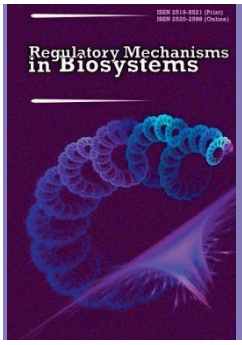

Regulatory Mechanisms in Biosystems

ISSN 2519-8521 (Print)
ISSN 2520-2588 (Online)
Regul. Mech. Biosyst.,
2024, 15(2), 189–197
doi: 10.15421/022428

Effect of growth promotors on morphogenesis, photosynthetic apparatus, productivity and residual substances content in sweet pepper (*Capsicum annuum*) fruits

V. V. Rohach*, V. G. Kuryata*, O. O. Stasik*, D. A. Kiriziy**,
I. H. Grabyk***, O. V. Kaitanyuk***, T. I. Rohach*, M. V. Tarasyuk**

**Mykhailo Kotsyubynskyi Vinnytsia State Pedagogical University, Vinnytsia, Ukraine*
***Institute of Plant Physiology and Genetics, National Academy of Sciences of Ukraine, Kyiv, Ukraine*
****Vinnytsia Research Expert Forensic Center of the Ministry of Internal Affairs of Ukraine, Vinnytsia, Ukraine*

Article info

Received 02.02.2024
Received in revised form 15.03.2024
Accepted 03.04.2024

Mykhailo Kotsyubynskyi Vinnytsia State Pedagogical University, Ostrozhskogo st., 32, Vinnytsia, 21000, Ukraine. Tel.: +38-098-59-44-832. E-mail: rogachv@ukr.net

Institute of Plant Physiology and Genetics, National Academy of Sciences of Ukraine, Vasylkivska st., 31/17, Kyiv, 03022, Ukraine. Tel.: +38-044-257-90-18. E-mail: phot-ecol@jfrg.kiev.ua

Vinnytsia Research Expert Forensic Center of the Ministry of Internal Affairs of Ukraine, V. Poryka st., 8, Vinnytsia, 21021, Ukraine. Tel.: +38-096-12-61-235. E-mail: kaytanyk1985@gmail.com

Rohach, V. V., Kuryata, V. G., Stasik, O. O., Kiriziy, D. A., Grabyk, I. H., Kaitanyuk, O. V., Rohach, T. I., & Tarasyuk, M. V. (2024). Effect of growth promotors on morphogenesis, photosynthetic apparatus, productivity and residual substances content in sweet pepper (Capsicum annuum) fruits. Regulatory Mechanisms in Biosystems, 15(2), 189–197. doi:10.15421/022428

The use of plant growth promoting substances in order to optimize crop performance is one of the leading tasks of modern plant physiology. The effect of 0.005% 1-naphthaleneacetic acid (1-NAA), 0.005% gibberellic acid (GA3), 0.005% 6-benzylaminopurine (6-BAP) foliar treatment on morphogenesis, leaf parameters, photosynthetic pigments content, photosynthetic apparatus activity indices, and residual amounts of the used substances in the sweet pepper *Capsicum annuum* L. variety Antey fruits was investigated. The field experiment was laid on plots of 32 m^2. The treatment of plants was carried out at the budding stage. Morphometric indices were determined at the flowering and fruit formation stages. The mesostructure of the middle tier leaves was studied at the fruit formation stage. The content of the chlorophylls was determined in the fresh leaf tissue by the spectrophotometric method. Indices of photosystem II (PSII) activity were determined by registration of chlorophyll fluorescence induction, using a portable single-beam fluorimeter. The assay of the residual content growth of promotors in the fruits was carried out on a Shimadzu GC gas chromatograph with a mass spectrometric detector – GCMS-QP2020 EI. It was revealed that treatment with GA3 increased plant height. All substances increased the number of leaves on the plant, their fresh and dry weight, the leaf area, and the canopy leaf area index. All growth promoting substances thickened leaf laminas due to the growth of chlorenchyma. GA3 and 6-BAP increased the volume of columnar parenchyma cells, and 1-NAA and GA3 increased the size of leaf spongy parenchyma cells. 6-BAP increased the total chlorophyll ($a + b$) content in the leaves, and under the GA3 treatment this index tended to decrease. 6-BAP increased the chlorophyll index of plants at the fruit formation stage, and GA3 decreased it at the flowering stage. The net photosynthetic rate increased under the influence of 1-NAA and 6-BAP, and decreased under GA3 treatment. GA3 reduced photorespiration and transpiration in leaves, 6-BAP increased transpiration, and 1-NAA decreased it. All substances increased dark respiration. Treatment with GA3 and 6-BAP reliably increased the maximum quantum efficiency of PSII photochemical reactions. 1-NAA and 6-BAP increased the actual quantum efficiency of PSII photochemistry. The coefficient of chlorophyll fluorescence photochemical quenching significantly increased at flowering under the application of 1-NAA, and tended to increase at the stage of fruit formation under the treatment of 1-NAA and 6-BAP. When GA3 was applied, the rate of chlorophyll fluorescence photochemical quenching was significantly reduced. The index of PSII reaction centers fraction that do not reduce the QB acceptor did not change with the use of 1-NAA and GA3 and tended to decrease after the use of 6-BAP. All growth promotors increased the whole plant dry weight. They reduced the net photosynthetic efficiency at the flowering stage, and at the stage of fruit formation it significantly increased under the treatment with 6-BAP. The 6-BAP caused the redistribution of plastic substances into the fruits due to the decrease in the dry weight of vegetative organs. Treatment with 1-NAA increased the proportion of root dry weight. 1-NAA, GA3 and 6-BAP increased the yield of fruits from one plant by 17%, 22% and 20% respectively due to the increase in the number of fruits per plant. The residual amounts of 1-NAA and 6-BAP in ripe sweet pepper fruits did not exceed the maximum permissible sanitary standards.

Keywords: *Capsicum annuum* L.; growth promoting substances; morphometry; leaf parameters; leaf mesostructure; crop cenotic indices; photosynthesis; crop capacity; residual amounts of growth promoting substances.

Introduction

One of the important tasks of modern plant physiology is the search for new ways and means of increasing the productivity of cultivated plants, and improving the quality of their products (Singh et al., 2017). A condition for obtaining significant achievements in this direction is the optimization of plants' genetic potential realization level together with the simultaneous minimization of influence of negative environmental factors during ontogenesis (Rai et al., 2017).

More effective and purposeful management of plant productivity is made possible by growth and development regulators, among which one of the earliest and most widely used in agricultural production is a group of promotors. By their nature, these substances are native phytohormones or their synthetic analogues. They have a wide range of effects on plants, and their use allows purposeful regulation of individual stages of plant growth and development in order to mobilize the potential capabilities of the plant organism, and above all to more fully use light energy for enhanced synthesis of organic substances with their subsequent redistribution to economically valuable tissues and organs (Aremu et al., 2017; Madzikane-Mlungwana et al., 2017). It is known that action of physiologically active substances leads to the restructuring of the plant assimilation apparatus, to change in the habit, the weight ratio of its organs, the appearance of ad-

ditional attracting centers, and the strengthening or weakening of the functioning of already existing ones, which indicates changes in the pattern of source-sink relations in the plant (Gao et al., 2017; Singh et al., 2017; Mohorović et al., 2023).

The effect of growth promotors is associated with the acceleration of cell division processes, stretching and differentiation with a simultaneous increase in the linear dimensions of plants (Aremu et al., 2017; Madzikane-Mlungwana et al., 2017; Rai et al., 2017), the assimilation surface area (Ren et al., 2017), increasing of the chlorophyll content (Luo et al., 2016; Ren et al., 2017) and, as a result, activation of photosynthetic processes (Rai et al., 2017; Ren et al., 2017) and increasing plant productivity (Gonzatto et al., 2016; Khalid et al., 2016; Alexopoulos et al., 2017). In the scientific literature, there is enough information about the use of native and synthetic growth promotors in order to activate the production process through morphometric changes in cereal (Luo et al., 2016; Zhao et al., 2017), legume (Xing et al., 2016), oilseed (Froschle et al., 2017), vegetable (Tubis et al., 2016; Alexopoulos et al., 2017), technical (Rai et al., 2017), fruit (Li et al., 2016), medicinal and decorative (Aremu et al., 2017; Madzikane-Mlungwana et al., 2017) cultures. Growth promotors also increase the resistance of agricultural crops to adverse abiotic and biotic environmental factors due to changes in the hormonal status, and activation of antioxidant systems in the plant organism (Morgun et al., 2019; Tubis et al., 2016; Xing et al., 2016).

Literary sources contain information on the use of gibberellins to regulate the growth, development and productivity of a number of cultivated plants. The effect of gibberellins is primarily associated with the activation of meristem tissues and, as a result, the formation of larger plants with a powerful assimilation apparatus and better potential opportunities for the formation of biological productivity (Alexopoulos et al., 2017). In addition, gibberellins delay senescence in leaves, and induce laying of more flowers on the plant. In particular, gibberellic acid (GA3) accelerated growth and elongation of stems and leaves, and increased plant fresh and dry weight and leaf area in wheat (Zhao et al., 2017). An increase in the dry weight of fruits and a decrease in their fresh weight were noted after the use of the GA on tangerine (Khalid et al., 2016). The use of GA on sugarcane plantations led to an increase in the linear dimensions of plants, the stems and number of internodes, the stem fresh weight and diameter, leaf area, leaf area index, net photosynthetic efficiency, and the photosynthetic period duration compared to the control. Such morphometric changes in plants caused an increase in the biological productivity of the crop (Rai et al., 2017).

