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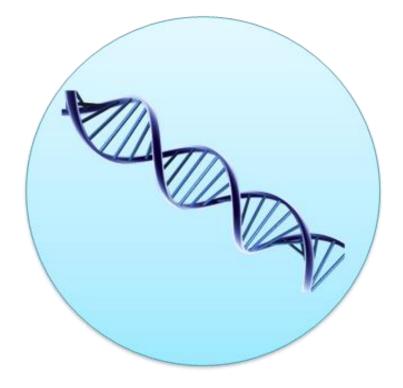
# СОВРЕМЕННАЯ БИОЛОГИЯ И ГЕНЕТИКА (МЕЖДУНАРОДНЫЙ НАУЧНЫЙ ЖУРНАЛ)

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UDC 57,578.2

# IDENTIFICATION OF CERTAIN BIOLOGICAL PROPERTIES AND RESERVOIR PLANTS OF TOMATO BROWN RUGOSE FRUIT VIRUS USING THE PCR METHOD

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*Summary.* In the cultivation of virus-free agricultural crops, primary attention should be given to the elimination of "natural reservoirs" of viruses. Various infected plants and plant residues in the soil serve as such natural sources of viral diseases. Identifying insect vectors and reservoir plants plays a significant practical and theoretical role in the development of virus control measures. Among the viral infections affecting tomatoes under protected cultivation conditions, the Tomato brown rugose fruit virus (ToBRFV), belonging to the family *Virgaviridae*, genus *Tobamovirus*, is considered a particularly dangerous pathogen. Importantly, the identification of ToBRFV in both cultivated tomatoes and perennial wild plants *Chenopodium quinoa*, *Taraxacum officinale*, and *Capsella bursa-pastoris* L. emphasizes the role of reservoir hosts in sustaining the virus during off-seasons and facilitating its re-emergence. The cosmopolitan nature and overwintering capacity of these weed species contribute to periodic viral circulation and increase the risk of widespread outbreaks in greenhouse environments.

Keywords: tomato, virus, ToBRFV, RT-PCR, RNA, reservoir, amplification.

# ОПРЕДЕЛЕНИЕ НЕКОТОРЫХ БИОЛОГИЧЕСКИХ СВОЙСТВ И РАСТЕНИЙ-РЕЗЕРВАТОРОВ ВИРУСА КОРИЧНЕВОЙ МОРЩИНИСТОСТИ ПЛОДОВ ТОМАТА МЕТОДОМ ПЦР

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Аннотация: При выращивании безвирусных сельскохозяйственных культур основное внимание следует уделять уничтожению «естественных очагов» вирусов. Различные поражённые растения и растительные остатки в почве служат такими «естественными очагами» вирусных заболеваний. В процессе разработки мер борьбы с вирусами важное практическое и теоретическое значение приобретает выявление переносчиков насекомых и резервуарных растений. Среди вирусных инфекций, поражающих томаты в условиях защищённого грунта, особо опасным патогеном считается вирус коричневой морщинистости плодов томата (*Tomato* 

brown rugose fruit virus, ToBRFV), относящийся к семейству Virgaviridae, род Tobamovirus. Важно отметить, что выявление вируса коричневого морщинистого плода томата (ToBRFV) как на культурных томатах, так и на многолетних диких растениях Chenopodium quinoa, Taraxacum officinale и Capsella bursa-pastoris L. подчеркивает важную роль резервуарных хозяев в сохранении вируса в межсезонный период и его повторном появлении. Космополитный характер и способность этих сорных растений перезимовывать способствуют периодической циркуляции вируса и повышают риск масштабных вспышек, особенно в условиях теплиц.

Ключевые слова: помидор, вирус, ToBRFV, RT-PSR, PHK, резервуар, амплификация.

#### Introduction

At present, special attention is being paid in the Republic of Uzbekistan to the expansion of areas allocated for the cultivation of agricultural crops. In particular, there is a steady trend toward increasing the sown areas of one of the most important cultivated plants tomato (Lycopersicum esculentum Mill.). Tomato holds a significant place in the structure of vegetable production in the country due to its high nutritional and dietary value, as well as its widespread use in both fresh consumption and processing. This crop is a rich source of vitamins, minerals, and antioxidants, especially lycopene, which has a pronounced health-promoting effect.

Tomato (*Lycopersicum esculentum Mill.*) is one of the main vegetable crops cultivated in Uzbekistan. Annually, approximately 60,000 hectares are planted with tomato in the country, with a total production volume of around 1.6 million tons. Between 2000 and 2019, tomato became one of the most important and widely grown vegetable crops worldwide, with global production increasing by more than 49 million tons [1].

Tomato plants infected by a complex of viral pathogens, including members of the genera Tobamovirus, Potexvirus, and Tospovirus, can suffer yield losses of up to 100%. On large cultivated areas, complete plant death before the end of the growing season is often observed [2]. Among the viral affecting tomatoes under infections protected cultivation conditions, the Tomato brown rugose fruit virus (ToBRFV), belonging to the family Virgaviridae and genus Tobamovirus, is considered a particularly dangerous pathogen [3, 4, 5].

Like other members of the Tobamoviridae family, Tomato brown rugose fruit virus (ToBRFV) is primarily transmitted mechanically over short distances, whereas infected seeds and fruits serve as the main means for longdistance spread. During various agronomic practices, the virus is transmitted through contact with workers' hands, clothing, footwear, and agricultural tools. Moreover, when ToBRFV is present in greenhouse conditions, the virus spreads rapidly. Even just two infected tomato plantswhich represent less than 0.5% of the total cultivation area-can contaminate up to 98.96% of the entire tomato crop within 9 months under normal growing conditions [6, 7]. ToBRFV is a seedtransmitted virus, with infected seeds playing a key role in the dissemination of the virus over considerable distances [8]. ToBRFV, like other tobamoviruses (TMV, ToMV, ToMMV), utilizes host cellular components to complete its life cycle. The viral cycle consists of three main stages: entry into the host cell, replication and translation of viral proteins in the cytosol, and movement and spread of the virus throughout the plant via the vascular system. Both viral and plant proteins play crucial roles at each of these stages. In terms of genome structure and sequences, ToBRFV is closely related to TMV; therefore, it is assumed that host factors required for the replication of TMV and ToMV also facilitate the replication of ToBRFV [9]. When cultivating agricultural crops free of viral diseases, primary attention should be given to eliminating "natural reservoirs" of viruses. Various infected plants and plant residues in the soil serve as such natural reservoirs for viral diseases [10, 11]. In developing virus management strategies, identifying

insect vectors and reservoir plants is of both practical and theoretical importance. Besides tomato, Tomato brown rugose fruit virus (ToBRFV) has also been detected in sweet pepper plants in Jordan and Sicily (Italy) [12, 13]. The host range of ToBRFV was studied on 31 plant species from seven botanical families, of which 20 species belonging to four families exhibited local symptoms or systemic virus presence in leaves [14]. The identified hosts include representatives of the following families: Solanaceae – petunia, pepper, tobacco, and tomato; Amaranthaceae — amaranth, guinoa, and lamb's quarters; Apocynaceae periwinkle; and Asteraceae - Indian lupine and crown chrysanthemum [14]. It is likely that many of these 20 species, well as numerous unidentified as potential hosts, will not be tested for the presence of ToBRFV due to the absence of pronounced symptoms or their lack of economic importance. Nevertheless, ornamental plants and weeds may act as sources of ToBRFV dissemination, contributing thereby to its epidemiological potential. In this regard, the main objective of the present study was to identify reservoir plants for Tomato brown rugose fruit virus (ToBRFV) using PCR under the climatic conditions of Uzbekistan, focusing on both wild and cultivated plant species.

## **Materials and Methods**

The study focused on various wild and cultivated plants that naturally harbor isolates of ToBRFV found in 6

Uzbekistan. A survey was conducted in a greenhouse farm located in the Kibray district of the Tashkent region, where symptoms on the leaves and fruits of the tomato cultivar 'Pink Paradise' were examined. From five sampling points, a total of 25 samples of leaves and fruits were collected, along with samples from different parts of symptomatic and asymptomatic weeds (leaves, stems, or roots) growing in the soil around greenhouses and along roadsides.

All samples were carefully packed into individual polyethylene bags. In the laboratory, the collected samples were thoroughly inspected and described in terms of symptomatology, followed by analysis using reverse transcription polymerase chain reaction (RT-PCR). The ToBRFV genome consists of single-stranded RNA, and its detection via RT-PCR involves several stages: extraction of viral RNA, synthesis of double-stranded cDNA from the RNA via reverse transcription, and amplification of the cDNA. Tomato samples testing negative via PCR were used as healthy controls. Extraction of Total RNA from Plant Tissues Total RNA was extracted from plant tissues using nucleic acid extraction kits from Agrodiagnostika (Russia) and the RNeasy Plant Mini Kit (Qiagen, Germany). When using the Agrodiagnostika kit, 1 gram of the plant sample was first weighed and then finely chopped into small pieces. For homogenization, the plant material was placed in a porcelain mortar, extraction buffer was added, and the sample was ground. The extraction thoroughly buffer was prepared in a 1:20 ratio, i.e.,

20 mL of buffer per 1 gram of plant material. The following components were used to prepare 1 liter of extraction buffer: KCl – 0.2 g, NaCl – 8 g, Na<sub>2</sub>HPO<sub>4</sub> 2.9 KH<sub>2</sub>PO<sub>4</sub> g, — 0.2 g, Polyvinylpyrrolidone (PVP) – 10 g, Bovine serum albumin (BSA) – 1 g, TWEEN-20 – 0.5 mL. From the resulting homogenate, 1.5 mL of sap was taken using an automatic pipette and transferred into Eppendorf tubes. The samples were then centrifuged at 2000 rpm for 5 minutes at 4°C. RNA extraction then was carried out according to the manufacturer's protocol.

RNA Extraction Using the **RNeasy Plant Mini Kit** For RNA extraction using the RNeasy Plant Mini Kit (Qiagen), plant tissues were ground either in liquid nitrogen or in 0.1 M phosphate buffer (Na<sub>2</sub>HPO<sub>4</sub>/KH<sub>2</sub>PO<sub>4</sub>, pH 7.2). The buffer-to-sample ratio was 1:10 (w/v) for leaf tissue and 1:20 for fruits and bees. From the resulting plant sap, 100 µL was mixed with 380 µL of RLT buffer from the RNeasy Plant Mini Kit. RNA extraction was then carried out according to the manufacturer's protocol. The total RNA concentration was measured using the QuantiFluor system (Promega). Amplification was carried out using primers developed by Rodríguez-Mendoza et al. (2019). This classical RT-PCR assay is suitable for detecting and identifying ToBRFV in tomato and pepper leaves, calyxes, and fruits. The primer pair targets the **RNA-dependent RNA** putative

polymerase gene (ToBRFV-IL isolate,

GenBank No. KX619418) (Table 1).

et

One-step **RT-PCR** (Loewe Biochemica GmbH) Prepare master mix according manufacturer's to instructions. Reverse transcription PCR parameters according cycling to manufacturer's The instructions. amplification protocol was: reverse transcriptase 42 °C 45 min, initial denaturation 95°C 12 min, than 35 cycles:95 °C 30 sec, 57°C 30 sec, 72°C 5 min and then final elongation 72 °C 5 min. The amplification was performed using a BioRad C1000 Thermal Cycler (USA). The resulting **c**DNA amplification products were separated by 1.5 % agarose gel electrophoresis at 100 V for 50 minutes, with migration from the anode (–) to the cathode (+).

The viral load in tomato fruit and leaf tissues, as well as in wild-growing weed plants, was determined by realtime RT-PCR using the One-step duplex real-time RT-PCR protocol based on ISF-ISHI-Veg (2020) was developed for the detection and identification of ToBRFV in leaf, sepal and fruit material.

#### Table 2

Primer Name	Sequence (5'–3')
CaTa28 Fw	5'-GGT GGT GTC AGT GTC TGT TT-3'
CaTa28 Rv	5'-GCG TCC TTG GTA GTG ATG TT-3'
CaTa28 Pr	5'-6FAM- AGA GAA TGG AGA GAG CGG ACG AGG-BHQ'1–3'
CSP1325 Fw	5'-CAT TTG AAA GTG CAT CCG GTT T-3'
CSP1325 Rv	5'-GTA CCA CGT GTG TTT GCA GAC A-3'
CSP1325 Pr	5'-VIC-ATG GTC CTC TGC ACC TGC ATC TTG AGA-BHQ'1-3'

**Real-time RT-PCR primers used in this study** 

After One-step duplex real-time RT-PCR protocol was run on these samples in a mix that contained  $0.75 \ \mu M$ each primer (CaTa28 Fw, CaTa28 Rv, CSP1325 Fw, CSP1325 Rv), 0.5 µM each probe (CaTa28 Pr, CSP1325 Pr) at a concentration of 10 pmol, 5

µMdeionized water, 10 µM master mix (GE50.5100 Xpert One-Step Fast Probe), 1 µM Rtase and 5 µM RNA. Total volume was 25 µM per sample. The amplification protocol was: reverse transcriptase 50 °C 15 min, initial

		J	
Primer Name	Sequence (5'–3')	Amplicon Size (bp)	Reference
ToBRFVFMX	AACCAGAGTCTTCCTATACTCGGA	475	Rodríguez-
ToBRFVRMX	СТСАССАТСТСТТААТААТСТССТ		Mendoza e
ΙΟΔΚΓΥΚΙνΙΛ	CICACCAICICITAAIAAICICCI		al. (2019)

Classical RT-PCR primers used in this study

denaturation 95°C 2 min, than 40 cycles:

95 °C 10 sec, 60°C 30 sec.

#### **Results and Discussion**

As the study, part of а diagnosis visual preliminary was conducted on the tomato cultivar 'Pink Paradise' grown in greenhouses of a farm located in the Qibray district of Tashkent region. During sample collection, plants exhibiting symptoms specific to ToBRFV, as described in the literature, were selected. In order to investigate the persistence of the virus under greenhouse conditions across sampling growing seasons, and monitoring of wild plant species (weeds) were also carried out. Environmental conditions and potential routes of plant infection were analyzed. For subsequent total RNA extraction, fruit and leaf tissue samples were collected according to a diagonal

sampling pattern from different areas of the cultivated field and placed in individual polyethylene bags. As negative controls, leaves and fruits of tomato plants confirmed to be free of ToBRFV by PCR were collected from the "New Agromax" farm located in the Zangiata district of Tashkent region.

During the analysis of leaf samples, symptoms such as chlorosis on young apical shoots, the formation of blister-like swellings on the leaf surface, mosaic patterns, discoloration, and tissue deformation were observed. In tomato fruits, the predominant symptoms included deformation of immature fruits, uneven ripening, irregular discoloration, and the appearance of chlorotic spots (Figure 1).





#### Fig. 1. Symptoms of viral infection on tomato fruits and leaves

Samples were homogenized using an autoclaved sterile porcelain mortar with the addition of liquid nitrogen or extraction buffer, after which the plant tissue was thoroughly ground. Total RNA was extracted using the RNeasy Plant Mini Kit (QIAGEN, Germany) and the RNA extraction kit from Agrodiagnostika (Russia), following the manufacturers' protocols. The extracted RNA was then mixed with primers using a specialized PCR master mix.

Samples of weed species such as *Taraxacum officinale* (dandelion), *Capsella bursa-pastoris* (shepherd's purse), and *Chenopodium quinoa* (quinoa), growing in the greenhouse, were also collected and analyzed using conventional RT-PCR. The results obtained are presented on the electropherogram (Figure 2).

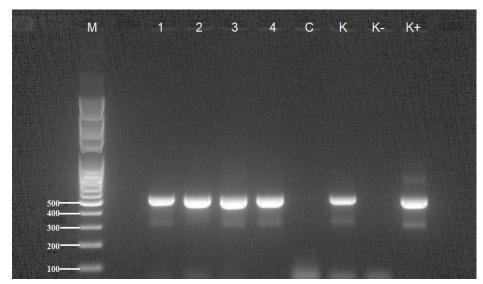


Fig. 2. Electrophoretic analysis of conventional PCR (RT-PCR) results. 1-tomato fruit (cv. 'Pink Paradise'), 2 – dandelion (*Taraxacum officinale*), 3 – shepherd's purse (*Capsella bursa-pastoris* L.), 4 – quinoa (*Chenopodium quinoa*), C – healthy tomato, K– infected tomato fruit, K– negative control (Loewe), K+ – positive control (Loewe).

The electropherogram shows that amplification of ToBRFV was detected both in the fruits of the tomato cultivar 'Pink Paradise' and in all studied weed species.

Analyses at the Central Phytosanitary Laboratory of the Agency for Plant Quarantine and Protection are carried out in accordance with international standards. According to these standards, confirmation of a detected quarantine organism requires verification using an alternative method. Therefore, the samples were analyzed using the "Real-Time PCR Detection Kit for Tomato brown rugose fruit virus" developed by the Russian company Agrodiagnostika, performed on the CFX96 Real-Time System (BioRad, USA).

The obtained results were interpreted using specialized bioinformatics software.

Table 3

Well	Fluor	Target	Content	Sample	Cq	Cq Mean	Cq Std. Dev
C10	FAM		Unkn	Tomato fruit «Pink Paradise»	31,52	31,52	0,000
C11	FAM		Unkn	Capsella bursa-pastoris L	31,62	31,62	0,000
C12	FAM		Unkn	Healthy Tomato fruit	N/A	0,00	0,000

# Quantification Data of Real-Time PCR detection of ToBRFV

D01	FAM	Unkn	Taraxacum officinale	35,85	35,85	0,000
D02	FAM	Unkn	Chenopodium quinoa	34,64	34,64	0,000
D03	FAM	Unkn	Internal control	N/A	0,00	0,000
D04	FAM	Neg Ctrl	Negative control	N/A	0,00	0,000
D05	FAM	Pos Ctrl	Positive control	26,01	26,01	0,000

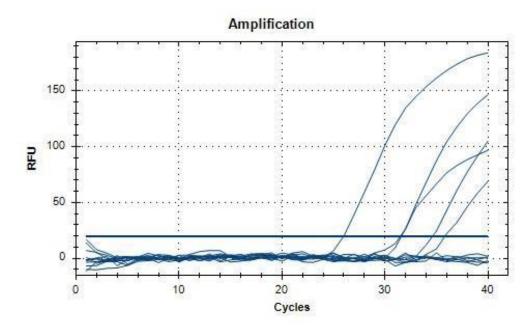


Fig. 3. Graphical representation of ToBRFV detection results using RT-PCR analysis.

To confirm the presence and strain composition of ToBRFV in RNA samples isolated from infected Pink Paradise tomato fruits and weeds growing in the greenhouse, RT-PCR was performed. The virus accumulation level in Pink Paradise tomato fruits was observed at a Cq value of 31.52, while in Capsella bursa-pastoris L. it was 31.62. In RNA samples isolated from Taraxacum officinale and Chenopodium quinoa, the Cq values were 35.85 and 34.64, respectively. For the positive control provided by the manufacturer, the Cq value was 26.01. The internal control and negative control both showed a Cq value of 0.

According to the analysis of reservoir plants of ToBRFV growing in greenhouses alongside tomato, the presence of the virus in Chenopodium quinoa has been previously reported in the literature. Additionally, the virus was detected in Taraxacum officinale and Capsella bursa-pastoris L., which have not been previously mentioned in the literature (Figure 4 a, b). Based on the conducted study, it was established that ToBRFV is present in varying concentrations in perennial plants such as Taraxacum officinale, Capsella bursapastoris L., and Chenopodium quinoa. This indicates that these plant species act as reservoir hosts for ToBRFV.



Fig. 4. Reservoirs of ToBRFV: a) Taraxacum officinale, b) Capsella bursa-pastoris L., c) Chenopodium album

Considering that *Taraxacum* officinale and *Capsella bursa-pastoris* L. are cosmopolitan species whose leaves overwinter under snow and resume

growth in spring, the persistence of ToBRFV in these reservoir plants facilitates periodic virus circulation and its spread over a wider area.

#### **Discussion and Conclusion**

In recent years, viruses infecting important agricultural plants in Uzbekistan have effective research is being conducted on it [15, 16, 17, 18], molecular identification of the virus [19, 20], obtaining specific serum for virus immunodiagnostics and its practical application [17] and studying the effect of the virus on some physiological properties of the plant research is being conducted on it [21].

Viral RNA isolated from Alamino variety Uzbeksiton climate NCBI of the isolate "Tomato brown rugose fruit virus Uzb\_4.1" for the first time from Shariot served as the basis for its deposit in the database with the number OR501605 [7].

The results of this study highlight the importance of early and accurate detection of *Tomato brown rugose fruit virus* (ToBRFV) in tomato production systems, particularly in greenhouse conditions where the virus can persist and spread via both cultivated and wild plants. The application host of molecular techniques such as RT-PCR has demonstrated high sensitivity and specificity, allowing for reliable identification of ToBRFV even at low viral concentrations. These methods provide significant advantages over traditional diagnostic approaches such as indicator plants and immunological assays, particularly in distinguishing between genetically similar viral strains.

Importantly, the identification of ToBRFV in both cultivated tomatoes and perennial wild plants-*Chenopodium quinoa, Taraxacum officinale,* and *Capsella bursa-pastoris* L.-emphasizes the role of reservoir hosts in sustaining the virus during off-seasons and facilitating its reemergence. The cosmopolitan nature and overwintering capacity of these weed species contribute to periodic viral circulation and increase the risk of widespread outbreaks in greenhouse environments.

In this context, implementing effective virus management strategies becomes critical. This includes the use of certified virus-free seeds and seedlings, enhancement plant of immune and responses, integrated pest management to control insect vectors. Moreover, regular phytosanitary monitoring and molecular screening of both cultivated and surrounding wild

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7. Munira Bakhtiyorova Tokhirbek Norov, Sevarakon Khodjaeva, Nasiba Botirova, flora should be integrated into greenhouse management practices to reduce the risk of infection and limit the economic losses associated with ToBRFV.

Overall, the integration of molecular diagnostic tools with comprehensive phytosanitary measures offers robust framework а for controlling the spread of ToBRFV and ensuring the production of healthy, high-quality tomato crops.

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# ALTERATIONS IN PHOTOSYNTHETIC PIGMENTS IN VIRUS-AFFECTED CHERRY LEAVES

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Summary. This study investigates the impact of Cherry Leaf Roll Virus (CLRV) infection on chlorophyll pigment content in cherry (Prunus spp.) leaves. Chlorophyll a, chlorophyll b, total chlorophyll, and carotenoid levels were measured spectrophotometrically after pigment extraction with 96% ethanol. Results show a significant decrease in chlorophyll pigments correlated with the severity of viral infection. Notably, chlorophyll b content declined by nearly 80% in severely infected leaves compared to healthy controls, indicating substantial impairment of photosynthetic capacity. These findings confirm that CLRV infection disrupts chloroplast function and reduces photosynthetic pigments, serving as a reliable physiological marker for viral disease progression in cherry trees. The combined use of pigment quantification and molecular diagnostics offers an effective approach for early detection and management of CLRV in orchards.

*Keywords:* Cherry Leaf Roll Virus (CLRV), chlorophyll content, photosynthesis, chlorophyll a and b, carotenoids, virus-induced stress, spectrophotometric analysis.

# ИЗМЕНЕНИЯ ФОТОСИНТЕТИЧЕСКИХ ПИГМЕНТОВ В ЛИСТЬЯХ ВИШНИ, ПОРАЖЁННЫХ ВИРУСОМ

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Аннотация. В настоящем исследовании рассматривается влияние инфекции вируса скручивания листьев вишни (Cherry Leaf Roll Virus, CLRV) на содержание хлорофилловых пигментов в листьях вишни (Prunus spp.). Содержание хлорофилла а, хлорофилла b, общего хлорофилла и каротиноидов определялось спектрофотометрически после экстракции пигментов 96% этанолом. Результаты показали значительное снижение уровня хлорофиллов, коррелирующее с тяжестью вирусного поражения. Особенно заметно снижение содержания хлорофилла b - почти на 80% в сильно поражённых листьях по сравнению с контрольными образцами, что свидетельствует о серьёзном нарушении фотосинтетической способности. Полученные данные подтверждают, что инфекция CLRV нарушает функционирование хлоропластов и снижает фотосинтетических пигментов, что уровень может служить надёжным физиологическим маркером прогрессирования вирусной инфекции у вишнёвых деревьев. Совместное использование количественного анализа пигментов и молекулярной диагностики представляет собой эффективный подход для раннего выявления и контроля CLRV в садах.

Ключевые слова: вирус скручивания листьев вишни (CLRV), содержание хлорофилла, фотосинтез, хлорофилл а и b, каротиноиды, стресс, вызванный вирусом, спектрофотометрический анализ.

#### Introduction

Viral infections in cherry trees (*Prunus* spp.) can significantly affect leaf physiology, particularly by reducing chlorophyll content, which plays a central role in plant metabolism and growth processes [2, 6]. The reduction of photosynthetic pigments not only leads to visible symptoms such as chlorosis, but also compromises overall plant productivity and fruit quality, posing a serious economic concern for fruit growers [7].

This study investigates changes in chlorophyll levels in cherry leaves following exposure to plant viruses, with a specific focus on Cherry Leaf Roll Virus (CLRV). CLRV is a nepovirus that has been reported in many Prunus species, and it is known to cause leaf rolling, chlorosis, and decline in photosynthetic efficiency [1, 8]. Infected leaves often exhibit a marked decline in total chlorophyll, especially chlorophyll and *b*, due to disruptions in а chloroplast structure and photosynthetic function [1]. These physiological effects are commonly linked to virus-induced damage to thylakoid membranes and altered expression of photosynthesisrelated genes [5, 9].

