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# THE INCREASE OF PHYSIOLOGICAL OPTIMUM OF NITROGEN FOR PLANTS BY INOCULATION AND USE OF PLANT GROWTH REGULATORS

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*As was established in greenhouse experiments using isotope dilution method and confirmed by field investigations, conducted on grey-forest and sod-podzol soils, the inoculation and (or) treatment of cereal grasses with growth stimulators enhances plants' physiological optimum in nitrogen and thus improves ecological state of agrocenoses by increasing fertilizers assimilation rate and their involvement into the constructive metabolism. At this, typically high doses of mineral nitrogen combined with plants growth stimulators become ecologically acceptable.*

Key words: *nitrogen, associative nitrogen fixation, physiological optimum, isotope dilution method, cereal grasses.*

As is well known, nitrogen fixation is a rational process which is not occurring at exceed of fixed nitrogen in the soil. However, plants make their own corrections into the functioning of associative nitrogen fixing bacteria. This appears in plants' assimilation of considerable amount of applied nitrogen and its subsequent use in constructive metabolism. At this point, the inhibitory effect of fixed nitrogen on the activity of associative diazotrophs is partially or completely vanished. Moreover, well developed plants can improve bacteria provision with root exudates and thus promote process of associative nitrogen fixation. Basing on the principles mentioned, almost at one time, M. Umarov et al. [1] and Ladha et al. [2] have defined conception of physiologically optimum doses of nitrogen for the plants, as the ones that stimulate nitrogen fixation.

Stimulation mechanism of associative nitrogen fixation activity with physiologically optimum doses might lay in the increase of the amount of root exudates used as the source of carbon and energy for rhizospheric nitrogen fixing bacteria. Thus, Mergel et al. [3] have shown the increase of root exudates under the application of mineral nitrogen in 9.5 times comparing to the ones in control plants (without fertilizers). At this, depending on the amount of applied fertilizers the root exudates will contain more or less amount of nitrogen compounds [4].

In our previous studies we have showed the sharp increase in number of nitrogen fixing bacteria under the application of mineral nitrogen. However, nitrogenase activity of root diazotrophs was first observed in variants with small doses of fertilizer. Nitrogen fertilizers in high concentrations repress synthesis of nitrogenase by microorganisms in spite of their high number. Increase of nitrogen fixing activity in such variants was observed later on after the application of fertilizers – with the decrease of concentration of fixed nitrogen to the optimum level [5]. In view of foresaid, we can consider that **doses of nitrogen fertilizers which promote nitrogen fixation in root zone – are doses, sufficient for constructive plant metabolism on the certain stage of organogenesis and intense excretion of root exudates containing nitrogen compounds in an amount which do not repress nitrogenase synthesis by associative diazotrophs**.

In our experiments we have used grass mixture "perennial ryegrass + awnless brome" as an example of determination of physiologically optimum nitrogen fertilization.

The experiments were conducted during three years on sod-podzol soils ($pH_{salt.}$ – 6,5; humus content – 1,1-1,2 %) on $P_{60}K_{60}$ background and rising doses of ammonium nitrate, applied separately – in early spring and after hay harvest. Nitrogen fixation activity was determined by means of acetylene method.

It was showed that separate application of mineral nitrogen even in small amounts initially represses nitrogen fixation or has no clear effect. Later on, at fertilizers' utilization the increase of nitrogen fixation activity was noticed in the variants with small doses of nitrogen. With time, stimulation of nitrogen fixation was observed in the variants with moderate doses of fertilizers. At this, the activity peak was shifting in dynamics from the variants with small doses of fertilizers to the ones with higher nitrogen concentrations – up to 160 kg/ha (table 1).

Estimation of nitrogen fixation productivity has revealed its highest point in the variant with 20 kg/ha of mineral nitrogen (table 2). Relatively high (higher than control indices) was nitrogen fixation productivity in variants with applied 10, 40 and 80 kg/ha doses of nitrogen fertilizers. Summary indices at application of mineral nitrogen in dose 120 kg/ha were on the control level. Further increase of fertilizers level had led to the decrease of nitrogen fixation productivity. Apparently, in spite of the noticed "splashes" of activity in variants with high doses of fertilizers the summary indices in this case were not very high because of the short-term stimulation of nitrogen fixation in these variants during the vegetative period.

