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Application of Pyrimidine and Pyridine Derivatives for Regulation of Chickpea (*Cicer arietinum* L.) Growth

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Abstract: The effect of new synthetic pyrimidine derivatives on the growth and development of the chickpea (*Cicer arietinum* L.) variety Mexican was studied.The growth-regulating activity of pyrimidine derivatives was compared with the activity of phytohormones auxins, derivatives of 1*H*-indole-3-acetic acid (IAA) and 1-naphthylacetic acid (NAA), as well as plant growth regulators, derivatives of sodium and potassium salts of 6-methyl-2-mercapto-4-hydroxypyrimidine (Methyur and Kamethur), N-oxide-2,6-dimethylpyridine (Ivin).The most biologically active synthetic compounds among pyrimidine derivatives were selected, which showed auxin-like, Ivin-like, Methyur-like and Kamethur-like regulatory effects on the growth and development of the shoot and root of chickpea plants. An analysis of the relationship between biological activity and the chemical structure of synthetic pyrimidine derivatives was conducted.To improve the growth and development of the chickpea (*Cicer arietinum* L.) variety Mexican, the application of selected synthetic pyrimidine derivatives, as well as plant growth regulators Methyur, Kamethur and Ivin is proposed.

Keywords: *Chickpea (Cicer arietinum L.), pyrimidine, pyridine, Methyur, Kamethur, Ivin.*

I. INTRODUCTION

Chickpea (*Cicer arietinum* L.) is one of the world's most important grain legumes grown on an area of more than 12 million hectares with world production of around 13 million tons [1 - 3]. Chickpea grains are enriched with protein (18.4 – 29.0 %), total carbohydrates (64 %, including 47 % starch, 6 % soluble sugar, 5% fat, 6 % crude fibre), ash (3 %), essential nutrients such as phosphorus (340 mg per 100g), calcium (190 mg per 100g), magnesium (140 mg per 100g), iron (7 mg per 100g) and zinc (3 mg per 100g) [1 - 5]. The main components of chickpea proteins are globulins (41.7 %) and albumins (16.1 %), characterized by high solubility and digestibility (89.0 %) [3]. The main components of chickpea lipid fraction (4.5–6.6%) are palmitic (10.8 %), oleic (33.5 %), linoleic (49.7 %), and linolenic (2.4 %) fatty acids, as well as tocopherols (230.3 mg/100 g of oil) and carotenoids (46.3 μg/100 g of flour), which exhibit antioxidant properties [3]. Fermented chickpeas products are a source of nutrients and bioactive compounds such as phenolic compounds, peptides, essential amino acid "Lysine", which is not normally found in dietary grains, and soluble vitamins with antioxidant, antihypertensive, and antidiabetic properties [4]. Chickpea seeds contain also saccharine, glucose, fructose, polysaccharides including starch, γ-galactan, levulose, P-galacto-araban, betaine, choline adenine, saponins, phytin, citric acid and malic acid etc., beneficial for human health, therefore 25% chickpea meal and 75% ground nut meal can be used both for correction in malnourished people, for the treatment of Kwaserkhor and other protein-deficient diseases, and for the treatment of patients with atherosclerosis due to the rich content of phosphorus [2]. Immature pods and fresh sporulated gram are enriched in phospho-enol-pyruvate carboxylase and contain biochanin-A, B and C, as well as vitamins-C (ascorbic acid), which are used to stimulate yeast cell growth and prevent scurvy in starving people.Chickpea seed flour is widely used to feed horses, while the leaves and stems are dried and used as cattle feed [2, 6]. Chickpea seed flour is also used in textiles sizing and adhesives [2].

Global climate change and environmental pollution are the most adverse environmental factors affecting chickpea (*Cicer arietinum* L.) growth and productivity.An urgent task of modern agriculture is the development of new effective and environmentally friendly plant growth regulators to improve the growth, increase the yield and drought tolerance of chickpea (*Cicer arietinum* L.) [2, 5 -10]. Currently, natural plant hormones, synthetic plant growth regulators and mineral fertilizers are used to increase chickpea growth and yield, and to improve the chickpea adaptation to global climate change, environmental pollution, and phytopathogens [2,5 - 10].

