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Assessment of growth and development of cowpea varieties based on phenological and morphological observations

Abstract. The search and selection of cowpea varieties with optimal economic characteristics is a key factor in the development of cultivation technologies to increase yields. The purpose of the study was to determine the phenological and morphological characteristics of cowpea varieties and establish the relationship between economically valuable indicators and weather conditions. The leading approaches to studying this problem are field method – to assess the growth and development of cowpea varieties; statistical – to conduct variance and correlation analysis of the assessment of morphological features and economically valuable indicators of cowpea. Five cowpea varieties were studied: Groik (Israel), Kafedralna (Ukraine), Gasson (Vietnam), American improved (USA), and U-Cha-Kontou (China). It was found that the duration of phenological phases of cowpea varieties depended on weather conditions. Varieties of cowpea were characterised by early maturation periods for the sum of effective temperatures of 192.0-207.5°C and precipitation of 30.54-31.65 mm. Earlier maturation periods were the varieties American improved, Kafedralna, Gasson with the duration of the period from germination to the beginning of harvesting ripeness of the pod – 58-62 days and the duration of the period from germination to biological ripeness of seeds – 92-94 days. For the vegetable industry, bush varieties Kafedralna and U-Cha-Kontou with a plant height of 48.5-54.9 cm were promising, which formed tender long beans with a weak parchment layer (22.8-23.6 cm). Groik and Gasson varieties proved valuable for growing seedlings due to the small light-coloured seeds with a plant height of 61.8-100.5 cm and the formation of a large number of short beans on the plant (12.8-15.6 cm). A strong relationship between morphological features and economically valuable indicators of cowpea has been established. The average bean yield had a direct strong relationship with the total number of beans per plant ($r=0.93$) and a strong inverse relationship with the length of the bean ($r=-0.88$). As the length of the beans increased, the average yield of beans and the number of beans on the plant decreased, as evidenced by a strong inverse relationship between these indicators ($r=-0.93$). The materials of the paper are of practical value for increasing the species diversity of vegetable crops and improving the provision of valuable food products to the population

Keywords: legumes, bean pods, flowering, harvesting ripeness, plant height, bean length, seeds

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INTRODUCTION

Vegetable growing in Ukraine has become a highly developed industry, the main task of which is to provide the population with food. It is necessary to introduce new technologies to intensify production. In turn, the criterion for evaluating any crop cultivation technology remains the yield, and its value must be economically justified and energetically confirmed. Along with the growth of yields, there is a constant expansion of vegetable diversity based on Ukraine's own production and imports. Among them, legumes are very valuable (Bobos et al., 2022).

In the vegetable growing industry of Ukraine for 2022, only a small number of species from the legume family are grown (Fabaceae) (Bobos et al., 2022). These are mainly vegetable beans, vegetable peas and, very rarely, vegetable beans. Even among these species, producers have not yet fully used them for the vegetable industry, including for obtaining unripe seeds, for green beans-pods, etc. The world variety of cultivated vegetable legumes is very large and includes more than 40 species. Vegetable farmers grow quite a lot of types – kidney and lima beans, sugar peas, dolichos, tetragonolobus, vegetable bean, trigonella (fenugreek, mushroom grass), cowpea, etc. (Bobos et al., 2022).

In Ukraine, domestic varieties have not yet been created for these species, and amateur vegetable growers widely grow local and accidentally introduced forms. Almost all crops have not developed technologies for their cultivation, both for one-time harvesting and for creating continuous production for a long period in supermarkets (Sysh & Bobos, 2011).

One of the most promising low-spread legumes is cowpea, which is valued for its content of easily accessible protein and vitamins, and also accumulates all the amino acids, calcium, phosphorus, and iron salts necessary for humans.

Morphological features of cowpea are very similar to beans. Genus *Vigna* (*Vigna* sp.) is characterised by a wide variety, which is collected at the International Institute of Tropical Agriculture of Nigeria (Sysh & Bobos, 2011). The State register of plant varieties has already included the first new climbing variety Kherson, which was created at the Institute of Southern Vegetable and Melon Production of the National Academy of Agrarian Sciences of Ukraine. In the steppe and forest-steppe of Ukraine, there is a sufficient sum of effective temperatures for growing cowpea (Sysh & Bobos, 2011).

Cowpea is grown to produce ripe seeds, green pods, and unripe “flageolet” seeds. In the United States, high-quality seedlings are obtained from the seeds of small-seeded varieties (Barros et al., 2021).

The optimal time for harvesting young beans on the cowpea pods falls on Day 7-10 after pollination. Especially valuable are varieties with black seeds that do not accumulate heavy metals – cadmium and lead.

At the Department of Indoor Vegetable Growing of the National University of Life and Environmental Sciences of Ukraine for the first time in the northern forest-steppe

during 2008-2010, samples of cowpea varieties were studied and evaluated, and their comparative assessment was carried out by precocity, morphological characteristics, productivity (Sysh & Bobos, 2011; Bobos et al., 2022). Valuable source material of cowpea was identified, which was used in breeding work as parent forms and the first bush plant of the Kafedralna variety was created. At the same time, there was a need for a comparative assessment of cowpea varieties by phenological and morphological characteristics, considering changes in climatic conditions.

The main value of cowpea is its high drought, heat, and salt resistance, which allows growing this crop for seeds, green manure, feed, and vegetables (Sysh & Bobos, 2011; Barros, 2021). However, an increase in temperature is associated with accelerated growth and the duration of the phenological phases of the crop. Once in stressful conditions, plants change their metabolism so as not to disrupt their normal development. Acceleration of development occurs due to the plant's response to stress, increased respiration (Schmidt et al., 2017). In this context, studying how cowpea plants respond to increased water scarcity and rising air temperatures, in the long run, can help growers change their management practices to maintain sustainable production (Barros, 2021).

Cowpea can grow and develop in a wide temperature range, from 18 to 37°C, but the optimal temperature value varies depending on the phenological stage of the plant. Temperatures above 35°C cause abortivity of flowers, stimulates the ageing of leaves, reducing photosynthetic ability, which affects the productivity of beans and cowpea seeds, since such an increase disrupts the physiological and biochemical processes in plants (Zandalins et al., 2018). In response, plants reduce the chlorophyll content, photosynthetic rate, and transpiration rate in an attempt to reduce moisture loss (Taiz et al., 2017).

Climate change is becoming a challenge for food security, as rising temperatures and changes in precipitation distribution have a negative impact on agricultural production (Gomes et al., 2019). In addition, forecasts for the future indicate that water scarcity can significantly affect plant growth and development on more than 50% of agricultural land by 2050, which would reduce crop yields worldwide (Deryng et al., 2011; Hasanuzzaman, 2019). Despite the fact that the main abiotic factor affecting the water balance of plants is a lack of soil moisture, the severity of this effect largely depends on air temperature (Jumrani & Bhatia, 2018; Havryliuk et al., 2022).

Numerous studies have shown that climate change has adverse effects on food production and food security, and the adverse effects are projected to increase in the future (Hansen, 2011; Iizumi et al., 2014). Climate variability, such as being too wet or too dry, also affects yields (Iizumi & Ramankutty, 2015).

Thus, the analysis of literature sources indicates the influence of climatic conditions on the economically valuable indicators of cowpea and the need to study the

variability of phenological and morphological features of crop varieties for the introduction of adaptive to growing conditions in production.

Purpose of the study is to determine the phenological and morphological characteristics of cowpea varieties and establish the relationship between economically valuable indicators and weather conditions.

MATERIALS AND METHODS

During 2014-2016, 5 varieties of cowpea were studied: Groik (Israel), Kafedralna (Ukraine), Gasson (Vietnam), American improved (USA), and U-Cha-Kontou (China). The research was conducted on the collection sites of the Department of Indoor Vegetable Growing in the Training Lab "Fruit and Vegetable Garden" of the National University of Life and Environmental Sciences of Ukraine, which is located in the northern part of the forest-steppe of Ukraine on sod-medium podzolic soils. The field method was performed according to one-factor experiment scheme (Bondarenko & Yakovenko, 2001). Repeatability – three times with randomisation. The accounting area – 5 m². The promising Ukrainian variety Kafedralna was taken as control.

Cowpea cultivation techniques are similar to those used for vegetable beans in production (Sysh, 2011). The sowing scheme was 70×25 cm. Seed embedding depth – 2-3 cm.

Seeds of the varieties under study and control were sown simultaneously on May 13. Immediately after sowing, the rows were mulched and the soil between the rows was loosened. Plant care consisted of systematic loosening, and control of weeds, diseases, and pests.

During the field experiments, the following phenological phases were noted: full shoots, the beginning and full flowering, the beginning of harvesting ripeness of pods and the economic ripeness of beans. Shoots were noted by the appearance of cotyledons on the surface

(Bondarenko & Yakovenko, 2001). In cowpea, the beginning of maturation was noted by the maturation of the predominant number of beans. The duration of the growing season was calculated from the date of sowing to economic ripeness. The sowing-germination period for selected varieties ranged from 12 to 17 days and took place with an average sum of effective temperatures of 70.2-105.7°C and precipitation of 42.8-56.8 mm.

Harvesting was carried out on all variants of the experiment simultaneously with the onset of the economic ripeness of beans – on September 6. Before harvesting, biometric measurements were made, namely, the length of the central stem was measured with a ruler and the number of side shoots on the plants was counted. During harvesting, the number and weight of beans per plant, the length of the bean, and the number of seeds per bean were determined. Simultaneously considering the yield, an average sample was selected from all varieties according to the variants, which was used to determine the weight of 1,000 seeds.

Variance (ANOVA) and correlation analysis were performed using the XLSTAT add-in in MS Excel software suite. The differences were considered significant for the validity of $\alpha=0.95$ (Nageswara, 2018).

RESULTS AND DISCUSSION

The study of phenological variability showed that cowpea varieties were characterised by early maturation periods. Earlier flowering was noted in the varieties Kafedralna, Gasson, and U-Cha-Kontou, with the beginning of flowering on 13.07. Late flowering dates were characterised by the Groik variety (16.07), which affected the beginning of harvesting ripeness of the pods, which was noted later than other varieties 02.08. The variability of plant growth and development affected the duration of phenological phases of cowpea varieties (Fig. 1).

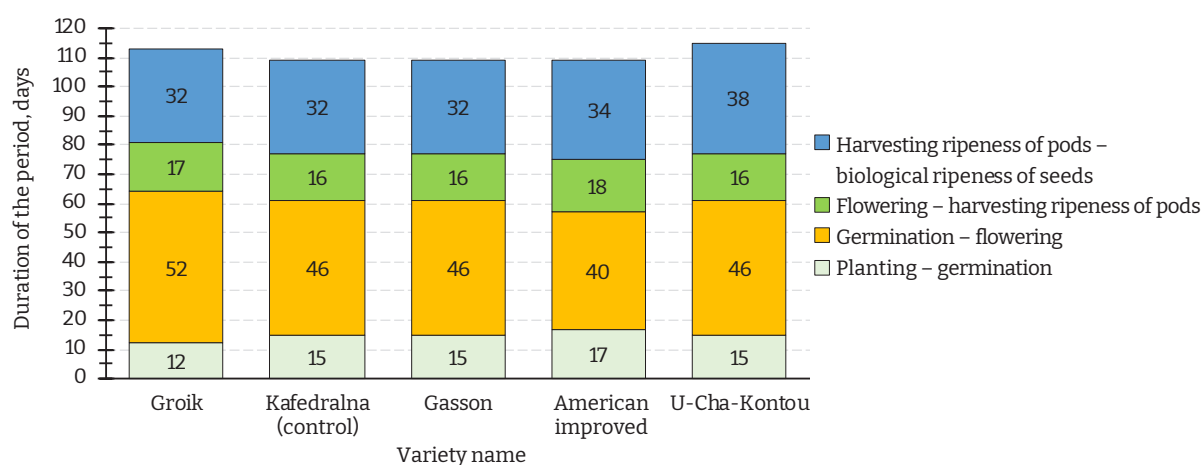


Figure 1. Duration of phenological phases of cowpea varieties, days (average for 2014-2016)

The shortest period from germination to flowering was distinguished by the American improved variety (Day 40), the longest period was observed in such varieties

as Groik (Day 52) and Gasson (Day 46). This period was held at an average sum of effective temperatures of 436.6-549.7°C and precipitation of 45.83-68.28 mm (Fig. 2, 3)

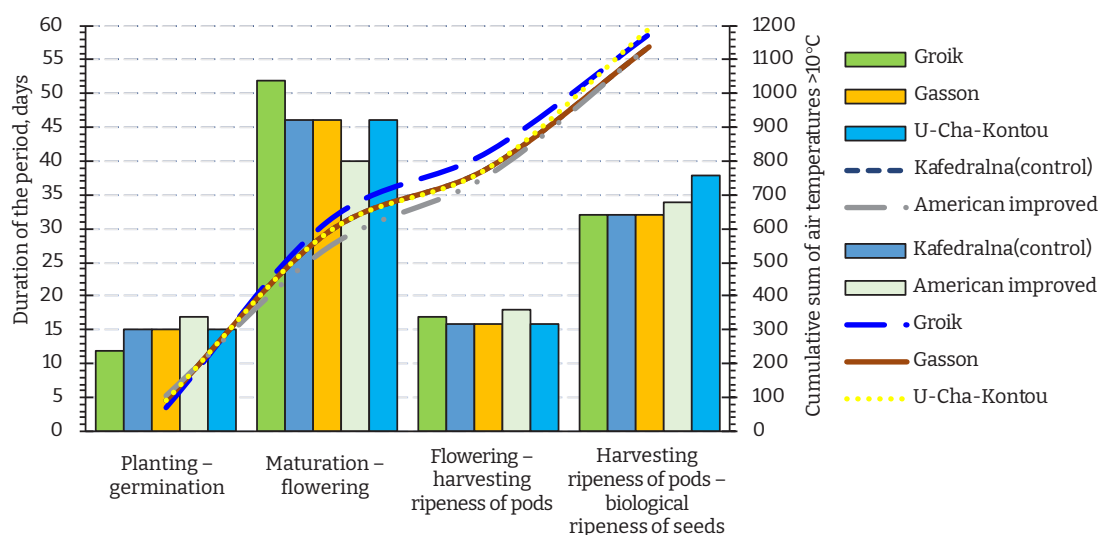


Figure 2. Influence of the sum of air temperatures over 10°C on the passage of phenological phases in cowpea varieties

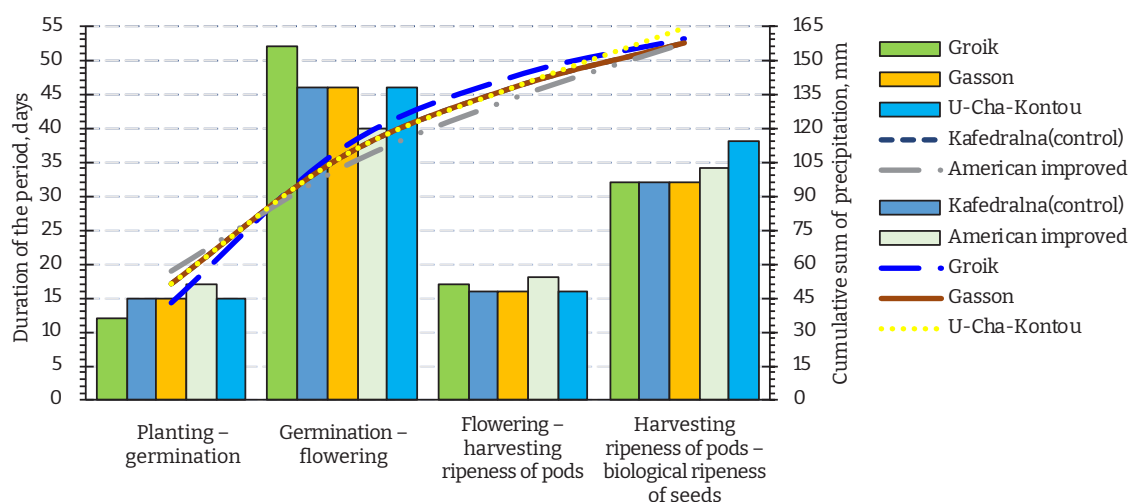


Figure 3. Influence of precipitation amount on the passage of phenological phases in cowpea varieties

The period from flowering to the beginning of harvesting ripeness of the pods was almost the same in all varieties and ranged from 16 to 18 days. The average sum of effective temperatures during this period was 192.0-207.5°C and the amount of precipitation was 30.54-31.65 mm. But it is the smallest in the control varieties Kafedralna, U-Cha-Kontou, and Gasson (16 days).

The period from germination to the beginning of harvesting ripeness of the pods was the smallest in the American improved variety (58 days), which characterises the variety with an early maturation period. This period took place with an average effective amount of 652.1°C and a precipitation amount of 76.4 mm. The longest period was observed in the Groik variety – 69 days.

Kafedralna, American improved, and Gasson varieties with a duration of 92-94 days from germination to biological ripeness of seeds distinguished themselves by their early maturation periods. The average sum of effective temperatures during this period was 1,034.6-1,047.0°C

and the sum of precipitation was 100.7-106.4 mm. The duration of the period from germination to biological ripeness of seeds in the Groik and U-Cha-Kontou varieties was 100-101 days, which indicates their average maturation time. This period was held at an average effective amount of 1,100.7-1,105.3°C and precipitation of 113.0-116.0 mm.

Cowpea varieties differed in morphological characteristics from each other (Table 1). Among the cowpea assortment that was tested, dwarf bush varieties with plant heights up to 50 cm and more than 50 cm are distinguished. Among the dwarf varieties, Kafedralna and American improved stand out. The average height of plants of these varieties reached 46.1-48.5 cm. Groik and Gasson varieties were characterised by intensive growth of the main and lateral shoots, due to which they can be grown on supports. The height of these varieties was 61.8-100.5 cm with the shortest length of beans – 12.8-15.6 cm. These varieties formed a large number of beans on the plant (34.6-58.3 pcs.), which affected their seed productivity.

Table 1. Evaluation of cowpea varieties by morphological parameters

Variety name	Plant height, cm				Number of beans per plant, pcs.				Bean length, cm			
	years			Average	years			Average	years			Average
	2014	2015	2016		2014	2015	2016		2014	2015	2016	
Groik	98.7	96.4	106.4	100.5	34.2	36.2	33.4	34.6	15.2	16.5	15.1	15.6
Kafedralna (control)	50.1	47.3	48.1	48.5	24.7	26.1	24.8	25.2	24.5	23.1	23.2	23.6
Gasson	60.9	58.9	65.6	61.8	57.9	60.1	56.9	58.3	13.1	12.5	12.8	12.8
American improved	47.2	45.2	45.9	46.1	13.1	12.5	12.5	12.7	27.2	26.4	25.9	26.5
U-Cha-Kontou	53.8	55.6	55.3	54.9	16.1	15.5	16.1	15.9	23.1	20.4	24.3	22.8
LSD ₀₅				5.36				1.93				1.59

Source: based on the conducted research

The main feature of pods is the absence of a parchment layer and fibres in the seams. The best quality vegetable varieties are those in which the beans do not thicken for a long time and are not characterised by a tendency to form a parchment layer and fibre during the entire harvesting period. Kafedralna and U-Cha-Kontou varieties were characterised by a plant height of 48.5-54.9 cm. In the Kafedralna variety, a larger number of beans were formed on the plant (25.2 pcs.) with strong peduncles and long beans (23.6 cm) (Fig. 4).

They are characterised by the establishment of a weak parchment layer and fibre, especially in conditions of high air temperature and delayed harvesting. However, the disadvantage of the Kafedralna and U-Cha-Kontou varieties is that the long beans (22.8-23.6 cm), which are laid on the soil, are polluted and therefore require mulching with straw. At the same time, the U-Cha-Kontou variety formed a small number of beans on the plant (15.9 pcs.) with a small number of seeds, which affected the low seed productivity of plants.

**Figure 4.** Kafedralna beans

A small number of beans on the plant was also established by the American improved variety, which on average for three years revealed the smallest plant height of 46.1 cm with fewer beans on the plant (12.7 pcs.) and a longer bean length (26.5 cm), which affected the productivity of plant seeds.

Groik and Gasson varieties belong to cereals. Their height is more than 50 cm. The beans are short with a well-defined parchment layer, which makes it impossible to use them to produce unripe pods. At the same time, these varieties are very valuable for growing seedlings due to the small light-coloured seeds. Semi-bush cowpea varieties Gasson and Groik do not need supports. Despite the high ability to form a large number of lateral shoots, they form a crop on strong vertical peduncles.

Using correlation calculations, the relationship between morphological features and economically valuable indicators of cowpea is established (Table 2).

Calculation of the correlation coefficient showed that the average bean yield has a direct strong relationship with the total number of beans per plant ($r=0.93$) and a strong inverse relationship with the bean length ($r=-0.88$). As the length of the beans increased, the average bean yield decreased. At the same time, as the bean length increased, the number of beans decreased, as evidenced by a strong inverse relationship between these indicators ($r=-0.93$).

In this study, the productivity of ripe seeds of a single plant has a strong direct relationship with the number of beans per plant ($r=0.96$) and the average yield over

three years ($r=0.91$) and the inverse strong relationship with bean length ($r=-0.79$). There is a clear pattern: varieties with a longer bean length produce fewer seeds, which affected the lower productivity of ripe seeds. The

same pattern exists for the yield of ripe seeds, which has a strong direct relationship with the number of beans on the plant ($r=0.96$), the average yield over three years ($r=0.91$), and the length of the bean ($r=-0.78$).

Table 2. Matrix of correlations between morphological features and economically valuable indicators of cowpea

Indicator	Plant height, cm	Number of beans per plant, pcs.	Bean length, cm	Average bean yield for three years, t/ha	Number of seeds in a bean, pcs.	Productivity of ripe seeds per one plant, g	Yield of ripe seeds, t/ha	Weight of 1,000 seeds, g
Plant height, cm	1							
Number of beans per plant, pcs.	0.38	1						
Bean length, cm	-0.66	-0.93	1					
Average bean yield for three years, t/ha	0.46	0.93	-0.88	1				
Number of seeds in a bean, pcs.	-0.25	0.18	0.00	-0.06	1			
Productivity of ripe seeds per one plant, g	0.14	0.96	-0.79	0.91	0.26	1		
Yield of ripe seeds, t/ha	0.13	0.96	-0.78	0.91	0.26	1.00	1	
Weight of 1,000 seeds, g	-0.82	-0.82	0.96	-0.77	-0.04	-0.63	-0.63	1

Source: based on the conducted research

In addition, a direct strong relationship was established between the weight of 1,000 seeds and the length of the bean ($r=0.96$) and an inverse relationship with the height of plants ($r=-0.82$), the number of beans per plant ($r=-0.82$), and the average yield of beans ($r=-0.77$).

Significant differences were found between the weight of 1,000 seeds and the length of the bean ($r=0.96$) and the productivity of ripe seeds of one plant with the number of beans per plant ($r=0.96$).

The results of the study show that abiotic factors play a very important role in the growth and development of cowpea. The results of the analysis of agrometeorological conditions during cowpea cultivation indicate that the duration of interphase periods depends on environmental factors such as heat and moisture. As a general pattern, it can be noted that at the beginning of the growing season of the studied crop, the rate of onset of development phases is significantly affected by the amount of precipitation. Knowledge of the duration of the growing season is necessary primarily to get an idea of the early ripening of the variety and establish the possibility of its cultivation in a particular soil and climatic zone.

Research conducted by J.L. Hatfeld and J.H. Prueger (2015) also shows that plant growth and development depend on temperature and that each variety has a characteristic temperature range expressed by minimum, maximum, and optimum. According to L.F. Oliveira (2018) individual phenological phases, such as flowering and formation of cowpea beans, largely depend on the availability of soil moisture. In addition, temperatures outside the optimal value can affect seed germination up to the final stage of development (Sehgal et al., 2018). For cowpea, the

reproductive stage is most sensitive to rising temperatures, which leads to the loss of flower buds and beans, and reduces seed productivity (Singh et al., 2010). Therefore, the selection of tolerant varieties in this context will be crucial to minimise losses due to climate change (Barros et al., 2021).

An increase in the temperature regime leads to an increase in vegetative development, since the optimal temperature for vegetative development is higher than the optimal temperature for reproductive development (Hatfeld & Prueger, 2015). Leaves adapt to high temperatures due to effective thermal resistance mechanisms that allow leaf temperature control through transpiration during periods with elevated temperatures (Sehgal et al., 2018). Plants tend to channel their resources to overcome heat stress by limiting photosynthesis, which is essential for reproductive development. Therefore, exposure to high temperatures on plants during the grain development phase, even for a short period, can accelerate leaf ageing, reduce the number and weight of seeds, and reduce crop yields (Sita et al., 2018). In addition, the root system is more sensitive to the stress effects of high temperatures than the aboveground part of plants, as indicated by an increase in the ratio of vegetative mass to roots (Barros et al., 2021).

