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# INFLUENCE OF NEW PHYSIOLOGICALLY ACTIVE SUBSTANCES OF NATURAL ORIGIN ON NITROGEN METHABOLISM OF WINTER WHEAT

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Вивчено фізіологічний вплив N-оксиду піридину Триман та фізіологічно активних речовин Емістим та Агростимулін на азотний метаболізм пшениці озимої сорту Поліська 90. Препарати сприяли підвищенню загального вмісту азоту і нітратредуктазної активності в надземних органах пшениці озимої та збільшенню вмісту білка в зернівці і, в цілому, загальній продуктивності рослин. Найефективнішим препаратом природного походження виявився Емістим. Обґрунтовано, що застосування цих препаратів сприяє покращенню фізіологічного статусу пшениці озимої, особливо за несприятливих умов навколишнього природного середовища, та є перспективним засобом екологізації сільськогосподарського виробництва.

*Ключові слова:* регулятор росту рослин, пшениця озима, азотистий метаболізм, урожайність.

Plant growth regulators (GR) play an important role in enhancement the cereals productivity. Application of such regulators permits to regulate the most important processes in plants. An important result of application the growth regulators is the increase of plant resistance of to unfavorable environmental conditions, such as excessively low or high temperatures, drought, phytotoxic action of pesticides, plague of infections and pests.

Institute of Bioorganic Chemistry and Petrol chemistry of Ukrainian National Academy of Sciences has conducted complex research of new plant growth regulators for the

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last years. In-depth research of physics-chemical properties of growth regulators based on N-oxides of pyridine derivatives proved that these regulators have a number of unique qualities, which explain their high ability to regulate growth processes. Such regulators include Ivin (N-oxide 2.6-dimethylpyridine), Triman (aqua-N-oxide 2-dimethylpyridine Mn), and their derivatives [1]. These regulators are highly sensitive to slow mechanic vibrations, vibrations of electric and magnetic fields and infra sound. Some of these properties are inherent to electronic or ion semiconductors, which allow them to transmit electronic and magnetic impulses and other signals. Study of test plants showed that Ivin has cytokinin and auxin activity.

In this study we used Triman (a synthesized substance derived from N-oxides of pyridine), Emistim and Agrostimulin. Emistim is a product of metabolism of the roots' epiphytes of *Panax ginseng* and *Hippophao rhamnoides.* It contains some amino acids. growth regulators and lipids. Emistim is a GR of natural origin. Agrostimulin is a mix of Ivin (a synthesized substance) and Emistim (a natural one). However the ratio of Ivin to Emistim is very low, which ensures ecological safety of this GR. The aim of our work was to study the physiological effect of Agrostimulin, Emistim and Triman on some stages of nitrogen metabolism and productivity of winter wheat in the conditions of Kyiv region; and to study the joint use of these growth regulators and fungicides during the pre-seedtime treatment of winter wheat seeds.

## MATERIALS AND METHODS

*Plants*: For our tests we used plants of winter wheat (*Triticum aestivim* L.) of sort «Polis'ka 90». The experiments were carried out in our test plots (managed by the Institute of Agriculture of the Ukrainian Academy of Agricultural Sciences, Chabany) in several stages.

*Experiment* –*Effect of the growth regulators in field conditions*: Each test plot had an area of 10 m2 with the light gray clay soil. The norm of sowing was 5 million grains per hectare. Each plot was fertilized with ammonium saltpeter  $(N_{120}P_{90}K_{90})$ . The growth regulators were introduced as solutions by spraving onto the plots in the following concentrations: Agrostimulin – 5 ml/ha, Emistim – 5 ml/ha and Triman 50 ml/ha at IV-V stage of organogenesis. Nitrate reductase activity in wheat leaves was determined *in vivo* by the method of Mulder. We used the Kyedal's method for determining the total content of nitrogen. However, we used spectrophotometric with the use of Nesler reagent instead of distillation. The quantity of nitrates in the plant leaves was determined by using the potentiometer method. The grain quality was evaluated with an «Infrapid» device (USA), which allows to measure the protein content in grain precisely.