Auxin growth promotors are quite diverse in terms of chemical structure, but with similar anatomical-morphological and physiological-biochemical effects on plants. There is information that synthetic auxin from the indolin group, indole-3-butyric acid, enhanced the rooting of *Eriocephalus africanus* seedlings (Madzikane-Mlungwana et al., 2017). Literature sources contain information on the use of halogen-containing derivatives of phenoxyacetic acid. In particular, treatment of tangerine plants with 3,5,6-trichloro-2-pyridyloxyacetic acid increased yield by increasing the size and weight of fruits without increasing their number (Gonzatto et al., 2016). An extremely active group of auxins are derivatives of naphthylcarboxylic acids and their salts. A number of researchers note the positive effect of 1-NAA on the growth, development and productivity of cultivated plants. For example, 1-NAA increased plant size and the length of wheat leaves, and the fresh and dry weight of the whole plant (Zhao et al., 2017). However, according to other data, the substance practically did not affect the morphometric parameters of *Lachenalia montana* (Aremu et al., 2017).

Literary sources contain information on the use of synthetic regulators of cytokinin action in agricultural practice. Thus, phenylurea derivatives – thidiazuron and N-(2-chloro-4-pyridyl)-N-phenylurea increased the number of buds in onion (Tubis et al., 2016). The most common representative of another group of cytokinins, N-oxide substituted pyridine, is 2,6-dimethylpyridine-1-oxide. The substance is widely used as a growth promotor in the cultivation of vegetable and industrial crops. In particular, the treatment of sunflower seeds with ivin increased the linear dimensions of the shoot and root, fresh weight of the whole plant, the content of chlorophylls *a*, *b* and carotenoids in leaves, increased the basket diameter, which could be a prerequisite for increasing the seeds yield (Tsygankova et al., 2023).

The most widely used group of synthetic cytokinins are analogues of natural purine cytokinins. The literature contains information on the use of 6-BAP on many agricultural crops. The substance inhibited the growth of young shoots in an apple tree, but enhanced the flowering of the plant (Li et al., 2016). The use of 6-BAP on maize had a positive effect on the leaf mesostructure, increased the number of chloroplasts and their chlorophyll content, and also increased the leaf area index and net photosynthetic efficiency. Such changes in the leaf apparatus had a positive effect on grain yield (Ren et al., 2017). Under the influence of 6-BAP an increase in chlorophyll content in wheat leaves and grain yield was also noticed (Luo et al., 2016). The productivity of *Jatropha curcas* increased after the application of 6-BAP and forchlorfenuron (Froschle et al., 2017).

There is information about changes in the indices of photosynthetic apparatus activity under the influence of growth promotors. In particular, 6-BAP significantly increased stomatal conductance, CO_2 concentration in intercellular spaces, CO_2 assimilation rate, photochemical activity of PSII in wheat leaves (Yang et al., 2017). Treatment of cucumber seedlings with the same substance increased the chlorophyll content, CO_2 assimilation rate, stomatal conductance, and the maximum quantum efficiency of PSII photochemical reactions (Fv/Fm) under shade conditions (Xiao-Tao et al., 2013). Another synthetic cytokinin, kinetin, at a rate of 10 μM increased Fv/Fm, as well as the rate of electron transport, coefficient of chlorophyll fluorescence photochemical quenching, and reduced the non-photochemical quenching of chlorophyll fluorescence in tomato leaves (Ahanger et al., 2018). The same kinetin concentration in the treatment of eggplant seedlings increased the quantum efficiency of PSII and photochemical quenching (Singh & Prasad, 2014). Synthetic auxin (3,5,6-trichloro-2-pyridyloxyacetic acid) at the rate of 15 mg/L reduced the quantum efficiency of PSII, the rate of linear electron transport during noncyclic photophosphorylation, and the photosynthetic processes rate in tangerine leaves during the first twenty days after treatment. After that, the efficiency of the plants' assimilation system was restored to the previous level and even increased (Mesejo et al., 2012).

Treatment of camellia plants with different GA3 concentrations (100, 200 and 300 mg/L) at the stage of generative bud formation increased the photosynthetic rate, chlorophyll content, and photochemical activity of PSII in the 1st and 6th leaves. The most effective dose of the substance was 100 mg/L. The photosynthetic CO_2 assimilation rate positively correlated with the total chlorophyll content and fluorescence parameters in the 1st and 6th leaves (Wen et al., 2018). GA3 and 6-BAP significantly increased the photosynthetic activity in cotton plants under pre-sowing treatment of seeds and foliar treatment of plants. Growth promotors simultaneously improved crop productivity (Fang et al., 2018). The use of IAA and 6-BAP on *Epipremnum aureum* plants led to an increase in the CO_2 assimilation rate in leaves and the net photosynthetic efficiency (Di Benedetto et al., 2015). Under the action of the substances, leaves thickened, and the relative share of the intracellular volume in the mesophyll layer increased. Treatment of tomatoes under controlled conditions with GA3 and 2-NAA increased the photosynthetic rate, the fresh weight of the whole plant, and enhanced the outflow of photoassimilates to generative organs (Starck et al., 1987). In other studies, IAA increased photosynthetic rate, transpiration rate, and dry weight of *Zizania latifolia* plants in contrast to the synthetic anti-auxin 2,3,5-triiodobenzoic acid (Li et al., 2019).

The efficacy and load of growth substances on the environment are largely dependent on soil and climatic conditions, species and variety peculiarities of plants, the development stage, and compliance with drug use regulations. Therefore, the search for optimal technologies for their application, taking into account the complex peculiarities of their action on various agricultural plants, is highly relevant.

There is information in the literature on the toxicological characteristics of the growth promoting substances used in our study, as well as on the determination of their residual quantities in the soil, plant organs, and animal organisms. In particular, GA3 increased the yield of tomatoes by 24.7%, and improved the marketable appearance of the products: the fruits had the same size, shape and color. Residual amounts of GA3 in tomato fruits were determined using liquid tandem chromatography with a mass spectrometer. The samples were extracted with methanol acidified with 1% formic acid. The limits of detection and quantification of the substance in fruits were 0.01 and 0.05 mg/kg. The content of residual amounts of

GA3 was significantly lower than the maximum permissible concentrateons (Mandal et al., 2021). 6-BAP and GA3 were used on grape plantations in recommended and doubled doses in order to optimize productivity. Residual amounts of drugs were determined using liquid tandem chromatography with a mass spectrometer. The samples were extracted with methanol acidified with 1% formic acid. The limits of quantitative determination of the substance in fruits were 0.0025–0.005 μg/g. Determination of residual quantities was carried out on the 12th and 32nd day after treatment. The detected residual amounts do not exceed the maximum permissible concentrations (Ugare et al., 2013).

Other researchers point to a rapid quantitative method for the determination of 6-BAP that has been developed using surface-enhanced Raman spectroscopy (SERS). Residual amounts of the substance were determined in seedlings of various cultures. The detection rate was 0.99 and the estimated relative standard deviation was below 10% over the concentration range of 0.1–5.0 μg/mL (Zhang et al., 2018). A simple and sensitive analytical method based on solid-phase extraction using liquid chromatography and tandem mass spectrometry was developed to determine cytokinin 6-BAP in bean sprouts. The value of the relative standard deviation was less than 3.3% at enrichment levels of 20 and 40 ng/g (n = 5). The limit of detection and the limit of quantification were 2.1–3.7 ng/g and 6.3–11.1 ng/g, respectively. Monitoring of 126 bean sprout samples collected from local markets in China detected 6-BAP at 15–20 ng/g in three samples (Kim et al., 2016).

A rapid and practical method for high-performance liquid chromatography has been developed with a using NCSi-modified silica gel as a solid-phase extraction sorbent for the simultaneous determination of thidiazuron, 1-NAA, dichlorophenoxyacetic acid and 6-BAP in fruits. High-performance liquid chromatography was performed on a Waters X Bridge C18 column (inner diameter 250 × 4.6 mm, 5 μm) with a mobile phase of methanol/0.1% H_3PO_4 (55 : 45, v./v.). At a detection wavelength of 220 nm, the linearity of concentrations of growth promotors was obtained in the range from 0.03 to 200 mg/kg ($R^2 \geq 0.999$). Lower detection limits (4.3–20.4 μg/kg), and quantification limits (12.9–64.0 μg/kg) were also determined, which can match the requirements of the trace analysis of the studied substances. In addition, the overall recovery in the extraction and purification steps ranged from 72.4% to 94.9%, and the relative standard deviation was less than 4.84%. The proposed method was recognized as practical and promising in determining the trace amounts' content of growth regulators in fruits (Wang et al., 2015).

To assess the environmental safety of using growth substances, it is important to have information about their toxicological effects on plants, animals, fungi and bacteria. In particular, the potential side effects of 6-BAP on zebrafish in vivo at a rate of 0.2–25.0 mg/L were investigated. The results showed that when exposure was restricted to early life (4–36 h post-fertilization), cytokinin at a concentration of 20 mg/L induced early hatching, abnormal spontaneous movements, and premature hyperactivity in zebrafish embryos and larvae. With constant exposure to 6-BAP at a concentration of 0.2 mg/L, it caused hyperactive locomotion and transcription of genes related to neurogenesis and the endocrine system. Quantitative determination of the substance using liquid chromatography with a mass spectrometer showed that the bioaccumulation of 6-BAP in zebrafish increases under dose of 0.2 or 20 mg/L. This indicates that 6-BAP can accumulate in aquatic organisms and disrupt the neuroendocrine system. Accordingly, its effect at concentration of 0.2 mg/L increased the production of estradiol, that is, this substance is estrogenic. Therefore, induction of estrogenic effects through potential interactions with hormone receptors or disruption of downstream transcriptional signaling was a possible mechanism underlying 6-BAP toxicity (Gong et al., 2022).