The weight of 100 seeds is one of the most important marketing characteristics, since the heavier the seeds, the higher the price for them. During processing, the weight of seeds is also important information for designing appropriate processing machines that can process the maximum weight of seeds (Gondwe et al., 2019). Seed size in cowpea plays a key role, because it directly indicates the yield of the variety and, together with colour standardisation,

determines the corresponding grain quality (Gunathilake et al., 2016). Seed size also reflects seed quality indicators, and therefore, plant growth is affected by the size of the seed that was sown (Ambika et al., 2014). Under appropriate conditions, germination and subsequent growth of varieties with large seeds will definitely provide more seedlings compared to small seeds, which may contribute to increased crop productivity (Krisnawati et al., 2015).

Protein accumulation in cowpea increases significantly from 10 to 20 days after flowering and the maximum content is observed during the period of full ripeness, as a result, farmers often cannot recognise the phase of increasing nitrogen content in cowpea seeds for better protein intake. Ishikawa et al. (2021) note that to increase protein intake from green beans, a visual method has been developed to determine bean elongation, namely, the relative length of beans, which allows estimating the nitrogen content of unripe seeds. This is a simple visual method for determining the optimal time to harvest green beans and is therefore very practical for farmers.

There are morphological differences between grain cowpea and spearhead in terms of bean characteristics, including bean length, bean tenderness, bean cracking, and the presence of fibres in the bean (Suanum et al., 2016).

Correlation analysis is an important tool for determining relationships between plant traits, but correlation coefficients can be misleading if the correlation between two traits is the result of indirect exposure to other traits (Bizeti et al., 2004).

Undensi et al. (2012) recorded a significant positive correlation between flowering and bean maturation in some cowpea varieties and concluded that there is a strong association between the plant growth period before flowering and the time of bean or seed development. The early start of flowering and ripening of beans is crucial for increasing yields in regions with a short period of moisture. Delayed flowering and maturation of beans can prevent the formation of proper quality beans and seeds in cowpea (Aliyu et al., 2022). Watcharatpong et al. (2020) note that the fibre content (cellulose, lignin, and haemicellulose) correlates with the tenderness (softness/hardness) and cracking of beans.

Important factors affecting cowpea yield are the number of beans per plant and the number of seeds in them (Srinivas et al., 2017).

Aliyu et al. (2019) indicate that a strong inverse relationship has been established between phenological traits and yield components.

Among the quantitative agronomic features associated with cowpea yield is bean length (Ogunkanmi et al., 2013).

The results obtained in this study show that high temperatures affect the metabolism of cowpea plants, causing changes in plant growth. However, the varieties had different physiological responses due to each variety's different need for thermal energy. Therefore, the selection of varieties that are resistant to high temperatures is an important measure. However, the influence of temperature combined with a lack of moisture can be more harmful, so it is necessary to understand the interaction of temperature and moisture in different varieties of cowpea to develop effective strategies for adapting cultivation technology.

CONCLUSIONS

Early maturation periods are characterised by the varieties American improved, Kafedralna, Gasson with the duration of the period from germination to the beginning of harvesting ripeness of the pod – 58-62 days and the duration of the period from germination to biological ripeness of seeds – 92-94 days. For the vegetable industry, bush varieties Kafedralna and U-Cha-Kontou with a plant height of 48.5-54.9 cm are promising, forming tender long beans with a weak parchment layer (22.8-23.6 cm). Groik and Gasson varieties are valuable for growing seedlings due to their small light-coloured seeds with a plant height of 61.8-100.5 cm and the formation of a large number of short beans on the plant (12.8-15.6 cm). The relationship between morphological features and indicators of the economic value of cowpea was revealed. The average bean yield had a direct strong relationship with the total number of beans per plant ($r=0.93$) and a strong feedback with the length of the bean ($r=-0.88$). The average yield of beans and the number of beans per plant decreased with increasing bean length, as evidenced by a strong inverse relationship between these indicators ($r=-0.93$).

Promising areas for research are the isolation and selection of cowpea genotypes with the desired agronomic properties for inclusion in future breeding programmes aimed at genetically improving seed yield.

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Оцінка росту та розвитку сортів вігні за фенологічними і морфологічними спостереженнями

Анотація. Пошук та відбір сортів вігні з оптимальними господарськими ознаками є ключовим фактором у розробці технологій вирощування для підвищення рівня врожайності. Мета досліджень полягала у визначенні фенологічних і морфологічних ознак сортів вігні кущової та встановленні взаємозв'язку між господарсько-цінними показниками та погодними умовами. Провідними методами дослідження цієї проблеми є польовий – для оцінки росту та розвитку сортів вігні, статистичний – для проведення дисперсійного та кореляційного аналізу оцінювання морфологічних ознак та господарсько-цінних показників вігні. Вивчено п'ять сортів кущової вігні: Гроїк (Ізраїль), Кафедральна (Україна), Гассон (В'єтнам), Американська покращена (США), У-Тя-Контоу (Китай). З'ясовано, що тривалість фенологічних фаз сортів вігні залежала від погодних умов. Сорти вігні кущової характеризувались ранніми строками досягання за суми ефективних температур 192,0-207,5 °C і суми опадів 30,54-31,65 мм. Більш ранніми строками досягання виявилися сорти Американська покращена, Кафедральна, Гассон з тривалістю періоду від сходів до початку збиральної стиглості лопатки 58-62 дів і тривалістю періоду від сходів до біологічної стиглості насіння – 92-94 доби. Для овочевого напрямку перспективними виявилися кущові сорти Кафедральна та У-тя-Контоу з висотою рослин 48,5-54,9 см, які формували ніжні довгі боби зі слабким пергаментним шаром (22,8-23,6 см). Сорти Гроїк та Гассон виявилися цінними для вирощування проростків завдяки дрібному світло забарвленому насінню з висотою рослин 61,8-100,5 см та формуванням великої кількості коротких бобів на рослині (12,8-15,6 см). Встановлений сильний взаємозв'язок між морфологічними ознаками та господарсько-цінними показниками вігні. Середня врожайність бобів мала прямий сильний зв'язок із загальною кількістю бобів на рослині ($r=0,93$) та сильний обернений зв'язок із довжиною бобу ($r=-0,88$). Із збільшенням довжини бобів зменшувалась середня урожайність бобів і кількість бобів на рослині, про що свідчить сильний обернений зв'язок цих показників ($r=-0,93$). Матеріали статті становлять практичну цінність для збільшення видового різноманіття овочевих культур та покращення забезпечення населення цінними харчовими продуктами

Ключові слова: бобові культури, боби-лопатки, цвітіння, збиральна стиглість, висота рослин, довжина бобу, насіння

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Influence of the fertiliser system on the soil nutrient regime and onion productivity

Abstract. Modern approaches to optimising the nutrition of vegetable plants often have low efficiency and negatively affect various components of the agrobiocoenosis (soil, microbiota). In this regard, the purpose of the study was to determine the influence of various fertiliser systems on the yield and quality of onions in irrigated vegetable-fodder crop rotation, changes in the main agrochemical and microbiological parameters of the soil. The study was conducted in the laboratory of agrochemical research and quality of products of the Institute of Vegetable and Melon Growing of NAAS of Ukraine. Research methods: long-term stationary field experiment; laboratory determination of agrochemical and microbiological parameters of soil and biochemical composition of bulbs; statistical data processing. It is determined that a significant increase in the content of nitrate nitrogen in the arable soil layer is provided by the use of mineral fertilisers, mobile compounds of phosphorus and potassium – mineral and systems. The use of mineral fertilisers leads to an increase in the number of fungi in the soil rhizosphere and a decrease in the potential activity of nitrogen fixation, which worsens the microbiological activity of the soil. The application of organic fertilisers in combination with mineral fertilisers leads to an increase in the number of nitrogen-fixing microorganisms and potential nitrogen-fixing activity, which improves soil fertility. The use of organo-mineral and mineral fertiliser systems leads to an increase in the yield of onions by 6.6–8.6 t/ha or 41.3–53.8%. When using biologised nutrition optimisation systems, the yield increases by

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19.4-23.8%. The maximum level of onion yield is indicated for the introduction of organo-mineral fertiliser systems (63 t/ha of humus + randomly $N_{45}P_{45}K_{45}$; 36 t/ha of humus + locally $N_{45}P_{45}K_{45}$); causing an increase in yield by 8.2-8.6 t/ha. There is a certain pattern of a decrease in the dry matter content in bulbs and an increase in the nitrate content under various fertiliser systems. The research materials are of practical value for the implementation of nutrition optimisation systems aimed at both sustainable growth in the yield of vegetable products and maintaining the level of soil fertility of vegetable agrobiocoenoses

Keywords: nutrition optimisation, vegetable and fodder crop rotation, yield, quality composition of the bulb

INTRODUCTION

Onions are a valuable vegetable plant that ranks first among other vegetable crops in terms of world production. Increasing gross production without expanding the area of sowing crops is possible only through the use of intensive cultivation technologies, where the key aspect is the system of nutrition optimisation (fertilisation). At the same time, the use of high fertiliser rates often has a number of negative consequences: deterioration of product quality, accumulation of nitrates in bulbs, and negative impact on the microbiota and soil fertility parameters (Horodniy & Bykina, 2000).

There are many opinions of researchers regarding the mechanisms of improving the nutrient regime of the soil and the efficiency of applying various types of fertilisers (Armstrong *et al.*, 2000; Kuts *et al.*, 2017; Novikova, 2020;). Some researchers suggest that the most effective factor in increasing the availability of mobile forms of nitrogen, phosphorus and potassium in the arable soil layer is the use of mineral fertilisers, especially the localisation of their application (Backes *et al.*, 2018; Bezabih & Girmay, 2020). A number of agricultural chemists point to a decrease in the mobility of nutrients at the end of the growing season of agricultural crops for the use of mineral fertilisers, and a more prolonged positive effect on the soil nutrient regime for the use of organic fertilisers (Diaz-Perez *et al.*, 2018). According to the results of a number of studies, it was found that the most effective way to improve the nutrient regime of the soil during the entire growing season of plants is to use the organo-mineral fertiliser system (Colla *et al.*, 2018; Moraes *et al.*, 2018; Moralet & Ansari, 2022). The level of soil fertility also depends on the fertiliser system, since fertilisers affect many parameters of fertility: nutrient regime, absorption capacity, structure and water-physical properties, microbiological activity, etc.

Different fertiliser systems have different effects on crop rotation productivity and soil fertility indicators (Moraes *et al.*, 2018; Bezabih & Girmay, 2020;). It is established that the effectiveness of fertilisers varies depending on the level of soil fertility, doses and methods of fertilisation, balance in nutrition elements, and distribution of fertilisers between crop rotations (Kuts *et al.*, 2017; Novikova, 2020;). Many studies have found that the use of a mineral fertiliser system contributes to an increase in soil degradation processes and a gradual decrease in crop rotation productivity. Thus,

in studies on acidic forest soils of the Western Forest-Steppe, long-term use of mineral fertilisers caused a decrease in crop rotation productivity relative to the option without fertilisers (Backes *et al.*, 2018;). Whereas systematic joint application of the organo-mineral fertiliser system on the liming background (10 t/ha of manure + $N_{65}P_{68}K_{68}$) contributes to the development of high crop rotation productivity at the level of 6.15-6.78 t/ha of grain units (Kaminsky *et al.*, 2012). Many researchers have proven that a combination of organic and mineral fertilisers plays an important role in optimising crop nutrition in crop rotation (Backes *et al.*, 2018; Colla *et al.*, 2018). But for the combination of applying organic and mineral fertilisers, the dose of the latter should be optimised considering the amount of nutrients in the composition of organic fertilisers, the availability of soil, and the crop rotation system (Backes *et al.*, 2018). The use of manure combined with mineral fertilisers increases the efficiency of the latter, promotes the growth of yield and product quality, increases the recycling of nutrients and carbon in agrocoenoses, ensures the preservation and reproduction of soil fertility (Backes *et al.*, 2018; Colla *et al.*, 2018; Erkalo *et al.*, 2022).

The efficiency of using mineral fertiliser systems in technological schemes for growing onions depends on the type of soil, technological growing conditions, and irrigation conditions. For each soil, climatic and technological growing conditions, a certain range of fertiliser rates can be distinguished, which is economically feasible. Thus, according to Ethiopian researchers, in irrigation conditions, it is economically feasible to apply the fertiliser for growing onions at a rate $N_{200}P_{220}S_{40}$, which provides a yield of 47.4 t/ha, causes an increase in the height of the plant, the length of leaves, the diameter and length of the bulb, its average weight, the content of dry soluble substances in the bulbs (Shura *et al.*, 2022). According to Iraqi researchers, by using conditionally suitable water for irrigation, an increase in the efficiency of fertilisers depends on the norm of potassium, which is economically feasible in the range of 60-80 kg/ha (Khalefah *et al.*, 2022).

Localisation of the use of mineral fertilisers is also a source of increasing their efficiency, which is especially important for nitrogen fertilisers. In the research by Mexican scientists, local application of nitrogen fertilisers provided a significant increase in bulb weight and

yield, but often created conditions for excessive supply of nitrogen to plants, which caused the appearance of a thick neck, germination and rotting of bulbs during storage (Kitila *et al.*, 2022).

In conditions of a shortage of organic fertilisers, a combination of green manure and microbiological fertilisers can be used as an alternative. Thus, the use of microbiological fertilisers (Azotovit, Phosphatovit) for growing onions contributes to an increase in the leaf surface area by 27–80 cm², increase in yield by 0.9–3.9/ha or by 6.0–26.3% (Kanevskaya, 2022). The range of microbiological preparations for optimising nutrition and enhancing plant growth processes is constantly growing, which allows introducing comprehensive solutions to the problem of mineral nutrition of plants.

Currently, the problem of studying the influence of various fertiliser systems on soil fertility, its agrochemical and microbiological state in vegetable agrobiocoenoses remains neglected. The solution of this problem would allow creating or correcting fertiliser systems for vegetable plants, which, in addition to increasing yields, will ensure the preservation or reproduction of soil fertility, reduce the chemical load on plant communities, and increase the economic efficiency of using fertilisers, especially mineral fertilisers.

The purpose of the study – to investigate the dynamics of agrochemical and microbiological parameters of the arable soil layer, the level of yield and qualitative composition of onion products under various fertiliser systems in the irrigated vegetable-fodder crop rotation of the Left-Bank Forest-Steppe of Ukraine.

MATERIALS AND METHODS

Scientific research was conducted at the Institute of Vegetable and Melon Growing of the National Academy of Agrarian Sciences of Ukraine in the laboratory of agrochemical research and quality of products, during 2018–2020. The soil of the experimental site is represented by typical low-humus heavy loamy chernozem on loessial soil (humus content – 4.3%; hydrolysed nitrogen – 139; mobile phosphorus – 135–352, and exchange potassium – 66–177 mg/kg of soil, pH of salt extract – 5.7).

The study is a continuation of the long-term stationary experiment that was started in 1968. Determination of the effectiveness of various fertiliser systems was carried out in an irrigated 9-field vegetable-fodder crop rotation: barley with the sowing of perennial grasses – alfalfa of the first and second years of use – cucumber – winter wheat – onion – tomato – white cabbage – table beet.

Field studies were conducted according to the experiment scheme:

1. No fertiliser (control)
2. Humus 36 t/ha (scattered)
3. N₉₀P₉₀K₉₀ (randomly)
4. N₃₂₀P₂₀₀K₁₃₀ (randomly) + foliar top dressing with Reakom–SO in three terms.

5. Humus 36 t/ha + N₄₅P₄₅K₄₅ (randomly)

6. Humus 63 t/ha + N₄₅P₄₅K₄₅ (randomly)

7. Humus 36 t/ha + N₄₅P₄₅K₄₅ (locally)

8. Humus 63 t/ha + N_{22.5}P_{22.5}K_{22.5} (locally)

9. Green manure system of onions includes ploughing 3 t/ha of straw with the addition of Ecostern stubble biodestructor (1 l/ha) + soaking seeds in a solution of Azotofit-r biological preparation (1 l/t) + foliar top dressing with a solution of Organik balans biological preparation (1 l/ha) in three terms

Biologised fertiliser system includes an aftereffect of 40 t/ha (applied to the predecessor – cucumber) + ploughing of 3 t/ha of straw with the addition of Ecostern stubble biodestructor (1 l/ha) + soaking of seeds in a solution of Azotofit-r biological preparation (1 l/r) + foliar top dressing with a solution of Organik balans biological preparation (1 l/ha) in three terms

Foliar top dressing was carried out in three periods: phase 3–4 of true leaves, phase 6–8 of true leaves, and the beginning of bulb formation.

Total plot area 58.3 m², accounting area – 19.6 m², repetition – four-fold, systematic placement of plots in two tiers (Bondarenko & Yakovenko, 2001).

In soil samples, the content of nitrate nitrogen was determined: in fresh – the content of nitrate nitrogen (DSTU 4729:2007, 2008) colourimetrically with disulfophenolic acid, in air-dry – mobile phosphorus compounds and exchange potassium according to the modified Chirikov method, followed by determination of phosphorus – photometrically, potassium – on a flame photometer (DSTU 4115:2002, 2003).

Determination of soil microbiological activity included determination of:

- the number of fungi using the cup method on an acidified Czapek medium (Zvyagintseva, 1980);
- the number of nitrogen-fixing microorganisms using the method of limit dilutions using acetylene test on Ashby semi-liquid media (Villemin *et al.*, 1974);
- potential activity of nitrogen fixation (activity that is noted for the formation of optimal carbon, temperature and water regimes for nitrogen-fixing microorganisms) for the use of the acetylene method in rhizospheric soil with the addition of glucose solution (Umarov, 1976; Bilay, 1982);
- the mineralisation coefficient (an indicator of the intensity of the processes of mineralisation and assimilation of nitrogen compounds in the soil, expressed by the ratio of the number of bacteria and streptomycetes that absorb mineral forms of nitrogen (cultivated on starch-ammonia agar), to the number of bacteria that consume nitrogen of organic compounds (cultivated on meat-peptone agar) (Zvyagintseva, 1980).

The quality of onion products was determined using the following methods: dry matter – (DSTU 7804:2015, 2016), total sugar – (DSTU 4954:2008, 2009), ascorbic acid – (DSTU 7803:2015, 2016), nitrates – (DSTU 4948:2008, 2009).

RESULTS AND DISCUSSION

Onions of the Tkachenkivska variety were grown according to the technology recommended for the zone (DSTU 8072:2015, 2017): cultivation – ploughing for 22–25 cm with continuous cultivation in autumn and early spring harrowing; sowing seeds in the third 10 days of March – first 10 days of April with a norm of 650 thousand units/ha; irrigation – sprinkling in 4 terms with a rate of 250–35 m³/ha; plant protection – a complex of recommended herbicides, fungicides, and insecticides; harvesting with mechanical digging and drying of bulbs in the field.

The results of studies show that the content of mobile forms of basic nutrients in 0–30 cm soil layer during the cultivation of onions significantly varied depending on fertiliser systems. Thus, in the control option, the amount of nitrate nitrogen was the lowest and ranged from 23.6–26.5 mg/100 g of soil. Application of high doses of mineral fertilisers (N₃₂₀P₂₀₀K₁₃₀ + top dressing “Reakom-SO”) caused the highest accumulation of nitrate nitrogen in 0–30 cm soil layer during all periods of soil analysis from 111.9 in June to 45.9 mg/kg before harvesting (Fig. 1).

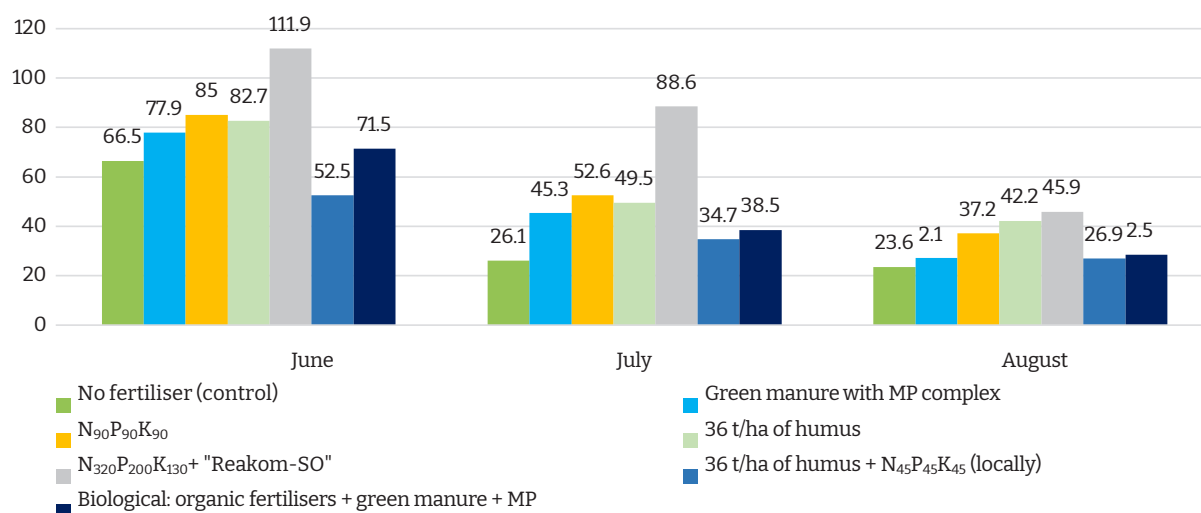


Figure 1. Content of nitrate nitrogen in the 0–30 cm soil layer, depending on the fertiliser system (average for 2018–2020)

A high level of soil supply with nitrogen compounds is also noted during the period of maximum increase in the vegetative mass of the bulb (June – July) according to the nutrition optimisation system for the use of straw as green manure in combination with microbial preparations (45.3–77.9 mg/kg), for the introduction of N₉₀P₉₀K₉₀ (52.6–85.0 mg/kg) and 36 t/ha of humus (49.5–82.7 mg/kg).

During the period of physiological ripeness, the amount of nitrate nitrogen sharply decreases to the level of 26.8–45.9 mg/kg of soil according to the fertiliser systems under study. The explanation for this phenomenon is, firstly, the active accumulation of nitrogenous substances by plants, and secondly, the migration of soil nitrogen compounds to the suborder layer of the soil. However, the pattern characteristic of previous periods is also noted in the future: the combined use of mineral and organic fertilisers causes high values of the indicator (37.2–45.9 mg/kg of dry soil); the use of straw ploughing in combination with microbial preparations provides a lower level of nitrogen regime in the soil (27.1–28.5 mg/kg of dry soil).

Thus, it is noted that the improvement of the nitrogen regime of the soil provides, in addition to the use of high doses of nitrogen fertilisers (N₃₂₀), also the use of organic fertilisers (humus). At the same time, there is no peak increase in the provision of the arable soil layer

with nitrate nitrogen in the initial periods of vegetation of the crop, as with the use of N₃₂₀P₂₀₀K₁₃₀, but this parameter increases with the introduction of organic and organo-mineral fertiliser systems relative to control (by 105–120%). Such data are noted in the research of South Korean scientists (Lee, 2010).

It is indicated that the joint application of organic and mineral fertilisers (locally N₄₅P₄₅K₄₅) creates the best conditions for phosphorous nutrition for onion plants (Fig. 2). Under such conditions, the content of mobile phosphorus compounds in the soil increases to 316–352 mg/kg of dry soil, depending on the development phases of onion plants.

Application of N₉₀P₉₀K₉₀ and N₃₂₀P₂₀₀K₁₃₀ in combination with microfertilisers Reakom-SO contributes to a high level of provision of the arable soil layer with mobile phosphorus compounds – 252–320 mg/kg of dry soil.

When using organic fertilisers and ploughing straw with microbial preparations, the content of mobile phosphorus was almost at the same level and amounted to 239–252 mg/kg for organic systems and 241–281 mg/kg for biological fertiliser systems, which significantly exceeded the control (142–156 mg/kg of dry soil). It is also noted that if the mineral and organic fertiliser systems of onions provide the soil with mobile phosphorus compounds during the growing season does not change, then

with the use of a biological fertiliser system (organic fertilisers + straw + microbial preparations) there is a certain

tendency to reduce the content of mobile phosphorus compounds at the end of the growing season of the crop.

