше пронајден вештачки радионуклид. Просечната вредност на активност на еквивалент на радиум во сите примероци беше $12,56 \text{ Bq kg}^{-1}$, што беше помало од максималната дозволена вредност од 370 Bq kg^{-1} . Вредностите на надворешните индекси на опасност за примероците од зеленчук, житарки и млеко варираат со просечна вредност од 0,11, што е помалку од една во сите примероци што укажува на нештетноста на примероците. Просечните концентрации на активност на ^{226}Ra , ^{40}K и ^{232}Th (Bq kg^{-1}) во примероците беа искористени за пресметување на стапката на апсорбирана доза чија средна вредност за сите примероци на храна беше $6,16 \text{ Bq kg}^{-1}$. Утврдено е дека измерените вредности се во рамките на глобално прифатените вредности, односно се доста пониски од податоците во литературата. Овие податоци би биле корисни за утврдување на основната линија за концентрациите на природна радиоактивност во прехранбените производи што се консумираат во Република Северна Македонија.

Клучни зборови: природна радиоактивност, храна, гама спектрометрија, ризик од зрачење



USE OF *Orius laevigatus* TO CONTROL *Frankliniella occidentalis* (THYSANOPTERA: THIRIPIDAE) POPULATION IN GREENHOUSE PEPPER

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Abstract

Although chemical pesticides play a vital role in controlling the number of harmful insects, they also contribute to accelerate pollution of soil, air and water. Due to the frequent use, insects become resistant to active ingredients very quickly; they destroy the natural enemies of the pests and have a harmful effect on humans. Accordingly, the application of biological protection, that is, the use of living organisms (predators and parasites) in plant protection programmes in protected areas, takes on a larger scale worldwide rather than the use of chemical pesticides.

The aim of our research was determining the effectiveness of pirate bug *Orius laevigatus* (Hemiptera: Anthocoridae) on reducing the population of Western flower thrips (*Frankliniella occidentalis*). The experiment was set in commercial greenhouses (3 unheated plastic tunnels, ca. 125 m², each), located in the area of Dabilje, Republic of Macedonia, during 2019 and 2020.

The results obtained correspond to our expectation in controlling the population of the trips. Predator proved to be effective in reducing the number of thrips population.

Key words: predators, biological control, western flower thrips, pirate bug

INTRODUCTION

Western flower thrips, *Frankliniella occidentalis* Pergande (Thysanoptera: Thripidae) is an important pest of pepper, and a broad range of vegetable and ornamental crops in greenhouses and fields (Tavella et al., 1991; Tommasini and Maini, 1995; Kirk and Terry, 2003). That very small insect, commonly hides in flowers, buds and leaf axils, and often go unnoticed until damage appears. Both larval and adult thrips have rasping mouthparts that they use to puncture the plant surface. They feed on the sap that is exuded from the resulting wound. Plants are also injured when female thrips lay their eggs in the plant tissue. Western flower thrips is of special importance because it transmits the tomato spotted wilt virus (TSWV) and impatiens necrotic spot virus (INSV), that are most epidemic on a wide range of the agricultural crops (Jones, 2005; Rotenberg et al., 2009).

Controlling greenhouse pests by chemical pesticides results with many problems, such as development of resistance in pests and rising environmental and health concerns (Arnaouty et al., 2020). Indeed, greenhouse crops are harvested frequently, at short intervals, and thus intensive use of chemicals becomes questionable because of the possible contamination of products with chemical residues. Furthermore, greenhouse vegetables are consumed fresh, which is another motivation for farmers to reduce intensive chemical control and to meet the consumers demands for offering products of high quality.

The possibility to apply biological control programmes against greenhouse pests is highly needed. It can overcome the abovementioned problems, and at the same time can provide an adequate pest control. They will not completely eliminate pest problems but can reduce pest

population, and damage to an acceptable level (under the economical threshold). Biological control generally requires more time than pesticides to bring a pest population under an acceptable control level (Arnaouty et al., 2020).

The aim of the present study was to

determine the effectiveness of the pirate bug *O. laevigatus* (Hemiptera: Anthocoridae) on reducing the population of Western flower thrips (*Frankliniella occidentalis*) at peppers, grown for commercial production in plastic, unheated tunnels, during 2019 and 2020.

MATERIAL AND METHODS

The study was conducted in commercial greenhouses (3 unheated plastic tunnels, ca. 125 m², each), located in the area of Dabilje, Republic of North Macedonia, during 2019 and 2020. One of the tunnels was used for biological control, one for chemical control and one was untreated tunnel (control). Each tunnel included 9 rows of 75 pepper plants (675 plants/tunnel) of Kurtovska kapija type. The transplanting of pepper started at the beginning of May, and the growing season extended to the end of September, 2019 and 2020, respectively.

Population density of *F. occidentalis* was estimated in intervals of 15 days throughout the growing season of pepper, counting nymphs and adults. Fifty randomized plants from each tunnel were chosen and the thrips were sampled from 50 plant flowers. The number of thrips was directly inspected on the plant using a special magnifying hand lens (x 10).

In the tunnel used for biological control, a blend of nymphs and adults of *O. laevigatus* were released at rate of 1 predator per m², when thrips appeared on the plants. Three following releases were carried out in the both 2019 and 2020 years of research (Table 1). *O. laevigatus* applied, came from Bioline Agrosiences Ltd., United Kingdom.

Table 1. Releasing rates and dates of tested *O.laevigatus* against *F. occidentalis* on pepper plants during 2019 and 2020.

<i>Oriuslaevigatus</i>	Rate of application	2019			2020		
		Date of first releasing	Date of second releasing	Date of third releasing	Date of first releasing	Date of second releasing	Date of third releasing
	1 individual/m ²	18.V	8.VI	1.VIII	17.V	7.VI	5.VII

In the chemical treated tunnel, 3 pesticides were applied against *F. occidentalis* and the timing and rate of applications of different

pesticides were determined by the grower, based on his assessment of pest populations (Table 2).

Table 2. A list of pesticides applied to control *F.occidentalis* on sweet pepper pests in the chemical treated tunnel during 2019 and 2020.

Application time	Active ingridient	Application rate/100 L
2019		
18.V	Acrinathrin 22,5 g/L +abamectn 12,6 g/L	100 ml
27.V	Abamectin 18 g/L	100 ml
5.VI	Pyrethrin 50 g/L	100 ml
14.VI	Acrinathrin 22,5 g/L +abamectn 12,6 g/L	100 ml
23.VI	Abamectin 18 g/L	100 ml
2.VII	Pyrethrin 50 g/L	100 ml
11.VII	Acrinathrin 22,5 g/L +abamectn 12,6 g/L	100 ml
20.VII	Abamectin 18 g/L	100 ml
30.VII	Pyrethrin 50 g/L	100 ml
8.VIII	Acrinathrin 22,5 g/L +abamectn 12,6 g/L	100 ml
17.VIII	Abamectin 18 g/L	100 ml
27.VIII	Pyrethrin 50 g/L	100 ml

2020		
17.V	Acrinathrin 22,5 g/L + abamectin 12,6 g/L	100 ml
26.V	Abamectin 18 g/L	100 ml
4.VI	Pyrethrin 50 g/L	100 ml
13.VI	Acrinathrin 22,5 g/L + abamectin 12,6 g/L	100 ml
22.VI	Abamectin 18 g/L	100 ml
1.VII	Pyrethrin 50 g/L	100 ml
10.VII	Acrinathrin 22,5 g/L + abamectin 12,6 g/L	100 ml
19.VII	Abamectin 18 g/L	100 ml
29.VII	Pyrethrin 50 g/L	100 ml
7.VIII	Acrinathrin 22,5 g/L + abamectin 12,6 g/L	100 ml
16.VIII	Abamectin 18 g/L	100 ml
26.VIII	Pyrethrin 50 g/L	100 ml

The number of thrips for each treatment was subjected to analysis of variance (ANOVA) (SPSS). The significance of differences among

the number of thrips in different treatments was tested with LSD test at $P \leq 0.05$ significance level.

RESULTS AND DISCUSSION

The first thrips were found on the plants during second half of May, in both 2019 and 2020. Most of the thrips were found on pepper flowers and occasionally on the leaves. Berlinger et al. (1997) found that *F. occidentalis* is mainly attracted by the flowers than leaves. Higgins (1992) found that, in British Columbia (Canada), more than 85% of *F. occidentalis* larvae were found on leaves, and the majority (84-95%) of adults in flowers was females.