Spectrophotometric analysis and SPAD chlorophyll meter readings have consistently confirmed a significant decrease in chlorophyll content across infected samples, making these techniques valuable tools in the early diagnosis of viral infections [3, 4]. The severity of chlorophyll degradation is influenced by multiple factors, including the virus type, stage of infection, and the host plant genotype [10].

Chlorophyll is indispensable for light harvesting and energy conversion during photosynthesis. Biotic stress, such as virus infection, can impair chlorophyll biosynthesis, promote chlorophyll degradation, and disrupt stomatal conductance, leading to lower photosynthetic rates and compromised plant vigor [6, 11]. In Prunus spp., common viral pathogens such as CLRV and Prunus Necrotic Ringspot Virus frequently co-occur (PNRSV) and exacerbate the physiological burden on infected plants, often resulting in stunted growth, reduced leaf area, and premature senescence [1, 12].

shows that Research viral infections can alter the expression of chlorophyll-binding proteins, induce oxidative stress, and promote reactive oxygen species (ROS) accumulation, further accelerating pigment breakdown [5, 9]. Because of this, chlorophyll concentration serves as a valuable physiological marker for assessing the impact of viral stress in orchard crops. Early detection via non-destructive methods such as SPAD or pigment

extraction analysis is crucial for managing viral diseases and minimizing crop loss [3, 13].

Despite its importance, relatively few studies have focused specifically on how viral infections affect chlorophyll dynamics in cherry leaves under natural field conditions. This study aims to bridge that gap by evaluating the

#### Materials and methods

Leaf samples were collected from cherry trees (*Prunus* spp.) located in Chirchik, Tashkent region. Samples included healthy leaves, as well as leaves exhibiting varying degrees of viral infection symptoms classified as mild, moderate, and severe. From each category, representative leaf samples were collected for pigment analysis.

To determine chlorophyll pigment content, 0,5 g of fresh leaf tissue was finely ground and extracted in 10 mL of 96% ethanol. Extraction was performed in a water bath at 60 °C for 30 minutes to ensure complete pigment changes in chlorophyll content in virusexposed cherry leaves, particularly those infected with CLRV. The findings provide insights into the physiological response of cherry trees to viral stress offer foundation and а for the development of diagnostic indicators effective orchard management and practices.

release. The samples were then centrifuged at 8000 rpm for 10 minutes to separate the supernatant.

The clear supernatant was analyzed using а UV-5100 spectrophotometer at wavelengths of 663 nm, 645 nm, and 470 nm. Absorbance values were recorded to quantify chlorophyll a, chlorophyll b, total chlorophyll, and carotenoid contents according the formulas to established by Arnon (1949):

Chlorophyll a (mg/L) =  $12.7 \times A_{663} - 2.69 \times A_{645}$ Chlorophyll b (mg/L) =  $22.9 \times A_{645} - 4.68 \times A_{663}$ 

Carotenoids (mg/L) =  $(1000 \times A_{470} - 1.82 \times A_{663} - 85.02 \times A_{645}) / 198$ 

Total Chlorophyll (mg/L) =  $20.2 \times A_{645} + 8.02 \times A_{663}$ 

Pigment concentrations werebiomass using the formula fromconverted to mg per gram of dryLichtenthaler (1987):

#### $C(mg/g)=C\times V/1000\times m$

where *C* is the pigment concentration in mg/L, *V* is the volume of extract (mL), and *m* is the dry weight of the leaf sample (g).

All measurements were performed in triplicate, and mean values were calculated. Differences in pigment content among healthy and infected leaves were statistically analyzed using ANOVA with significance set at p < 0.05.

# **Results and Discussion**

Chlorophyll pigment contents were quantified in healthy, mildly infected, moderately infected, and severely infected cherry leaf samples. The data are summarized in Table 3.1. A clear trend of chlorophyll degradation was observed as the severity of viral infection increased.

Table 1

Sample Type	Chlorophyll a	Chlorophyll b	Total Chlorophyll
Healthy	0.524	0.959	1.499
Mildly Infected	0.520	0.634	1.230
Moderately Infected	0.510	0.595	1.120
Severely Infected	0.414	0.198	0.612

Notably, **chlorophyll b content decreased sharply** with disease severity. In healthy samples, chlorophyll b averaged **0.959 mg/g**, while in severely infected samples it dropped to **0.198 mg/g**, representing nearly an **80% reduction**. Chlorophyll a also showed a decline, though less drastic, with values ranging from **0.524 mg/g** in healthy leaves to **0.414 mg/g** in severely infected samples. Total chlorophyll content decreased from **1.499 mg/g** in healthy samples to **0.612 mg/g** in the most affected leaves.

These reductions indicate a clear physiological response to **Cherry Leaf Roll Virus (CLRV)** infection, particularly a degradation of photosynthetic pigments essential for plant metabolism.

The quantitative assessment of pigment content under different CLRV infection levels confirms a consistent trend of degradation in photosynthetic pigments. As detailed in Table 1, chlorophyll b showed the steepest 18 decline (~79.3%), reflecting its location within light-harvesting complex II (LHCII), which is highly vulnerable to virus-induced oxidative stress [1, 5]. The degradation of chlorophyll a and total chlorophyll was less drastic but still biologically significant.

reduction The marked in chlorophyll b relative to chlorophyll a may suggest selective destabilization of LHCII antenna complexes, particularly the CP29 and CP26 proteins, which are chlorophyll b-binding subunits sensitive to oxidative damage and membrane disintegration [5, 6]. Additionally, the reduction in carotenoid content (data not shown) would also compromise photoprotection, making chloroplasts more vulnerable to photoinhibition and ROS-mediated injury.

From a physiological perspective, the decline in chlorophyll pigments likely mirrors underlying disruptions in chloroplast ultrastructure, including granal stacking and envelope membrane integrity. Studies using electron microscopy have confirmed such alterations in virus-infected tissues [6]. Moreover, stress-induced downregulation of genes such as *CHLH* (encoding Mg-chelatase H subunit) and *CAO* (chlorophyllide a oxygenase), essential for chlorophyll biosynthesis, may further contribute to pigment loss [2, 8].

The observed pigment depletion supports the hypothesis that CLRV infection triggers a systemic alteration in source-sink relationships. Virus-infected cells often reprogram their metabolism to favor viral replication, deprioritizing anabolic processes such as photosynthesis. This metabolic shift may be further amplified by hormonal changes, especially increased abscisic acid (ABA) levels that reduce stomatal aperture and carbon assimilation [10, 13].

The data obtained in this study **CLRV** confirm that infection significantly impairs chlorophyll biosynthesis and stability in cherry leaves. The reduction in both chlorophyll a and b suggests that the virus disrupts chloroplast structure and function, resulting in decreased light absorption efficiency and suppressed photosynthetic capacity.

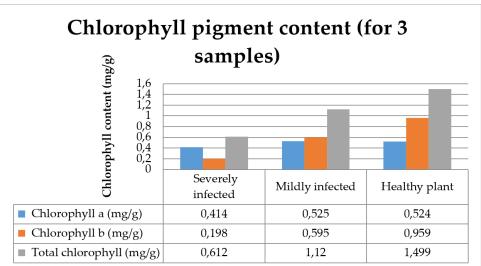


Fig.1. Graph illustrating variations in chlorophyll *a* and *b* levels.

Chlorophyll b was more severely affected than chlorophyll a, consistent with previous studies such as Bilal et al. (2022), which reported that CLRV specifically disrupts light-harvesting complex II, where chlorophyll b is predominantly located. The near 80% reduction in chlorophyll b content in severely infected samples illustrates the virus's profound effect on the plant's photosynthetic machinery. These findings are in agreement with those of Lichtenthaler (1987) and Arnon (1949), who emphasized that chlorophyll content serves as a sensitive indicator of plant stress, particularly under viral attack. The progressive reduction in chlorophyll with increasing disease severity further supports the use of pigment quantification as an early diagnostic tool in virus-infected orchards. Moreover, the application of spectrophotometric analysis provides a reliable framework for understanding the physiological impact of CLRV. This dual approach allows for both pathogen confirmation and the assessment of its functional consequences in the host plant.

Overall, the sharp decline in chlorophyll levels, especially under severe infection, indicates that CLRV exerts strong

physiological pressure on cherry trees, ultimately compromising their productivity. Monitoring chlorophyll content may serve not only as a physiological indicator of plant health but also as a complementary method in the molecular surveillance of CLRV infections in Prunus species.

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# COMPARATIVE ANALYSIS OF THE AMINO ACID COMPOSITION OF THE COAT PROTEIN OF *PLUM POX VIRUS* ISOLATES COLLECTED FROM THE ECOLOGICAL CONDITIONS OF TASHKENT REGION M.S. Sattorov, V.B. Fayziev

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*Summary.* This article focuses on the comparative analysis of the amino acid composition of the coat protein of isolates identified as the D strain of Plum Pox Virus. The coat protein (CP) gene is considered one of the essential structural components of the virus. In a simplified representation, the virus particle can be envisioned as a nucleic acid encased by a protein shell. To investigate this issue, TT-PCR analysis, nucleotide polymorphism in genomic regions, and comparative analysis of nucleotide sequences with closely related isolates are discussed.

*Keywords*: Plum, virus, pox, amino acids, nucleotides, comparative, strain, capsid, gene, isolate.

# СРАВНИТЕЛЬНЫЙ АНАЛИЗ АМИНОКИСЛОТНОГО СОСТАВА ОБОЛОЧЕЧНОГО БЕЛКА ИЗОЛЯТОВ ВИРУСА ОСПЫРЫ, ПОЛУЧЕННЫХ ИЗ ЭКОЛОГИЧЕСКИХ УСЛОВИЙ ТАШКЕНТСКОЙ ОБЛАСТИ

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Аннотация. В данной статье основное внимание уделяется сравнительному аминокислотного состава белка оболочки анализу ИЗОЛЯТОВ, идентифицированных как штамм D вируса оспы. Ген белка шерсти (СР) считается структурных компонентов вируса. В упрощенном ОДНИМ ИЗ важных представлении вирусную частицу можно представить как нуклеиновую кислоту, покрытую белковой оболочкой. Для исследования данного вопроса обсуждаются ТТ-ПЦР-анализ, нуклеотидный полиморфизм в областях генома и сравнительный анализ нуклеотидных последовательностей с близкородственными изолятами.

*Ключевые слова*: Слива, вирус, оспа, аминокислоты, нуклеотиды, сравнительный, штамм, капсид, ген, изолят.

#### Introduction

Plum pox virus is classified among highly pathogenic plant viruses and is considered an RNA-containing virus. It is transmitted mechanically from infected plants to healthy ones [3]. Alongside other viruses, it causes significant damage to agricultural crops. Developing plum pox virus-resistant cultivars is considered the most economical and effective method of combating this disease. However, the development of such cultivars is timeconsuming, and a single amino acid mutation in the viral genome can lead to the emergence of new strains [4].

As mentioned above, the viral coat is referred to as the "capsid," and its subunits constituent are called capsomers or morphological subunits that form the capsid [1, 4]. The entire particle is known as the virus nucleocapsid. In simple viruses, such as the tobacco mosaic virus, the protein layer of the virus consists of a single type of polypeptide chain with a uniform structure. The coat protein is the important one of structural components of the virus, and in a simplified form, the virus particle can be envisioned as a shell encasing the nucleic acid [4, 7].

In complex viruses (such as Teven bacteriophages), where dozens of proteins may be present, determining the complete amino acid composition becomes more challenging due to the heterogeneity of the proteins. To address this issue, TT-PCR analysis was conducted, which is based on identifying nucleotide polymorphisms in genomic regions [6,8], particularly those responsible for the synthesis of the coat protein (CP), or on detecting a single nucleotide polymorphism in the gene encoding the coat protein [2, 9].

The PCR method allows for the selective amplification of a specific segment of DNA millions of times within a few hours. The specificity of this method lies in the use of synthetic oligonucleotide primers, which amplify the nucleotide sequence of interest. With 22

the help of PCR, it is possible to detect even minute quantities of pathogens present in soil, plant tissues, plant sap, or other extracts [2, 8].

Purified nucleic acid, undamaged cells or tissues, or their mixtures are used as samples. The material analyzed may include not only DNA but also RNA molecules, as many plant viruses are RNA-containing viruses and serve as carriers of genetic information [11]. Theoretically, PCR is sensitive enough to detect even a single molecule of nucleic acid from the target object in the sample. Undoubtedly, to amplify the desired region of nucleic acid, at least minimal knowledge of the nucleotide sequence is necessary to synthesize appropriate primers for the process. The use of two different primers increases the specificity and sensitivity of the reaction [5, 12].

Each newly synthesized DNA molecule serves as a template for synthesizing additional DNA using a different primer. To achieve this, it is necessary to denature the DNA molecules produced in the first stage of the reaction, allow the primers to bind to the DNA, and then perform elongation [13]. These three operations constitute the PCR cycle and result in the doubling of the DNA quantity in the sample.

The TT-PCR method has begun to be used in many countries to detect new strains and recombinants of Plum Pox Virus. While indicator plants and serological methods allow for the identification of plum pox virus strains, it has been found that these methods are insufficient for distinguishing differences between new strains and complex recombinants of the virus [4, 6]. For example, plum pox virus and its strains cannot be differentiated using IID or indicator plants, as they share the same serological properties and, in some cases, exhibit identical phenotypic symptoms in indicator plants [7, 8, 9].

A multiplex TT-PCR system was developed based on the participation of several primer pairs in the reaction, enabling the detection of viruses in multiple samples simultaneously [5]. Furthermore, the multiplex TT-PCR system is used to determine the strain composition of Plum Pox Virus [10].

In this case, the composition of each coat protein is analyzed. Therefore, in the following analysis, the amino acid composition of the coat protein of Plum Pox Virus isolates MT038048 Uz1, MT038050 Uz2, and MT038049 Uz3, collected from the ecological conditions of Tashkent region, was compared with several other isolates [10].

#### Materials and methods

Several methods have been developed to determine the amino acid composition of proteins, which differ in terms of the equipment used and the level of complexity [10]. In determining the amino acid composition of the viral coat protein, the nucleotide sequence of the gene responsible for the synthesis of this coat protein was sequenced, and this method is outlined below [3].

For the diagnosis of Plum Pox Virus using polymerase chain reaction cDNA synthesized (PCR), through reverse transcription was used in the PCR mixture (for 1 reaction): 16.5 µL MilliQ water, 2.5 µL 10x buffer for Taq DNA polymerase (Evrogen), 0.5 µL of 10 mM dNTP mixture, 2 µL of 25 mM MgCl2; 0.5 µL each of forward and reverse primers (10 pM/ $\mu$ L), and 0.5  $\mu$ L DNA Taq polymerase (PK113L, Evrogen) were added. To prevent evaporation, mineral oil was added on top.

The PCR amplification was carried out using the "Tersik" PCR amplifier (DNA-Technology, Russia). The PCR conditions included denaturation of the sample (94°C, 3 minutes), amplification for 30-35 cycles, strand reannealing at 72°C for 5-10 and storage at 4°C. For minutes, sequencing, the PCR product was obtained using an analogous method, but the reaction was performed in a 50 μL volume, and the required components calculated were accordingly and mixed. For precise replication, Encyclo DNA polymerase (Evrogen) was used. The annealing temperature of the primers and the duration of the elongation process were determined by the characteristics of the primers and the length of the PCR product being synthesized [11, 13].

The PCR products were analyzed by electrophoresis on an agarose gel prepared in 1x tris-acetate buffer (TAE, Thermo Scientific) and stained with ethidium bromide. To 10 µL of the sample, 3 µL of dye (6x DNA loading dye, Fermentas) was added, and the mixture was loaded into the agarose well. DNA markers of known sizes (GeneRuler Plus 100 bp DNA ladder or GeneRuler Plus 1 kb DNA ladder, Fermentas) were used as standards. Electrophoresis was performed at 80V for 40-60 minutes using a horizontal electrophoresis system SE-1 (Xelikon, Russia). The gel was analyzed under a transilluminator TFP-M/WL (Vilber Lourmat, France) at a wavelength of 312 nm, and images were captured using a gel documentation system MultiDoc-It (UVP, UK) [1, 3, 5, 8].

Determining the Genome 3' End Sequence of PPV. The PCR product intended for sequencing was separated 1-2% agarose gel. After on а electrophoresis, the gel was illuminated using a transilluminator (TFP-M/WL), and the required region was cut out with a scalpel, then transferred to a 1.5 mL centrifuge tube. The DNA was extracted from the agarose using a spin column (Cleanup Standard, cat. No. Evrogen) BC022, according to the manufacturer's instructions. The purified DNA was sequenced using the Sanger method at Evrogen with forward and reverse primers. The sequences of the primers used are provided in the table.

Table 1

Primers	5'-3' Sequence	Number of Nucleotides
Forward (F)	CTTGAATGGGACAGATCAAATGA	23
Reverse (R)	GAGAAAAGGATGCTAACAGGAATC	24

The sequence of the PCR products was determined based on the sequencing results and analyzed using the EditSeq and MegAlign programs (DNASTAR Lasergene, USA).

NucleotideandAminoAcidSequenceAnalysis.Thenucleotidesequences and the corresponding aminoacidsequenceswerecomparedtheClustalWv.2.1program

(<u>http://clustalw.ddbj.nig.ac.jp</u>) or the version available in the BioEdit package.

In the conducted studies, the amino acid composition of the coat protein of the identified isolates was analyzed using FASTA bioinformatics software and compared with a closely related isolate from Kazakhstan [7, 8, 10].

#### **Results and discussion**

Today, Plum Pox Virus is widespread worldwide and causes several issues in the cultivation of stone fruit plants and the harvesting of their products. Molecular genetic identification, sequencing, and bioinformatics analyses for early detection of the virus are of significant practical importance in determining the amino acid composition of the virus.

Molecular genetic identification of the virus coat protein (CP) (Cter) NIb-CP-3'-NCR genomic region was performed using the specially designed primer NIbF/4CPR1. Based on the sequencing results, three isolates were identified, which differ in molecular genetic characteristics from those of the Uzbekistan climate conditions.

These isolates were named PPV Uz1, Uz2, and Uz3, and registered in the International GenBank - NCBI with ID numbers MT038048.1 (Uz1), MT038050.1 (Uz2), and MT038049.1 (Uz3).

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NGMEPICASMVEAWGYKELLREIRKFYSWVLEQAPYNALSKDGKAPYIAETALKKLYTDT
   MT038050.1
               -----MVEAWGYKELLREIRKFYSWVLEQAPYNALSKDGKAPYIAETALKKLYTDT
   MT038049.1
   MT038048.1
               -RMEAICASMVEAWGYKELLREIRKFYSWVLEQAPYNALSKDGKAPYIAETALKKLYTDT
   LT600780.1 8308-9610
RLE<mark>A</mark>ICASMVEAWGYKELLREIRKFYSWVLEQAPYNALSKDGKAPYIAETALKKLYTDT
             *****
               EASETEIERYLEAFYNDINDDGESNVVVHQADEREDEEEVDAGKPIVVTAPAATSPILQP
   MT038050.1
               EASETEIERYLEAFYNDINDDDESNVVVHQADEREDEEEVDAGKPIVVTAPAATSPILQP
   MT038049.1
               EASETEIERYLEAFYNDINDDGESNVVVHQADEREDEEEVDAGKPIVVTAPAATSPILQP
   MT038048.1
   LT600780.1_8308-9610
EASETEIERYLEAFYNDINDDGESNVVVHQADEREDEEEVDAGKPIVVTAPAATSPILQP
           ***********
               PPVIQPAPRTTAPMFNPIFTPATTQPATKPVSQVSGPQLQTFGTHGNEDASPSNSNALVN
   MT038050.1
   MT038049.1
               PPVIQPAPRTTAPMFNPIFTPATTQPATKPVSQVSGPQLQTFGTHGNEDASPSNSNALVN
               PPVIQPAPRTTAPMFNPIFTPATTQPATKPVSQVSGPQLQTFGTHGNEDASPSNSNALVN
   MT038048.1
   LT600780.1 8308-9610
PPVIQPAPRTTAPMFNPIFTPATTQPATKPVSQVSGPQLQTFGTHGNEDASPSNSNALVN
           *****
               TNRDRDIDAGSIGTFTVPRLKAMISKLSLPKVKGKAIMNLNHLAHYSPAQVDLSNTRAPQ
   MT038050.1
               TNRDRDIDAGSIGTFTVPRLKAMTSKLSLPKVKGKAIMNLNHLAHYSPAQVDLSNTRAPQ
   MT038049.1
   MT038048.1
               TNRDRDVDAGSIGTFTVPRLKAMTSKLSLPKVKGKAIMNLNHLAHYSPAQVDLSNTRAPQ
   LT600780.1 8308-9610
TNRDRDVDAGSIGTFTVPRLKAMTSKLSLPKVKGKAIMNLNHLAHYSPAQVDLSNTRAPQ
           *******
   MT038050.1
SCFOTWYEGVKRDYDVTDDEMSIILNGLMVWCIENGTSPNINGMWVMMDGETOVEYPIKP
               SCFQTWYEGVKRDYDVTDDEMSIILNGLMVWCIENGTSPNINGMWVLMDGETQVEYPIKP
   MT038049.1
   MT038048.1
SCFQTWYEGVKRDYDVTDDEMSIILNGLMVWCIENGTSPNINGMWVMMDGETQVEYPIKP
   LT600780.1 8308-9610
SCFQTWYEGVKRDYDVTDDEMSIILNGLMVWCIENGTSPNINGMWVMMDGETQVEYPIKP
           ***********
   MT038050.1
               LLDHAKPTFRQIMAHFSNVAEAYIEKRNYEKAYMPRYGIQRNLTDYSLARYAFDFYEMTS
               LLDHAKPTFRQIMAHFSNVAEAYIEKRNYEKAYMPRYGIQRNLTDYSLARYAFDFYEMTS
   MT038049.1
   MT038048.1
               LLDHAKPTFRQIMAHFSNVAEAYIEKRNYEKAYMPRYGIQRNLTDYSLARYAFDFYEMTS
   LT600780.1_8308-9610
LLDHAKPTFROIMAHFSNVAEAYIEKRNYEKAYMPRYGIORNLTDYSLARYAFDFYEMTS
           ******
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MT038050.1
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TTPVRAREAHIQMKAAALRNVQNRLFGLDGNVGTQEEDTERHTAGDVNRNMHNLLGVRGV
MT038049.1

TTPVRAREAHIQMKAAALRNVQNRLFGLDGNVGTQEEDTERHTAGDVNRNMHNLLGVRGV MT038048.1

TTPVRAREAHIQMKAAALRNVQNRLFGLDGNVGTQEEDTERHTAGDVNRNMHNLLGVRGV LT600780.1\_8308-9610

TTPVRAREAHIQMKAAALRNVQNRLFGLDGNVGTQEEDTERHTAGDVNRNMHNLLGVRGV

\*\*\*\*\*\*\*\*\*\*\*\*\*\*\*\*\*\*

CLUSTAL 2.1 multiple sequence alignment

#### Fig. 1. Comparative analysis of the amino acid composition of the coat protein of PPV isolates

Based on the results of this study, a comparative analysis of the amino acid composition of the coat protein of PPV isolates was conducted. According to the above-mentioned results, it is important to study the effect of observed changes in the nucleotide sequence on the composition of amino acids in the protein. Therefore, in subsequent studies, the amino acid composition of the coat protein of the identified isolates was determined by comparing it with the closely related Kazakhstan isolate (LT600780.1) through a comparative analysis (Figure 1).

А phylogenetic tree was constructed based on the coat protein gene of the isolates, and their relatedness to other isolates was determined. As a result, the Uzb1 (MT038048.1) isolate showed 99.23% homology with LT600780.1 (Kazakhstan), the Uzb2 (MT038050.1) isolate showed 99.31% homology, the LT600781.1 (Kazakhstan) isolate showed 99.16% homology, Uzb3 the (MT038049.1) isolate showed 99.14% homology, the LT600782.1 (Kazakhstan) isolate showed 99.08% homology, and

the GU461890.1 (Slovakia) isolate 99.00% showed homology. This suggests that these isolates share a common ancestor. Additionally, а comparative nucleotide analysis of the Uzbekistan's isolates identified in climate conditions and the closest Kazakhstan isolate (LT600780.1) using the FASTA program revealed the presence of 14 mutation sites, which, in turn, necessitates the study of the amino acid composition of the coat protein.

**Bioinformatics** analysis conducted on the amino acid the virus isolates composition of revealed that out of the 14 mutation observed in the nucleotide sites sequence, 4 changes were found in the protein amino acid composition. Specifically, at the first site, the Uz3 isolate contained Aspartate (D), while the remaining isolates had Glycine (G). At the second site, Valine (V) was present in the Uz1 and Kazakhstan isolates, while Isoleucine (I) was found in the Uz2 and Uz3 isolates.

At the third site, the Uz2 isolate contained Isoleucine (I), while the remaining Uz1, Uz3, and Kazakhstan isolates had Threonine (T). At the fourth site, the Uz2, Uz1, and Kazakhstan isolates contained

In general, based on the results of this study, the following conclusion can be drawn: the Uz1, Uz2, and Uz3 isolates, isolated in our country, as well as the closely related Kazakhstan isolate, were studied in terms of the amino acid composition of the protein shell. When analyzing the amino acid composition of the virus isolates, it was found that out Methionine (M), while the remaining Uz3 isolate had Leucine (L).

