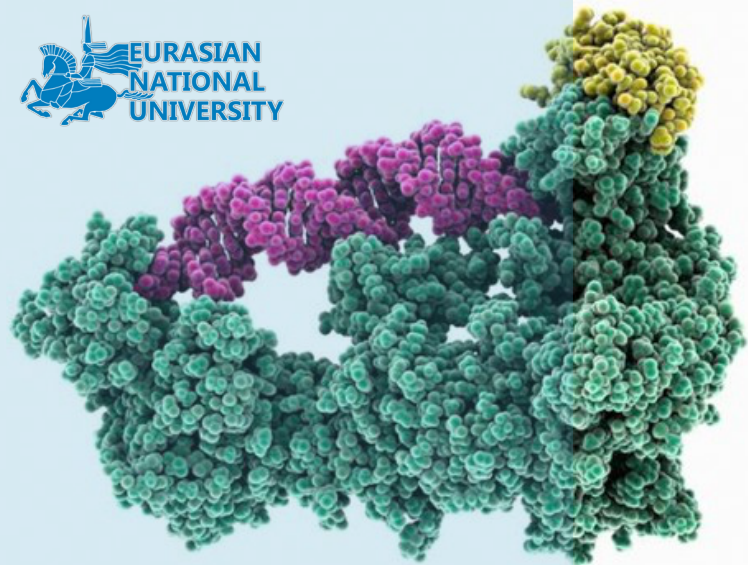


ҒЫЛЫМ ЖӘНЕ ЖОҒАРЫ БІЛІМ МИНИСТРЛІГІ
МИНИСТЕРСТВО НАУКИ И ВЫСШЕГО ОБРАЗОВАНИЯ



Л. Н. ГУМИЛЕВ АТЫНДАҒЫ
ЕУРАЗИЯ ҰЛТТЫҚ УНИВЕРСИТЕТІ

ЕВРАЗИЙСКИЙ НАЦИОНАЛЬНЫЙ
УНИВЕРСИТЕТ ИМЕНИ
Л. Н. ГУМИЛЕВА

АСТАНА, ҚАЗАҚСТАН
11 СӘУІР 2024 ЖЫЛ

АСТАНА, КАЗАХСТАН
11 АПРЕЛЯ 2024 ГОД

"ОМАРОВ ОҚУЛАРЫ: ХХІ
ҒАСЫРДЫҢ БИОЛОГИЯ ЖӘНЕ
БИОТЕХНОЛОГИЯСЫ" АТТЫ
ХАЛЫҚАРАЛЫҚ ҒЫЛЫМИ
ФОРУМНЫҢ БАЯНДАМАЛАР
ЖИНАҒЫ

СБОРНИК МАТЕРИАЛОВ
МЕЖДУНАРОДНОГО НАУЧНОГО
ФОРУМА "ОМАРОВСКИЕ ЧТЕНИЯ:
БИОЛОГИЯ И БИОТЕХНОЛОГИЯ
ХХІ ВЕКА"

УДК 57 (063)
ББК 28.0
Ж 66

Жалпы редакцияны басқарған т.ғ.д., профессор Е.Б. Сыдықов
Под редакцией д.и.н., профессора Е.Б. Сыдыкова

Редакция алқасы:
Редакционная коллегия:

Ж.К. Масалимов, А.Б. Курманбаева, Ж.А.Нурбекова, Н.Н. Иқсат.

«Омаров оқулары: ХХІ ғасыр биология және биотехнологиясы» халықаралық ғылыми форумының баяндамалар жинағы. – Астана: Л.Н. Гумилев атындағы Еуразия ұлттық университеті, 2024. – 284 б., қазақша, орысша, ағылшынша.

Сборник материалов международного научного форума «Омаровские чтения: Биология и биотехнология ХХІ века». – Астана. Евразийский национальный университет имени Л.Н. Гумилева, 2024. – 284 с., казахский, русский, английский.

ISBN 978-601-337-977-7

Жинақ «Омаров оқулары: ХХІ ғасыр биология және биотехнологиясы» атты халықаралық ғылыми форумна қатысушылардың баяндамаларымен құрастырылған. Бұл басылымда биология, биотехнология, молекулалық биология және генетиканың маңызды мәселелері қарастырылған. Жинақ ғылыми қызметкерлерге, PhD докторанттарға, магистранттарға, сәйкес мамандықтағы студенттерге арналған.

Сборник составлен по материалам, представленным участниками международного научного форума «Омаровские чтения: Биология и биотехнология ХХІ века». Издание освещает актуальные вопросы биологии, биотехнологии, молекулярной биологии и генетики. Сборник рассчитан на научных работников, PhD докторантов, магистрантов, студентов соответствующих специальностей.