*Table 1.* ***Dynamics of nitrogen fixation activity in root zone of grain crops depending on nitrogen fertilizers doses,*** *μg N/m² per hour*

| Nitrogen doses, kg/ha | Days after application of fertilizers | | | | | | |
|---|---|---|---|---|---|---|---|
| | spring application | | application after 1st hay-harvest | | application after 2nd hay-harvest | | |
| | 20 days | 35 days | 9 days | 58 days | 11 days | 23 days | 45 days |
| 1 | 64,7 | 77,5 | 303,3 | 224,1 | 582,5 | 582,5 | 111,6 |
| 10 | 70,0 | **212,0*** | 283,3 | 201,6 | 481,7 | 572,5 | 304,2 |
| 20 | 86,8* | **168,4*** | 246,6 | **1681,7*** | 459,2 | **4144,2*** | **1254,2** |
| 40 | **185,0*** | **155,0*** | 200,0 | 212,5 | 414,2 | **1792,5*** | **1512,5*** |
| 80 | 50,8 | 134,2 | 225,0 | 111,7 | 370,0 | 1243,0 | **1275,0*** |
| 120 | 50,8 | **187,5*** | 108,3 | 90,0 | 370,0 | 851,6 | **570,8*** |
| 160 | 28,3 | 134,2 | 100,0 | 90,0 | 335,8 | 526,6 | **335,8*** |
| 200 | 28,3 | 111,6 | 141,6 | 78,3 | 324,2 | 526,6 | 111,7 |
| 240 | 33,3 | 78,3 | 100,0 | 78,3 | 370,0 | 515,0 | 100,8 |
| 280 | 28,3 | 55,8 | 75,0 | 78,3 | 290,8 | 526,6 | 78,3 |
| $LSD_{05}$ | 20,0 | 63,3 | 75,0 | 1375,0 | 93,3 | 788,3 | 223,3 |

According to the data obtained we may conclude that application of 20 kg/ha is an ecologically optimum dose. Doses, not exceeding 120 kg/ha of mineral nitrogen are ecologically acceptable. Application of 160 kg/ha might be considered as the ecological threshold or even superfluous.

These data were obtained without using plants' inoculation techniques. Hence, we have the question rised: how does mineral nitrogen influence on the development of introduced nitrogen fixing microorganisms?

As known from literature, the successful development and functioning of microorganisms introduced into the plants' root zone requires presence of some easily accessible nitrogen in the soil. Numerous studies on the efficiency of plants' bacterization with associative nitrogen fixing microorganisms were given consideration due to the showed yield increase resulting from the inoculation performed on the nitrogen fertilizers background.

*Table 2*. ***Nitrogen fixation productivity, grass vegetative mass yield and nitrates contents depending on the nitrogen fertilizers doses***

| Nitrogen doses, kg/ha | Total productivity of nitrogen fixation, kg/ha/150 days | Nitrates content*, mg/kg | Grasses vegetative mass yield, c/ha |
|---|---|---|---|
| 0 | 9,19 | 27,5 | 124,6 |
| 10 | 17,07 | 24,5 | 137,6 |
| 20 | 34,69 | 30,0 | 150,2 |
| 40 | 22,51 | 30,2 | 173,6 |
| 80 | 17,97 | 81,2 | 214,1 |
| 120 | 10,44 | 112,0 | 246,1 |
| 160 | 7,34 | 118,0 | 269,6 |
| 200 | 5,81 | 575,0 | 284,5 |
| 240 | 5,51 | 575,0 | 290,9 |
| 280 | 5,87 | 497,0 | 288,8 |
| $LSD_{05}$ | | | 36,5 |

* – BPC (boundary possible concentration) in grasses vegetative mass equals 500 mg/kg.