Today an important task of agriculture is the search for new growth regulators of chickpea plants. In this direction, the new plant growth regulators based on low molecular weight heterocyclic compounds, similar in activity to phytohormones auxins and cytokinins were developed at the V.P. Kukhar Institute of Bioorganic Chemistry and Petrochemistry, National Academy of Sciences of Ukraine [11, 12]. Among such compounds, the most promising are synthetic compounds, derivatives of pyridine and pyrimidine, which are widely used in agriculture as plant growth regulators and herbicides [13 - 19].

The best-known new plant growth regulators based on pyrimidine and pyridine derivatives are Methyur (sodium salt of 6-methyl-2-mercapto-4-hydroxypyrimidine), Kamethur (potassium salt of 6-methyl-2-mercapto-4-hydroxypyrimidine) and Ivin (N-oxide-2,6-dimethylpyridine) [20]. Our earlier studies of plant growth regulators Methyur, Kametur and Ivin showed that these chemical compounds significantly improve the growth and development of important agricultural, industrial, horticultural and ornamental crops (corn, barley, oats, wheat, sorghum, miscanthus, cherry and rose) in laboratory, field and under *in vitro* culture of isolated plant cells and tissues[11, 20 - 30]. In addition, the use of these plant growth regulators contributed to increased yield and improved adaptation of plants to stressors of abiotic origin [24, 25, 27, 28].

The molecular mechanisms of the growth regulatory activity of Methyur, Kamethur and Ivin are of considerable interest.We found that the regulatory effects of plant growth regulators Methyur, Kamethur and Ivin is associated with their stimulating effect on the processes of elongation, division and differentiation of the plant cell, which are the main processes of formation and development of shoot and root meristems in plants grown in laboratory, field and under *in vitro* culture of isolated plant cells and tissues [20 - 30].

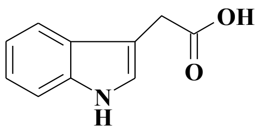
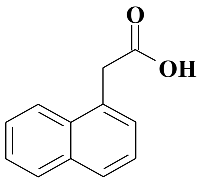
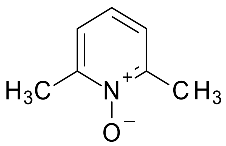
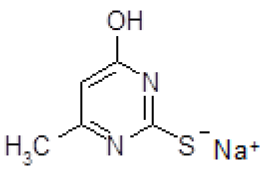
Studies of the molecular mechanisms of action of Methyur and Ivin show that these plant growth regulators have a stimulatory effect on changes in gene expression at the level of transcription and translation of genetic information, due to which the time of plant ontogenesis is almost halved [31]. In addition, synthetic plant growth regulators Methyur and Ivin can influence plant growth indirectly, through the endogenous pool of phytohormones in plant cells [32].

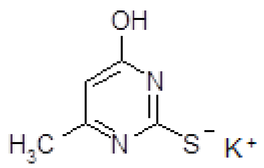
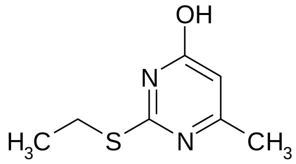
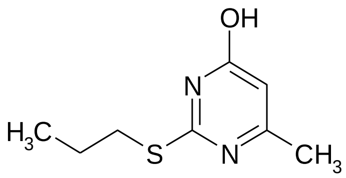
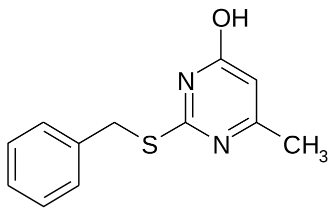
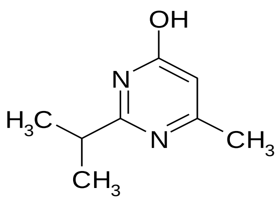
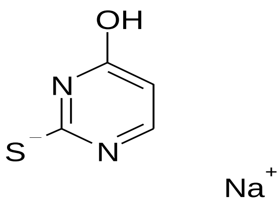
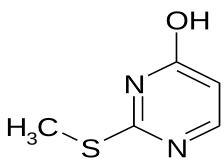
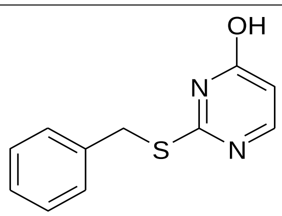
The aim of the work is to study the effect of new synthetic pyrimidine derivatives in comparison with the effect of phytohormones auxins IAA and NAA, as well as plant growth regulators Methyur, Kamethur and Ivin on the vegetative growth of the chickpea (*Cicer arietinum* L.) variety Mexican.