The mean number of thrips/flowers is (31.34 in 2019 and 32.16 in 2020) at the last week of the growing season (Table 3).

presented in Table 3. The numbers of thrips/flowers in the biological controlled tunnel and in chemical treated tunnel were lower than in the untreated greenhouse (control). Thrips infestation started in May, in both years of research 2019 and 2020, when the number of thrips/flowers was similar in the three experimental greenhouses. In the control, the population density of the thrips increased and continued to grow until the end of the season to reach the highest number of thrips/flower

Table 3. Number of thrips/50 flowers in the sweet pepper tunnels during 2019/2020.

Days of inspection	Control	Biologically controlled tunnel	Chemically controlled tunnel
2019			
17.V	9.04	8.66	8.48
31.V	13.16	7.72	6.50
7.VI	16.90	4.12	5.50
21.VI	19.56	6.10	4.26
5.VII	22.18	5.16	3.68
19.VII	24.12	3.88	3.16
2.VIII	27.56	4.56	2.26
16.VIII	28.64	3.08	1.94
30.VIII	31.34	2.16	1.48
2020			
16.V	12.98	12.58	12.68
30.V	14.68	9.94	10.26
6.VI	18.70	8.08	9.38
20.VI	22.86	11.36	7.50

4.VII	25.30	8.34	5.26
18.VII	26.46	6.50	4.46
1.VIII	28.36	7.74	3.56
15.VIII	30.62	4.20	2.04
29.VIII	32.16	2.50	1.70

O. laevigatus proved to be an efficient predator in maintaining the number of thrips under the economic threshold. According to Ramchandra and Chang (2013) the number of thrips under the economic threshold is 4.9 individuals/flower. In our research, the lowest number of thrips/flowers, recorded in the biologically controlled tunnel was 2.16 thrips/flower in 2019 and 2.50 thrips/flower in 2020. In the chemically controlled tunnel, the lowest number of thrips/flowers was 1.48 thrips/flower and 1.70 thrips/flower in 2020, what was expected after the application of the insecticides.

In the biologically controlled tunnel, the number of thrips/flower reduced to the economic threshold in the third week after the first release of *O.laevigatus*, at the rate of 1 adult/m² in 2019. After the second and the third release of *O.laevigatus*, the number of thrips/flower continued declines. (Graph. 1). In 2020 the number of thrips/flower reached 4.20, which is below the economic threshold according to Ramchandra and Chang (2013), after the third release of 1 adult/m² of *O.laevigatus* (Graph.2). Similar results were obtained by Keçeci and Gürkan, in 2013 Arnó et al. (2008) reported that *Orius* species could serve as an important biological control agent for use in sweet pepper.

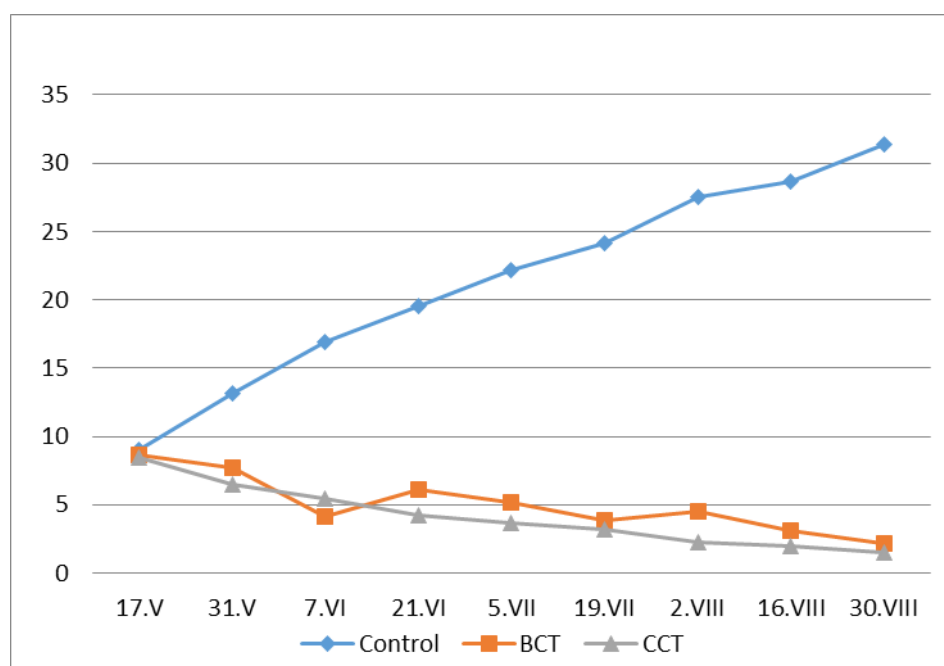


Figure 1. Mean number of thrips/50 flowers for different treatments in 2019

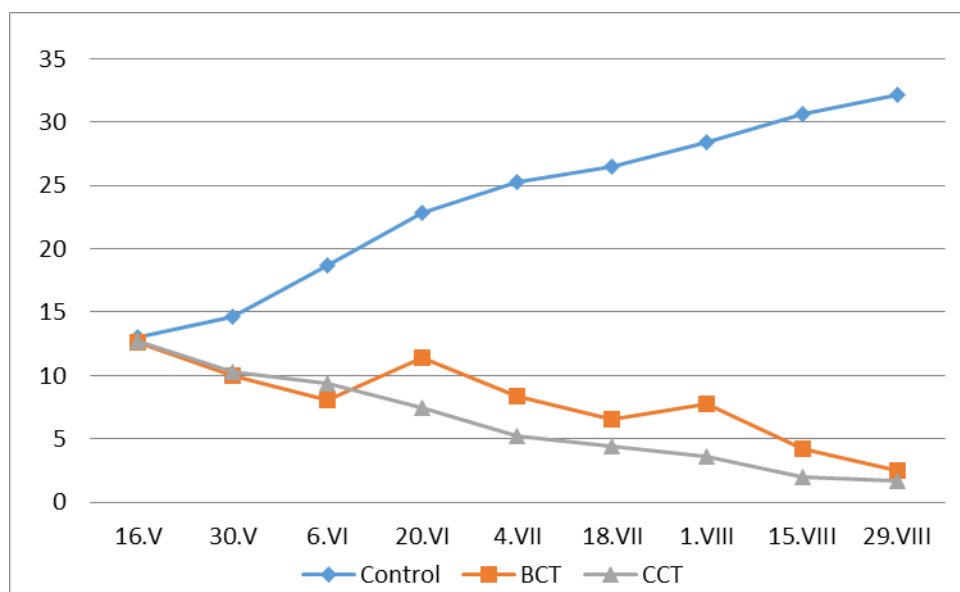


Figure 2. Mean number of thrips/50 flowers for different treatments in 2020

Statistically, insignificant differences were found between the biologically and chemically controlled tunnels in both years of research. There was significant difference between the

control and the chemically controlled tunnel and between control and biologically controlled tunnel in years of research, 2019 and 2020 (Table 4).

Table 4. Analysis of variance (SPSS) and multiple comparisons between the treatments and the number of thrips/50 flowers in 2019 and 2020.

ANOVA						
		Sum of Squares	df	Mean Square	F	Sig.
Number of thrips/50 flowers for different treatments in 2019	Between Groups	1696.150	2	848.075	39.108	<.001
	Within Groups	520.449	24	21.685		
	Total	2216.598	26			
Number of thrips/50 flowers for different treatments in 2020	Between Groups	2039.879	2	1019.940	38.172	<.001
	Within Groups	641.264	24	26.719		
	Total	2681.143	26			
MULTIPLE COMPARISONS						
LSD						
Dependent Variable	(I) Treatments	(J) Treatments	Mean Difference (I-J)	Std. Error	Sig.	
Number of thrips/50 flowers for different treatments in 2019	Control	BCT	16.34056*	2.19521	<.001	
		CCT	17.24944*	2.19521	<.001	
	BCT	Control	-16.34056*	2.19521	<.001	
		CCT	.90889	2.19521	.683	
	CCT	Control	-17.24944*	2.19521	<.001	
		BCT	-.90889	2.19521	.683	
Number of thrips/50 flowers for different treatments in 2020	Control	BCT	18.77333*	2.43672	<.001	
		CCT	18.08444*	2.43672	<.001	
	BCT	Control	-18.77333*	2.43672	<.001	
		CCT	-.68889	2.43672	.780	
	CCT	Control	-18.08444*	2.43672	<.001	
		BCT	.68889	2.43672	.780	

*The mean difference is significant at the 0.05 level

CONCLUDING REMARKS

In the present study releasing *O. laevigatus* showed to be effective and safe compared to the chemical control program under the same circumstances. The pirate bug reduced *F. occidentalis* individuals below the economic threshold and can be used effectively to decrease or even to completely replace the chemical treatments in pepper production in plastic tunnels.