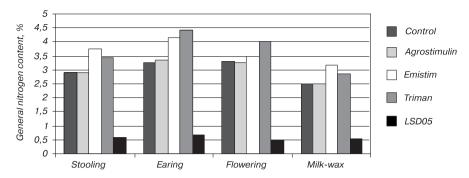
*Statistical Analysis*: Data were analyzed using repeated measures analysis of variance. The statistical processing of our data was carried out using the dispersion analysis. All evaluations were made using an alpha level of 0.05.

## **RESULTS AND DISCUSSION**

In the first part of our work we studied the physiological influence of these growth regulators on the nitrate-reducing ability of leaves, the nitrate content, the total nitrogen content and on grain productivity and quality.

The analysis of the dynamics of total nitrogen accumulation in the flag leaf during the 1-st year of vegetation for example, (Fig. 1) shows that the regulators promote the nitrogen content increase. Action of Emistim and Triman is the strongest during the earing stage, which is crucial for the formation of wheat productive process.

Since the regulators show cytokinin activity, they can induce the synthesis of nitrate reductase [2] regardless of the quantity of substrate and promote launching of the synthesis of various enzymes involved in biosynthesis of RNA and proteins [3, 4]. Wheat is known to belong to an intermediate group of plants in which the nitrate reductase activity is distributed between roots and leaves [5–7]. According to our data, Agrostimulin, Emistim and Triman stimulated the increase of level of induced nitrate reductase activity



**Fig. 1.** Influence of physiologically active substances on total nitrate content in flag leaf of winter wheat Polis'ka 90,% (1-st year of vegetation)

in the flag leaf and particularly in its sheath. Sheath, that is the leaf basis, which envelops the intercalary meristem zone, appeared to be the place of the most intensive nitrate reductase activity.

The most significant impact of Agrostimulin, Emistim and Triman on the nitrate reductase activity was observed in the 2-nd year of vegetation (Fig. 2), which was characterized by high 24-hours temperature during the stage of earing (24-hours temperature varied from 18 to 25°C, the maximum reached 36°C) and moisture deficiency.

As already mentioned, the highest nitrate reductase activity was detected in sheath. The most effective among the regulators was Agrostimulin during the stage of earing (the enzyme activity increased by 132.5%). The nitrate reductase activity in sheath of the Emistim-treated plants at the stage of earing rose by 123.6% compared to the control plants.

We also studied the nitrate content in the test plants. According to our data (Fig. 3), under growth regulators' action, the nitrate content in the flag leaf grew, especially at the stage of stooling (under Agrostimulin action it grew by 79.2% and under the action of Emistim it grew by around 400%). This may be due to somewhat excessively dense sowing of wheat, which may have caused insufficient illumination of photosynthesizing organs, which, in turn, may lead to accumulation of free unassimilated NO<sub>3</sub><sup>-</sup> ions

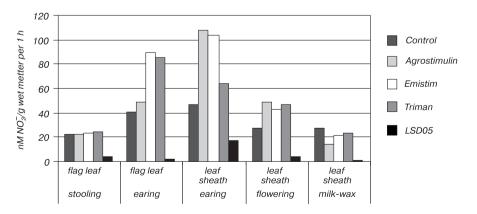


Fig. 2. Influence of physiologically active substances on nitrate reductase activity in flag leaf and sheath of winter wheat Polis'ka 90 (nM  $NO_2^{-}/g$  of wet matter per 1 hour), 2-nd year of vegetation

