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The effect of biopreparations on the adaptive properties of soybean in organic farming under hydrothermal stress

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Abstract. In the context of organic farming and hydrothermal stress, the use of biopreparations – particularly arbuscular mycorrhiza, rhizobial bacteria, and phytohormonal treatments – has gained special significance as a means of enhancing the adaptive properties of soybean. Their application helps to mitigate the negative effects of abiotic factors and maintain crop productivity. This study aimed to assess the adaptive potential of soybeans under organic farming conditions through the application of mycorrhizal, bacterial, and phytohormonal biopreparations during periods of hydrothermal stress. Based on a field study conducted in the Poltava Region, the influence of biopreparations of various origins on the biochemical, physiological-morphological, and stress-indicative parameters, as well as the productivity of the Khorol soybean variety, was analysed. The application of biopreparations significantly improved biochemical and physiological-morphological indicators, as well as the productivity of soybeans, indicating an improvement in the overall physiological condition of the plants. The highest concentrations of chlorophylls *a* and *b* were recorded in 2023 under the three-component treatment (Mycofriend with Profix and Violar), where the average increase in Chl *a* over the years of research was 42.3%, and Chl *b* was 26.7% compared with the control. The assimilative leaf area reached an average of 29.24 thousand m^2/ha, while yield was recorded at 3.23 t/ha, exceeding the control by 28.9% and 49.0% respectively. In the drought affected 2024 season, the adaptive application of biopreparations contributed to a reduction in leaf surface area loss within the range of 16.4-20.0% and yield reduction of 17.1-20.0%, compared with the control, where losses amounted to 22.1% and 24.4%, respectively. The use of biopreparations under hydrothermal stress in 2024 also resulted in an increase in proline concentration (up to 7.27 mg/g) and a decrease in MDA content (down to 7.14 mg/g), indicating reduced oxidative stress and improved osmoregulation. This effect is attributed to the synergistic action of mycorrhiza formation, nitrogen fixation, and phytohormonal activity. The practical significance of the findings lies in demonstrating the potential of integrated biological treatments to enhance the yield and stress tolerance of soybeans under the conditions of organic farming and climate-related challenges

Keywords: arbuscular mycorrhizal fungi; rhizobial bacteria; phytohormones; chlorophyll; leaf surface area; yield; proline; malondialdehyde

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INTRODUCTION

In the current context of climate instability, agricultural production is increasingly challenged by intensifying hydrothermal stress, which leads to significant disruptions in the physiological and biochemical processes of plants, ultimately reducing their productivity and resilience. The organic farming system, which relies on natural mechanisms of crop growth regulation and protection, is particularly sensitive to such changes. Against this backdrop, the search for agrobiological solutions capable of enhancing plant adaptability to adverse environmental conditions is of growing relevance. Among these solutions is the application of mycorrhizal fungi, rhizobial bacteria and phytohormonal treatments, which can synergistically support the physiological condition of plants, improve nutrient uptake efficiency and modulate stress responses. Investigating the effectiveness of such biopreparations in soybean cultivation under organic farming conditions is of considerable scientific and practical interest, especially in light of the urgent need to strengthen crop adaptability during drought-prone years.

As noted by G. Di Capua & S. Rahmstorf (2023), there is a global trend towards increasing frequency and intensity of abiotic stress factors, which adversely affect the growth, development and productivity of major agricultural crops. Under such conditions, the studies of A. Muhammad *et al.* (2024) have identified disruptions in the physiological and biochemical processes in plants, resulting in suppressed growth compared with optimal cultivation conditions. According to S. Chattaraj *et al.* (2025), this situation is causing growing concern in the agricultural sector and is driving global demand for effective, environmentally safe strategies to mitigate the negative effects of stress factors. In this context, T. Sa (2024) highlighted the ability of beneficial microorganisms to enhance plant resistance to abiotic stress, emphasising the crucial role of plant-microbiome interactions as a fundamental prerequisite for stable agricultural production in the face of climate change.

Alongside these challenges, the growing demand for environmentally friendly products, climate change, biodiversity loss and soil degradation are driving the development of organic farming as a strategy to reduce the anthropogenic pressure on agroecosystems. According to J. Sanders *et al.* (2025), organic farming can contribute to addressing current environmental and resource-related issues and is rightly regarded as a key approach to sustainable land use. Within this system, the use of biological preparations to enhance crop productivity and stress tolerance plays an especially important role. Research by T. Chaika *et al.* (2023) indicated that soybean (*Glycine max* (L.) Merr.), as a leading protein and oilseed crop, holds a central position in the structure of organic production in Ukraine, accounting for 24.7% of the EU's demand. This places Ukraine second among the countries exporting organic soybeans to the European market.