#### Conclusion

of points 14 mutation in the nucleotide composition, 4 changes were observed in the protein amino acid composition. Therefore, based on the results of the comparative analyses, it was concluded that while the isolates were similar, they were identical to the previously not identified isolates.

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## IDENTIFICATION OF SYMPTOMS OF POTATO VIRUS Y (PVY) IN INDICATOR PLANTS

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Summary. The issue of identifying the symptoms of Potato Virus Y (PVY) in indicator plants is of great importance for the successful cultivation of potato crops and for maximizing their yield in agriculture. Potato crops are widely distributed worldwide as a major source of food, and their production plays a key role in the economies of many countries. However, potato viruses especially Potato Virus Y pose a significant threat to these crops. This virus contributes to the spread of viral diseases in various plants and crops, including potatoes, leading to a decline in both the quality and quantity of the yield. Potato Virus Y (PVY) is highly widespread among potato plants and causes disease that manifests through various symptoms. Detecting the virus in potato plants particularly identifying viral infections at early stages is critically important for preventing the spread of infection. The symptoms caused by the virus lead to numerous physiological changes in the plants, including mosaic patterns on the leaves, chlorosis at leaf tips, vein deformation, darkening or rotting of tissues, and stunted growth. These effects, in turn, significantly reduce crop yield. This article discusses Potato Virus Y (PVY), its characteristics, and the extent of its spread in regions of Tashkent province, Republic of Uzbekistan. It also explores the cultivation of indicator plants for PVY, identification of disease symptoms in those plants, and the main manifestations of the virus, as well as the physiological effects it causes in host plants. Various methods were employed for this purpose, including visual observation and laboratory techniques. As a result of the conducted studies, mechanical inoculation of the virus was carried out on indicator plants such as Datura metel, Nicotiana glutinosa, Nicotiana samsun, Nicotiana virginiana, Nicotiana barley, Dahlia, Chenopodium amaranticolor, Physalis, and Solanum nigrum. It was found that disease symptoms appeared in some plants within 7-8 days, while in others symptoms were mainly observed after 12–14 days.

*Keywords*: virus, Potato Virus Y, indicator plants, pathogen, symptom, isolation, infection, inoculation, suspension, diagnostics, RT-PCR.

# ИДЕНТИФИКАЦИЯ СИМПТОМОВ Ү ВИРУСА КАРТОФЕЛЯ (ҮВК) У ИНДИКАТОРНЫХ РАСТЕНИЙ

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Аннотация. Выявления и анализ симптомов Y вируса картофеля (YBK) у растений-индикаторов имеет большое значение для успешного выращивания картофельных культур и для максимизации их урожайности в сельском хозяйстве. Картофельные культуры широко распространены во всем мире как основной источник продовольствия, а его производство играет ключевую роль в экономике многих стран. Однако вирусы картофеля, особенно Y вирус картофеля, представляют значительную угрозу для этих культур. Этот вирус способствует распространению вирусных заболеваний различных растений и культур, включая картофель, что приводит к снижению как качества, так и количества урожая.

У вирус картофеля широко распространен среди растений картофеля и вызывает заболевание, которое проявляется различными симптомами. Выявление вируса у растений картофеля, особенно выявление вирусных инфекций на ранних стадиях, имеет решающее значение для предотвращения распространения инфекции. Симптомы, вызываемые вирусом, приводят к многочисленным физиологическим изменениям в растениях, включая мозаичные узоры на листьях, хлороз на кончиках листьев, деформацию жилок, потемнение или гниение тканей и задержку роста. Эти эффекты, в свою очередь, значительно снижают урожайность.

В этой статье обсуждается Y вирус картофеля, его характеристики и степень его распространения в регионах Ташкентской области Республики Узбекистан. В ней также исследуется выращивание растений-индикаторов YBK, выявление симптомов заболевания у этих растений и основные проявления вируса, а также физиологические эффекты, которые он вызывает у растений-хозяев. Для этой цели применились различные вирусологические методы, включая визуальное наблюдение и лабораторные методы. В результате проведенных исследований была проведена механическая инокуляция вируса на такие растения-индикаторы, как Datura metel, Nicotiana glutinosa, Nicotiana samsun, Nicotiana virginiana, Nicotiana barley, Dahlia, Chenopodium amaranticolor, Physalis и Solanum nigrum. Было установлено, что симптомы заболевания у некоторых растений появлялись в течение 7–8 дней, а у других симптомы в основном наблюдались через 12–14 дней.

*Ключевые слова:* вирус, Y-вирус картофеля, растения-индикаторы, возбудитель, симптом, изоляция, заражение, инокуляция, суспензия, диагностика, ОТ-ПЦР.

#### Introduction

At present, viral diseases are considered one of the main threats to the sustainable development of global agriculture, causing significant economic losses each year. It is well known that many vegetatively propagated food crops, fodder plants, and ornamental species are chronically infected by viruses, which in turn leads to substantial yield losses and a marked decline in the quality of agricultural products [1]. Nearly half of all newly identified infectious plant diseases over the past decade have been of viral origin [2].

Today, various viruses that infect potato crops exist worldwide, among Virus (PVY) which Potato Y is considered the most widespread and one of the most economically damaging. Studying PVY is crucial for the production of high-quality food products, assessing the virus's spread under the climatic conditions of our country, identifying naturally resistant plant strains, and developing effective control measures against the virus.

This issue is highly relevant for the sustainable development of the potato industry and food security in our country, making it necessary to conduct extensive scientific research in this area.

Since gaining independence, systematic measures have been implemented in Uzbekistan to protect plants from various diseases and pests. In this regard, the Republic of Uzbekistan has adopted a number of regulatory documents laws and governing plant quarantine and phytosanitary control. Notably, the Law on Plant Quarantine and related subordinate legislation serve as the primary legal framework for ensuring protection of plants against diseases and pests [12].

These legal regulations play a crucial role in maintaining plant health, preventing the spread of viruses and other pathogens, and implementing necessary preventive and monitoring activities within the country.

Potato Virus Y (PVY) is one of the most dangerous plant viruses transmitted by aphids. This virus significantly damages plants of the Solanaceae family, particularly crops such as potato (Solanum tuberosum), tobacco (Nicotiana tabacum), tomato (Solanum lycopersicum), and pepper (Capsicum annuum). PVY infection can reduce potato yields by 30% to 80%, negatively affecting both the quality and storage life of the crop [3].

Currently, Potato Virus Y is widespread globally and has become a major problem especially in potatogrowing regions of Asia, Europe, South America, and Africa. The spread of this virus causes direct economic losses to potato producers, as productivity declines significantly in infected areas. For instance, in some regions of Europe, PVY infection has led to nearly a 50% reduction in potato yield [4]. Viral infections represent an economic threat to the agricultural industries of many developed and developing countries. Moreover, PVY not only reduces potato quality but can also increase the price of agricultural exports, adversely affecting trade [5].

Potato Virus Y (PVY) is primarily transmitted by aphids, with Myzus being persicae the main vector responsible for spreading the virus to healthy plants. In addition to aphid transmission, the virus can also be spread through mechanical inoculation

of plants. In this method, a suspension prepared from virus-infected plants is rubbed onto the leaves of healthy plants, introducing the infection under laboratory conditions. Mechanical inoculation is used to study the spread and transmission of the virus in controlled environments [6].

Using indicator plants is an effective and rapid method for detecting Potato Virus Y. Plants such as Nicotiana tabacum, Chenopodium amaranticolor, and Datura metel are sensitive to the virus, showing distinct infection symptoms on their leaves, including spots, curling, deformation, chlorosis, and necrosis. These symptoms allow for the identification of the virus. For example, Nicotiana tabacum (tobacco) infected with Potato Virus Y typically develops ringshaped spots and necrotic symptoms on the leaves. Chenopodium amaranticolor exhibits symptoms such as leaf curling, necrotic spots, mosaic patterns, and deformation, while Datura metel leaves show changes such as chlorosis, mosaic, and deformation. One of the main advantages of detecting disease using indicator plants is the method's speed cost-effectiveness. If and infection symptoms are observed on the plants, the virus can be rapidly identified [7].

Molecular diagnostic methods such as ELISA and RT-PCR are also effectively used to detect Potato Virus Y. The ELISA test is widely applied for detecting viral antigens or antibodies, allowing precise confirmation of the virus presence in potato plants. RT-PCR helps identify the virus's genetic material, offering highly accurate and reliable results. When combined with detection using indicator plants, molecular diagnostic techniques significantly enhance the efficiency of virus identification and control of its spread [8].

In modern breeding and seed certification systems, detecting Potato Virus Y is a crucial step in preventing the virus's spread. Developing genetically resistant potato varieties and preventing viral dissemination depend heavily on these methods. Creating genetically resistant potato cultivars can effectively reduce the transmission of Additionally, this virus. advanced biotechnological approaches are being in increasingly used contemporary agriculture for effective virus management [9].

Overall, potato is susceptible to many pathogens and pests, with over fifty viruses and viroids described. Since potatoes are propagated vegetatively, this facilitates the transmission of subsequent generations, viruses to making the crop highly vulnerable to viral diseases [10]. This study aims to inoculate healthy plants mechanically phytopathogenic viruses with and observe changes in growth based on infection symptoms.

#### Materials and methods

To conduct the study, a range of indicator plants was selected to detect phytoviruses in potato plants exhibiting viral disease symptoms. The test indicator plants included species such as Datura metel, Nicotiana glutinosa, Nicotiana tabacum cv. Samsun, Nicotiana verjena, Nicotiana barley, Dahlia spp., Chenopodium amaranticolor, Physalis spp., and Solanum nigrum. These plants serve as biological indicators for the presence and pathogenicity of phytoviruses.

The research was carried out using the laboratory and greenhouse facilities of the Department of Biology, Faculty of Natural Sciences, Chirchiq State Pedagogical University. During the study, potato fields in the Qibray, Yuqori Chirchiq, and Bostanliq districts of Tashkent region were surveyed, and disease symptoms closely matching those caused by Potato Virus Y (PVY) were specifically selected. Leaf samples were collected from these symptomatic plants.

The use of indicator plants for viral disease diagnostics, particularly through mechanical inoculation of the virus in laboratory conditions, is a widely employed method. In this study, mechanical inoculation of Potato Virus Y into indicator plants was performed following generally accepted protocols [13].

Initially, 100 g of leaf samples were accurately weighed using an analytical balance from potato plants infected with PVY. The collected samples were finely chopped using sterilized scissors in a porcelain mortar and then ground mechanically. To stabilize the virus and ensure its transmission, the sample was mixed with 0.01 M phosphate buffer solution (K<sub>3</sub>PO<sub>4</sub>, pH 7.4) in a 1:1 ratio to form a homogenate. The resulting homogenate was filtered through four layers of sterile gauze and collected in sterilized test tubes under aseptic conditions.

The uniformity of the homogenate volume was checked using a precision balance. The homogenate in the test tubes was centrifuged at 4000 rpm for 20 minutes, and the supernatant (the liquid above the sediment) was carefully separated into a clean test tube. The resulting supernatant was prepared for inoculation, while the sediment was discarded.

This method facilitates the transmission of the virus to the indicator plants and subsequently aids in confirming the presence of the virus by observing symptom development.

To infect the test indicator plants with the viral infection, the separately collected supernatant liquid was used as infectious material. The inoculation procedure was carried out on the upper leaf surfaces of several indicator plants susceptible to Potato Virus Y, including *Nicotiana tabacum, Chenopodium amaranticolor,* and *Datura metel,* which were grown under laboratory and greenhouse conditions.

Initially, the experiment was conducted on Nicotiana tabacum cv. *Samsun*. Fine corundum powder (Al<sub>2</sub>O<sub>3</sub>) was gently dusted onto the upper surface of the leaf. Then, three to four drops of viral sap were applied to the leaf surface using a sterile dispenser. Using clean, washed, and air-dried fingers, the viral sap was carefully rubbed onto the leaf surface. This process created micro-wounds on the leaf epidermis, facilitating the entry of viral particles into the mesophyll tissue. After inoculation, each plant was tagged with labels indicating the sample name,

inoculation date, and other relevant information. Approximately two hours later, excess  $Al_2O_3$  particles and viral liquid were washed off from the leaf surface with distilled water.

The test indicator plants inoculated with viral sap were maintained for 24 hours under cool, low-light conditions to minimize stress effects. Following mechanical inoculation, the characteristic symptoms of the virus were visually monitored within a time frame ranging from 2-3 days up to 35-40 days, depending on the plant species [56; Figures 9–14].

#### **Results and discussion**

The symptomatology and phenotypic characteristics of Potato Virus Y vary among different plant species belonging to the Solanaceae family. According to the literature, the virus can induce a broad spectrum of clinical signs from asymptomatic infections to mosaic patterns, necrosis, and even plant death depending on the plant species, cultivars, viral strains, and isolation conditions [11].

During the research, potato fields in the Qibray, Yuqori Chirchiq, and Bo'stonliq districts of Tashkent region were selected as study sites to investigate the spread of Potato Y virus (PYV).

Initial visual observations were conducted in the potato fields of Qibray district, with the research process subsequently continued in the Yuqori Chirchiq and Bo'stonliq districts. Throughout these observations, 34 particular attention was paid to the characteristic symptoms of Potato Y virus infection in potato plants, based on information derived from scientific literature.

As a result of these observations, distinctive disease symptoms were identified in the potato fields. Leaf samples from various potato plants exhibiting these symptoms were collected and transported the to further study. laboratory for The analysis of the spread of Potato Y virus and its effects on the growth of potato plants provided deeper insight into the disease's distribution.

Furthermore, the analysis and observations revealed variability in the indicators of Potato Y virus infection across the different districts, with distinct symptoms and infection patterns recorded for each district. The outcomes of this study hold significant importance for the development of effective strategies to prevent the spread of the disease and to manage potato cultivation more efficiently. Below (Figure 1), different potato plants exhibiting symptoms characteristic of Potato Y virus infection observed in the fields are presented.

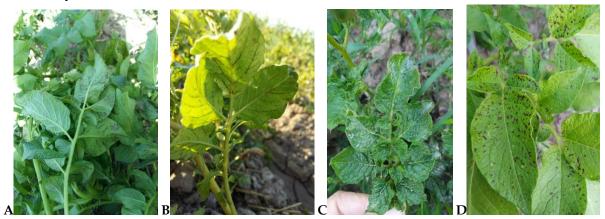


Fig. 1. Characteristic disease symptoms caused by the virus observed in potato plants. In the image: A, B – darkening of veins on the underside of the leaves; C – leaf curling; D – mottled mosaic pattern on the leaves.

The incubation period of Potato Y virus in *Nicotiana tabacum cv. Samsun* typically ranges from 7 to 14 days. During this period, symptoms such as vein constriction, whitening of the young terminal leaves, and yellow mosaic patterns on the leaf surface develop. Disease symptoms significantly intensify between 24 and 40 days, becoming clearly defined and characteristic.

During the study, samples showing distinct disease symptoms were taken from *Nicotiana tabacum cv*. Samsun mechanically plants and inoculated onto Nicotiana glutinosa plants. Within 7 to 15 days postinoculation, general chlorosis (light vellowing) of the leaves was observed. Between 20 and 40 days, systemic and distinct mosaic symptoms became clearly visible on the plants. To confirm the accuracy of the study, the presence of Potato Y virus in the infected Nicotiana glutinosa plants was verified using the immunostrip (immunoassay) method, which yielded a positive result (Figure 2).



## Fig. 2. Test results of Nicotiana glutinosa plant analyzed using immunostrip assay.

To continue the research, samples taken from the infected *Nicotiana glutinosa* plants were mechanically inoculated onto various test-indicator plants, including Datura metel, Nicotiana samsun, Nicotiana verjenya, Nicotiana barley, Dahlia, Chenopodium amaranticolor, Physalis, and Solanum nigrum (Figure 3).



Fig. 3. Newly inoculated test-indicator plants with Potato Y virus 1 - Physalis; 2 - Chenopodium amaranticolor

The appearance and development of disease symptoms in these plants were regularly monitored. Below are several test-indicator plants inoculated with Potato Y virus that exhibited clear disease symptoms (Figure 4).





Fig. 4. A series of test-indicator plants inoculated with Potato Y virus, exhibiting distinct disease symptoms: 1-N.barley; 2-N.glutinosa; 3-Physalis; 4,6,12-N.samsun; 5-Ch.amaranticolor; 7-Dahlia; 8-N.verjenya; 9-D.metel;10-Lactuca serriola; 11-Artemisia vulgaris.

Studies on the effectiveness of using test-indicator plants for the detection of phytopathogenic viruses have shown that early-stage identification of viral infections plays a

# Conclusion

Based on the results of the conducted scientific research and the analysis of existing literature, it was determined that Potato Y virus (PYV) can cause yield losses ranging from 10% to 80% in plants. Field observations carried out within the scope of this study in the Qibray district of Tashkent region revealed a significantly high proportion of infected potato plants. crucial role in preventing the spread of infection. This significance has been widely noted in numerous scientific sources [Fayziyev 2020; Jovliyeva 2023].

This indicates the rapid regional spread of the virus and underscores its recognition as a critical phytopathological issue for the agricultural sector.

Considering the damage caused by Potato Y virus to plants and its economic importance, there is a need for an in-depth study of the virus's biological characteristics, factors influencing its spread, and its interactions with ecological factors. At the same time, the use of indicator plants for early-stage detection of viral infection is of great significance. During this research, test-indicator plants such as Datura metel, Nicotiana glutinosa, Nicotiana tabacum (Samsun, Barley, Verjenva varieties), Chenopodium amaranticolor, Dahlia spp., Physalis spp.,

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Therefore, to prevent the widespread dissemination of Potato Y virus, reduce its negative impact on yield, and establish an effective monitoring system, it is urgent to develop integrated approaches based on the use of indicator plants.

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# SOME PHYSIOLOGICAL AND BIOCHEMICAL PROPERTIES OF FOREIGN AND DOMESTIC SAMPLES OF ARACHIS HYPOGAEA L.

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*Summary:* The article presents the results of a scientific study conducted to study some physiological and biochemical characteristics of foreign and local samples of peanut Arachis hypogaea L. It was found that the total water content in leaves and the transpiration rate differed in experimental variants of different varieties and collection varieties of peanuts, both in the control and during pre-sowing seed treatment with biopreparations. As a result of the studies, it was found that foreign and local samples of peanut Arachis hypogaea L. differ from each other in the total protein content in seeds.

*Keywords:* Peanut, Arachis hypogaea L., foreign and domestic samples, total leaf water content, transpiration rate, total protein content, biopreparations.

# НЕКОТОРЫЕ ФИЗИОЛОГИЧЕСКИЕ И БИОХИМИЧЕСКИЕ СВОЙСТВА ЗАРУБЕЖНЫХ И ОТЕЧЕСТВЕННЫХ ОБРАЗЦОВ ARACHIS HYPOGAEA L. У.А. Уразбаева<sup>1</sup>, И.Дж. Курбанбаев<sup>2</sup>.

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Аннотация. В статье представлены результаты научного исследования, проведенного изучения некоторых физиолого-биохимических С целью особенностей зарубежных и местных образцов арахиса Arachis hypogaea L. Установлено, ЧТО общее содержание воды в листьях и интенсивность транспирации различались в опытных вариантах разных сортов и коллекционных сортов арахиса, как в контроле, так и при предпосевной обработке семян биопрепаратами. В результате проведенных исследований установлено, что зарубежные и местные образцы арахиса Arachis hypogaea L. отличаются друг от друга по общему содержанию белка в семенах.

Ключевые слова: Apaxuc, Arachis hypogaea L., зарубежные и отечественные образцы, общая оводненность листа, интенсивность транспирации, общее содержание белка, биопрепараты.

## Introduction

The common peanut (Arachis hypogaea L.) or peanut is an annual herbaceous plant belonging to the legume family (*Fabaceae* or *Leguminosae*). The peanut is native to South America (Argentina and Bolivia). Peanuts are cultivated in approximately 120 countries, with a total area of 24.6 million hectares worldwide, and world production is 38.2 million tons. Peanut seeds contain 40-60% oil and 20-37% protein [1].

Peanut (Chinese peanut) is a valuable oilseed and food crop. Today, increasing the yield and seed quality of oilseed crops, including peanut (Arachis hypogaea L.), is of great importance in meeting the world's food needs. According to world statistics for 2018, Uzbekistan ranks 51st in the world in peanut production and 1st in productivity. The largest production is accounted for by India, China, and the United States, and 60-80% of the produced product is processed in these countries. The highest yields in the world are recorded in China (2.5-3.0 t/ha) and the United States (3.0-4.0 t/ha). In our republic, this indicator is on average 1.5-1.8 t/ha when cultivated as a repeated crop, and 3.5-4.0 t/ha when grown as a main crop on advanced farms [2].

The study of some physiological and biochemical characteristics of the peanut plant collection is of great importance, allowing the collection samples to be evaluated for their positive character traits. in peanut (*Arachis hypogaea* L.), the transpiration rate and total water content in the leaves directly affect the water use efficiency, drought tolerance, and productivity of the plant. transpiration is the process of evaporation of water from the leaves of 40 a plant, which not only cools the plant, but also helps to transport nutrients from the roots to the leaves. the transpiration rate in peanut varies genotype depending on the and environmental conditions. for example, some varieties close the stomata of the leaves early in drought conditions, reducing water loss, which helps to save water [3]. Relative water content (rwc) in leaves reflects the water status of a plant. high relative water content is essential for photosynthesis, cell turgor, and growth processes. peanut has a high relative water retention capacity under drought conditions compared to other crops, which increases its drought tolerance [4].

M.K. Mahatma et al., (2016) compared the nutritional composition and antioxidant properties of peanut samples from Spain and Virginia. More than 60 peanut genotypes belonging to the Spanish (30) and Virginia (30) groups were characterized by the content of protein, fat, phenols and antioxidant activity in the seeds, as well as the content of fatty acids and sugars. The antioxidant activity of Virginia genotypes was found to be from 12.5 to 16.5  $\mu$ M, while in Spanish genotypes this indicator was from 6.8 to 15.2  $\mu$ M. Among the Virginia genotypes, the highest content of oleic/linolenic acids was observed in sample NRCG 12312 and amounted to 2.38, while in sample NRCG 12731 belonging to the Spanish group this indicator was 1.24. The sugar content in the Virginia group ranged

from 38.5 to 69.0 mg/g, while the Spanish genotypes ranged from 27.9 to 53.3 mg/g. The authors concluded that the Virginia peanut genotypes were more effective in terms of antioxidant activity and fat content than the Spanish genotypes [5].

In Uzbekistan, many studies have been conducted to study the morphological, economic and valuable characteristics of peanuts. However, scientific studies on the study of its physiological and biochemical properties are insufficient. In this regard, in the Laboratory of Genetics, Breeding and Seed Production of

The research was conducted at the field experimental site of the Durmon Scientific and Experimental Station of the Institute of Genetics and Experimental Biology of Plants of the Academy of Sciences of Uzbekistan. The following 10 varieties from the collection brought from various foreign countries were used in the experiments in the laboratory of the Institute "Genetics. selection and seed production of legumes, oilseeds and medicinal plants": Uganda Erect SB 33 (Uganda); Virginia Improved (Israel); Zac Trang (Vietnam); Philippin Pink (Portugal); Namuno (Portugal); R 30 (Israel); Var Cuba (Japan); Dessertny (Russia); PL #1 (India) and Hippagi 2-20 (India). In addition, the local varieties of Polvon (a variety peanut called "Polvon"), Kibray, Lider, Mumtoz and

Legumes, Oilseeds and Medicinal Plants of the Institute of Genetics and Experimental Biology of Plants of the Academy of Sciences of Uzbekistan, some physiological and biochemical properties of peanuts of foreign and local varieties, including: transpiration rate, total water content in leaves, were analyzed the control of the in experiment, in the variants in which the seeds were treated with Bradyrizobium japonicum sp D 24-1 and Bionitrogen biopreparations before sowing, and scientific studies were conducted to study the total protein content in the seeds.

## Material and methods

Salomat, as well as 5 local samples were used in the scientific research.

After 10 days of the flowering phase, the physiological indicators of water balance were evaluated - leaf water content and transpiration rate. To determine the total water content in the leaves of the plant, 5 leaves were taken from each sample and weighed on an electronic balance and placed in room conditions. After a 2-hour exposure, their weight was measured again. Then, they were dried in a drying cabinet at 100-105 0C for 3 days, the dry weight of the leaves was measured, and thus the total water content in the leaves was determined. The data obtained were statistically analyzed using the method of B.A. Dospekhov [6]. Transpiration activity was determined by L.A. Ivanov [7], and the total water content in the leaf by N.N. Tretyakov et al. [8].

One of the methods for determining the amount of total proteins is the Keldal method. It consists in calculating the amount of total protein by determining the amount of nitrogen. The essence of the method is to hydrolyze the organic substances in with the help the sample of concentrated sulfuric acid (amine the protein) to form groups in sulfate After ammonium salts. the hydrolysis was complete, the ammonium sulfate formed was treated with sodium hydroxide to convert it to ammonia.

The ammonia or ammonium hydroxide sulfate formed as a result of neutralization is dissolved in an acid solution. The remaining acid is titrated with an alkaline solution. The amount of

nitrogen calculated from is the calculated amount of ammonia. An accurate sample is taken from а medium-sized homogeneous sample of the sample under study into a test tube for analysis, the error level should not exceed 0.1%. The sample is quantitatively transferred to a Kjeldahl flask. The subsequent experiment is carried out according to the instructions [9]. Processing of the results obtained: the mass fraction of nitrogen (X) in the analyzed sample is calculated by the formula as a percentage of the sample mass by titrating the volume after the amount of ammonia passed through dilute sulfuric acid.