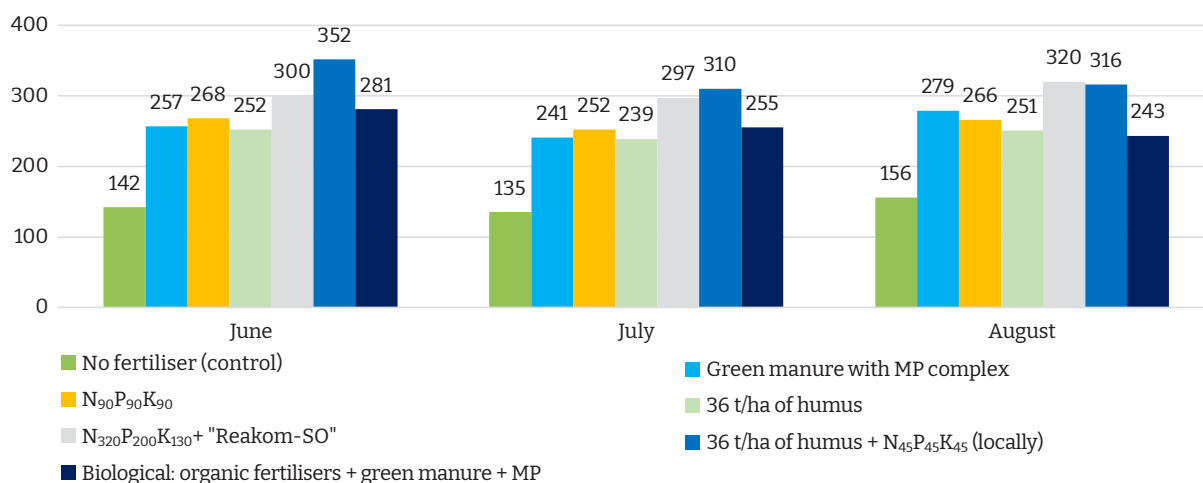


Figure 2. Dynamics of mobile phosphorus in 0-30 cm soil layer depending on the fertiliser system of Tkachenkivska onion variety (average for 2018-2020)

Similar patterns are also inherent in the supply of potassium (Figure 3). During the growing season, a high level of soil supply with potassium mobile compounds is indicated for organo-mineral (36 t/ha humus + locally N₄₅P₄₅K₄₅) and biologised systems for optimising plant nutrition (with the content of exchange potassium in

0-30 cm soil layer at the level of 148-177 mg/kg of soil). For the green manure fertiliser system with a complex of microbial preparations, the content of exchange potassium in the soil significantly exceeded the control – 66-79 mg/kg of dry soil, although it was less than for the biological fertiliser system (98-106 mg/kg).

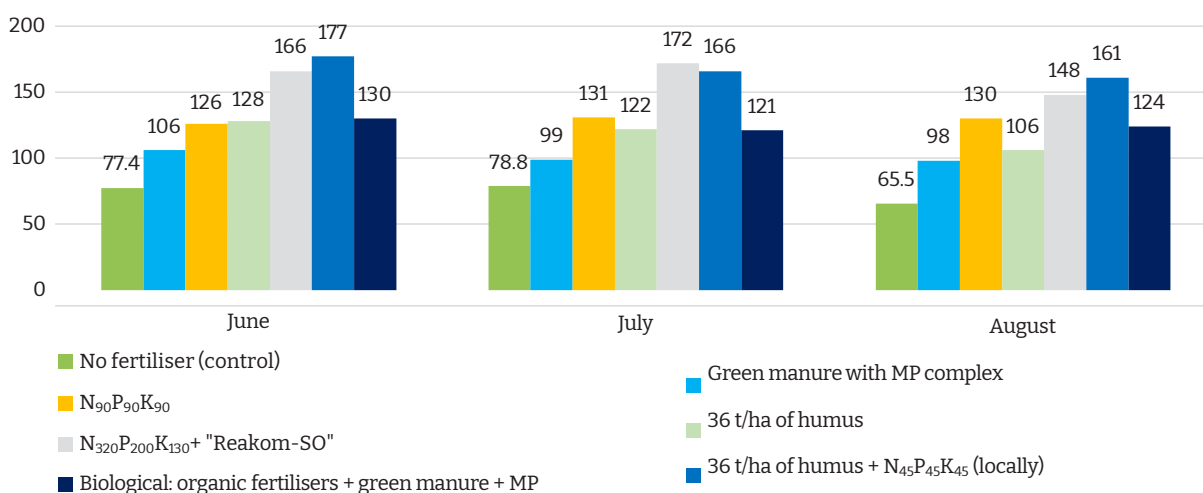


Figure 3. Content of exchange potassium in 0-30 cm soil layer, depending on the onion fertilisation system (average for 2018-2020)

Thus, the introduction of biologised and green manurefertiliser systems with a complex of microbial preparations that can be actively involved in the systems of growing agricultural plants according to the principles of organic farming, determine the optimal level of supply of plants with mobile compounds of phosphorus (241-281 mg/kg) and potassium (121-130 mg/kg) with an excess of 69-75% of the corresponding indicators on the control variant. The effectiveness of a complex of microbial preparations in combination with organic and green

manure fertilisers has also been proven by studies conducted in Venezuela (Blanco *et al.*, 2021), in India Parray *et al.*, (2016), Spain-Barea *et al.*, (2013).

Analysing the influence of onion fertilisation systems on the microbiological activity of the soil, the dependence of the content of nitrate nitrogen in the soil on the number of nitrogen-fixing microorganisms is indicated. A certain increase in the number of nitrogen-fixing microorganisms (46-71 thousand colony-forming units (CFU)/g of dry soil) and potential

nitrogen-fixing activity (28.6-34.2 nmol C_2N_2 /g of dry soil per hour) according to the green manure, biological, and organo-mineral fertiliser system (Table 1). An increase in the number of nitrogen-fixing micro-

organisms was also noted when using an organic fertiliser system with the introduction of humus under the culture; the number of microorganisms was 68.4 thousand. CFU/g of dry soil.

Table 1. Changes in soil microbiological activity, onion growth and development phase of 6-8 true leaves, average for 2018-2019

Fertilisation system	Number of microorganisms, thous. CFU/g of dry soil		Potential nitrogen fixation activity, nmol C_2N_2 /g of dry soil per hour	Mineralisation coefficient
	Fungi	Nitrogen-fixing microorganisms		
Without fertilisers (control)	18.8	38.8	21.8	0.38
Green manure: straw 3 t/ha + stubble biodestructor (1 l/ha) + seed treatment with Azotophyte-r (1l/t) + foliar top dressing with Organik balans (1 l/ha)	23.6	46.3	28.6	0.52
36 t/ha of humus (scattered)	20.7	68.4	30.5	0.41
$N_{90}P_{90}K_{90}$ (randomly)	22.3	42.5	24.1	0.47
36 t/ha of humus + $N_{45}P_{45}K_{45}$ (randomly)	19.3	52.3	28.6	0.41
Biological: aftereffect 40 t/ha of manure + straw 3 t/ha + stubble biodestructor (1 l/ha) + seed treatment with Azotophyte-r (1l/t) + foliar top dressing with Organik balans (1 l/ha)	21.1	71.5	34.2	0.44
Least significant difference (LSD) _{0.95}	1.32	5.12	4.55	-

The use of mineral fertilisers causes a certain decrease in the potential activity of nitrogen fixation relative to other fertiliser systems to the level of 24.1 nmol C_2N_2 /g of dry soil per hour.

It is indicated that the number of soil fungi for the use of humus both separately and in combination with mineral fertilisers (19.3-20.7 thous. CFU/g of dry soil) is significantly reduced relative to other fertiliser systems and is within the values in the control variant (18.8 thous. CFU/g of dry soil). According to the green manure, biological, and mineral fer-

tiliser systems, the number of soil fungi increases. This does not correspond to the results of studies obtained by Serbian researchers, where for the use of microbial preparations (*Azotobacter chroococcum*, *Pseudomonas fluorescens*) there is a decrease in the number of soil fungi (Čolo *et al.*, 2014).

Improvement of agrochemical and microbiological parameters of the soil with the use of organic and mineral fertilisers provides an increase in the yield of onions and an improvement in the biochemical composition of products (Table 2).

Table 2. Influence of fertiliser systems on the yield and quality of onions of the Tkachenkivska variety (average for 2018-2020)

Onion fertiliser system	Yield		Biochemical parameters of bulbs			
	t/ha	increment	dry substance, %	total sugar, %	ascorbic acid, mg/100 g	nitrites, (MS ¹ 80 mg/kg)
Without fertilisers (control)	16.0	-	12.8	9.6	7.5	35.5
Organic system						
36 t/ha of humus (scattered)	22.3	6.3	13.1	9.8	7.5	66.0
Mineral system						
$N_{90}P_{90}K_{90}$ (randomly)	22.6	6.6	11.5	9.4	7.2	124.4
$N_{300}P_{200}K_{130}$ (randomly) + Reakom-SO	23.6	7.6	11.4	11.2	7.5	136.0

Table 2, Continued

Onion fertiliser system	Yield		Biochemical parameters of bulbs			
	t/ha	increment	dry substance, %	total sugar, %	ascorbic acid, mg/100 g	nitrates, (MS ¹ 80 mg/kg)
Organo-mineral system						
36 t/ha of humus + N ₄₅ P ₄₅ K ₄₅ (randomly)	22.8	6.8	13.3	9.5	7.4	60.1
63 t/ha of humus + N ₄₅ P ₄₅ K ₄₅ (randomly)	24.6	8.6	12.5	9.4	7.3	59.0
36 t/ha of humus + N ₄₅ P ₄₅ K ₄₅ (locally)	24.2	8.2	11.8	10.6	7.7	72.1
63 t/ha of humus + N _{22.5} P _{22.5} K _{22.5} (locally)	22.8	6.8	11.9	10.1	7.5	111.7
Biological fertiliser system						
Green manure (straw 3 t/ha + stubble biodestructor (1 l/ha) + seed treatment with Azotophyte-r (1l/t) + foliar top dressing with Organik balans (1 l/ha)	19.8	3.8	12.8	10.2	7.6	57.0
Biologised (aftereffect 40 t/ha of manure + straw 3 t/ha + stubble biodestructor (1 l/ha) + seed treatment with Azotophyte-r (1l/t) + foliar top dressing with Organik balans (1 l/ha)	19.1	3.1	13.1	10.0	7.8	59.1
LSD _{0.95}	2.17		0.94	1.25	0.76	15.8

Note: ¹ MS – molar solution

A high level of yield is noted for organo-mineral fertiliser systems. Thus, for the introduction of 63 t/ha of humus with N₄₅P₄₅K₄₅ randomly or less than the humus rate (36 t/ha), but with the use of mineral fertilisers locally, the yield of onions increases by 8.2-8.6 t/ha or 51.3-53.8%.

Using only mineral fertilisers at a rate N₉₀P₉₀K₉₀ randomly provides a yield increase of 41.3%. Increasing the norm to N₃₀₀P₂₀₀K₁₃₀ (to ensure a yield of 30 t/ha) does not significantly increase the yield of onions.

The use of a system for optimising plant nutrition with the involvement of green manure fertilisers and microbial preparations causes the smallest increase in yield at the level of 3.1-3.8 t/ha or 19.4-23.8%. The corresponding results were obtained by other researchers. Thus, in the conditions of Ethiopia, inoculation of seeds with preparations containing bacteria from genera *Pseudomonas*, *Bacillus*, *Azotobacter*, *Enterobacter* and *Azospirillum* was effective for growing onions (Mekonnen & Kibret, 2021). According to Indian researchers, the use of a consortium of microorganisms *Azotobacter* sp. + *Sphingobacterium* sp. + *Burkholderia* sp. provides an increase in crop yield by 12.0% (Tinna et al., 2020). The combination of microbial preparations with mineral fertilisers is also effective, which reduces the amount of the latter (Devi & Ado, 2005; Rafique et al., 2019).

On average, over the years of research, there was a certain pattern of reducing the dry matter content in bulbs under different fertiliser systems. The dry matter content was significantly reduced for mineral fertiliser systems, as well as for the use of 36 t/ha of humus +

locally N₄₅P₄₅K₄₅; the indicator was 11.4-11.8%. At the same time, using only organic fertilisers (36 t/ha of humus) and with the joint application of 36 t/ha of humus and randomly N₄₅P₄₅K₄₅ or according to the biological fertiliser system, there is a tendency to increase this biochemical parameter (13.1-13.3%).

According to the effect on the total sugar content in bulbs, a variant with an estimated dose of mineral fertilisers is distinguished, which provides a significant increase in this indicator to the level of 11.2% relative to the control with a sugar content of 9.6%. According to other fertiliser systems, the total sugar content in bulbs ranged from 9.4-10.6%.

The content of ascorbic acid (vitamin C) in bulbs was not significantly affected by the fertiliser system. At the same time, the value of the indicator ranged from 7.2-7.8 mg/100 g with a vitamin content of 7.5 mg/100 g in the control.

The use of different types and rates of fertilisers leads to an increase in the content of nitrates in bulbs. There are quite high values of this parameter for the use of mineral fertiliser systems, as well as for organo-mineral fertiliser systems, where mineral fertilisers are applied locally. At the same time, the nitrate content in the bulbs increases to the level of 72.1-136.0 mg/kg of raw weight. For organo-mineral fertiliser systems, where mineral fertilisers are used randomly, as well as for green manure and biologised fertiliser systems, the nitrate content increases significantly relative to the control, but within smaller limits (up to the level of 57.0-60.1 mg/kg

of raw weight), which does not exceed the maximum allowable level (80 mg/kg of raw weight).

CONCLUSIONS

The maximum growth of the main parameters of the soil nutrient regime is provided by the use of mineral and organo-mineral fertiliser systems, as well as a biologised system, which includes the use of a complex of microbial preparations for the aftereffect of organic fertilisers in crop rotation.

Deterioration of some parameters of soil microbiological activity causes the use of mineral fertilisers (the number of fungi in the rhizospheric soil increases, the potential activity of nitrogen fixation decreases). The introduction of organic and organo-mineral systems for optimising the nutrition of onion plants provides an increase in the number of nitrogen-fixing microorganisms (52.3–71.5 thousand CFU/g of dry soil) and potential nitrogen fixation activity (46.3–71.5 nmol S_2N_2 /g of dry soil per hour).

The maximum level of onion yield was noted for the introduction of organo-mineral fertiliser systems (63 t/ha of humus + randomly $N_{45}P_{45}K_{45}$; 36 t/ha of humus + locally $N_{45}P_{45}K_{45}$); which contribute to an increase in yield in the range of 8.2–8.6 t/ha or 51.3–53.8%.

The use of organo-mineral fertiliser systems with the introduction of mineral fertilisers randomly, and the use of green manure and biologised fertiliser systems ensure the production of vegetable products with a high content of biologically active substances.

The continuation of these studies is aimed at solving two problems: 1) investigation of the transformation of the main parameters of chernozem soils of the forest-steppe of Ukraine under various fertiliser systems in vegetable agrobiocoenoses; 2) development of systems for optimising the nutrition of vegetable plants with a shortage of organic fertilisers and high cost of mineral fertilisers.

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Вплив системи удобрення на поживний режим ґрунту та продуктивність цибулі ріпчастої

Анотація. Сучасні підходи щодо оптимізації живлення овочевих рослин часто мають низьку ефективність та негативно впливають на різні компоненти агробіоценозу (ґрунт, мікробіоту). У зв'язку з цим метою досліджень було визначення впливу різних систем удобрення на формування врожайності та якості цибулі ріпчастої в зрошуваній овоче-кормовій сівозміні, зміну основних агрохімічних та мікробіологічних параметрів ґрунту. Дослідження проведено в лабораторії агрохімічних досліджень та якості продукції Інституту овочівництва і баштанництва НААН України. Методи досліджень: довготривалий стаціонарний польовий дослід, лабораторне визначення агрохімічних та мікробіологічних параметрів ґрунту, біохімічного складу цибулин, статистична обробка даних. Визначено, що істотне підвищення вмісту нітратного азоту в орному шарі ґрунту забезпечує використання мінеральних добрив, рухомих сполук фосфору та калію – мінеральна та органо-мінеральна системи удобрення. Використання мінеральних добрив зумовлює збільшення чисельності грибів у ризосфері ґрунту і зменшення потенційної активності азотфіксації, що погіршує мікробіологічну активність ґрунту. Внесення органічних добрив в поєднанні з мінеральними добривами, призводить до зростання кількості азотфіксувальних мікроорганізмів та потенційної активності азотфіксації, що покращує родючість ґрунту. Використання органо-мінеральних та мінеральних систем удобрення зумовлює підвищення урожайності цибулі ріпчастої на 6,6-8,6 т/га або 41,3-53,8%. За використання біологізованих систем оптимізації живлення урожайність зростає на 19,4-23,8%. Максимальний рівень урожайності цибулі ріпчастої зазначено за впровадження органо-мінеральних систем удобрення (63 т/га перегною + врозкид $N_{45}P_{45}K_{45}$; 36 т/га перегною + локально $N_{45}P_{45}K_{45}$); зумовлюючи підвищення урожайності на 8,2-8,6 т/га. Відмічена певна закономірність зниження в цибулинах вмісту сухої речовини та підвищення умісту нітратів за різних систем удобрення. Матеріали досліджень становлять практичну цінність для впровадження систем оптимізації живлення, що спрямовані як на стаке зростання урожайності овочевої продукції, так і збереження рівня родючості ґрунтів овочевих агробіоценозів

Ключові слова: оптимізація живлення, овоче-кормова сівозміна, врожайність, якісний склад цибулини

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Identification of chlorogenic acid in cotyledonous leaves and husks of common sunflower (*Helianthus annuus* L.)

Abstract. The relevance of the study is conditioned by the need to optimise the production process of the sunflower farming ecosystem by increasing environmental sustainability. Among the by-products of processing, the use of which is still not effective, the release of the powerful antioxidant chlorogenic acid has not been sufficiently studied. In this regard, this paper is aimed at revealing the patterns of accumulation and identification of phenolic compounds in cotyledons and sunflower husks. The purpose of the study is to identify phenolic compounds in cotyledonous leaves and husks of various genotypes of common sunflower (*Helianthus annuus* L.) by thin-layer chromatography. In the course of research on this problem, the authors used field, biochemical, and statistical research methods. To identify chlorogenic acid, a solvent system was used: chloroform – glacial acetic acid – methanol – water (60:32:12:8). Chromatography analysis was performed in the SORBFLIL TLC software suite. The object of research is the Auris common sunflower hybrid. It is established that the qualitative composition of the phenolic complex and the number of individual compounds has a pronounced variety specificity. On the chromatogram, this substance has an R_f of ~0.60 (for sunflower husks) and an R_f of ~0.74 (for cotyledonous leaves). It was found that the most effective solvent for chlorogenic acid is aqueous ethanol. Extraction of chlorogenic acid with 40% ethanol provides up to 10 mg/g of chlorogenic acid. In addition to chlorogenic acid, a small amount of caffeic acid was found in the alcohol extract. The materials of the study are of practical value for improving the processes related to the development of the productivity of sunflower crops and the rational use of processing by-products

Keywords: oilseeds, phenols, antioxidants, caffeic acid, chromatogram, photodensitogram

Suggested Citation:

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INTRODUCTION

At the current stage of crop production development as a science, increasing the ecological stability of varieties and plant communities is one of the important factors for intensifying crop cultivation technologies. In the conditions of increasing the number of technogenic means used to optimise the growing conditions of agricultural crops (fertilisers, pesticides, etc.), the role of ecological stability of varieties and agrocoenoses in the implementation of their potential productivity is constantly increasing. The maximum possibilities of plant adaptation to negative environmental factors are determined by the genotype and functional features of the synthesised metabolites, which, in particular, include polyphenolic compounds. Numerous studies show that phenolic compounds perform a wide range of regulatory and protective functions (Pedrosa *et al.*, 2000; Voitsekhivska *et al.*, 2015), directed against various negative environmental factors (infections, insects, UV radiation, temperature stress, etc.) (Levytskyi *et al.*, 2010; Makarenko & Levytskyi, 2013). These properties of polyphenols can determine the varietal characteristics of cultivated plants. The implementation of the stress-protective effect is based on the antioxidant properties of these compounds (Andersen & Markham, 2005; Alexieva *et al.*, 2013).

The biological activity of chlorogenic acid is primarily conditioned by its strong antioxidant effect (Parr & Bolwell, 2000). Its biosynthesis occurs exclusively in plants and, along with phenolic compounds, is widespread in the plant world (Clifford, 1999; Nakatani *et al.*, 2000). Chlorogenic acid is often found in higher dicotyledonous plants, ferns, and many medicinal plants, which are called "plant gold" in China (Liang Wang *et al.*, 2022). It has strong antioxidant, antifungal, antibacterial, and antiviral properties. A number of researchers have established anti-cancer and hepatoprotective effects, effects on the regulation of glucose metabolism, and positive effects on blood vessels (Levytskyi *et al.*, 2010; Liang Wang *et al.*, 2022). This property can be used both in medicine, in agriculture, and the food industry. Chlorogenic acid plays an important role in plant respiration, is a regulator of growth processes, occupies an important place in ensuring plant immunity, and performs protective functions. The content of chlorogenic acid depends on the varietal characteristics of plants, natural and climatic factors, and on the method of determination (Makarenko & Levytskyi, 2013; Voitsekhivska *et al.*, 2015; Labeiko, 2021).

Sunflower is one of the main industrial crops in Ukraine. Taking into account processing, it is considered a valuable oilseed crop using by-products of processing, mainly for feed purposes. However, it can be used as a source of production of various types of biofuels, potash fertilisers, paper, hexose and pentose sugar, etc. Common sunflower is also an important source of natural antioxidants and, compared to other crops, its seeds are characterised by the highest potential (Halvorsen *et al.*, 2002). The husk of sunflower seeds contains only 0.7-5.4% of the total amount of its phenolic compounds, while the rest is found in the kernel (Pedrosa *et al.*, 2000; Grana *et al.*, 2017). The predominant

phenolic compound of the sunflower kernel is chlorogenic acid, which is 43-73 % of all phenolic compounds (Weisz *et al.*, 2009) and has strong antioxidant, antifungal, and antibacterial properties (Levytskyi *et al.*, 2010). Scientific studies in most cases relate to the production and use of sunflower protein, and the production of the antioxidant chlorogenic acid from sunflower meal is rather neglected (Labeiko, 2015).

Chlorogenic acid is one of the most valuable compounds that is in great demand among consumers, primarily due to its antioxidant properties. The use of chlorogenic acid is promising in the fat and oil industry as an antioxidant of vegetable oils as a substitute for synthetic substances (tributylhydroquinone, butylhydroxytoluene, etc.) (Shapovalova *et al.*, 2013). However, the use of methanol to produce chlorogenic acid, which in the future can be used in food and supplements, is not desirable. In addition, pre-extraction of chlorogenic acid from meal would allow obtaining plant protein without phenolic compounds and using it for the production of light-coloured food (Labeiko, 2015).

In this regard, the purpose of the study was to identify composite phenolic complexes in the husk and cotyledon leaves of various genotypes of common sunflower (*Helianthus annuus* L.) by thin-layer chromatography.

MATERIALS AND METHODS

The study was conducted during 2017-2018 in the laboratory of biotechnology and cell engineering of the Department of Ecobiotechnology and Biodiversity of NUBiP of Ukraine. Field experiments were carried out in the stationary crop rotation of the Department of Crop Growing in the special subdivision of NUBiP "Agronomic Experimental Station" on typical low-humus chernozems.

The object of research was a hybrid of common sunflower (*Helianthus annuus* L.) Auris (resistant to herbicides based on tribenuron-methyl (SU), listed in the State register of varieties suitable for distribution in Ukraine (2022).

In the course of the study, a field research method was used to investigate the interaction of the research object with biotic and abiotic factors in the field.

Experiment scheme – fertiliser options: 1. $N_{60}P_{50}K_{90}$ (control); 2. $N_{60}P_{50}K_{90} + S_{30}$; 3. $N_{90}P_{75}K_{135} + S_{30}$; 4. $N_{120}P_{100}K_{180} + S_{30}$. Mineral fertiliser application rates were calculated using the balance method for the planned yield, considering the content of the main elements of mineral nutrition in the soil. Plot area 25 m². Repetition – four times. Placement of options is sequential.

The technology of growing sunflower seeds is generally accepted for the forest-steppe zone. Its predecessor was winter wheat. After harvesting the predecessor, stubble ploughing was carried out. In the first-second ten days of October, according to the experiment scheme, 50% of the annual norm of nitrogen fertilisers (ammonium nitrate, 33.4%), 100% of phosphorous and potash fertilisers (double superphosphate, 46%; potassium chloride, 62%) were applied and winter ploughing was

carried out to a depth of 25–27 cm. In spring, when the soil was physically ripe, mulching was carried out using a toothed harrow. On the day of sowing, according to the experiment scheme, 50% nitrogen fertilisers and 100% sulphur (ammonium sulphate) were applied and pre-sowing treatment was carried out with a combined unit. Seeds were sown to a depth of 4–5 cm at a soil temperature of 10°C at a depth of 6–8 cm (second 10 days of April). In the post-sowing period (pre-emergence), a soil herbicide (312.5 g/l S – metolachlor + 187.5 g/l Terbutylazine) was applied. The fungicide (80 g/l Ciproconazole + 200 g/l Azoxystrobin) was applied when the first signs of the disease appeared, and the second application – if necessary with an interval of 18–22 days. All elements of the cultivation technology, with the exception of the subjects, were the same throughout the experiment.