ISBN 978-601-337-977-7



УДК 57
ББК 28
О-58

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UDC 581.1.03

Influence of combined drought and temperature stresses on the levels of water- and fat-soluble vitamins in plants

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The antioxidant and cellular reductant properties of vitamin C (ascorbic acid) and vitamin E (tocopherols) play essential roles in plant physiology. They also have a range of additional functions in the growth and development of plants, as well as their ability to regulate multiple cellular systems in reaction to environmental challenges. The influence of combined temperature and drought-induced stress on the levels of water- and fat-soluble vitamins in barley was investigated in this study, which used barley as a model. To achieve this, plants were grown in the growth chambers with high (+40°C) and low (+10°C) temperatures with and without water supply for 5 days. The amounts of antioxidants other than enzymes were measured, as well as the levels of vitamin E and vitamin C. A significant increase in the concentration of vitamin E was seen at high temperatures, but an increase in the amount of vitamin C was observed at low temperatures, according to the findings. Using the information gathered, researchers may be able to develop new transgenic crops that are more resistant to a broader variety of environmental challenges.

Keywords: abiotic stress, reactive oxygen species (ROS), drought, temperature, biomolecule, vitamin E, vitamin C, antioxidative defense

Introduction

Thermal stress (both heat and cold) has a detrimental impact on plant development, and metabolism since these processes have optimum temperature limitations for every plant species. Heat stress is a significant constraint on plant development because it impairs normal physiological activities such as photosynthesis, respiration, membrane integrity, and protein metabolism. Low temperatures may have a detrimental effect on many elements of crop growth, including survival, cell division, photosynthesis, water transport, development, and yield. Drought (abnormally low water levels) is also a critical abiotic element in the ecosystem. Drought may deplete the leaf's water potential. This results in a decrease in the turgor of plant cells and, therefore, a drop in agricultural yields.

Abiotic stressors primarily induce damage via reactive oxygen species (ROS) production, including superoxide radicals, hydrogen peroxide, and hydroxyl radicals. Increased ROS production results in oxidative damage to biological macromolecules. Abiotic stressors seldom occur independently, and plants are often subjected to several kinds of stress concurrently. Despite the abundance of accessible data, research on the development of oxidative stress because of temperature and water shortage is mostly incomplete. Furthermore, the cross-effects of these variables on the impact of oxidative stress in plants have received little attention.

Plants have developed many defensive mechanisms to combat oxidative stress. Antioxidative defense systems detoxify or remove free radicals from the plants [1]. Antioxidants are classified into two types: enzymatic and non-enzymatic antioxidants, of which the latter involves vitamin C, vitamin E [2].

Fat-soluble vitamin E acts as an essential redox buffer system. Vitamin E is usually synthesized in chloroplasts and protoplasts and is localized in cell membranes. The antioxidant activity of tocopherols has been shown to have two leading roles. This compound regulates the level of singlet oxygen, removing it, and provides protection against lipid peroxidation by removing the peroxide radical [3, 4-6].

Another non-enzymatic antioxidant is vitamin C, which is an important compound of the plant defense system. The amount of vitamin C (ascorbic acid) is high in fruits, leaves, and flowers, while the concentration in roots and stems is low [7].

It is now recognized that abiotic stressors may induce cell dehydration and the generation of reactive oxygen species, resulting in cellular damage to membranes and the photosynthetic system. Most of the molecular processes involved with plant responses to a combination of heat stress and water shortage, such as alterations in gene expression, signal transduction, and regulatory networks, are, nevertheless, mostly understood. As a result, this may be the first effort to study the combined impact of several abiotic variables, such as temperature and water shortage (drought), on plant oxidative burst.

Materials and methods

Growth conditions

To pre-sterilize the seeds, they were treated for 10 minutes with a 50 percent aqueous solution of sodium hypochlorite (NaClO) (50 ml distilled water, 50 ml NaClO), then incubated for 60 seconds in 70% ethanol and rinsed three times with distilled water. Following drying, each pot was planted with 30 seeds in 150 g of soil wet with 40 ml of water. In the greenhouse, we utilized soil that had been sterilized in an autoclave (TerraVita, Russia) and included vermicompost, as well as basic nutrients such as nitrogen (NH₄ + NO₃) - 150 mg / l, phosphorus (P₂O₅) - 270 mg / l, and potassium (K₂O) - 300 mg / l. Additionally, vermiculite was applied to the soil at a ratio of 10 g to 150 g soil during planting. The plants were then cultivated for two days in a greenhouse equipped with white, fluorescent lights Econ 4200K, 230V, a timer set to a 16-hour photoperiod, and a temperature regime of 25/20°C (day and night) with an 80 percent relative humidity.