II. MATERIALS AND METHODS

The chemical structure of phytohormones auxins IAA and NAA produced by Sigma-Aldrich, USA, plant growth regulators Methyur, Kamethur and Ivin, and new chemical compounds, pyrimidine derivatives (compounds № 1 – 7), synthesized in the Department for Chemistry of Bioactive Nitrogen-Containing Heterocyclic Compounds, V.P. Kukhar Institute of Bioorganic Chemistry and Petrochemistry of the National Academy of Sciences of Ukraine, are listed in Table 1.

Table1: Chemical structure and relative molecular weight of auxins IAA and NAA, plant growth regulators Methyur, Kamethur and Ivin, and pyrimidine derivatives

Chemical compound №	Chemical structure	Chemical name and relative molecular weight (g/mol)
IAA		1*H*-Indol-3-ylacetic acid MW=175.19
NAA		1-naphthylacetic acid MW=186.21
Ivin		N-oxide-2,6-dimethylpyridine MW=125.17
Methyur		Sodium salt of 6-methyl-2-mercapto-4-hydroxypyrimidine MW=165.17

Kamethur		Potassium salt of 6-methyl-2-mercapto-4-hydroxypyrimidine MW=181.28
1		2-ethylsulfanyl-6-methylpyrimidin-4-ol MW=170.23
2		6-methyl-2-propylsulfanyl-pyrimidin-4-ol MW=184.26
3		2-benzylsulfanyl-6-methylpyrimidin-4-ol MW=232.31
4		2-isopropyl-6-methyl-pyrimidin-4-ol MW=152.20
5		Sodium salt of 4-hydroxypyrimidine-2-thiolate MW=149.14
6		2-methylsulfanylpyrimidin-4-ol MW=142.18
7		2-benzylsulfanylpyrimidin-4-ol MW=218.28

III. PLANT GROWING CONDITIONS

In laboratory conditions, the effect of phytohormones auxins IAA and NAA, plant growth regulators Methyur, Kamethur and Ivin, and new synthetic pyrimidine derivatives (compounds № 1 – 7) on the growth and development of the chickpea (*Cicer arietinum* L.) variety Mexican was studied.For this, chickpea seeds were sterilized with 1 % KMnO4 solution for 3 min, then treated with 96 % ethanol solution for 1 min, and then washed three times with sterile distilled water. After this procedure, chickpea seeds were placed in cuvettes (10 seeds each) in perlite moistened with distilled water (control), or water

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solutions of auxins IAA and NAA, or plant growth regulators Methyur, Kamethur and Ivin,or new synthetic pyrimidine derivatives (compounds № 1 – 7), which were used at a concentration of 10^{-7}M. Then the chickpea seeds were placed in a thermostat for germination in the dark at a temperature of 22°C for 48 hours, then the germinated chickpea seeds were placed in a climate chamber, where chickpea plants were grown for 3 weeks in a 16/8 light/dark mode, at a temperature of 20-22 °C, illumination of 3000 lux, and an air humidity of 60-80 %.

Comparative analysis of morphometric parameters of 3-week-old chickpea plants (average length of shoots and roots (mm), average number of roots (pcs)) was carried out according to the methodology [33].

IV. STATISTICAL ANALYSIS

Statistical processing of the experimental data, performed in triplicate, was carried out according to the Student's t-test with a significance level of P≤0.05; mean values ± standard deviation (± SD) [34].