The results showed that there are statistically significant differences between the population of *F. occidentalis* in the tunnel

with performed biological control using *O. laevigatus* and the control, as well as between the population of *F. occidentalis* in the tunnel with performed chemical treatments and the control. No statistically significant differences were observed between the population of *F. occidentalis* in tunnels with biological and chemical control.

So, we can recommend *O. laevigatus* for controlling *F. occidentalis* on pepper plantations in the greenhouses.

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**УПОТРЕБА НА *Orius laevigatus* ЗА КОНТРОЛА НА ПОПУЛАЦИЈАТА НА *Frankliniella occidentalis*
(THYSANOPTERA: THIRIPIDAE) КАЈ ПИПЕРКА ВО ЗАШТИТЕН ПРОСТОР**

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Резиме

Иако хемиските пестициди играат витална улога во контролирањето на бројот на штетни инсекти, тие исто така придонесуваат за забрзување на загадувањето на почвата, воздухот и водата. Поради честата употреба инсектите многу брзо стануваат отпорни на активни материи, ги уништуваат природните непријатели на штетниците, а штетно влијаат и врз луѓето. Соодветно на тоа, примената на биолошката заштита, односно употребата на живи организми (предатори и паразити) во програмите за заштита на растенијата во заштитените подрачја, зазема поголем обем во светски рамки отколку употребата на хемиски пестициди.

Целта на нашето истражување беше да се утврди ефикасноста на предаторот *Orius laevigatus* (Hemiptera: Anthocoridae) за намалување на популацијата на западниот цветен трипс (*Frankliniella occidentalis*). Експериментот беше поставен во комерцијални оранжерии (три тунели без греење, околу 125 m², секој), лоцирани во областа Дабиле, Република Македонија, во текот на 2019 и 2020 година.

Добиените резултати одговараат на нашите очекувања во контролирањето на популацијата на патувањата. Предаторот се покажа како ефикасен во намалувањето на бројот на популацијата на трипсот.

Клучни зборови: *предатори, биолошка контрола, западен цветен трипс, пиратска бубачка*



FEEDING OF LAMBS WITH IMPROVED PELLETED FEED

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Abstract

The feed costs are the major expenses for the farmers and the main input that influenced the economics of livestock production. The physical form of diet is one of the main drivers for better digestibility of feed and improved growth performance of farm animals. This study aimed to evaluate the effect of using two different physical forms of diet (ground feed mixtures and pelleted feed mixtures) on the growth performance of fattening lambs. Seventy-two fattening lambs were randomly assigned to two groups with 36 fattening lambs to assess the effects of feeding with pelleted (test group) and ground feed mixtures (control group) on growth performance. Two physical forms (pelleted vs. ground) were equal in ingredient and chemical composition. The duration of the experiment was 42 days. At the beginning of the trial, the lambs in the test group had lower average live body weight and daily gain compared with the lambs in the control group. However, at the end of the feeding trial, the lambs feeding with a pelleted feed mixture had achieved a higher average live body weight and better daily gain. The results from the ANOVA for growth performance of lambs depending from the physical form of diet indicate that there was a statistically significant difference ($p < 0.05$) between the test and control groups of lambs in the 5th and the 6th week of the feeding trial. These results support the possibility of using pelleted feeds in intensive fattening of lambs for improving the growth performance, increasing average daily gain and reducing the duration of the fattening period.

Keywords: *fattening lambs, diet, growth performance*

INTRODUCTION

Sheep breeding and fattening of lambs for export have a long tradition in North Macedonia. It is an essential source of income for sheep farmers that require minimum investment, care and management. According to data from the Statistical Yearbook of the Republic of North Macedonia (2021), the sheep population in North Macedonia is around 700.000 sheep with the production of around 3.183 tons of sheep meat. Several factors affect the economy of sheep husbandry. One of them is nutrition, which in the total production costs per unit of livestock product contributes up to 80%. The healthy breeding of livestock and the production of high-quality animal products require a stable supply of complete feed (Xu et

al., 2017). In the current intensive rearing system for lambs in many countries are usually fed ground concentrates and roughages in separate or a loose total mixed ration (TMR) (Joy et al., 2008; Rodríguez et al., 2008). The productivity of lambs can be increased by improving nutrition through supplementation of concentrates or compound feed. Complete pelleted feed is a good option in this regard to ensure balanced nutrition for the lambs in intensive rearing system since pellet feeding gives nutrition to lambs in a balanced form (Islam et al., 2017; Zhong et al., 2018). Additionally, seasonal oscillations and climatic variability also affected forage quality, quantity and availability. Moreover, low-quality feed supply has hindered lambs' capacity to

achieve their productivity potential (Nardone et al., 2010; Sultana et al., 2011). The ration's characteristics, such as ingredients, shape, smell, taste, and particle size, could affect the palatability and animals' feed intake. In addition, the physical form of rations is extremely important for efficiency and can affect rumen fermentation. Compared with ground feed, pelleted feed has been regarded as an efficient form for improving the intake, digestibility, feed conversion, and reducing animal ingredient selection. Thus, better digestion and usability of nutrients enhance animal growth and lead to better conversion into animal products (Blanco et al., 2014). Pelletized feed is defined as "agglomerated feedstuff" formed by grinding and extruding individual feed or mixtures (Zimonja et al., 2007), by compaction and passing through sieve openings in a mechanical process. Mechanical pressure can partly break down complicated fibre structures and promote starch gelatinization resulting in increases of feed voluntary intake and nutritional digestibility. These pellet products

are easier to handle, tastier, more digestible and usually result in improved feeding results when compared to non-pelleted animal feed (Zhao et al., 2011). Long-term conditioning of pellets (85°C, for 3 minutes) has a positive effect on their physical quality, reducing power fraction (Svihus and Zimonja, 2011), and improving the hygienic quality of pelleted feeds (Peisker, 2006; Jones, 2011).

Due to the limited natural resources in North Macedonia, and in order to achieve the benefit of the produced compound feed, the present experiment was designed to develop a total mixed ration based on complete pelleted feed and evaluate its utilization for commercial lamb fattening under intensive or stall-fed condition. It was hypothesized that feeding the lambs with complete pelleted feed (pelleted Total Mixed Ratio - PTMR) can enhance growth performance by improving the daily feed intake, average daily gain and feed conversion ratio compared to feeding lambs with ground fodder mixtures (unpelleted Total Mixed Ratio – UPTMR).

MATERIALS AND METHODS

After the total mixing of ingredients, the preparation of the pelleted feed was done in the pelleting machine. The two dietary treatments were PTMR and UPTMR. Both diets had the same chemical and ingredient composition. The approximate diameter of the complete pellet was 6-8 mm.

The feed chemical analysis was done at the Faculty of Agricultural Sciences and Food, Ss. Cyril and Methodius University in Skopje, Republic of North Macedonia. Before the chemical analysis pellet and other feed samples of the experimental diets were dried and ground in a grinding mill through 2-mm sieve. The crude protein (CP) content of the experimental diet was determined using the Kjeldahl method (MKC EN ISO 5983- 1:2010 corrigendum). The moisture content of the feed was determined by drying the samples at 105°C overnight (MKC ISO 6496:2012), while ash was measured by burning further at 500°C for 4 hours (MKC ISO 5984:2012). Solvent extraction methods were used for crude fat analysis (MKC ISO 6492:2012). Crude fibre determination in animal feed was done according to the standard MKC EN ISO 6865:2010.