At the same time, as noted by S. Gouli *et al.* (2024), climate change and increasingly erratic moisture conditions during the growing season are critical risk factors, posing a growing threat to crop yields and the overall resilience of agriculture. This highlights the need for effective adaptive solutions to stabilise the productivity of organic soybeans under abiotic stress, as drought conditions can lead to yield losses of up to 100%. In the context of organic farming, the use of arbuscular mycorrhizal (AM) fungi, rhizobial bacteria and phytohormones represents a promising agrobiotechnological strategy, particularly in soybean cultivation. R.K. Ravi *et al.* (2024) reported that AM fungi promote crop growth by enhancing nutrient uptake and play a key role in regulating water balance and improving plant nutrition under adverse conditions. According to the studies of M.P. Inbaraj (2021) and W. Sun & M.H. Shahrajabian (2023), the combined inoculation of soybean with AM fungi and rhizobial bacteria – particularly *Bradyrhizobium japonicum* – enhances the efficiency of biological nitrogen fixation and improves the overall physiological condition of the plants.

In their study, Z. Liao *et al.* (2025) stated that phytohormones play a pivotal role in the system of adaptive mechanisms that counteract drought stress. Acting as endogenous signalling molecules, they coordinate the expression of drought-resistance genes, regulate osmotic balance, modify the architecture of both root systems and aerial plant parts, and induce the biosynthesis of protective compounds, thus enabling a comprehensive physiological adaptation to moisture deficiency. Such biological approaches align with the principles of organic farming by supporting soil fertility and reducing the need for chemical fertilisers, thereby lowering the environmental impact on agroecosystems. As a result, the ecological intensification of crop production through biopreparations – combining the functions of growth stimulators, pathogen antagonists, and stress resistance inducers – represents a promising strategy for improving crop productivity, particularly in soybean cultivation. At the same time, comprehensive studies on the synergistic interaction of arbuscular mycorrhizal fungi, rhizobial bacteria and exogenous phytohormones in soybean agrocenoses under organic farming conditions remain largely unexplored. This highlights the high scientific and practical relevance of the present study, particularly in the context of intensifying hydrothermal stress and the urgent need to enhance the adaptive potential of agricultural crops.

This research aimed to assess the adaptive potential of organically grown soybeans under the influence of mycorrhizal, bacterial and phytohormonal biopreparations across years with differing moisture and temperature conditions. To achieve this aim, the following objectives were pursued: to evaluate the biochemical, physiological-morphological, stress-indicator parameters and productivity of soybean concerning the application of mycorrhizal, bacterial and phytohormonal biopreparations; and to analyse the effectiveness of integrated biological stimulation of soybean stress tolerance under organic farming conditions depending on the climatic conditions of the growing season.

MATERIALS AND METHODS

Experimental research was carried out from May to August during the years 2022-2024 on trial plots located in Khudoliivka Village, Kremenchuk District, Poltava Region. The research was conducted following ethical requirements. Experimental studies on plants, including the collection of plant material, followed institutional and international guidelines. The research also complied with the standards of the Convention on Biological Diversity (1992).

The experimental site is characterised by high environmental quality due to the presence of nearby forest plantations and Lake Sudebske. The soil cover of the trial plots is represented by residual-solonetz chernozem on loess deposits, with a medium level of nitrogen and phosphorus availability and a high potassium content. Agrochemical parameters of the soil were determined in the arable layer (0-20 cm) using a Palintest SK500 multiparameter photometer (Palintest Ltd., United Kingdom). According to the analysis, the soil had the following properties: acidity (pH KCl) – 6.3 (slightly acidic reaction); humus content – 5.2%; total nitrogen – 58.6 mg/kg; available phosphorus – 78.3 mg/kg; exchangeable potassium – 138.4 mg/kg. The total area of the experimental plot was 0.3 ha, with the accounting area covering 0.1 ha.

The research material consisted of the early-maturing Ukrainian soybean variety Khorol, developed by the Soybean Research Institute LLC. Field experiments were conducted in triplicate with randomised placement of treatment variants. Standard agronomic practices for the region were applied during the cultivation process. Soybeans were sown after spring barley as a preceding crop. Sowing was carried out at optimal dates, under the climatic conditions of each research year, to a depth of 5 cm, with row spacing of 38 cm and a seeding rate of 700,000 viable seeds per hectare.

Soil cultivation techniques, adapted to the principles of organic farming, included a sequence of operations: deep autumn ploughing using a mouldboard plough, early spring harrowing with heavy harrows to conserve soil moisture, pre-sowing cultivation with a stubble cultivator, and mechanical weed control through the use of mesh harrows both before and after crop emergence, followed by two inter-row cultivations. To provide biological protection of the crops from pests, *Trichogramma* entomophages were manually released three times at a rate of 100,000-200,000 individuals per hectare, distributed across 50 locations per hectare: before sowing, 30 days after the first application, and additionally if a high level of pest infestation was observed.