It was shown that high doses of GA3 reduced the uterus size in female mice, caused the degeneration of ovarian follicles, and blood stagnation in vessels. Embryos exposed to GA3 had histopathological changes in the liver, kidneys, and skin tissues (Abu Amra et al., 2020). Other researchers studied the effect of native and synthetic auxins (IAA and 1-NAA) on the structure of membrane complexes of plant (*Arabidopsis*) and animal (white rat) organisms. It was revealed that auxins cause membrane destabilization, changes in morphology, reduce their condensation and weaken the interaction of molecules. The lipid part of membrane complexes underwent the most significant changes. Synthetic auxin 1-NAA had significant negative effects on rat hepatocyte membranes (Hac-Wydro & Flasiński, 2015). When white rats were orally administered IAA at a dose of 500 mg/kg for 14 days, a decrease in the red blood cells and hemoglobin content in them, which caused persistent anemia, leucopenia, and lymphopenia, was noted. At the same time, liver tests worsened, the condition of the myocardium and skeletal muscles, the condition and functioning of the kidneys deteriorated, and the content of testosterone and gonadotropins in the blood serum of male white rats decreased (Ismail, 2022). The introduction of native phytohormones IAA and kinetin through water at a dose of 100 mg for 21 days caused a decrease in the activity of antioxidant and immune enzyme systems (Celik et al., 2006). IAA in a dose of 1 mM caused cytotoxic effects in neutrophils and lymphocytes of rats during 24 hours of exposure. The growth regulator led to the initiation of apoptotic processes in cells (Melo et al., 2004). Damage to liver morphology and functions, and oxidative stress was observed in rats when synthetic auxins 2-NAA and 4-CPA (4-chlorophenoxyacetic acid) were used at doses of 10 and 20 mg/kg (Ozok & Celik, 2012). A decrease in antioxidant protection in rats also occurred when another auxin, indolylbutyric acid, was administered at doses of 25 and 50 ppm for 20 and 45 days (Topalca et al., 2009).

Therefore, the literature analysis showed that use of growth promotors optimizes growth of agricultural crops, and increases productivity due to beneficial changes in the plants' leaf apparatus and photosynthetic processes. However systematic studies of the effect on growth rate, morphogenesis, leaf apparatus formation, and photosynthetic processes in sweet pepper plants under the action of different groups of growth promotors are practically absent in literary sources. The details of ecologically safe methods of application of native and synthetic growth promotors, as well as the determination of their residual quantities content in fruits of the Solanaceae family agricultural crops, also remain undetermined.

In connection with the above, the aim of work was to study the effect of growth promotors 1-NAA, GA3 and 6-BAP on morphogenesis, leaf parameters, pigment content, indices of photosynthetic activity and productivity of sweet pepper *Capsicum annuum* L. plants, as well as determination of their residual amounts content in fruits.

Materials and methods

Small-scale field experiments were conducted on the lands of the farm "Berzhan P. G." village Gorbanivka, Vinnytsia district, Vinnytsia region in the growing seasons 2013–2017. Seedlings of the sweet pepper Antey variety were planted by the tape method according to the formula 80 + 50 + 50 × 25. The plots area was 32 m^2, the repetition was fivefold. Also, plants were grown under the conditions of a pot experiment in soil culture in 10 L opaque plastic pots (one plant per pot) under natural lighting. The soil was gray forest, podzolized, coarse-grained and medium-loamy. The soil-sand mixture for filling pots was prepared in a ratio of 3:1. Soil moisture was maintained at the level of 60% FC throughout the growing season.

The plants were treated at the budding stage using a sprinkler CO-12 "Marolex" (Poland) until the leaves were completely wetted with a 0.005% solution of 1 naphthaleneacetic acid (1-NAA), a 0.005% solution of gibberellic acid (GA3) and 0.005% solution of 6-benzylaminopurine (6-BAP). Control plants were treated with tap water. Plant height, plants and leaves fresh and dry weight, leaves area were determined at the flowering and fruit formation stages on ten plants.

The leaf area was determined by the die cutting method (Latimer, 2010). The leaf anatomical analysis was carried out at the period of carpogenesis on the middle tier leaves, which had completely finished growing. The leaf material was stored in a mixture of ethyl alcohol, glycerin, and water (1:1:1) with addition of 1% formalin. The chlorenchyma cells sizes were determined after partial maceration of leaf tissues by 5% solution of acetic acid in hydrochloric acid (2 mol/L). To determine the size of anatomical elements a MB-130 40×–1600× LED Mono microscope (SIGETA, Ukraine) with MOV-1-15× eyepiece micrometer was used. The repetition of measurements was thirty-five.

The photosynthesis, photo- and dark respiration rates were recorded under controlled conditions on a facility with the infrared gas analyzer GIAM-5M (RF), switched on differential scheme. The intact middle tier

leaves were placed in a temperature-controlled chamber (25 °C) and illuminated with a TA-1150 W LED spotlight with color temperature of 5200 K. The illumination at the chamber level was 1500 μmol/(m^2 • s) photosynthetically active radiation. Conditioned (humidity 9.5–10 mbar) atmospheric air flow speed through the chamber was 1 L/min. The net CO_2 assimilation rate was measured 50 min after the start of leaf illumination, when the indices of gas exchange stabilized. To assess the photorespiration rate, the CO_2 burst from the leaf was recorded within 1 min after turning off the light. The dark respiration rate was recorded after 10 min exposition of leaf in the dark. The CO_2 gas exchange rate was calculated by the changes in its concentration in the air after the chamber compared to the atmospheric one. The transpiration rate was recorded under controlled conditions with gas analyzer EGM-5 (PP Systems, USA), and was calculated on the difference in air humidity at the inlet and outlet of the leaf chamber. The leaf gas exchange indices were calculated according to the methods described in (Laisk & Oja, 1998). The repetition of gas exchange measurements was three times.

The total chlorophyll content was measured in the fresh leaf tissue by the spectrophotometric method on a ULAB 102UV spectrophotometer (Shanghai Metash Instruments Co., China) in five repetitions (Latimer, 2010). The net photosynthetic efficiency (NPE) was determined as the increment in the dry weight of leaf area unit per unit of time, the leaf area index – as the total leaf area on the unit of ground area, the chlorophyll index – as the product of total chlorophyll content in leaves on their area, and the specific leaf area – as the ratio of leaves dry weight to their area.

The chlorophyll fluorescence induction indices were measured by a portable single-beam fluorimeter "Floratest" (V. M. Hlushkov Institute of Cybernetics, NAS of Ukraine). The remote optoelectronic sensor includes a light-emitting diode with wavelength of irradiation 470 ± 15 nm on the area 15 mm^2, and illuminance not less than 5000 mcd. The optoelectronic sensor perceives the signal of fluorescence intensity in spectral range 670–770 nm; the reception window area – 9 mm^2; the photodetector sensitivity 650 nm – 0.45 A/W. Exposure time was 240 s with recording data 90 times according to the $5\sqrt{t}$ law. The time dynamics of the chlorophyll fluorescence intensity was obtained in the form of a Kautsky curve (Brayon et al., 2000; Gol'cev et al., 2016; Rohach et al., 2023).

The residual amounts of growth promotors 1-NAA and 6-BAP in tomato fruits were determined as follows. The homogenate (250 g) was placed in a separatory funnel (500 cm^3) with 30 cm^3 of dichloromethane and shaken for 1 min. After complete phase separation, the lower organic layer was discarded. To the aqueous phase in the separatory funnel 1 cm^3 of sodium tetrafluoroborate solution and 30 cm^3 of dichloromethane were added, and shaken for 1 min. The lower organic layer was transferred in a 200 cm^3 flat-bottom flask. The extraction operation was repeated with a new portion (30 cm^3) of dichloromethane. The settled organic layer was added to the previously obtained extract.

The total extract was placed in a 250 cm^3 separatory funnel, 25 cm^3 of a 2 M HCl solution was added, and it was shaken for 1 min. The lower organic layer was separated and discarded. Then 25 cm^3 of dichloromethane was added to a separatory funnel, and shaken for 1 min. The lower organic layer was separated and discarded. The aqueous phase was placed in a 150 cm^3 round-bottom flask, and evaporated to a dry fraction on a vacuum rotary evaporator at a temperature of 60 °C. To the flask with the dry extract, deionized water (25 cm^3) was added, and evaporated to dryness again. The evaporation with an addition of 25 cm^3 deionized water was repeated twice (for complete removal of HCl). The residue in the flask was dissolved in 10 cm^3 of acetonitrile-methanol mixture (95:5) and transferred into the prepared column. The flask was washed with 5 cm^3 of this mixture, and also introduced into the column. After absorption of the solution, the substance was eluted from the column in a 250 cm^3 evaporating flask with 85 cm^3 acetonitrile-methanol mixture (95:5) at a flow rate of 1–2 drops per second. The obtained solution was dried on a rotary vacuum evaporator at a temperature of 45 °C.