Seventy-two healthy lambs from the indigenous Ovchepolian breed at approximately 15 to 20 days of age with similar body live weights were randomly allocated into two pens with 36 lambs in each pen. The pens were randomly assigned to two dietary treatments. The control treatment group was fed with unpelleted feed while the test treatment group received a pelleted feed. This experiment consisted of a 42 days fattening period for data collection. Experimental diets were formulated to meet the lambs' nutrient requirements for growing and fattening lambs (Feeding Standard for lambs in growing and fattening according to the Rule book for feed quality in the Republic of North Macedonia, Official Gazette 54/2014). The diet compositions and nutritional contents are shown in Table 1. Sufficient diets were made in one batch to make sure there was no batch effect on dietary treatments. During the whole experimental period, all lambs had free access to the assigned diets. Diets were offered ad libitum. All the animals had free access to fresh tap water. During the whole experimental period, all lambs in a pen were individually measured on days 0, 7, 14, 21, 28, 35 and 42.

Average daily gain (ADG) was calculated for day 0-7 (week 1); 7-14 (week 2); 14-21 (week 3); 21-28 (week 4); 28-35 (week 5) and 35-42 (week 6) by dividing the difference of measured weights by the period interval. The consumed feed by groups were calculated to determine average daily feed intake ADFI. Feed conversion ratio

(FCR) was calculated by dividing ADFI with ADG.

Using one-way analysis of variance (ANOVA), the statistically significant differences were tested in the growth performance of lambs in test and control group, depending from the physical form of diet.

RESULTS AND DISCUSSION

Table 1 shows the diet compositions and nutritional contents that were used for fattening

lambs during the trial. The pelleted and ground feed had the same ingredient composition.

Table 1. Ingredient composition of experimental mixtures.

Diet Ingredients (% or gr/100gr)	Diet for growing and fattening lambs under 15 kg live weight	Diet for growing and fattening lambs from 15-30 kg live weight
Maize	37.00	36.00
Barley	30.00	42.00
Soybean meal	23.00	12.00
Alfalfa hay	8.00	8.00
Vitamin-mineral premix	2.00	2.00
Total (%)	100.00	100.00
Metabolizable energy, OU/kg DM	1.33	1.31
Crude protein	18.11	14.13

Table 2. Chemical composition of experimental mixtures

Feed	Chemical composition of mixture				
	Moisture, %	Ash, %	Crude proteins, %	Crude fat, %	Crude fiber, %
Un-pelleted ground feed	12.71	4.17	18.16	4.02	4.93
Pelleted feed	12.65	7.37	18.11	4.10	5.08

The lambs in the test group were included in the experiment at the approximate age of 15.80 days, while the lambs in the control group were older 5 days in average (20.68 days).

Respectively, the lambs in the control group at the start of the experiment were a little bit heavier than the lambs in the test group (Table 3 and Table 4).

Table 3. Structure of the lamb population involved in the trial.

Period	Groups	N	Days of age	Days in feeding
Initial point	Test	36	15.80	0
	Control	36	20.68	
week 1	Test	36	22.80	7
	Control	36	27.68	
week 2	Test	36	31.80	14
	Control	36	36.68	
week 3	Test	36	38.80	21
	Control	36	43.68	
week 4	Test	36	45.80	28
	Control	36	50.68	
week 5	Test	36	52.80	35
	Control	36	57.68	
week 6	Test	36	59.80	42
	Control	36	64.68	

Table 4. Growth performance of lambs.

Period	Group of lambs	Average lamb live weight (kg)	Weekly weight gain of group (kg)	Daily weight gain of lamb (kg)
Initial point	Test	13.30±1.79		
	Control	13.76±2.21		
week 1	Test	14.90±2.35	57.46	0.32
	Control	14.98±2.28	58.32	0.34
week 2	Test	16.98±2.50	75.02	0.30
	Control	17.27±2.39	82.37	0.33
week 3	Test	19.28±2.03	82.94	0.33
	Control	19.24±1.89	76.61	0.28
week 4	Test	21.59±3.01	82.94	0.33
	Control	21.55±2.71	77.43	0.33
week 5	Test	24.73±2.06	98.71	0.39
	Control	24.17±1.97	92.04	0.37
week 6	Test	27.87±3.34	118.87	0.47
	Control	27.14±3.50	111.94	0.42

However, at the end of the experiment, the lambs in the test group achieved a higher daily weight gain and better feed conversion rate than the lambs in the control group (Table 4 and Table 5). Higher daily weight gain was

observed in the pelleted-fed group indicating that pelleting increased the digestibility and non-selectivity in feed intake. A similar result was also found by Roy et al. (2010) and Ahmed et al. (2020).

Table 5. Feed intake and feed conversion rate in groups of lambs.

Period	Group of lambs	Total weekly feed intake by group (kg)	Total daily feed intake by group (kg)	Daily feed intake by lamb (kg)	Feed conversion rate (kg)
S t a r t point	Test	88.20	12.60	0.35	
	Control	100.80	14.40	0.40	
week 1	Test	88.20	12.60	0.35	1.54
	Control	100.80	14.40	0.40	1.73
week 2	Test	88.20	12.60	0.35	1.18
	Control	100.80	14.40	0.40	1.22
week 3	Test	141.75	20.25	0.56	1.71
	Control	148.68	21.24	0.59	1.94
week 4	Test	141.75	23.46	0.65	1.98
	Control	153.72	25.66	0.71	2.32
week 5	Test	166.95	34.27	0.95	2.43
	Control	181.44	36.29	1.01	2.76
week 6	Test	177.03	49.08	1.36	2.89
	Control	189.00	49.89	1.38	3.12

The results from the ANOVA for the growth performance of lambs depending from the physical form of diet indicate that there was a statistically significant difference ($p < 0.05$)

between the test and control group of lambs in the 5th and the 6th week of the feeding trial (Table 6).

Table 6. Effects of feeding treatment on growth performance of fattening lambs.

Dependent variable: Groups of lambs related to the type of diet			
Source of variation	df between groups	df in groups	F-value
Initial control	1	70	0.428 ^{NS}
Control week 1	1	70	0.896 ^{NS}
Control week 2	1	70	0.681 ^{NS}
Control week 3	1	70	0.955 ^{NS}
Control week 4	1	70	0.651 ^{NS}
Control week 5	1	70	0.046*
Control week 6	1	70	0.045*

Ruminants are able to select rations according to their needs or preferences (Askar et al., 2006). Feeding PTMR is a great method to prevent lambs selecting rations when comparing to traditional feeding systems (feeding concentrate and long fibre separately). Although pelleting increases feed production costs by at least 10% (Jahan et al., 2006), it greatly expands potential sources of fibrous feedstuff, saves storage space and labour costs, which promotes development of a precision animal feeding industry. The farmer can choose locally available, cheaper roughage for pelleting according to season, region, and market to reduce production costs.

Feeding PTMR increased growth rate of fattening lambs in the present study, largely attributed to better FCR. The increased ADG (50

g/d) was consistent with the findings of Coufal-Majewski et al. (2017) and Zhang et al. (2019), in which they found that fattening lambs fed completely pelleted mixtures had around 60 g/d greater ADG than those fed mash diets. Consistent with the results of Blanco et al. (2014, 2015) obtained from fattening lambs, the increased ADG and daily feed consumption could reduce the duration of the fattening period, the most direct way to improve efficiency of sheep production in an extensive system. The results of this study demonstrated that feeding fattening lambs PTMR enhanced their growth performance in terms of ADG and better FCR. Zhong et al. (2018) found that feeding PTMR increased the feed consumption and growth rate of fattening lambs.

CONCLUDING REMARKS

In the intensive feedlot rearing system, providing stable and high-quality rations is necessary for improving animal growth performance and producing high-quality animal products. The use of ground feed mixtures in animal nutrition is associated with certain weaknesses that reduce the profitability of livestock breeders. The production of pelleted feed mixtures increases the competitiveness of both the feed producer and the animal breeder. Non-floury (pellets, flakes, popcorn forms) products have advantages in homogeneity,

density-hectolitre mass, and also have a longer shelf life, due to the reduced initial number of microorganisms (saprophytic and pathogenic) as a result of the technological process of pelleting, but also the smaller surface of the pellets which is exposed to contamination. Feeding pelleted instead of mash diet increased dry matter intake and average daily gain of fattening lambs without causing any health issues, consequently reducing the duration of the fattening period. This feeding regime is beneficial to increase profitability.

FUNDING

This article is based as a result of the research within the project funded by the Fund for innovation and technology development

of the Republic of North Macedonia under the instrument for Co-financing grants for newly established companies „start-up“ and „spin-off“.