The experimental design was implemented as follows: Variant 1 – control (seed and plant treatment with an equivalent volume of water); Variant 2 – seed treatment with Mycofriend®-sc (BTU-Centre, Ukraine), using a diluted solution (1:10) at a rate of 1.5 L/t of seed, applied one hour before sowing; Variant 3 – seed treatment using the dry method with Mycofriend-wp (BTU-Centre, Ukraine) at a rate of 1.5 kg/t. After 30 minutes, seeds were inoculated with Profix® (Certis Belchim, Belgium) using the dry method, 30 minutes before sowing, at a rate of 1.25 kg per 500 kg of seed; Variant 4 – dry seed treatment with Mycofriend-wp at a rate of 1.5 kg/t. During the budding to early flowering stage, plants were sprayed with a diluted solution of Violar® (Innovation Company Bioinvest-Agro LLC, Ukraine) – 100 mL diluted in 200 L of water per hectare; Variant 5 – dry seed treatment with Mycofriend-wp at a rate of 1.5 kg/t. After 30 minutes, seeds were inoculated with Profix using the dry method 30 minutes before sowing at a rate of 1.25 kg per 500 kg of seed. During the budding to early flowering stage, soybean crops were sprayed with the phytohormone complex preparation Violar at a rate of 100 mL/ha, diluted in 200 L of water.

To obtain representative data on the effectiveness of biopreparations in organic soybean cultivation, products with different mechanisms of action on plant physiological processes and various forms of symbiotic interaction were selected. Particular attention was paid to the potential synergistic effects of interactions between components when choosing the biopreparations. In Mycofriend-wp and Mycofriend-sc, synergy is achieved through the mutually reinforcing effects of mycorrhizal fungi and rhizosphere bacteria. Specifically, *Pseudomonas fluorescens* secretes metabolites that stimulate hyphal development in *Glomus sp.*, while the mycorrhizal fungi improve phosphorus availability for *Bacillus* species. The *Bradyrhizobium* strains in Profix exhibit functional complementarity due to differing colonisation dynamics of root tissues and distinct temperature optima for nitrogenase activity, ensuring stable nitrogen fixation across a wide range of agro-ecological conditions. In Violar, synergism is realised through a balanced ratio of phytohormones from various classes, enabling comprehensive regulation of growth and adaptive processes in the plant organism.

Observations of weather conditions and precipitation dynamics were carried out annually during the growing season (May-August), enabling an assessment of the impact of hydrothermal conditions on soybean performance. The degree of moisture during each month of the growing season was evaluated using the hydrothermal coefficient (HTC) developed by G.T. Selyaninov (Koval & Bräuning, 2024). Weather conditions during the 2022-2024 soybean growing seasons were characterised by significant variability in the hydrothermal regime, with a marked deterioration in cultivation conditions in 2024 (Fig. 1).

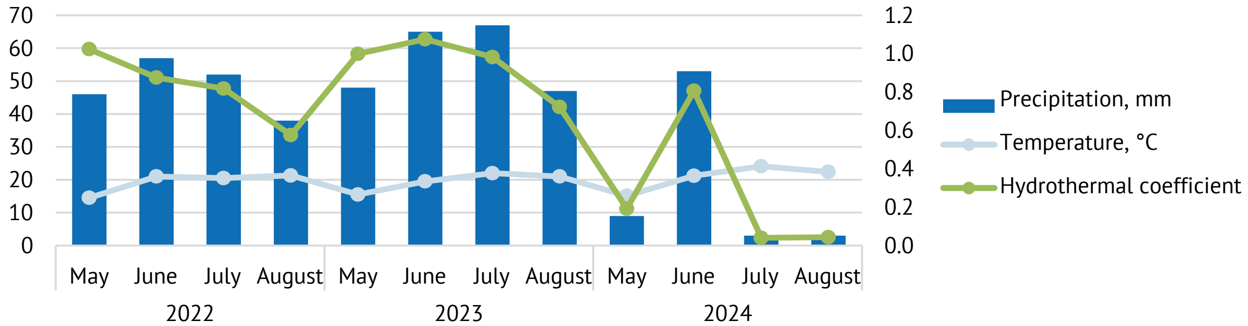

Figure 1. *Dynamics of precipitation, average monthly temperature, and hydrothermal coefficient during the soybean growing season, 2022-2024*
Source: *developed by the authors*