## **Result and discussion**

The total water content in peanut leaves was found to be different in the control and experimental variants of the study. In the control variant of the experiment, the total water content in the leaf was observed to be from 70.95% to 80.72%. The highest results for this indicator were observed in the samples of the peanut varieties Salomat, 1-local, Uganda Errect (Uganda) and Virginia Improved (Israel), while the lowest indicator was observed in the Qibray variety, with a total water content of 70.95%. In the variant of the experiment, in which the seeds were treated with the Bradyrizobium japonicum sp D 24-1 biopreparation before sowing, the total water content in the leaf was found to be 42

slightly higher than in the control variant and ranged from 70.56% to 84.01%. The highest indicators were observed in the samples of the peanut varieties Salomat, 1-local and R-30 (Israel). In the variant of the experiment, in which the seeds were with treated the Bradyrizobium japonicum sp D 24-1 biopreparation before sowing, low indicators of total leaf water content were observed in the samples of the Kibray and PL#1 (India) varieties, which were 70.56 and 70.64%, respectively. In the variant of the experiment, in which the seeds were with treated the Bionitrogen biopreparation before sowing, no significant difference observed was compared to the control variant, and the

total leaf water content in the studied samples ranged from 69.50% to 82.31%. High indicators for this sign were observed in the samples of the 1st local, Salomat and Hippagi 2-20 (India) varieties, which were 80.46, 81.46 and 82.31%, respectively. The lowest total water content in the leaf was observed in the sample of the Dissertation (Russia) variety, which was 69.50% (Table 1).

## Table 1

		Total water content (%)		
Nº	Sample name	Control	Bradyrizobium japonicum sp D 24-1	Bionitrogen
1	Polvon	73,21	72,95	72,76
2	Qibray	70,95	70,56	71,60
3	Lider	74,32	72,92	73,22
4	Mumtoz	77,27	74,12	75,25
5	Salomat	80,72	81,18	81,46
6	1-mahalliy	80,56	84,01	80,46
7	2-mahalliy	78,35	77,95	77,97
8	4-mahalliy	77,29	74,97	77,95
9	6-mahalliy	77,36	79,54	78,97
10	7-mahalliy	75,31	79,48	75,95
11	Uganda Errect (Uganda)	80,06	73,25	74,30
12	Virginia Improved (Isroil)	80,56	76,76	75,95
13	Zag Trang (Vetnam)	79,27	76,14 75,52	
14	Phillipine Pink (Portugaliya)	76,77	76,91	75,15
15	Namuno (Portugaliya)	79,19	78,72	76,76
16	R-30 (Isroil)	75,52	80,65	78,33
17	Var Cubo (Yaponiya)	79,89	75,75	75,64
18	Dessertniy (Rossiya)	74,81	77,73	69,50
19	PL#1 (Indiya)	74,55	70,64	75,64
20	Hippagi 2-20 (Indiya)	77,43	79,32	82,31

During the research, the transpiration rate of plants in the general flowering phase was analyzed in the control and pre-sowing variants of the field experiment of local varieties of peanuts and foreign and local samples of the collection, in which the seeds were treated with Bradyrizobium japonicum sp D 24-1 and Bionitrogen biopreparations. In the control variant of the experiment, the highest

transpiration rate was observed in the varieties of peanuts Lider, Dessertny (Russia) and Kibray and amounted to 190.48 mg/g, 193.55 mg/g and 196.72 respectively. The lowest mg/g, transpiration rate was observed in the sample Namuno (Portugal) and was determined to be 74.07 mg/g. In the variant of the experiment, in which the with seeds were treated the Bradyrizobium japonicum sp D 24-1

biopreparation before sowing, it was observed that the transpiration rate in plants increased slightly. This can be seen from the fact that the transpiration rate in the peanut variety Leader was 208.33 mg/g. In this variant of the experiment, the lowest transpiration rate was observed in the peanut variety Kibray and amounted to 93.75 mg/g. In the variant of the experiment, in which seeds were treated with the the Bionitrogen biopreparation before sowing, it was found that the transpiration rate in plants increased in one or another variety of samples. In this variant of the experiment, the highest transpiration rate was observed in the Zag Trang (Vietnam) sample and

amounted to 206.9 mg/g. The lowest transpiration rate was observed in the Uganda Errect (Uganda) sample from Uganda and was found to be 65.57 mg/g. During the conducted studies, an increase in the transpiration rate in the experimental variants compared to the control variant of the experiment was observed in the 1st local and R-30 (Isroil) samples from Israel. It was found that the transpiration rate in these varieties was higher in the variants of the experiment in which the seeds were treated with Bradyrizobium japonicum sp D 24-1 and Bionitrogen biopreparations before sowing, compared to the control variant (Table 2).

Table 2

	Sample name	Transpiration rate			
N⁰		Control	Bradyrizobium japonicum sp D 24-1	Bionitrogen	
1	Polvon	107,14	131,15	103,45	
2	Qibray	196,72	93,75	139,53	
3	Lider	190,48	208,33	111,11	
4	Mumtoz	178,57	169,49	142,86	
5	Salomat	163,93	196,72	135,59	
6	1-mahalliy	85,71	115,94	123,08	
7	2-mahalliy	126,98	187,50	161,29	
8	4-mahalliy	142,86	137,93	131,15	
9	6-mahalliy	163,93	163,93	95,24	
10	7-mahalliy	140,85	125,0	93,75	
11	Uganda Errect (Uganda)	153,85	158,73	65,57	
12	Virginia Improved (Isroil)	157,73	137,93	122,45	
13	Zag Trang (Vetnam)	161,29	140,35	206,90	
14	Phillipine Pink (Portugaliya)	140,85	151,52	88,24	
15	Namuno (Portugaliya)	74,07	137,93	101,69	
16	R-30 (Isroil)	136,99	158,73	163,93	
17	Var Cubo (Yaponiya)	121,21	163,93	103,45	
18	Dessertniy (Rossiya)	193,55	190,48	161,29	
19	PL#1 (Indiya)	142,86	98,36	103,45	
20	Hippagi 2-20 (Indiya)	117,65	178,57	88,24	

Transpiration rate of peanut (*Arachis hyupogaea* L.) plant (mg H<sub>2</sub>O/1g wet leaf x 1 hour)

In subsequent studies, the amount of total protein in the seeds of foreign and local samples of the peanut Arachis hypogaea L. collection was determined (Table 3). The data presented show that the foreign and local varieties of the peanut Arachis L. collection hypogaea differ significantly in the total protein content of their seeds. The total protein content

of the studied peanut varieties ranged from 19.50% to 30.38%. In particular, high indicators of total protein content were observed in PL#1 (India) 30.38%, Var Cuba (Japan) 28.13%, Virginia (Israel) 26.88%, while Improved relatively low indicators were observed in R 30 (Israel) 22.44%, Polvon (Uzbekistan) 20.75%, Philippine Pink (Portugal) 19.50% (Table 3).

Table 3

Arachis hypogaea L. collection						
N⁰	Accession name	Total protein content (%)	Difference from control Polvon, ±%			
1.	Uganda Erect Sb 33 (Uganda)	22,63%	+1,88			
2.	Virginia Improved (Israel)	26,88%	+6,13			
3.	Zac Trang (Vietnam)	25,13%	+4,38			
4.	Philippine Pink (Portugal)	19,50%	-1,25			
5.	Namuno (Portugal)	26,06%	+5,31			
6.	R 30 (Israel)	22,44%	+1,69			
7.	Var Cuba (Japan)	28,13%	+7,38			
8.	Dessertny (Russia)	22,56%	+1,81			
9.	PL#1 (India)	30,38%	9,63			
10.	Hippagi 2-20 (India)	23,44%	2,69			
11.	Nazorat Polvon (Uzbekistan)	20,75%	-			
12.	*LSD <sub>0•05</sub>	2.54%	-			

**Note**:  $LSD_{0.05}$  (*Least Significant Difference at* 5% *level*) is a statistical criterion used to determine the minimum difference required between variants to consider the variation statistically significant. If the observed difference between variants exceeds  $LSD_{0.05}$  or  $LSD_{0.01}$ , it is regarded as a true difference.

The peanut varieties studied in terms of protein content can be divided into 4 groups. The first group includes Philippine Pink (Portugal) and Polvon (Uzbekistan), which have a low protein content in their seeds, i.e. 19.50% -20.75%. These varieties, especially Polvon (Uzbekistan), are distinguished by their large seeds. From this point of view, this variety can be used as a

valuable material for creating highyielding peanut varieties in the future.

The second group includes the varieties R 30 (Israel), Dessertny (Russia), Uganda Erect Sb 33 (Uganda) and Hippargi 2-20 (India), whose seed protein content ranges from 22.44% to 23.44%. Most of these varieties are distinguished by their creeping growth. As we know, creeping growth of peanuts leads to an increase in pods

under the soil, which, in turn, increases yield. These varieties can also be widely used in the selection process to identify fertile peanut forms.

According to the results of the protein content, we can include the following varieties rich in protein in groups 3 and 4 of the study: Zac Trang (Vietnam) (25.13%), Namuno (Portugal)

Thus, it was found that the total water content in leaves and transpiration rate of different varieties of peanuts and collection varieties were different in the control and pre-sowing treatment of seeds with biopreparations. It was observed that the total water content in leaves was slightly higher in the treated with variants biopreparations compared to the control variant of the experiment. This indicates that field experiments depend on the amount of irrigation of plants and the geographical origin of peanut varieties, as well as the different reactions of different varieties to biopreparations. biopreparations provide Because intensive supply of biological nitrogen and other compounds, which leads to an acceleration of cell division, tissue formation, and also an increase in the intensity of physiological processes.

The results of statistical analysis showed that the studied peanut

(26.06%), Virginia Improved (Israel) (26.88%), Var Cuba (Japan) (28.13%), PL#1 (India) (30.38%). These varieties showed higher protein content than other studied research sources. In the future, these varieties will play an important role in creating peanut varieties with improved grain quality and high protein content.

## Conclusion

collection samples were superior to the control Polvon variety in terms of oxalic acid content, except for Philippine Pink, introduced from Portugal. In particular, Virginia Improved, Var Cuba and PL#1 collection samples were noted to be 6.13-9.63% more oxalic acid than the control variety.

As a result of the conducted studies, it was found that samples of foreign and local varieties of peanut Arachis hypogaea L. collection differ from each other in terms of the total protein content in the seed. According to the results of the study, samples with high results in terms of the total protein content in the seed composition were isolated. The selected varieties PL#1 (India) 30.38%, Var Cuba (Japan) 28.13%, Virginia Improved (Israel) 26.88% will serve as valuable material for the future creation of high-yielding varieties of peanuts with a high protein content in their seeds

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# IN VITRO EVALUATION OF SALINITY AND DROUGHT STRESS RESISTANCE IN COMMON BEAN (PHASEOLUS VULGARIS L.) VARIETIES

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*Summary.* Drought and soil salinity remain among the most limiting environmental factors affecting agriculture worldwide. An important solution to this problem is the selection of plant species that yield high crops even in saline soil conditions. Common bean (*Phaseolus vulgaris* L.) is one of the most important food legume crops in the world. For the study, we selected local varieties of *Phaseolus vulgaris* L. "Ravot" and "Mahsuldor". The germination and germination factors of these varieties were analyzed under drought and salt stress conditions in vitro. The germination and germination rates of the "Ravot" and "Mahsuldor" bean varieties were compared with each other. In this case, the "Ravot" variety showed higher resistance indicators in both environments compared to the "Mahsuldor" variety.

*Keywords*: *Phaseolus vulgaris* L., salinity, drought, Ravot, Mahsuldor, germination.

# ОЦЕНКА УСТОЙЧИВОСТИ К ЗАСОЛЕНИЮ И ЗАСУХЕ У СОРТОВ ФАСОЛИ ОБЫКНОВЕННОЙ (*PHASEOLUS VULGARIS* L.) В УСЛОВИЯХ *IN VITRO*

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Аннотация. Засуха и засоление почв остаются одними из наиболее значимых ограничивающих факторов окружающей среды, влияющих на сельское хозяйство во всем мире. Важным решением этой проблемы является отбор видов растений, которые дают высокие урожаи даже в условиях засоленных почв. Фасоль обыкновенная (*Phaseolus vulgaris* L.) является одной из важнейших продовольственных бобовых культур в мире. Для исследования нами были отобраны местные сорта *Phaseolus vulgaris* L. «Равот» и «Махсулдор». Всхожесть и факторы прорастания этих сортов были проанализированы в условиях засухи и солевого стресса *in vitro*. Показатели всхожести и прорастания сортов фасоли «Равот» и «Махсулдор» сравнивались между собой. В данном случае сорт «Равот» показал более высокие индикаторы устойчивости в обеих средах по сравнению с сортом «Махсулдор».

*Ключевые слова: Phaseolus vulgaris* L., засоление, засуха, Равот, Махсулдор, прорастание.

# Introduction

One of the major challenges facing global agriculture is the need to produce 70% more food crops by 2050 to feed an additional 2.3 billion people worldwide [1]. Drought and soil salinity the are among most limiting environmental factors affecting agriculture on a global scale. Even moderate water scarcity or salt stress can lead to a 50-70% reduction in average yield for most crops [2]. Soil salinization is becoming а major constraint for agriculture in many regions of the world, particularly in arid and semi-arid areas. More than 20% of the world's cultivated land is affected by soil salinity, and this proportion is increasing daily [3]. However, this trend is primarily driven by anthropogenic factors such as irrigation with saline inefficient water and the and unregulated use of chemical fertilizers [4].

Therefore, selecting plant species that can maintain high productivity under saline soil conditions has become a crucial agricultural priority.

Common bean (*Phaseolus vulgaris* L.) is considered the most important food legume crop globally. In world agriculture, the common bean (*P. vulgaris*) type, native to Central and South America, is the most widespread. Additionally, species such as runner bean (*P. coccineus*), lima bean (*P. lunatus*), tepary bean (*P. acutifolius*), adzuki bean (*Vigna angularis*), rice bean (*Vigna umbellata*), and other types are also cultivated [5]. Globally, the area cultivated with beans reached 22 million hectares (as of 1999), with large areas grown in India, Brazil, and China [6]. In Uzbekistan, the common bean has been cultivated since ancient times.

This staple food contains high levels protein and of significant of amounts fiber. complex carbohydrates, vitamins (such as folic acid), and minerals (Cu, Ca, Fe, Mg, Mn, Zn) that cover daily nutritional needs. Its grain contains 20-31% protein, 0.7-3.6% oil, approximately 50% starch, 2.3-7.1% fiber, and organic acids [7]. The stems are considered good fodder for livestock. Concurrently, anti-nutritional factors such as phytic acid, lectins, and saponins have also been identified in the pods and dry seeds of this crop [8]. Furthermore, bean protein is rich in essential amino acids and bioactive peptides, thereby exhibiting functional The main bioactive properties. compounds reported in common bean include phenolic acids (ferulic acid, pcoumaric acid, sinapic acid, and gallic acid) and flavonoids (kaempferol, quercetin, catechin, and proanthocyanidins). Nutrition experts encourage the consumption of beans for the prevention of chronic diseases [5].

For our research, we selected the 'Ravot' and 'Mahsuldor' cultivars of *Phaseolus vulgaris* L. We analyzed the germination capacity and germination factors of these cultivars under *in vitro* drought and salt stress conditions. The primary objective of our research was to obtain relevant data concerning the biochemical mechanisms underlying plant tolerance to salinity and drought, and specifically, to determine which *Phaseolus* cultivars demonstrate the greatest suitability (tolerance) for coping

greatest suitability (tolerance) for coping **Materials** a The research was conducted in 2025 at the Plant Genetic Resources Research Institute, in the Laboratory of Plant Physiology and Immunity, under in vitro conditions. The study was carried out on local varieties of common bean (*Phaseolus vulgaris* L.), namely

During the experiments, different concentrations of NaCl (0 (control  $H_2O$ ), 50, 100, 150, 200 mM) and PEG-6000 (0 (control  $H_2O$ ), 10%, 15%, 20%) solutions were used. PEG-6000 was employed to simulate a drought environment.

Ravot and Mahsuldor.

Prior to germination, seeds were treated with 5% sodium hypochlorite for 5 minutes and then rinsed with distilled water. The seeds were subsequently placed in Petri dishes (11 cm in with salt and drought stress. We hypothesized that the observed differences in stress tolerance among the selected cultivars could be attributed to variations in the efficiency of regulating ion transport and maintaining cellular osmotic balance.

# Materials and Methods

diameter) lined with two layers of filter paper. Ten seeds were placed in each Petri dish. A 10 mL aliquot of the prepared solutions was added to the seeds [9], and the dishes were sealed with parafilm before being incubated in complete darkness at  $25 \pm 1$  °C to allow germination. Seeds were considered germinated when the radicle reached a length of 2 mm. The final germination count was recorded on the seventh day. The germination rate (%), root length (cm), shoot length (cm), and number of root branches were measured.

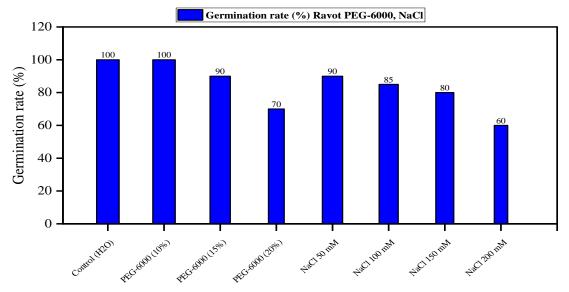
The germination rate was calculated using the following formula:

# $Germination \ rate \ (\%) = \frac{Number \ of \ germinating \ seeds}{Total \ number \ of \ seeds} * 100$

A bean seed consists of two cotyledons attached to the hypocotyl and an epicotyl that grows between the two cotyledons to form the shoot [10,11]. The radicle-hypocotyl junction was identified based on the color of the emerging seedling (the white part was considered the radicle, while the other section was classified as the hypocotyl). The experiment was designed as a completely randomized trial with two replications [12]. All data were subjected to statistical analysis. Statistically significant mean values were grouped using the LSD Multiple Comparison Test [13].

**Results and Discussion** 

This study investigated the in vitro tolerance of common bean (Phaseolus vulgaris L.) cultivars 'Ravot' and 'Mahsuldor' to salinity and drought stress. The effects of these stress factors on seed germination rate, root and shoot length, and seedling vigor index were evaluated (Figures 1, 2, 3, 4, 5, and 6).

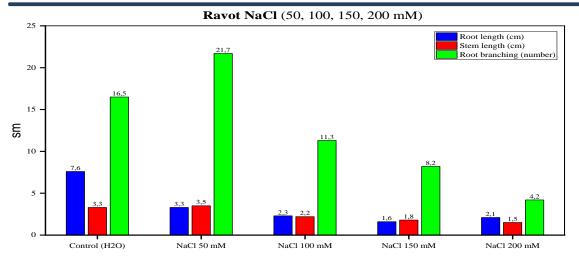


**Fig. 1.** Ravot length, shoot length (cm), and root branching (number) of 7-day-old seedlings of the local bean variety "Ravot" under the influence of control and drought (PEG-6000).

The germination rate of 'Ravot' cultivar under drought stress, relative to the control, was observed to be 100%, 90%, and 70% in PEG-6000 solutions at concentrations of 10%, 15%, and 20%, respectively. Under salinity stress, the germination rates in NaCl solutions at concentrations of 50, 100, 150, and 200 mM were 90%, 85%, 80%, and 60%, respectively (Figure 1).

The effects of varying levels of drought stress (10%, 15%, 20% PEG-6000) on root and shoot length, and root branching in 7-day-old 'Ravot' common bean seedlings were investigated. In the control group (H<sub>2</sub>O), seedlings exhibited the highest growth parameters: root length (7.6 cm), shoot length (3.3 cm),

and root branching (16.4 branches). Under 10% PEG-6000 treatment, the drought initial level of stress significantly suppressed seedling growth, resulting in reduced root length (3.8 cm), shoot length (2.1 cm), and root branching (11.7 branches) compared to the control. As drought stress increased with 15% PEG-6000, growth parameters further declined, with root length, shoot length, and root branching decreasing to 3.275 cm, 1.6 cm, and 6.4 branches, respectively. The highest level of drought stress (20% PEG-6000) severely impacted seedling growth, leading to the lowest recorded values: root length (1.5 cm), shoot length (0.43 cm), and root branching (1.9 branches) (Figure 2).

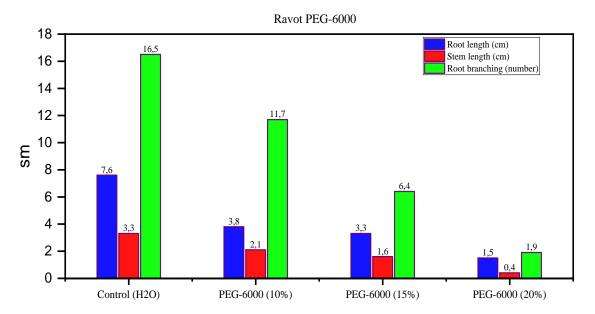


**Figure 2** Germination (%) of the local bean variety "Ravot" under the influence of control, saline NaCl, and drought PEG-6000.

The effect of salinity stress on seedling growth was characterized as follows. In the control group (H<sub>2</sub>O), representing a salt-free environment, seedlings exhibited optimal growth, with root length (7.6 cm), shoot length (3.3 cm), and root branching (16.4 This indicates that the absence of salinity is conducive to optimal growth. An initial salinity concentration of 50 mM NaCl slightly affected growth, resulting in root length (3.2 cm) and shoot length (3.5 cm) that were marginally lower than the control, while root branching slightly increased to 21.7 branches.

As salinity increased, growth parameters further declined. At 100 mM NaCl, root length (2.3 cm), shoot length (2.1 cm), and root branching (11.3 branches) significantly decreased. At 150 mM NaCl, growth parameters were further reduced, with root length (1.5 cm), shoot length (1.7 cm), and root branching (8.2 branches). The highest salinity concentration (200 mM NaCl) severely inhibited seedling growth, yielding the lowest recorded values: 52 root length (2.06 cm), shoot length (1.5 cm), and root branching (4.2 branches) (Figure 3).

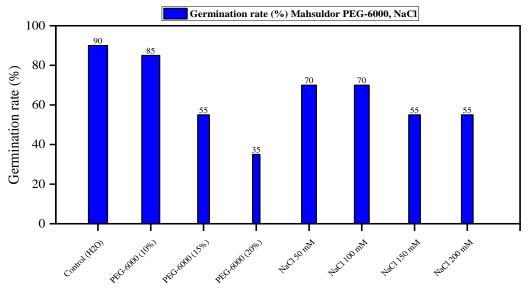
Changes germination in the percentage of the 'Mahsuldor' common bean cultivar under various stress factors, specifically drought (PEG-6000) and salinity (NaCl), are illustrated in figure 4. In the control group  $(H_2O)$ , where no stress factors were applied, the germination percentage was 90%, indicating a high germination capacity of this bean cultivar under optimal conditions. A significant decline in germination percentage was observed with increasing PEG-6000 concentration. 10% PEG-6000, At germination decreased to 85%, while at 15% PEG-6000, it further declined to 55%. Under the highest level of drought stress (20% PEG-6000), germination sharply decreased to 35%. These results confirm that drought stress negatively impacts seed germination in beans, with a pronounced reduction in germination capacity as stress intensity increases.



**Fig. 3.** Root length, shoot length (cm), and root branching (number) of 7-day-old seedlings of the local bean variety "Ravot" under the influence of control and saline (NaCl) stress

Under salinity stress, germination percentage also decreased with increasing NaCl concentration, albeit to a lesser extent compared to drought stress. At 50 mM and 100 mM NaCl, germination was recorded at 70%, while at 150 mM and 200 mM NaCl, it

decreased to 55%. These findings indicate that salinity also stress negatively affects seed germination in beans. However, at lower NaCl concentrations (50 mM and 100 mM), the impact of salinity stress was less severe compared to drought stress.



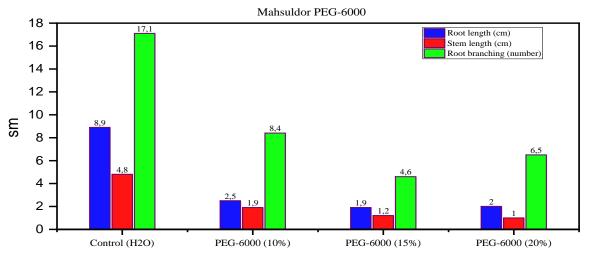
**Fig. 4**. Germination (%) of the local bean variety "Mahsuldor" under the influence of control, saline NaCl, and drought PEG-6000

Changes in 7-day-old seedlings of the 'Mahsuldor' cultivar were investigated under artificially induced drought (10%, 15%, and 20% PEG-6000)

and salinity stress (50, 100, 150, and 200 mM NaCl). Under control conditions (H<sub>2</sub>O), seedlings exhibited optimal growth, with root length measuring 8.9 cm, shoot length 4.8 cm, and root branching at 17.1 branches. However, with increasing PEG-6000 concentration,

a progressive decline in growth parameters was observed.

In the 10% PEG-6000 treatment, root length decreased to 2.8 cm, shoot length to 1.9 cm, and root branching to 8.4 branches.



**Fig. 5.** Root length (cm) of 7-day-old seedlings of the local Mahsuldor bean cultivar under control and drought (PEG-6000) conditions.