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ИСХРАНА НА ЈАГНИЊА СО ПОДОБРЕНИ ПЕЛЕТИРАНИ КРМНИ СМЕСИ

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Резиме

Исхраната на фармските животни претставува најголемиот трошок за фармерите и главен фактор кој влијае врз економиката на сточарското производство. Физичката форма на добиточната храна има значајно влијание врз искористливоста на хранливите материи и постигнување на добри производни резултати во сточарското производство. Главна цел на истражувањето беше воведување на подобрена и избалансирана пелетирана крмна смеса за гоеење на јагниња. За следење на производните резултати во гоеењето на јагнињата беа формирани две групи по 36 јагниња. Јагнињата во тест групата беа хранети со пелетирана крмна смеса, а јагнињата во контролната група со брашнеста крмна смеса. Товот на јагнињата во експериментот траеше 42 денови. Забележително беше дека на почеток на експериментот јагнињата во тест групата имаа помала просечна маса и помал прираст во споредба со јагнињата во контролната група. Меѓутоа, на крајот од експериментот јагнињата во тест групата постигнаа поголема просечна телесна тежина и подобар прираст во споредба со јагнињата во контролната група. Анализата на варијанса (ANOVA) за производните показатели на јагнињата, зависно од исхраната со пелетирани или брашнести крмни смеси, покажа дека постои статистички значајна разлика ($p < 0,05$) во средните вредности од производните показатели меѓу групите јагниња при направената контрола во петтата и шестата недела од експериментот. Употребата на пелетирани крмни смеси во овчарското производство, во интензивни услови на одгледување и тов на јагниња, овозможува подобрување на производните перформанси, зголемување на дневниот прираст и скратување на времето на гоеење на јагнињата.

Клучни зборови: исхрана, гоеење на јагниња, производни резултати



CONDITIONS AND CHALLENGES IN WHEAT, BARLEY AND CORN PRODUCTION IN REPUBLIC OF NORTH MACEDONIA IN THE PERIOD DURING THE NEW WORLD ECONOMIC CRISIS

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Abstract

Wheat, barley and corn are the most common cereals in crop production in Republic of North Macedonia. In 2021, 243,676 tons of wheat, 151,600 tons of barley and 130,769 tons of corn were produced. The other cereal species, like rye, rice and oats are produced in much smaller quantities. The total production of wheat, barley and corn grain in 2021 was 526,045 tons. The average production for the three-year period (2019/21) of grain from these crops was 529 956 tons.

The production obtained from these grains does not satisfy the domestic need and demand. Wheat is dominant and strategic cereal crop, followed by barley and corn. The domestic production meets about 55 to 60% of needs. The remaining amount is still provided by imports. In order to overcome these conditions and become import-independent for wheat, barley and corn grain, it is necessary to take certain bolder steps in the organizational part of production.

Key words: *production, cereals, yield, crops, grain, measures*

INTRODUCTION

Cereal crops are very important group of agricultural crops (Vasilevski, 2004). Cereal plants have many uses for human nutrition, domestic animals and the processing industry (Ilievski, 2014). The production of wheat, barley and corn grain does not meet the need in Republic of North Macedonia, although they have a strategic importance in production and are the most common crops grown in our country. These conditions are the result of a number of inconsistencies in production, such as:

- using uncertified seed material,
- inappropriate fertilization according to the requirements of cereal crops,
- fertilization with insufficient amounts of fertilizers;
- impossibility of interventional irrigation in critical phenophases of the growth and development of cereal plants as a result of outdated or non-functional irrigation

systems;

- use of old and inappropriate machinery in production;
- poor application of plant protection products during production, etc.

The soil and climate conditions of the region, as well as the degree of the agricultural technology used, greatly affect the achievement of high and stable production.

Of the total agricultural area in Republic of North Macedonia (1,263,000 ha), 40.46% is arable land (511,579 ha). Almost 81% of the arable land is used for agricultural production, of which 38.98% or 160,988 ha are with cereal crops. There are small differences in the achieved average yields of wheat, barley and maize between agricultural enterprises and family farms (State statistics office of the Republic of Macedonia (2012/07)). Wheat is in first place by area of all cereal crops.

Natural conditions for cereal plant production

The natural conditions in Republic of North Macedonia provide the opportunity for the cultivation of all cereal crops. At the same time, winter forms of grain withstand much lower temperatures compared to spring ones (Ilievski, 2014). In the case of winter forms, the optimum for spending the first vegetation period is from 0 to 15°C. In the later period of development, and especially in the phase of formation of the generative organs and flowering, they need higher temperatures, with an optimum of 16 to 22°C (Jevtić, 1992). The most resistant cereal crops to low temperatures are rye and wheat. They tolerate from -20 to -25°C without a snow cover, and maybe more (Ilievski, 2014). The most resistant to low temperatures are the winter forms of grain in the tillering phenophase. Millet, sorghum, rice and buckwheat can hardly tolerate temperatures below 0°C, while corn in the phenophase of germination tolerates negative temperatures from -2 to -3°C. Cereal

crops have a great need for water and can hardly bear the lack of it (Ilievski, 2014). One of the main problems in the production of cereal crops is precisely this factor. The most sensitive to lack of water are oats and rye, followed by wheat and barley, in the phenophases of grain filling and milk maturity.

Cereals are mostly grown on all soil types. According to the reaction of the soil solution, the cereals are divided into two groups: cereals that normally grow and vegetate at a neutral or slightly acidic reaction (pH 6-7): wheat, barley and corn; and cereals that tolerate a wider pH interval value: rye, oats, millet and buckwheat (Jevtić, 1992).

Wheat production in Republic of North Macedonia

Table 1 provides data on the representation, yield and realized production of wheat in the 2019, 2020 and 2021 production years and the average for the three-year period.

Table 1. Wheat production on agricultural arable land in Republic of North Macedonia 2019-2021.

Year	Area (ha)	Yield (kg/ha)	Production (t)
2019	68 959	3 485	239 916
2020	69 902	3 527	246 031
2021	70 515	3 463	243 676
Average 2019/21	69 702	3 492	243 208

Data in Table 1 show that the areas with wheat are constant, with small oscillations from year to year, but the data on the average yield per unit area show a tendency to increase.

Representation of high-yielding varieties and sowing quality seed material is one of the main factors for achieving high wheat yields (Georgievski et al., 2004/2005).

The average production of wheat grain for the three-year period is 243 208 tons. This

amount meets about 55 to 60% of needs. The remaining quantity is still provided by imports.

Barley production in Republic of North Macedonia

Table 2 provides data on the representation, yield and realized production of barley in the production years 2019, 2020 and 2021 and the average for the three-year period.

Table 2. Barley production on agricultural arable land in Republic of North Macedonia 2019-2021.

Year	Area (ha)	Yield (kg/ha)	Production (t)
2019	44 098	3 151	138 453
2020	45 011	3 283	147 711
2021	47 890	3 179	151 600
Average 2019/21	45 667	3 204	145 835

Data in Table 2 show that the areas with barley and the total production tend to increase slightly from year to year. The average

production of barley is 145 835 tons for the three-year period (2019/21).

Corn production in Republic of North Macedonia

Table 3 provides data on the representation,

yield and realized production of corn in the production years 2019, 2020 and 2021 and the average for the three-year period.

Table 3. Corn production on agricultural arable land in Republic of North Macedonia 2019-2021.

Year	Area (ha)	Yield (kg/ha)	Production (t)
2019	34 123	4 277	145 528
2020	32 013	4 589	146 434
2021	30 425	4 327	130 769
Average 2019/21	32 187	4 398	140 910

The data show that the areas under corn are decreasing from year to year, but the data for the average yield per unit area is constantly above 4000 kg/ha. The average corn grain production for the three-year period is 140,910 tons. Maize is produced on an area of 32 187 hectares with an average yield of about 4 398 kg/ha.

Total grain production of wheat, barley and corn

Table 4 provides data on the total production in tons of wheat, barley and corn for the years 2019, 2020 and 2021 and the three-year average.

Table 4. Total production of wheat, barley and corn in Republic of North Macedonia 2019-2021.

Year	Wheat (t)	Barley (t)	Corn (t)	Total (t)
2019	239 916	138 453	145 278	523 647
2020	246 031	147 711	146 434	540 176
2021	243 676	151 600	130 769	526 045
Average 2019/21	243 207	145 835	140 910	529 956

From the data in Table 4, it can be stated that Republic of North Macedonia produced an average of 243,208 tons of wheat, 145,835 tons of barley and 140,910 tons of corn for the period 2019/2021. The largest total grain production of these three cereal crops in the year 2020, amounts to 540,176 tons. In 2021, there is a slight decrease in the amount of production achieved, compared to the previous year.