Study of mineral nitrogen influence on the development of introduced into the plants' root zone microorganisms testifies their increase in root spheres in 1-2 orders comparing to the nitrogen-free background. The results of calculation of number of *Azospirillum lipoferum* C-1 bacteria in root spheres of English ryegrass (small pot experiment on sod-podzol soils) are given below (table 3). Grass seeds were infected with the named strain prior to their sowing. The strain was adapted in advance to the high concentrations of streptomycin sulfate in order to introduce marking factor to the cells. Data presented testifies the considerably higher level of colonization of root spheres of ryegrass grown on the nitrogen background – number of bacteria has increased in 10-100 times subject to studied root sphere. Thereby, the analogy between the influence of mineral nitrogen on the indigenous and introduced nitrogen fixing bacteria can be drawn. Moreover, introduced nitrogen fixing bacteria have another interesting feature – they enhance assimilation of nutrients and mineral fertilizers by inoculated plants grown in certain (the optimum the better) agrichemical background [6-9].

*Table* ***3. Number of introduced azospirills into the root spheres of English ryegrass under the influence of mineral nitrogen***

| Nitrogen doses, kg/ha | Root sphere | Number of cells, tsnd/g |
|---|---|---|
| 0 | rhizosphere soil | 2,2 |
| | washed roots | 552,0 |
| 40 | rhizosphere soil | 18,2 |
| | washed roots | 10773,0 |

The scientific society agreed on this phenomenon as on the effect of physiologically active substances of microbial preparations on plants ability to form active root adsorbing surface and increase of whole root system. Besides, infected plants undergo considerable changes in penetrability of root tissues membranes revealed by measurement of the intensity of protons flow, segregated by wheat seedlings after inoculation [10]. It was also observed that inoculation activates specific enzymatic systems which can effect on the assimilation of biogenic elements. Thus, in particular, it was shown that inoculation activates plant nitratereductases – key enzymes of nitrogen exchange [11]. Bacterial nitratereductase has an additional effect on the assimilation of e.g. nitrates. This possible aspect of microorganisms influence on nitrogen assimilation by plants was noticed by R. Boddey et al. [12]. So, at least in case of nitrates we have observed an extra effect of microorganisms introduced to the agricoenosis as the overall action of plant and bacterial nitratereductases which also confirms that in this case we are talking not only on the nitrates assimilation but also on their introduction to the metabolism of plants.

Further investigations in this area have resulted in discovering of some interesting facts. Thus, we have studied the efficiency of presowing seeds inoculation (microbial preparation Diazobacterin based on the *Azospirillum brasilense* 410) in field experiments with annual ryegrass, conducted on grey forest podzolic soil ($pH_{salt}$ – 5,1; humus contents – 1,6-1,9 %) on $P_{60}K_{60}$ background and rising nitrogen doses of fertilizers (0, 40, 80, 160 і 200 kg/ha). Nitrogen fixation activity as well as grass productivity was studied in dynamics – in 30, 40 and 50 days after the application of fertilizers. It was shown that physiologically optimum dose of fertilizer for ryegrass on grey forest soils in the absence of bacterization is 40 kg/ha. This variant has demonstrated authentic nitrogen fixation increase by all observed stages (fig. 1).