With 15% PEG-6000, these parameters further declined, with root length measuring 1.9 cm, shoot length 1.2 cm, and root branching 4.9 branches. The highest level of drought stress (20% PEG-6000) resulted in a severe reduction in growth, with root length recorded at 2.02 cm, shoot length at 0.99 cm, and root branching at 6.5 branches (Figure 5).

From the above results, it can be concluded that when artificial drought conditions are induced using PEG-6000, the growth rate of plants decreases as the PEG-6000 concentration increases, with significant variations observed in root and shoot length. Under salinity stress (NaCl), while seedlings in the control group (H<sub>2</sub>O) exhibited optimal growth, a decline in seedling growth parameters was observed with increasing NaCl concentration. At 50 mM NaCl, root length was 3.0 cm, shoot length 3.4 cm, and root branching was 11.6 branches. At 100 mM NaCl, root length was 3.1 cm, shoot length decreased to 1.2 cm, and root branching declined to 8.6 branches. Exposure to 150 mM

NaCl further reduced growth parameters, with root length measuring 1.5 cm, shoot length 1.8 cm, and root branching decreasing to 5.7 branches. At the highest salinity level (200 mM NaCl), seedling growth parameters reached their lowest values, with root and root branching at 4.6 branches length at 1.2 cm, shoot length at 1.3 cm, (Figure 6).

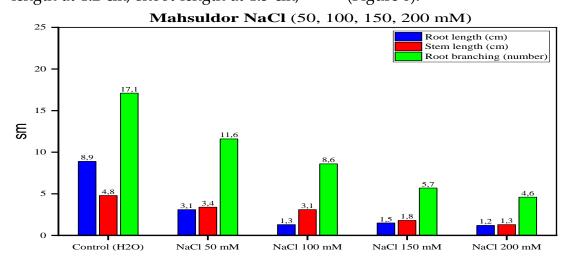


Figure 6. Root length (cm) of 7-day-old seedlings of the local *Mahsuldor* bean cultivar under control and salinity (NaCl) conditions

This indicates that as NaCl concentration increases, root and shoot length, as well as root branching, significantly decrease. Salinity is a stress factor for plants, substantially affecting their ability to absorb water and nutrients.

Based on the research findings, it is challenging to definitively determine which stress factor, drought or salinity, the plants exhibited greater tolerance to, as both significantly negatively impacted growth parameters. However, several observations provide insights into the differential responses of the cultivars to these stress conditions.

A comparative analysis of germination and seedling growth between the 'Ravot' and 'Mahsuldor' bean cultivars revealed that 'Ravot' generally demonstrated higher tolerance under both stress conditions. Under severe drought conditions (20% PEG-6000), 'Ravot' maintained a germination

## Conclusion

rate of 70%, whereas 'Mahsuldor' exhibited a significantly lower rate of 35%. Similarly, under high salinity stress (200 mM NaCl), the germination rate was 60% for 'Ravot' and 55% for 'Mahsuldor'.

These results suggest that while both cultivars are sensitive to drought and salinity stress, 'Ravot' shows a relatively higher tolerance compared to 'Mahsuldor'. Nevertheless, to comprehensively assess plant stress tolerance, further research is warranted, including investigation the of physiological biochemical and parameters.

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# TOTAL WATER CONTENT INDICATORS OF THE SUBSPECIES GOSSYPIUM HERBACEUM L. AND GOSSYPIUM ARBOREUM L.

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*Summary.* This article presents data on the total water content of *Gossypium herbaceum* L. and *Gossypium arboreum* L. subspecies, analyzing the impact of this indicator on productivity and economically important traits. Furthermore, the article provides a detailed review of scientific studies, literature, and findings conducted by researchers in the field concerning the total water content in cotton species. Through this analysis, information is presented on the water requirements of different cotton subspecies and the relationship between their water content and agrobiological characteristics.

*Keywords:* Water efficiency, water storage capacity, agronomic practices, biotic and abiotic stress factors, photosynthesis and respiration processes, soil moisture.

# ОБЩИЕ ПОКАЗАТЕЛИ СОДЕРЖАНИЯ ВОДЫ У ПОДВИДОВ GOSSYPIUM HERBACEUM L. И GOSSYPIUM ARBOREUM L.

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Абстракт. В данной статье представлены данные о общем содержании воды у подвидов Gossypium herbaceum L. и Gossypium arboreum L., а также проанализировано влияние данного показателя на урожайность и хозяйственно ценные признаки. Кроме того, подробно рассматриваются научные исследования, литература и результаты, проведённые специалистами в области изучения общего содержания воды в видах хлопчатника. На основе анализа приводится информация о потребности различных подвидов хлопчатника в воде и взаимосвязи между содержанием влаги и их агробиологическими особенностями.

*Ключевые слова:* Эффективность использования воды, Способность к накоплению воды, Агрономические приёмы, Биотические и абиотические стрессовые факторы, Процессы фотосинтеза и дыхания, Влажность почвы.

## Introduction

Cotton (*Gossypium* spp.) with its diverse economic properties provides raw materials for many industries, such as natural fiber for the textile industry, cottonseed for the oil sector, and cellulose for the livestock industry [4, 6, 8, 5]. This industrial enterprise, which directly and indirectly affects the livelihoods of many people around the world, is drought-resistant and, due to its warm climate, is cultivated by about 30 countries, primarily China, India, the United States, Pakistan, and Brazil [22, 25]. At the same time, intensive cultivation also leads to the emergence of various biotic diseases and pests and abiotic stress factors. Abiotic stress factors such as drought and salinity prevent cotton from absorbing water, while biotic stress factors such as Verticillium and Fusarium cause the xylem vessels to become clogged and the plant to not receive enough water, which leads to a decrease in turgor pressure that ensures the verticality of plants and causes leaf wilting and drop [15]. Drought is defined as a prolonged period of water shortage for plants globally due to the effects of greenhouse gases caused by human and non-human activities. Drought is one of the most important abiotic stress factors, causing 73% of yield losses in cotton [23].

Although cotton is more drought tolerant than other cultivated field crops, prolonged drought stress can lead to reduced yield, reduced biomass and stem weight, stunted plant growth, reduced fiber quality, and the development of small pores [19]. Drought has been shown to negatively affect Gossypium spp., among other plants, by limiting plant height, leaf weight, node number, transpiration rate, photosynthetic rate, and stomatal 28, permeability [19, 2, 21, 7]. Furthermore, when subjected to prolonged drought stress, it closes its stomata, curls its leaves, attempts to achieve osmotic regulation, and attempts to access water/moisture from within deep the soil [12], and photosynthetic rates decrease [9]. 58

Under water deficit conditions, short-term drought during the seedling stage of the root system increased the ability of cotton roots to reach deep water and led to a reduction in root diameter [18]. Studies have shown that cotton roots are more affected by drought than by root distribution [17]. Insufficient soil moisture reduces root elongation [3, 20] and a reduced root system develops approximately 42-70 days after cotton emergence [17]. Root growth is an important process that is cultivated at a stable temperature [16, 11]. Zohid et al. [27] reported that 22–30 °C is the optimal temperature and root/shoot (R/S) ratio for cotton root and shoot growth, but temperatures between 32 and 40 °C limit the spread and growth of the root/shoot system. A finding similar was reported by Koevoets et al. [13]. Optimum temperature promotes an increase in the root/shoot ratio (R/S ratio); however, temperatures above the optimum temperature reduce the uptake of water and plant nutrients by the root system of cotton plants, resulting in a weakened root system against drought [14].

The reduction in soil water content through evaporation significantly reduces root activity, but hydraulic uplift and transpiration by roots can increase the groundwater level to levels close to the soil surface (0–40 cm soil depth) [10].

The total water content in plants is important. Water performs a number of important functions for plants, which

further increases its relevance. The main roles in this are metabolic processes, photosynthesis, respiration, osmotic balance, osmotic pressure, growth and development, cell growth, tissue and development, transpiration, organ thermoregulation, minerals and nutrients. In particular, A. Kholliyev noted that if a plant is provided with a sufficient amount of water, the physiological and biochemical processes taking place in its body are activated [26]. The amount of water in the soil composition above or below the optimal level has a negative effect on the course of the above processes.

In order to analyze the waterrelated properties of legumes and how efficiently they use water, indicators such as total water content, water storage capacity and plant water consumption were measured in laboratory and field conditions. The water storage capacity of plants (LWC-Leaf Water Content) was observed to vary from 50% to 70%. In addition, it was found that the water storage capacity of plants is related to soil moisture and genetic differences between plants. It is recommended to consider the water resistance of plants and environmental conditions, apply water efficient use and agronomic practices to increase the water-use capacity [24]

The impacts of cotton production on the environment are easily visible and have different faces. On the one hand there are the effects of water

depletion, on the other hand the effects on water quality. In many of the major textile processing areas, downstream riparians can see from the river what was the latest colour applied in the upstream textile industry. The Aral Sea is the most famous example of the effects of water abstractions for irrigation. In the period 1960-2000, the Aral Sea in Central Asia lost approximately 60% of its area and 80% of its volume as a result of the annual abstractions of water from the Amu Darya and the Syr Darya-the rivers which feed the Aral Sea-to grow cotton in the desert.

About 53% of the global cotton field is irrigated, producing 73% of the global cotton production. Irrigated cotton is mainly grown in the Mediterranean and other warm climatic regions, where freshwater is already in short supply. Irrigated cotton is mainly located in regions: dry Egypt, Uzbekistan, and Pakistan. The province Xinjiang of China is entirely irrigated, whereas in Pakistan and the North of India a major portion of the crop water requirements of cotton are met by supplementary irrigation. As a result, in Pakistan already 31% of all irrigation water is drawn from ground water and in China the extensive freshwater use has caused falling water tables. Nearly 70% of world's the cotton crop production is from China, USA, India, Pakistan and Uzbekistan (USDA, 2004). Most of the cotton productions rely on a furrow irrigation system. Sprinkler and 59

drip systems are also adopted as an irrigated method in water scarce regions. However, hardly about 0.7% of land in the world is irrigated by this method.

As you know, when the plants are provided with sufficient amounts of water The physiological and biochemical processes that take place in their bodies are intensified. The amount of water in the soil is higher than the optimal level or low The presence of a negative impact on the passage of these processes [16, 2, 15, 12]. H.S. Samiev [27]The water of plants One of the most important indicators in the balance shared water on the leaves The amount is.

## Material and methods

Research sources A number of forms of the species G.herbaceum subsp. pseudoarboreum, subsp. pseudoarboreum f. subsp. *frutescens*, G.arboreum harga, subsp. obtusifolium var. indicum, subsp. neglectum, subsp. neglectum f. sanguineum, subsp. perenne, subsp. nanking (white fiber) belonging to the Gossypium L. genus, obtained from the collection of the unique object "Cotton gene pool" of the Institute of Genetics

and Experimental Biology of Plants of the Academy of Sciences of the Republic of Uzbekistan, were studied [1] (Table 1). In the studies, important of physiological indicators water exchange in foreign collection samples belonging to Lathyrus sativus L. species were determined using the following methods:

Total water content in leaves (Tretyakov et al., 1990).

## Table 1

Carborneries	Total water content (%)				
Subspecies	$\overline{x} \pm \mathbf{S} \overline{x}$	S	V %		
Gossypium herbaceum					
subsp. pseudoarboreum	74,3 ± 2,88	4,98	6,69		
subsp. pseudoarboreum f. harga	$72,4 \pm 2,84$	4,91	6,78		
subsp. <i>frutescens</i>	65,5 ± 2,70	4,67	7,13		
Gossypium arboreum					
subsp. obtusifolium var. indicum	68,5 ± 2,76	4,78	6,97		
subsp. perenne	68,8 ± 2,77	4,79	6,96		
subsp. <i>neglectum</i>	71,6 ± 2,83	4,89	6,82		
subsp. neglectum f. sanguineum	71,6 ± 2,82	4,89	6,82		
subsp. nanking (whiett fiber)	$68,4 \pm 2,76$	4,78	6,98		

## Total water content indicators in the subspecies G.herbaceum L. and G.arboreum L.

## **Results and discussion**

The total water content was studied in the sources obtained from these subspecies *G.herbaceum* L. and *G.arboreum* L. The total water content was analyzed in the leaf of the plant during the leafing phase. In the course of our research, we studied the total water content of plants in the budding phase of the subspecies *G.herbaceum* L. and *G.arboreum* L. In the *G.herbaceum* L.

varieties, a clear dominance in terms of total water content in the combing phase was observed in the subspecies subsp. pseudoarboreum (74.3  $\pm$  2.88), and the corresponding coefficient of variation was 6.69%. The average total water observed in subsp. content was pseudoarboreum f. harga compared to other samples. Its content was on average 72.4 ± 2.84, the coefficient of variation was 6.78%. In the budding phase, the *G.herbaceum* L. subsp. frutescens subspecies showed a low coefficient of variation of  $65.5 \pm 2.70\%$ , while the coefficient of variation was 7.13%.

Among the subspecies *G.arboreum* L., the highest indicator of total water content was observed in the form of

Studies on the total water content of the subspecies *G.herbaceum* L. and *G.arboreum* L. show that there are differences in total water content in both species, and these differences affect the plants' tolerance to water deficit and yield potential.

In conclusion, the subspecies *G.herbaceum* L. subsp. *pseudoarboreum* 

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The average value compared to other samples was determined for the subspecies subsp. *perenne* (68.8  $\pm$  2.77). The coefficient of variation for this sample was 6.96%. The lowest index was observed in the subspecies subsp. nanking (white fiber) (68.4  $\pm$  2.76). The coefficient of variation was found to be 6.98%.

Analysis of the results showed that the total water content in the studied sources varied. The highest values were observed in the subspecies *G.herbaceum* subsp. *pseudoarboreum* (74.3  $\pm$  2.88), *G.arboreum* subsp. *nanking* (68.4  $\pm$ 2.76). This increases the tolerance to water deficit and creates the basis for obtaining high yields.

# Conclusion

and G.arboreum L. subsp. neglectum good tolerance to drought show conditions and efficient water storage capabilities with high total water However, content. the subspecies G.herbaceum L. subsp. frutescens and G.arboreum L. subsp. nanking have lower water retention capacity, which may limit their drought tolerance.

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# BIOLOGICAL AND ECONOMIC IMPORTANCE OF SESAME (SESAMUM INDICUM L.)

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*Summary.* This article examines the increasing demand for plant-based raw materials and medicinal products due to the current global population growth. The genetics and breeding of sesame (*Sesamum Indicum* L.) are crucial for developing new varieties with high yields, resistance, and quality oil. This paper also includes a literature review on the use of compounds found in sesame seeds. The prospects for sesame cultivation in arid regions, including Central Asian countries, are highlighted, with a focus on optimizing agronomic practices, expanding cultivated areas, and developing processing industries. The economic aspects of sesame production, its export potential, and its applications in the food, pharmaceutical, and feed industries are also considered.

*Keywords*: Sesamum indicum, oil, antioxidant, sesamin, sesamolin, chlorosesamone, vitamin, protein, carbohydrate.

# БИОЛОГИЧЕСКОЕ И ХОЗЯЙСТВЕННОЕ ЗНАЧЕНИЕ КУНЖУТА (SESAMUM INDICUM L.)

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Абстракт. В этой статье рассматривается растущий спрос на растительное сырье и лекарственные препараты в связи с текущим ростом населения во всем мире. Генетика и селекция кунжута (*Sesamum Indicum* L.) имеют решающее значение для выведения новых сортов с высокой урожайностью, устойчивостью и качественным маслом. Эта статья также включает обзор литературы об использовании соединений, содержащихся в семенах кунжута. Освещаются перспективы выращивания кунжута в засушливых регионах, включая страны Центральной Азии, с акцентом на оптимизацию агрономических методов, расширение посевных площадей и развитие перерабатывающих производств. Также рассматриваются экономические аспекты производства кунжута, его экспортный потенциал и применение в пищевой, фармацевтической и комбикормовой промышленности.

**Ключевые слова:** сезам индийский, масло, антиоксидант, сезамин, сезамолин, хлорсезамон, витамин, белок, углевод.

## Introduction

Myanmar is the world's leading ranks second with 769,000 tons. producer of sesame seeds, with an However, in terms of cultivated area, annual output of 861,573 tons. India India takes the first place, growing 64 sesame on 1,780,000 hectares, which is more than Myanmar's 1,584,000 hectares [15].

Historically, sesame has been used in the food industry as a source of oil, flour, and seasoning. In medicine, sesame oil was used to treat skin diseases and boost immunity. In culture and religion, in Ancient Egypt, sesame was used in rituals, while in China, it was considered a symbol of longevity. Today, sesame is cultivated in more than 40 countries worldwide, including India, China, Sudan, Nigeria, Burma, Ethiopia, and Pakistan. The largest cultivated areas are in Africa and Asia; however, in recent decades, interest in sesame cultivation has been growing in the CIS countries as well [12].

Sesame is one of the earliest ancient plants used by humans, with information about it recorded in historical texts. For instance, Egyptians called sesame "SESEMT" and included it in their list of medicinal and healing plants 3600 years ago. Babylonian and Assyrian manuscripts (4300 years ago) contain detailed information about the sesame plant and its uses. Reports on archaeological research in Turkey indicate that sesame was used for various purposes (consumption, fuellubricant products) during the Urartu Empire, 2750 years ago. According to scientific sources, sesame was introduced to Central Asia in the early 17th-18th centuries and is now cultivated in almost all republics [2].

(Sesamum indicum L.) Sesame belongs to the family *Pedaliaceae*. It is an herbaceous plant that grows to a height of 60-100 cm [5]. Wild sesame species are divided into three types based on chromosome number: 2n=26 (S. indicum, S. alatum), 2n=32 (S. protratum, S. angolense), and 2n = 64 (S. radiatum, S. schinzianum) [9]. Cultivated sesame is an annual herbaceous plant. Its taproot is thickened at the top, penetrates 120 cm deep into the soil, and produces numerous lateral branches. The stem grows upright, is 4 or 8-angled, and reaches an average height of 100-110 cm in irrigated areas and 50-60 cm in rained areas. It is densely or sparsely covered with green hairs. The stem branches from its lower part, where there are no capsules, producing 4-6 long branches that grow upwards. The number of branches can reach 10-12, or it may branch sparsely (forms with 2 branches or entirely unbranched forms also exist).

According to FAO, global sesame production is around 7 million tons per year, with the largest exporters being India, China, Sudan, Ethiopia, Myanmar, and Nigeria [12]. However, demand for sesame and its processed products continues to grow rapidly, particularly in Europe, the USA, and the Middle East.

The leaves are petiolate, solitary or oppositely arranged, and hairy. The leaves on the lower part of the plant are entire, while those in the middle part are entire or lobed, depending on the variety. The leaves on the upper part of the stem are usually lanceolate.

The flowers are large, shortstalked, emerging singly or in groups of three from the leaf axils, distinguishing between single-flowered and threeflowered forms of sesame. The flowers are pentamerous. The corolla is fivelobed, with petals fused together, tubular, bilabiate, and swollen at the throat. The color of the corolla ranges from pink and purple to white, depending on the sesame form. There are five stamens, one of which usually remains undeveloped. The ovary has a long style and a four-lobed stigma.

Considering the growing demand for healthy and natural products, the production of sesame oil in CIS Uzbekistan, countries, including Kazakhstan, Tajikistan, and has promising opportunities for expansion and export.

The fruit elongated, Is an flattened capsule, hairy. The capsule consists of 2 or 4 carpels, whose edges curl inwards, forming a false septum. In some forms of sesame, the false septa underdeveloped or completely are absent. When the capsules mature, they dehisce, and the valves open. If the transverse septa are underdeveloped or absent, the seeds easily scatter. However, if open capsules with septa are inverted downwards, the seeds easily fall out because the top of each locule in the carpel is open. Nowadays, there are forms of sesame with closed capsules. The capsule is four or eight-66

celled, averaging 4 cm in length and 0.9 cm in width. A single plant can have 20 to 100 capsules. Sesame seeds are small, flat, egg-shaped, measuring 2.7-4 mm in length and 1.9 mm in width. The weight of 1000 seeds ranges from 2 g to 5 g, averaging 3 g. The color of the seeds is often light or brown, sometimes white or black [16].

The homeland of sesame Is Southwest Africa. It is cultivated in Central Asian republics, Transcaucasia, Crimea, Krasnodar, Ukraine, and Moldova [4].

According to scientists, the sesame plant grows very slowly during the first twenty to twenty-five days of its growth period. It takes 35-40 days from germination to the appearance of the first flowers. Sesame is a thermophilic plant; seedlings germinate quickly and uniformly when the soil temperature is 18-20°C. The optimal temperature for plant growth and development is considered to be 22-25°C. Short-term cool temperatures (2-3°C) in autumn are also dangerous for the plant and immature capsules [3].

Globally, approximately 70% of cultivated sesame seeds are processed for oil and meal production. Annual consumption is divided into 65% for oil extraction and 35% for food purposes. Sesame meal, after oil extraction, contains 35-50% protein, making it a valuable feed for poultry and livestock.

Sesame has several important industrial applications. For example, African peoples have used perfumes

and aromatic substances made from sesame flowers. The sesamin compound in sesame has bactericidal and insecticidal properties and acts as an antioxidant: this compound inhibits cholesterol absorption and cholesterol synthesis by the liver. Sesamolin has an insecticidal effect and is used as a synergist for pyrethrum-based pesticides [13].

The meal remaining after cold pressing sesame seeds contains 40% protein and 8% oil, and it is widely used confectionery industry. Oil in the obtained from hot pressing sesame seeds is used for technical purposes, and its soot, produced by burning it, makes excellent quality ink. The meal resulting from hot pressing sesame seeds is a nutritious feed for livestock, with 100 kg of it containing 132 feed units. Sesame seeds contain 48-65% oil. 16-19% protein, and 15.7-17.5% soluble carbohydrates [16].

Sesame oil has been used for cooking and as a flavoring additive in Asian and Western countries since ancient times [18]. This oil is also valued as a dietary and therapeutic agent. Sesame seeds are sometimes called immortality," "seeds of which is explained by their resistance to oxidation and rancidity, maintaining their quality even when stored at room temperature [6].

Studies have shown that sesameseeds have antioxidant and anti-cancerproperties[8].phytocomponentsextracted from the

seeds, such as sesamin and sesamolin, have cholesterol-lowering and blood pressure-regulating effects in the human body. Furthermore, they are rich in trace elements like copper, manganese, and calcium, which have been found useful in reducing osteoporotic pain (bone fragility) and swelling in rheumatoid arthritis [15].

Defatted sesame meal contains almost 50% protein, while the seed hull is rich in oxalic acid and fiber [1]. Sesamin and sesamolinlignans, located in the non-glycerin fraction of sesame oil, play an important role in its oxidative stability and antioxidant activity [14].

Sesame seeds contain up to 60% oil, vitamin Ε, factor-T, sesamin, sesamone, sesamin, and other substances. Sesame oil is a yellow liquid that solidifies at 3-6°C and belongs to semi-drying oils. Its specific gravity is 0.9197-0.9260, refractive index 1.4731-1.4760, acid value 1.4-10, saponification value 185.5-195, and iodine value 103-116. The oil contains glycerides of oleic, linoleic, palmitic, stearic, arachidic, and lignoceric acids [4].

In addition, sesame seeds are rich in minerals such as calcium, phosphorus, magnesium, and potassium. They also contain important vitamins like niacin, thiamine, riboflavin, and vitamin B<sub>6</sub> [15].

Sesame oil is used as a solvent, a fatty carrier for drugs, an emollient for the skin, and in the production of margarine and soap. The oil is primarily

used in cooking, salad preparation, and margarine production. Furthermore, it is widely used in the production of cosmetics, pharmaceuticals, paints, and insecticides. Chlorosesamone isolated roots is active from sesame an agent. antifungal Sesamin and sesamolin compounds have been found to increase the rate of fatty acid oxidation in liver mitochondria and peroxisomes. Consumption of sesame seeds increases plasma gammatocopherol levels and enhances vitamin activity, which is important in Ε preventing cancer and heart diseases. In pharmaceuticals, sesame oil is used as a solvent for intramuscular injections and has nourishing, emollient, and protective properties, also acting as a laxative [17].

Sesame oil is used as an antibacterial mouthwash. Sesame seeds are added to bread, rolls, cookies, healthy snacks, and breakfast products. The seeds are consumed raw or roasted, salted, and also mixed with lemon and honey, often ground into a sweet paste with sugar [15].

The global demand for sesame oil is steadily increasing. In this regard, it is crucial to develop new sesame varieties resistant to abiotic and stress factors and to expand the area under sesame cultivation. In recent decades, sesame (*Sesamum indicum* L.) has been gaining increasing significance in global agriculture due to its high nutritional and economic value, drought resistance, and potential for industrial processing. 68

The prospects for sesame cultivation are associated with genetic improvement, the advancement of agronomic practices, the expansion of cultivated and the development areas, of processing technologies [7]. The use of drip irrigation increases sesame yield by 25-30% compared to traditional methods [10]. Modern sesame varieties, such as Toshkent-122, Qora Shaxzoda, Seraxs-470, demonstrate and high productivity with minimal water consumption, which is particularly important under climate change conditions [11]. Genetic studies have identified genes responsible for drought tolerance and disease resistance, enabling the accelerated breeding of new adapted varieties [20]. The use of molecular markers and CRISPR-Cas9 technology allows for an increase in yield up to 3.5–4.0 t/ha, which is 20–30% higher than that of traditional varieties [19].