Recommendations and measures for the improvement of cereal crops production Republic of Macedonia

Cereal crops production in the period of the new world economic crisis does not satisfy the domestic demand, although they have a strategic importance and are the most represented crops in plant production in our country. Domestic production meets about 55 to 60% of needs. The remaining quantity is still provided by imports.

In order to overcome these conditions and become market independent for cereal grains, it is necessary to take certain bolder steps in the organization of production. One

of the possibilities is to increase the areas with cereal crops. But in such market conditions, realistically, it is more difficult to do. Another possibility is to increase the average yield per unit area of all cereal crops. The realization of the second possibility requires a series of steps that must be followed and implemented.

Those steps are as follows:

1. Use of certified seed material by all producers;
2. Timely and sufficient amount of fertilizers for plant nutrition according to the needs of the crops and the soil conditions of the agricultural plots;
3. Precise nutrition of the crops in appropriate organogenetic stages to increase the generative organs in the grain, and thus the yield;
4. Alleviation of dry periods in the critical phenophases of the growth and development of cereal plants with interevent irrigation, especially in the phenophases of filling and ripening of the grain;

5. State investments in the improvement of obsolete and ineffective irrigation systems and the construction of new agro-ameliorative systems;
 6. Support and investment in small and medium-sized farms and family producers for the purchase of new and appropriate machinery;
 7. Increased and improved use of plant protection products;
 8. Increased support in the area of subsidies for strategic cereal crops;
- Agrotechnical measures, with which application higher yields can be achieved, among others, are: correct selection of the

pre-crop; growing in crop rotation; avoiding cultivation in monoculture and pre-crops that are harvested late and make it impossible to adhere to the optimal sowing dates; correct selection of varieties; quality tillage; use of optimal amounts of seed per sowing unit; harmonizing additional plant nutrition with natural soil fertility; breeding genotypes resistant to pests, diseases and weeds etc (Glemoćlija, 2004).

These moments are not fully observed in the production of cereal crops in Republic of North Macedonia, which leads to a direct decrease in average yields per unit area.

CONCLUDING REMARKS

Based on the above, the following conclusions and findings can be drawn:

- The cereal crops production in Republic of North Macedonia (2019/21) is realized with the production of wheat grain (243 207t), barley (145 835 t) and corn (140 910 t).
- Rye, rice and oats are produced in much smaller quantities.
- Of the total agricultural area in Republic of North Macedonia (1,263,000 ha), 40.46% is arable land (511,579 ha). Almost 81% of the arable land is used for agricultural production, of which 38.98% or 160,988 ha are with cereal crops.
- The differences in average yields achieved between agricultural enterprises and cooperatives and family farms are small.
- The cereal crops production in the period

of economic crisis does not satisfy the domestic demand, although they have a strategic importance and are the most represented crops in plant production in our country.

- Domestic production meets about 55 to 60% of needs. The remaining quantity is still provided by imports.
- To achieve a higher and stable production to a large extent in addition to the soil and climate conditions of the region, it is necessary to improve the degree of applied agricultural technology.
- In order to overcome the dependence on wheat, barley and corn grain imports, all efforts and measures should be aimed at increasing the average yield per unit area.

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СОСТОЈБИ И ПРЕДИЗВИЦИ ВО ПРОИЗВОДСТВОТО НА ПЧЕНИЦА, ЈАЧМЕН И ПЧЕНКА ВО РЕПУБЛИКА МАКЕДОНИЈА ВО ПЕРИОДОТ НА НОВАТА СВЕТСКА ЕКОНОМСКА КРИЗА

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Резиме

Пченицата, јачменот и пченката се најзастапени житни култури во растителното производство во Република Македонија. Во 2021 година се произведени 243 676 тони пченица, 151 600 тони јачмен и 130 769 тони пченка. Од останатите видови житарки, како на пример 'ржта, оризот и овесот, се произведуваат во многу помали количини. Вкупното производство на зрно од пченица, јачмен и пченка во 2021 година изнесува 526 045 тони. Просечното производство за тригодишниот период (2019-2021 година) на зрно од овие култури е 529 956 тони.

Добиеното производство од овие жита не ја задоволува домашната потреба и побарувачка. Пченицата е доминантна и стратешка житна култура, потоа следат јачменот и пченката. Домашното производство задоволува околу 55 до 60% од потребите. Останатата количина сè уште се обезбедува со увоз. За да се надминат овие состојби и да станеме увозно независни за зрно од пченица, јачмен и пченка, потребно е да се направат одредени посмели чекори во организацискиот дел на производството.

Клучни зборови: производство, жита, принос, површина, култури, зрно, мерки



MICROPROPAGATION OF POTATO SEED TUBERS (*Solanum tuberosum* L.) UNDER *IN VITRO* CONDITIONS

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Abstract

In this paper the results of influence of phytohormone gibberellic acid GA₃ on sprout formation in *in vivo* conditions and *in vitro* microtuberization of the potato varieties Agria, Agata, Sunshine, Ultra and Marabel are presented. The tubers from all varieties utilized in the experiment were certified potato seed tuber material.

The experiment in *in vitro* conditions was established on sprout explants and nodal segment explants on the MS medium (Murashige & Skoog, 1962) with addition of different combination and concentration of auxins and cytokines. Microtuberization was stimulated by rising the concentration of sucrose in MS medium from 40g/l to 60 g/l and 80g/l, respectively.

The *in vivo* tuber treatment with 30mg/l GA₃ was the most effective treatment for all potato varieties in proliferation of sprouts. All tubers that were treated with GA₃ resulted in *de novo* sprouting of tubers.

The variety Agata resulted with 100% of microtuberization from nodal segment explants on MS medium supplied with 40g/l and 80g/l sucrose. Microtuberization of the variety Sunshine was stimulated with addition of 80 g/l sucrose in MS medium.

The developed microtubers were detached from the nodal segments and subcultured on new MS medium supplied with BAP 4mg/l, KIN 4mg/l and 8% of sucrose to increase their weight.

Key words: phytohormones, microtuberization, gibberellic acid, sucrose, variety of seed potatoes

INTRODUCTION

Potato (*Solanum tuberosum* L.) is widely grown worldwide because of its rich nutrition, easiness of cultivation and high yield performance (Wang et al., 2020). Nowadays it is the fourth most important food crop in the world, after wheat, rice and maize, cultivated on 19.3 million hectares with yield of 388.2 million tons of potato tubers (Waqas et al., 2021). Potato (*Solanum tuberosum* L.) is grown in more than 100 countries and feeds more than a billion people worldwide (Islam et al., 2020).

The formation of the tubers is a very complex process, but it can be stimulated in *in vitro* conditions, process known as microtuberization (Abbott and Belcher, 1986; Apichai, 1988; Dodds et al., 1992; Coleman et al., 2001).

Previous studies have shown that micropropagation of potato seed tubers depends on the biological value of cultivars,

explant type (leaf, nodal segments, shoot tip), type of culture medium, season, temperature, photoperiod and balanced combination of plant growth regulators (PGRs) in the culture media (Akhtar et al., 2006; Dhital et al., 2010). GA₃ participates in cell elongation and GA₃ addition in MS medium enhances shoot growth (Camara et al., 2018; Rizza et al., 2017).

Osmotically active solutes have shown that sucrose acts as a carbon source and osmotic regulator. Sucrose and sucrose concentration are important factors for potato microtuberization and they have a profound effect on tuber growth (Azar et al., 2013). It acts as an energy for growth and biosynthetic processes and may influence growth in *in vitro* conditions (Ferreira et al., 2011). Sucrose is also closely related to stomatal density and photosynthetic pigment content, as well as development induction in some plant tissues, such as vascular and support

tissues (Mohamed and Alsadon, 2010; Iarema et al., 2012). The rise of sucrose concentration in medium can enhance the microtuber production to some extent (Khan et al., 2018). However, high concentration of sucrose in the medium may decrease the photosynthetic ability of *in vitro* potato plants (Fuentes et al., 2005).

MATERIAL AND METHODS

The research was conducted in the Laboratory of Plant Biotechnology, Faculty of Agriculture, Goce Delcev University – Stip, Republic of North Macedonia. As starting material seed tubers from the potato varieties Agata, Marabel, Ultra, Sunshine and Agria were used.