An analysis of HTC dynamics indicates that the 2022 and 2023 seasons provided mostly favourable conditions for soybean growth and development, with HTC values generally ranging between 0.6 and 1.1. In contrast, 2024 was marked by a critical decline in HTC to 0.0-0.2 in July and August, accompanied by a rise in average daily temperatures to 22°C-24°C and a severe precipitation deficit (2-8 mm). This combination of extremely high temperatures and exceptionally low moisture indicates the presence of pronounced hydrothermal stress during the critical phases of soybean development (flowering and pod formation). Under organic farming conditions, this stress was partially mitigated through the use of bacterial preparations from the PGPB (plant growth-promoting bacteria) group, which are capable of stimulating root system development, improving the plant water regime, and inducing physiological and biochemical mechanisms of drought resistance. This contributed to the preservation of the soybean's adaptive potential and the stabilisation of yields, even under conditions of severe hydrothermal stress.

Freshly collected plant material was used to analyse photosynthetic pigments, with pigment extraction carried out in a 96% ethanol solution. The quantitative determination of chlorophyll *a* (Chl *a*) and chlorophyll *b* (Chl *b*) was performed using direct spectrophotometry of ethanol extracts, without prior chromatography. Optical density was measured at wavelengths of 665 nm (for Chl *a*) and 649 nm (for Chl *b*) using a ULAB 108 UV spectrophotometer (ULAB, China). Pigment content in the experimental samples was calculated using the formulas proposed by A.R. Wellburn (1994). To determine the leaf assimilation area, the Easy Leaf Area (n.d.) software was used. The integral indicator of leaf surface area per unit of sown area (m^2/ha) was calculated by multiplying the average leaf area per plant by the number of plants per hectare. The concentrations of proline and malondialdehyde (MDA) in soybean leaf tissues were analysed in accordance with the methodological guidelines developed by M.K. Fatema *et al.* (2023). Soybean yield was determined by fully harvesting the accounting plots manually, following the preliminary cutting of aboveground phytomass.

The significance of the experimental data was assessed using analysis of variance (ANOVA) with the Statistica 12.0 software package (StatSoft Inc., USA, 2013). Mean values and standard deviations (SD) were calculated using Tukey's test ($P < 0.05$) with Bonferroni correction. To test statistical hypotheses regarding functional relationships between the experimental indicators, correlation analysis was carried out using Pearson correlation coefficients. The strength and direction of relationships between variables were interpreted according to the standard scale (r = ±0.1-0.3 – weak; ±0.3-0.7 – moderate; ±0.7-1.0 – strong). The statistical significance of the correlation coefficients was determined at a significance level of $P < 0.05$. Statistical analysis was performed using the Statistica 12.0 software.

RESULTS AND DISCUSSION

The selection of biological products for the experimental study was based on a comprehensive analysis of their composition, with particular attention to the presence of various functional groups of plant growth-promoting bacteria (PGPB) and the specificity of their biological action. Figure 2 presents a summarised scheme of the functional groups of microorganisms included in the selected biopreparations and the directions of their biological activity, which contribute to growth stimulation and enhanced plant resilience.

Thus, the biopreparations chosen for the study represent a representative selection of modern microbial inoculants with diverse mechanisms of action and compositional features, as well as a metabolic biopreparation based on biologically active metabolites. This selection enables a comprehensive evaluation of their effectiveness and supports the development of optimal application strategies in agricultural production. The results of the quantitative analysis of photosynthetic pigments, physiological parameters, productivity, and stress indicators for the soybean cultivar Khorol are presented in Tables 1-3.

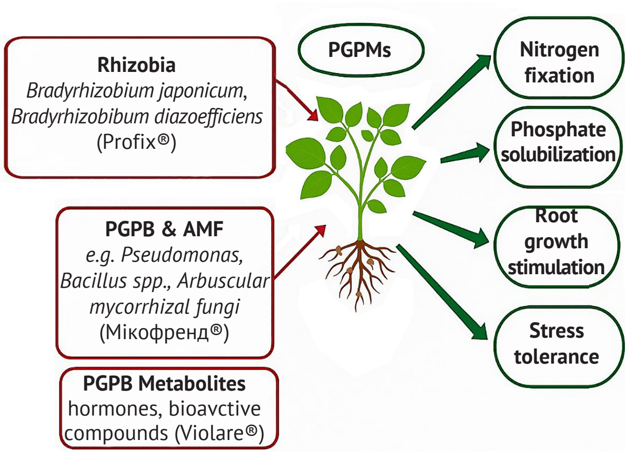

Figure 2. *Effects of PGPMs in soybean*
Source: *developed by the authors*

Table 1. *Dynamics of Chl a and Chl b content in soybean leaves depending on the method of biopreparation application, 2022-2024 (x ± SD, n = 10)*