Sampling of cotyledons in each variant was carried out in the full ripeness phase on 10 plants. The results obtained for each of the indicators were tested for normality according to the Shapiro-Wilk criterion (Razali & Wah, 2011). In the case when the variational series did not correspond to the normal distribution, the number of samples was increased. The seed yield was determined by collecting all seeds from the registered plot and converting the value in t/ha to the basic seed moisture index of 8%.

A biochemical research method was used to determine the content of chlorogenic acid. The qualitative and quantitative composition of phenolic compounds was studied by thin-layer chromatography on sunflower seedlings grown in the laboratory and the husk of sunflower seeds grown in the field. Extraction of phenolic compounds from plant material depends on the type and polarity of the solvent. Ethanol was chosen as the solvent for the isolation of chlorogenic acid. Identification was performed by thin-layer chromatography (TLC) using plates measuring 100×150 mm, sorbent – Silicagel 60 F254 (Merck). Solvent systems were used to identify chlorogenic acid: chloroform – glacial acetic acid – methanol – water (60 : 32 : 12 : 8). To enhance the glow of substances under ultraviolet illumination, UV (365 Nm) plates with chromatographs were treated with 5% EtOH solution AlCl₃ with subsequent heating (5 min at 105°C). The chromatogram was kept for 7–10 minutes at a temperature of 110°C to the appearance of characteristic spots. Photo documentation of the materials was carried out using the AxioVision 40V Carl Zeiss software suite. Digital data processing was performed in the Image Pro Premier 9.0 (Trial version) software suite. Chromatography analysis was performed in the SORBFLIL TLC (Kovalov *et al.*, 2003). To determine the chemical nature of substances, chromatograms were treated with developing reagents. R_f indicators were determined photodensitometrically using *Sorbfil TLC* software suite. The presence of coumarins in methanol (MeOH) extracts was detected by lactone breakdown and qualitative reaction to diazotated sulfonylic acid (Kovalov *et al.*, 2003). The statistical

research method was used to analyse experimental data, determine the reliability of data, and identify dependencies between the studied indicators. Statistical data processing, principal component analysis (PCA), correlation and cluster analysis were performed in the XLSTAT software suite (Addinsoft Inc., USA, 2010).

RESULTS AND DISCUSSION

The qualitative composition of the phenolic complex and the number of individual compounds has a pronounced variety specificity, which is confirmed by the results of the conducted studies.

Based on thin-layer chromatography in methanol leaf extracts by the nature of the glow in the UV region (365 Nm), performed by a colour reaction to a FeCl₃ solution and the R_f value, a substance that can be identified as chlorogenic acid was discovered. On the chromatogram, this substance has an R_f of ~0.60 (for sunflower husks) and an R_f of ~0.74 (for cotyledonous leaves) (Fig. 1). Thus, it is established that secondary metabolites, including oxybenzoic acids, accumulate in the vegetative organs of plants, which play an important role in the formation of their constitutional stability. The general state of the plant organism and its survival strategy depend on the activity of the synthesis of phenolic substances.

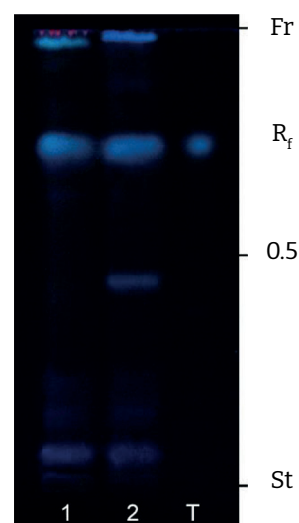


Figure 1. Chromatography of MeOH extracts: 1 – cotyledonous leaves; 2 – husks of Auris sunflower hybrid; T – chlorogenic acid (system: chloroform – acetic acid – methanol – water (60 : 32 : 12 : 8))

The effect of phenolic compounds on plant resistance against negative environmental factors is confirmed by the findings of other researchers (Levytskyi *et al.*, 2010; Gai *et al.*, 2020). Phenolic compounds are crucial for the antioxidant potential of sunflower seeds (Karamac *et al.*, 2012). The possibility of using phenolic compounds obtained from extracts of the aboveground part of sunflower as antioxidants is evidenced by scientific studies by Gai *et al.* (2020).

Given that the content of chlorogenic acid is quite high not only in cotyledonous leaves, but also in pericarpia, it is justified to consider them as a source of a valuable chemical compound (Table 1). Chlorogenic acid was detected by

thin-layer chromatography in 40% of water-alcohol extraction of cotyledonous leaves and husks, as evidenced by the area of peaks in the fluorescence intensity of individual compounds according to photodensitometry results.

Table 1. Area of fluorescence intensity peaks of individual compounds based on photodensitometry results

Rf Indicators	Peak area, S · 10 ³ (c.u.)									
	0.02	0.07	0.16	0.22	0.45	0.64	0.74	0.88	0.92	0.97
Cotyledonous leaves	27.5	128.4	60.4	56.1	0	35.3	367.5	0	18.8	224.1
Husk	0	133.4	57.4	39.9	111.6	0	370.2	36.2	0	201.7
Chlorogenic acid							104.7			

Obtaining extracts with high antioxidant activity from sunflower seed husks is confirmed by Szydłowska-Czeraniak *et al.* (2011).

Chlorogenic acid is highly soluble in ethanol and can therefore be extracted from the husk, where its content for the Auris hybrid ranges from 10 to 25 mg/g. The content of chlorogenic acid in sunflower husks varied depending on the zonal placement of seeds in the basket, which is a phenomenon of its matrix diversity. The content of chlorogenic acid in the seed husk, which is located in the centre of the flower (within a radius of 50.0 mm), is almost twice as high as in the seeds of its peripheral zone. This is conditioned by the different intensity of its development during certain periods (from flowering, fertilisation of flowers to seed formation) – in the flower, seeds are most intensively formed from the periphery to the

centre. Thus, in the peripheral zone, the seed has a larger weight of 1,000 seeds than in the central zone, which even with the same absolute content of chlorogenic acid, in terms of fractions per unit mass, shows its lower content. This allows considering the possibility of recycling waste that remains during the seed cleaning process.

According to the densitogram, the average content of chlorogenic acid in cotyledon leaf and husk extracts almost did not differ (Fig. 2). The use of ethanol as a solvent does not allow for complete extraction of the phenolic compound, but this method is the most optimal in terms of price/quality. In addition, unlike other solvents, it does not exclude the further use of raw materials, and when used as a substrate for fungi, it improves the effectiveness of sterilisation against pathogenic microflora and spores of competing fungi.

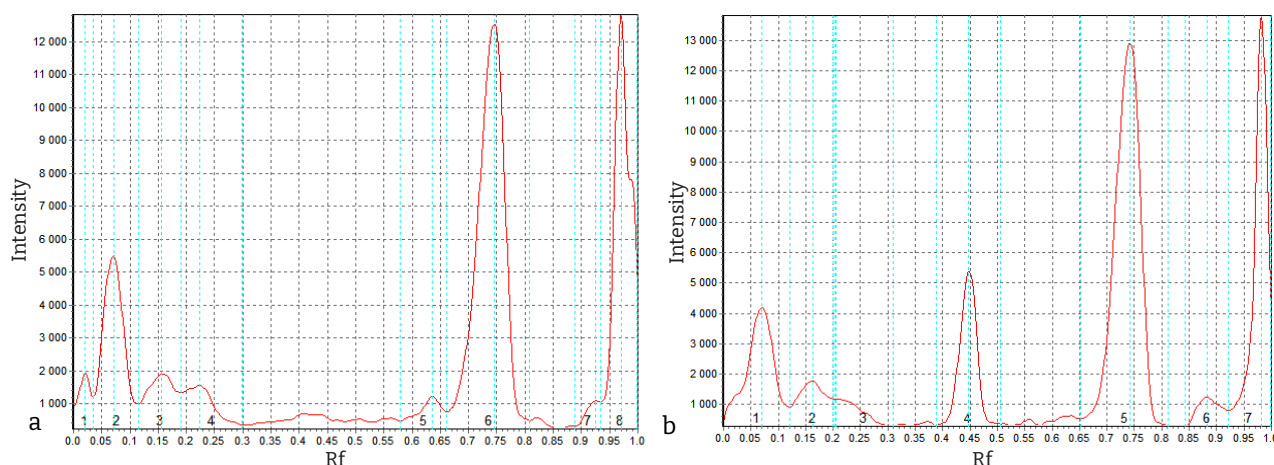


Figure 2. Photodensitogram of MeOH extracts: a – cotyledonous leaves; b – husks of the Auris sunflower hybrid

According to the results of studies, extraction of chlorogenic acid with 40% ethanol provided up to 10 mg/g of chlorogenic acid. Moreover, in addition to chlorogenic acid, a small amount of caffeic acid was found in the alcohol extract (Fig. 3), as well as minor traces of cinnamic, coumaric, ferulic, and sinapic acids.

The confirmed biological activity suggests that the green parts of sunflower plants can be considered a source of natural antioxidants.

Similar results were obtained by Pedrosa *et al.* (2000), where in addition to chlorogenic acid, up to 10 components were identified in the sunflower phenolic complex, including caffeic, cinnamic, coumaric, ferulic, sinapic, traces of vinyl and oxybenzoic acids.

The use of aqueous ethanol as a solvent is confirmed by the results obtained by Kovalov *et al.*, 2008, Labeiko, 2021. However, the use of ethanol as a solvent at concentrations of 40% compared to 50% and 70% in studies by

Labeiko et al. (2019) and Náthia-Neves & Alonso (2021) provides a high yield of this compound and reduces the cost of obtaining it.

Field and laboratory studies have shown that the content of chlorogenic acid varies depending on

environmental factors, varietal characteristics, and cultivation technology. All modern sunflower hybrids are characterised by a high fat content, and therefore, a higher content of chlorogenic acid compared to wild forms, which is confirmed by studies by Pomenta & Burns (1971).

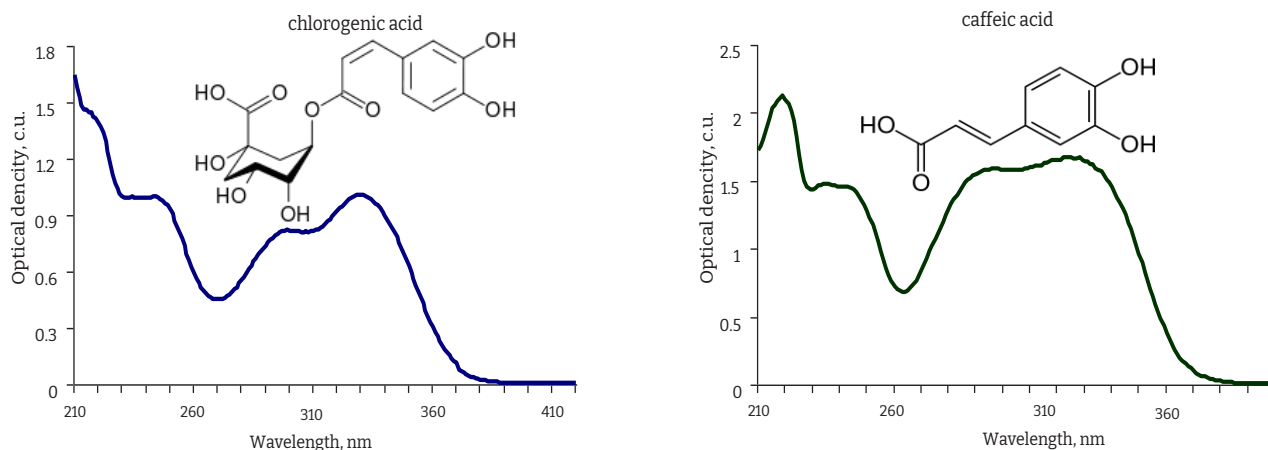


Figure 2. Electronic spectra of chlorogenic and caffeic acids extracted with 40% ethyl alcohol from the Auris sunflower husk

The application of mineral fertilisers has a significant effect on the synthesis of chlorogenic acid: a balanced mineral diet increases its content (Table 2). With an insufficient amount of phosphorus and potassium against the background of high nitrogen application, there is a significant decrease in its content in the husk. Foliar application of growth stimulants in combination with trace elements in chelated form leads to an increase in the content of phenolic compounds in sunflower leaves by 2.3-2.5 times compared to the control variant, which has a positive effect on the accumulation of chlorogenic acid in the achenes. Microelements are an integral part of enzymatic systems, which are biological catalysts that increase the activity of biochemical processes in plants. Due to the synergy of introduced growth regulators and trace elements,

plants are able to use nutrients from the soil more efficiently, and plant resistance to adverse factors increases.

The technology of growing sunflower seeds has a direct impact on the estimated gross yield of chlorogenic acid per unit area: the application of mineral fertilisers significantly affected the gross yield of this phenolic compound.

The highest content and the gross yield of chlorogenic acid was obtained on variants with full mineral fertiliser. Unbalanced mineral nutrition and unilateral application of nitrogen fertilisers against the background of phosphorus and potassium deficiency leads to a decrease in crop yield and a decrease in the antioxidant content in sunflower husks, and as a result – a smaller estimated yield of chlorogenic acid.

Table 2. Crop productivity and estimated gross yield of chlorogenic acid for sunflower cultivation (average for 2017-2018)

Fertiliser options	Yield, t/ha	Huskiness, %	Estimated gross yield of chlorogenic acid, kg/ha
$N_{60}P_{50}K_{90}$ (control)	3.19	21.9	6.71
$N_{60}P_{50}K_{90}+S_{30}$	3.46	21.6	7.47
$N_{90}P_{75}K_{135}+S_{30}$	3.80	22.1	8.31
$N_{120}P_{100}K_{180}+S_{30}$	3.85	22.8	8.60
LSD ₀₅	0.26	1.1	

It was found that the total gross yield of chlorogenic acid depends on the productivity of sunflower seeds. The highest biological yield of the studied hybrid is formed by applying $N_{90}P_{75}K_{135}+S_{30}$ – 3.8 t/ha. A further increase in the rate of application of mineral fertilisers does not lead to a significant increase in crop yield. The

huskiness index of cotyledons, depending on the background of mineral nutrition, is 21.6-22.8 %. Thus, the estimated gross yield of chlorogenic acid for growing Auris sunflower hybrid with the application of mineral fertilisers at a rate $N_{90}P_{75}K_{135}+S_{30}$ is 8.31 kg/ha, $N_{120}P_{100}K_{180}+S_{30}$ – 8.60 kg/ha.

Chlorogenic acid has a stronger antioxidant effect than ascorbic acid, caffeic acid, or vitamin E. The prospects for chlorogenic acid eradication in medicine are confirmed by Liang Wang *et al.* (2022); Cao *et al.* (2020). Preliminary extraction of chlorogenic acid from sunflower processing products expands the possibilities of using plant proteins, which is confirmed by the results of research by M. Labeiko *et al.* (2019); M. Labeiko & Z. Fediakina (2017); W. Jia *et al.* (2022). Even crude chlorogenic acid is of considerable commercial interest, and phenolic compounds extracted from the husk do not reduce, but even slightly improve its properties, which can be used in production: for example, adding to the substrate for growing oyster mushrooms and other edible mushrooms, in medicine, food industry.

CONCLUSIONS

As a result of the analysis of the current state of research on the identification, content, and influence of phenolic compounds, it was established that the qualitative composition of the phenolic complex and the number of individual compounds has a pronounced variety specificity. Chlorogenic acid was detected in water-alcohol (40%) extraction of cotyledon leaves and husks by thin-layer chromatography. Chlorogenic acid can be extracted from the sunflower husk by 40% ethanol, where its content in plants of the Auris hybrid is 10-25 mg/g.

Seeds placed in the centre of the basket within a 50.0 mm radius contain almost twice as much chlorogenic acid as seeds placed closer to the periphery. The application of mineral fertilisers has a significant effect on the synthesis of chlorogenic acid: a balanced mineral diet increases its content.

The total gross yield of chlorogenic acid depends on sunflower productivity and seed huskiness. The highest biological yield of the studied Auris hybrid is formed by applying $N_{90}P_{75}K_{135}+S_{30} - 3.80$ t/ha.

Extraction of 40% ethanol provides up to 10 mg/g of chlorogenic acid. Estimated gross yield of chlorogenic acid for growing Auris sunflower hybrid with the application of mineral fertilisers at a rate $N_{90}P_{75}K_{135}+S_{30}$ is 8.31 kg/ha, $N_{120}P_{100}K_{180}+S_{30} - 8.60$ kg/ha.

It can be argued that even crude chlorogenic acid is of considerable commercial interest, and phenolic compounds extracted from the husk do not reduce, but even improve its properties.

However, it is necessary to further investigate the dynamics of chlorogenic acid content in different periods of growth and development of sunflower plants and its effect on plant resistance against biotic and abiotic factors. To increase the yield of chlorogenic acid from the products under study, it is necessary to investigate the use of various solvents. Ultimately, the use of ethanol as a solvent does not allow for the complete extraction of the phenolic compound.

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Ідентифікація хлорогенової кислоти в сім'ядольних листках і лушпинні соняшника однорічного (*Helianthus annuus* L.)

Анотація. Актуальність досліджень зумовлена необхідністю оптимізації продукційного процесу агроценозів соняшника за рахунок підвищення екологічної стійкості. Серед побічних продуктів переробки, використання яких дотепер не є ефективним, виділення потужного антиоксиданта хлорогенової кислоти вивчено недостатньо. У зв'язку з цим дана стаття спрямована на розкриття закономірностей накопичення та ідентифікацію фенольних сполук в сім'ядольних листках і лушпинні соняшника. Мета досліджень – ідентифікація фенольних сполук в сім'ядольних листках і лушпинні різних генотипів соняшнику однорічного (*Helianthus annuus* L.) методом тонкошарової хроматографії. У процесі наукових пошуків за цією проблемою застосовували польові, біохімічні та статистичні методи досліджень. Для ідентифікації хлорогенової кислоти використовували систему розчинників: хлороформ – льодяна оцтова кислота – метанол – вода (60 : 32 : 12 : 8). Аналіз хроматографії проводили в програмі Sorbfil TLC. Об'єкт досліджень – гібрид соняшнику однорічного Ауріс. Встановлено, що якісний склад фенольного комплексу і кількість індивідуальних сполук має виражену сортоспецифічність. На хроматограмі дана речовина має $R_f \sim 0,60$ (для лушпиння соняшника) та $R_f \sim 0,74$ (для сім'ядольних листків). Встановлено, що найбільш ефективним розчинником для хлорогенової кислоти є водний етанол. Екстракція хлорогенової кислоти 40 % етанолом дозволяє отримати до 10 мг/г хлорогенової кислоти. Окрім хлорогенової кислоти у спиртовому екстракті у незначній кількості виявлено кавову кислоту. Матеріали статті становлять практичну цінність для вдосконалення процесів, пов'язаних із формуванням продуктивності посівів соняшника, а також раціональному використанню побічних продуктів переробки

Ключові слова: олійні культури, феноли, антиоксиданти, кавава кислота, хроматограма, фотоденситограма

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The level of intensity of soft winter wheat varieties infection by *Fusarium* link pathogens and their identification on grain

Abstract. In modern conditions, an important task of agricultural production is to increase the yield and improve the quality of agricultural products. A reliable and environmentally beneficial factor for a sustainable increase in yield and improvement of grain quality is the creation of new varieties with group resistance to diseases and a high level of adaptability. The effectiveness of breeding for immunity directly depends on the diversity of donor resistance genes, and the search for new sources of resistance has been and remains a topical issue. The purpose of the study was to establish the level of infection of grain of soft winter wheat varieties with pathogens of *Fusarium* head blight and their identification. Winter wheat varieties from different breeding institutions of Ukraine were used in the research: V.M. Remeslo Myronivka Institute of Wheat of NAAS (MIW), Breeding and Genetic Institute – National Centre for Seed Breeding and Variety Research of NAAS (SGI-NCNS), V.Ya. Yuriev Plant Production Institute of NAAS (PPI), Institute of Plant Physiology and Genetics of NASU (IPPG), Institute of Plant Protection of NAAS (IPP), Institute of Irrigated Agriculture of NAAS (IA), National Research Centre “Institute of Agriculture” of NAAS (NRC IA). Phytopathological analysis was used to determine the species composition of the *Fusarium* Link pathogen, and the proportion of species was determined by the ratio of infected wheat grain samples to their total number. A higher (15-18%) level of intensity of *Fusarium* blight infection was observed in the varieties Doskonala, Turunchuk, Ovidii, Vodohrai,

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and Myroliubyva. Winter wheat grain in the years of research was colonised by such types of *Fusarium* head blight as: *F. sporotrichiella*, *F. monilifopme*, *F. graminearum*, *F. culmorum*, *F. avenaceum*, *F. gibbosum* *F. sambucinum* and *F. oxysporium*. *F. monilifopme* and *F. oxysporium* species dominated the situation, with their prevalence on wheat grain being 16.2 and 1.7%, respectively. Winter wheat varieties with the lowest (1.2–4.5%) damage by pathogens of the genus *Fusarium* are recommended for use in wheat breeding for immunity. The use of a stable starting material in practical breeding will help contain the increase in infection of pathogens without the use of chemical protective equipment

Keywords: *Triticum aestivum* L., source material, seed infection, *Fusarium* head blight, fungi of the genus *Fusarium*, species composition, dominant species

INTRODUCTION

Wheat is grown in more than 80 countries around the world and is the main food product in 43 of them (Bakumenko *et al.*, 2019). It is one of the most productive and valuable food crops, (Shpaar *et al.*, 2012; Leonov *et al.*, 2016) are convinced that in the future the importance of wheat will constantly grow and this particular crop will become the most important on the globe. Pathogenic microorganisms accompany soft winter wheat from the moment of sowing to harvesting and storage. Even minimal damage to its diseases leads to large overall crop losses (Kirichenko, 2016). Diseases significantly reduce the yield and quality indicators of grown products. Losses of the gross grain harvest annually amount to about 25–30 % (Savary *et al.*, 2019). As evidenced by M. Kayim *et al.* (2022) “almost 22% of wheat crop losses were caused by diseases. These percentages will increase over time due to mutations and diversity of virulent strains.” Success in addressing these issues mainly depends on the effectiveness of genetic improvement of wheat varieties (Mukha & Murashko, 2019). Selection of soft winter wheat has reached a level where its potential yield is largely related to the stability of the varieties grown. Selection for wheat resistance against phytopathogens allows reducing their harmful effect. Creating and growing disease-resistant varieties allows protecting the crop, preserving the environment, and saving money, so one of the most effective measures is to use resistant varieties (Deutsch *et al.*, 2018).

The effectiveness of immunity breeding depends on the availability of a well-studied source material and a scientifically based approach to its use. The main components of the methodology for creating such material are regular monitoring of the pathogenic complex, morphophysiological properties of plants and populations of particularly dangerous pathogens, objectivity in assessing the immunological properties of varieties, establishing relationships in the plant-host-pathogen system, identifying and selecting highly effective sources and donors adapted to zonal conditions (Bushulian, 2018). In Ukraine, work on the creation of raw materials for wheat breeding with group resistance against different species of powdery mildew, rust, septoria, *Fusarium* blight, stinking smut, and cercospora leaf spot is successfully conducted at the Institute of Plant Protection of NAAS, V.M. Remeslo Myronivka Institute of Wheat (MIW) and Breeding and Genetic Institute –

National Centre for Seed Breeding and Variety Research of NAAS (SGI-NCNS) *etc.* (Kovalyshina *et al.*, 2018).

Grain crops can be affected by more than 300–350 species of various organisms. These are insects, fungi, bacteria, viruses, nematodes, rodents, weeds, etc. But the main threat to crop loss, especially in the initial stages of development, in the autumn–winter period is represented by pathogens of fungal infections, among which one of the leading places is occupied by micromycetes of the genus *Fusarium* (Ryabovol *et al.*, 2019). The widespread distribution of *Fusarium* fungi, their variability, and indisputable evidence of the danger posed to human and animal health by mycotoxins, generate the interest of the scientific community (Ostrovsky *et al.*, 2017; Janik *et al.*, 2020). Plant damage involves a complex of fungi of this genus, which differ in biology and adaptation to the conditions of the biocenosis in different areas of grain cultivation (Nazarenko & Lykholat, 2018; Birr *et al.*, 2020). They cause damage at all stages of organogenesis of winter wheat plants, reduce the energy of germination and field germination of seeds, and worsen the baking properties of flour. The danger of *Fusarium* blight infection lies not only in a decrease in the grain yield, but also in contamination of grain with *fusarium* toxins, which leads to a weakening of plants during their further development, it can only be detected by mycological analysis (Topi *et al.*, 2020).