The residue was transferred from the flask to a vial by 3 portions (1 cm^3) of methanol under a stream of nitrogen. The vial was placed in a thermostat at 45–50 °C with addition of 50 mm^3 10% NaOH solution. The vial was closed with a metal cap with a silicone spacer, which was crimped with a crimper and additionally reinforced with wire. The vial was heated to 200 °C for 15 min in a thermostat. After that, it was cooled to room temperature. The gas phase was analyzed using a Shimadzu GC gas chromatograph with a mass spectrometric detector GCMS-QP2020 EI under the following conditions: capillary column – Rxi-5ms (Serial No. 1544328), length – 30 m, diameter – 0.25 mm, phase – 0.25 μm, constant flow – 1.2 mL/min, carrier gas – helium. Injector – auto-injector AOS-20i+s, Split 20:1, evaporator temperature T = 250 °C. Thermostat – Tstart = 100 °C (2 min), heating – 15 °C/min to Tend = 280 °C (10 min). The detector was mass-selective, the temperature of the interface – 280 °C, ionization by electron impact, the temperature of the ion source – 280 °C. Sample – 5.0 μL, automatic input. Full ion current chromatograms were registered (m/z = 50–550). Mass chromatograms were analyzed using LabSolutions software and the NIST14.lib mass spectral database.

The percentage content of the analyzing substance was calculated according to the formula:

$$\omega = (c/r) * (s/p) * 100\%,$$

where ω – percentage content of the analyzing substance (%), c – concentration of the solution of the analyzing substance standard sample (mg/cm^3), r – the ratio of the analyzing substance weight to the volume of the extractant (mg/cm^3), s – the area of the peak of the analyzing substance (c.u.), p – peak area of the standard sample of the analyzing substance (c.u.).

The mass of the analyzing substance in the test sample was calculated according to the formula:

$$m = (\omega * n)/100\%,$$

where m – the mass of the analyzing substance in the test sample (g), ω – percentage content of the analyzing substance (%), n – the mass of the test sample (g).

The results obtained were processed statistically with the Statistica 6.0 (StatSoft Inc., USA) computer program. Univariate analysis of variance was used (differences between mean values were calculated by ANOVA with Bonferroni's correction, they were considered significant at $P < 0.05$) (Van Emden, 2008).

Results

Foliar treatment of sweet pepper at the budding stage with aqueous solutions of 1 NAA, GA3 and 6-BAP affected the linear growth of plants. GA3 reliably increased the height of pepper plants at the flowering and fruit formation stages by 24% and 18%, respectively (Table 1).

The main source of plastic substances in a plant is the leaves, so it is important to study the effect of growth promotors on the plants' leaf apparatus. It was revealed that 1-NAA, GA3, and 6-BAP increased the leaves' number on the plant by 19–38%, the leaves' fresh weight – by 19–34%, and the leaves dry weight – by 17–36% (Table 1).

One of the main parameters that affect the yield of agricultural crops is the leaf area on an individual plant and the leaf area index. It was found that these parameters under the treatment of 1-NAA increased at the flowering and fruiting stages by 24% and 32%, after the application of GA3 – by 19% and 24%, and under the action of 6-BAP – by 19% and 12%, respectively (Table 1). The specific leaf weight indicates its provision with structural elements involved in photosynthetic processes on a leaf surface unit, and indirectly related to the leaf lamina's thickness. The results of our research show that all growth promotors tend to increase this index or did not change it. The highest value of the specific leaf weight was noticed at the stage of fruit formation under the treatment with 6-BAP (raise by 13% compared to control) (Table 1).

These results are to a certain extent consistent with the data of analysis of leaf lamina mesostructure (Table 2). It was revealed that treatment of plants with growth promotors 1-NAA, GA3 and 6-BAP increased the thickness of chlorenchyma by 5%, 23% and 13%, respectively. At the same time, the thickness of the upper and lower epidermis increased significantly only under the use of GA3. It was found that growth regulators GA3 and 6-BAP increased the volume of columnar parenchyma cells by 34% and 16%, respectively. An increase in the length and width of spongy parenchyma cells was also noticed. These indices increased most significantly under treatment with 1-NAA (by 34% and 39%), and under GA3 – by 20% and 30%, respectively.

Table 1
Effect of foliar treatment at the budding stage with growth promoting substances on morphometric and cenotic indices, and pigment content in leaves of *Capsicum annuum* L. cv. Antey (x ± SE, n = 10)

Index	Control		1-NAA		GA_3		6-BAP	
	1	2	1	2	1	2	1	2
Plant height, cm	38.7 ± 1.9	47.8 ± 2.3	42.6 ± 2.1	50.6 ± 2.4	48.0 ± 2.4*	56.5 ± 2.8*	40.7 ± 2.0	49.4 ± 2.4
The number of leaves on a plant, pcs.	80.5 ± 4.0	98.7 ± 4.9	99.9 ± 4.9*	117.2 ± 5.6*	107.3 ± 5.2**	126.6 ± 6.3**	111.2 ± 5.3**	128.2 ± 6.2**
Leaves fresh weight, g	60.0 ± 3.0	97.2 ± 4.2	76.4 ± 3.4**	113.9 ± 5.7*	71.3 ± 3.5*	118.4 ± 5.9*	76.8 ± 3.6*	130.0 ± 6.4**
Leaves dry weight, g	12.92 ± 0.58	20.11 ± 0.98	17.63 ± 0.88**	23.35 ± 1.11	16.51 ± 0.77**	24.80 ± 1.18*	17.42 ± 0.85**	25.78 ± 1.28**
Leaf area, cm^2	1735 ± 83	1939 ± 96	2143 ± 106*	2549 ± 122**	2065 ± 101*	2399 ± 119*	2056 ± 101*	2174 ± 108
Leaf index, m^2/m^2	1.16 ± 0.05	1.29 ± 0.06	1.43 ± 0.07*	1.70 ± 0.08**	1.38 ± 0.06*	1.60 ± 0.08*	1.37 ± 0.06*	1.45 ± 0.07
Leaf specific weight, mg/cm^2	7.74 ± 0.38	10.97 ± 0.55	8.65 ± 0.41	10.92 ± 0.53	8.40 ± 0.41	10.73 ± 0.52	8.72 ± 0.42	12.83 ± 0.61*
The total chlorophyll ($a + b$) content, % f.w.	0.488 ± 0.012	0.504 ± 0.014	0.510 ± 0.015	0.524 ± 0.016	0.460 ± 0.011	0.473 ± 0.013	0.538 ± 0.017*	0.553 ± 0.016*
Chlorophyll index, g/m^2	1.85 ± 0.08	2.31 ± 0.11	1.95 ± 0.09	2.47 ± 0.12	1.59 ± 0.06*	2.14 ± 0.11	2.02 ± 0.09	3.01 ± 0.14**

Note: differences between mean values were calculated using the ANOVA test, with Bonferroni's correction, which is considered significant at * – P < 0.05; ** – P < 0.01; *** – P < 0.001 compared to the control at this stage of vegetation; 1 – flowering stage; 2 –fruit formation stage; 1-NAA – 0.005% solution of 1- naphthaleneacetic acid, GA3 – 0.005% solution of gibberellic acid, 6-BAP – 0.005% solution of 6-benzylaminopurine.

It was established that only under the action of 6-BAP (by 10%) was there a significant increase in the amount of chlorophylls (a + b) in sweet pepper leaves. Under GA3 treatment, the pigments content tended to decrease, and under 1-NAA, it tended to increase (Table 1). The plants' chlorophyll index under 6-BAP treatment significantly increased (by 30%) at the fruit formation stage. Under the GA3 treatment, this index decreased significantly (by 14%) at the flowering stage, and tended to decrease at the fruit formation stage. In other treatments, the tendency to increase of this cenotic index was observed (Table 1).

Gas exchange processes in the leaves are important parameters of plants' photosynthetic apparatus activity. The data of measuring the gas exchange parameters of sweet pepper leaves indicate that 1-NAA significantly (by 36%) increased the net photosynthetic rate at the flowering stage, while under the action of 6-BAP only a tendency to increase of this index was observed (Table 3). At the same time, at the beginning of fruit formation stage under the influence of all growth promotors, except for GA3, a steady trend towards an increase in the photosynthetic rate was observed. GA3 during both stages of vegetation significantly reduced CO_2 assimilation (by 21% and 43%).

At the flowering stage and at the beginning of fruit formation stage, 1-NAA simultaneously with the intensification of photosynthetic CO_2 assimilation increased the photorespiration rate (Table 3). Under GA3 treatment, photorespiration decreased (by 35%) simultaneously with a decreese in the photosynthetic rate at the fruit formation stage. At the fruit formation stage, an increase up to 30% in the dark respiration rate under the influence of all growth promotors was observed (Table 3).

Table 2
Effect of foliar treatment at the budding stage with growth promoting substances on leaf mesostructural parameters of *Capsicum annuum* L. cv. Antey fruit formation stage (x ± SE, n = 35)

Index	Control	1-NAA	GA_3	6-BAP
Leaf thickness, μm	263 ± 2.7	274 ± 4.1**	307 ± 6.5***	299 ± 5.7***
Upper epidermis thickness, μm	23.32 ± 0.62	22.87 ± 0.57	31.08 ± 0.21***	28.71 ± 0.73***
Chlorenchyma thickness, μm	216 ± 1.7	228 ± 2.9**	247 ± 5.8***	245 ± 4.1***
Lower epidermis thickness, μm	23.92 ± 0.49	23.96 ± 0.62	29.63 ± 0.53***	25.07 ± 0.85
Columnar parenchyma cells volume, μm^3	19857 ± 896	20637 ± 817	26689 ± 1117***	23059 ± 1147*
Spongy parenchyma cells length, μm	33.28 ± 0.95	42.75 ± 0.74***	39.81 ± 0.78***	34.06 ± 1.30
Spongy parenchyma cells width, μm	24.95 ± 0.75	33.35 ± 0.82***	32.43 ± 0.89***	26.92 ± 1.04

Note: differences between mean values were calculated using the ANOVA test, with Bonferroni's correction, which is considered significant at * – P < 0.05; ** – P < 0.01; *** – P < 0.001 compared to the control; 1-NAA – 0.005% solution of 1- naphthaleneacetic acid, GA3 – 0.005% solution of gibberellic acid, 6-BAP – 0.005% solution of 6-benzylaminopurine.