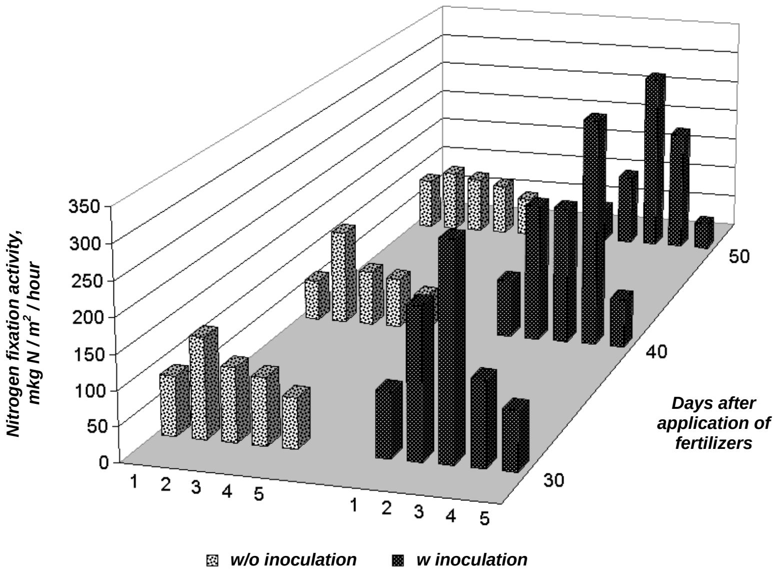


*Fig. 1. Influence of mineral nitrogen and inoculation on the dynamics of nitrogen fixation in root zone of annual ryegrass (1-$N_0$; 2-$N_{40}$; 3-$N_{80}$; 4-$N_{160}$; 5-$N_{200}$).*

Bacterization has resulted in the unexpected increase of ecological optimum of mineral nitrogen in comparison to the variants without inoculation. Thus the biggest value of nitrogen fixation activity was observed in the variant with application of 80 kg/ha of fertilizer. Moreover, even dose 160 kg/ha has initially showed tendency to the enhancement of nitrogen fixation activity and has authentically ensured its increase later on. So, presowing seed bacterization considerably increases ecological optimum of fixed nitrogen for the plants. Well, but how it can be explained if considering that increase of nitrogen fixation leads to the raising of biological nitrogen inflow to the plants? Understanding of this arises in data comparison in variants without inoculation and the ones treated with biopreparation (table 4).

*Table 4.* ***Influence of mineral nitrogen and inoculation on vegetative mass yield of annual ryegrass,*** *field experiment*

| Nitrogen doses, kg/ha | Total yield (2 hay-harvest), c/ha (dry matter) | Inoculation increase | |
|---|---|---|---|
| | | c/ha | % |
| without inoculation | | | |
| 0 | 60,0 | – | – |
| 40 | 66,4 | – | – |
| 80 | 74,9 | – | – |
| 160 | 82,7 | – | – |
| 200 | 94,4 | – | – |
| with inoculation | | | |
| 0 | 66,2 | 6,2 | 10 |
| 40 | 79,1 | 12,7 | 19 |
| 80 | 101,1 | 26,2 | 35 |
| 160 | 104,0 | 21,3 | 26 |
| 200 | 115,3 | 20,9 | 22 |

$LSD_{05}$ by experiment 3,3
for nitrogen 2,4
for inoculation 1,3

Bacterization provides significant increase of crop yield (might be due to the initial influence of physiologically active substances of bacterial nature). To ensure biomass formation plants certainly require bigger amount of nitrogen. Their demand is partially satisfied by activation of nitrogen fixation and additional biological nitrogen supply. But initiated plants are not able to satisfy their needs from the molecular nitrogen fixation only (at inoculation, for example, on $N_{80}$ background plants have formed same vegetative mass as is under the 200 kg/ha dose). In this case we have observed enhancement of mineral nitrogen assimilation aimed on the formation of additional nitrogen. It is obvious that in this case concentration of fixed nitrogen in soil will be decreased influencing the level of nitrogen fixation activity, which in turn will increase due to the sufficient provision of root diazotrophs with carbon and optimal nitrogen content.

All the conclusions made were proved in our field experiments with grass mixture "perennial ryegrass + awnless brome", performed under the conditions mentioned above.