Biopreparation application	Chl *a*, mg/g			Chl *b*, mg/g		
	2022	2023	2024	2022	2023	2024
Variant 1	2.22 ± 0.066^{a}	2.40 ± 0.070^{a}	1.78 ± 0.052^{a}	0.86 ± 0.029^{a}	1.05 ± 0.029^{a}	0.68 ± 0.018^{a}
Variant 2	2.49 ± 0.068^{b}	2.69 ± 0.077^{b}	1.99 ± 0.059^{b}	0.92 ± 0.026^{b}	1.12 ± 0.027^{b}	0.73 ± 0.017^{b}
Variant 3	2.82 ± 0.082^{c}	3.05 ± 0.088^{c}	2.26 ± 0.070^{c}	0.99 ± 0.028^{c}	1.21 ± 0.037^{c}	0.78 ± 0.023^{c}
Variant 4	2.89 ± 0.088^{c}	3.12 ± 0.086^{c}	2.31 ± 0.071^{c}	1.01 ± 0.029^{c}	1.24 ± 0.038^{c}	0.80 ± 0.021^{c}
Variant 5	3.15 ± 0.089^{d}	3.41 ± 0.100^{d}	2.53 ± 0.077^{d}	1.08 ± 0.030^{d}	1.32 ± 0.041^{d}	0.86 ± 0.029^{d}

Notes: *1. Biopreparation application: Variant 1 – control; Variant 2 – Mycofriend-sc (1.5 L/t); Variant 3 – Mycofriend-wp (1.5 kg/t) with Profix (1.25 kg/500 kg seed); Variant 4 – Mycofriend-wp (1.5 kg/t) with Violar (100 mL/ha); Variant 5 – Mycofriend-wp (1.5 kg/t) with Profix (1.25 kg/500 kg seed) and Violar (100 mL/ha). 2. Letters a, b, c and d indicate mean values that differ significantly within each row of the table (between biopreparation application variants), based on pairwise comparisons using Tukey's test (p < 0.05) with Bonferroni correction*
Source: *developed by the authors*

Based on the data presented in Table 1, a statistically significant positive effect of biopreparations on the accumulation of Chl *a* and Chl *b* in soybean leaves was established. The results of three years of observations indicated that the application of Mycofriend had a beneficial impact on the functional state of the photosynthetic apparatus compared to the control. Specifically, the average increase in Chl *a* and Chl *b* content was 12.2% and 7.0%, respectively, relative to the control. Each successive combination of biological agents in the experimental variants contributed to a further increase in pigment concentration, indicating the presence of a synergistic effect. When Mycofriend was applied in combination with Profix, the average content of Chl *a* and Chl *b* increased by 13.4% and 7.6%, respectively, compared to Mycofriend alone. A similar trend was observed in the variant combining Mycofriend with Violar, where the increase reached 15.9% for Chl *a* and 10.9% for Chl *b*.

The highest pigment levels were recorded in the variant with the three-component treatment, where the average increase in Chl *a* and Chl *b* content reached 26.8% and 18.5%, respectively, compared to the application of Mycofriend alone. These results confirm the high effectiveness of the combined action of biological agents and demonstrate a consistent trend towards increased levels of key photosynthetic pigments in soybean leaves under their influence. The dynamics of the measured indicators over the three-year study period indicate that 2023 was the most favourable year for the development of the photosynthetic apparatus. This is evidenced by the highest recorded values of Chl *a* and Chl *b* under all treatment variants (Table 1): Mycofriend – 2.69 and 1.12 mg/g; Mycofriend with Profix – 3.05 and 1.21 mg/g; Mycofriend with Violar – 3.12 and 1.24 mg/g; Mycofriend with Profix and Violar – 3.41 and 1.32 mg/g, respectively.

In contrast, a noticeable decrease in these values was observed in 2024 across all experimental variants, due to critical hydrothermal stress, which inhibited chlorophyll synthesis. However, even under these unfavourable weather conditions, the combined application of biopreparations led to a significant increase in Chl *a* and Chl *b* content compared to the control: by 11.8% and 7.4% with Mycofriend; by 27.0% and 14.7% with Mycofriend and Profix; by 29.8% and 17.6% with Mycofriend and Violar; and by 42.1% and 26.5% with Mycofriend, Profix and Violar. Thus, the combined use of biopreparations – particularly Mycofriend with Profix and Violar – ensured optimal functioning of the soybean photosynthetic apparatus, even under hydrothermal stress. Analysis of the data in Table 2 revealed clear trends in the influence of biopreparations and weather conditions on leaf surface area and yield of the Khorol soybean variety over the 2022-2024 period.