***In vivo* treatment of potato seed tubers with GA₃**

The tubers from different varieties were treated with GA₃ with concentration of 10, 20

Potato starch has some unique physicochemical characteristics compared to starches from other sources as high phosphate content, absence of internal lipids and proteins in granules (Burlingame et al., 2009; Romano et al., 2016).

and 30 ppm. Control treatment, where the tubers were not treated with GA₃ was used to determinate whether GA₃ had effect on emergence of sprouts (Fig. 1).

The GA₃ treatment was used for induction of germination and rapid emergence of sprouts. After GA₃ treatment, one week old sprouts were detached from the potato tubers, and they were used as starting explants for further *in vitro* cultivation on MS medium supplemented with different concentrations of phytohormones.

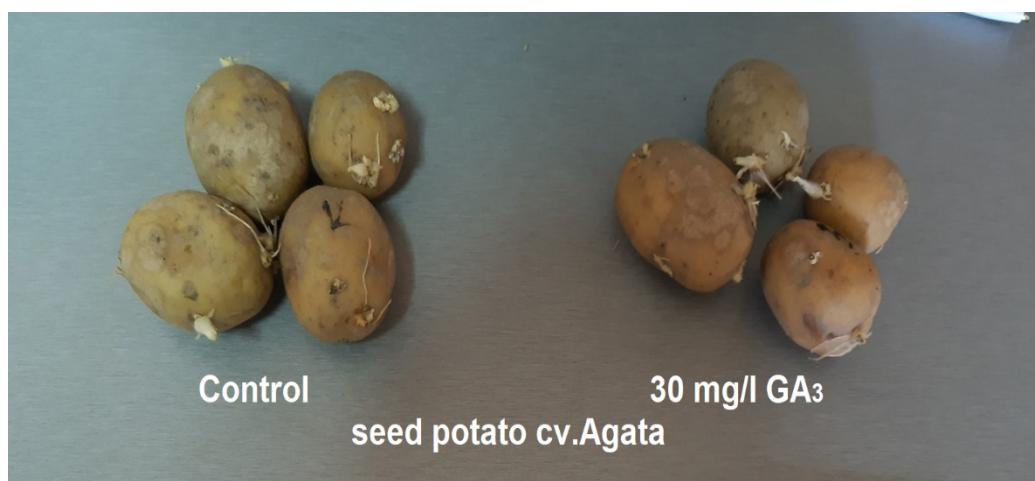


Figure 1. The effect of 30 mg/l GA₃ treatment for rapid sprouting and de novo proliferation of sprouts in variety Agata compared to the control.

Sterilization of initial explants (sprouts)

The sprouts were surface cleaned by washing them under tap running water for 10-15 minutes and rinsing them in distilled water several times followed by surface sterilization of sprouts surface by immersion in:

- 70% C₂H₅OH for 2 minutes.
- 0.5% HgCl₂ for 3-5 minutes or 0.1% NaClO for 10 minutes and
- several times rinsing with sterile water.

Initial explants – sprouts cultivated in *in vitro* conditions

The sprouts as initial explants were cultivated in MS medium supplemented with 30 g/l sucrose, 0.7% agar, 100 g/l myoinositol, 200 g/l casein enzymatic hydrolysate, 0.1mg/l thiamine, 1.0 mg/l pyridoxine and 0.5 mg/l nicotinic acid. The MS medium pH was adjusted to 5.8.

The MS media supplemented with different concentration of cytokinins and/or auxins were used for induction of shoots culture from different potato varieties:

- Sprouts > MS + 2mg/l BAP
- Sprouts > MS + 2mg/l KIN
- Sprouts > MS + 2mg/l BAP + 1mg/l NAA
- Sprouts > MS + 2mg/l KIN + 1mg/l NAA

The sprouts developed into shoots with different number of nodes within a month. The shoots were divided into nodal segments and subcultured on:

- Shoot nodal segments > MS + 3mg/l BAP + 1mg/l NAA
- Shoot nodal segments > MS + 3 mg/l KIN + 1mg/l NAA

These media were used for stimulation of nodal segments growth.

When the explants have reached 15-20 mm length, they were divided into nodal segments and subcultured on MS supplemented with different concentration of BAP, NAA, and sucrose for induction of microtubers. The following media were used for induction of microtubers

in different potato varieties:

- Nodal segments > MS + 2mg/l BAP + 2mg/l NAA + 4% sucrose
- Nodal segments > MS + 4mg/l BAP + 2mg/l NAA + 6% sucrose
- Nodal segments > MS + 6mg/l BAP + 2mg/l NAA + 8% sucrose

Maintenance of cultures in the climate chamber

All explants, sprouts and nodal segments, were incubated in a climate chamber under the following conditions: temperature $25 \pm 10^{\circ}\text{C}$; relative humidity 50%; photoperiod: 16/8 hours light/dark; illumination of 50 cd.

Data analysis

All data were subjected to statistical analysis with statistical package IBM SPSS Statistical 29, one-way ANOVA and Duncan post hoc test, with the level of significance 0.05%.

RESULTS AND DISCUSSION

All tubers treated with gibberellic acid GA_3 resulted in *de novo* germination of sprouts from the tuber eyelets. The treatment with 30 ppm GA_3 was the most effective for all potato varieties. The application of 30 ppm GA_3 as the highest dose of gibberellic acid resulted in 100% formation of sprouts from the tubers of potato varieties Ultra, Sunshine and Agria. The results presented in Table 1 show that all potato varieties have good response to gibberellic acid treatments, regardless of applied concentration. The variety Sunshine has shown 100% of formation of sprouts when treated with 10, 20 and 30 ppm GA_3 .

The initiation of sprouts was the key factor to induce microtuberization. The subcultured sprouts on MS medium supplied with different concentrations and combinations of auxins and cytokinins proliferated into shoots. The nodal segments from regenerated shoots were used for induction of microtubers (Fig. 2a).

The nodal segments from shoots were subject of subcultivation on MS medium supplied with cytokinins and auxins and sucrose in concentration of 4, 6 and 8%. The sucrose was added in order to initiate higher rate of formation of microtubers. The results of microtuberization of seed potatoes are shown in Table 2.

Different researchers agreed that higher percent of sucrose in the medium had positive results on microtuberization process and increased the number and quality of microtubers (Farran and Mingo-Castel, 2006; Motallebi-Azar and Kazemiani, 2012; Ahmed et al., 2013). This confirms our findings during this research.

The culture of nodal segments was incubated in controlled climate chamber under dark conditions to initiate formation of microtubers. (Fig. 2b).

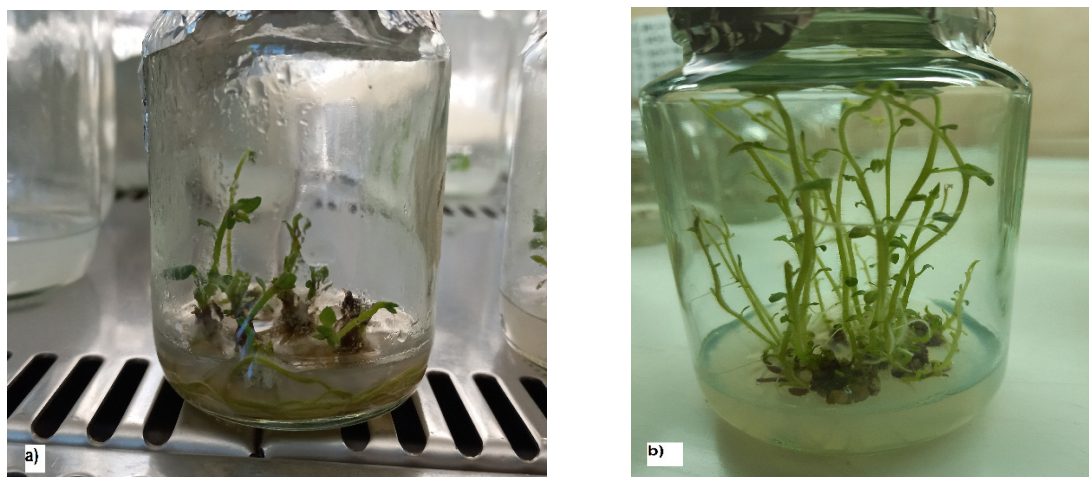


Figure 2. a) Culture of shoots b) Culture of nodal segments.

Table 1. Varieties of potato tubers treated with GA₃ and control.