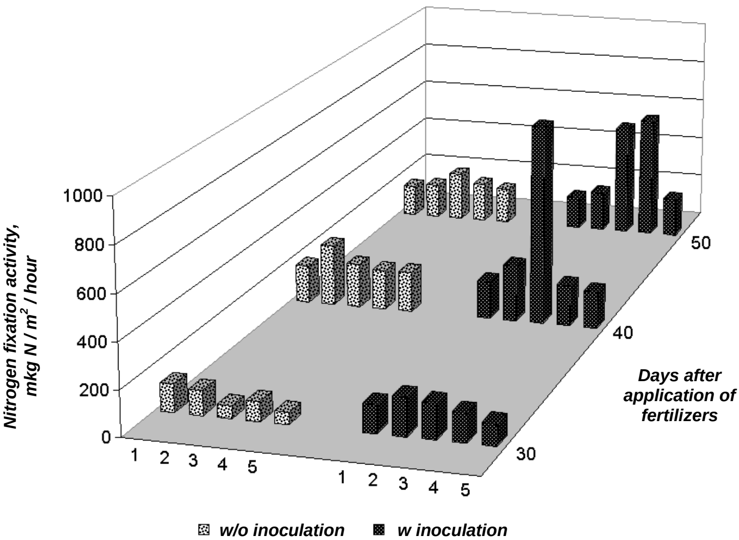


*Fig. 2. Dynamic of nitrogen fixation in root zone of cereal grasses depending on the level of nitrogen fertilizers and inoculation (1-$N_0$; 2-$N_{40}$; 3-$N_{80}$; 4-$N_{160}$; 5-$N_{200}$).*

Here, the most optimal doses by inoculation have fallen within 80-160 kg/ha of mineral nitrogen. Shifting of the activity peak from the variants with the small doses of nitrogen to the bigger ones was clearly showed in the studies of the process dynamics. At the same time both lower activity and less intensive changes were observed in the variants without inoculation: the peak of nitrogen fixation activity had moved to the variant with 80 kg/ha of mineral nitrogen to the end of the experiment (fig. 2). The same as it was shown in the previous experiment, comparison of yield indices in variants with and without inoculation had testified that formation of plants' biomass initiated by bacterization requires much more nitrogen compounds. Use of mineral nitrogen in the processes of constructive metabolism of plants leads to the decrease of the concentration of this element in root zone, thus stimulating nitrogen fixation process.

The one should notice that we have not only promoted additional atmospheric nitrogen supply to the bacterized plants but have reduced dangerous (under the typical conditions) from the ecological point of view doses of mineral nitrogen to the ecologically safe ones.

Our results were proved in the experiments on ryegrass using $^{15}N$ labeling (table 5). Data analysis by first hay harvest (30 days of plants growing) had showed authentic rise of ryegrass vegetative mass, nitrogen content and increase of heavy isotope from inoculation with *Azospirillum sp.* on the 7,5 mg $(NH_4)_2SO_4$ background. Assimilation of mineral nitrogen in this variant was three times bigger than in control one. Second hay harvest had not revealed yield differences between the variants. Increase of $^{15}N$ content was also inauthentic. However, general nitrogen content was higher than in control variant, which is due to the $^{14}N$ inflow resulted from the activation of associative nitrogen fixation.

*Table* 5. ***Influence of inoculation on the ryegrass yield, accumulation of root mass and nitrogen utilization depending on the dose of nitrogen fertilizer,*** *green-house experiment*

| Variants | Yield, g/pot | Increase by inoculation, g/pot | $N_{gen.}$, mg/pot | Increase by inoculation, mg/pot | $^{15}N$, mg/pot | $^{15}N$ increase by inoculation, mg/pot |
|---|---|---|---|---|---|---|
| 1st hay harvest | | | | | | |
| 7,5 mg $(NH_4)_2SO_4$ | 0,328 | – | 15,88 | – | 0,22 | – |
| Same + inoculation | 0,368 | 0,040* | 17,87 | 1,99* | 0,67 | 0,45* |
| 32,5 mg $(NH_4)_2SO_4$ | 0,360 | – | 16,42 | – | 2,20 | – |
| Same + inoculation | 0,380 | 0,020 | 17,85 | 1,43 | 2,20 | – |
| 2nd hay harvest | | | | | | |
| 7,5 mg $(NH_4)_2SO_4$ | 0,984 | – | 17,13 | – | 0,48 | – |
| Same + inoculation | 0,953 | – | 19,99 | 2,86* | 0,68 | 0,20 |
| 32,5 mg $(NH_4)_2SO_4$ | 0,985 | – | 18,69 | – | 1,30 | – |
| Same + inoculation | 1,126 | 0,141* | 21,55 | 2,86* | 2,25 | 0,95* |
| Roots | | | | | | |
| 7,5 mg $(NH_4)_2SO_4$ | 1,254 | – | 11,84 | – | 0,26 | – |
| Same + inoculation | 1,432 | 0,178 | 12,83 | 0,99 | 0,23 | – |
| 32,5 mg $(NH_4)_2SO_4$ | 1,206 | – | 12,11 | – | 0,75 | – |
| Same + inoculation | 1,570 | 0,364* | 17,14 | 5,03 | 1,10 | 0,35* |

