

Nanopreparations in technologies of plants growing

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Abstract. The use of engineered nanomaterials in sustainable agriculture has demonstrated a completely new way of food production that can potentially overcome uncertainty in the agricultural sector with limited available resources. Nanoparticle engineering is one of the latest technological innovations which demonstrate unique target characteristics.

During 2013–2020, research on the directions and effectiveness of nanopreparations in plant growing: nutrient source, activation of photosynthesis, immunocorrectors, stimulators of seed germination, plant growth and development, multivalent drugs for increasing plant resistance to stress was conducted. Monoparticles, nanoparticle combinations, and chelate complex of nano fertilizers on crops of soybean were tested. Field research was conducted in a stationary field experiment of the Plant Science Department of the National University of Life and Environmental Sciences of Ukraine. The soil of the stationary experiment is typical chernozem. In research was used soybean (*Glycine max* (L.) Merr.) of early-ripening variety Horol. Over the years of research, weather conditions varied, but were within the typical for zone of research. Average monthly temperatures were close or higher the perennial average indicators.

The purpose of the research is to find out the influence of pre-sowing seed treatment and fertilizing of crops by nano-preparations Avatar (microfertilizer of carboxylates of natural acids), Iodis-concentrate (immunomodulator - stimulator of growth processes), and Super Micro Plus (nanochelate fertilizer) on leaf formation - rate, the activity of symbiotic nitrogen fixation and yield of soybean variety Horol.

Nanopreparations were used for pre-sowing seed treatment and fertilizing - spraying during the growing season in several doses. The use of nanopreparations, as seed treatment in combination with inoculation and as fertilizer, intensified formation of the leaf surface area, symbiotic apparatus activity of soybean plants. The introduction of nanofertilizers complex in the top-dressing helped to increase yields and change the functional quality of crop products which indicates their unconditional effectiveness. Soybean yield significantly depended on weather conditions, varying from 1.23 to 3.48 t ha⁻¹ depending on the weather conditions and the combination of seed inoculation and nanofertilizer. Soybean yield under favourable weather conditions in 2016, depending on the use of preparation combination ranged from 2.27 to 3.48 t ha⁻¹.

As a result of the research, it was found that the use of nanopreparations Avatar, Jodis-concentrate and Super Micro Plus for seed treatment and fertilizing intensified leaf surface formation and symbiotic apparatus activity of soybean plants. The obtained results confirm that application of nanofertilizers complex Jodis-concentrate, Avatar and nano chelate fertilizer Super Micro Plus in the soybean fertilizing helped to increase the yield, which testifies to their unconditional effectiveness. The highest efficiency of nanofertilizers was shown by inoculation and seed treatment by Avatar and fertilizing by Avatar + nano chelate fertilizer Super Micro Plus, providing the formation of 52.4 thousand m² ha⁻¹ of leaf surface area of soybean varieties Horol, 69.7 pcs per plant of root nodules, 785 mg per plant of their weight and yield at the level of 2.79 t ha⁻¹ an average of five years.

Key words: nano fertilizers, forms of fertilizers, soybean, yields.

INTRODUCTION

To decide the growing challenges of sustainable production and food security, significant technological advances and innovations have been achieved in recent years (Dijk & Meijerink, 2014; Kou et al., 2018). The growing demand for food in the world causes an increase in the intensity of production in plant growing, which leads to a chain of negative consequences – soil depletion, reduced crop yields, a significant increase in economic and energy costs (Kirchmann et al., 2020). About 40% of agricultural land in the world, due to intensive production, degraded, this led to significant losses of soil fertility (Kale & Gawade, 2016).

Sustainable agriculture involves the minimal use of agrochemicals, which has a positive impact on the environment, conservation of species from extinction, improving management tactics and conservation by reducing the cost of agricultural resources (Dubey & Mailapalli, 2016; Dwivedi et al., 2016). Energy use in agricultural production has been increasing faster than in many other sectors of the world economy. Owing to high energy consumption during the production of agricultural inputs, with mineral nitrogen fertilizers in particular, it is often questioned as to whether agricultural production is still energy efficient. The compared systems differed in nitrogen fertilization rates and the level of fungicide protection. The highest output/input ratio was noticed growing winter triticale in low-input production system. The most energy-consuming operation during winter triticale production in the compared systems was mineral fertilization. The high-input production system was significantly lower energy efficiency than the other systems (6.21, medium-input 5.95, low-input 8.19) (Bielski et al., 2019).

Nanotechnology could become a key technology in the twenty-first century. Over the last two decades, a significant amount of research on nanotechnology and their use in agriculture has been conducted (Khan & Rizvi, 2014; Chen et al., 2016; Kozyrskyi et al., 2019). One of the ways to achieve food security can be the development of innovative technologies for plants growing (Makarenko et al., 2015; Eremenko et al., 2019).

Advances in the production of nanomaterials of various sizes and shapes have given a wide range of applications in medicine, ecology, agriculture and food industry (Godfray et al., 2010; Panpatte et al., 2016; Worrall et al., 2018). The use of engineered nanomaterials has shown a completely new way of food production that can potentially overcome uncertainty in the agricultural sector with limited available resources (Lopatko et al., 2009; Godfray et al., 2010). Nanotechnologies have many promising areas of use in crop production: nutrient source, activation and growth of photosynthesis,

immunocorrectors, antistressors, nanopesticides, specific root effectors for rooting shoots and tissue crops, multivalent drugs to increase plant resistance to stress (Kwak et al., 2017; Prasad et al., 2017).

An important role is given to nanofertilizers and growth-stimulating complexes (Kou et al., 2018.). It is estimated that at least a third of crop productivity is accounted for by fertilizers, and the rest depends on the efficiency of other agricultural resources (Dubey & Mailapalli, 2016). However, the efficiency of nutrient use from traditional fertilizers does not exceed 30–40% (Dijk & Meijerink, 2014) and largely depends on the final concentration fertilizer that reaches the target place (Solanki et al., 2015). And reaches much lower than the minimum required concentration, due to the loss of chemicals during leaching, application, runoff, hydrolysis, evaporation, degradation (Sabir et al., 2014; Miao et al., 2015; Wang et al., 2015). Repeated use of excessive amounts of fertilizers negatively affects the inherent balance of soil nutrients, water purity, drinking water pollution, development of flora and fauna (Makarenko et al., 2015).