Table 2. *Dynamics of leaf surface area and soybean yield depending on the method of biopreparation application, 2022-2024 (x ± SD, n = 10)*

Biopreparation application	Leaf surface area, thousand m²/ha			Yield, t/ha		
	2022	2023	2024	2022	2023	2024
Variant 1	23.40 ± 0.688[a]	25.10 ± 0.747[a]	19.55 ± 0.571[a]	2.24 ± 0.066[a]	2.46 ± 0.070[a]	1.86 ± 0.059[a]
Variant 2	25.99 ± 0.765[b]	28.11 ± 0.824[b]	22.49 ± 0.688[b]	2.73 ± 0.077[b]	3.01 ± 0.088[b]	2.41 ± 0.070[b]
Variant 3	27.63 ± 0.813[c]	30.09 ± 0.887[c]	24.28 ± 0.718[c]	2.93 ± 0.088[c]	3.24 ± 0.095[c]	2.65 ± 0.077[c]
Variant 4	27.81 ± 0.818[c]	30.34 ± 0.895[c]	24.85 ± 0.736[c]	3.02 ± 0.088[d]	3.36 ± 0.100[d]	2.76 ± 0.082[d]
Variant 5	28.92 ± 0.854[d]	32.03 ± 0.942[d]	26.78 ± 0.788[d]	3.15 ± 0.088[d]	3.57 ± 0.095[d]	2.96 ± 0.088[d]

Notes: *1. Biopreparation application: Variant 1 – control; Variant 2 – Mycofriend-sc (1.5 L/t); Variant 3 – Mycofriend-wp (1.5 kg/t) with Profix (1.25 kg/500 kg seed); Variant 4 – Mycofriend-wp (1.5 kg/t) with Violar (100 mL/ha); Variant 5 – Mycofriend-wp (1.5 kg/t) with Profix (1.25 kg/500 kg seed) and Violar (100 mL/ha). 2. Letters a, b, c and d indicate mean values that differ significantly within each row of the table (between biopreparation application variants), based on pairwise comparisons using Tukey's test ($p < 0.05$) with Bonferroni correction*
Source: *developed by the authors*

The dynamics of changes in leaf assimilatory surface area demonstrate a significant influence of biopreparations, both in comparison with the control and with the application of Mycofriend alone, which resulted in an average increase in leaf surface area of 12.6%. The use of combined treatment schemes – specifically, Mycofriend in combination with Profix and Violar – led to an additional increase in leaf surface area of 7.1% and 8.4% on average, respectively, compared to the Mycofriendonly treatment. The highest values for assimilatory surface area were recorded in the variant involving the combined application of all three biopreparations, averaging 29.24 thousand m²/ha, which exceeded the Mycofriend-only variant by 14.5%. This confirms the positive impact of integrated biopreparation use and highlights their synergistic effect.

The positive impact of biopreparations is also clearly evident in the yield dynamics. The use of Mycofriend contributed to an average increase in soybean yield of 24.2% compared to the control. Combined application of Mycofriend with Profix and Violar resulted in an additional yield increase of 8.1% and 12.1%, respectively, relative to the Mycofriend-only treatment. The highest yield values over the three-year period were recorded in the variant involving the three-component treatment, which averaged 3.27 t/ha and exceeded the yield achieved with Mycofriend alone by 20.2%. This indicates the substantial effectiveness of their combined use and their synergistic effect. Correlation analysis confirms a direct relationship between the development of the assimilatory surface area and yield formation, and prevailing climatic conditions. In 2024, a decrease in precipitation – particularly during July and August – was associated with a reduction in both leaf area and yield. In 2022, the leaf surface area ranged from 23.4 to 28.92 thousand m²/ha depending on the treatment (with the minimum in the control and the maximum under the three-component treatment); in 2023, it ranged from 25.10 to 32.03 thousand m²/ha; and in 2024, from 19.55 to 26.39 thousand m²/ha.

The increase in leaf surface area in 2023 averaged 8.7% compared to 2022 and closely correlated with the application of biopreparations: in the control plots – 7.3%; with Mycofriend – 8.2%; with the combination of Mycofriend and Profix – 8.9%; with Mycofriend and Violar – 9.1%; and with the combined use of Mycofriend, Profix and Violar – 10.8%. Thus, the application of biopreparations of various types contributed to the activation of morphogenetic processes in the plant organism and the optimisation of the functioning of the soybean photosynthetic apparatus. This was reflected in a progressive increase in the assimilatory surface area and, consequently, a rise in the crop's potential productivity due to the intensification of photosynthetic processes.