* – Authentic values at 95 % level of significance

No positive results were obtained in variants with inoculation by azospirills on the higher doses of mineral nitrogen background within first hay harvesting. Nevertheless, the second hay harvesting (60 days of plants growing) had resulted in the increase of all studied indices.

Overall data values obtained during the experiment are showed in table 6. As is seen, inoculation has promoted mineral nitrogen utilization at 1.3 mg/pot upon the application of larger dose and at 0.62 – upon application of the lower one.

Inoculation on the background of higher doses of nitrogen fertilizer promotes both utilization of mineral nitrogen and assimilation of $^{14}N$ nitrogen (atmospheric nitrogen) by plants.

Thus, inoculation of cereal grasses stimulates nitrogen fixation activity; increases grass yield and assimilation level of mineral nitrogen used for the formation of additional output. Notice, that at this the output quality was nor worsening but improving. Reduced nitrates amount accompanied with increased protein contents was observed in vegetative mass of the grasses due to the activation of plant's enzyme systems of nitrogen cycle.

*Table 6.* ***Influence of inoculation and mineral nitrogen fertilizers on the nitrogen isotopes content in English ryegrass plants***

| Variants | General nitrogen, mg/pot | $^{15}N$ carry-over, mg/pot | Utilization of $N_{fertilizer}$, % | Atmospheric nitrogen ($^{14}N$) inflow, mg/pot |
|---|---|---|---|---|
| 7,5mg $(NH_4)_2SO_4$ | 44,85 | 0,96 | 60,0 | 3,24 |
| Same + inoculation | 50,69 | 1,58 | 98,8 | 8,44 |
| 32,5 mg $(NH_4)_2SO_4$ | 47,22 | 4,25 | 61,6 | 2,32 |
| Same + inoculation | 56,54 | 5,55 | 80,4 | 10,34 |
| $LSD_{05}$ | 2,04 | 0,26 | 16,3 | |

Previously, studying the influence of various factors on the course of nitrogen fixation, we had observed stimulatory action of plant hormones of auxin and cytokinin nature as well as their synthetic analogues on this process [13]. We had assumed certain similarity in action of microbial preparations and some growth regulators on the ecological optimum of mineral fertilizers, since both inoculation and use of plant growth regulators had resulted in the initiation of plants' development and yield increase.

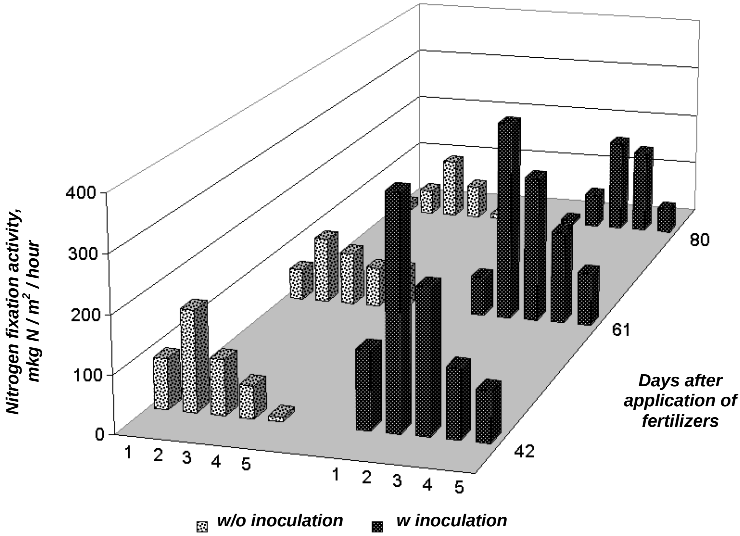


*Fig. 3. Influence of mineral nitrogen and Triman-1 on nitrogen fixation activity in root zone of cereal grasses (1-$N_0$; 2-$N_{40}$; 3-$N_{80}$; 4-$N_{160}$; 5-$N_{200}$).*