The purpose of the study was to determine the level of infection of grain of soft winter wheat varieties with pathogens of *Fusarium* head blight and identify them. According to Mostoviyak *et al.*, (2020) questions of morphology, biology, biochemistry, physiology, and genetics of fungi of the genus *Fusarium*, and the search for ways to limit their number in the agrobiocenoses of field crops and reduce the number today is of considerable interest to both Ukrainian and foreign researchers. A number of studies have been devoted to the issues of wheat resistance to *Fusarium* head blight depending on the sensitivity of the variety and meteorological factors, grain quality, and mycotoxin contamination. Góral *et al.* (2018); Zhu *et al.* (2019); Birr *et al.* (2020); Martínez *et al.* Karellov *et al.* (2021) recommend the use of a polymerase chain reaction test (PCR test) for more accurate quantitative and qualitative genus and species-specific identification of *Fusarium* on durum wheat. Bahadur (2022) found that new populations of *Fusarium* will continue to

appear due to microevolution and the invention of exotic pathogens. The researcher recommends the use of resistant varieties and biological control agents for disease control (*Trichoderma* spp. and *Pseudomonas* spp.). D. Naguib (2018) investigated the effect of treatment of wheat grains with defensin (low-molecular-weight positively charged proteins with a high cysteine content) on the resistance of cultivated plants against *Fusarium* wilt as one of the methods of biological protection of crops. It was found that priming improves the antioxidant system of plants, initiates the regulation of protective signalling molecules, such as jasmonic and abscisic acids, which, in turn, activate the expression of several protective genes for a number of diseases, including *Fusarium* blight. Another biosecurity method is proposed by L. Shcherbakova et al. (2018), who found that the extract purified from the mycelium of the strain *Fusarium sambucinum* controlling tomato wilt can protect wheat from *Fusarium* blight and common root rot.

Scientists are very interested in identifying and studying *Fusarium* blight pathogens in other field crops. In particular, research by B. Kalman et al. (2020) conducted in north-eastern Israel for the first time identified and characterised species of *Fusarium* spp., pathogens of basal rot of onions: *F. proliferatum*, *F. oxysporum* F. sp. *cepae* and two species that are less well known as pathogens of this disease, *F. Acutatum* and *F. anthophilium*. The research was a necessary step towards identifying the mycoflora of diseased onion plants and developing a preventive programme that would reduce the harm from the disease. Due to insufficient information on the molecular epidemiology of *Fusarium* wilt of the important yams root crop in China (*Dioscorea polystachya* Thunb.) a number of studies by a group of Chinese researchers F. Dongzhen et al. (2020) were aimed at identifying species and strains of fungi of the genus *Fusarium* and establishing links between pathogen strains and populations. The attention of researchers to the important scientific problem of *Fusarium* head blight infection and the search for ways to protect determines the relevance of the conducted research, which consisted in studying *Fusarium* blight on an artificial infectious background and identifying sources of resistance among winter wheat varieties.

MATERIALS AND METHODS

The research was conducted during 2021-2022 at the winter wheat breeding laboratory of the V.M. Remeslo Myronivka Institute of Wheat of NAAS of Ukraine (Tsentralne village, Obukhiv district, Kyiv oblast) in the northern part of the Right-Bank Forest-Steppe. The object of research was grain of soft winter wheat varieties grown in the field on an artificial infectious background of *Fusarium* head blight harvest 2021-2022 from different breeding institutions of Ukraine: V.M. Remeslo Myronivka Institute of Wheat of NAAS (MIW), Breeding and Genetic Institute – National Centre for Seed Breeding and Variety Research of NAAS (SGI-NCNS), V.Ya. Yuriev Plant

Production Institute of NAAS (PPI), Institute of Plant Physiology and Genetics of NASU (IPPG), Institute of Plant Protection of NAAS (IPP), Institute of Irrigated Agriculture of NAAS (IA), National Research Centre “Institute of Agriculture” of NAAS (NRC IA). Distribution of grain *Fusarium* blight pathogens and species composition of fungi of the genus *Fusarium* Link was studied on 23 varieties of winter wheat from different breeding centres of Ukraine (Doskonala, Pryvablyva, Stolychna, Fermerka, Mriya, Povernia (PPI), Turunchuk, Zvytiha, Kosovytsia, Viktoria, Bezmezhna (SGI-NCNS), Vodohrai, Myroliubyva, Polisiaanka (NRC IA), Voloshkova (MIW, IPPG), Chornyava, Tsyhanka (IPPG), Blaho, Ovidii (IA)), and sources of stability – Myronivska early ripening/CATALON, MV 20-88/Smuhlianka, BILINMEVEN 49/Natalka, Donskoyprosotor/Slavna, (Mikon/ALMA)/Legenda Myronivska (MIW).

To identify species of *Fusarium* Link in laboratory conditions, phytopathological analysis of grain of soft winter wheat varieties was carried out in accordance with the methods of conducting phytopathological studies and the methodology for assessing the resistance of wheat varieties against pests and pathogens (Volkodav et al., 2001; Tribel et al., 2010; Methods..., 2016). Seeds (100 pcs.) each of the 23 studied varieties of winter wheat was disinfected for 5 minutes in a 0.5% solution of KMnO₄, then it was washed with sterile water and dried between sheets of filter paper, flamed in the flame of an alcohol lamp and laid out in 10 pieces on a sterile nutrient medium (potato-glucose agar (PGA) with the addition of an antibiotic of antibacterial action – streptomycin sulphate). Cups of grain were incubated in a thermostat at 25°C for five days. After this period, each micromycete isolated from the seed to the surface of the nutrient medium was transplanted into a separate tube with PGA for further identification of the pathogen type.

To determine the species composition of fungal pathogens (seed infection) on the grain of the 2021-2022 harvest, phytopathological analysis of grain samples was carried out in the laboratory conditions of the institute. Isolates were examined at Day 5-7, recording the presence or absence of microconidia. The final identification of pathogens was carried out by microscopic examination, considering morphological features, the presence or absence of chlamydospores of the pathogen (Volkodav et al., 2001; Tribel et al., 2010; Methods..., 2016). The species name of the fungi was determined according to Bilai et al. (1988). The frequency of the species was determined by the ratio of grain samples where it was found to the total number of samples studied and expressed as a percentage.

RESULTS AND DISCUSSION

The average air temperature in the period August 2021 – July 2022 was 9.3°C, which is 0.4°C more than the long-term average (Table 1). In August and November 2021, the average monthly air temperatures exceeded the long-term average by 0.1 and 2.4°C, respectively, and in September and October they were

lower by 1.7 and 1.1°C. In the spring-summer growing season of wheat, the average monthly temperatures

were 0.1-1.5°C lower than long-term ones, only in June they were 1.4°C higher.

Table 1. Meteorological conditions during the years of research, Myronivka

Year	Month	Air temperature, °C			Amount of precipitation, mm			
		actual	multi-year	±*	actual	multi-year	±*	%*
2021	August	20.5	20.4	0.1	109	55	54	198
	September	13.2	14.9	-1.7	27	58	-31	46
	October	7.6	8.7	-1.1	26	43	-17	60
	November	4.8	2.4	2.4	41	40	1	101
	December	-1.1	-1.8	0.7	94	42	52	223
2022	January	-1.2	-3.7	2.5	33	39	-6	86
	February	1.7	-2.5	4.2	10	33	-23	32
	March	2.3	2.4	-0.1	13	39	-26	32
	April	8.3	9.8	-1.5	143	41	102	349
	May	14.7	15.7	-1.0	42	61	-19	70
	June	20.7	19.3	1.4	58	87	-30	66
	July	20.4	21.1	-0.7	68	75	-7	91
Per year		9.3	8.9	0.4	663	615	48	108

Source: observation logs of the Myronivka meteorological station

Note: * ± – difference to multi-year; % – percentage to multi-year

From August 2021 to July 2022, 663 mm of precipitation fell (108% of the long-term average). Precipitation in August (109 mm, 198% of the long-term amount) contributed to the production of uniform wheat shoots. In the spring-summer growing season of winter wheat, a sufficient amount of moisture was observed, although the amount of precipitation was 7-30 mm lower than the long-term one, only in April 349% of the average long-term amount fell. The duration of the period from germination to the end of vegetation in 2021 was at the level

of long-term (59 days), the dormant period was shorter by 8 days. The level of hydrothermal coefficient (HTC) and its dynamics in the spring-summer growing season are shown in Figure 1. According to the indicator of water supply, the reporting year refers to a year with a weak drought (HTC=0.9). In the spring-summer growing season of winter wheat, insufficient moisture was observed (HTC was 0.8). The average duration of winter wheat development periods and the distribution of precipitation depending on these periods are shown in Table 2.

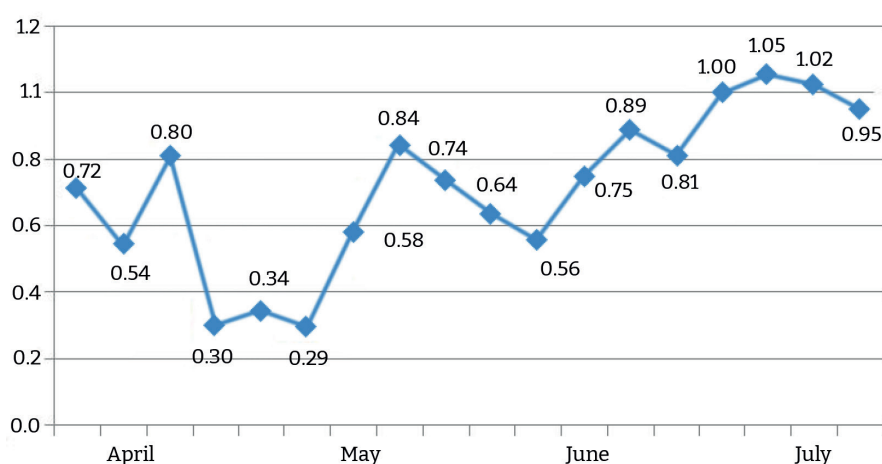


Figure 1. Dynamics of the hydrothermal coefficient in the spring-summer vegetation period of winter wheat, 2022

Table 2. Hydrothermal regime of winter wheat development by period (2020-2021)

Year termination of vegetation		Periods of winter wheat development							Amount		Number of days from sowing to	
		beginning of the dormant period – restoration of vegetation	restoration of vegetation – stem elongation	stem elongation – earing	earring – milk ripeness	milk ripeness – waxy ripeness	waxy ripeness – threshing			waxy ripeness	threshing	
		Precipitation, mm										
2021/22		53.2	161.7	148.5	36.3	3.0	55.1	67.4	525.2		–	–
multi-year		124.4	245.8	60.6	62.4	77.1	32.5	73.2	675.9		–	–
		Sum of active temperatures of 5 °C and more										
2021/22		346.3	64.3	511.4	206.8	361.2	286.2	503.8	2,280.0		–	–
multi-year		479.1	77.2	298.1	399.8	484.3	312.5	342.9	2,359.9		–	–
		Average daily air temperature, °C										
2021/22		6.6	-0.01	9.9	14.8	20.1	20.4	20.2	–		–	–
multi-year		8.9	-1.9	9.1	15.4	19.3	21.0	20.9	–		–	–
		Duration of individual periods, days										
2021/22		58	118	54	14	18	15	24	–		276	301
multi-year		59	126	37	27	25	15	17	–		290	307

Source: observation logs of the Myronivka meteorological station

As a result of the conducted studies, it was found that the average intensity of winter wheat *Fusarium* blight infection was 9.3% (Table 3). This indicator in the studied varieties differed significantly, and according to the level of damage intensity, all the studied varieties can be divided into several groups. Lowest percentage of damage (1.20-4.5 %) was noted in the varieties Bezmezhna (SGI-NCNS), Poverna (PPI), Polisianka (NRC-IA), the second group included varieties – Zvytiha, Kosovytsia (SGI-NCNS), Pryvablyva, Stolychna (PPI), Tsyhanka (IPPG), Voloshkova (MIP, IPPG) (8.1-11.5%), the third – Chorniava (IPPG), Mriya (PPI), Viktoria (SGI-NCNS), Blaho (IA), Fermerka (PPI) (5.1-7.5%).

According to the resistance to this pathogen on an artificial infectious background in the field, MIW identified in previous years (2016-2020): ((MV – 20-88/Smuglyanka), BILINMEVEN-49/Natalka, Donskoy prostor/Slavna, Mikon/ALMA)/Legenda Myronivska, Myronivska early-maturing/Catalon), they are involved in direct and reverse crosses as parent forms when creating new hybrid combinations for resistance against *Fusarium graminearum*. The highest intensity of damage was observed in wheat varieties Natula (POL), Doskonala (PPI), Turunchuk, Ovidii (IA), Vodohrai, Myroliubyva (NRC IA) (15.1-25.1 %).

Table 3. Characteristics of the best varieties of soft winter wheat in terms of resistance against pathogen *Fusarium graminearum*, (average for 2021-2022)

Variety, breeding line	Country of origin	Intensity of damage, %	Intensity of damage, %± to the vulnerable variety
Natula (vulnerable variety)	POL	25.1	–
Doskonala	PPI	15.2	+9.9
Turunchuk	SGI-NCNS	16.3	+8.8
Ovidii	IA	15.5	+9.3
Vodohrai	NRC IA	15.1	+10.0
Myroliubyva	NRC IA	15.3	+9.8
Zvytiha	SGI-NCNS	10.2	+14.9
Kosovytsia	SGI-NCNS	11.5	+13.6
Pryvablyva	PPI	8.1	+17.0
Tsyhanka	IPPG	6.6	+18.5

Table 3, Continued

Variety, breeding line	Country of origin	Intensity of damage, %	Intensity of damage, %± to the vulnerable variety
Voloshkova	MIW, IPPG	9.1	+16.0
Stolychna	PPI	8.3	+16.8
Chorniava	IPPG	7.5	+17.6
Mriya	PPI	5.2	+19.9
Viktoria	SGI-NCNS	6.4	+18.7
Blaho	IA	5.1	+20.0
Fermerka	PPI	5.3	+19.8
Bezmezhna	SGI-NCNS	4.5	+20.6
Povernna	PPI	3.3	+21.8
Polisianka	NRC IA	3.1	+22.0
Myronivska early ripening/CATALON	MIW	1.2	+23.9
MV 20-88/Smuhlianka	MIW	1.2	+23.9
BILINMEVEN49/Natalka	MIW	1.3	+23.8
Donskoy prostor/Slavna	MIW	1.3	+23.8
(Mikon / ALMA)/Legenda Myr	MIW	1.2	+23.9

Source: field journals of phenological observations of the winter wheat breeding laboratory of the V.M. Remeslo Myronivka Institute of Wheat of NAAS for 2021-2022

The species composition of fungi of the genus *Fusarium* Link on soft winter wheat grain is established (Fig. 2), which included the presence of eight species, the predominant of which were three: *sporotrichiella* (14.4%), *oxysporium* (11.3%) *culmorum* (8.1%).

The pathogen *F. sporotrichiella* was detected on the grains of 16 varieties of winter wheat, and the highest

damage was observed in varieties Turunchuk, Pryvablyva, Voloshkova, and Blaho (36.1-46.9%). *F. oxysporium* affected only 11 varieties, the most vulnerable – the Doskonala variety (50%). The third most common type of *Fusarium* infection was *F. culmorum* with a damage rate of 8.1%, in other species, the proportion of the intensity of wheat grain damage was noted in the range from 2.2 to 6.5% (Fig. 2).

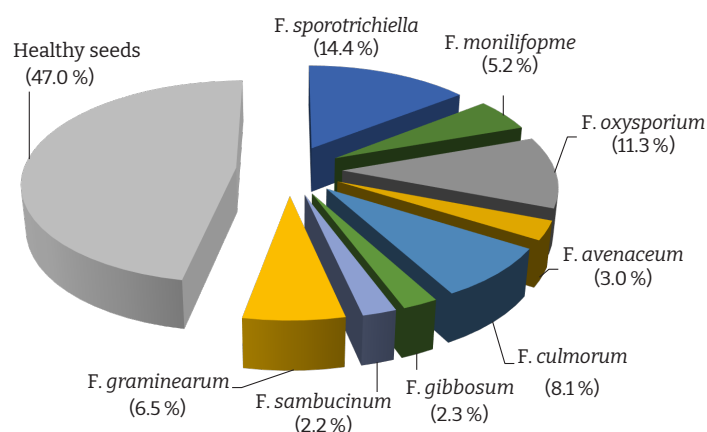


Figure 2. Species composition of fungi of the genus *Fusarium* Link on soft winter wheat grain (MIW, average for 2021–2022)

High resistance against the causative agent of *Fusarium* blight was observed in sources (lines) created in MIW – Myronivska early-maturing/CATALON, MV 20-88/Smuglyanka, BILINMEVEN49/Natalka, Donskoy prostor/Slavna and (Mikon/ALMA)/Legenda Myronivska,

the percentage of healthy grains reached 76.6%. The average level of damage intensity was identified as wheat varieties Kosovytsia, Stolychna, Mriya, and Bezmezhna, the percentage of healthy grains was within 55.5–65.6% (Table 4).

Table 4. Species composition of *Fusarium* blight infection on winter wheat grain, average for 2021-2022

Variety, breeding line	Proportion of fungi of the genus <i>Fusarium</i> on grain, %								Healthy grains, %
	<i>F. sporotrichiella</i>	<i>F. moniliforme</i>	<i>F. oxysporum</i>	<i>F. avenaceum</i>	<i>F. culmorum</i>	<i>F. gibbosum</i>	<i>F. sambucinum</i>	<i>F. graminearum</i>	
Natula (vulnerable variety)	18.0	–	25.1	–	25.0	–	–	10.2	21.7
Doskonala	10.1	14.9	50.1	–	–	–	–	9.1	15.8
Turunchuk	39.9	–	30.1	–	–	10.3	–	–	19.7
Kniahynia Olha	25.2	4.8	25.0	–	25.1	–	–	–	19.9
Vodohrai	–	–	29.3	–	22.3	–	15.4	–	33.0
Myroliubyva	18.5	20.5	–	10.2	10.8	5.1	–	–	34.9
Zvytiaha	12.1	9.9	–	–	15.9	–	–	10.1	52.0
Kosovytsia	–	8.5	–	–	8.1	3.9	6.1	7.8	65.6
Pryvablyva	38.1	–	6.9	–	10.1	10.3	–	–	34.6
Tsyhanka	17.2	–	36.1	17.5	–	–	–	–	29.2
Voloshkova	46.9	–	–	–	18.1	–	–	–	35.0
Stolychna	12.2	6.3	11.1	–	14.9	–	–	–	55.5
Chorniava	24.5	–	25.5	–	–	–	–	5.5	44.5
Mriya	18.2	12.1	–	10.2	–	–	–	–	59.5
Viktoria	–	–	10.4	5.6	–	15.0	–	22.3	46.7
Blaho	36.1	10.2	–	–	14.1	–	2.7	–	36.9
Fermerka	–	–	–	–	–	–	8.5	40.3	51.2
Bezmezhna	–	10.3	–	–	25.1	–	–	–	64.6
Poverna	32.1	–	15.5	–	–	8.3	–	–	44.1
Polisianka	–	15.1	–	14.5	–	–	–	20.3	50.1
Myronivska early ripening/CATALON	–	5.1	9.8	4.2	5.1	–	–	10.1	65.7
MV 20-88/Smuhlianka	9.8	8.5	–	–	–	–	10.1	–	71.6
BILINMEVEN49/Natalka	3.5	–	2.4	–	–	–	8.2	9.3	76.6
Donskoy prostor/Slavna	–	5.6	–	6.7	–	3.5	–	8.9	75.3
(Mikon / ALMA)/Legenda Myr	–	–	6.4	–	9.6	–	2.9	10.8	70.3
Isolation frequency, %	14.4	5.2	11.3	3.0	8.1	2.3	2.2	6.5	–

Source: field journals of phenological observations of the winter wheat breeding laboratory of the V.M. Remeslo Myronivka Institute of Wheat of NAAS for 2021-2022

Compared to the variety Natula (25.1%) susceptible to *Fusarium* infection, the proportion of healthy grains was low in the varieties Doskonala (15.8%), Turunchuk (19.7%), Ovidii (19.9%), and Tsyganka (29.3%), indicating a high level of *Fusarium* infection.

Climate changes caused by global warming lead to sharp changes in air temperature both in winter and summer, which cause long periods of drought in summer and sudden frosts during the spring growing season of field crops. Miedaner & Juroszek (2021), Juroszek & Tiedemann (2013) point out that global climate change threatens wheat productivity in the context of the significant impact of air temperature and humidity on the development of pathogens. These weather changes negatively affect not only cultivated plants, but also the development of pathogens (Jarroudi *et al.*, 2017; Jarroudi *et al.*, 2018). Scope of variation in the intensity of damage to varieties by *Fusarium* pathogens can be determined not only by the characteristics of

genotypes, but also by such non-random factors as the influence of environmental conditions. Meteorological conditions during the years of research were not very favourable for the development of the pathogen *Fusarium graminearum* Schwabe, the average intensity of winter wheat *Fusarium* infection was 9.3 %.

F. Doohan research *et al.* (2003) proved that the establishment of the species composition and the ratio of species is significantly influenced by weather conditions, in particular, the humidification regime. The main role here is played not by all precipitation during the season, but only by those that fell during the flowering period – grain maturation, they determine the level of development and spread of the disease. These findings are consistent with the results of studies conducted in the context of the regime of moisture supply during the flowering period – grain maturation on the species composition of fungi of the genus *Fusarium* Link. During the years of research, the amount of precipitation during

the flowering and ripening period of winter wheat grain was significantly lower than the long-term indicator.

N. Carlo *et al.* (2021) argue that in the field it is not always possible to create a levelled artificial infectious background due to the influence of environmental conditions, as a result – a decrease in the intensity of the manifestation of the disease, and the resulting intensity of the damage may not correspond to the actual assessment of the stability of an individual sample. The conducted studies indicate that the intensity of damage to soft winter wheat plants by the *Fusarium* pathogen was determined within 1-35%, the average development of the disease was 15.8%. Hybridisation can significantly increase the availability of genotypic diversity in terms of resistance. M. Figueroa *et al.* (2018) consider it appropriate to create an artificial background of pathogens in the field and laboratory to differentiate the initial breeding material by the corresponding trait under controlled conditions. This, in turn, would contribute to the selection of stable forms, adjust the further areas of breeding work, and accelerate the creation of stable source material.

21 species of fungi of the genus *Fusarium* are described in the scientific literature, which are most often present in the pathogenic complex of Ukraine. According to S. Choni (2014), depending on weather conditions, grain crops are affected by 5-10 species, the most common and most dangerous among which are *F.graminearum* and *F.culmorum*. Frequency of isolation of pathogens typical of Ukrainian conditions, such as *F. graminearum* and *F. sulmorum*, gradually decreases, and other types of pathogens that can develop in dry weather conditions begin to dominate. The danger of these pathogens lies in the fact that it is quite difficult to visualise them at the stage of grain development in the conditions of each specific experiment, which is confirmed by the research of Mykhailska *et al.* (2019). Two types of fungus dominated the experiment, the first of which was *F. sporotrichiella*, the frequency of its dominance was in the range from 3.5 to 39.9 %, and the second-found species – *F. oxysporium* – the dominance of which was from 2.4 to 50.1 %. In addition to the immediate threat to seed material and its quality, pathogens *F. sporotrichiella*, which predominate in the pathogenic complex, produce trichothecin mycotoxins. These substances can cause acute poisoning in humans and animals, in addition, studies note

that in laboratory conditions, strains of *F. sporotrichiella* at a temperature of +26-28°C is able to synthesise and accumulate mycotoxins three times faster (Awuchi *et al.*, 2021). One of the most effective ways to prevent the spread of *Fusarium* head blight disease is to create and introduce wheat varieties resistant to *Fusarium* blight and proper agricultural techniques of crops into production.

CONCLUSIONS

Fusarium head blight is one of the most common and harmful diseases among winter wheat diseases, which is one of the leading diseases of winter wheat in terms of its prevalence and significance. At the same time, the climatic changes towards aridity that were observed on the territory of Ukraine in recent years do not contribute to the loss of a significant part of the crop from *Fusarium* blight, since only in wet and warm weather during the flowering period of winter wheat *Fusarium* wilt can develop so much that quantitative losses will be very significant. However, the result of the vital activity of *Fusarium* fungi is a whole set of mycotoxins that can significantly degrade the quality of grain, so one of the most effective ways to prevent the spread of *Fusarium* blight without the use of chemical means of protection is to use a stable starting material when creating new varieties.

The smallest damage (1.2-4.5%) by the *Fusarium* blight was characterised by winter wheat varieties Bezmezhna, Povernna, Polisiaanka and sources of resistance (lines) – Myronivska early-maturing/CATALON, MV 20-88/Smuglianka, BILINMEVEN 49/Natalka, Donskoy prostor/Slavna and (Mikon/ALMA)/Legenda Myronivska (1.2-4.5%), which are recommended for use in breeding for immunity. The highest isolation frequency was characterised by the type *F. sporotrichiella* (14.4%), second place – *F. oxysporium* (11.3%), third – *F. culmorum* (8.1%), and *F. graminearum* (6.5%) shared fourth place with *F. monilifopme* (5.2%). Partially observed species *F. sambucinum* (2.2%), *F. avenaceum* (3.0%), and *F. gibbosum* (2.3%).