Table 3
Effect of foliar treatment at the budding stage with growth promoting substances on the photosynthetic, photo- and dark respiration, and transpiration rates of *Capsicum annuum* L. cv. Antey leaves at the end of flowering (1) and the beginning of fruit formation stages (2) (x ± SE, n = 3)

Treatment	Net photosynthesis, $\mu mol\ CO_2/(m^2 \cdot s)$		Photorespiration, $\mu mol\ CO_2/(m^2 \cdot s)$		Dark respiration, $\mu mol\ CO_2/(m^2 \cdot s)$		Transpiration, $mmol\ CO_2/(m^2 \cdot s)$	
	1	2	1	2	1	2	1	2
Control	7.56 ± 0.38	10.87 ± 0.51	0.79 ± 0.07	1.42 ± 0.07	0.47 ± 0.03	0.63 ± 0.03	2.25 ± 0.09	2.96 ± 0.12
1-NAA	10.24 ± 0.52*	11.18 ± 0.58	1.10 ± 0.08	1.58 ± 0.08	0.79 ± 0.07*	0.79 ± 0.07	1.42 ± 0.06**	2.13 ± 0.09*
GA_3	5.99 ± 0.26*	6.14 ± 0.28**	0.79 ± 0.06	0.95 ± 0.05**	0.47 ± 0.02	0.79 ± 0.06	1.42 ± 0.06**	1.78 ± 0.08**
6-BAP	8.03 ± 0.33	11.03 ± 0.52	0.95 ± 0.08	1.26 ± 0.06	0.63 ± 0.03*	0.79 ± 0.07	2.49 ± 0.11	2.73 ± 0.12

Note: see Table 1.

The treatment of sweet pepper plants with growth promotors also affected the transpiration rate. 1-NAA and GA3 significantly reduced this index during both investigated stages of ontogenesis (by 37% and 28%, and 37% and 40%, respectively), and after the application of 6-BAP it tended to increase during flowering and to decrease during the fruit formation stage (Table 3).

Important indices of the photosynthesis efficiency are the parameters of the Kautsky curve, which characterize the level of chlorophyll fluorescence, light photosynthetic reaction rate, the photochemical and non-photochemical fluorescence quenching rate, the PSII efficiency and quantum yield, the electron transport rate in photosystems. The results of our research show that growth promotors changed these parameters (Table 4). At the flowering stage GA3 and 6-BAP decreased minimal fluorescence (F_0) level, and 1-NAA did not change it. At the fruit formation stage GA3 increased it, and other substances did not affect this parameter. At the flowering stage, 1-NAA, and during both vegetation stages, GA3 increased the fluorescence level at the time of reaching a temporary slowdown in its increase ($F_=$), the steady-state terminal fluorescence (F_s), and the maximal fluorescence level (F_m) compared to the control. Under treatment with 6-BAP, the F_p, F_s and F_m indices did not differ significantly from the control. The level of variable fluorescence (F_v) under GA3 treatment significantly exceeded the control both at the flowering stage (by 2.7 times) and at the fruiting stage (by 1.6 times). When using 1-NAA and 6-BAP, F_v increased reliably only at the flowering stage – by 50% and 34%, respectively.

The maximum quantum efficiency of PSII photochemical reactions (Kl) is the most important parameter of the chlorophyll fluorescence induction. It characterizes the efficiency of plants' photosynthetic apparatus

with optimal value about 0.83. A significant increase in the Kl occurred at the flowering stage under GA3 (by 43%) and 6-BAP (by 25%) treatment. In other treatments, a tendency towards an increase in this index was mainly observed.

The coefficient of chlorophyll fluorescence decay (Kg), which correlates with CO_2 assimilation rate, significantly exceeded the control after treatment with 6-BAP and 1-NAA both at the flowering (by 88% and 98%, respectively) and fruit formation stages (by 55% and 65%). Under the influence of GA3, Kg tended to increase at the fruit formation stage.

The rate of linear electron transport (Kf), indicates the activity of photosynthetic processes in the leaf. It was found that Kf increased reliably under the treatment of 1 NAA (by 55%) and 6-BAP (by 51%) at flowering, and tended to increase at fruiting stage. Under the action of GA3, this index did not change significantly compared to the control.

Table 4

Effect of foliar treatment at the budding stage with growth promoting substances on chlorophyll fluorescence parameters in leaves of *Capsicum annuum* L. cv. Antey at the stages of flowering (1) and fruit formation (2) (x ± SE, n = 3)

Index	Control		1-NAA		GA_3		6-BAP	
	1	2	1	2	1	2	1	2
Minimal fluorescence (relative units) (F_0)	424 ± 19.2	144 ± 6.8	512 ± 25.5	144 ± 7.1	312 ± 15.5*	192 ± 9.1*	296 ± 14.4*	144 ± 6.9
The level of fluorescence at the time of reaching a temporary slowdown in its increase (F_p)	636 ± 31	416 ± 19	880 ± 39*	416 ± 19	960 ± 45**	576 ± 29*	656 ± 33	432 ± 21
Maximal fluorescence (relative units), which is proportional to the total amount of chlorophyll (F_m)	1020 ± 50	1632 ± 81	1408 ± 70*	1632 ± 80	1904 ± 91*	2512 ± 122**	1096 ± 54	1344 ± 67
Steady-state terminal fluorescence (relative units), which is defined by the dynamic balance of photosynthetic processes (F_s)	728 ± 36	560 ± 28	785 ± 39	560 ± 27	1316 ± 66**	1056 ± 51**	624 ± 30	480 ± 23
Variable fluorescence – the level of which characterizes the activity of primary photochemical processes in PSII ($F_v = F_m - F_0$)	596 ± 29	1448 ± 72	896 ± 44*	1488 ± 71	1592 ± 78**	2320 ± 111**	800 ± 39*	1200 ± 59
Maximum quantum efficiency of PSII photochemical reactions (index of the photosynthesis light phase overall efficiency) ($Kl = F_v/F_m$)	0.584 ± 0.028	0.910 ± 0.045	0.636 ± 0.032	0.912 ± 0.044	0.836 ± 0.038*	0.924 ± 0.041	0.731 ± 0.036*	0.893 ± 0.043
Index of the PSII reaction centers fraction that do not reduce Q_B acceptor ($Kn = (F_p - F_0)/Fv$)	0.664 ± 0.032	0.812 ± 0.039	0.589 ± 0.028	0.817 ± 0.038	0.593 ± 0.029	0.834 ± 0.041	0.550 ± 0.027	0.760 ± 0.036
The coefficient of chlorophyll fluorescence decay, which correlates with CO_2 assimilation rate ($Kg = (F_m - F_s)/F_s$),	0.401 ± 0.019	1.163 ± 0.055	0.795 ± 0.039**	1.914 ± 0.089**	0.447 ± 0.022	1.379 ± 0.063	0.756 ± 0.037**	1.800 ± 0.088**
The actual quantum efficiency of PSII photochemistry, which characterizes the rate of linear electron transport ($Kf = (F_m - F_s)/F_m$)	0.286 ± 0.012	0.538 ± 0.025	0.443 ± 0.022**	0.657 ± 0.032	0.309 ± 0.014	0.580 ± 0.028	0.431 ± 0.021**	0.643 ± 0.032
The coefficient of chlorophyll fluorescence photochemical quenching, which characterizes the share of PSII open reaction centers ($Kq = (F_m - F_s)/(F_m - F_0)$)	0.490 ± 0.023	0.591 ± 0.029	0.696 ± 0.032*	0.720 ± 0.035	0.369 ± 0.018*	0.628 ± 0.028	0.590 ± 0.027	0.720 ± 0.035

Note: see Table 1.

The coefficient of chlorophyll fluorescence photochemical quenching, which characterizes the share of PSII open reaction centers (Kq), significantly increased at flowering after the treatment with 1-NAA (by 42%), and tended to increase at the stage of fruit formation under the treatment of 1-NAA and 6-BAP. Under GA3 treatment, the Kq decreased significantly (by 25%) at flowering.

The index of the PSII reaction centers fraction that do not reduce QB acceptor (Kn) characterizes the relative number of inactive reaction centers that do not participate in the electron transport to the pool of plastoquinones. It was revealed that Kn practically did not change under the treatment of sweet pepper plants with 1-NAA and GA3, and tended to decrease at both stages of ontogenesis after the treatment with 6-BAP (by 17% and 6%). The increase in the growth processes rate, leaf apparatus formation, and changes in photosynthetic processes affected the net photosynthetic efficiency and accumulation of dry matter by plants. All growth promotors increased the whole plant dry weight both at the flowering and fruit formation stages. At the end of the studied period, this index after the application of 1-NAA, GA3 and 6-BAP increased compared to the control by 19%, 32% and 26%, respectively (Table 5).

An important parameter that indicates the plants' photosynthetic apparatus performance is the net photosynthetic efficiency. The results showed that at flowering stage, growth promotors 1-NAA, GA3, and 6-BAP reduced this index value by 28%, 19%, and 17%, respectively, and under treatment with GA3 and 6-BAP, it significantly increased at the fruit formation stage compared to the control by 32% and 19%, respectively.

The analysis of dry weight ratio of vegetative and generative organs of sweet pepper revealed that treatment with synthetic cytokinin 6-BAP led to the redistribution of plastic substances towards fruits due to a decrease in the dry weight primarily of shoots and roots, especially at the fruit formation stage (Fig. 1). The application of 1-NAA increased the proportion of roots dry weight compared to the control.