Our experiments were conducted on the grey forest podzol soils ($pH_{salt.}$ – 5.1; humus contents – 1,6-1,9 %) on the fields with cereal grass stand (perennial ryegrass + awnless brome + timothy-grass + meadow fescue grass). Field experiments were conducted under the same scheme used in our previous studies, with the exception of microbial preparation substituted with plant growth stimulator – Triman-1.

Same as it was shown before we have observed considerable effect of small doses of mineral nitrogen on the nitrogen fixation activity in root zone of cereal grasses (first, it was stimulated in the variant with 40kg/ha dose, while at the end of vegetative period its indices were higher in the one with 80 kg/ha). Use of growth stimulator had resulted in almost same effect as the use of microbial preparation – the highest activity values were noticed under the 80 and 160 kg/ha doses of mineral nitrogen (fig. 3). Influence of plant growth regulators is probably alike effect of physiologically active substances of bacterial nature, produced by the introduced into the plants root zone bacteria. At this, active development of root system and whole plants causes high needs in fixed nitrogen due to the formation of considerably bigger biomass (table 7). This results in the increase of physiological (ecological) optimum of nitrogen for the plants.

*Table 7.* ***Interaction efficiency of mineral nitrogen and plant growth stimulator Triman-1****, field experiment*

| Dose of nitrogen, kg/ha | Yield, total for 3 hay harvest (dry matter), ц/ha | Increase by Triman-1 | |
|---|---|---|---|
| | | c/ha | % |
| without Triman-1 | | | |
| 0 | 54,4 | – | – |
| 40 | 75,5 | – | – |
| 80 | 98,1 | – | – |
| 160 | 122,6 | – | – |
| 200 | 148,7 | – | – |
| with Triman-1 | | | |
| 0 | 53,4 | – | – |
| 40 | 91,7 | 15,9 | 21 |
| 80 | 118,9 | 20,9 | 21 |
| 160 | 132,8 | 10,1 | 8 |
| 200 | 168,6 | 19,9 | 13 |
| $LSD_{05}$ experiment | 5,7 | | |
| for Triman-1 | 2,3 | | |
| for nitrogen | 4,1 | | |

All considerations made were proved in our experiments using isotopic dilution method. Ryegrass plants (Drogobytska cultivar) were grown in the climatic camera conditions. The scheme of experiment was as follows:

1. Control;
2. Inoculation *Azospirillum lipoferum*, 4014;
3. Triman-1, presowing seed treatment;
4. Triman-1, treatment of vegetative plants.

Illumination conditions – 20 thousand lux on plant level, photoperiod – 16 hours; air temperature – 26 ± 1°C, sand humidity – 70 % of total water capacity. Mixture of labeled and regular (not labeled) nitrogen fertilizers were applied into the pots (650 ml) with washed and roasted sand. Isotopic nitrogen contents in leaves, plants stems and roots as well as in the substrate was analyzed on mass-spectrometer МИ-1201.

In strictly controlled conditions we have observed positive influence of inoculation and use of plant growth stimulator on grass yield and nitrogen uptake. Moreover, increased content of mineral and atmospheric nitrogen was revealed in plants initiated either by bacteria or growth stimulators (table 8).

Increased mineral nitrogen uptake upon inoculation or use of growth stimulators is not standing that plant are accumulating inorganic nitrogen. As was shown in our experiments use of named agricultural techniques considerably promotes nitrogen fixing enzymes' activity in plants, and nitratereductase, in particular (table 9).

Nitratereductase activity in our experiments was increased by 1,7-2,4 depending on the plants ontogenesis stage while its action was observed during prolonged period of time (100 days).