In the article "Sesame breeding in the post-genomic era: applying new tools to old challenges" [8], modern breeding methods aimed at enhancing sesame resistance to drought and high temperatures are discussed. The study "Effect of sowing methods and nitrogen fertilization on growth and yield of sesame" [21] demonstrates that the proper selection of sowing methods and nitrogen fertilization levels can significantly increase crop yield. The article "Potential of sesame (Sesamum *indicum* L.) cultivation in temperate regions: A review" [5] analyzes the

possibilities of cultivating sesame in

non-traditional climatic zones.

## Conclusion

Sesame (Sesamum indicum L.) is one of the most valuable oilseed crops, thanks to its high oil, protein, and compound bioactive content. Its drought resistance, adaptability to various soil and climatic conditions, and diverse applications make it a promising crop for further development. Enhancement of breeding programs focus on developing early-maturing, high-oil-content, and disease-resistant varieties, which will help expand

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sesame cultivation regions. Application modern selection methods of Molecular breeding techniques and physical mutagenesis can accelerate the development of new varieties, improving productivity and resilience. Possibilities for introduction into new regions – Expanding sesame cultivation areas is feasible through adaptation to new climatic zones. Its high drought resistance makes it an attractive crop for water-deficient regions.

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# ANALYSIS OF MICRO AND MACRO ELEMENTS IN THE SEED COMPOSITION OF FOREIGN COLLECTION SAMPLES OF THE SPECIES LATHYRUS SATIVUS L.

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*Summary.* This article analyzes the quantitative indicators of micro and macro elements in the seed composition of *Lathyrus sativus* L. collection samples (Bio 520 x Bio) x273, prateek x IG140034, Ratan x 2125, Jabbouleh, 1330x2125, Ratan x1307, RatanxIG 135481, Bio 520 x Bio) x274, PrateekxIG 140035, Bio520x1330) obtained from the international organization ICARDA, and in the local varieties of common chickpea (Lalmikor, Polvon) from Uzbekistan. In particular, the highest indicator for the microelement Fe was observed in the 1330x2125 sample (110 mg/kg,  $\mu$ g/g), while the highest indicator for the microelement Al was found in the Pratek x IG 140034 sample (83 mg/kg,  $\mu$ g/g). Among the studied field pea collection samples, the highest indicator for the macroelement Mg was observed in the Ratanx2125 sample (3900 mg/kg, g/t), while the highest indicator for the macroelement Ca was found in the Ratan x 2125 sample (4500 mg/kg,  $\mu$ g/g), and the highest indicator for the macroelement K was recorded in the Ratan x 2125 sample (22000 mg/kg,  $\mu$ g/g).

*Keywords: Lathyrus sativus* L., sample, collection, microelement, macroelement, autoclave, mass spectrometry.

# АНАЛИЗ СОДЕРЖАНИЯ МИКРО- И МАКРОЭЛЕМЕНТОВ В СОСТАВЕ СЕМЯН ЗАРУБЕЖНЫХ КОЛЛЕКЦИОННЫХ ОБРАЗЦОВ ВИДА LATHYRUS SATIVUS L.

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## Introduction

Due to the increasing global population, the problem of malnutrition is being observed. The growing demand plants rich in microfor and macroelements and proteins, especially in African countries, coupled with a scarcity of ecologically adapted plants, is increasing the risk of hunger. Increasing the production of leguminous plants, which are rich in

protein and consumed in large quantities by poor people as a substitute for meat, is of significant importance [7, 11].

One of the important microelements for plants is zinc and iron. Foliar spraying of *Lathyrus sativus* L. leaves with solutions of these elements significantly increases plant height, leaf area, number of branches, harvest index, and seed yield, and ensures ripening in a shorter period compared to the vegetative period. Maintaining a balance between root and shoot growth allows for sufficient yield and nutritional value. It has also been found that spraying Lathyrus leaves with 1.0% ZnSO4 significantly increases the Zn concentration in the seeds [2; 9].

Minerals are chemical components in food. Each mineral has its own specific properties, and together they ensure important processes in the human body (5, 10).

The optimal level of micro- and macroelements in *Lathyrus sativus* L. affects the grain quality of plants. Many microelements play a very important role in the physiological processes of plants, participating in the regulation of catalytic and hormonal functions [6, 13].

Field pea seeds are rich in minerals, and it has been studied that the concentration of some minerals

Macro and microelements were determined by inductively coupled plasma mass spectrometry (ICP-MS). This method was used to determine the elements calcium, phosphorus, magnesium, iron, and iodine in food products. For this purpose, 0.0500-0.500 g of the test substance was weighed on an analytical balance and placed in a Teflon vessel of the autoclave, then the appropriate of purified amount concentrated mineral acids (nitric acid and hydrogen peroxide) was added. The autoclave was closed and placed in a 72

(calcium, magnesium, and phosphorus) is high [12].

Mineral substances do not have energy value, but the body cannot function without them. These minerals perform a building function in human life, participate in the metabolic processes in every tissue of the human body, and in the construction of bone tissue [3, 4].

Zinc deficiency in the soil has a comprehensive effect on plants, including the chemical composition of their seeds and causing an increase in the concentration of neurotoxins [8].

**Microelements** are active compounds vital for the body, influencing blood formation, tissue respiration, and the growth and development of the organism [1].

## Materials and methods

Berghof programmed (MWS-3+) microwave digester. Depending on the type of substance being tested, the appropriate program was selected. After the substances in the autoclave were digested, they were transferred to 50 or 100 ml volumetric flasks and brought to the required mark with 0.5% nitric acid.

The determination of substances was carried out by ICP-MS or a similar inductively coupled argon plasma emission spectrometer.

The following equipment was used to perform the above analyses:

ICP-MS NEXION-2000 or similar mass spectrometer, microwave digesters (Germany) or similar Teflon autoclave: flasks of various sizes.

Reagents used: Multielement standard No. 3 (29 elements for MS).

The research object included collection samples of *Lathyrus sativus* L. belonging to the species obtained from the international organization ICARDA: Bio 520 x Bio) x273, prateekxIG 140034, Ratanx2125, Jabbouleh, 1330x2125,

Standards: mercury, nitric acid, hydrogen peroxide, distilled water, and argon (gas purity 99.995%).

# Material and metods

Ratanx1307, RatanxIG 135481, Bio 520 x Bio)x274, PrateekxIG 140035, Bio 520x1330, as well as local varieties of common chickpea (Lalmikor, Polvon) from Uzbekistan.

## **Results and discussion**

The amount of micro and macro elements in the seeds of foreign samples belonging to the species Lathyrus sativus L. and local common chickpea varieties was analyzed biochemically. The amount of 61 micro and macro elements in the seed composition was determined. The results of the biochemical analysis showed that the of metals amount some was significantly high, while a large amount of some elements was found in the control samples (Table 1). In particular, the indicator for the highest microelement Li was found in the 1330x2125 sample (6.40 mg/kg, g/t), while the local Lalmikor variety showed a good indicator for this trait (3.50 mg/kg, µg/g), and low indicators were observed in the remaining samples. The amount of the microelement Fe varied in the field pea samples, with the highest indicator observed in the 1330x2125 sample (110 mg/kg, µg/g), while the lowest indicator for this trait was recorded in the Bio 520xBio??x273 sample (51 mg/kg,  $\mu$ g/g). In addition, the highest indicator for the microelement Al was found in the Pratek x IG 140034 sample (83 mg/kg,  $\mu$ g/g), and the lowest result for this microelement was observed in the Bio 520x1330 sample (8.90 mg/kg,  $\mu$ g/g).

The amount of macroelements varied in the field pea collection samples. For example, the highest indicator for the macroelement Mg was observed in the Ratanx2125 sample (3900 mg/kg, g/t), while a relatively low indicator was recorded in the Bio 520xBio??x273 sample (1400 mg/kg,  $\mu g/g$ ). The macroelement Ca, which Is important for living organisms, is also one of the main sources of nutrition. The results of the analysis of the Ca macroelement in the seeds of field pea plants showed that a high indicator (4500 mg/kg,  $\mu$ g/g) was found in the Ratan x 2125 sample selected for the study, and a low indicator for this

macroelement was recorded in the Bio 520xBiox273 sample (1300 mg/kg, μg/g). At the same time, the macroelement K is very important for plants, maintaining pressure in plant osmotic cells. improving water absorption from the and increasing resistance soil, to drought stress. The highest indicator for this element was found in the Ratan x 2125 sample (22000 mg/kg, µg/g), and the lowest indicator was found in the Bio 520xBio??x273 sample (10000 mg/kg,  $\mu$ g/g). In addition, the highest Indicator for the macroelement P was found in the Ratan x 2125 sample (12000 mg/kg,  $\mu$ g/g), while the Bio 520xBio??x273 sample showed a lower indicator (4200 mg/kg,  $\mu$ g/g) compared to the others (Table 1).

# Conclusions

The analysis of the obtained results showed that macro and microelements are found in free form in the seeds of field pea plants. As a result of the conducted research, it was found that among the foreign collection samples, the 1330x2125 sample had a higher content of Li, B, Na, Cu, As in its seeds; the Ratanx2125 sample had a

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	Co	0,10-4000	0,110	0,110	0,240	0,100	0,190	0,320	0,190	0,140	0,140	0,110	0,150	0,081	In	0,006-	30%	<0,005	<0,005	<0,005	<0,005	<0,005	<0,005	<0,005	<0,005	<0,005	<0,005	<0,005	-
	Fe*	0,006- 30%	51,0	99,0	100	72,0	110	73,0	82,0	56,0 0	88,0 (	65,0 (	95,0 (	93,0 (	-	0,0	30	~0	~0 ,	<0>-	<0>	<0,	<0,	<0,	<0,	<0,	<0,	<0,	
8/1)	Mn	0,002- 0 10% 3	13,0	33,0	30,0	32,0	27,0	30,0	31,0	17,0	19,0	17,0	58,0	13,0	Cd	0,002-	10%	0,032	0,040	0,061	0,058	0,046	0,040	0,075	0,051	0,039	0,027	0,052	
· ( 111 g/ g)	C	1,0- 0 4000 1	<1,0	<1,0	1,10	<1,0	<1,0	<1,0	1,10	<1,0	<1,0	<1,0	<1,0	<1,0	Ag	1,0-	4000	0,740	42,0	0,078	0,050	0,085	0,010	0,012	0,730	0,190	0,083	0,068	-
2	>	0,10- 4000	0,068	0,096	0,110	0,110	0,180	0,064	0,070	0,055	0,092	0,061	0,092	0,069	Mo	0,10-	4000	10,0	14,0	18,0	20,0	15,0	15,0	24,0	9,90	14,0	12,0	18,0	-
	Ti*	0,0006- 9%	14,0	19,0	23,0	14,0 (	17,0 (	14,0 (	18,0 (	12,0 (	18,0 (	15,0 (	17,0 (	13,0 (	Nb	0,0006-	9%	0,008	0,012	0,008	0,010	0,008	0,006	0,005	0,019	0,004	0,043	0,004	
	Sc	0,10- 0 4000	0,028	0,059	0,068	0,081	0,082	0,042	0,037	0,046	0,046	0,045	0,047	0,046	$\mathrm{Zr}^*$	0,10-	4000	0,110	0,320	0,059	0,068	0,080	0,087	0,039	0,085	0,031	0,087	0,060	-
6 ~~~	$Ca^*$	0,005- 28%	1300	2800	4500	2100	2100	2200	2100	1900	2100	2200	2600	4300	γ	0,005-	28%	<0,10	$<\!0,\!10$	<0,10	<0,10	<0,10	<0,10	<0,10	<0,10	<0,10	<0,10	<0,10	
in me seens of concentron sumpres of me runny as suntas in (mg g, g, c)	K*	0,008- 30%	10000	20000	22000	15000	19000	15000	20000	13000	19000	12000	21000	14000	Sr	0,008-	30%	6,10	9,90	16,0	8,40	7,90	9,20	8,20	7,90	7,50	10,0	17,0	
mon la	P*	1,0- 4000	5200	9000	12000	6500	8300	7500	9800	6300	9500	7200	8400	6200	Rb	1,0-	4000	5,90	10,0	9,50	8,50	8,40	7,40	11,0	5,80	10,0	7,10	5,40	-
	Al*	0,002- 20%	17,0	83,0	26,0	40,0	34,0	10,0	14,0	12,0	18,0	8,90	49,0	27,0	Se	0,002-	20%	0,450	0,800	0,750	0,670	0,710	0,540	0,700	0,540	0,830	0,510	0,630	
	Mg*	0,004- 11%	1400	3200	3900	2500	2900	2200	2800	1600	2800	1800	3200	2000	$\mathbf{As}$	0,004-	11%	0,910	0,440	1,50	0,670	1,60	0,390	0,210	0,280	0,750	0,360	0,610	
	Na*	0,004- 11%	150	420	610	200	470	240	300	190	370	180	280	230	Ga	0,004-	11%	0,023	0,064	0,089	0,031	0,071	0,046	0,043	0,036	0,041	0,046	0,071	-
	B*	0,10- 4000	6,30	15,0	13,0	8,30	13,0	9,60	12,0	8,50	11,0	9,00	15,0	7,10	Zn	0,10-	4000	26,0	44,0	48,0	41,0	37,0	36,0	47,0	29,0	50,0	33,0	37,0	-
	Be	0,05- 4000	<0,05	<0,05	<0,05	<0,05	<0,05	<0,05	<0,05	<0,05	<0,05	<0,05	<0,05	<0,05	Си	0,05-	4000	7,20	13,0	14,0	11,0	15,0	9,80	13,0	8,80	18,0	9,20	13,0	-
1 man	E	0,05- 4000	0,790	3,10	2,10	0,960	6,40	1,70	0,880	1,80	1,30	0,850	3,50	2,30	Ż	0,05-	4000	1,00	2,10	2,50	1,10	1,90	2,10	2,00	1,30	1,70	1,60	1,40	-
	Elements	Measurement range elements	Bio 520xBio??x273	Pratek x IG 140034	Ratan x 2125	Jobbouleh	1330x2125	Ratan x 1307	Ratan IG 135481	Bio 520xBio??x274	Pratek x dg 140035	Bio 520x1330	Lalmikor	Polvon	Elements	Measurement range	elements	Bio 520xBio??x273	Pratek x IG 140034	Ratan x 2125	Jobbouleh	1330x2125	Ratan x 1307	Ratan IG 135481	Bio 520xBio??x274	Pratek x IG 140035	Bio 520x1330	Polvon	
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õ	Elements	Sn	Sb Sb	Te	C	Ba	La	Ce	Pr	PN	Sm	Eu	Gd	Tb	Dy	Ηo
	Measurement range elements	0,05- 4000	- 0,05- 0 4000	0,10- 4000	0,004-	0,004- 11%	0,002- 20%	1,0- 4000	0,008- 30%	0,005- 28%	0,10- 4000	0,0006- 9%	0,10- 4000	1,0- 4000	0,002- 10%	0,006- 30%
-	Bio 520xBio??x273	<0,10	-	_		-	0,011	0,028	<0,01	0,010	<0,01	<0,01	<0,01	<0,01	<0,01	<0,01
6	Pratck x IG 140034	<0,10	0 < 0, 10	) <0,30	0,051	5,70	0,018	0,034	<0,01	0,010	<0,01	<0,01	<0,01	<0,01	<0,01	<0,01
3	Ratan x 2125	<0,10	0 < 0,10	) <0,30	0,100	8,50	0,015	0,031	<0,01	0,013	<0,01	<0,01	<0,01	<0,01	<0,01	<0,01
4	Jobboulch	<0,10	0 < 0,10	) <0,30	0,024	3,20	0,027	0,040	<0,01	0,019	<0,01	<0,01	<0,01	<0,01	<0,01	<0,01
5	1330x2125	<0,10	0 < 0,10	) <0,30	0,097	5,70	0,080	0,220	0,019	0,081	0,013	<0,01	<0,01	<0,01	<0,01	<0,01
9	Ratan x 1307	<0,10	0 < 0, 10	) <0,30	0,023	4,90	<0,01	0,016	<0,01	<0,01	<0,01	<0,01	<0,01	<0,01	<0,01	<0,01
7	Ratan IG 135481	<0,10	0 < 0,10	) <0,30	0,041	4,10	<0,01	0,024	<0,01	<0,01	<0,01	<0,01	<0,01	<0,01	<0,01	<0,01
8	Bio 520xBio??x274	<0,10	0 < 0,10	) <0,30	0,018	4,60	<0,01	0,019	<0,01	<0,01	<0,01	<0,01	<0,01	<0,01	<0,01	<0,01
6	Pratck x IG 140035	<0,10	0 < 0,10	) <0,30	0,050	4,10	0,011	0,027	<0,01	<0,01	<0,01	<0,01	<0,01	<0,01	<0,01	<0,01
10	Bio 520x1330	<0,10	0 < 0,10	) <0,30	0,018	5,50	<0,01	0,017	<0,01	<0,01	<0,01	<0,01	<0,01	<0,01	<0,01	<0,01
11	Lalmikor	<0,10	0 < 0, 10	) <0,30	0,027	2,80	0,026	0,049	<0,01	0,019	<0,01	<0,01	<0,01	<0,01	<0,01	<0,01
12	Polvon	<0,10	0 < 0,10	) <0,30	0,017	8,40	0,020	0,036	<0,01	0,011	<0,01	<0,01	<0,01	<0,01	<0,01	<0,01
ŝ	Elements	Er	Tm	Υb	Γn	Hf	Ta	M	Re	Pt*	Au*	II	Pb	B	Th	n
Σ	Measurement range elements	0,05- 4000	0,05- 4000	0,10- 4000	0,004- 11%	0,004- 11%	0,002- 20%	1,0- $4000$	0,008- 30%	0,005- 28%	0,10- 4000	0,0006- 9%	0,10- 4000	1,0- 4000	0,002- 10%	0,006- 30%
	Bio 520xBio??x273	<0,01	<0,01	<0,01	<0,01	<0,05	<0,04	<0,08	<0,01	<0,05	<0,05	<0,05	0,620	<0,0	<0,01	0,010
2	Pratek x IG 140034	<0,01	<0,01	<0,01	<0,01	<0,05	<0,04	<0,08	<0,01	<0,05	<0,05	<0,05	0,700	<0,01	<0,01	0,013
3	Ratan x 2125	<0,01	<0,01	<0,01	<0,01	<0,05	<0,04	<0,08	<0,01	<0,05	<0,05	<0,05	0,600	<0,01	<0,01	0,013
4	Jobbouleh	<0,01	<0,01	<0,01	<0,01	<0,05	<0,04	<0,08	<0'01	<0,05	<0,05	<0,05	0,770	<0,01	0,012	0,010
5	1330x2125	<0,01	<0,01	<0,01	<0,01	<0,05	<0,04	<0,08	<0'01	<0,05	<0,05	<0,05	0,770	<0,01	0,049	0,018
6	Ratan x 1307	<0,01	<0,01	<0,01	<0,01	<0,05	<0,04	<0,08	<0,01	<0,05	<0,05	<0,05	0,230	<0,01	<0,01	<0,01
7	Ratan IG 135481	<0,01	<0,01	<0,01	<0,01	<0,05	<0,04	<0,08	<0,01	<0,05	<0,05	<0,05	0,260	<0,01	<0,01	<0,01
8	Bio 520xBio??x274	<0,01	<0,01	<0,01	<0,01	<0,05	<0,04	<0,08	<0,01	<0,05	<0,05	<0,05	0,290	<0,01	0,029	0,011
6	Pratek x IG 140035	<0,01	<0,01	<0,01	<0,01	<0,05	<0,04	<0,08	<0,01	<0,05	<0,05	<0,05	0,530	<0,01	<0,01	<0,01
10	Bio 520x1330	<0,01	<0,01	<0,01	<0,01	<0,05	<0,04	<0,08	<0,01	<0,05	<0,05	<0,05	0,200	<0,01	<0,01	<0,01
11	Lalmikor	<0,01	<0,01	<0,01	<0,01	<0,05	<0,04	<0,08	<0,01	<0,05	<0,05	<0,05	0,380	<0,01	0,011	0,010
12	Polvon	<0,01	<0,01	<0,01	<0,01	<0,05	<0,04	<0,08	<0,01	<0,05	<0,05	<0,05	0,340	<0,01	0,006	0,010

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# COLLECTION OF LANDRACE WHEAT VARIETIES BRED IN DIFFERENT ECOLOGICAL ZONES OF UZBEKISTAN AND STUDY OF VALUABLE ECONOMIC TRAITS

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*Summary.* Expansion of area under cultivation of intensive high productive commercial wheat cultivars brought decreasing of areas on which wheat landraces had been cultivated. However, wheat landraces were being kept by local farmers in remote districts where no intensive cultivars had been spread widely. Employment of valuable genetic resources of locally originated cultivars is im portant in plant breeding programs. The aim of current study is to characterize landraces of winter wheat of Uzbekistan by morphological and quality traits (e.g. plant heights, yielding, etc.) with geo-information data on the points of landraces origin, and also to analyze elements of crop productivity. Wheat landraces such as Kzyl-bugday, Ak-bugday, Tyuya-Tish, unnamed landrace and also Surhak (created from local landrace) were used in this study.

**Keywords:** Wheat landraces, morphological analysis, GPS-navigation, quantitative traits.

# КОЛЛЕКЦИЯ СТАРОДАВНИХ МЕСТНЫХ СОРТОВ ПШЕНИЦЫ ВОЗДЕЛЫВАЕМЫХ В РАЗНЫХ ЭКОЛОГИЧЕСКИХ ЗОНАХ УЗБЕКИСТАНА И ИЗУЧЕНИЕ ЦЕННЫХ ХОЗЯЙСТВЕННЫХ ПРИЗНАКОВ.

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Аннотация. Расширение площадей под интенсивными высокоурожайными сортами привело к уменьшению территорий, на которых высевают стародавние сорта. Эти сорта сохранялись лишь местными фермерами в где не проводилось широкого отдаленных частях страны, внедрения коммерческих сортов. Использование ценных генетических ресурсов местного происхождения исключительно важно для селекционных проектов. Цель настоящей работы охарактеризовать стародавние сорта мягкой пшеницы Узбекистана по морфологическим и количественным признакам (высота растений, продуктивность) с учетом геоинформационных характеристик мест возделывания, а также провести анализ элементов структуры урожая. Объектами исследования служили стародавние местные сорта пшеницы Кзыл-бугдай, Акбугдай, Тюя-Тышь, безымянный местный сорт, а также сорт Сурхак, выведенный из популяции местного стародавнего сорта.

*Ключевые слова*: Стародавнего сорта пшеница, морфологический анализ, GPS-навигация, количественные признаки.

# Introduction

Over the past decade, wheat has been grown on about 218.5 million hectares worldwide, with an average annual yield of 740 million tons and an average yield of 3.4 tons per hectare [7].

About 95% of the world's wheat is soft wheat and the remaining 5% is durum wheat [11; 13], which is the main food crop of Central Asian peoples.

Crop breeding is a major challenge because of the very narrow genetic diversity of most crops. The solution to this problem may be the involvement of genetic materials of wild close ancestors and species, i.e. overcoming erosion by adding the secondary gene pool to the primary gene pool. At the same time, reduction of the natural range of potential donor species and storage of a small number of seeds of their few representatives in genebanks leads to reduction of their polymorphism and, as a result, to the loss of the secondary gene pool, which inevitably limits the opportunities for increasing biodiversity [17].

Modern wheat varieties are mostly genetically similar and have a narrow genetic base. The genetic base of local varieties developed by farmers through natural selection for yield is broader and may include important aspects for breeding. Such varieties are characterized by tolerance to local stress factors, stable productivity, wide diversity in protein content and differ from commercial varieties in grain quality, so these varieties are valuable gene pools and play an important role as an initial source for developing new varieties [15].

Jaradat A.A. and others [6] note that landrace wheat varieties were developed by farmers as a result of many years of natural and artificial selection and adapted to local conditions. As distinct а plant population, ancient local varieties were planted by the population to fulfill social, economic, cultural and ecological needs.

Newton's [10] research shows that depletion of genetic resources and genetic erosion are serious problems in agriculture in many countries. Today, the replacement of ancient indigenous wheat varieties with varieties resistant to pests, diseases, biotic and abiotic factors, developed with the help of modern agriculture and its huge investments, has led to a decline in genetic diversity. He noted that it could be observed that in areas where indigenous wheat had been cultivated since ancient times, its potential had not been fully utilized.

Harlan J.R. [14] in his scientific paper describes ancient local varieties as "balanced populations, that is, variable and genetically dynamic, in equilibrium with the environment and pathogens". It was emphasized that if the seeds of a INTERNATIONAL SCIENTIFIC JOURNAL "MODERN BIOLOGY AND GENETICS" 2025 №2 (12) ISSN: 2181-3396

variety have been planted in a region for at least one generation, they are considered "local" [8].

When describing "landrace" varieties, A. S. Zeven emphasizes that due to their complex and ambiguous nature it is impossible to give a precise definition, but he offers the following: "landrace varieties are resistant to biotic and abiotic stresses, resulting in stable low input yields under and economically marginal agricultural systems" [6].

Landrace wheat varieties have diverse populations and usually get their names from local people. These varieties are adapted to the environmental conditions of the growing region (resistant to biotic and abiotic stresses) and their yields need to be improved [2].

Rodriguez et al. observed that landrace wheat varieties have considerable diversity in terms of protein content and some grain quality traits compared to modern commercial varieties [12].

#### Materials and methods

In modern grain production, modern intensive commercial varieties have displaced landrace wheat varieties, which have survived only in small farms, for example, in the mountainous regions of Uzbekistan.