The results of our research indicate changes in the elements of sweet pepper productivity (Table 6). Growth promotors 1-NAA, GA3 and 6-BAP increased the fruits number per plant by 15%, 28% and 21%, respectively. The average weight of one fruit did not change reliably, and the fruits' weight from one plant increased by 17%, 22% and 20%, respectively.

The analysis of growth regulators residual amounts content in sweet pepper fruits showed that content of 1-NAA was 0.0004 mg/kg, and after the application of 6-BAP, the substance was contained in the fruits in trace amounts. In accordance with State Sanitary Rules and Regulations 8.8.1.2.3.4-000-2001, residual amounts of these substances in agricultural products should not exceed 0.01 mg/kg. Thus, no excess of the maximum permissible concentration of growth promotors was found.

Discussion

The plant functioning as a source-sink self-regulating system depends on a number of exogenous and endogenous factors, among which regulation by native hormones and their synthetic analogues or modifiers is quite significant, since changes in growth, physiological and biochemical processes determine the restructuring of the entire plant organism (Stasik & Kiriziy, 2011). Stimulation of growth and development processes is associated with mobilization of plant genetic potential, and the directing of assimilative resources to increase biological productivity, in contrast to the effects caused by growth inhibitors, although the action of the latter, as is known, can also be accompanied by an increase in yield due to the redistribution of plastic substances between plant organs.

Table 5

Effect of foliar treatment at the budding stage with growth promoting substances on biological productivity of *C. annuum* L. cv. Antey (x ± SE, n = 10)

Index	Control		1-NAA		GA_3		6-BAP	
	1	2	1	2	1	2	1	2
Plant dry weight, g	61.6 ± 3.0	83.7 ± 4.2	74.9 ± 3.7*	99.8 ± 4.9*	75.8 ± 3.7*	111.3 ± 5.6**	74.5 ± 3.7*	105.2 ± 5.3*
Net photosynthetic efficiency, g/(m^2 · day)	3.62 ± 0.18	2.21 ± 0.11	2.62 ± 0.12**	2.17 ± 0.09	2.92 ± 0.14*	2.91 ± 0.14**	3.02 ± 0.15*	2.63 ± 0.12*

Note: see Table 1.

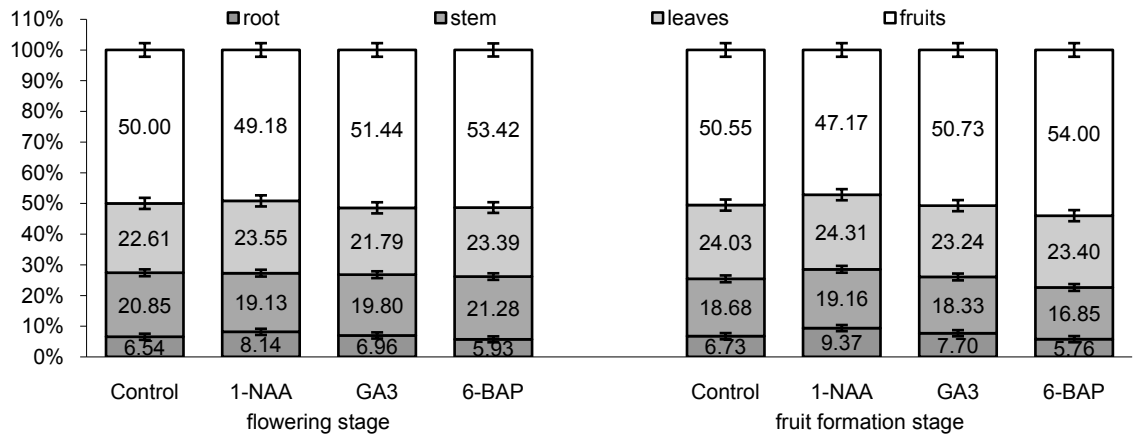

Fig. 1. Effect of foliar treatment at the budding stage with growth promoting substances on the whole plant organs' dry weight ratio in *Capsicum annuum* L. cv. Antey (x ± SE, n = 10): 1-NAA – 0.005% solution of 1-naphthaleneacetic acid, GA3 – 0.005% solution of gibberellic acid, 6-BAP – 0.005% solution of 6-benzylaminopurine

Table 6
Effect of foliar treatment with growth promoting substances on plant productivity elements of *Capsicum annuum* L. cv. Antey (x ± SE, n = 10)

Index	Control	1-NAA	GA_3	6-BAP
Number of fruits per plant, pcs.	6.08 ± 0.28	7.01 ± 0.32	7.81 ± 0.38**	7.36 ± 0.35*
The average weight of one fruit, g	88.7 ± 4.2	90.1 ± 4.4	83.9 ± 4.1	88.2 ± 4.4
Weight of fruits from one plant, g	539 ± 25	631 ± 29*	655 ± 32*	649 ± 32*

Note: treatment of plants at the budding stage, determining of indices at the fruit ripening stage; * – P < 0.05; ** – P < 0.01; comparison to control was carried out by the method of difference between average values calculated according to the ANOVA criterion with Bonferroni's correction; 1-NAA – 0.005% solution of 1-naphthaleneacetic acid, GA3 – 0.005% solution of gibberellic acid, 6-BAP – 0.005% solution of 6-benzylaminopurine.

A significant increase in the linear size of pepper plants was observed only when GA3 was applied. Other growth promotors did not noticeably change plant height. Similar morphological effects after treatment with this growth substance were revealed on eggplant (Rogach et al., 2020) and tomato (Rogach et al., 2022; Rohach et al., 2023).

The main source of assimilates in the plant is the leaf. It is changes in the structure and functioning of the leaf apparatus as a source of plastic substances that are key in the plant's "performance." Increased activity of all types of meristematic tissues under the influence of growth promotors contributed to the formation of larger plant (Aremu et al., 2017; Madzikane-Mlungwana et al., 2017), in which a more powerful leaf apparatus was formed accordingly (Rogach et al., 2017). The laying of more leaves under the influence of growth promotors, and the increase in their area and fresh and dry weight led to the activation of photosynthetic processes, and strengthened the source function of the leaf. Similar changes in the leaf apparatus structure under the influence of gibberellins and cytokinins were noted by other researchers (Madzikane-Mlungwana et al., 2017; Rai et al., 2017).

A separate direction of influence of growth promotors is the leaf mesostructural organization. The mitotic activity strengthening under the action of these substances contributed to the thickening of leaf laminas due to the growth of assimilation tissue. This manifested in an increase in the number of mesophyll cells in all treatments, and the size of spongy cells, as well as the volume of columnar parenchyma cells. Such effect of growth promotors on the mesostructure of sweet pepper leaf laminas can create prerequisites for increasing the photosynthetic performance of the culture. The thickening of leaves due to the growth of chlorenchyma, the increasing of spongy parenchyma cells size and the volume of columnar parenchyma cells under the influence of growth promotors was noted in our earlier works in small-scale field experiments with eggplant (Rogach et al., 2017), and in pot experiments with sweet pepper (Rogach et al., 2021). Optimizing the anatomical-morphological and mesostructural characteristics of the leaf apparatus after treatment with growth promotors, as expected, resulted in positive changes in photosynthetic processes in the plant.

A significant intensification of photosynthesis under 1-NAA treatment and a tendency to its increase under 6-BAP treatment enhanced photorespiration (Table 3). A decrease in the photosynthetic rate under GA3 treatment led to a decrease in photorespiration. This can be explained by the fact that CO_2 uptake due to photosynthetic assimilation and its release from the leaf due to photorespiration are manifestations of the Rubisco carboxylase and oxygenase activity. In addition to this, the above results show that treatment of pepper plants with growth promotors in most cases increased the photosynthetic apparatus capacity at all levels of plant organism. This should have increased its supply by assimilates, primarily carbohydrates, which are the main substrate of respiratory processes. The reliable increase in the dark respiration rate under the influence of growth promotors both at the flowering and fruit formation stages indirectly confirms this suggestion. The fact that high indices of the plant's biological productivity were accompanied with high photo- and dark respiration rate also attracts attention.

A decrease in the transpiration rate against the background of an increase in the photosynthesis means an increase in the water use efficiency, which is also an important index of optimizing the plant's production process. The influence of growth promotors on the photosynthetic apparatus also manifested itself at the level of chloroplast thylakoids, as evidenced by the changes in fluorescence induction parameters. Under the influence of all substances, most of the important indices characterizing the PSII photosynthetic activity significantly increased, tended to increase, or at the very least did not decrease compared to the control. At the same time, the Kn index, which characterizes the relative number of inactive reaction centers, tended to decrease or did not change reliably compared to the control. This can indicate the enhancement of using the potential capabilities of the sweet pepper photosynthetic apparatus under treatment with growth promotors.

The net photosynthetic efficiency, which largely depends on the total leaf surface area, the leaves' functional state, and the demand for assimilates from the attractive centers, is an integral index of the photosynthetic apparatus activity (Stasik & Kiriziy, 2011). The higher values of this parameter were revealed under GA3 and 6-BAP treatment, which clearly match with the higher yield of the sweet pepper culture.

The analysis of dry weight ratio of whole plant organs shows that during the stage of fruit formation – the main sink of assimilates in plants, their share increased under the action of growth promotors. At the same time, the mass share of assimilates sources – leaves, at this stage of ontogenesis, practically did not change, while the share of another powerful sink – stems, tended to decrease. Enhanced accumulation of dry matter by plants, and a tendency towards an increase in the net photosynthetic effi-

ciency after the application of growth promotors were earlier noted by us (Rogach et al., 2022; Rohach at al., 2023) and other researchers (Rai et al., 2017; Ren et al., 2017). Growth promotors contributed to the initiation of a greater number of fruits.