V. RESULTS AND DISCUSSION

The study of the growth-regulating activity of new synthetic pyrimidine derivatives (compounds № 1 – 7, listed in Table 1) showed that they exhibit an effect similar to the auxins IAA and NAA, and plant growth regulators Methyur, Kamethur and Ivin, when used to treat chickpea seeds in a concentration of 10^{-7}M. Improvement of growth and development of roots and shoots of chickpea plants was observed within 3 weeks (Figure 1).

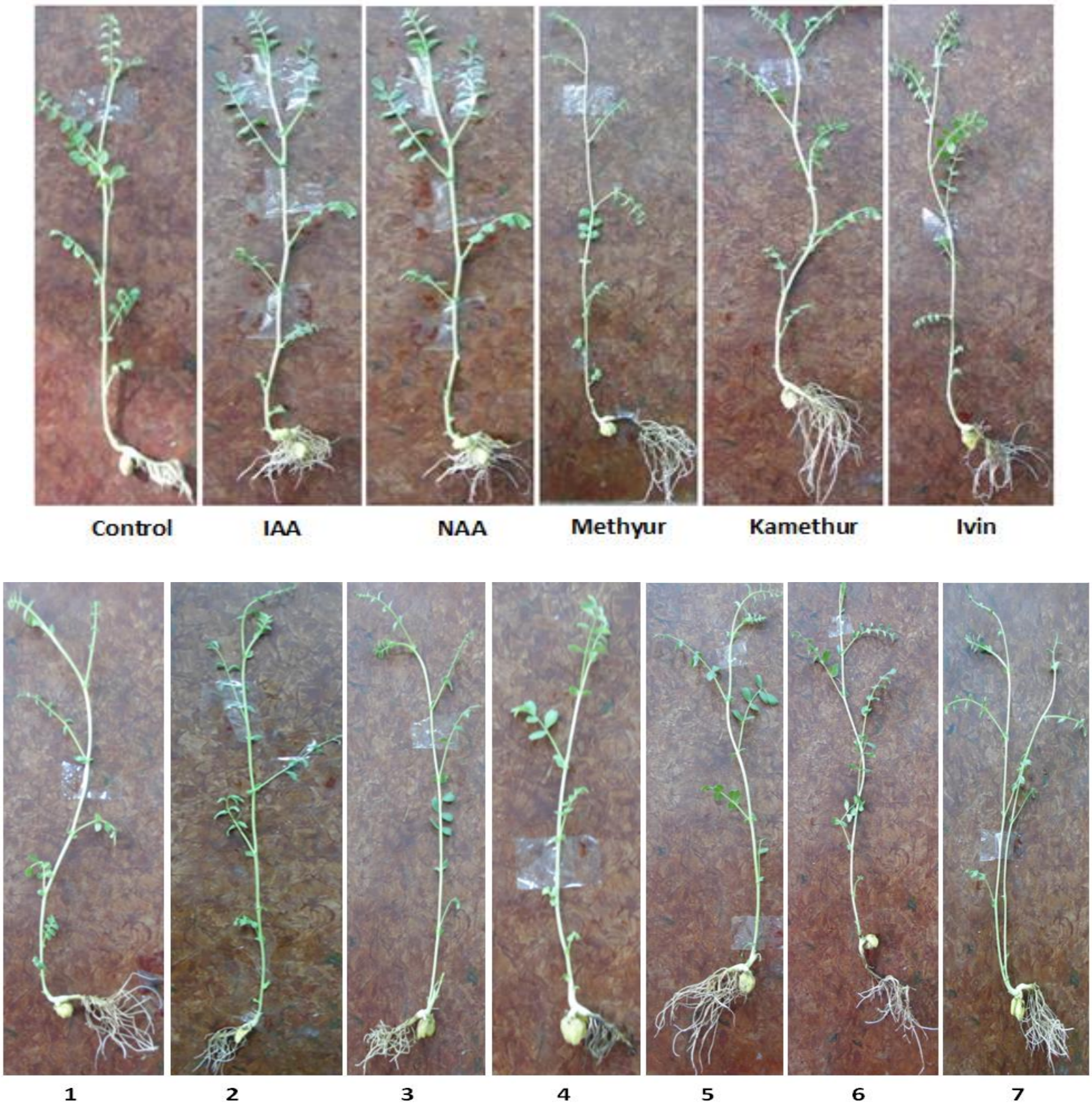

Fig. 1: The effect of auxins IAA and NAA, plant growth regulators Methyur, Kamethur and Ivin, as well as new synthetic pyrimidine derivatives (compounds № 1 – 7) at a concentration of 10^{-7}M on the growth and development of shoots and roots of chickpea (*Cicer arietinum* L.) variety Mexican for 3 weeks

Statistical analysis showed that synthetic pyrimidine derivatives,the compounds № 1, 2, 5, 6, and 7 show the highest activity. The morphometric parameters of 3-week-old chickpea plants grown on water solution of synthetic

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pyrimidine derivatives, the compounds № 1, 2, 5, 6 and 7, used at a concentration of 10^{-7}M, were equal to or exceeded similar parameters of chickpea plants grown on water solution with plant growth regulators Methyur, Kamethur and Ivin, as well as auxins IAA and NAA, used in a similar concentration of 10^{-7}M (Figures 2, 3 and 4)).

The parameters of the average length of the shoots (mm) of chickpea plants grown on water solution of auxins IAA and NAA increased by 55.26 % and 44.74 %, respectively, compared with similar parameters of control chickpea plants grown on distilled water (Figure 2).

The parameters of the average length of the shoots (mm) of chickpea plants grown on water solution of plant growth regulators Methyur, Kamethur and Ivin increased by 83.33 %, 79.82 %, and 89.47 %, respectively, compared with similar parameters of control chickpea plants grown on distilled water (Figure 2).

The parameters of the average length of the shoots (mm) of chickpea plants grown on water solution of compounds № 1, 2, 5, 6 and 7 increased by 48.68 % – 93.68 %, respectively, compared with similar parameters of control chickpea plants grown on distilled water (Figure 2).