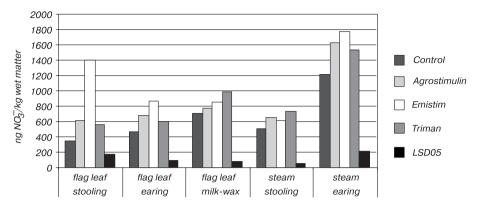


Fig. 3. Influence of physiologically active substances on nitrate content in flag leaf of winter wheat Polis'ka 90 (mg  $NO_3^-/kg$  of wet matter), 1-st year of vegetation

in leaves [8]. The level of nitrates especially increased in the stems at the earing stage: 1631.4 mg of  $NO_3^-$  per kg of wet matter for Agrostimulin, 1773.83 mg of  $NO_3^-$  per kg of wet matter for Emistim and 1215.9 mg of  $NO_3^-$  per kg of wet matter for Triman compared to the control plants. There is a direct correlation between the nitrate content in wheat stem as the stages of stooling and earing and the content of protein. Therefore, these data on the nitrate content can be used for predicting the protein content in grain [9].

In the following Tab. 1 we summarized our data for the crops of all vegetation years. The increase of yields as well as the increase of protein content in grain was recorded every year for the plants treated with any of the three growth regulators.

In 2-nd year of vegetation, which was a year with unfavorable weather conditions because of prolonged draught, the increase in yields was especially significant for the plants treated with Agrostimulin and Emistim, which are the growth regulators of natural origin. In particular, the yield of the plants treated with Agrostimulin exceeded by 36% the yield compared to control plants. For plants treated with Emistim, the figure was 40%. The notable increase in the quality of grain and productivity of the plants treated with Agrostimulin and Emistim may suggest

Table 1

	1-st year vegetation		2-nd year vegetation		3-rd year vegetation	
	Content of proteins, %	Yield, kg per ha	Content of proteins, %	Yield, kg per ha	Content of proteins, %	Yield, kg per ha
Control*	13.43	2980	14.02	3130	10.61	3470
Agrostimulin	14.20	3580	15.03	4270	11.37	3880
Emistim	14.30	3400	14.78	4340	11.83	3970
Triman	14.62	3550	14.50	3780	12.20	4370
LSD <sub>05</sub>	-	370	-	415	-	390

Influence of Agrostimulin, Emistim and Triman on the protein content of grain and productivity of winter wheat (Polis'ka 90)

\* Control plants, as well as the test plants, were grown on the plots fertilized with ammonium saltpeter  $(N_{120}P_{90}K_{90})$ .

that these growth regulators are capable to improve the plants' ability to resist stress.

During the next years we carried out studies on the joint use of these growth regulators and fungicides during the pre-seedtime treatment of the winter wheat seeds. Real possibility to reduce fundamentally (by 25% and more) the fungicide expenditure without fungicide bioprotective effect being declined but with harvest being increased and its quality improved was demonstrated.

No less effective may be use of the growth regulators in parallel with the prosecution of the phyto-sanitary treatments of winter wheat sowings against the deseases. The results of the phyto-sanitary treatment of winter wheat against the leaf deseases show that Emistim C failed to worsen the Tilt (propiconazole) biological effeciency relative to the extent of farinose dew and septoriose expansion: in the variants with fullscale Tilt dose and with it being reduced by 50% the progression of the farinose dew is retarded by 15%, that of septoriosis - by 21%. Both variants with the Emistim C use showed statistically significant harvest increase. Multiannual tests for Emistim C and Agrostimulin in various groundclimatic zones of Ukraine showed that these may improve the wheat endurance not only to diseases and pests lesions, but to the other stress factors as well – drought, low temperatures, soil salinization.

## CONCLUSION

Our results show that treatment with new growth regulators, especially with the growth regulators of natural origin, promotes increase in the grain yield and quality, improves adaptive abilities of wheat by enhancing the activity of nitrate-reducing system, which is proved by increase in the grain protein content, as well as favour the decreased fungicide amounts used (by 25%) during the pre-seedtime treatment of the winter wheat seeds, being effective upon these substances use simultaneously with the prosecution of the wheat phyto-sanitary treatments against the leaf diseases.

The use of ecologically safe growth regulators as a means for biologization of new technologies to increase the grain culture productivity and extend the employment of the new kinds of the biological approaches to plant protection, especially under unfavorable environment conditions is very urgent in modern sustainable agriculture.

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