Study of the prevalence and presence of the species composition of fungi of the genus *Fusarium* allows screening of wheat varieties and detection of samples less infected with *fusarium* infection. This is important for breeding programmes when choosing optimal wheat varieties for resistance against *Fusarium* blight pathogens.

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Рівень інтенсивності ураження сортів пшениці м'якої озимої збудниками *Fusarium* link та їх ідентифікація на зерні

Анотація. В умовах сьогодення важливим завданням аграрного виробництва є підвищення рівня врожайності та покращення якості продукції сільськогосподарських культур. Надійним та екологічно вигідним фактором стійкого збільшення врожайності та поліпшення якості зерна є створення нових сортів з груповою стійкістю проти хвороб та високим рівнем адаптивності. Ефективність селекції на імунітет прямо залежить від різноманіття складу генів стійкості донорів, топу пошук нових джерел стійкості було і залишається актуальним питанням. Мета досліджень полягала у встановленні рівня інфікованості зерна сортів пшениці м'якої озимої збудниками фузаріозу колоса та їх ідентифікації. В дослідженнях використовували сорти пшениці озимої різних селекційних установ України: Миронівського інституту пшениці імені В. М. Ремесла НААН (МІП), Селекційно-генетичного інституту – Національного центру насіннезнавства та сортовивчення НААН (СГ-НЦНС), Інституту рослинництва ім. В. Я. Юр'єва НААН (ІР), Інституту фізіології рослин і генетики НАНУ (ІФРГ), Інституту захисту рослин НААН (ІЗР), Інституту зрошуваного землеробства НААН (ІЗЗ), Національного наукового центру «Інститут землеробства» НААН (ННЦ ІЗ). Для визначення видового складу збудника *Fusarium* Link використовували фітопатологічний аналіз, частку видів – співвідношенням інфікованих зразків зерна пшениці до їх загальної кількості. Вищий (15-18 %) рівень інтенсивності ураження фузаріозною інфекцією спостерігали у сортів Досконала, Турунчук, Овідій, Водограй, Миролубива. Зерно пшениці озимої у роки досліджень колонізувалось такими видами фузаріозу як: *F. sporotrichiella*, *F. moniliforme*, *F. graminearum*, *F. culmorum*, *F. avenaceum*, *F. gibbosum*, *F. sambucinum* та *F. oxysporium*. Домінуючу ситуацію займали види *F. moniliforme* та *F. oxysporium*, їх поширеність на зерні пшениці становила 16,2 та 1,7 % відповідно. Виокремлені сорти пшениці озимої з найменшим (1,2-4,5 %) ураженням збудниками роду *Fusarium* рекомендовано для використання в селекції пшениці на імунітет. Використання в практичній селекції стійкого вихідного матеріалу дозволить утримувати наростання інфекції збудників хвороб без застосування хімічних засобів захисту.

Ключові слова: *Triticum aestivum* L., вихідний матеріал, насіннева інфекція, фузаріоз, гриби роду *Fusarium*, видовий склад, домінуючі види

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Optimal time of plant growth regulator application to Sorghum canopy according to BBCH and Kuperman crop growth scales

Abstract. The relevance of the study is determined by the problem of choosing the optimal scale for identifying plant growth phases to determine the best terms for conducting agrotechnical operations for the care of sorghum plants. The purpose of the experiment was to apply a plant growth regulator with higher efficiency during crop growth stages 21, 31, 37 (BBCH) and III, IV, VI-VII on the Kuperman scale. Field and laboratory research methods were used in the study. The experiment was conducted in the Forest-Steppe zone of Ukraine, two sorghum varieties of *Sorghum bicolor* and *Sorghum saccharatum* were treated with PGR. Foliar application of PGR (0.5 L/ha) in the BBCH stages 21 and 31 provides 2.8 and 4.9% better results than in stages III and IV of the Kuperman scale (the latter is based on the complex morphophysiological analysis to identify I to VII CGS). Foliar application of the PGR in microstage 21 (BBCH) promoted faster development and an increase in grain yield in sorghum varieties (0.19 t/ha in Odeskyi 205 and 0.12 t/ha in Lan 59) compared to application in stage III (Kuperman). A similar effect of the PGR applied in microstage 21 (BBCH) compared to stage III (Kuperman) was recorded for two sweet sorghum hybrids: in Dovista, an increase in the yield of biomass was 1.6t/ha, dry matter – 0.7t/ha, and sugar content in the stem juice – 0.0%, while these indices in Huliver amounted to 1.6t/ha, 0.7t/ha, and 0.2% respectively. Therefore, the BBCH scale is recommended to adopt for PGR application, and foliar application of PGR should be conducted in the 21 and 31. The practical value of the study

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lies in the selection of the scale of growth and development of sorghum crops and the terms of application of foliar fertilisation, optimal for the conditions of Ukraine. The study is useful from the practical standpoint of conducting foliar fertilisation of plants in production and as a theoretical assessment of plant growth and development scales for students and researchers

Keywords: foliar application of fertilisers, identification of plant development stage, organogenesis stages, the yield of grain, biomass yield

INTRODUCTION

Correct identification of phenological stages in common and sweet sorghum is important for optimising cultivation technology since many agronomic measures of tending crops, especially the foliar application of fertilisers, plant protection products, and plant growth regulators require strict identification of the plant physiological state in different growth stages (Maw et al., 2017; Chapke, 2019; Cuevas & Prom, 2020). The application of protective and stimulative products in inappropriate stages can harm plants and reduce crop yield. Other researchers state that besides proper management, the application of plant growth regulators (PGR) in the optimal stage of growth ensures maximum production (Sory et al., 2017; Thapa et al., 2017; Maiti & Singh, 2019). Similarly, other agronomic measures should be rationally applied based on the scale of growth and development of sorghum.

Several scales of growth and development for cereals have been developed over the years, and they were compared in earlier studies (Kalenska et al., 2018). Dutch agronomist Willem Feekes is the author of the Feekes scale, which was introduced in 1941 (Maity et al., 2021). This scale is used more often in the US, while other scales, such as the Zadoks or the BBCH, are less used. The Feekes scale is especially useful in stages 6 to 10.5 (stem elongation), which is relevant when it is necessary to decide on the best timing for fungicide application. The Keller and Baggiolini scale was developed in Switzerland in 1954. This scale is an extended version of the Feekes scale, with growth stages being coded with letters. In 1962, another scale was developed under the leadership of F.M. Kuperman. The scale describes 12 stages of plant growth and development into three periods: I-IV stages – embryonic period and youth (i), V-VIII stages – maturation and reproduction (ii), and IX-XII stages – senescence (iii) (Thomas, 2014; Maity et al., 2021). The Haun scale is focused on leaf formation and describes the length of each emerging leaf. The Zadoks scale describes only critical plant development stages (Gulumser & Mut, 2016; Rasti et al., 2022). Based on the Zadoks scale, BBCH uses a decimal code system and includes macro- and micro-stages (Thomas, 2014). BBCH scale is often used to determine the optimum timing for fertilisation, pesticide treatment, and other agronomic measures.

Today, in Ukraine, the following species are available: common sorghum (*Sorghum bicolor*), soryz (*S. oryzoidum*), sweet sorghum (high sugar content variety of *S. bicolor*), Sorghum-Sudan grass hybrid (*S. bicolor* x

Sorghum × *drummondii*), broom sorghum (*S. vulgare* var. *technicum*), perennial sorghum (*S. alnum* Parodi), and Sudan grass (*S. bicolor* subsp. *drummondii*) (State Register of Plant Varieties..., 2020). Of them, the most common species are common sorghum and sweet sorghum (Fedorchuk et al., 2017) grown on an area of 47 000 hectares. A considerable increase in sown area is observed in the Forest-Steppe zone (20%), which is characterised by unstable soil moisture. Meanwhile, sorghum has been conventionally grown in the Steppe zone characterised by insufficient soil moisture (Tretiakova et al., 2022).

In the early stages, sorghum plants demonstrate slow growth of the stem and leaf. Because of this, they cannot compete with weeds, especially in conditions of the Forest-Steppe, where weed species diversity and their seed reserves in soil are considerable (Fedorchuk et al., 2017). The lack of mobile forms of nutrients in the soil, along with drought, pests, and diseases can also decrease the yield potential of the crop (Marinov-Serafimov et al., 2018; Kraig et al., 2019). It is important to find effective ways of activating certain phenological stages of sorghum. Such agronomic measure may help avoid the stress in plants caused by adverse environmental factors during critical growth stages (Reddy et al., 2014).

S.S. Dhaliwala, V. Sharma, & A.K. Shuklab (2022) showed the effectiveness of the foliar application of fertilisers in sorghum. In the field experiments, crop yield increased from 15 to 25% due to the use of proper commercial micronutrient fertiliser. When the fertiliser was applied 4 and 6 weeks after sowing, biomass yield was 7-11% higher compared to the control (Stipešević et al., 2018). Application of spray foliar fertiliser Boost Extra 30, 60, and 90 days after planting at a rate of 2 L/ha ensured the highest yield of sorghum (Ogundare, et al., 2015). D. Vasundhara & V. Chhabra (2021) reported that the optimum timing of foliar nutrient application for corn and sorghum is in the 5-7 leaf stage, and the second application – after 14 days. In winter wheat, PGR is applied before and/or during the stem elongation stage (BBCH 30-32) aiming to produce lower internodes with shorter lengths, thicker stems and, stem cell walls (Gandee et al., 1997). Experiments by B. Bodson & M-H. Durdu (1996) on winter wheat showed that PGR can be applied not only in the period from the end of tillering to the beginning of stem elongation as conventionally recommended but also in more advanced stages of the vegetative period: 1st or 2nd node and even flag leaf emergence. Late applications do

not cause any substantial reduction in yield compared to applications at beginning of the stem elongation stage. In field experiments with spring wheat, the optimum timing for trinexapac ethyl application is Zadoks growth stages (GS) 30, 32, or 37. The application of trinexapac-ethyl in GS 37 resulted in less crop damage, shorter stands, and more erect plants than in GS 30 or 32 (Wiersma *et al.*, 2011). Early application of growth regulators in tillering stage increased the number of tillers, ears, and grain yield of winter wheat (Ghuman & Ram 2021). In experiments by Z. Hussain & M.H. Leitch (2007) on applications of PGR in spring wheat in growth stages 30 and 32, the grain yield was unaffected. However, the stem length decreased, while the specific stem weight increased.

Studies by many researchers contain a variety of information on the timing of the foliar feeding of sorghum. Among them, calendars of foliar applications of fertilisers (Ogundare *et al.*, 2015; Stipešević *et al.*, 2018; Dhalwala *et al.*, 2022) and certain phenological stages are identified (Vasundhara, & Chhabra 2021; Sebrina *et al.*, 2020). Thus, there is no consensus on the correct timing of the foliar feeding (Sebrina *et al.*, 2020).

Now, the most frequently used scales are Keller & Baggiolini, BBCH, Zadoks, and Haun, Feekes (Thomas, 2014; Lambright, 2019; Rasti *et al.*, 2022; López-Sandin *et al.*, 2022). In Ukraine, it is common to use the Kuperman scale (Kalenska *et al.*, 2018). The BBCH scale, which is based on more precise knowledge of plant development physiology, can hardly be compared to simpler scales (Kuperman, 1984; Reddy, 2017; Lambright, 2019).

The scientific originality of this study consists of the theoretical analysis of the practice of using plant growth and development scales and the practical selection of optimal scales of growth and development of sorghum for use in agronomic operations of tending crops, specifically, optimisation of the plant growth regulator application.

The purpose of this study is to evaluate the effectiveness of two scales (BBCH and Kuperman) for plant growth and development for the determination of the

most suitable time of PGR application to increase the yield of two common sorghum varieties and two sweet sorghum hybrids.

MATERIALS AND METHODS

Experimental site and soil condition

The study was conducted in the years 2012–2020 in two field experiments (two locations): state farm Salyvinkivske (Ksaverivka 2, Vasylkiv district, Kyiv region) and Bila Tserkva Research and Breeding Station (Mala Vilshanka. Bilotserkivskyi district, Kyiv region). Both locations belong to the Central Forest-Steppe of Ukraine, the zone of unstable moisture.

The soil of the state farm Salyvinkivske is podzolic chernozem, with the following characteristics, determined by methods of agrochemical analysis (Hospodarenko, 2013): the content of humus is 3.21% (by Turin) (DSTU 4289:2004, 2005), pH_{sal} – 6.4, alkaline hydrolysed nitrogen – 156 mg/kg (by Kornfeld) (DSTU 7863:2015, 2016), mobile phosphorus and potassium – 77 mg/kg and 89 mg/kg (by Chirikov) (DSTU 4115-2002, 2002). The soil of the Bila Tserkva Research and Breeding Station is typical low-humus chernozem, the content of humus is 3.50% (DSTU 4289:2004, 2005), pH_{sal} – 6.2, alkaline hydrolysed nitrogen – 134 mg/kg (DSTU 7863:2015, 2016), mobile phosphorus and potassium – 76 mg/kg and 98 mg/kg, respectively (DSTU 4115-2002, 2002).

Weather conditions

The analysis of weather conditions and their variability in the research years 2012–2020 compared to the average long-term data was performed based on the formula:

$$K_s = \frac{(X_1 - X_2)}{S}, \quad (1)$$

where K_s is the significance coefficient of deviations of the agrometeorological regime; X_1 is the element of the current weather; X_2 is the indicator of the long-term average value (1980–2020); S is the standard deviation (Rozhkov *et al.*, 2016).

Table 1. Significance coefficient (K_s) of deviations of precipitation and average monthly temperatures from long-term averages, 2012–2020

Months	Precipitation									Air temperature								
	2012	2013	2014	2015	2016	2017	2018	2019	2020	2012	2013	2014	2015	2016	2017	2018	2019	2020
April	0.8	0.4	0.4	-1.5	0.9	-1.0	-1.6	-0.1	-1.5	2.5	1.0	1.0	0.7	2.9	1.4	0.5	1.2	0.6
May	-0.5	2.1	2.1	-0.2	1.8	-0.4	-0.1	0.2	1.7	2.2	1.1	1.1	1.2	0.3	0.5	2.5	1.4	-0.8
June	-0.5	0.2	0.2	-1.5	-2.4	-1.7	0.4	0.2	-0.5	1.4	-0.4	-0.4	1.1	0.6	1.2	2.8	2.6	2.1
July	-1.1	0.3	0.3	-0.4	-1.3	-0.7	-2.1	-1.4	-0.2	2.4	1.4	1.4	1.4	2.3	0.9	2.7	0.3	1.1
August	1.2	-0.3	-0.3	-1.6	-0.9	-1.5	-1.2	-1.2	-0.4	0.8	1.4	1.4	1.4	1.7	2.3	3.0	1.2	0.9
September	-0.4	0.3	0.3	-0.5	-1.8	-1.7	0.5	-1.0	-0.5	1.5	0.2	0.2	2.2	1.3	1.8	0.7	0.8	1.8

Note: green indicates conditions close to normal (K_s 0–1), gray – conditions that differ substantially from the average perennials (K_s 1–2), orange – conditions close to the extreme (K_s >2)

In 2012, 2015, 2016, 2017, and 2018 the most substantial deviations in precipitation were observed. In 2015 and 2017, all the months of the vegetation period corresponded to insufficient precipitation. Extreme conditions of the average daily air temperature were in the years 2012 (April, May, July) and 2018 (May, June, July, August). The systematic lack of precipitation in 2015-2016 resulted in a moisture shortage in 2017 and the productivity of sorghum was rather low.

Experimental scheme and treatments

Field experiment I with two common sorghum varieties (Sorghum bicolor) was conducted at the state farm Salvyvinkivske in 2012-2015: *Factor A*: varieties Odeskyi 205 and Lan 59; *Factor B*: the timing of foliar fertilisation: stages 21, 31, and 37 (BBCH) and III, IV, and VI-VII (Kuperman). The PGR used in the study was Vympel 2 (0.5 L/ha; active ingredients: polyatomic alcohols 300 ± 30 g/L, humic acids 30 ± 0.3 g/L, carboxylic acids 3.0 ± 0.3 g/L). In the control treatment, the plants were treated with water only. The application rate of the working solution was 200 L/ha. The single plot area was 35 m²; the accounting area was 25 m².

Field experiment II with two sweet sorghum hybrids (Sorghum saccharatum Jakushev.) Huliver and Dovista was conducted at the Bila Tserkva Research and Breeding Station in 2016-2020. The scheme of the experiment: *Factor A*: hybrids: Huliver and Dovista; *Factor B*: foliar application of fertilisers: stages 21, 31, and 37 (BBCH) and III, IV, and VI-VII (Kuperman). Plants were treated with the PGR (application rate 0.5 L/ha; active ingredients: polyatomic alcohols 300 ± 30 g/L, humic acids 30 ± 0.3 g/L, carboxylic acids 3.0 ± 0.3 g/L). Control: the plants were treated with water only, as described in Experiment I. The single plot area was 35 m²; the accounting area was 25 m².

The experiment was established in a randomised plot scheme were planned according to the general methodology of experiments in agronomy (Rozhkov et al., 2016; Tkachyk, 2015).

Plant treatment

The cultivation technology sorghum was typical for the Right-Bank Forest-Steppe of Ukraine. Mineral fertiliser $N_{45}P_{60}K_{60}$ was applied in the experiment. Phosphate-potassium fertiliser was incorporated before tillage, while nitrogen fertiliser – before seedbed preparation. The seeds were sown to a depth of 2-4 cm at soil temperature $>12^{\circ}\text{C}$. Common sorghum was sown with a plant density of 160-180 plants per hectare (at harvest) at a row spacing of 70 cm. Sweet sorghum was sown in wide rows at a row spacing of 45 cm, at a seeding rate of 10-12 seeds/m and plant density of 190-230 thousand plants per hectare (at harvest). Common sorghum was harvested at grain humidity of $<20\%$ using a combine harvester, sweet sorghum was harvested using silo-harvesters in the stage of middle dough (Fedorchuk et al., 2017).

The seeds were treated with the antidote Concep® III (Syngenta, USA), which is approved for pre-emergence application to ensure efficient weed control. Post-emergence treatment of crops was performed using preparations based on S-metolachlor (Primextra TZ Gold 500 SC, Primextra Gold 720 SC, Dual Gold 960 EC, all Syngenta, USA). Dicotyledonous weeds were controlled by post-emergence herbicides of the 2.4 D group (Prima SE, Peak 75 WG, all Syngenta, USA) before the five-leaf stage (Fedorchuk et al., 2017). Before sowing, the seeds were treated with the fungicide Maxim XL 035 FS (Syngenta, USA). During the growing season, Karate Seon 050 SC and Engio 247 SC (all Syngenta, USA) were used to control such pests as aphids, European corn borer (*Ostrinia nubilalis*), and beet webworm (*Loxostege sticticalis*).

PGR Vympel 2 (Dolyna-Tsentr LLC, Ukraine) contains polyatomic alcohols (300 g/L), humic acids (30 g/L), carboxylic acids (3.0 g/L), and natural growth promoters-adaptogens. The product accelerates plant growth and development, activates nutrient consumption and leaf surface formation (Storozhyk & Muzyka, 2017).

Biochemical seed quality

Biochemical indicators of seed quality were determined by the following methods: protein content by the method of mineralisation with sulfuric acid in the presence of a catalyst according to Stein-Moore; starch – by the method of dissolution in a hot solution of hydrochloric acid followed by a polarimeter examination. The content of dry matter was determined by drying in a drying chamber at a temperature of 105°C for 4-6 h. The total sugar content in the juice of sugar sorghum stems was determined by the Luff-Schoorl method (Tkachyk, 2015).

Data analysis

The statistical processing of the experimental data was performed using ANOVA at LSD_{05} using the Statistica 10.0 package (StatSoft). The share of influence of the factors was determined based on the results of variance analysis (ANOVA) with the display of only reliable variances in the graphs (pie chart graph).

The summary statistics of the analysis of variance, specifically the sum of squares (SS), were used to build a graph of factor influence (Urdan, 2010). Only the influence of the effects at a substantiality level of $p < 0.05$ was used, setting this parameter when building the graph. Therewith, factors with values of $p > 0.05$ were automatically considered as an experimental error; thus the distribution of influencing factors was not disturbed.

RESULTS AND DISCUSSION

Tables 2 and 3 demonstrate the specific features of the impact of the PGR (0.5 L/ha) on the productivity of two sorghum varieties and two sorghum hybrids when applied in different growth stages by the BBCH and the Kuperman scales.

Table 2. The impact of the growth regulator (0.5 L/ha) on the productivity of two common sorghum (*Sorghum bicolor* L.) varieties when applied in different growth stages by the BBCH and the Kuperman scales (average for 2012-2015)

Variety	Stage	PGR	Duration of vegetation period (day)	Yield of grain (t/ha)	Protein content (%)	Starch content (%)
Odeskyi 205	BBCH microstage 21	Control	110	4.70	10.8	73.5
		GR	102 (-8)	5.26 (+ 0.56)	10.9 (+0.1)	73.8 (+0.3)
	Kuperman stage III	Control	110	4.73	10.9	73.5
		GR	103 (-7)	5.10 (+0.37)	10.8 (-0.1)	73.6 (+0.1)
	BBCH microstage 31	Control	110	4.68	10.8	73.6
		GR	104(-6)	5.05 (+0.37)	11.0 (+0.2)	73.9 (+0.3)
	Kuperman Stage IV	Control	110	4.71	10.7	73.5
		GR	104 (-6)	5.01 (+0.3)	11.0 (+0.3)	73.9 (+0.4)
	BBCH microstage 37	Control	110	4.74	10.8	73.2
		GR	107(-3)	4.92 (+0.18)	11.2 (+0.4)	74.1 (+0.9)
	Kuperman stage VI-VII	Control	110	4.70	10.9	73.3
		GR	108 (-2)	4.90 (+0.2)	11.3 (+0.4)	74.2 (+0.9)
Lan 59	BBCH microstage 21	Control	110	5.22	11.0	75.1
		GR	103 (-7)	5.70 (+0.48)	11.2 (+0.2)	75.3 (+0.2)
	Kuperman stage III	Control	110	5.20	11.1	75.0
		GR	104 (-6)	5.56 (+0.36)	11.1 (+0.0)	75.2 (+0.2)
	BBCH microstage 31	Control	110	5.25	11.0	74.8
		GR	105 (-5)	5.50 (+0.25)	11.4 (+0.4)	75.2 (+0.4)
	Kuperman stage IV	Control	110	5.18	10.9	74.9
		GR	105 (-5)	5.44 (+0.26)	11.5 (+0.6)	75.3 (+0.4)
	BBCH microstage 37	Control	110	5.17	11.2	75.0
		GR	108 (-2)	5.35 (+0.18)	11.6 (+0.4)	75.7 (+0.7)
	Kuperman stage VI-VII	Control	110	5.22	11.1	75.1
		GR	109 (-1)	5.32 (+0.1)	11.6 (+0.5)	75.9 (+0.8)
ANOVA (LSD ₀₅)			1	0.12	0.3	0.8

The application of the PGR in BBCH microstage 21 and Kuperman stage III reduced the vegetation period of Odeskyi 205 by 8 and 7 days, respectively, and Lan 59 – by 7 and 6 days. Application of PGR in later stages for common sorghum practically did not affect the duration of the vegetation period. The differences between these varieties and control treatments after the application of the regulator in BBCH microstage 37 and Kuperman stage VI-VII were 1-2 days for Lan 59 variety and 2-3 days for Odeskyi 205.

Faster growth and development of common sorghum plants promoted the foliar application of the PGR in BBCH microstage 21 and Kuperman stage III, which resulted in higher grain productivity – in Odeskyi 205 by 0.56 t/ha and 0.37 t/ha, respectively, and in Lan 59 – by 0.48 t/ha and 0.36 t/ha, respectively.

Application of the PGR in later stages of common sorghum (*Sorghum bicolor* L.) is interesting from the standpoint of the quality of the obtained harvest. Foliar fertilisation in BBCH microstage 31 and Kuperman stage IV in varieties Odeskyi 205 increased the content of pro-

tein by 0.2-0.33%, respectively, in Lan 59 by 0.4-0.6%, respectively, compared to control. Similarly, the content of starch was higher by 0.3-0.4 and 0.4%, respectively. In the case of foliar application of PGR in BBCH microstage 37 and Kuperman stages VI-VII, the yield of Odeskyi 205 was higher by 0.18 t/ha and 0.20 t/ha, respectively, compared to the control, and Lan 59 – by 0.18 t/ha and 0.10 t/ha, respectively. This data only slightly exceeded the indices of the experimental error. However, the quality characteristics of sorghum harvest increased considerably compared to the earlier application of the PGR. For example, the fertilisation in BBCH microstage 31 ensured an increase in protein content of 0.2% and starch – of 0.3% in Odeskyi 205 and 0.3% and 0.4%, respectively, in Lan 59. The variant with the fertilisation in Kuperman stage IV yielded 0.3% higher content of protein and 0.6% higher content of starch in Odeskyi; in Lan 59, the increase was 0.6% and 0.4%, respectively.