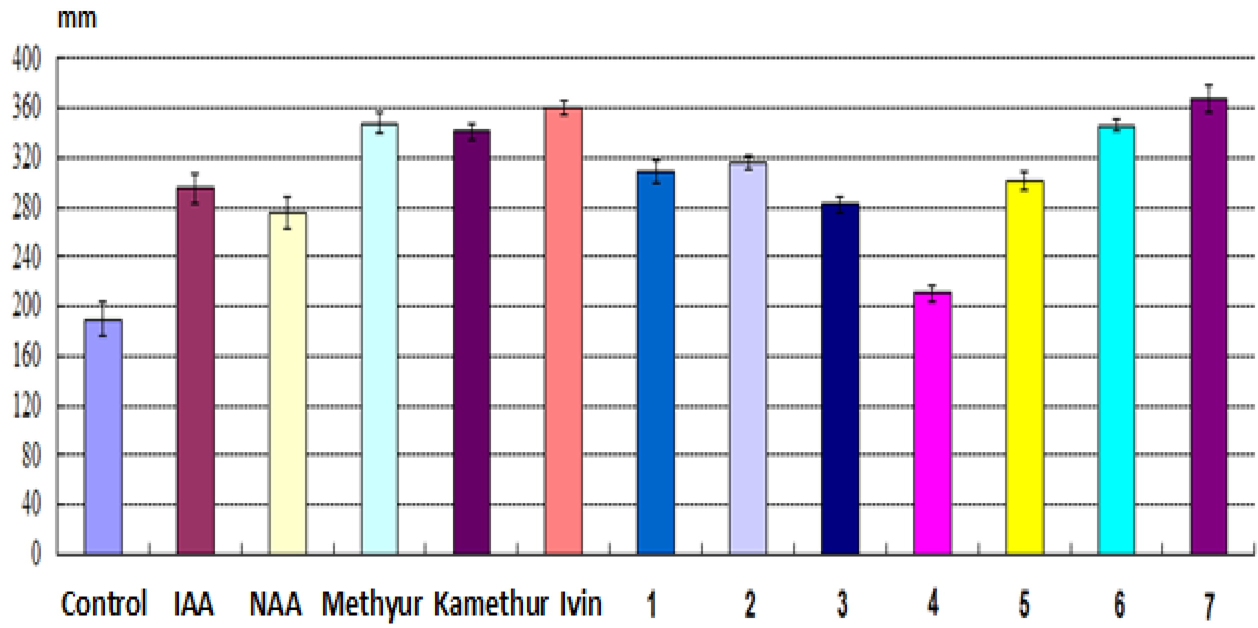

Fig. 2: The effect of auxins IAA and NAA, plant growth regulators Methyur, Kamethur and Ivin,and new synthetic pyrimidine derivatives (compounds № 1 – 7) at a concentration of 10^{-7}M on the average length of shoots (mm) of 3-week-old chickpea (*Cicer arietinum* L.) variety Mexican

Synthetic compound № 4 showed less activity, parameters of the average length of the shoots (mm) of chickpea plants grown on water solution of compound № 4 increased by 11.05 %, compared with similar parameters of control chickpea plants grown on distilled water (Figure 2).

The parameters of the average length of the roots (mm) of chickpea plants grown on water solution of auxin IAA increased by 20 %, compared with similar parameters of control chickpea plants grown on distilled water (Figure 3).

The parameters of the average length of the roots (mm) of chickpea plants grown on water solution of plant growth regulators Methyur, Kamethur and Ivin increased by 30%, 23.33%, and 100 %, respectively, compared with similar parameters of control chickpea plants grown on distilled water (Figure 3).

The parameters of the average length of the roots (mm) of chickpea plants grown on water solution of compounds № 1, 2, 5, 6 and 7 increased by 40 % – 63.33 %, respectively, compared with similar parameters of control chickpea plants grown on distilled water (Figure 3).

Auxin NAA and synthetic compounds № 3 and 4 showed less activity, parameters of the average length of the roots (mm) of chickpea plants grown on water solution of compounds № 3 and 4 were lower than similar parameters of control chickpea plants grown on distilled water (Figure 3).