Nanofertilizers synthesized specifically for the controlled release of nutrients depending on the crops needs, minimizing differential losses, have great potential (Sabir et al., 2014; Solanki et al., 2015; Batsmanova et al., 2020). Controlled released nanoparticles improve plant growth and development, increase yields and improve product quality (Vermeulen et al., 2012; Kale & Gawade, 2016; Shcherbakova et al., 2017). The approach to targeted delivery of nutrients based on nanosized particles is used to optimize the production process, aimed at changing its passage, through certain areas of their functional efficiency (Sabir et al., 2014; Wang et al., 2015; Gogos et al., 2020).

Fertilizer nanoformulas synchronize the ‘emission’ of nanoparticles with the demand of the culture, which prevents nutrient loss through direct use by plants, avoiding the interaction of nutrients with soil, water, air and microorganisms (Miao et al., 2015; Dubey & Mailapalli, 2016).

An important problem in the growing crops efficiency is the use of high quality seeds. The search for ways of reducing seed injury during harvesting and post-harvest cleaning of crop seeds is quite relevant (Bulgakov et al., 2020). There are ongoing studies on ways to reduce seed injury and increase its sowing properties, which may also be associated with the use of nanopreparations or monoelements for pre-sowing seed treatment (Shcherbakova et al, 2017).

MATERIALS AND METHODS

Field research was conducted in 2016–2020 in a stationary field experiment at the ‘Agronomic Research Station’ of NULES of Ukraine in a 10-field field crop rotation and on the basis of educational and scientific laboratory ‘Demonstration collection field of agricultural crops’ of the Plant Science Department of NULES of Ukraine. The soil of the experimental site is typical low-humus chernozem, coarse-dusty-medium-loamy in granulometric composition. The humus content is 4.39–4.53%; pH of salt extract - 6.9–7.3; absorption capacity - 30.7–32 mg-eq. per 100 g of soil. The content of total nitrogen (according to Keldal) is 0.27–0.31%, total phosphorus is 0.15–0.25%, and potassium is 2.3–2.5%. The content of mobile phosphorus (according to Machigin) is 4.5–5.5 mg per 100 g of soil, and of exchangeable potassium is 9.8–10.3.

In research was used soybean (*Glycine max* (L.) Merr.) variety Horol. It’s early ripening variety of grain direction of use. Yield: 3.0–3.3 t ha⁻¹. Resistance to diseases -

up to 1 point. Plant is 75–85 cm height with flower purple and yellow seeds. The weight of 1,000 seeds is 180–190 g, oil content - 19–20%.

Soybean was sown with soil warming at the depth of seeding to +8 °C. In all years of research, the calendar date of sowing corresponded to the first decade of May.

In accordance with the set goal, research program and field experiment scheme were developed, which included options for seed treatment (Table 1).

Table 1. The effectiveness of nanopreparations and pre-sowing seed treatments (scheme of the experiment)

Pre-sowing treatment <i>factor A</i>	Designation of a variant	Drugs use <i>factor B</i>	Designation of a variant
Seed inoculation (<i>control</i>)	A1	Seed treatment by water (<i>control</i>)	B1
Seed inoculation + Iodis concentrate	A2	Iodis concentrate	B2
Seed inoculation + Avatar	A3	Nano Chelate fertilizer Super Micro Plus	B3
		Avatar	B4
		Avatar + Jodis concentrate	B5
		Avatar + Nano Chelate fertilizer Super MicroPlus	B6

Seeds before sowing were treated by wet method according to the developed scheme.

The area of the accounting plot is 50 m², repetitions is quadruple. The placement of plots is systematic. Seeds sowing rate is 600 thousand seeds ha⁻¹. HighStik (4 kg per 1 ton of seeds) was used as inoculum. Preparative form is sterilized peat. Active substance is Bradyrhizobium japonicum (strain 532 C).

Over the years of research, weather conditions varied, but were within the typical for zone of research. Average monthly temperatures were close or higher the perennial average indicators, except May 2020, which is was colder (Fig. 1). In general, years of research characterized by the warm spring and hot summer.

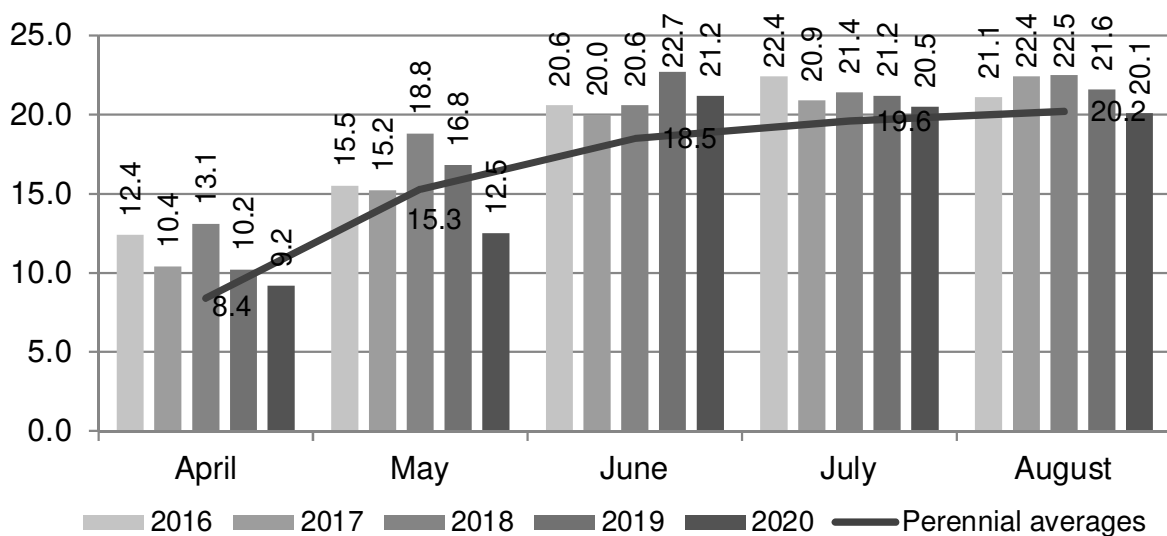


Figure 1. The average monthly temperature for the years of research during the growing season, °C.