The reduction in leaf surface area under the abiotic stress conditions of 2024, compared to the favourable conditions of 2023, varied depending on the biopreparations used: control – 22.1%; Mycofriend – 20.0%; Mycofriend with Profix – 19.3%; Mycofriend with Violar – 18.1%; and Mycofriend with Profix and Violar – 16.4%. These findings confirm the effectiveness of biopreparations in promoting leaf development even under abiotic stress, particularly when all three biopreparations of different natures were used in combination. An analysis of yield under varying weather conditions revealed that the highest levels were recorded in 2023, ranging from 2.46 to 3.57 t/ha, while the lowest were observed in 2024, ranging from 1.86 to 2.96 t/ha – an average decrease of 19.2% compared to the previous year and 10.0% compared to 2022 (Table 2). It is noteworthy that under the favourable conditions of 2023, yield increases were dependent on the application of biopreparations. Specifically, the control variant showed a 9.8% increase compared to 2022, while the use of Mycofriend resulted in a 10.3% increase, Mycofriend with Profix – 10.6%, Mycofriend with Violar – 11.3%, and Mycofriend with Profix and Violar – 13.3%. In 2024, under more stressful conditions for plant growth due to hydrothermal stress, particularly during the critical stages of soybean vegetation, yield reductions compared to 2023 also varied depending on the biopreparation treatment: control – 24.4%, Mycofriend – 20.0%, Mycofriend with Profix – 18.2%, Mycofriend with Violar – 17.9%, and Mycofriend with Profix and Violar – 17.1%.

The observed variation in soybean adaptive responses confirms the role of biopreparations as effective tools for mitigating the effects of abiotic stress and highlights the potential of combined biopreparations to enhance crop stress tolerance under increasingly unstable climatic conditions. According to the data presented in Table 3, there was a clear trend in the concentration of proline and malondialdehyde (MDA) in soybean leaves, depending on both the weather conditions of the study years and the biopreparation treatment schemes. Both indicators are important biochemical markers that reflect plant adaptive responses to stress factors, particularly soil moisture deficit.

Table 3. *Dynamics of proline and MDA content in soybean plants depending on the biopreparation application method, 2022-2024 ($x \pm SD$, $n = 10$)*

Biopreparation application	Proline, mg/g			MDA, mg/g		
	2022	2023	2024	2022	2023	2024
Variant 1	5.10 ± 0.147[a]	5.42 ± 0.159[a]	6.87 ± 0.206[a]	9.40 ± 0.279[a]	9.17 ± 0.272[a]	17.93 ± 0.530[a]
Variant 2	5.72 ± 0.165[b]	5.88 ± 0.177[b]	7.39 ± 0.217[b]	8.19 ± 0.242[b]	7.87 ± 0.236[b]	14.85 ± 0.442[b]
Variant 3	6.12 ± 0.173[c]	6.25 ± 0.186[c]	7.98 ± 0.236[c]	7.26 ± 0.217[c]	6.87 ± 0.206[c]	12.55 ± 0.371[c]
Variant 4	6.28 ± 0.185[d]	6.35 ± 0.188[d]	8.27 ± 0.247[d]	6.92 ± 0.200[d]	6.44 ± 0.191[d]	11.15 ± 0.324[d]
Variant 5	6.51 ± 0.195[e]	6.58 ± 0.202[e]	8.71 ± 0.254[e]	6.28 ± 0.193[d]	5.76 ± 0.170[d]	9.37 ± 0.276[d]

Notes: *1. Biopreparation application: Variant 1 – control; Variant 2 – Mycofriend-sc (1.5 L/t); Variant 3 – Mycofriend-wp (1.5 kg/t) with Profix (1.25 kg/500 kg seed); Variant 4 – Mycofriend-wp (1.5 kg/t) with Violar (100 mL/ha); Variant 5 – Mycofriend-wp (1.5 kg/t) with Profix (1.25 kg/500 kg seed) and Violar (100 mL/ha). 2. Letters a, b, c and d indicate mean values that differ significantly within each row of the table (between biopreparation application variants), based on pairwise comparisons using Tukey's test ($p < 0.05$) with Bonferroni correction*

Source: *developed by the authors*

Over the period from 2022 to 2024, there was a steady increase in proline concentration across all treatment variants, showing a clear dependence on hydrothermal conditions and the applied biopreparations (Table 3). For instance, in the control variant, the average proline content was 5.80 mg/g, while the use of Mycofriend resulted in 6.33 mg/g – an increase of 9.1%. When Mycofriend was applied in combination with Profix and Violar, proline levels increased by 16.9% and 20.2%, respectively, compared to Mycofriend alone. The highest proline concentration was recorded under the three-component biopreparation treatment, with an average annual value of 7.27 mg/g – 14.8% higher than in the Mycofriend-only treatment.