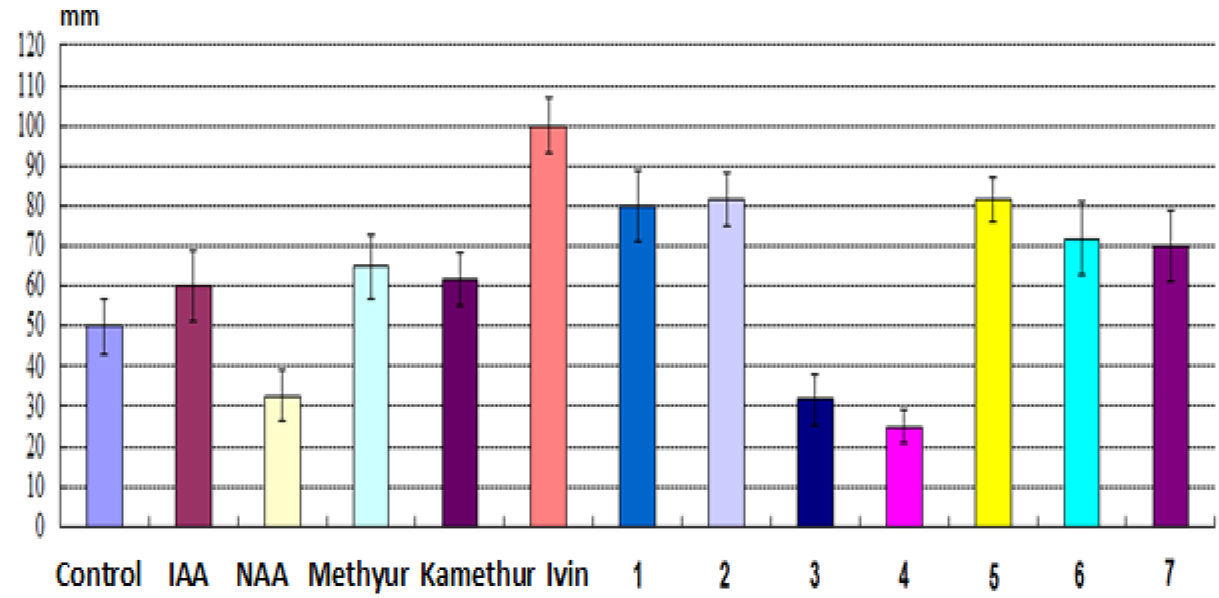

Fig. 3: The effect of auxins IAA and NAA, plant growth regulators Methyur, Kamethur and Ivin, and new synthetic pyrimidine derivatives (compounds № 1 – 7) at a concentration of 10^{-7}M on the average length of roots (mm) of 3-week-old chickpea (*Cicer arietinum* L.) variety Mexican

The parameters of the average number of roots (pcs) of chickpea plants grown on water solution of auxins IAAand NAA increased by 50 % and 35.71 %, compared with similar parameters of control chickpea plants grown on distilled water (Figure 4).

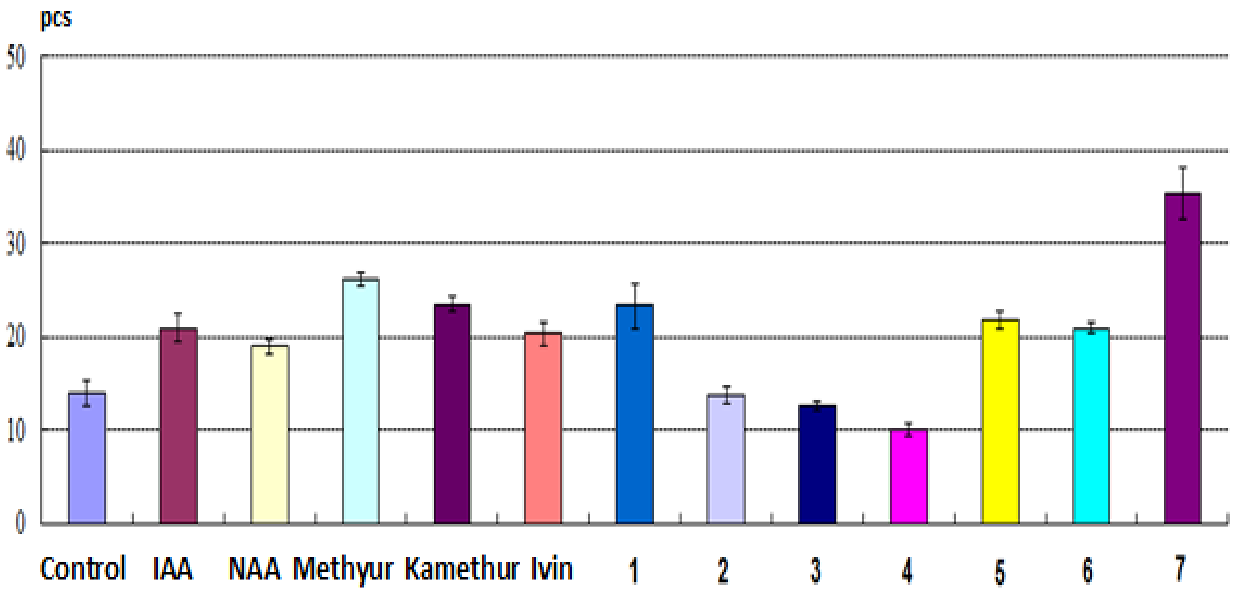

Fig. 4: The effect of auxins IAA and NAA, plant growth regulators Methyur, Kamethur and Ivin, and new synthetic pyrimidine derivatives (compounds № 1 – 7) at a concentration of 10^{-7}M on the average number of roots (pcs) of 3-week-old chickpea (*Cicer arietinum* L.) variety Mexican

The parameters of the average number of roots (pcs) of chickpea plants grown on water solution of plant growth regulators Methyur, Kamethur and Ivin increased by 86.74 %, 67.86 %, and 45.24 %, respectively, compared with similar parameters of control chickpea plants grown on distilled water (Figure 4).