During the vegetations of 2016–2020, precipitation was unevenly distributed (Fig. 2).

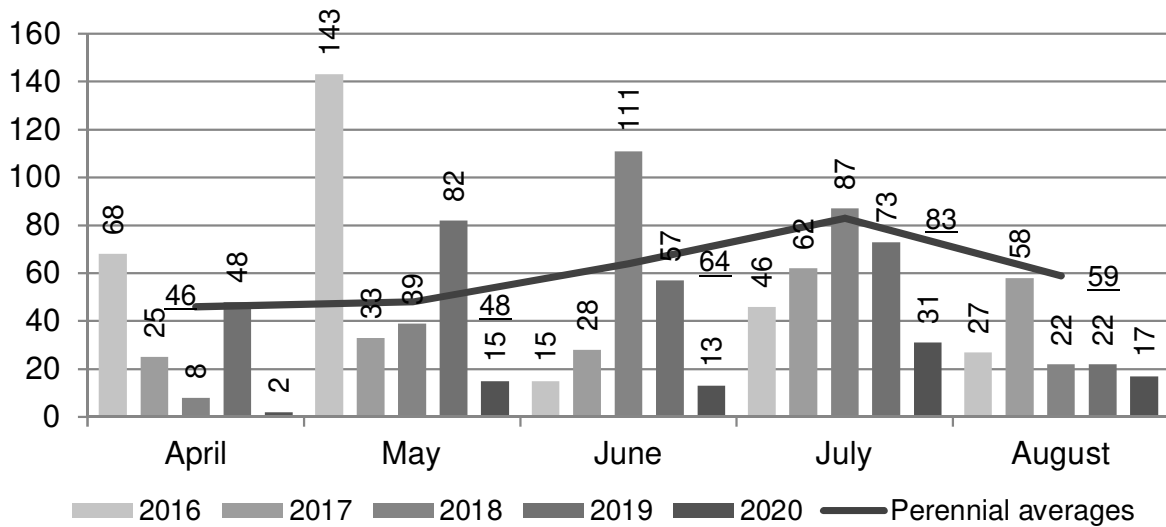


Figure 2. The average monthly precipitation during the growing season, mm.

During the spring months of 2016, the plants were sufficiently provided with moisture, and in May, even in excess. Index of precipitation was much higher average perennial. The similar situations were observed in June and July 2018 and in May 2019.

These conditions had both positive and negative effects on the development of soybean plants.

Next preparations were used in our research:

Complex microfertilizer Avatar (Co - 0.0001–0.0025%, Cu - 0.01–0.08, Zn - 0.001–0.007, Fe - 0.0015–0.008, Mn - 0.0005–0.005, Mo - 0.00001–0.0025, Mg - 0.01–0.08%), contains a complex of citrate chelates obtained from colloidal solutions of metals of such important trace elements as zinc, magnesium, copper, manganese, iron, cobalt, molybdenum. The complex microfertilizer contains microelements chelated by natural organic acids - carboxylates, necessary for plant growth and development.

Iodis concentrate is an immunomodulatory drug. Aqueous solution containing biologically active iodine (70 mg dm⁻³). The total mineralization is not more than 1,000 mg per dm³. Chemical composition, mg per dm³ (not more): Na + K - 10–100; Ca - 50–150; Mg - 10–100; chlorides - < 50; hydrocarbons -300–600.

Nanochelate fertilizer Super Micro Plus contains Fe - 4.5% (FeO), (FeS) - 9% of the total quantity), Zn - 8% (ZnO), (ZnS) - 13% of total), Mn - 0.8% (MnO), (MnS) - 3.2% of the total), K - 3% (KO) - 5% of the total), Mg - 6% (MgO), (MgS) - 9.5% of the total), Cu - 0.65% (CuO) - 2.7% of the total), N - 5% (NO), (N₂H₄O₃) - 10.3% of the total), P - 3% (PO) - 5% of the total), P - 3% (P O) - 5% of the total), Mo - 0.1% (MoO) - 2.1% of the total), Ca - 6% (CaO), (CaS) - 10.5% of the total), B - 0.65% (BO) - 2.7% of the total).

The leaf surface area was determined by scanning the leaf surface and determined their area using the computer program IpSquare, yield by accounting method, statistical data processing was performed using the program SAS 9.4 and IBM SPSS.

RESULTS AND DISCUSSION

Soybean is extremely demanding to environmental factors during the growth and development of plants and especially to weather conditions, nutrient supply. Weather conditions during the years of research differed significantly in both temperature and rainfall. The most favorable conditions for plant growth and development were in 2016 and 2018 - sufficient rainfall and optimal temperatures. Weather conditions in 2017 and 2019 were characterized by a deficit of moisture in certain periods of plants' growth and development at relatively high average daily air temperatures. At the same time, the weather conditions in 2020 were extremely unfavorable and close to extreme conditions for moisture supply in combination with critically high air temperatures, which negatively affected the growth and development of plants.

The leaf area of soybean crops significantly depended on the weather conditions of the year (Figs 3, 4). Pre-sowing seeds inoculation in combination with nano preparation used for seed treatment and fertilizing during the growing season provided differentiation of plants in terms of growth and development, which could primarily be identified by the intensity of leaf surface area formation.