*Table 8.* ***Isotopic nitrogen contents in vegetative mass, roots of ryegrass and substrate***

| Variants | $^{14}N$ and $^{15}N$ contents, mg/pot | | | | | |
|---|---|---|---|---|---|---|
| | vegetative mass | | roots | | substrate | |
| | $^{14}N$ | $^{15}N$ | $^{14}N$ | $^{15}N$ | $^{14}N$ | $^{15}N$ |
| Control | 2,11 | 1,77 | 1,40 | 1,34 | 0,98 | 0,05 |
| Inoculation | 2,21 | 2,82 | 1,98 | 1,52 | 0,64 | 0,03 |
| Triman-1, seeds treatment | 2,21 | 2,20 | 1,85 | 1,42 | 1,07 | 0,03 |
| Triman, plants treatment | 2,31 | 2,63 | 1,88 | 1,20 | 1,09 | 0,09 |

It was established that activation of plants' nitrogen fixing enzymes results in the decrease of nitrates quantity which is quite obvious since in this case nitrates from reserve pool are involved into the plants' metabolism.

*Table 9.* ***Dynamics of nitratereductase activity in leaves of ryegrass under the inoculation*** *(green-house experiment)*

| Variants | Nitratereductase activity, µg $NO_3$/g raw mass per 30 minutes | | |
|---|---|---|---|
| | 70 days after grass standing | 85 days after grass standing | 100days after grass standing |
| Control | 7,50 | 9,10 | 9,93 |
| Diazobacterin | 18,24 | 18,59 | 16,81 |
| Triman-1, presowing seeds treatment | 19,50 | 18,15 | 15,44 |
| $LSD_{05}$ | 1,42 | 2,30 | 2,57 |

Thus inoculation and (or) plants treatment with growth stimulators promotes rise of ecological nitrogen optimum for plants, and improves ecological state of agrocoenosis due to the increase of fertilizers uptake level and their use in plant metabolism as the result. At this considerably high under the typical conditions doses of mineral nitrogen used together with the bacterial preparations or plant growth stimulators are proceed to the category of ecologically appropriate ones.

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## ЗБІЛЬШЕННЯ ФІЗІОЛОГІЧНОГО ОПТИМУМУ АЗОТУ ДЛЯ РОСЛИН ЗА ІНОКУЛЯЦІЇ ТА ЗАСТОСУВАННЯ СТИМУЛЯТОРІВ РОСТУ

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*У вегетаційних дослідах за використання методу ізотопного розбавлення та у польових, проведених на сірому лісовому та дерново-підзолистому ґрунтах, показано, що інокуляція та (або) обробка злакових трав стимуляторами росту сприяє зростанню фізіологічного оптимуму азоту для рослин, і внаслідок цього – поліпшенню екологічної обстановки в агроценозах через зростання ступеню засвоєння добрив і залучення їх до конструктивного метаболізму. При цьому високі за типових умов дози мінерального азоту, застосовані у поєднанні з бактеріальними препаратами або стимуляторами росту рослин, переходять у розряд екологічно доцільних.*

Ключові слова: *азот, асоціативна азотфіксація, фізіологічний оптимум, метод ізотопного розбавлення, злакові трави.*

## УВЕЛИЧЕНИЕ ФИЗИОЛОГИЧЕСКОГО ОПТИМУМА АЗОТА ДЛЯ РАСТЕНИЙ ПРИ ИНОКУЛЯЦИИ И ПРИМЕНЕНИИ СТИМУЛЯТОРОВ РОСТА

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*В условиях вегетационных опытов при использовании метода изотопного разбавления и в полевых, проведенных на серой лесной и дерново-подзолистой почвах, показано, что инокуляция и (или) обработка злаковых трав стимуляторами роста способствует увеличению физиологического оптимума азота для растений, и вследствие этого – улучшению экологической обстановки в агроценозах при увеличении степени усвоения удобрений и вовлечения их в конструктивный метаболизм. При этом высокие в типичных условиях дозы минерального азота, примененные в сочетании с бактериальными препаратами или стимуляторами роста растений, переходят в разряд экологически приемлемых.*

Ключевые слова: *азот, ассоциативная азотфиксация, физиологический оптимум, метод изотопного разбавления, злаковые травы.*