Production of sprouts								
Variety	Treatment with GA ₃	Number of tubers	Number of eyelets per tuber	Number of sprouts per tuber	Length of sprouts (mm)	Width of sprouts (mm)	Number of sprouts per eyelet	% of sprout proliferation
Agata	Control	20	1.89a	0.93ab	2.31b	2.04b	1.37ab	85
	10 ppm	22	1.47a	1.00a	2.41b	2.02a	1.40bc	100
	20 ppm	22	1.66a	1.00a	2.49b	2.00b	1.35a	100
	30 ppm	22	1.52b	0.96a	2.64bc	2.15ab	1.21ab	90.90
Marabel	Control	18	1.45a	0.94ab	2.50a	2.09b	1.52a	83.33
	10 ppm	18	1.67a	1.00a	2.40b	2.05a	1.50a	55.55
	20 ppm	18	1.34b	0.89b	2.64b	2.02b	1.30a	77.77
	30 ppm	18	1.32b	0.95a	2.65bc	2.02b	1.22ab	94.44
Ultra	Control	17	1.47a	1.00a	3.37a	2.23a	1.08b	100
	10 ppm	18	1.47a	0.94a	3.38a	2.14a	1.13bc	88.88
	20 ppm	18	1.45ab	1.00a	3.47a	2.33a	1.13ab	100
	30 ppm	18	1.24b	1.00a	3.28a	2.24a	1.32a	100
Sunshine	Control	15	1.81a	0.97ab	2.57a	2.00b	1.18ab	93.33
	10 ppm	15	1.45a	1.00a	2.71b	2.07a	1.05b	100
	20 ppm	15	1.46ab	1.00a	2.60b	2.02b	1.04b	100
	30 ppm	15	1.31b	1.00a	2.97ab	2.09ab	1.04b	100
Agria	Control	17	1.87a	0.87b	2.22b	2.00b	1.40ab	88.23
	10 ppm	17	1.50a	0.94a	2.62b	2.09a	1.14bc	88.23
	20 ppm	16	1.42ab	0.96a	2.57b	2.27a	1.28a	93.75
	30 ppm	16	2.00a	1.00a	2.35c	2.10ab	1.21ab	100

Means within each column having different letters are significantly different according to Duncan's test at $p < 0.05$.

The rise of sucrose concentration in MS medium from 40g/l to 80 g/l increased the percentage of formation microtubers from 42.85% (4% sucrose) to 58.33% (8% sucrose) in the variety Ultra (Fig. 3a).

The variety Agata resulted with 100% microtuberization of nodal segments when cultivated on MS medium with 40g/l and 80 g/l sucrose. Higher microtuberization rate of the variety Sunshine was achieved with 80 g/l

sucrose in MS medium (33.33%) as compared to 40 g/l (16.16%).

The developed microtubers were detached from nodal segments and subcultured on the

new MS medium enriched with BAP 4mg/l, KIN 4mg/l and 8% of sucrose in order to increase their weight (Fig. 3b).

Table 2. Effect of different concentration of BAP, NAA, and sucrose on microtuberization in potato nodal segments.

Explants – nodal segments						Formation of microtubers			
Variety	MS medium with cytokinins and auxins	% of sucrose	Number of nodal segments	Length of nodes (mm)	Thickness of nodal segments (mm)	Number of microtubers per explant	Length of tubers (mm)	Width of tubers (mm)	Microtuberization (%)
Agata	2mg/l BAP + 2mg/l NAA	4%	4	7.50b	1.62a	4	3.50a	3.25a	100
Agata	6mg/l BAP + 2mg/l NAA	8%	3	13.33a	1.50a	3	5.00a	3.00a	100
Ultra	2mg/l BAP + 2mg/l NAA	4%	7	13.00a	1.07b	3	2.14a	1.42ab	42.85
Ultra	6mg/l BAP + 2mg/l NAA	8%	36	11.21a	1.14a	21	1.94b	1.44b	58.33
Sunshine	2mg/l BAP + 2mg/l NAA	4%	6	12.50a	1.16b	1	1.00a	0.33a	16.66
Sunshine	MS+6mg/l BAP + 2mg/l NAA	8%	24	11.91a	1.43ab	8	1.33b	0.75b	33.33

Means within each column having different letters are significantly different according to Duncan's test at $p < 0.05$.

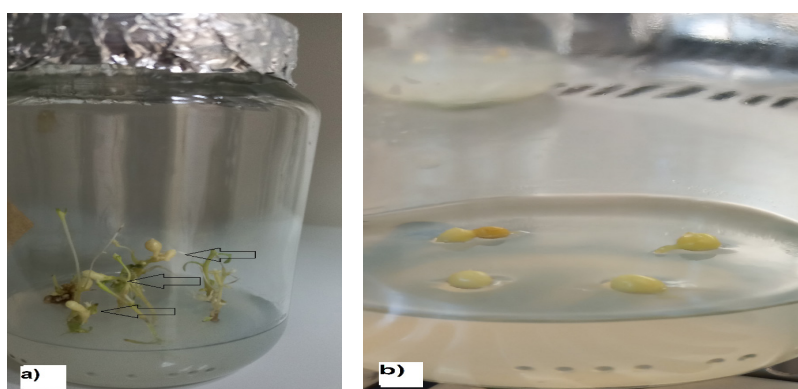


Figure 3. a) Microtuberization b) Culture of microtubers.

CONCLUDING REMARKS

Micropropagation is an alternative method for conventional breeding of potatoes. Methods of *in vitro* propagation using sprouts and nodal segments are more reliable to maintain the integrity of the genetic and breeding material.

Microtuberization is an important process for the production and storage of potatoes.

Microtubers obtained by *in vitro* culture of nodal segments are suitable for manipulation, storage and distribution of healthy germplasm.

The results presented in this paper have proven that potato seed tubers have regenerative power and good potential for microtuberization.

Nodal segments of the variety Agata cultured on medium MS+6mg/l BAP + 2mg/l NAA+8% sucrose responded with 100% microtuberization.

The high concentration of sucrose acts as a stimulation signal leading to the accumulation of starch in microtubers.

The nodal segment culture of the variety Ultra resulted with 42.85% microtuberization when cultured on MS medium supplemented

with 40 g/l sucrose, while rising the concentration of sucrose from 40 g/l to 80 g/l resulted in increase of microtuberization from 42.85% to 58.33%.

Microtuberization of the variety Sunshine was stimulated with higher concentration of sucrose 80 g/l in the medium and it resulted in microtuberization response of 33.33% of nodal segments.

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МИКРОПРОПАГАЦИЈА НА СЕМЕНСКИ КОМПОР (*Solanum tuberosum* L.) ВО IN VITRO УСЛОВИ

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Резиме

Во овој труд се прикажани резултатите од влијанието на фитохормонот гибберелинска киселина GA₃ врз формирање на 'ртулци во *in vivo* услови и *in vitro* микротуберизацијата на неколку генотипови компор Agria, Agata, Sunshine, Ultra и Marabel. Клубените од сите генотипови користени во овој експеримент беа сертифициран семенски компор.

Експериментот во *in vitro* услови беше поставен со 'ртулци и нодии на MS медиум (Murashige & Skoog, 1962) со додавање на различни концентрации и комбинации на ауксини и цитокинини. Микротуберизацијата беше стимулирана со зголемување на процентот на сахароза во MS медиумот од 40 g/l сахароза на 60 и 80 g/l.

Третирањето на клубените во *in vivo* услови со 30mg/l GA₃ се покажа како најефикасен за сите испитувани семенски генотипови за добивање на 'ртулци. Кај сите генотипови третирани со GA₃ резултираше со *de novo* 'ртулци од окцата на клубените.

Генотипот Agata резултираше со 100% микротуберизација од нодиите на MS медиум со 40 g/l и 80g/l сахароза. Микротуберизацијата кај генотипот Sunshine беше стимулирана со додавање на 80 g/l сахароза во MS медиумот.

Формираните микроклубени беа одделени од нодалните сегменти и пасажирани на нов MS медиум збогатен со BAP 4mg/l, KIN 4mg/l и 8% сахароза за зголемување на нивната тежина.

Клучни зборови: фитохормон, микротуберизација, гибберелинска киселина, сахароза, генотипови на семенски компор

Journal of Agriculture and Plant Sciences, JAPS, Vol 20, No. 2

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ISSN 2545-4447 print
ISSN 2545-4455 on line
Vol. 20, No.2 , Year 2022