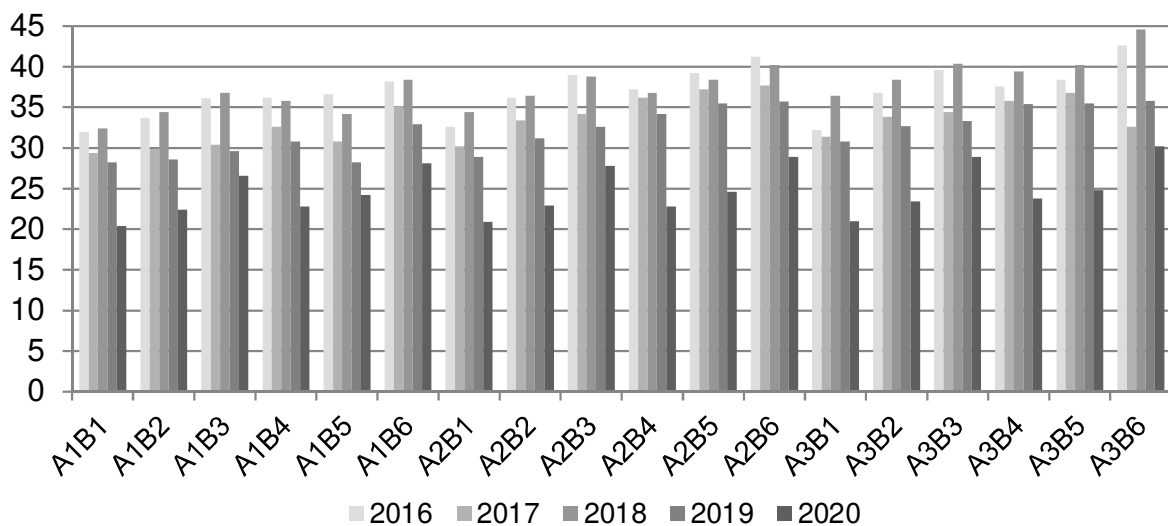


Figure 3. The leaf surface area of soybean crops in the flowering phase (BBCH 65-70) depending on the plants and seeds treatment by nano preparations, thousand $m^2 ha^{-1}$.

The advantage of foliar feeding by the liquid complex of nano fertilizers in comparison with the variant without fertilizing in terms of leaf surface area is obvious.

The use of nano preparation for seed treatment and fertilizing intensified leaf surface area formation of soybean plants. The maximum leaf surface area - 50.4 thousand $m^2 ha^{-1}$ was formed by seeds inoculation, seed treatment, and fertilizing by Avatar + Nano Chelate fertilizer Super Micro Plus.

Soybeans, unlike many field crops, continue to actively form the leaf surface after the flowering phase. The determination of leaf area confirmed that the maximum size of the leaf surface of soybean crops was formed at 78–82 microstages on the BBCH scale (Fig. 4). The maximum area of the leaf surface of the crop was formed by the combined treatment of seeds by inoculant and Avatar, and fertilizing plants by solution of Avatar

+ Nano Chelate fertilizer Super Micro Plus - 52.4 thousand $m^2 ha^{-1}$ on average for 2016–2020. The leaf surface area in 2016 and 2018, with this combination of nano preparation, was 58.0 and 58.8 thousand $m^2 ha^{-1}$, respectively.

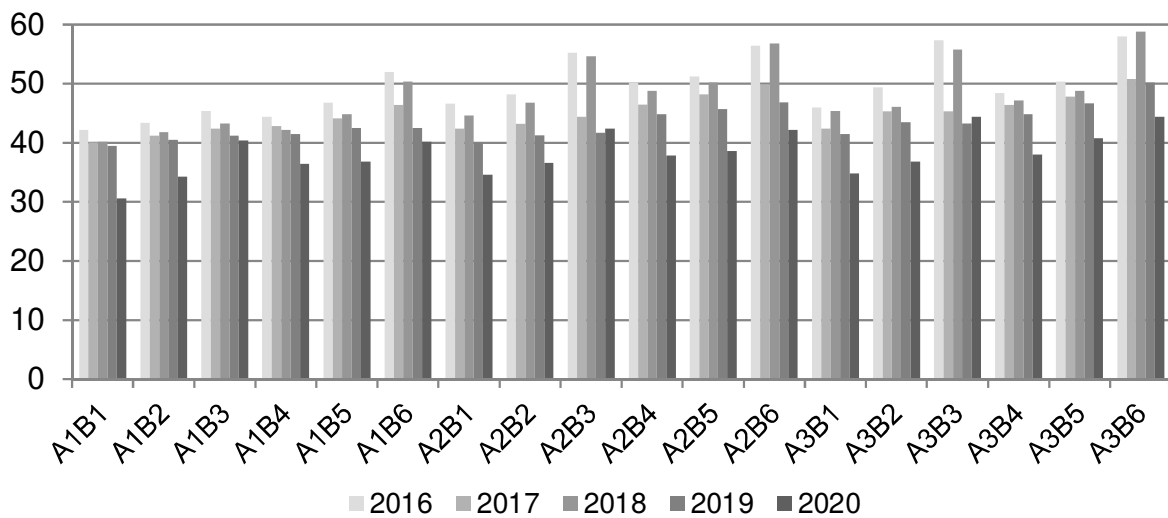


Figure 4. The leaf surface area of soybean crops in the beginning of ripening (BBCH 78–82) depending on the plants and seeds treatment by nano preparations, thousand $m^2 ha^{-1}$.

The leaf surface area in favorable for the growth and development of soybean 2016 and 2018 with this combination of nano preparation, was 58.0 and 58.8 thousand $m^2 ha^{-1}$, respectively. The use of nano preparation, both for pre-sowing seed treatment and for foliar feeding, helps to increase the resistance of plants to abiotic stresses. During the combined drought in 2020, soybean crops formed a leaf surface area from 30.6 to 44.4 thousand $m^2 ha^{-1}$, depending on the drugs' combinations.

When the seeds were treated only by HighStick inoculant, the leaf surface area was significantly lower compared to other applications of nano preparations. During the years of research, it varied in the range of 30.6–42.2 thousand $m^2 ha^{-1}$. Among the studied combinations of nano preparations for soybean crops fertilizing on the background of inoculation, higher efficiency was noted in the variant of experiment A1B6, with the combination of inoculation and Avatar + Nano Chelate fertilizer Super Micro Plus. The leaf surface area, meanwhile, varied between 40.2–52.0 thousand $m^2 ha^{-1}$.