The subjects of the study were soft spring wheat landraces such as Kzyl-bugday, Ak-bugday (Graecum), Tyuya-Tish, Pashmak, Khivit, Boboki, Muslimka, Kairaktash, Kzyl-Shark, Unnamed, and Surkhak (bred from a local landrace).

During scientific expeditions organized over a number of years in the Table 1). mountainous and foothill areas of Surkhandarya, Kashkadarya and Jizzak regions of the Republic, samples of ancient local wheat varieties grown on the lands of individual peasant farms in these regions were collected and studied for morphological traits (ear length, grain, ear color). These varieties were included in the collection as 24 major ancient local wheat varieties based on distribution their and breeding significance (see

# **Results and discussion**

It is noted in the literature that the study of landrace wheat varieties, to which no breeding methods have been applied, as a promising source material 80 is undoubtedly an urgent problem of genetics and plant breeding [1; 3; 4; 5;9;].

Over thousands of years of farming in the Central Asian region,

wheat varieties sufficiently resistant to drought and hot weather conditions have been developed. These wheat varieties are characterized by early maturity and relatively high resistance to heat in the last stages of development. In regions with different altitudes, differences in soil moisture levels during the growing season are observed due to different weather conditions. In particular, specific features of wheat varieties grown in plain, foothill and mountainous areas are related to the coarseness of plant components, i.e. the grain is covered with a dense outer shell, it is difficult to crush, and the cob on the cob breaks easily, etc. Unique natural and climatic conditions of Central Asia annually create favorable conditions for intensive development of various diseases and entoparasitic pests in agriculture, and local wheat varieties are exposed to unfavorable external conditions under these conditions. In particular, under conditions of intensive agrotechnics it was noted that the stem of wheat varieties is not strong, the degree of brittleness is high, and the tendency to lodging is high.

In particular, most of the above traits are observed in wheat varieties in irrigated fields, wheat grown varieties grown in arid conditions show relative stem strength and formation of a peculiar type of ear. Wheat varieties used for irrigated agriculture in the highland areas of Central Asia are mesophilic characterized and by relatively soft ears.

During the research, expeditions to a number of districts of Uzbekistan were organized to collect and study landrace wheat varieties. Collection of landrace wheat varieties was conducted by surveying the population in mountainous, steppe and semi-desert areas of the republic. A special GPS device was used to determine the place of sample collection and a special form prepared for the survey was filled in. This form collected information such as location and time of sample collection, name of the village or farmer, local name of the variety, time of planting and harvesting, planting method and criteria, origin of the seeds taken for planting, where and how the seeds were stored, how they were prepared for planting, and the purpose of planting these local varieties. From each field, 200 ears were randomly sampled. Each sample was morphologically analyzed based on the color and shape of the ear and grain to determine to which species or variety it belonged. The composition of weeds and other species in each field was approximated and recorded.

Two expeditions to Surkhandarya province and one expedition to Jizzak province were organized during the research. The first expedition was organized at the end of July in Altynsoy district of Surkhandarya province. They went to the village of Loka, which is located at an altitude of 1,300 meters above sea level. In this village, it was found that an ancient variety of wheat belonging to 81

Kzyl-bugday variety is mainly the cultivated. We drove through the village to Vakhsh village which is located at an altitude of 1400 meters above sea level. Surveys were conducted among the local villagers living in the countryside regarding the methods of growing wheat and other crops in their fields. In the region, it is found that an oldgrowth wheat variety belonging to the Red Wheat variety is cultivated, the yield of this wheat variety averages 25 tons/ha. The wheat fields are sown mainly by hand, the crop is also harvested by hand, and machinery is used on more than 20 hectares of land. The second expedition was organized in September. In this case they went to Uzun district, which is 1600-1700 meters above sea level. The population of the area is mainly engaged in cattle breeding. Old varieties of wheat are grown in the region - "Pashmak", "Kzyl-bugday", and in some districts "Khivit" is also grown. This variety of is interesting because wheat its vegetation period lasts almost a year. The wheat is sown in June-July and harvested the following year in July, sometimes in August.

This variety of wheat differs from other ancient varieties in the quality of bread baked from it, yield, large grain size, shell strength and, most importantly, extreme frost resistance.

During the trip to Bakhmal and Zamin districts of Jizzak oblast, we continued collecting samples of old wheat varieties. However, 82 unfortunately, we failed to find new wheat varieties in this region. In these districts, the ancient variety "Surkhak" and only in some places "Ak-bugday" are grown almost everywhere. Samples of wheat varieties collected from the region were planted in an experimental plot for comparison with previously collected varieties. In this case, many wheat varieties have the same name, so during our research we selected wheat ears for each individual wheat variety identified and grew them under neardry conditions, i.e. without irrigation throughout the growing season.

Thus, samples of 31 landace wheat varieties were collected and their geographical distribution and phenological indices were studied (Table 1).

The table shows that most of these varieties are spring wheat sown in March, but in some regions they are also sown in the fall, indicating that these varieties are double-cropped and have adapted to these regions through many years of reseeding. Despite the fact that these varieties have been grown only in peasant farms for several years and no breeding work has been done, it has been observed that in most cases they consist of pure, homogeneous rows and in some varieties up to 15-20% of other varieties and barley are admixed. Since the population paid more attention to grain color when naming varieties, most varieties were named after red or white wheat.

Field experiments were conducted on samples of old local varieties during three years with the same agronomic measures. In the first year of experiments, 10 ears were taken from each sample, planted separately on a plot of 1 m2 and their valuable economic characteristics (ear length, number of spikelets in an ear, number of grains in an ear, grain weight) were studied. The most efficient ears were selected and planted the following year (Table 2).

If we consider the results of statistical analysis of yield elements of the second year, the number of spikelets per ear averaged 18 in red wheat and 17 in white wheat, and the number of grains averaged 44 and 38, respectively. The weight of 1000 grains of most of the studied varieties was found to be 45-50 grams and their grains were larger than those of commercial wheat varieties.

In the third year, samples of oldgrowth local varieties were planted in spring and phenological observations were made, including evaluation of wheat varieties for rust resistance, lodging, and yield performance. The wheat varieties included in this nursery were planted in parallel for the third year in the field in the fall. Analysis of the results obtained (Table 3) When samples sown under fall and spring conditions were compared, the earing phase in wheat varieties sown in the fall occurred 117-122 days from January 1, whereas when sown under spring conditions, it occurred in 60 days for early maturing samples and 68 days for relatively late maturing samples. These analyses also showed that redgrained wheat tended to mature later than white-grained wheat.

When landrace wheat varieties were planted under irrigated conditions, severe yellow rust infestation was observed. All wheat varieties planted in fall showed heavy infestation, while spring samples showed relatively lower infestation.

Grain size or fullness is mainly determined by the weight of 1000 grains. When analyzed for this trait, the weight of 1000 grains of red wheat varieties planted in fall were all above 50 grams, whereas these varieties were significantly smaller with an average of 42 grams when planted in spring. Nevertheless, it was observed that the total grain yield was almost similar in fall and spring planting and averaged 350-450 grams.

# Conclusions

Thus, the traditional landraces of Uzbekistan have many traits useful for selection. The collection and study of these landraces are of great importance for preserving the wheat gene pool in the natural conditions.

It was observed that there was a clear difference in the analyzed parameters, namely: the height of wheat 83 plants planted in autumn was relatively high, and there was a higher probability of stem lodging and a higher incidence of yellow rust. In contrast, wheat plants in spring fields had relatively average height, low probability of stem lodging, and low incidence of yellow rust.

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Geogra	phical location and d	escription of traditional	Geographical location and description of traditional wheat landraces sampled		Table 1
Latitude, longitude; elevation, m	Cultivar	Sowing time	Pure line or impurity (%) and impurity type in a mixture	Grain color	Plant height, cm
	Surkhandaryinskaya	inskaya region Baisunsky District Duoba Kishlak.	ct Duoba Kishlak.		
38°32'121"N, 67°38'181"E;1391	Kzyl-bugday	March	Mixture, 2 %, various	Red	120
		Kurgancha Kishlak	-		
38°37'921"N, 67°41'462"E;1633	Kzyl-bugday	March	Mixture, 5 %, having red ears	Red	120
		<b>Gumatak Kishlak</b>			
38°35'699"N, 67°37'737"E; 2136	Kzyl-bugday	March	Pure line	Red	130
38°35'986"N, 67°07'070"E;2174	Surkhak	March	Mixture, 5 %, barley	Red	130
38°35'057"N, 67°42'538"E;2143	Kalbugday	March	Pure line	Red	100
		Pulkhokim Kishlak			
38°16'484"N, 67°38'905"E;1050	Buhkar bobo	October	Pure line	Red	140
38°16′496″N, 67°38′715″E;1180	Kzyl bashak	October	Mixture, 5 %, various	White	120
38°16'600"N, 67°38'760"E;1190	Ak bashak	October	Mixture, 20 %, having red	Red	100
			ears		
38°16′645″N, 67°38′750″E;1090	Boysun Tura-1	October	Pure line	White	110
38°16'580''N, 67°38'565''E;1100	Boysun Tura-2	October	Pure line	Red	120
38°16'500"N, 67°38'800"E;1125	Kairaktash	March	Mixture, 15 %, having red	White	100
			ears		
k	ashkadaryinskaya re	Kashkadaryinskaya region Yakkabagsky District Guldara Kishlak	ict Guldara Kishlak		
38°78'582"N, 66°81'014"E;1159	Ak-bugday	March	Pure line	White	120
38°77'369″N, 66°82'451″E;1270	Graecum	March	Mixture, 20 %, Surkhak	White	120
			landrace		

		Terakli Kishlak			
38°75'540"N, 66°81'783"E;1634	Unnamed	March	Mixture, 20 %, various	Red	100
38°75'934"N, 66°82'558"E;1500	Korakiltik	November	Pure line	Red	120
	Kamasł	Kamashinsky District Kuga Kishlak	hlak		
38°66'376"N, 66°92'626"E;2249	Ak-bugday	March	Pure line	White	120
38°63'243"E, 66°94'461"E;1988	Kzyl-Shark	March	Pure line	Red	90
38°64'701"N, 66°93'114"E; 1731	Kzyl-bugday	March	Pure line	Red	120
		<b>Kzyltom Kishlak</b>			
38°61'663"N, 66°93'731"E;1753	Tyuya-Tish	March	Mixture, 15 %, having red	Red	120
			ears		
38°66'376"N, 66°92'626"E;2249	Unnamed	March	Mixture, 20 %, having red	Red	100
			ears		
38°65'243"N, 66°90'205"E;2147	Muslimka	March	Mixture, 20 %, various	Red	100
38°59'266"N, 66°91'480"E;1317	Surkhak	March	Pure line	Red	130
	Dzhizakskaya regioi	region Bakhmalsky District Muzbulak Kishlak	fuzbulak Kishlak		
39°71'376"N, 68°12'882"E;1520	Graecum	October	Pure line	White	115
	Gallyaara	lyaaralsky District Yonbosh Kishlak	ishlak		
39°70'017"N, 68°19'329"E;1763	Surkhak	March	Mixture, 10 %, barley	Red	120
	Surkhandaryi	Surkhandaryinskaya region Saryasiysky District	ky District		
38°33'086"N, 67°65'667"E;1301	Kzyl-bugday	October	Pure line	Red	105
38°34'088"N, 67°65'678"E;1361	Kzyl-Shark	March	Mixture, 20 %, various	Red	110
		Uzunsky District			
38°61'500"N, 67°58'411"E; 2008	Pashmak	October	Pure line	White	90
38°60'202"N, 67°56'589"E; 1650	Khivit	August	Pure line	White	90
38°57'685"N, 67°58'622"E;1558	Kzyl-bugday	March	Pure line	Red	110
38°61'500"N, 67°58'521"E; 1710	Surkhak	October	Mixture, 15 %, having red	Red	110
			ears		

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				Terakli Kishlak	shlak				
Unnamed	16,5±0,34	10,2±0,47	34,9±2,58	1,9±0,15	16,9±0,41	10,3±0,21	37,7±1,14	$1,8\pm 0,08$	36,7±2,21
Korakiltik	16,2±0,28	11,1±0,56	36,2±1,23	2,0±0,12	16,0±0,75	11,0±0,32	35,6±1,23	1,9±0,06	35,6±2,13
			Kam	ashinsky Distri	Kamashinsky District Kuga Kishlak	-			
Ak-bugday	13,6±0,35	8,2±0,62	32,0±2,01	$1,7\pm 0,23$	16,2±0,63	11,2±0,26	34,5±2,60	2,0±0,18	46,8±2,36
Kzyl-Shark	13,4±0,43	8,3±0,33	31,0±2,46	1,3±0,13	16,4±0,34	11,7±0,75	31,2±1,58	1,8±0,12	46,9±2,23
Kzyl-bugday	$14,5\pm 0,27$	9,1±0,23	34,0±1,47	1,9±0,10	18,2±0,65	12,5±0,75	38,1±4,01	1,2±0,23	29,1±3,5
	-	-	-	Kzyltom Kishlak	ishlak	-	-	-	-
Tyuya-Tish	$14,4\pm 0,22$	9,0±0,21	27,0±1,77	1,5±0,15	18,2±0,39	12,6±0,43	35,1±1,99	1,6±0,12	45,7±2,78
Unnamed	$13,6\pm 0,40$	7,8±0,20	27,4±1,16	1,7±0,08	18,5±0,48	13,1±0,57	45,7±2,04	1,9±0,17	41,7±2,38
Muslimka	15,2±0,36	9,3±0,33	33,1±1,86	1,8±0,15	19,3±0,56	13,9±0,66	46,0±3,01	1,9±0,21	41,1±2,49
Surkhak	12,2±0,25	7,1±0,18	28,5±1,62	1,3±0,08	17,2±0,61	11,1±0,60	42,4±3,07	2,1±0,19	48,0±1,31
	-	D	Dzhizakskaya reg	jon Bakhmalsk	a region Bakhmalsky District Muzbulak Kishlak	ulak Kishlak	_	-	-
Graecum	$14,6\pm 0,34$	9,3±0,42	38,0±1,90	$1,7\pm 0,10$	18,8±0,55	12,5±0,45	52,8±3,47	2,5±0,28	46,0±2,84
		-	Gallya	aralsky District	Gallyaaralsky District Yonbosh Kishlak	k	-	-	-
Surkhak	14,6±0,33	9,2±0,26	30,2±2,01	$1,6\pm 0,09$	16,0±0,62	11,2±0,31	38,6±1,02	2,0±0,13	44,2±1,56
			_						

UDC:631.5

#### VALUABLE ECONOMIC TRAITS OF TRITICALE

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*Summary.* In Uzbekistan, during a period of rapid population growth, the creation and implementation of high-yielding varieties of cereal crops with high protein and energy content, adapted to all climatic conditions and with high yield potential, is a pressing task. Triticale grain contains a higher amount of essential amino acids that cannot be freely substituted in wheat, such as lysine, aspartic acid, arginine, and others. Therefore, the biological value of triticale is higher than that of wheat. According to its biological characteristics, winter triticale is cold-resistant, which is why the distribution of triticale is gradually expanding.

*Keywords:* Triticosecale, productivity, disease, pest, short-stemmed, amphidiploid, gluten, drought

# ЦЕННЫЕ ХОЗЯЙСТВЕННЫЕ СВОЙСТВА ТРИТИКАЛЕ

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#### Introduction

Scientific research is being conducted worldwide to create new highyielding, disease and pest-resistant, and adaptable to various soil-climatic and ecological conditions selection varieties of cereal crops with high protein content and good nutritional quality, including triticale, an artificially created plant by breeder scientists, to ensure global food security [2].

Triticale is a new grain crop obtained by crossing wheat and rye. It is an amphidiploid of wheat-rye hybrids and is currently classified as a separate independent polymorphic botanical genus Triticosecale. The name Triticale is formed by combining the first parts of the words Triticum (wheat) and Secale (rye).

Triticale belongs to the wheat-rye hybrid amphidiploids - a grain crop that embodies the characteristics of two different plants. Many characteristics of triticale have not yet been deeply studied, which significantly hinders the improvement of complex intergeneric hybrids. For example, incomplete ear development in octoploid forms of triticale, the tendency of some forms to cross-pollination, shriveled grains, and brittle ears. At the same time, triticale significantly surpasses wheat in disease resistance, protein and lysine content in the grain, greater winter hardiness, and spring forms of triticale are much more droughtresistant than spring wheat. Today, the main countries cultivating triticale are Germany, Belarus, France, Russia, China, Hungary, Spain, and Australia. The first wheat-rye hybrid was created in 1875 by the Scottish scientist Wilson [3].

The first fertile wheat-rye amphidiploids were obtained by the German breeder Rimpau in 1888 by crossing the F-1 wheat-rye hybrid generation with their ancestors [4, 6].

In 1921, G.K. Meister observed occasional pollination of wheat plants by rve pollen from neighboring plots. However, these hybrids were mostly male sterile, and rarely were fertile hybrids created due to spontaneous doubling of The next fundamental chromosomes. success occurred in 1937 when botanists learned that colchicine could "double" the chromosomes in newly formed cells. The introduction of in vitro embryo rescue and chemical treatment of two chromosomes with colchicine made it possible to regularly produce viable seeds (Oettler 2005).

The person who laid the foundation for obtaining wheat-rye hybrids was Y. Chermak, who, starting in 1902, crossed wild and cultivated rye species with of wheat to obtain various types productive wheat-rye hybrids of great economic importance. Currently, octoploid 56-chromosome hybrids of triticale by crossing winter and spring soft wheat with hexaploid and 42-chromosome rye, hybrids by crossing durum wheat with rye have been created [1].

Hexaploid hybrids have many grains in a high-quality ear, with a higher protein content in the grain and a higher yield compared to octoploid hybrids. In recent years, 3 types of hybrids have been created (soft wheat, durum wheat, and rye), which combine the hereditary characteristics of durum wheat, soft wheat, and rye [3].

In Russia, V.E. Pisarev conducted selection for the creation of 56chromosome triticale. He created a 56chromosome amphidiploid triticale by crossing soft wheat with rye. By treating the wheat-rye hybrid with colchicine, the number of chromosomes was doubled to create a 56-chromosome amphidiploid.

At the Ukrainian Institute of Plant Breeding, Genetics and Selection, A.F. Shulindin created a 42-chromosome amphidiploid – triticale – by crossing triticale with rye [5].

In triticale breeding, the world gene pool collection nursery of varieties and samples with distant geographical origin of triticale is used to create initial sources and evaluate introduced varieties based on the results of ecological variety trials in regional conditions. The world gene pool collection nursery includes 185 varieties and samples of triticale. The collection nursery mainly consists of world gene pool varieties and samples of triticale obtained from the P.P. Lukyanenko Krasnodar National Grain Center (Krasnodar Territory), the Nemchinovka Agricultural Research Institute (Moscow Region) (Russia), the Institute of Plant Genetic Resources, as well as the international

N⁰	Geographic origin	Country names	Items, quantity
1.	Eastern Europe	Russia, Hungary, Serbia	88
2.	Western Europe	France, Italy, Denmark	30
3.	Central Asia	Uzbekistan, Kyrgyzstan, Kazakhstan	20
4.	Front Asia	Turkey, Syria	37
5.	Central America	Mexico	10
	Total:	12	185

scientific centers CIMMYT, ICARDA, and ISRISAD.

At the Bosh Botanical Garden, amphidiploids with 56 chromosomes were crossed with forms having 42 chromosomes. For the wheat-rye amphidiploids, A21 and A24 with 2n=56 served as the initial forms. These were well winter-hardy, resistant to fungal diseases, and matured at the same time as the PPG-599 winter wheat. These forms crossed with wheat-rye were amphidiploids brought from Hungary, in the origin of which T. Vusalle turgidum var. and Madyarovari rye were involved. As a result of such crosses, heterotic hybrid forms of amphidiploids with well-filled ears were obtained. The somatic cells of the new hybrid forms contained 42 chromosomes.

Their chromosome set was formed as a result of the combination of the AV genome of triticale (14 chromosomes), the AB genome of soft wheat (14 chromosomes), and two genomes of rye (14 chromosomes). Some of the resulting amphidiploids were not inferior to the regionalized wheat varieties in terms of winter hardiness, contained 29-30 spikelets and 65-70 grains in the ear, had a 1000grain weight of 48-51 g, and their protein content was 3-4% higher than that of the regionalized winter wheat varieties, with some forms having up to 19.2% protein in the grain. Subsequent selection involved crossing the octoploid amphidiploids Myunsing and V.Y. Pisarev's AD 778-79, and in the second generation of offspring obtained from crossing the hexaploid Meksikanskiy-131, ears with 90 grains were observed. In the cross AD 119 x AD Derjavin, many plants had ears with 70 grains, whereas in the control winter wheat, this figure averaged 48.

## Table 2

Table 1

		(0)
Amino acids	Wheat flour	Triticale
Lysine	17,9	19,6
Valine	27,6	24,2
Leucine	45,0	41,2
Isoleucine	20,4	18,7

Triticale grain quantity indicators, (Umirov 2016)

Methionine	9,4	6,0
Threonine	18,3	19,6
Tryptophan	6,8	6,3
Phenylalanine	28,2	28,6
Cystine	15,9	7,9
Tyrosine	18,7	19,5
Arginine	28,8	38,2
Histidine	14,3	13,3
Alanine	22,6	25,8
Aspartic acid	30,8	41,6
Glutamic acid	186,6	152,8
Glycine	25,4	26,5
Proline	62,1	52,1
Serine	28,7	25,0

The grain of triticale contains more essential lysine, aspartic acid, arginine, and other amino acids compared to wheat, therefore the biological value of triticale is higher than that of wheat (Table 1). The composition of triticale grain is: water 14.0%, protein \_ 12.8%, carbohydrates – 68.6%, fat – 1.5%, fiber -3.1%, and ash -2.0%. The endosperm contains 26-28% water-soluble proteins, 7-8% salt-soluble proteins, 25-26% alcohol-soluble proteins, and 18-20% acetic acid-soluble proteins, which is why the biological value of triticale is higher than that of wheat. Triticale grain has a higher ash content, а lower content of carbohydrate components, and is distinguished by the presence of rye's specific trifructose carbohydrate. The protein in triticale grain contains an average of 5-10% albumin, 6-7% globulin, 60-37% prolamine, and 15-20% glutamine. The main part of

triticale grain is starch, which accounts for <sup>3</sup>/<sub>4</sub> of the grain weight.

In terms of the content of glutenforming proteins, it approaches wheat. However, due to the presence of rye proteins, the quality of its gluten is often lower. Although the selection of triticale crops began in Uzbekistan in 1974, currently 4 varieties have been created and regionalized. In Uzbekistan, the grain yield of the regionalized Prag and Bahodir varieties in 1989 at the Fergana variety testing station was 81 and 72.7 c/ha, and the mass of dry matter was 224.5 and 225 c/ha.

In addition, when assessing lodging and winter hardiness, and resistance to diseases before harvesting, the samples received 9 points, while the standard variety received 7 points. The grain weight (1000 grains) of these promising triticale samples was 43.2-53.2 g, while in the standard it was 40.4 g. Furthermore, these samples are resistant to drought and high temperatures. According to its biological characteristics, winter triticale is coldtherefore the distribution of resistant, triticale is gradually expanding. Triticale does not die during the winter when the ground freezes under the snow. The cause of plant death varies in different regions, depending on living conditions, factors, and characteristics. The varietal main susceptible tillering node. Injury to the tillering node is equivalent to the death of the plant.

With the onset of the spring growing season, severe frosts sometimes occur. This causes significant damage to the plant, especially if the plant is not cold-resistant, which often happens in years with unexpected severe frosts in the spring. Cold resistance varies depending on the growth phase and development of triticale varieties. Drought is sometimes associated with heat resistance. This is also related to the stage of development. The booting and heading stages are the most critical periods for drought. Heat resistance is most critical during the heading and milk development stages. Triticale is a moderately droughtresistant crop, so it is mainly grown on irrigated land.

In Uzbekistan, triticale is mainly irrigated land. grown on Irrigation contributes to good growth and development of the plant, increases the number of leaves, tillering, and the number of grains in the ear, but accelerates lodging. Increased tillering and yield can be observed under irrigation. Triticale has moderate resistance to soil salinity. In saline soils, seedlings emerge late, and the growing season is shortened. In such 92

conditions, it is necessary to study the response of varieties to salinity. The response of varieties to salinity becomes the initial stages apparent in of development. Winter triticale has been shown to be less susceptible to salinity. Our scientists have determined the salt tolerance of triticale, which makes it possible to create salt-tolerant, high-yielding promising triticale varieties.

Scientific research conducted by P. Prosviryak (2009) in the soil and climatic conditions of Ukraine showed that moderate fertilization during the spring cultivation of triticale allows for additional yield. Also, due to the high resistance of spring triticale varieties to fungal diseases, seed treatment before sowing is not required to protect against fungal diseases. Boboyorov (1991) notes that selection achievements in creating high-yielding triticale varieties are associated with taking into account genetic factors and, of course, the semi-dwarfism factor. The problem of lodging resistance can be solved in two ways: by breeding non-lodging varieties through selection and by using chemical agents that shorten the stem length (Shomurodov et al., 1998; Oripov, 1989; Rashidov, 1986). Lodging resistance often depends on the anatomical structure of the stem, which determines the stem's elasticity in relation to fracture. The anatomical structure of the stem is genetically determined, but light strongly influences the development of mechanical elements of the root and stem tissues. Assessing lodging resistance, considering only the underground part of the plant, is not

entirely complete, since the structure of the root system is also of great importance in this regard.