Test system for exposure to stress factors

To simulate drought, the test plants were not watered after their emergence. After two days, and immediately upon the appearance of the first seedlings, the plants were moved to temperature chambers set to a temperature regime of 10°C and 40°C. Control samples were kept at 25°C in a greenhouse. Every day, at a particular time of day, all plants with a scheduled watering routine were watered. Five days of appropriate circumstances provided long-term exposure to stress stimuli.

Determination of water-soluble (vitamin C) and fat-soluble (vitamin E) antioxidants

The spectrophotometric technique developed by Hewitt et al. [8] was used to determine water-soluble antioxidants of vitamin C. 3 ml of a 2 percent metaphosphoric

acid solution was used to homogenize plant material (0.3-0.5 g of raw leaves). The homogenate was transferred to a measuring tube and adjusted to 10 ml with a combination of HPO₃ and Na₃PO₄ in a 3: 2 (V / V, pH 7.3-7.4) ratio. The extract was centrifuged at 3000 g for 15 minutes. On a spectrophotometer at 265 nm, the optical density of the solution was measured against a standard - the above solutions of HPO₃ and Na₃PO₄ were taken in the same ratio (3: 2).

The spectrophotometric technique was used to determine the quantity of tocopherols (vitamin E) in plants. Total extracts were produced to determine the quantitative number of tocopherols (vitamin E) in the investigated samples. The extractant was selected to be Vaseline oil (the use of vegetable oils was unacceptable since they "a priori" contain various forms of tocopherols). 1 g of plant leaves were put into extractants in a 1:5 raw / extractant ratio and maintained in flasks with ground stoppers for 120 hours. After the infusion time was up, the liquid was decanted, and the resultant extract was centrifuged at 8000 rpm for 15 minutes to remove mechanical impurities. UV spectra were acquired on a spectrophotometer for the pure extracts using the matching extractants as reference solutions at 292 nm against a standard.

Results and discussion

Many vitamins have been shown to be powerful antioxidants, capable of neutralizing the superoxide radical and converting it into hydrogen peroxide [8]. It has been shown in a few studies [9, 10, 11] found that several vitamins are anti-mutagenic. The presence of non-enzymatic antioxidants under the impact of combined drought and temperature stressors was discovered as a result of this research.

Ascorbic acid (also known as ascorbate or vitamin C) is an important chemical found in substantial concentrations in plant tissues, where it plays a vital role in plant growth and development. Numerous biochemical parameters in crops, including growth, tissue and organ formation, and metabolism, are significantly influenced by it. One of the functions of vitamin C is to restore numerous free radicals while simultaneously reducing the damage caused by oxidative stress during aerobic metabolism, vitamin C is generated, and it subsequently interacts with O₂, singlet oxygen, and ozone (chemically), as well as with H₂O₂ (enzymatically) to counteract the toxicity of these toxins. Vitamin C is also important in the regeneration of antioxidant pigments, such as carotenoids and vitamin E [11].

We discovered that higher temperatures resulted in a substantial rise in vitamin C content. No substantial variations in vitamin C concentrations were seen under drought circumstances (Figure 1A). Ascorbic acid has been demonstrated in many studies to have a critical function in increasing plant resilience to environmental stresses. It is believed that exogenous ascorbate plays the most crucial function in protecting lipids and proteins from oxidation when exposed to abiotic stressors [12,13, 14]. It has been demonstrated that ascorbate is effective in protecting plants from a variety of environmental stresses such as drought, salinity, ozone, low/high temperatures, and high light intensity [15-20]. However, several recent studies have demonstrated that drought stress leads to the downregulation of vitamin C content in Labiatae species [21] and soybean (*Glycine max*) [22].