The activity of symbiotic nitrogen fixation reached a maximum during the period of their greatest physiological activity, during the flowering period - the maturation of beans (BBCH 65-70 - 75–80). Prior to the start of beans maturation, there was an active root nodules formation and an increase in their mass in all studied variants, with the next phase the mass began to slowly decrease to full maturity of plants. The use of Avatar and Super Micro Plus nano fertilizers for foliar feeding proved to be quite effective for the formation and activity of the symbiotic apparatus of soybean plants.

Among the studied variants of soybean seed treatment, a smaller number of root nodules, to 40.1 pcs per plant per vegetation, were formed on the plants' root system with seeds inoculation and treatment by Iodis-concentrate fertilizer. It should also be noted that the arid weather conditions in 2019 did not contribute to the significant root nodules formation on soybean roots and in the experiment with seed inoculation without

additional fertilizing of crops, the number of root nodules per growing season did not exceed 12.0 pcs per plant.

Seed treatment by Avatar and Iodis-concentrate nano fertilizers in combination with inoculation and without additional fertilizing of crops increased the number of root nodules during the growing season by 7–11%. Foliar application of Avatar micro fertilizer and Super Micro Plus nano chelate fertilizer contributed to the growth of the root nodules number by 11–18%.

The main criteria for evaluating the efficiency of photosynthesis, biological nitrogen fixation, and plant productivity formation are indicators of individual plant productivity and soybean yield. The results of the research made it possible to establish the positive effect of the use of Avatar fertilizer, Jodis-concentrate, and Nano Chelate fertilizer Super Micro Plus in the formation of individual productivity indicators. Soybean yield significantly depended on weather conditions, varying from 1.23 to 3.48 t ha⁻¹ depending on weather conditions and the combined use of seed inoculation and nano fertilizer. The most favorable conditions for the growth and development of soybeans were in 2016. Yields, depending on the use of drugs combination ranged from 2.27 to 3.48 t ha⁻¹ (Fig. 5).

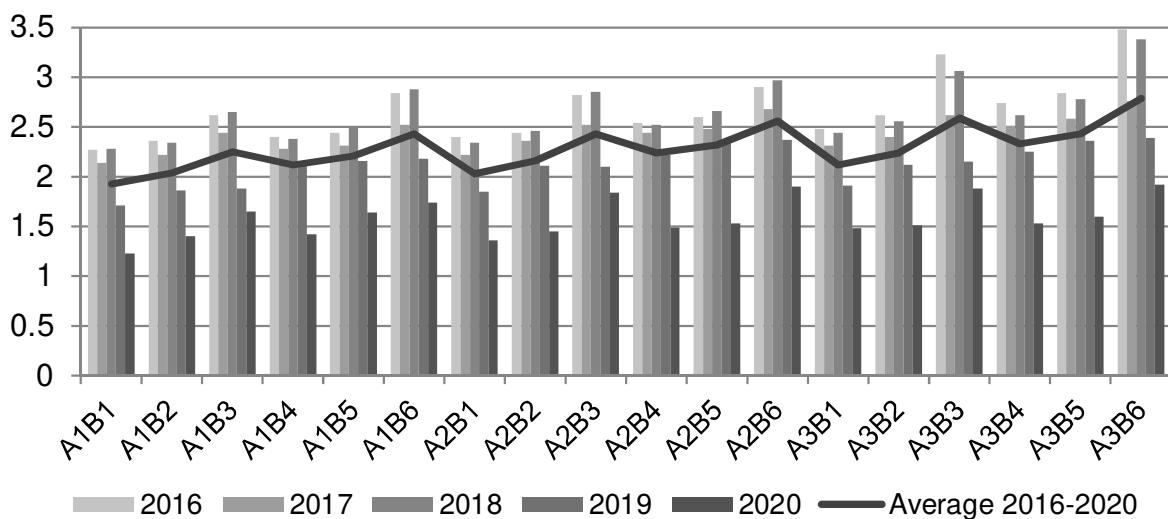


Figure 5. Soybean yield depending on the combined use of nano preparations, t ha⁻¹.

The yield of soybean in the control variant without fertilizing (control) and seed treatment by inoculant only on average for 2016–2020 amounted to 1.93. Pre-sowing treatment of seeds by Iodis-concentrate or Avatar, in addition to inoculation, increased yield by 0.10–0.19 t ha⁻¹. At the same time, the effectiveness of nano preparations increased with the combined use of Avatar and Nano Chelate fertilizer Super Micro Plus. The soybean yield in the control variant (A1B1) in 2019 was 1.71 t ha⁻¹, and in the variants with fertilizing during vegetation and combined pre-sowing seed treatment (A2B1; A3B1) the yield varied from 1.86 to 2.36 t ha⁻¹. The use of Avatar + Nano Chelate fertilizer Super Micro Plus for fertilizing increased the crop yield to 2.18 (on background of A1B1); 2.37 (at background A2B1); 2.39 t ha⁻¹ (on background of A3B1).

The highest yield of soybeans in all years of the research was formed by the use of nano preparations during plant vegetation, formed from seeds treated before sowing by inoculant and the drug Avatar.

The combination of pre-sowing seed treatment with inoculant and Avatar (A3B1) and fertilizing during vegetation by Avatar + Nano Chelate fertilizer Super Micro Plus(A3B6) was more effective -2.79 t ha^{-1} on average for 2016–2020 over the range of yield changes by years from 1.92 to 3.48.

The significance of yield difference depending on the drugs combination and application methods is shown in Table 2.

Cluster analysis is a sequential process of combining into one group of the most similar, related combinations of drugs on a set of characteristics, performed using the IBM SPSS methodology. Signs are the value of soybean yield, which is formed depending on the combination and methods of nanopreparations' application. The Hierarchical method of cluster analysis and the method of proximity between clusters are used: the square of Euclidean distances.