The results of the analysis of the factors that affect common sorghum yield are shown in Figure 1.

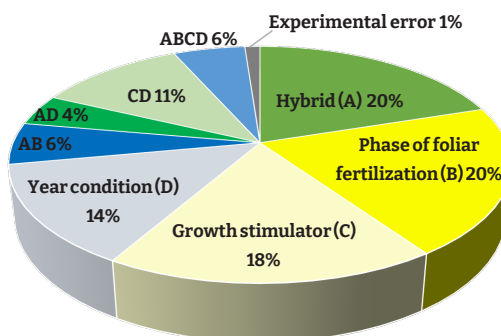


Figure 1. The influence of factors on the formation of common sorghum yield (only substantial factors of influence, $p < 0.05$ are defined)

It was identified that varietal characteristics and stage of foliar fertilisation are the most effective factor in sorghum productivity (20%). The PGR allows influencing the harvest formation at the level of 18%. Weather conditions contributed to the deviations in sorghum yield with a share of influence of 14%, with

interaction with the factor of PGR (CD) of 11% and variety (AD) – 4%.

The impact of the PGR on the productivity of the two sweet sorghum hybrids when applied in different growth stages by the BBCH and the Kuperman scales is shown in Table 3.

Table 3. The impact of the PGR (05 L/ha) on the productivity of two hybrids of sweet sorghum (*Sorghum saccharatum* Jakushev) when applied in different growth stages according to the BBCH and the Kuperman scales (average for 2016-2020)

Hybrid	Stage	PGR	Duration of vegetation period (day)	Biomass yield (t/ha)	Dry matter yield (t/ha)	Total sugars (%)
Huliver	BBCH microstage 21	Control	114	88.5	17.6	14.5
		GR	105 (-9)	93.5 (+5.0)	18.9 (+1.3)	14.9 (+0.4)
	Kuperman stage III	Control	114	88.7	17.8	14.6
		GR	107 (-7)	92.1 (+3.4)	18.4 (+0.6)	14.8 (+0.2)
	BBCH microstage 31	Control	114	88.4	17.8	14.5
		GR	108 (-6)	90.8 (+2.4)	18.0 (+0.2)	15.1 (+0.6)
	Kuperman stage IV	Control	114	88.9	17.8	14.5
		GR	108 (-6)	90.9 (+2.0)	18.2 (+0.4)	15.0(+0.5)
	BBCH microstage 37	Control	114	88.6	17.7	14.5
		GR	112 (-2)	89.9 (+1.3)	18.2 (+0.5)	15.3 (+0.8)
	Kuperman stage VI-VII	Control	114	88.5	17.6	14.6
		GR	113 (-1)	89.7 (+1.2)	18.0 (+0.4)	15.2 (+0.6)
Dovista	BBCH Microstage 21	Control	136	92.9	18.5	15.1
		GR	129(-7)	98.8 (+5.9)	19.8 (+1.3)	15.7 (+0.6)
	Kuperman stage III	Control	136	93.0	18.7	15.0
		GR	130 (-6)	97.3 (+4.3)	19.3 (+0.6)	15.6 (+0.6)
	BBCH microstage 31	Control	136	92.6	18.6	15.0
		GR	132 (-4)	96.5 (+3.9)	19.3 (+0.7)	15.8 (+0.8)
	Kuperman stage IV	Control	136	92.9	18.4	15.1
		GR	132 (-4)	96.4 (+3.5)	19.2 (+0.8)	16.0 (+0.9)
	BBCH Microstage 37	Control	136	93.2	18.5	15.0
		GR	133 (-3)	95.2 (+2.0)	19.1(+0.6)	16.5 (+1.5)
	Kuperman stage VI-VII	Control	136	93.3	18.6	15.1
		GR	134 (-2)	94.8 (+1.5)	18.9 (+0.3)	16.4 (+1.3)
ANOVA (LSD ₀₅)			1	1.3	0.5	0.4

When PGR was applied in BBCH microstage 21, Huliver hybrid showed a decrease in vegetation period of 9 days and Dovista hybrid – 7 days; when applied in Kuperman

stage III, the decrease was 7 and 6 days. After application of the PGR in BBCH microstage 37 and Kuperman stage VI-VII, the vegetation period was shorter, compared to

control, by 2 and 1 day, respectively, in Huliver and by 3 and 2 days, respectively, in Dovista. This proves the low efficiency of the application of PGR in the later stages for sweet sorghum.

As for the common sorghum varieties, foliar application of the PGR in BBCH microstage 21 and Kuperman stage III also stimulated the yield and quality of the crop (Table 3). The yield of Huliver biomass was 5.0 t/ha higher and dry matter – 1.3 t/ha higher. The total sugar content in the stem juice increased by 0.4% but it was statistically insubstantial. In Dovista, an increase amounted to 5.9 t/ha, 1.3 t/ha, and 0.6% respectively. When the PGR was applied in Kuperman stage III, the above-mentioned indices were in Huliver 3.4 t/ha, 0.6 t/ha, and 0.2%, respectively, and in Dovista 4.3 t/ha, 0.6 t/ha, and 0.6%, respectively which was statistically insubstantial. Thus, the optimal timing for the PGR application (BBCH microstage 21) contributed to obtaining higher productivity of the studied cultivars: in Dovista, biomass yield was high-

er by 1.6 t/ha, dry matter yield – by 0.7 t/ha and there was no increase in the sugar content of juice; while in Gulliver, these indicators were 1.6 t/ha and 0.7 t/ha, respectively, with the sugar content in juice increase of 0.2%.

After the application of the growth promoter in BBCH microstage 31, the increase in biomass yield in Dovista was 3.9 t/ha, dry matter 0.7 t/ha, and an increase in total sugar content 0.8% as compared to the control, and in Huliver the increase was 2.4 t/ha, 0.2 t/ha, and 0.6% respectively.

When the PGR was applied later, in BBCH microstage 37 and Kuperman stage VII, the biomass yield increase in Huliver was 1.3 t/ha and 1.2 t/ha, respectively, and in Dovista 2.0 t/ha and 1.5 t/ha, respectively. In Huliver, the increase was statistically insubstantial, while in Dovista it was. Therefore, it can be concluded that the application of PGR in later stages is less efficient for common sorghum varieties.

The results of the analysis of the factors that affect sweet sorghum biomass yield are shown in Figure 2.

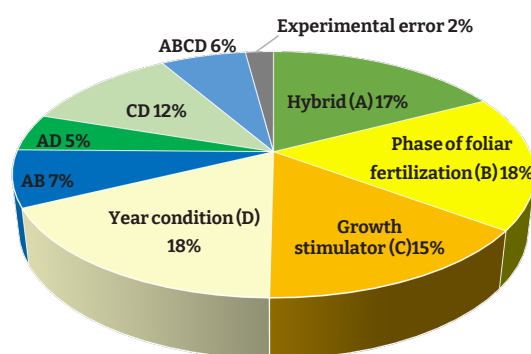


Figure 2. The influence of factors on the formation of sugar sorghum biomass yield (only substantial factors of influence, $p < 0.05$ are defined)

It was identified that the stage of foliar fertilisation (18%) and varietal characteristics (17%) are the most effective factor in sorghum productivity. The PGR allows influencing the harvest formation at the level of 15%. Weather conditions contributed to the deviations in sorghum yield with a share of influence of 18%, with interaction with the factor of PGR (CD) of 12% and variety (AD) – 5%.

Quality characteristics of sweet sorghum more substantially differed from the control in the case of foliar application of the PGR in BBCH microstage 37 and Kuperman stages VI-VII. The total sugar content in stem juice in Huliver increased by 0.8% and 0.6%, respectively, compared to control, and in Dovista – by 1.5% and 1.3%, respectively. The obtained data clearly shows that the foliar applications of the PGR (0.5 L/ha) in common sorghum and sweet sorghum sowings during the period of active growth and development considerably improve the quality and quantity of the obtained produce due to the activation of growth processes.

Nowadays, the investigation of the efficiency of growth and formation of common and sugar sorghum yield has gained new emphasis in connection with the

active use of the crop as a feedstock for bioenergy. However, the papers of researchers are mainly focused on determining the efficiency of the main fertilisation, in particular, nitrogen fertilisation (Pannacci & Bartolini 2018; Bartzialis *et al.*, 2020). However, in Ukraine, on the most common soil types, the optimal doses of fertilisers are quite well investigated (Fedorchuk *et al.*, 2017), while the optimal timing of application of foliar fertilisers needs clarification (Kalenska *et al.*, 2018).

This study on the correspondence of stages and microstages of sorghum was conducted on the background of long-term experiments on the foliar application of fertilisers as the method of intensification of plant growth at the beginning of their vegetation. It is known that sorghum plants grow slowly during the first 40-50 days after germination, which affects their adaptation to adverse conditions of cultivation (Reddy, 2017; Marinov-Serafimov *et al.*, 2018). In the case of foliar fertilisation with a PGR, the stage of plant growth and development is very important as everything that is applied, penetrates the plant very fast and there is no buffer concentration in the soil.

The specific features of the growth of sorghum and the ways of enhancing the efficiency of its cultivation through the optimisation of agronomic measures were investigated by a number of researchers (Calvino & Messing, 2012; Saadat & Homaei, 2015; Masaka *et al.*, 2019). The application of PGR was shown in earlier studies to be an efficient agrotechnical practice for the reliable intensification of plant growth, consequently, the yield and quality of the sweet sorghum crop, ensuring an average increase in the total sugar content in stem juice by 1.4% (Storozhyk & Muzyka, 2017).

Results in many aspects also correspond to other recent studies, proving that PGR affects the rate of photosynthesis processes, productivity, and general resistance to adverse cultivation conditions (Narayanan *et al.*, 2013; Stipešević *et al.*, 2018; Sebrina *et al.*, 2020). Therewith, the examples of using the BBCH and the Kuperman scales in the determination of the time for efficient application of PGR have not been identified in the scientific literature.

In conclusion, the application of the PGR as a component of cultivation technology in the early stages of sorghum plant development can be easier when using the BBCH scale because the Kuperman scale requires morphophysiological analysis to identify certain growth stages. Moreover, applications of the PGR (0.5 L/ha) in the BBCH 21 and 31 stages appear to give slightly better results than its applications in the Kuperman stages III and IV. In reality, the application dates based on the BBCH scale differed by 5-7 and 2-3 days compared to the Kuperman scale over the years of the study. This could be further investigated in more detail.

CONCLUSIONS

Foliar application of PGR (0.5 L/ha) in the early stages of sorghum growth and development requires accurate identification of stages and microstages of organogenesis. Foliar application of the PGR in Kuperman stage III ensured an increase in sorghum grain yield, compared to the control, by 0.37 t/ha in Odeskyi 205 and 0.36 t/ha in Lan 59. With the application of PGR in BBCH microstage 21, the obtained increase in yield was 0.56 t/ha in Odeskyi 205 and 0.48 t/ha in Lan 59. Similarly, the increase in the biomass yield of sweet sorghum was 3.4 t/ha in Huliver and 4.3 t/ha in Dovista using of the Kuperman scale, and 5.0 t/ha and 5.9 t/ha using the BBCH scale. In the early stages it is easier and more profitable to use relevant microstages by the BBCH scale to plan the application of a PGR, than equivalent ones of the Kuperman scale.

It was identified that application of the PGR at a rate of 0.5 L/ha in BBCH microstage 37 provided an increase in protein content in common sorghum grain of 0.4% in Odeskyi 205 0.4% in Lan 59, and starch content – 0.9% and 0.7%, respectively. Similarly, the application of the PGR at a rate of 0.5 L/ha in sweet sorghum hybrids lead to an increase in total sugar content in stem juice of 0.8% in Huliver and 1.5% in Dovista.

In this study, only two scales of growth and development, the most widely used in Ukraine, were considered. However, the value of simple scales lies in their ease and accuracy of use; therefore, the issue of the effectiveness of identifying plant growth and development stages using scales other than the BBCH and the Kuperman scales is relevant.

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Оптимальний час застосування регулятора росту рослин на посівах сорго за шкалою BBCH та Купермана

Анотація. Актуальність досліджень зумовлена проблемою вибору оптимальної шкали ідентифікації фаз росту рослин для визначення кращих строків проведення агротехнічних операцій догляду за рослинами сорго. Метою експерименту було визначення ефективного часу застосування регулятора росту рослин на 21, 31, 37 фазах (шкала BBCH) та III, IV, VI-VII (шкала Куперман). У роботі використовували польові та лабораторні методи досліджень. Дослід проводився в зоні Лісостепу України, два сорти сорго *Sorghum bicolor* і *Sorghum saccharatum* були оброблені регулятором росту рослин. Позакореневе підживлення регулятором росту рослин (0,5 л/га) на стадіях 21 і 31 BBCH дає на 2,8 та 4,9 % кращі результати, ніж на стадіях III і IV за Куперман (остання шкала базується на комплексному морфофізіологічному аналізі для ідентифікації I-VII періодів росту рослин). Позакореневе підживлення регулятором росту рослин на мікростадії 21 (BBCH) сприяло швидшому розвитку рослин сорго та збільшенню врожаю зерна сортів сорго (0,19 т/га сорту «Одеський 205» та 0,12 т/га сорту «Лан 59») порівняно з застосуванням в III стадії (Куперман). Подібний ефект регулятора росту рослин, застосованого на мікростадії 21 (BBCH) порівняно з III фазою (Куперман), зафіксовано для двох гібридів сорго цукрового: у сорту «Довіста» приріст урожайності біомаси становив 1,6 т/га, сухої речовини 0,7 т/га та цукристості стеблового соку 0,0%, тоді як у сорту «Гулівер» ці показники становили 1,6 т/га, 0,7 т/га та 0,2% відповідно. Таким чином, шкалу BBCH рекомендується використовувати для застосування регулятора росту рослин, а позакореневе підживлення слід проводити в мікростадії 21 і 31. Практична цінність роботи полягає у виборі, оптимальної для умов України, шкали росту та розвитку соргових культур та строків застосування позакореневого підживлення. Робота корисна з практичної точки зору проведення позакореневих підживлень рослин у виробництві так, а також як теоретичне дослідження шкал росту і розвитку рослин, для студентів та науковців

Ключові слова: позакореневе підживлення, визначення стадії розвитку рослин, етапи органогенезу, урожайність зерна, вихід біомаси

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Nutritional components of fresh and frozen fruits of highbush blueberries (*Vaccinium corymbosum* L.)

Abstract. The relevance of the study is conditioned by the significant popularity of highbush blueberries among consumers, which they have gained due to their balanced taste and the content of nutrients and bioactive substances that are useful for the human body. Therefore, the purpose of the study was to establish the suitability of fruits of the varieties 'Duke', 'Patriot', 'Chanticleer' for freezing and storage in a frozen state. Biometric, laboratory, analytical, and statistical research methods were used to achieve this goal. As a result of the conducted studies, it was established that the frozen fruits of highbush blueberries of the studied varieties 'Duke', 'Patriot', 'Chanticleer' met the requirements of the highest commercial grade according to DSTU 4837:2007. Defrosted berries of the 'Duke' variety had a better appearance than the other two varieties under study, were unfrozen with a wax coating inherent in this type of fruit, their weight loss during freezing was 0.20%, and juice loss after defrosting – 0.71%. Highbush blueberries of the varieties 'Duke', 'Patriot', 'Chanticleer' in the forest-steppe of Ukraine accumulated from 17.6 to 19.0 mg/100 g of vitamin C, 378-545 mg/100 g of polyphenolic substances, 127.2-176.8 mg/100 g of flavonoids, 59.4-162.9 mg/100 g of anthocyanins, and 15.6-32.5 mg/100 g of chalcones. Defrosted 'Duke' berries contained the largest amount of nutrients, while losses during freezing and defrosting were: vitamin C – 2.7 mg/100 g, total polyphenols – 102 mg/100 g,

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anthocyanins – 41.8 mg/100 g. The data obtained are of practical value for gardeners when choosing a variety for creating plantings of highbush blueberries, the fruits of which can be used both for fresh consumption and freezing

Keywords: polyphenols, vitamin C, anthocyanins, chalcones, flavonoids

INTRODUCTION

Fruit and berry crops are excellent sources of nutrients. They contain a significant amount of biologically active compounds, in particular, ascorbic acid, anthocyanins, flavanols, flavonoids, phenols, and tannins (Prokhorchuk, 2019; Galat, 2021). Fruit consumption improves the overall condition of the body, in particular, it helps strengthen the immune system, the functioning of the cardiovascular and digestive systems (Skrovankova *et al.*, 2015). Considering the data of the State Statistics Service of Ukraine, which indicates that the amount of fruit products consumed by one person per year from 2005 to 2021 increased by 29.4 kg and amounts to 58.7 kg, it can be assumed that functional food products have become a trend and are actively in demand in recent years (Balances and consumption..., 2019). The main reason for the increase in demand for fruit products is a healthy lifestyle, rational nutrition, and vegetarianism (Dhalaria *et al.*, 2020).

The prospects of highbush blueberries (*Vaccinium corymbosum* L.) are evidenced by the ever-growing volume of production of its fruits (Manganaris *et al.*, 2014; Dhalaria *et al.*, 2020). Their global production increased from 262 tonnes in 2006 to 556 tonnes in 2016 (Aliman *et al.*, 2020). According to FAOSTAT, the leaders in the production of highbush blueberry fruits are the United States (269 tonnes) and Canada (179 tonnes). In Ukraine, over the past 10 years, highbush blueberries have moved from a little-known niche crop to the status of one of the widely cultivated ones. As of 2017, Ukraine became one of the 5 largest European producers of highbush blueberries. The area under highbush blueberries for the period from 2007 to 2017 increased almost 12 times, from 130 to more than 1,500 hectares, and at the end of 2018, it was expected to expand to 2,000 hectares (Shevchuk *et al.*, 2021). Berries of *Vaccinium corymbosum* L. contain a large amount of valuable nutrients that have a positive effect on the human body and are part of a balanced diet (Szajdek & Borowska, 2008). A significant antioxidant capacity of its fruits is provided by the high content of vitamin C, anthocyanins, and phenolic compounds (Szajdek & Borowska, 2008; Skrovankova *et al.*, 2015; Coronel *et al.*, 2019). The fruits of highbush blueberries are consumed both fresh and processed, in particular, in the form of defrosted after freezing, dried or freeze-dried, as well as in the form of jams, jellies, juices, canned fruit and purees (Michalska & Łysiak, 2015; Balances and consumption..., 2019).

However, the short shelf life of highbush blueberries is the main problem in providing the population with products of this type. Therefore, producers are faced with the problem of choosing a processing method that would

ensure maximum preservation of the nutritional components of this type of fruit. One of such methods is freezing.

The main task of the research was to establish the influence of the freezing process, the duration of storage of berries in the frozen state and defrosting on the content of bioactive substances, as well as changes in physical and sensory indicators of their quality.

A number of researchers claim that processing highbush blueberries by freezing causes a significant decrease in its nutritional components (Brownmiller *et al.*, 2008). On the other hand, some researchers argue that certain freezing processes have a positive effect on the functional properties of raw materials, since they can promote the release of bound compounds, in particular, phenols and anthocyanins (Mullen *et al.*, 2002; Leong *et al.*, 2012; Neri *et al.*, 2020). R. Veberic (2014) noted an increase in the content of bioactive substances in berries after their long-term storage in a frozen state. Such ambiguous conclusions of the researchers gave rise to the study of the effect of freezing on the preservation of the nutritional values of highbush blueberry fruits. Therefore, the purpose of the study was to identify varieties of highbush blueberries, the fruits of which, according to the complex of preserved quality indicators, are an excellent raw material for freezing. The scientific originality of the study lies in the fact that for the first time, the physical, sensory, and bioactive quality indicators of fruits of the 'Duke', 'Patriot', 'Chanticleer' varieties grown in the forest-steppe of Ukraine were investigated and their suitability for freezing was established.

LITERATURE REVIEW

Highbush blueberry berries, according to researchers from Ukraine (Shevchuk *et al.*, 2021), France (Brat *et al.*, 2006) and other countries of the world (Smrke *et al.*, 2021), are a source of nutritional biologically active substances that are useful for the human body. They contain a significant number of compounds that have an antioxidant effect, in particular: vitamin C, polyphenols, flavonoids, anthocyanins, and chalcones. Anthocyanin substances, which are more than 500 in highbush blueberries, provide its fruits with a characteristic black colour. Anthocyanins have become the subject of many scientific studies, mainly for two reasons: their beneficial effects on human health and their use as potential sources of natural food colouring.

Natural extracts of these pigments are used in the food industry as a natural dye, which is an environmentally friendly alternative to synthetic ones (Mullen *et al.*,

2002). In addition to anthocyanins, blueberries contain flavonoids, which are a subgroup of polyphenols. It is the latter that are of particular interest to human health researchers because of their antioxidant activity (Brat *et al.*, 2006). The largest and most diverse group of polyphenols are flavonoids, which are found in the form of free molecules or bound to sugars. Flavonoids are characterised by a wide range of health benefits and are used in the pharmaceutical, medical, and cosmetics industries. This is conditioned by their antioxidant, antiinflammatory, antimutagenic, and anticarcinogenic properties, combined with their ability to modulate key functions of cellular enzymes. They are used to fight diseases such as cancer, Alzheimer's disease, and atherosclerosis (Panche *et al.*, 2016).

Chalcone substances are another subclass of flavonoids. They are also known as open-chain flavonoids. Chalcones, like flavonoids, have anticancer, antioxidant, antibacterial, antiinflammatory, antitumor, cytotoxic, and immunosuppressive properties. But due to their biological and morphological characteristics, the fruits of highbush blueberries are not endowed with good shelf life. The maximum period during which they can maintain their quality indicators is 1-3 months, so producers of highbush blueberries are faced with the task of extending the shelf life of this type of product while maximally preserving sensory and bioactive indicators of its quality.

One of the most effective ways to store fruits for a long time, which minimally affects their quality, is freezing (Neri *et al.*, 2020). With this method of processing, a significant amount of biologically valuable substances containing fruits is preserved, which allows the population to consume natural and useful products throughout the year (Michalska & Łysiak, 2015). Freezing inhibits many metabolic processes in fruits, slows down the growth kinetics of microorganisms, and ultimately prevents degradation reactions of important biological substances, which mainly affect the deformation of the texture and structure of the product and worsens the characteristic colour of berries (Neri *et al.*, 2020).

Processing and storage conditions cause a gradual decrease in the content of nutritionally valuable components, including vitamin C in berries (Skupień, 2006). The vitamin C content of berries is influenced by numerous factors: genetic variation, ripeness, climate, methods of cultivation, harvesting, and storage (Davey *et al.*, 2000), therefore, the content of this vitamin in berries is highly variable (Skupień, 2006).

Based on the considerable amount of scientific literature, it is established that the issue of changes in the content of biologically active substances in the fruits of highbush blueberries during freezing and defrosting is rather underinvestigated, so the studies performed are relevant and necessary. They will be useful both for scientific breeders to further conduct the breeding process for creating new varieties of highbush blueberries, and for producers when choosing raw materials for freezing.

MATERIALS AND METHODS

The study was conducted during 2020-2021. The objects of the study were the fruits of highbush blueberries – varieties of American selection 'Duke', 'Patriot', 'Chanticleer'. Fruits for laboratory tests were selected at the experimental sites of the laboratory of selection and technology of berry crops cultivation of the Institute of Horticulture of the National Academy of Agrarian Sciences (IH NAAS) of Ukraine. The weight of one sample was one kilogramme. Its geographical location is in the forest-steppe zone of Ukraine, altitude above sea level – 187 m (50° 27'16"N, 30° 13'25"W), distance to Kyiv – 4.0 km. Year of creation of plantings – 2016, planting scheme 1×3 m, the soil of the site is grey forest medium loamy. The system of soil retention in a row is mulching with sawdust, in the row spacing – sodification. The experiment was laid without irrigation, the care of plantings is recommended for the forest-steppe zone of Ukraine (Balabak *et al.*, 2017).

The zone's climate is temperate continental. According to long-term data, the average air temperature was +9.7°C and the amount of precipitation for the year was 466.3 mm. The minimum average long-term temperature – negative 28.4°C (February), the maximum – 37.7°C (August).

Analytical research

Analytical research was conducted in the laboratory of post-harvest quality of fruit and berry products of the Institute of Horticulture of the National Academy of Sciences of Ukraine. On the day of fruit collection, the content of vitamin C, total polyphenols, anthocyanins, flavonoids, and chalcones was determined. Laboratory tests were conducted on blueberries in a state of consumer ripeness with a characteristic shape and colour for each of the studied varieties according to the 'Methods of assessing the quality of fruit and berry products' (Kondratenko *et al.*, 2008). Crushed analytical samples of fruits to determine the content of biologically active substances in them were prepared using a laboratory homogeniser. The berries were weighed on an analytical scale with an accuracy of up to the second sign. Biochemical studies were performed in threefold repetition.