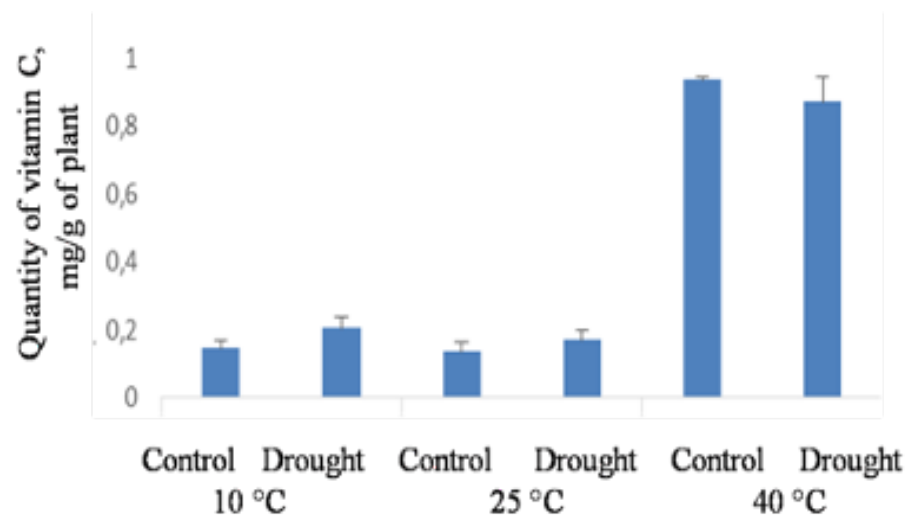


Figure 1A) Changes in vitamin C concentrations under the effect of temperature-induced oxidative stress,

As regards vitamin E concentrations, it rises significantly at both low and high temperatures. At a temperature of 25°C, however, this tendency was not observed. Most likely, changes in vitamin concentration are directly related to stressful conditions (Figure 1B). It has been shown that plants resistant to abiotic stress show an increased level of tocopherol, while sensitive plants show a reduced level under stress conditions, which leads to oxidative damage [21]. Also, some studies have shown that stress factors such as drought, salinity, and temperature lead to increased tocopherol levels as a response to the antioxidant system [23-25].

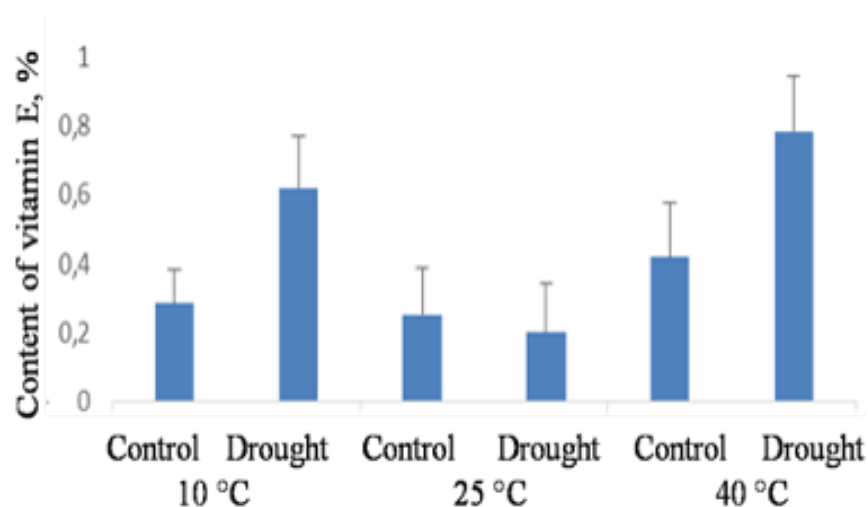


Figure 1B) Changes in vitamin E concentrations under the effect of temperature-induced oxidative stress

Low molecular weight antioxidants such as ascorbate (vitamin C), glutathione, and tocopherols (vitamin E) can mitigate the adverse effects of increased ROS accumulation [26]. On the one hand, they can influence the expression of genes associated with abiotic stresses. On the other hand, these antioxidants function as redox buffers that interact with ROS and act as a metabolic intermediate that modulates the corresponding induction of climatic reactions or programmed cell death [27,28]. These investigations showed that ascorbate is essential for plant growth and development, as well as for plant tolerance to abiotic stress.

Briefly, antioxidant enzymes are the main chemical molecules that protect plants from various stresses and keep ROS homeostasis.

Conclusion

In conclusion, we found out that the composition of biological macromolecules, such as water-soluble vitamins and fat-soluble vitamins, had changed as non-enzymatic antioxidants. A considerable rise in the content of vitamin C was detected when the temperature was raised to high levels. The content of vitamin C did not vary much under dry circumstances, according to the research. A significant rise in the content of vitamin E is found at both lower and higher temperatures. At a temperature of 25 degrees Celsius, this tendency, on the other hand, was not seen.

Funding

This work was funded by the Kazakhstan National Grant Program “Zhas galym” 2023-2025 of the Ministry of Science and Higher Education of the Republic of Kazakhstan (grant no. AP19175117)

Conflict of interests

The authors declare no conflict of interests regarding the publication of this article.

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УДК 578.1

TBSV P19 мутанттары мен ауыр металдардың өсімдіктерде ОБТ жинақталу деңгейіне әсері

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