The cluster analysis of the effectiveness of drugs combinations and methods of their use for each vegetation year, allowed us to identify the most effective combinations (Figs 6–10). We present an analysis of the formed clusters in contrasting weather conditions - 2016 and 2020; and in 2017, 2018 and 2019, similar dependencies are observed.

In 2016, favourable by the weather conditions, which formed the highest level of yield, according to the dendrogram results and analysis of the distance between clusters among 18 combinations of drugs and methods of their application can be divided into 3 large clusters and one much smaller cluster, each of which combines options combinations of drugs that cause formation of close yields. The following combinations are combined into these clusters (Fig. 6).

I cluster: A1B6, A3B5, A2B3, A2B6, A3B4 - yield in the range of $2.74\text{--}2.90 \text{ t ha}^{-1}$;

II cluster: A1B3, A3B2, A2B5, A2B4 - $2.54\text{--}2.62 \text{ t ha}^{-1}$;

III cluster: A1B5, A2B2, A3B1, A1B4, A2B1, A1B2, A1B1 - yield in the range of $2.27\text{--}2.48 \text{ t ha}^{-1}$;

IV cluster: A3B3 and A3B6 - $3.23\text{--}3.48 \text{ t ha}^{-1}$.

Thus, in 2016, the highest level of yield was formed by pre-sowing seed treatment in both combinations with inoculant and Avatar and additional feeding by Nano Chelate fertilizer Super Micro Plus (B3) and Nano Chelate fertilizer Super Micro Plus + Avatar (B6).

Table 2. Significance of yield difference depending on the drugs combination and application methods

Variant	Year					Average
	2016	2017	2018	2019	2020	
A1B1	d	e	e	e	e	e
A1B2	d	e	e	d	d	d
A1B3	c	c	cd	d	b	bc
A1B4	d	d	e	c	d	c
A1B5	d	d	d	c	b	c
A1B6	b	b	c	c	a	bc
A2B1	d	e	e	d	d	d
A2B2	d	c	d	c	c	c
A2B3	b	b	c	c	a	bc
A2B4	c	c	d	b	c	bc
A2B5	c	b	cd	a	c	c
A2B6	b	a	b	a	a	b
A3B1	d	d	d	d	c	c
A3B2	c	c	cd	c	c	bc
A3B3	a	b	b	c	a	b
A3B4	b	b	cd	b	c	bc
A3B5	b	b	c	a	b	bc
A3B6	a	a	a	a	a	a

Significant differences ($p < 0.05$) between the mean values of different seed strengths in one line are indicated by different letters.

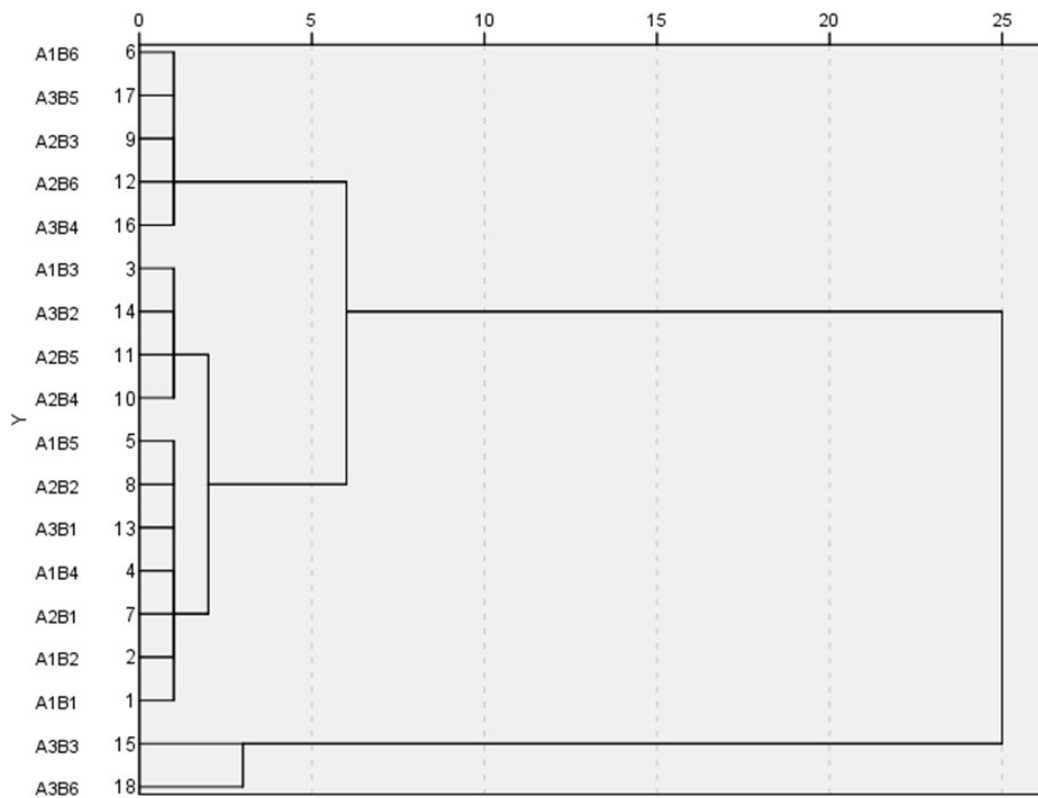


Figure 6. Dendrogram of yield using the method of intergroup connections (year of research 2016).