Highbush blueberries were frozen in a Samsung RZ32M7110SA/UA freezer, at a temperature of -24°C, in plastic containers. They were stored at a temperature of negative 18-20°C for six months. At the end of storage, weight loss in frozen fruits was determined. After that, the fruit was thawed at room temperature (+20°C) for 6 hours. Juice losses were determined in defrosted berries. Weight and juice losses were measured by weighing on analytical scales with accuracy up to the second sign, expressed in % of the weight of the fresh sample.

Determination of the content of nutrients was carried out in fresh and defrosted berries.

Vitamin C content (ascorbic acid) was determined according to the method based on its extraction from berry

samples with a mixture of 2% aqueous oxalic solution and 1% aqueous hydrochloric acid solution (80:20). 5 g of crushed fruit was ground using broken glass and transferred to a measuring flask with a capacity of 100 ml. The contents of the flask were brought to the mark with a mixture of 2% oxalic and 1% acetic acids (80:20) and left for extraction for 10 minutes. The resulting extract was filtered through a cotton wool filter into a 100 ml flat-bottomed flask. In a glass with a capacity of 50 ml, 10 ml of the extract was added and titrated from the microburette with a solution of 2.6 – dichlorophenol-indophenol with sodium salt (Tilmans paint) until a slightly pink colour appeared, which did not disappear for 1 min. According to the amount of paint that was used for oxidation, the content of vitamin C is determined, and a mixture of oxalic and acetic acids is used for control (Kondratenko *et al.* 2008).

Total polyphenol content was determined according to a method based on the extraction of phenolic substances from berry samples with ethyl alcohol. To do this, 5 g of crushed berry samples were added to a porcelain mortar, a small amount of ethyl alcohol was added and filtered under vacuum on a Buchner funnel through a blue ribbon paper filter into a Bunsen flask. The residue on the filter was washed with a small amount of ethyl alcohol until the sample was completely discoloured. The number of washes depended on the intensity of staining of the sample and was 3-5 times. Record the volume of alcohol used in ml. 7.9 ml of distilled water, 0.1 ml of Extract, 1 ml of Folin-Denis reagent were added to the test tube, mixed and after 3 minutes 1 ml of saturated sodium carbonate solution was added and mixed again. For an hour, the optical density of the contents of test tubes was recorded on an ULAB 102UV spectrophotometer at a wavelength of 640 Nm. For control, a mixture prepared from 8 ml of distilled water, 1 ml of Folin-Denis reagent and after 3 minutes, 1 ml of saturated sodium carbonate solution was added.

Total polyphenol content in the sample was calculated by equation (1):

$$X = \frac{A \cdot V \cdot 100}{V_1 \cdot M}, \quad (1)$$

where: X – total polyphenol content in the sample, mg/100 g; A – amount of chlorogenic acid in the volume taken for spectrophotometric determination, found on the calibration graph, mcg; V – total volume of the final extract of the sample, ml; V₁ – volume of aliquot of the final extract of the sample taken for analysis, ml; M – sample weight, g (Kondratenko *et al.*, 2008).

Anthocyanin and chalcone content was determined by spectrophotometric method (ULAB 102UV spectrophotometer). The absorption of extracts was determined at wavelengths of 530 and 364 Nm, respectively, using an alcohol extract from plant homogenate acidified with 3.5% hydrochloric acid (Kryventsov, 1982).

Flavonoid content was determined by absorption spectrophotometry by measuring the absorption of a flavonoid complex with a 3% aluminium chloride solution. An ULAB 102UV spectrophotometer with a wavelength of 410 Nm was used for the measurement (Vronska, 2018).

Statistical analysis

Statistical data processing was performed using STATISTICA 13/1 (StatSoft, Inc., USA). The results are presented as averages with their standard errors as the mean ± standard error (x±SE). Differences between repetitions and relative to the average intersort value were determined using ANOVA statistical model. The results of the research are presented at the level of reliability in P<0.05. Two-factor variance analysis of the materiality of the influence of the genotype of the variety and weather and climatic factors on the content of biologically active substances in the fruits of highbush blueberries and correlation analysis were performed in Excel software suite, Data Analysis tab.

RESULTS AND DISCUSSION

1. Sensory and physical indicators of the quality of frozen and defrosted highbush blueberry fruits

Weight loss of fruits during frozen storage and juice after their defrosting is a physical indicator that depends on the freezing temperature, defrosting rate, and varietal characteristics (Odarchuk *et al.*, 2020). It is from these physical characteristics of fruits that their appearance depends, which is normalised by the requirements of DSTU 4837:2007 (2008). According to the specified standard, 95% of frozen blueberries should be not cracked, and after defrosting without excessive juicing, the number of frozen blueberries should not exceed 10% of the total volume of the sample, and the colour should be uniform and characteristic of the berries of this variety, taste without extraneous flavour. According to the results of organoleptic analysis, the fruits of all the varieties under study were of the highest grade, they were whole, not deformed, and without foreign taste and smell.

In the process of freezing and storing in a frozen state, the weight loss of blueberry fruits ranged from 0.20 ('Duke') to 0.37% ('Patriot'), an intermediate value (0.21%) was found in the 'Chanticleer' variety (Fig. 1). These figures are significantly lower than those of red currants, where, according to research conducted by Y. Tereshchenko (2018), they were 1.5-1.8%.

Highbush blueberries of the 'Duke' variety after their defrosting had not only the smallest weight loss, but also the smallest juice loss (0.71%), they were more significant in the 'Chanticleer' variety (1.3 %), slightly smaller – in 'Patriot' (0.87%) (Fig. 1).

Considering the data obtained and comparing them with the results on juice losses of fruits of various crops, in particular, currants (2.7-4.9%) (Tageshchepko, 2018), cherries (4.8-7.1%) (Voitok, 2018), strawberries – 10.7% (Simakhina, 2019) and 16.9% (Odarchuk *et al.*,

2020), as well as sweet cherries of different maturation periods (13.6-22.9%) (Ivanova *et al.*, 2019), blueberries

from among all the above fruits have the least juice loss after their defrosting.

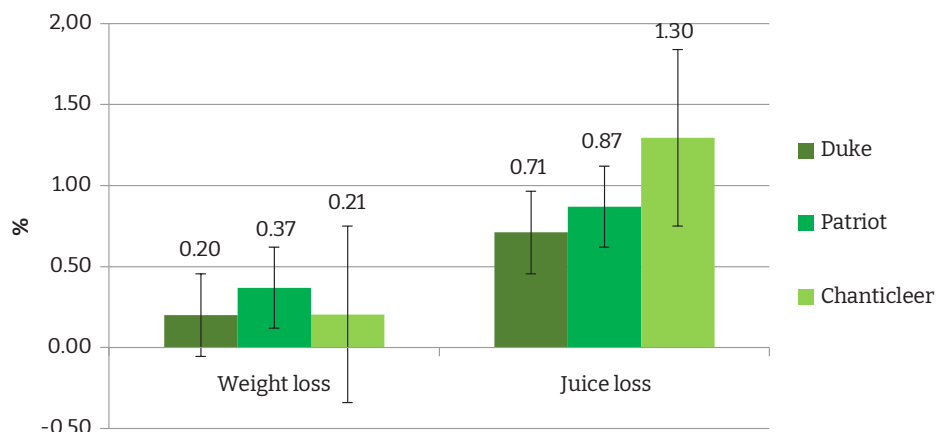


Figure 1. Weight and juice loss of highbush blueberries, % to fresh fruits (average for 2020–2021)

2. Vitamin C content of defrosted blueberries

According to Rafał Nadulski *et al.* (2019), highbush blueberries are not considered a source rich in vitamin C. They found a low content of this vitamin in fruits – 6.75 ± 1.06 mg/100 g of weight. Similar data were obtained by Latvian researchers. The fruits of highbush blueberry varieties studied by them contained from 7.43 to 9.98 mg/100 g by weight of ascorbic acid (Ozola & Duma, 2019). Blueberries grown in Romania accumulated this vitamin from 18.40 mg/100 g ('Blueray') to 13.79 mg/100 g 'Duke', average value – 16.28 mg/100 g (Ciucu-Paraschiv & Hoza., 2021). While blueberry fruits of the 'Bluecrop' variety grown in Turkey had a vitamin C content of 39.10 mg per 100 g of raw weight, which is significantly higher than fruits from Poland and Romania, as well as berries of wild forms of this crop, which accumulated this vitamin from 4 to 8 mg/100 g of raw weight (Celik *et al.*, 2018). According to B. Silenko and S. Marchenko (2013), fruits of highbush blueberries under grow-

ing conditions in the Right-Bank part of the western forest-steppe of Ukraine can accumulate from 15.19 ('Patriot') to 34.72 ('Dzhonni') mg/100 g of ascorbic acid.

According to the conducted studies, the fruits of highbush blueberries in the conditions of the forest-steppe of Ukraine accumulate 18.24 mg/100 g of raw weight of vitamin C (average for varieties), which is at the level of data obtained by Romanian researchers (Bunea *et al.*, 2011) and more than the indicators obtained by Polish researchers (Szajdek & Borowska, 2008). The highest amount of it was observed in fresh fruits of the 'Duke' variety – 19.0 mg/100 g of raw weight, the lowest – in 'Chanticleer' berries (17.6 mg/100 g of raw weight) (Fig. 2). Somewhat similar data were obtained in the laboratory of the horticultural Institute, but for other varieties, in particular, the amount of vitamin C in fresh blueberries ranged from 20.0 mg/100 g of raw weight ('Reca') to 27.0 mg/100 g of raw weight ('Elisabeth') with an intermediate value of 22.5 mg/100 g of raw weight ('Bluegold') (Shevchuk *et al.*, 2021).

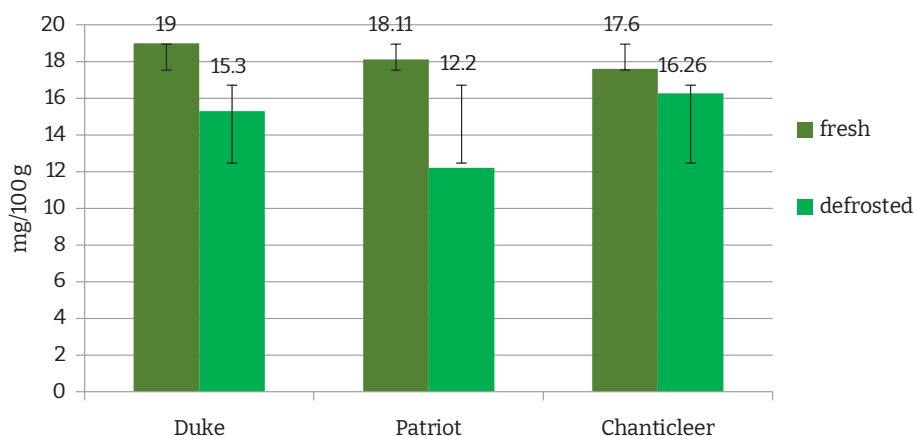


Figure 2. Vitamin C content in fresh and defrosted blueberry fruits, mg/100 g of raw weight (average for 2020–2021)

The fact of a decrease in the amount of vitamin C during fruit storage and defrosting has been established by many researchers (Skupień, 2006; Simakhina&Khalapsina, 2015). In particular, K. Scupien *et al.* (2006), argue that as the shelf life of frozen berries increases, the loss of this vitamin increases. As a result of the conducted studies, confirmation of previously obtained data was found, namely, the content of ascorbic acid in the studied defrosted blueberry fruits remained at the level of 62-76%, depending on the variety (Poiana *et al.*, 2010b). The fruits of the studied varieties of highbush blueberries, after storage in a frozen state and defrosting, quite significantly lost their vitamin C content. The lowest losses of this vitamin were in the variety 'Chanticleer' (8%), twice as high in 'Duke' (19%), and the largest in 'Patriot' (34%). The significance of the difference in the obtained data is explained by the different strength of fruit cells of different varieties, which during freezing causes cryogenic damage to them, and in the future, the process is accompanied by oxidation of ascorbic acid. The data obtained largely do not agree with the results of studies by Polish researchers, who note that blueberries lost from 57% ('Bluecrop') to 72% ('Jersey') of vitamin C during 6 months of storage (Skupień, 2006). Similar data were provided by G. Simakhina *et al.* (2019), which noted that the loss of vitamin C in highbush blueberries, blackberries, and mountain ash is 55.6-71.0%.

3. Content of total polyphenols and flavonoids in fresh and defrosted highbush blueberry fruits

Findings of L. Shevchuk (2021) showed that the content of total polyphenols in highbush blueberries ranges from 280.4 to 494.4 mg/100 g of fresh fruit weight, similar indicators of the content of these substances were obtained by W. Zheng and S.Y. Wang (2003) for blueberries of the 'Sierra' variety – 410 mg/100 g. According to research held in Romania, their number in the fruits of the 'Bluecrop' and 'Duke' varieties was 424.84-952.27 mg/100 g (Bunea *et al.*, 2011). According to K. Skupień (2006), similar to the amount of vitamin C, the highest total polyphenol content was found in 'Bluecrop' berries, and the lowest – in 'Blueray' and 'Jersey'. According to the data obtained, the fruits of highbush blueberries contained a higher amount of total polyphenols than indicated in the studies by a number of researchers, in particular, R. Moyer (2002), W. Zheng and S. Wang (2003). The content of polyphenolic substances in fresh fruits of the studied varieties was 482 ± 83 mg/100 g, significantly less than these substances accumulated in the 'Chanticleer' variety (378 ± 9.5 mg/100 g). Varietal difference of the polyphenolic component was at the average level, the variation coefficient was 17% (Table 1).

Table 1. Content of total polyphenols and flavonoids in fresh and defrosted highbush blueberries, mg/100 g of raw weight (average for 2020-2021)

Variety	Polyphenols			Flavonoids		
	fresh	defrosted	% change	fresh	defrosted	% change
Duke	524±9.8	422±2.5	-19	176.8±4.5	182±9.1 ^a	28
Patriot	545±7.4	417±8.1	-23	171.8±4.4	160.3±5.9 ^a	-7
Chanticleer	378±9.5 ^b	344±8.5 ^b	-9	127.2±4.0 ^b	106.0±3.4 ^b	-17
Average by variety	482±83	394±43	-18	158.6±24.8	149.4±3.4	-6
min	368	336	-	123.1	91.7	-
max	564	447	-	187.0	189.9	-
Variation coefficient	17	11	-	16	23	-

Note: the upper indices (a and b) in the lines next to the indicators indicate significantly different values of polyphenols and flavonoids relative to the average indicator (x) for the study group at $P < 0.05$

After freezing and defrosting, the greatest loss of polyphenols was observed in 'Patriot' variety (23%), while the lowest loss was observed in 'Chanticleer' (9%). Thus, the polyphenolic component of the studied defrosted fruits of highbush blueberries was the highest in the 'Duke' variety (422), slightly less contained in 'Patriot' (417), and the least – 'Chanticleer' (394 mg/100 g), which did not significantly change the picture regarding the amount of these substances that was in fresh fruits (Table 1). Significantly greater losses of the above substances in the fruits of blue honeysuckle were noted by

R. Khattab *et al.* (2015). Scientists have found that storing frozen at negative 18°C for six months reduces the content of total polyphenols by 37.08%. Other researchers have also observed the process of reducing polyphenolic substances in the fruits of highbush blueberries in their studies (Skupień, 2006). K. Skupień (2006) proved that the materiality of losses can depend on both freezing conditions and varietal characteristics. M.-A. Poiana, D. Moigradean (2010b) and K. Skupień (2006) proved that after 10 months of storage, there is an accelerated degradation of anthocyanin substances, which are a

component of polyphenols, in particular, in highbush blueberries by 13% compared to the amount that was in fresh berries. This is confirmed by the data of the conducted studies. Significantly greater losses of the above substances were noted by R. Khattab *et al.* (2015), but in the fruits of blue honeysuckle. He found that storing frozen at -18 °C for 6 months reduced the total phenol content of the fruit by 37.08%.

In general, the variation in the content of total polyphenols in fresh blueberries was average, the coefficient of variation was 17, and in defrosted blueberries it was insignificant, the corresponding indicator was 11 % (Table 1).

Fresh fruits of highbush blueberries of the varieties under study contained 158.6 mg/100 g of flavonoids. Significantly less than the average value for the studied varieties, their value was accumulated by berries of the 'Chanticleer' variety (127.2 mg/100 g), the amount of flavonoids in the other two varieties was higher, and was equal to: 171.8 ('Patriot') and 176.8 mg/100 g ('Duke'). In the process of freezing and defrosting, there was a significant reformatting of the biochemical component of the fruit, which resulted in a significant increase in the

flavonoid component in the fruits of the 'Duke' variety (by 28%), a decrease in their number, by 17%, was in the 'Chanticleer' variety and by 7% – in 'Patriot' (Table 1).

4. Anthocyanin and chalcone content in fresh and defrosted highbush blueberry fruits

Compared to other crops, blueberries are a significant source of anthocyanins (Grace *et al.*, 2019; Smrke *et al.*, 2021). According to K. McGee and M.C. Walton (2007), 100 g of its fruit can provide up to several hundred mg of anthocyanins. According to the conducted studies, the 'Duke' berry (162.9 mg/100 g) was significantly higher in anthocyanin content among the studied varieties. The other two varieties contained them from 59.4 ('Chanticleer') to 86.9 mg/100 g ('Patriot'), these data indicate a significant varietal difference, the coefficient of variation is 47%. Storage for 6 months of highbush blueberries of the 'Duke' and 'Patriot' varieties in a frozen state and after defrosting caused a decrease in the anthocyanin content by 26 and 22%, respectively. The opposite trend was observed in blueberry fruits of the 'Chanticleer' variety – the anthocyanin content increased by 15% and amounted to 68.5 mg/100 g (Table 2).

Table 2. Anthocyanin and chalcone content in fresh and defrosted highbush blueberry fruits, mg/100 g of raw weight (average for 2021-2022)

Variety	Anthocyanins			Chalcones		
	fresh	defrosted	% change	fresh	defrosted	% change
Duke	162.9±2.1 ^a	121.1±2.1 ^a	-28	32.5±1.5 ^a	34.7±2.4 ^a	7
Patriot	86.9±2.1	68.1±2.0	-22	25.5±1.2	30.6±1.6	20
Chanticleer	59.4±1.3	68.5±1.8	26	15.6±1.5 ^b	23.5±1.7 ^b	51
Average by variety	103.1±47.0	85.9±27.9	-17	24.5±7.6	29.6±5.7	21
min	58.1	56.35	-	14.1	21.7	-
max	164.9	123.05	-	33.9	36.0	-
Variation coefficient	47	33	-	32	19	-

Note: the upper indices (a and b) in the rows next to the indicators display significantly different values of anthocyanins and chalcones relative to the average indicator (x) for the study group at P<0.05

Long-term frozen storage of blueberries did not significantly affect the total polyphenol content, anthocyanins, or antioxidant activity (Ścibisz & Mitek, 2007). However, a number of researchers who have conducted research with other cultures indicate the opposite, in particular A. Hartmann *et al.* (2008) found that the amount of vitamin C and anthocyanins in defrosted strawberries was significantly lower than in fresh ones. Significant losses of the same substances, but in frozen pomegranate juice, were reported by H. Mirsaedghazi *et al.* (2014), in cherry fruit – by A. Chaovanalikit & R.E. Wrolstad (2004), in frozen strawberry, cherry and cherry juices – by M.A. Poiana *et al.* (2010a). The degradation effect is explained by the storage of fruits in a frozen state and the defrosting process (Celli *et al.*, 2016). These statements are also consistent with the results obtained in the study on strawberries (Poiana *et al.*, 2010a),

cherries and mazzard (Poiana *et al.*, 2010a), and blue honeysuckle berries (by 46.34-59.24%) (Khattab *et al.*, 2015).

L.M. Shevchuk (2019) found that in apple fruits, the amount of chalcones varies from 4.3 to 11.8 mg/100 g and prevails over the amount of anthocyanins, which is not typical for red-coloured fruits, in particular, their content in black currants is 40-100 mg/100 g, in strawberries – 15-28 mg/100 g, while the proportion of chalcones does not exceed 10 mg/100 g. A small number of chalcones (1.5-7.0 mg/100 g) was observed in red-coloured raspberries. The lowest content of these substances in fresh blueberry fruits was found in the 'Chanticleer' variety (15.6), and most of them were contained in the 'Duke' variety (32.5 mg/100 g). Under the influence of low freezing and storage temperatures, as well as the defrosting process, the number of chalcones in all the studied varieties increased significantly. In particular,

in 'Chanticleer' – by 51% and equal to 15.6 mg/100 g, in 'Patriot' – by 20%, in 'Duke' – by 7% (Table 2).

CONCLUSIONS

According to the results of organoleptic analysis, frozen fruits of highbush blueberries of the varieties 'Chanticleer', 'Patriot', and 'Duke' met the requirements of the highest commercial grade. The lowest weight loss after long-term storage in the frozen state and juice after defrosting was in the fruits of blueberries of the 'Duke' variety – 0.20 and 0.71%, respectively. Fresh berries of this variety in the process of growth in the forest-steppe of Ukraine accumulated the greatest amount of vitamin C (19.00 mg/100 g), polyphenols (524 mg/100 g), and anthocyanins (162 mg/100 g). In the process of freezing and defrosting, the losses of the first two of these substances were 19% each, anthocyanins – 28%, and the flavonoid and chalcone components of 'Duke' berries were also the highest (176.8 and 32.5 mg/100 g) compared to other varieties.

Blueberries of the 'Chanticleer' variety, during 6 months of storage in a frozen state at a temperature of negative 18-20°C, lost 0.21% of their weight. The loss of ascorbic acid after defrosting was 8%, polyphenols – 17%, but the

content of anthocyanins and chalcones increased by 51 and 26%, respectively, compared to the content in fresh berries.

The content of vitamin C, flavonoids, and anthocyanins in highbush blueberries of the 'Patriot' variety during freezing storage decreased by 34.7 and 22%, respectively, and the chalcone part increased (+20% to its original content) and amounted to 30.6 mg/100 g.

The result of the above is that the most suitable for freezing and storage in a frozen state, according to a set of quality indicators, is the 'Duke' variety. Therefore, this variety should be recommended to producers for creating plantings, the intended purpose of which is processing by freezing. 'Duke' and 'Patriot' varieties are distinguished by a significant content of common polyphenols, which is a valuable component of the nutritional quality of fruits. Therefore, both of these varieties can be involved as donors of economically valuable traits for further breeding work.

In order to clearly understand the adaptability of highbush blueberries to the conditions of Ukraine, it is necessary to continue studying the quality of fruits, including their size, shape index, crushing force, and the content of basic organic components and trace elements, which would be valuable not only for the breeding process but also for the food industry and pharmaceutical industry.

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Нутрієнтноцінна складова свіжих та заморожених плодів чорниці високорослої (*Vaccinium corymbosum* L.)

Анотація. Актуальність досліджень обумовлена значною популярністю чорниці високорослої серед споживачів, яку вона здобула завдяки збалансованому смаку, та вмісту корисних для організму людини поживних і біоактивних речовин. Тому метою проведених досліджень було встановлення придатності плодів сортів 'Duke', 'Patriot', 'Chanticleer' до замороження та зберігання у замороженому стані. Задля досягнення мети були застосовані біометричні, лабораторні, аналітичні та статистичні методи дослідження. В результаті проведених досліджень встановлено, що заморожені плоди чорниці високорослої досліджуваних сортів 'Duke', 'Patriot', 'Chanticleer' відповідали вимогам вищого товарного сорту згідно ДСТУ 4837:2007. Дефростовані ягоди сорту 'Duke' вирізнялися кращим зовнішнім виглядом ніж два інших досліджуваних сорти, були незморщеними з притаманним даному виду плодів восковим покриттям, втрати їх маси у процесі замороження становили 0,20%, а соку після дефростації – 0,71%. Чорниця високоросла сортів 'Duke', 'Patriot', 'Chanticleer' в умовах Лісостепу України накопичувала від 17,6 до 19,0 мг/100 г вітаміну С, 378-545 мг/100 г поліфенольних речовин, 127,2-176,8 мг/100 г флавоноїдів, 59,4-162,9 мг/100 г антоціанів та 15,6-32,5 мг/100 г халконів. Найбільшу кількість нутрієнтноцінних речовин містили розморожені ягоди сорту 'Duke', при цьому втрати в процесі замороження та дефростації становили: вітаміну С – 2,7 мг/100 г, загальних поліфенолів – 102 мг/100 г, антоціанів – 41,8 мг/100 г. Дані отриманих досліджень мають практичну цінність для садівників при виборі сорту для створення насаджень чорниці високорослої, плоди яких можна використовувати як для споживання у свіжому вигляді, так і замороження.

Ключові слова: поліфеноли, вітамін С, антоціани, халкони, флавоноїди

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