Conclusions

Thus, treatment of *Capsicum annuum* L. cv. Antey with growth promotors – 0.005% 1 naphthaleneacetic acid (1-NAA), 0.005% gibberellic acid (GA3), and 0.005% 6-benzylaminopurine (6-BAP) solutions at the budding stage changed the plants' morphogenesis, structure and functioning of photosynthetic apparatus, and increased the crop productivity.

The plants' linear dimensions increased under GA3 treatment. 1-NAA, GA3, 6-BAP increased the number of leaves per plant, fresh and dry leaf weight, leaf surface area, and plantation leaf area index. 6-BAP increased the leaf specific weight. All growth promotors thickened leaf laminas due to the growth of assimilating parenchyma cells. Under the influence of GA3 and 6-BAP, the volume of columnar parenchyma cells enlarged, and after the 1-NAA and GA3 treatment, the spongy parenchyma cell size increased. 6-BAP increased the amount of total chlorophyll in leaves, while GA3 decreased it.The plants'chlorophyll index under treatment with 6-BAP increased, and after the application of GA3 decreased.

The whole plant fresh weight increased after treatment with all growth promotors. The net photosynthetic efficiency exceeded the control at the fruit formation stage after 6-BAP treatment. Under the influence of 1-NAA and 6-BAP, the net CO2 assimilation rate increased, and under GA3 treatment, it decreased compared to the control. GA3 reduced photorespiration and transpiration. 6-BAP increased transpiration, and 1-NAA decreased it. All substances tended to increase dark respiration.

GA3 and 6-BAP reliably increased the maximum quantum efficiency of PSII. 1-NAA and 6-BAP increased the rate of linear electron transport. The chlorophyll fluorescence photochemical quenching significantly increased at flowering after the application of 1-NAA, and tended to increase at the fruit formation stage under the treatment with 1-NAA and 6-BAP. Under GA3 treatment, this index was significantly reduced at flowering. The fraction of PSII inactive reaction centers (that do not reduce QB acceptor) did not change under the 1-NAA and GA3 treatment, and tended to decrease after the 6-BAP application.

The treatment with all three growth promotors significantly increased fruit yield. The residual amounts of 1-NAA and 6 BAP in ripe fruits did not exceed the maximum permissible.

Thus, anatomical-physiological changes in sweet pepper plants under the action of growth promoting substances resulted in the increase in the rate of of growth processes, leaf weight and area, the gain of photosynthetic apparatus activity due to the chlorophyll content rise (6-BAP) and PSII photochemical processes enhancement (1-NAA, GA3 and 6 BAP), which increased the plants' biological productivity. The results obtained provide a new practical approach for increasing the sweet pepper yield. With that, certain questions regarding the action mechanisms on molecular and physiological levels of the growth promotors used require further study.

The research was carried out within the framework of the state budget topic "Development of technological solutions for the redistribution of photoassimilates to economically valuable organs of agricultural crops with the help of hormonal status modifiers" No. 0121U109625, which was funded by the Ministry of Education and Science of Ukraine.

The authors declare no conflict of interest.

References

Abu Amra, E. S., Lashein, F. E. D. M., Seleem, A. A., & Saleh, M. M. (2020). The protective role of olive oil against gibberellic acid-induced embryotoxicity at prenatal stages of mice. The Journal of Basic and Applied Zoology, 81(1), 44.

Ahanger, M. A., Alyemeni, M. N., Wijaya, L., Alamri, S. A., Alam, P., Ashraf, M., & Ahmad, P. (2018). Potential of exogenously sourced kinetin in protecting *Solanum lycopersicum* from NaCl-induced oxidative stress through up-regulation of the antioxidant system, ascorbate-glutathione cycle and glyoxalase system. PLoS One, 13(9), e0202175.

Alexopoulos, A. A., Karapanos, I. C., Akoumianakis, K. A., & Passam, H. C. (2017). Effect of gibberellic acid on the growth rate and physiological age of tubers cultivated from true potato seed. Journal of Plant Growth Regulation, 36(1), 1–10.

Aremu, A. O., Plackova, L., Masondo, N. A., Amoo, S. O., Moyo, M., Novak, O., Dolezal, K., & Staden, J. V. (2017). Regulating the regulators: responses of four plant growth regulators during clonal propagation of *Lachenalia montana*. Plant Growth Regulation, 82(2), 305–315.

Brayon, O. V., Kornieiev, D. Y., Sniegur, O. O., & Kytaiev, O. I. (2000). Instrumentalne vyvchennia fotosyntetychnogo aparatu za dopomogoiu induktsii fluorestsentsii khorofilu [Instrumental study of photosynthetic apparatus by chlorophyll fluorescence induction]. Kyiv University, Kyiv (in Ukrainian).

Celik, I., Tuluce, Y., & Turker, M. (2006). Antioxidant and immune potential marker enzymes assessment in the various tissues of rats exposed to indoleacetic acid and kinetin: A drinking water study. Pesticide Biochemistry and Physiology, 86(3), 180–185.

de Melo, M. P., De Lima, T. M., Pithon-Curi, T. C., & Curi, R. (2004). The mechanism of indole acetic acid cytotoxicity. Toxicology Letters, 148, 103–111.

Di Benedetto, A., Galmarini, C., & Tognetti, J. (2015). Effects of combined or single exogenous auxin and/or cytokinin applications on growth and leaf area development in *Epipremnum aureum*. The Journal of Horticultural Science and Biotechnology, 90(6), 643–654.

Fang, S., Gao, K., Hu, W., Wang, S., Chen, B., & Zhou, Z. (2018). Foliar and seed application of plant growth regulators affects cotton yield by altering leaf physiology and floral bud carbohydrate accumulation. Field Crops Research, 231, 105–114.

Froschle, M., Horn, H., & Spring, O. (2017). Effects of the cytokinins 6-benzyladenine and forchlorfenuron on fruit-, seed- and yield parameters according to developmental stages of flowers of the biofuel plant *Jatropha curcas* (Euphorbiaceae). Plant Growth Regulation, 81(2), 293–303.

Gao, Z., Liang, X. G., Zhang, L., Lin, S., Zhao, X., Zhou, L. L., Shen, S., & Zhou, S. L. (2017). Spraying exogenous 6-benzyladenine and brassinolide at tasseling increases maize yield by enhancing source and sink capacity. Field Crops Research, 211, 1–9.

Gol'cev, V. N., Kaladzhi, H. M., Paunov, M., Baba, V., Horachek, T., Mojski, J., Kocel, H., & Allahverdiev, S. I. (2016). Ispol'zovanie peremennoj fluorescencii hlorofilla dlja ocenki fiziologicheskogo sostojanija fotosinteticheskogo apparata rastenij [Variable chlorophyll fluorescence and its use for assessing physiological condition of plant photosynthetic apparatus]. Fiziologija Rastenij, 63(6), 881–907 (in Russian).

Gong, G., Kam, H., Chen, H., Chen, Y., San Cheang, W., Giesy, J. P., Zhou, Q., & Lee, S. M. Y. (2022). Role of endocrine disruption in toxicity of 6-benzylaminopurine (6-BA) to early-life stages of zebrafish. Ecotoxicology and Environmental Safety, 232, 113287.

Gonzatto, M. P., Böettcher, G. N., Schneider, L. A., Lopes, Â. A., Silveira Júnior, J. C., Petry, H. B., Oliveira, R. P., & Schwarz, S. F. (2016). 3,5,6-trichloro-2-pyridinyloxyacetic acid as effective thinning agent for fruit of 'Montenegrina' mandarin. Ciência Rural, 46(12), 2078–2083.

Hąc-Wydro, K., & Flasiński, M. (2015). The studies on the toxicity mechanism of environmentally hazardous natural (IAA) and synthetic (NAA) auxin – the experiments on model *Arabidopsis thaliana* and rat liver plasma membranes. Colloids and Surfaces B: Biointerfaces, 130, 53–60.

Ismail, H. T. H. (2022). Assessment toxic effects of exposure to 3-indoleacetic acid via hemato-biochemical, hormonal, and histopathological screening in rats. Environmental Science and Pollution Research, 29(60), 90703–90718.

Khalid, S., Malik, A. U., Khan, A. S., Razzaq, K., & Naseer, M. (2016). Plant growth regulators application time influences fruit quality and storage potential of young kinnow mandarin trees. International Journal of Agriculture and Biology, 18, 623–629.

Kim, K. G., Park, D. W., Kang, G. R., Kim, T. S., Yang, Y., Moon, S. J., Choi, E. A., Ha, D. R., Kim, E. S., & Cho, B. S. (2016). Simultaneous determination of plant growth regulator and pesticides in bean sprouts by liquid chromatography – tandem mass spectrometry. Food Chemistry, 208, 239–244.

Laisk, A., & Oja, V. (1998). Dynamics of leaf photosynthesis: Rapid response measurements and their interpretations. CSIRO Publishing, Collingwood.

AOAC International, Horwitz, W., & Latimer, G. W. (2010). Official methods of Analysis of Association of Analytical Chemist International. Association of Official Analytical Chemists, Gaithersburg.

Li, J., Guan, Y., Yuan, L., Hou, J., Wang, C., Liu, F., Yang, Y., Lu, Z., Chen, G., & Zhu, S. (2019). Effects of exogenous IAA in regulating photosynthetic capacity, carbohydrate metabolism and yield of *Zizania latifolia*. Scientia Horticulturae, 253, 276–285.