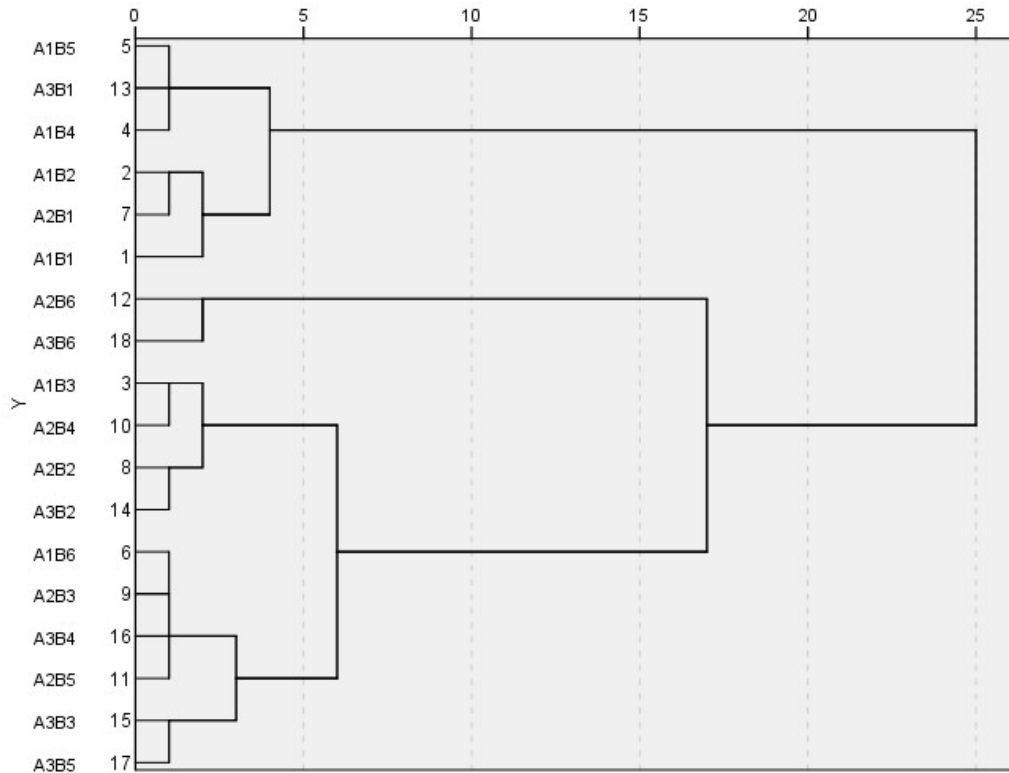


Figure 7. Dendrogram of yield using the method of intergroup connections (year of research 2017).

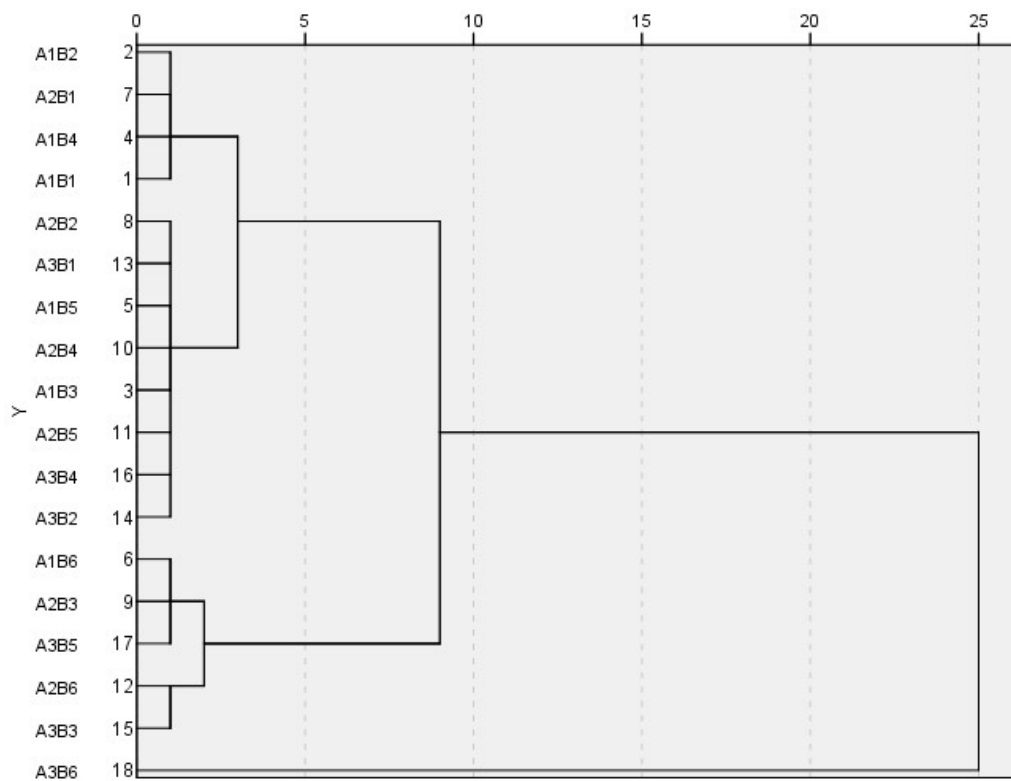


Figure 8. Dendrogram of yield using the method of intergroup connections (year of research 2018).