In modern, high-yielding varieties, according to V.N. Remeslo (1996), there is a tendency for an increasing inverse relationship between yield and protein content. This is interpreted as meaning that more productive forms (10-15 c/ha) require more soil nitrogen to increase yield (Sobirov, 1991; Soatov et al.; Khotamov,

1994). Triticale is resistant to many diseases characteristic of grain crops. It is practically not affected by powdery mildew, hard and smut, brown rust. loose and The disadvantages of triticale include sharp fluctuations in yield from year to year, susceptibility to lodging, sprouting of grain in the ear, and the incomplete filling of grain in some forms, late maturity, and susceptibility to snow mold and root rot diseases.

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# THE ROLE OF MIRNAS IN SALT STRESS TOLERANCE IN COTTON (GOSSYPIUM SPP.) SPECIES

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*Abstract.* MicroRNAs (miRNAs) are a class of endogenous non-coding small RNAs (20–24 nucleotides in length) that regulate gene expression by degrading target mRNAs or inhibiting their translation. During plant vegetative growth, miRNAs act as post-transcriptional regulators responding to various abiotic and biotic stresses. miRNAs control gene expression not only through direct binding to target genes but also indirectly via transcription factors. They play key roles in plant growth and development, morphogenesis, and stress responses. Typically, the expression of miRNAs is upregulated under stress, leading to the degradation of target mRNAs or increased expression of positive regulatory genes. While plant miRNAs were first discovered between 2001 and 2004, their investigation in Gossypium species began around 2007. Advances in technology have significantly accelerated the study of miRNAs in cotton, especially those associated with responses to environmental stresses such as salinity and drought. This article reviews miRNAs identified in Gossypium species under salt stress conditions.

*Keywords:* miRNA, Gossypium hirsutum, microRNA expression analysis, expression level, overexpression, short tandem target mimic (STTM).

# РОЛЬ МИКРОРНК В УСТОЙЧИВОСТИ ВИДОВ ХЛОПЧАТНИКА (GOSSYPIUM SPP.) К СОЛЕВОМУ СТРЕССУ

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Аннотация. МикроРНК (миРНК) - это класс эндогенных некодирующих малых РНК (длиной 20-24 нуклеотида), которые регулируют экспрессию генов путем деградации мРНК-мишеней или ингибирования трансляции. В процессе вегетации растений миРНК играют важную роль в реакции на различные абиотические и биотические стрессоры, действуя на посттранскрипционном уровне. Они регулируют экспрессию генов как напрямую, связываясь с мРНК, так и опосредованно — через транскрипционные факторы. миРНК участвуют в регуляции роста и развития растений, морфогенеза и ответа на стрессовые условия. Как правило, при стрессовом воздействии экспрессия миРНК усиливается, что приводит к деградации мРНК-мишеней или усилению экспрессии положительных регуляторов. Хотя миРНК были открыты в растениях в период 2001–2004 годов, у видов рода Gossypium их начали активно исследовать с 2007 года. Развитие технологий позволило активизировать исследования миРНК у хлопчатника, особенно тех, которые связаны с ответом на засоление и засуху. В данной статье приведён обзор миРНК, идентифицированных у представителей рода Gossypium в условиях солевого стресса.

**Ключевые слова:** миРНК, Gossypium hirsutum, анализ экспрессии микроРНК, уровень экспрессии, сверхэкспрессия, короткий тандемный мимик-мишень (STTM).

## Introduction

Soil salinization is one of the major ecological problems expanding globally. According to FAO data, based on research conducted in 118 countries, more than 424 million hectares of the world's topsoil (0-30 cm) and 833 million hectares of subsoil (30–100 cm) are affected by varying levels of salt Salinization contamination [1]. growth, negatively impacts plant development, and productivity, posing a serious challenge to crop cultivation. When salt concentration exceeds normal levels, the water potential in the soil decreases, reducing water uptake by plant roots. The effects of salinity stress on plants are classified into three main levels: hyperosmotic stress, ion toxicity, and oxidative stress [2].

To enhance salt tolerance in plants, classical transgenic approaches typically involve boosting endogenous defense mechanisms, often through a single-gene strategy. This usually includes enhancing the synthesis of compatible osmolytes, antioxidants, maintaining polyamines, hormonal homeostasis, manipulating transcription factors, transporters, or modification enzymes. In some cases, genetic manipulation of regulatory proteins or phytohormone levels can improve salt tolerance; however, these alterations

may also undesirably reduce plant growth or yield.

In the past decade, numerous studies have clearly demonstrated that microRNAs (miRNAs) play crucial roles as post-transcriptional regulators in controlling all of the above biological processes [3,4]. While the physiological, molecular, and transcriptional responses to salt stress are well characterized in many plant species [5,6], the mechanisms by which miRNAs mediate salt stress tolerance in plants remain insufficiently understood [7]. A single miRNA can interact with gene products, mRNAs, transcription factors, and their derivatives during the plant stress response. In addition to their involvement in plant development, miRNAs regulate the expression of genes responsible for plant responses to both abiotic and biotic stress factors [8]. Understanding the identification of the miRNA target genes and mechanisms through which they function can aid in the development of plants with improved resistance to abiotic and biotic stresses [9, 10].

To date, the genomes of 71 plant species have been studied, leading to the identification of 872 miRNAs belonging to 42 families [11]. A significant proportion of these miRNAs have been found in the model plant *Arabidopsis thaliana* (A. thaliana). In recent years, with the advancement of next-generation sequencing (NGS) technologies, miRNAs responsive to salt stress have been identified not only in model plants but also in various agricultural crops such as alfalfa [12], cotton [13], apple [14], switchgrass [15], artichoke [16], tamarisk trees [17], cereal crops [18], and others.

То identify salt-responsive miRNAs in these plants, experiments have been conducted by exposing plants different concentrations to NaCl (ranging from 80 to 600 mM) for varying durations (from 3 hours to 15 days) [19]. Based on these studies, miRNAs have been identified in various plant organs, including leaves, roots, stems, flowers, and fruits, as well as across different developmental stages. The number of miRNAs detected under salt stress conditions varies widely-from severalfold to over a hundredfold-depending

**Bioinformatics and validation** of miRNA functions. Such studies not only deepen understanding of miRNAs but also provide foundational data for constructing miRNA regulatory networks [21]. Using bioinformatics tools, miRNAs and their target genes can be identified, and their expression profiles validated by qRT-PCR to provide primary data for practical applications (Table 1). For instance, miR169 in A. thaliana showed strong under expression 300 mΜ NaCl treatment [22, 23]. Conversely, in the leaves of G. hirsutum, exposure to 50-300 NaCl resulted in mΜ decreased expression of miR169 [24, 25, 26].

By analyzing miRNA expression under various environmental on the plant species, tissue specificity, and developmental stage [20].

However, large-scale identification of miRNAs under salt stress is essential and critical, as miRNA expression differs significantly depending on plant species and organ. example, For under salt stress conditions, the expression level of miR393 is upregulated in A. thaliana, Triticum aestivum, Agrostis stolonifera, and Hordeum spontaneum, but downregulated in Oryza sativa, Gossypium hirsutum, Gossypium raimondii, and Spartina alterniflora. A similar trend is observed for miR396, which is downregulated in A. thaliana, O. sativa, G. raimondii, and S. alterniflora, but upregulated in Solanum lycopersicum, Nicotiana tabacum, G. hirsutum, Α. stolonifera, Raphanus sativus, and Panicum virgatum. These changes in miRNA expression patterns vary not only across species but also between different plant organs.

conditions, it is possible to identify genotypes resistant to specific stresses. miR156, classified as a salt stressresponsive miRNA, showed similar expression levels in both monocot and dicot plants. In Α. thaliana, S. lycopersicum, G. hirsutum, and Τ. aestivum seedlings treated with 100-300 mM NaCl, miR156 expression increased while its target gene, Squamosapromoter binding protein-like (SPL), was downregulated [22, 23, 24, 27]. Enhancing expression of miR156 confers salt stress tolerance in plants [28].

These studies highlight that miRNA expression varies not only among species but also depending on the duration of stress exposure (Table 1).

	r	Identif	ied microRNA	s in Cotton (G. hir	sutum) Under Sa	linity Stress	1
Plant Type	Local Name	Identified microRNA	Applied Technology	Target Genes	NaCl Concentration (mM)	Response	Reference
		miR156	microRNA expression analysis	Squamosa promoter- binding-like protein (SPL2,3,4,9,10)	50–300	Increased expression observed	24
		miR157	microRNA expression analysis	Unknown	100	Decreased expression observed	25
		miR159	microRNA expression analysis	TCP transcription factor (TCP3)	200	Decreased expression observed	26
		miR160	microRNA expression analysis	ADP Ribosylation Factor 1 (ARF1)	100	Increased expression observed	25
		miR162	microRNA expression analysis	Dicer-like; Drought 1 (DCL1)	200	Decreased expression observed	26
G. hirsutum	Upland cotton	miR166	microRNA expression analysis	Unknown	100	Decreased expression observed	25
G. <i>h</i>	Uplar	miR167	microRNA expression analysis	Auxin response factor (ARF6)	100	Decreased expression observed	25
		miR168	microRNA expression analysis	Unknown	100	Decreased expression observed	25
		miR169	microRNA expression analysis	Heme activator protein complex (HAP2)	50–300	Decreased expression observed	24
		miR172	microRNA expression analysis	Apetala 2 (AP2)	100	Decreased expression observed	25
		miR393	microRNA expression analysis	F-box	100	Increased expression observed	25
		miR394	microRNA expression analysis	F-box	100	Decreased expression observed	25

Identified microRNAs in Cotton (G. hirsutum) Under Salinity Stress

**Functional characterization of miRNAs.** The functions of miRNAs identified through bioinformatics can be confirmed by overexpressing or knocking down/out these miRNAs or their target genes (Table 2). For example, overexpression of miR414c in *G. hirsutum* increased salt tolerance [13], while overexpression of miRNVL5 caused salt sensitivity in cotton [29].

These findings indicate that manipulating miRNA expression and

Table 1

their target genes can be a powerful strategy to develop salt-tolerant genotypes. Studying miRNA functions also helps elucidate the roles of the genes they regulate.

Table 2

		Overexp	ression of microR	NAs in Cotton (	G. hirsutum) Und	ler Salt Stres	S
Plant Type	Local Name	Identified microRNA	Applied Technology	Target Genes	NaCl Concentration (mM)	Tolerance	Reference
G. hirsutum	Cotton	miR414c	Overexpression	Fibronectin type III and SPRY domain- containing protein 1 (FSD1)	0–400	Sensitive	13
		miRNVL5	Overexpression	GhCHR	50-400	Highly sensitive	29

# Conclusion

Developing salt-tolerant plants to mitigate soil salinity effects is among the most cost-effective and efficient biological improvement strategies. Since the early 2000s, advances in understanding plant microRNAs-small regulatory molecules of 18 - 24nucleotides-have demonstrated their crucial roles in plant growth, development, and environmental stress responses.

Unraveling the molecular mechanisms of miRNAs, especially under abiotic stresses like salinity, offers valuable tools for enhancing crop resilience amid climate change and environmental challenges. Highthroughput sequencing technologies

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have enabled identification of numerous stress-responsive miRNAs in major crops and model plants, while bioinformatics allows prediction of their targets. Nonetheless, experimental validation overexpression via or knockdown remains essential to understand their precise regulatory roles.

miRNAs not only enhance stress tolerance but also coordinate plant growth by modulating multiple target genes simultaneously. Investigating miRNA activity under stress and harnessing this knowledge for breeding offers great potential for sustainable agriculture.

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# ASSESSMENT OF SALT STRESS TOLERANCE IN LOCAL COTTON VARIETIES UNDER LABORATORY CONDITIONS

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*Abstract.* Cotton (*Gossypium hirsutum*) is a globally cultivated crop of economic importance, serving as a primary source of natural fiber and oil. However, various stress factors (both abiotic and biotic) negatively affect its normal growth, development, yield, and quality. One major stress factor is soil salinity. In this study, we evaluated the salt stress tolerance of 24 local cotton varieties from Uzbekistan currently cultivated in field conditions. The varieties were tested under phytotron conditions with four different concentrations of NaCl (50 mM, 100 mM, 150 mM, 200 mM). As a result, the varieties were classified into four groups. Four representative varieties from each group were selected for further experiments.

Keywords: cotton, NaCl, Relative Stress Index, Friedman test, two-way ANOVA

# ИЗУЧЕНИЕ УСТОЙЧИВОСТИ МЕСТНЫХ СОРТОВ ХЛОПЧАТНИКА К СОЛЕВОМУ СТРЕССУ В ЛАБОРАТОРНЫХ УСЛОВИЯХ

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Аннотация. Хлопчатник (Gossypium hirsutum) — это важная сельскохозяйственная культура, широко выращиваемая во всем мире, основной источник натурального волокна и масла. Однако различные стрессовые факторы (абиотические и биотические) оказывают негативное влияние на его рост, развитие, урожайность и качество. Одним из таких факторов является засоление почвы. В данной работе исследована устойчивость 24 местных сортов хлопчатника, выращиваемых на полях Узбекистана, к солевому стрессу в условиях фитотрона при воздействии NaCl в четырёх концентрациях (50 мМ, 100 мМ, 150 мМ, 200 мМ). В результате сорта были классифицированы на 4 группы. Из каждой группы были отобраны по 4 сорта для дальнейших экспериментов.

*Ключевые слова:* хлопчатник, NaCl, относительный индекс стресса, критерий Фридмана, двухфакторный дисперсионный анализ.

# Introduction

Cotton is one of the most producing high-quality natural fiber. Its important industrial crops, known for products serve not only the textile

industry but also provide raw materials for various other sectors. The Gossypium genus includes 54 wild, semi-wild, and cultivated species [1]. Among them, four species have been domesticated, with *G*. hirsutum (upland cotton) accounting for about 90% of the world's cotton cultivation area. In Uzbekistan, this species is also the main cultivated type, occupying approximately 1 million hectares of irrigated land and yielding a total production of 3.5 million tons. However, the increasing level of soil salinization across Uzbekistan is leading to a decline in yield and deterioration in fiber quality. This downward trend in productivity is linked to multiple factors, with one of the most significant being climate change, which contributes to rising soil salinity [2].

Soil salinization is considered one of the most widespread abiotic factors affecting plant stress life. According to some estimates, 53% of agricultural land in Uzbekistan (around 2.28 million hectares) is saline, with 47% of this land suffering from medium to high levels of salinity. The issue of soil salinization in Uzbekistan is further exacerbated by irrigation: under high temperatures, excess water evaporates, leaving behind concentrated salts. The primary effects of salt stress include osmotic stress and secondary ionic imbalance, both of which can ultimately lead to plant death. Therefore, there is an urgent need to develop salt-tolerant cotton varieties [3].

Conventional breeding methods

aimed at developing salt-tolerant, highyielding, and improved fiber quality varieties are time-consuming. Moreover, the multigenic nature of salt tolerance and the low genetic variability in cotton reduce the efficiency of traditional breeding. Hence, innovative approaches involving biotechnology and molecular genetics have become increasingly important in recent years for developing salt-tolerant plant varieties.

Plants employ various mechanisms to cope with salt stress, including salt exclusion, ion compartmentalization in tissues. vacuoles, or older leaves, and the production of osmolytes. Stress tolerance in plants is regulated by specific genes present in the genome. Numerous genes responsible for salt tolerance have been identified and studied in various plants by researchers worldwide [13–18], but in cotton, these thoroughly genes have not been explored.

Based the above on considerations, we decided to assess the salt tolerance of agricultural cropsspecifically G. hirsutum varieties-using modern biotechnological approaches alongside traditional breeding methods. Salt-tolerant varieties will be selected based on experimental results and subsequently future used in biotechnological applications. This study aims to statistically evaluate the salt tolerance levels of cotton varieties currently cultivated in Uzbekistan's cotton fields.

# **Materials and Methods**

The experiment was conducted during 2024–2025 at the Center for Genomics and Bioinformatics of the Academy of Sciences of the Republic of Uzbekistan under controlled greenhouse (phytotron) conditions. Cotton (*Gossypium hirsutum*) varieties were selected based on their diversity, genetic origin, adaptability to local conditions, productivity, and widespread cultivation across the country (Table 1).

As control samples for the experiment, the *G. hirsutum* line TM-1 and the *G. barbadense* line Pima 3-79 were used.

Table 1

N⁰	Variety Name	Species	Origin
1	Buxoro-10	G. hirsutum	Uzbekistan
2	Buxoro-102	G. hirsutum	Uzbekistan
3	Buxoro-6	G. hirsutum	Uzbekistan
4	Buxoro-8	G. hirsutum	Uzbekistan
5	C-4727	G. hirsutum	Uzbekistan
6	C-6524	G. hirsutum	Uzbekistan
7	C-6550	G. hirsutum	Uzbekistan
8	Doʻstlik-2	G. hirsutum	Uzbekistan
9	Gulbaxor-2	G. hirsutum	Uzbekistan
10	Ishonch	G. hirsutum	Uzbekistan
11	Kelajak	G. hirsutum	Uzbekistan
12	Koʻpaysin	G. hirsutum	Uzbekistan
13	Namangan-102	G. hirsutum	Uzbekistan
14	Namangan-34	G. hirsutum	Uzbekistan
15	Namangan-77	G. hirsutum	Uzbekistan
16	Nasaf	G. hirsutum	Uzbekistan
17	Navbaxor-2	G. hirsutum	Uzbekistan
18	Omad	G. hirsutum	Uzbekistan
19	Porloq-1	G. hirsutum	Uzbekistan
20	Ravnaq-1	G. hirsutum	Uzbekistan
21	Sulton	G. hirsutum	Uzbekistan
22	Toshkent-6	G. hirsutum	Uzbekistan
23	Surxon-103	G. hirsutum	Uzbekistan
24	Baraka	G. hirsutum	Uzbekistan
25	TM-1 (control)	G. hirsutum	USA
26	3-79 (control)	G. barbadense	USA

#### Selected Cotton Varieties for Evaluating Tolerance/Sensitivity to Salt Stress

**Evaluation of plant response to salt stress.** The salt tolerance of cotton plants was evaluated using a method originally developed by Rodriguez-Uribe and modified for this study [4]. To determine the natural salt stress 102 tolerance of Gossypium hirsutum, four different concentrations of sodium chloride (NaCl) aqueous solutions—50 mM, 100 mM, 150 mM, and 200 mM were used.

Plant samples were sown in polyethylene pots  $(30 \times 14 \text{ cm})$  in three technical replicates following а randomized design. Before sowing, cotton seeds were soaked in sterile distilled water for 24 hours. In each ten seeds replicate, were sown approximately 2 cm deep into soil-filled polyethylene pots.

After sowing, plants were irrigated with one of the four NaCl solutions (50–200 mM), while control plants were irrigated with distilled neutral water. The experiment was carried out under controlled phytotron

Root and hypocotyl length measurements. After 21 days of treatment, the seedlings were carefully removed from the polyethylene pots and washed under running water to eliminate particles soil without damaging the main root or its lateral branches. The length of the hypocotyl, epicotyl, and the total seedling length were recorded. Measurements were carried out using ImageJ software version 1.53e, by analyzing photographs

where:

• LsL\_sLs = average length of plants in the control group,

• LtL\_tLt = average length of plants in the salt-stress group.

where:

• SItSI\_tSIt = stress index of the NaCl-treated group,

conditions: daytime temperature was maintained at approximately 35 °C, nighttime at around 27 °C. The photoperiod consisted of 16 hours of light and 8 hours of darkness, with a light intensity of 2400 lux. The experimental duration was 21 days.

To avoid seedling rot while maintaining adequate soil moisture, irrigation with 100 ml of either NaCl solution or distilled water (for control) was carried out every other day.

of seedlings placed on a millimeter-scale background [5].

The fresh weights of roots, hypocotyls, and green shoots were measured using a laboratory balance.

The salt tolerance of each cotton variety was further assessed by calculating the Stress Index (SI), which represents the proportional decrease in seedling length due to NaCl treatment. The SI was calculated using the following formula:

# SI = ((Ls - Lt) / Ls)

After calculating the Stress Index, a Relative Stress Index (RSI) was derived to compare the tolerance of plants subjected to different NaCl concentrations. The RSI was calculated using the following equation:

# $RSI = (SIt / SIn) \times 100$

• SInSI\_nSIn = stress index of the untreated control group.

The RSI is expressed as a percentage and indicates the relative

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level of stress experienced by treated plants compared to the control group.

Based on RSI values, the plants were classified into four tolerance categories:

- Highly tolerant (0–20%),
- Tolerant (20–40%),
- Moderately tolerant (40–
- Susceptible (60–100%) [6].

# **Results and discussion**

60%),

Statistical Analysis of Varietal Tolerance to Salt Stress

The statistical analysis of the collected data was conducted using the R Core software environment. In this

Data Processing and Statistical Analysis

То evaluate the tolerance of abiotic stress cotton genotypes to conditions, data obtained from the experiments were processed using a range of statistical methods. Microsoft Core Excel and the R software environment were employed for data analysis and visualization [7].

study, the significance level was set at  $\alpha$  = 0.05. The average values for seedling height and green biomass weight were calculated across all tested varieties (Table 2).

Table 2.

NaCl concentration	Length, cm	Weight, g
Control	22.6±0.397	1.72±0.040
50	22.2±0.252	1.38±0.018
100	19.8±0.254	1.14±0.016
150	17.6±0.234	0.94±0.015
200	13.6±0.306	0.68±0.018

Average height and weight values of the cotton varieties under study

Based on the obtained data, a two-way analysis of variance (ANOVA) was performed, which demonstrated the significance of differences in seedling height and biomass weight between the varieties and treatment groups. Significant differences in seedling length were observed only between groups grown under high salt concentrations (150 and 200 mM) and those under low concentrations (control and 50 mM). Variations in green biomass values were also evident in treatments with 100–200 mM salt concentrations.

In this study, we selected hypocotyl length as a criterion for salt tolerance or sensitivity. According to the results of the two-way ANOVA, a 150 mM concentration of NaCl solution was chosen as a critical threshold for evaluating salt stress. Subsequently, data were normalized by calculating changes in seedling length as a percentage relative to the control. The selected threshold value corresponded to a 25% reduction compared to the control. If seedling length under 150 mM salt treatment did not decrease by more than 25%, the plant was classified as salt-tolerant.

A posteriori analysis using the Friedman test showed statistically significant differences only between groups subjected to high salt concentrations (150 and 200 mM) and those grown under lower concentrations (control and 50 mM). This led to the conclusion that varieties with а reduction in seedling length up to 150

mM NaCl were considered weakly tolerant, and if seedling length significantly decreased even at low salt concentrations, the variety was considered sensitive.

Thus, the degree of salt tolerance (or sensitivity) of 24 cotton varieties studied during the experiment was determined. Based on the results, the varieties were classified into the categories of tolerant, moderately tolerant, weakly tolerant, and sensitive (Table 3).

Table 3.

		e/sensitivity of studied		5
Variety Name	Tolerant	Moderate	Weak	Sensitive
Buxoro-10				+
Buxoro -102				+
Buxoro -6			+	
Buxoro -8		+		
C-4727	+			
C-6524		+		
C-6550		+		
Dustlik-2		+		
Gulbaxor-2			+	
Ishonch				+
Kelajak	+			
Kupaysin		+		
Namangan-102				+
Namangan -34		+		
Namangan -77			+	
Nasaf		+		
Navbaxor-2			+	
Omad			+	
Porloq-1		+		
Ravnaq-1		+		
Sulton		+		
Toshkent-6		+		
Surxon-103			+	
Baraka	+			
TM-1	+			
3-79		+		

Assessment of the tolerance/sensitivity of studied varieties to soil salinity

For subsequent experiments, we selected four contrasting varieties from each of the salt-tolerance categories: tolerant, moderately tolerant, and sensitive.

# Conclusions

Cotton is a major agricultural crop widely cultivated for its natural textile fiber and cottonseed oil. Commercial cotton varieties are grown 80 in over countries, occupying approximately 32-34 million hectares, with a total annual production of about 25.65 million metric tons [8]. However, one of the main challenges for cotton production in arid regions is soil salinization, which significantly reduces crop yields. The increase in the proportion of salinized agricultural land is associated with climate change and the application of modern irrigation which salts techniques, cause to accumulate due to leaching from the deeper soil layers [9].

Salt stress adversely affects physiological processes in plants, ultimately reducing their productivity. High salt concentrations in the soil delay plant growth and development, inhibit enzymatic activity, and reduce the intensity of photosynthesis [10, 11, 12]. There is genetic variability in salt sensitivity both within and between plant species. Plants employ various mechanisms to tolerate salt stress. In response to osmotic stress, plants produce compatible solutes-such as glycine betaine, trehalose, and proline – that protect them from dehydration or protein denaturation.

In light of these facts, one of the most pressing objectives of modern biotechnology is to study the molecular and genetic basis of salt tolerance in agricultural crops under extreme environmental conditions.

In the first phase of this effort, we evaluated 24 local cotton varieties under model (phytotron) conditions for salt stress tolerance at different NaCl concentrations (50, 100, 150, and 200 mM). Based on the results, the varieties were classified into four categories according to their level of salt tolerance: tolerant, moderately tolerant, weakly tolerant, and sensitive.

Three varieties (Baraka, Surxon-103, and Ishonch) were identified as having contrasting salt tolerance traits. Among them, the variety Baraka demonstrated the highest level of tolerance and was selected as а promising donor of salt-tolerance genes. The other two varieties, classified as sensitive or weakly tolerant, will be used as contrasting genotypes in further studies on the genetic basis of salt stress resistance.

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