Li, Y., Zhang, D., Xing, L., Zhang, S., Zhao, C., & Han, M. (2016). Effect of exogenous 6-benzylaminopurine (6-BA) on branch type, floral induction and initiation, and related gene expression in Fuji apple (*Malus domestica*). Plant Growth Regulation, 79(1), 65–70.

Luo, Y., Yang, D., Yin, Y., Cui, Z., Li, Y., Chen, J., Zheng, M., Wang, Y., Pang, D., Li, Y., & Wang, Z. (2016). Effects of exogenous 6-BA and nitrogen fertilizers

with varied rates on function and fluorescence characteristics of wheat leaves post anthesis. Scientia Agriculturalura Sinica, 49(6), 1060–1083.
Madzikane-Mlungwana, O., Moyo, M., Aremu, A. O., Plíhalova, L., Doleal, K., Staden, J. V., & Finnie, J. F. (2017). Differential responses to isoprenoid, N6-substituted aromatic cytokinins and indole-3-butyric acid in direct plant regeneration of *Eriocephalus africanus*. Plant Growth Regulation, 82(1), 103–110.
Mandal, S., Poi, R., Banerjee, K., Ansary, I., Bhattacharyya, S., Hazra, D. K., Ghosh, R., & Karmakar, R. (2021). Bioefficacy, residue dynamics and dietary risk assessment of gibberellic acid in improving the potential yield of tomato (*Solanum lycopersicum* L.). Environmental Monitoring and Assessment, 193, 652.
Mesejo, C., Rosito, S., Reig, C., Martínez-Fuentes, A., & Agustí, M. (2012). Synthetic auxin 3,5,6-TPA provokes *Citrus clementina* (Hort. ex Tan) fruitlet abscission by reducing photosynthate availability. Journal of Plant Growth Regulation, 31, 186–194.
Mohorović, P., Vaughan-Hirsch, J., Ceusters, J., & Van de Poel, B. (2023). The role of ethylene in photosynthate partitioning and source-sink modulation in crops. In: Khan, N. A., Ferrante, A., & Munné-Bosch, S. (Eds.). The plant hormone ethylene. Academic Press. Pp. 23–39.
Morgun, V., Pryadkina, G., Stasik, O., & Zborivska, O. (2019). Relationships between canopy assimilation surface capacity traits and grain productivity of winter wheat genotypes under drought stress. Agricultural Science and Practice, 6(2), 18–28.
Ozok, N., & Celik, I. (2012). Effects of subacute and subchronic treatment of synthetic plant growth regulators on liver damage serum biomarkers tissue antioxidant defense systems and lipid peroxidation in rats. Journal of Drug Metabolism and Toxicology, 3(4), 124.
Rai, R. K., Tripathi, N., Gautam, D., & Singh, P. (2017). Exogenous application of ethrel and gibberellic acid stimulates physiological growth of late planted sugarcane with short growth period in sub-tropical India. Journal of Plant Growth Regulation, 36(2), 472–486.
Ren, B., Zhang, J., Dong, S., Liu, P., & Zhao, B. (2017). Regulations of 6-benzyladenine (6-BA) on leaf ultrastructure and photosynthetic characteristics of waterlogged summer maize. Journal of Plant Growth Regulation, 36(3), 743–754.
Rogach, V. V., Kuryata, V. G., Kosakivska, I. V., Voitenko, L. V., Shcherbatiuk, M. M., & Rogach, T. I. (2021). Morphogenesis, pigment content, phytohormones and productivity of sweet pepper under the action of gibberellin and tebuconazole. Regulatory Mechanisms in Biosystems, 12(2), 294–300.
Rogach, V. V., Kuryata, V. G., Kosakivska, I. V., Voitenko, L. V., Shcherbatyuk, M. M., & Rogach, T. I. (2022). Morphogenesis, pigment content, phytohormones and yield of tomatoes under the action of gibberellin and tebuconazole. Biosystems Diversity, 30(2), 150–156.
Rogach, V. V., Voitenko, L. V., Shcherbatiuk, M. M., Kosakivska, I. V., & Rogach, T. I. (2020). Morphogenesis, pigment content, phytohormones and productivity of eggplants under the action of gibberellin and tebuconazole. Regulatory Mechanisms in Biosystems, 11(1), 129–135.
Rohach, V. V. (2017). Vpliv stimuljatoriv rostu na fotosintetichnij aparat, morfogenez i produkcijnij proces baklazhana (*Solanum melongena*) [Influence of growth stimulants on photosynthetic apparatus, morphogenesis and production process of eggplant (*Solanum melongena*)]. Biosystems Diversity, 25(4), 297–303 (in Ukrainian).
Rohach, V. V., Kuryata, V. G., Kiriziy, D. A., Sytnyk, S. K., Grabyk, I. H., Kaitanyuk, O. V., Tarasyuk, M. V., & Rohach, T. I. (2023). Effect of antigibberellins on morphogenesis, photosynthetic apparatus, productivity and their residual content in tomato fruits. Biosystems Diversity, 31(2), 191–201.
Singh, S. K., Nath, V., Marboh, E. S., & Sharma, S. (2017). Source-sink relationship in litchi verses mango: A concept. International Journal of Current Microbiology and Applied Sciences, 6(3), 500–509.
Singh, S., & Prasad, S. M. (2014). Growth, photosynthesis and oxidative responses of *Solanum melongena* L. seedlings to cadmium stress: Mechanism of toxicity amelioration by kinetin. Scientia Horticulturae, 176, 1–10.
Starck, Z., Stahl, E., & Witek-Czupryńiska, B. (1987). Responsiveness of tomato plants to growth regulators depends on light and temperature conditions. Journal of plant physiology, 128, 121–131.
Stasik, O. O., & Kirizii, D. A. (2011). Rehuliatomi zviazky i limituvalni chynnyky v systemi fotosyntez – produktsiinyi protses ta perspektyvy yikh optymizatsii. [Regulatory mechanisms and limiting factors in the photosynthesis – productivity relationships and prospects for their optimization]. Fiziologija i Biohimija Kul'turnyh Rastenij, 43(3), 226–238 (in Ukrainian).
Topalca, N., Yegin, E., & Celik, I. (2009). Influence of indole-3-butyric acid on antioxidant defense systems in various tissues of rats at subacute and subchronic exposure. Food and Chemical Toxicology, 47(10), 2441–2444.
Tsygankova, V. A., Voloshchuk, I. V., Kopich, V. M., Pilyo, S. G., Klyuchko, S. V., & Brovarets, V. S. (2023). Studying the effect of plant growth regulators ivin, methyur and kamethur on growth and productivity of sunflower. Journal of Advances in Agriculture, 14, 17–24.
Tubic, L., Savic, J., Mitic, N., Milojevic, J., Janosevi, D., Budimir, S., & Zdravkovic-Korac, S. (2016). Cytokinins differentially affect regeneration, plant growth and antioxidative enzymes activity in chive (*Allium schoenoprasum*). Plant Cell, Tissue and Organ Culture January, 124(1), 1–14.
Ugare, B., Banerjee, K., Ramteke, S. D., Pradhan, S., Oulkar, D. P., Utture, S. C., & Adsule, P. G. (2013). Dissipation kinetics of forchlorfenuron, 6-benzyl aminopurine, gibberellic acid and ethephon residues in table grapes (*Vitis vinifera*). Food chemistry, 141(4), 4208–4214.
Van Emden, H. F. (2008). Statistics for terrified biologists. Blackwell, Oxford.
Wang, F., Zhang, W., Chen, Y., Xu, G., Deng, Z., Du, H., Wang, Y., Zhang, S., & Zhao, W. (2015). Simultaneous determination of six plant growth regulators in fruits using high performance liquid chromatography based on solid-phase extraction and cleanup with a novel mixed-mode functionalized calixarene sorbent. Analytical Methods, 7(15), 6365–6371.
Wen, Y., Su, S. C., Ma, L. Y., & Wang, X. N. (2018). Effects of gibberellic acid on photosynthesis and endogenous hormones of *Camellia oleifera* Abel. in 1st and 6th leaves. Journal of Forest Research, 23(5), 309–317.
Xiao-Tao, D., Yu-Ping, J., Hong, W., Hai-Jun, J., Hong-Mei, Z., Chun-Hong, C., & Ji-Zhu, Y. (2013). Effects of cytokinin on photosynthetic gas exchange, chlorophyll fluorescence parameters, antioxidative system and carbohydrate accumulation in cucumber (*Cucumis sativus* L.) under low light. Acta Physiologiae Plantarum, 35(5), 1427–1438.
Xing, X., Jiang, H., Zhou, Q., Xing, H., Jiang, H., & Wang, S. (2016). Improved drought tolerance by early IAA- and ABA-dependent H_2O_2 accumulation induced by α-naphthaleneacetic acid in soybean plants. Plant Growth Regulation, 80(3), 303–314.
Yang, D., Dong, W., Luo, Y., Song, W., Cai, T., Li, Y., Yin, Y., & Wang, Z. (2017). Efects of exogenous 6-BA on photosynthetic characteristics and endogenous hormone content in wheat leaves under two nitrogen application levels at seedling stage. Scientia Agricultura Sinica, 50(20), 3871–3884.
Zhang, P., Wang, L. M., Zheng, D. W., Lin, T. F., Wei, X. D., Liu, X. Y., & Wang, H. Q. (2018). Surface-enhanced Raman spectroscopic analysis of N6-benzylaminopurine residue quantity in sprouts with gold nanoparticles. Journal of Environmental Science and Health, Part B, 53(9), 561–566.
Zhao, H., Cao, H., Ming-Zhen, P., Sun, Y., & Liu, T. (2017). The role of plant growth regulators in a plant aphid parasitoid tritrophic system. Journal of Plant Growth Regulation, 36(4), 868–876.