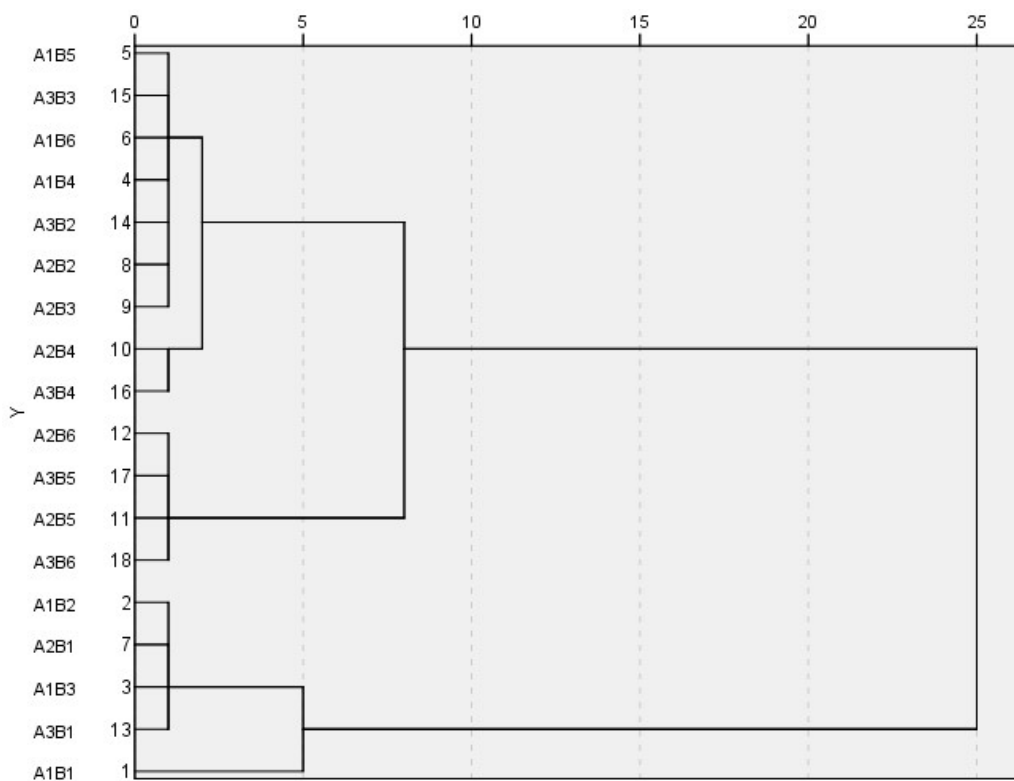


Figure 9. Dendrogram of yield using the method of intergroup connections (year of research 2019).

In 2020, due to extremely unfavourable weather conditions because of lack of moisture, the lowest level of yield was formed. According to the results of cluster analysis, the following clusters and yield levels were identified (Fig. 10):

I cluster: A2B5, A3B4, A3B2, A2B4, A3B1, A2B2 – 1.45–1.53 t ha⁻¹;

II cluster: A1B2, A1B4, A2B1 - 1.36–1.42 t ha⁻¹;

III cluster: A1B3, A1B5, A3B5 - 1.60–1.65 t ha⁻¹;

IV cluster: A2B6, A3B3, A3B6, A2B3 - 1.84–1.90 t ha⁻¹.

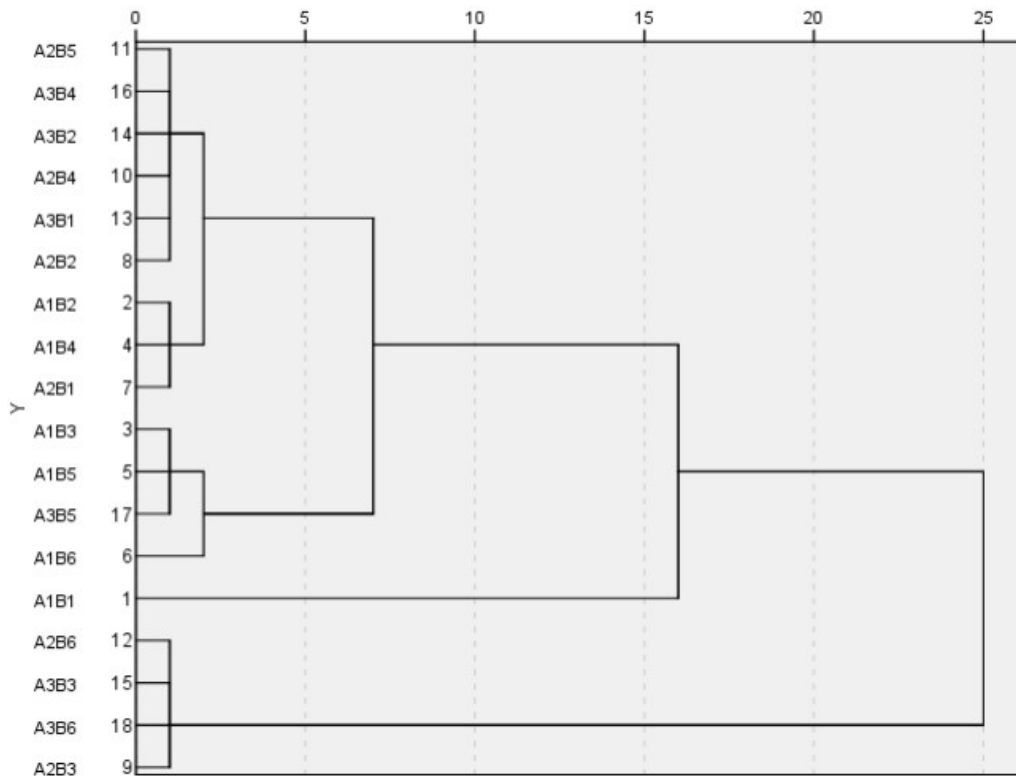


Figure 10. Dendrogram of yield using the method of intergroup connections (year of research 2020).

Analysis of the formed clusters shows that the highest level of yield (cluster IV) is also formed by combined pre-sowing treatment of seeds by inoculant and Iodis concentrate (A2) or Avatar (A3), and fertilizing by Nano Chelate fertilizer Super Micro Plus (B3) or combination of Nano Chelate fertilizer Super Micro Plus + Avatar (B6).

Feeding of soybean crops by nanofertilizer in combination with the drug Avatar with using for sowing seeds, inoculated and treated by the Avatar, and in adverse weather conditions and by Jodis concentrate, provide a significant increase in yield, which is observed in groups of formed clusters.

CONCLUSIONS

Based on the research, it was found that the use of nano preparations Avatar, Jodis-concentrate, and Super Micro Plus for seed treatment and fertilizing intensified the formation of the leaf surface area and the activity of the symbiotic apparatus of soybean plants. The obtained results confirm that application of the complex of nano fertilizers

Jodis-concentrate, Avatar, and Nano Chelate fertilizer Super Micro Plus in the soybean fertilizing helped to increase the yield, which testifies to their unconditional effectiveness. The highest efficiency of nano fertilizers was shown by inoculation and seed treatment by Avatar and co-fertilization Avatar + Nano Chelate fertilizer Super Micro Plus, ensuring the formation of 52.4 thousand m² ha⁻¹ of leaf surface area of soybean varieties Horol, 69.7 pcs plant⁻¹ root nodules, 785 mg plant⁻¹ of root nodules weight and yield at the level of 2.79 t ha⁻¹.

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