The parameters of the average number of roots (pcs) of chickpea plants grown on water solution of compounds № 1, 5, 6 and 7 increased by 50 % – 152.86%, respectively, compared with similar parameters of control chickpea plants grown on distilled water (Figure 4).

Synthetic compounds № 2, 3 and 4 showed less activity, parameters of the average length of the roots (mm) of chickpea plants grown on water solution of compounds № 2, 3 and 4 did not statistically significantly differ or were lower than similar parameters of control chickpea plants grown on distilled water (Figure 4).

Analyzing the relationship between the chemical structure and biological activity of new synthetic pyrimidine derivatives, compounds № 1, 2, 5, 6 and 7, it can be assumed that their plant growth-regulating activity is related to the presence of substituents in their chemical structure: compound № 1 contains an ethylthio group in position 2, a hydroxyl group in position 4 and a methyl group in position 6; compound № 2 contains a propylthio group in position 2, a hydroxyl group in position 4 and a methyl group in position 6; compound № 5 is the sodium salt of 4-hydroxypyrimidine-2-thiolate; compound № 6 contains a methylthio group in position 2 and a hydroxyl group in position 4; compound № 7 contains a benzylthio group in position 2 and a hydroxyl group in position 4 (Table 1).

At the same time, the decrease in plant growth-regulating activity of synthetic pyrimidine derivatives, compounds, №3 and 4, can be explained by the presence of substituents in the chemical structures of these compounds: compound № 3 contains a benzylthio group in position 2, a hydroxyl group in position 4 and a methyl group in position 6;compound № 4 contains an isopropyl substituent in position 2, a hydroxyl group in position 4, and a methyl group in position 6 (Table 1).

Summarizing the obtained data, it should be noted that high growth-regulating activity of the synthetic compounds № 1, 2, 5, 6 and 7 is associated with their auxin-like, Methyur-like, Kamethur-like and Ivin-like effects on the proliferation, elongation and differentiation of plant cells, which leads to improved growth and development of shoots and roots of chickpea (*Cicer arietinum* L.) varietyMexican [20 – 27; 29, 30, 35 – 39].

It should also be noted thatplant growth regulator Kamethurcontains the macronutrients potassium (K) and sulfur (S),while plant growth regulator Methyur and synthetic compounds № 1, 2, 5, 6 and 7 contain the macronutrient sulfur (S), which are necessary for the growth and reproduction of chickpea plants and take an important part in the regulation of metabolic processes in chickpea plant cells [2].

In addition, it was previously shown that the plant growth regulator Methyur containing the chemical element sodium (Na), plays an important role in plant adaptation to salt and osmotic stresses, which will be of great practical importance when growing chickpea plants under salt and water stress conditions [28; 40, 41].

The proposed molecular mechanisms of action of new plant growth regulators based on synthetic low molecular weight heterocyclic compounds, directly, through the network of signaling pathways of plant hormones, or indirectly, that is, affecting the biosynthesis, metabolism and signaling of endogenous auxins and cytokinins in plant cells,were discussed in our previously published works [12, 18, 19, 26, 30, 31 - 32] and the works of other authors [42 - 50].

VI. CONCLUSION

The study of the plant growth-regulating activity of synthetic pyrimidine derivatives (listed in Table 1) showed that their use at a concentration of 10^{-7}M for the treatment of chickpea seeds improves the growth and development of chickpea (*Cicer arietinum* L.) variety Mexican. The growth-regulating activity of these compounds was similar to or exceeded that of auxins IAA and NAA, as well as plant growth regulators Methyur, Kamethur and Ivin, used at a similar concentration of 10^{-7}M. The practical use of the selected most active synthetic compounds № 1, 2, 5, 6 and 7, as well as plant growth regulators Methyur, Kamethur and Ivin to improve the development of shoots and roots of chickpea (*Cicer arietinum* L.) variety Mexican is proposed.

STATEMENT OF CONFLICT OF INTEREST

The authors are declared that they have no conflict with this research article.

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