In the moisture-favourable conditions of 2023, the increase in proline concentration was minimal, particularly in the variants with biopreparation treatments: in the control – 6.3%; with Mycofriend – 2.8%; with the combination of Mycofriend and Profix – 2.1%; with Mycofriend and Violar – 1.1%; and with the combined application of Mycofriend, Profix, and Violar – 1.1%. Under hydrothermal stress in 2024, the highest values were recorded (ranging from 6.87 to 8.71 mg/g of fresh weight), along with a significant increase in proline concentration across all treatment variants (on average 28.6%). However, the intensity of this increase varied, reflecting the specific influence of the biopreparations applied: in the control – 26.8%; with Mycofriend – 25.7% (attributed to the protective effect of mycorrhiza); with Mycofriend and Profix – 27.7% (resulting from the activation of both symbiotic systems in response to stress); with Mycofriend and Violar – 30.2% (a synergistic effect of mycorrhiza and phytohormones in regulating osmotic potential); and with the combined application of Mycofriend, Profix, and Violar – 32.4%, which was the highest value, resulting from the integrated activation of all adaptive mechanisms.

Thus, biopreparations activate various adaptive mechanisms: mycorrhiza enhances water uptake, nitrogen fixation improves nitrogen nutrition, and phytohormones regulate metabolic processes. Together, these factors create conditions conducive to the synthesis of increased amounts of proline, an adaptive osmoprotectant, which enables plants to better withstand stress by maintaining cellular water balance. This dynamic in proline content not only demonstrates the successful activation of the plant's defence mechanisms through the use of biopreparations but also confirms their capacity to stimulate a more effective adaptive response to hydrothermal stress. In contrast, the content of malondialdehyde (MDA) (Table 3) showed an opposite trend – a decrease in average values depending on the biopreparation applied. In the control, the MDA level was 12.17 mg/g, whereas with biopreparation treatments it decreased significantly: with Mycofriend – 10.30 mg/g; with Mycofriend and Profix – 8.89 mg/g; with Mycofriend and Violar – 8.17 mg/g; and with the combined application of Mycofriend, Profix, and Violar – 7.14 mg/g.

The temporal dynamics of MDA also revealed a significant increase during the drought conditions of 2024 across all treatment variants, with an average level of 13.17 mg/g, compared with 7.61 mg/g in 2022 and 7.22 mg/g in the more favourable conditions of 2023. In the hydrothermally balanced year of 2023, a reduction in MDA concentration was observed for all treatments relative to 2022: in the control – 2.8%; with Mycofriend – 3.9%; with Mycofriend and Profix – 5.4%; with Mycofriend and Violar – 6.9%; and with the combined application of Mycofriend, Profix, and Violar – 8.3%. The stress conditions in 2024 led to a sharp increase in MDA content, although with varying intensity depending on the treatment compared to 2023: in the control – 96.2%; with Mycofriend – 88.7% (a result of the protective effect of mycorrhiza); with Mycofriend and Profix – 82.7% (reflecting enhanced antioxidant activity due to the function of both symbiotic systems); with Mycofriend and Violar – 72.4% (more effective protection of membrane structures through phytohormonal stimulation of antioxidant systems); and with the combined application of Mycofriend, Profix, and Violar – 62.7% (the highest effectiveness in protecting cellular membranes).

Thus, the biopreparations demonstrated a clear ability to reduce oxidative stress effectively through a range of complementary mechanisms: mycorrhizae improve water supply and mineral nutrition; nitrogen-fixing bacteria enhance nitrogen uptake and the synthesis of protein components of the antioxidant system; and phytohormones (particularly those in Violar) activate both enzymatic and non-enzymatic defence mechanisms. This combined action resulted in reduced lipid peroxidation intensity, as confirmed by the dynamics of MDA content, which decreased under conditions of effective adaptation. This trend corresponded with proline levels and indicated the formation of more efficient antioxidant systems, contributing overall to improved soybean resilience under hydrothermal stress.

To enable deeper analysis of the results and to visualise the variability of the experimental indicators, box plot diagrams were constructed based on statistical data from Tables 1-3 (Figs. 3-5). These plots display medians, interquartile ranges, and extreme values for each treatment variant, offering a multidimensional view of the experimental results and their statistical significance. This graphical format allows for a clear assessment of the distribution of the measured parameters and supports the evaluation of the effectiveness of different biopreparation combinations in relation to key physiological and biochemical indicators as well as crop performance. Such an approach enhances data interpretation and helps reveal patterns that may remain unnoticed in tabular presentation alone.

Analysis of Figure 3(a) revealed a clear trend towards increased Chl *a* content in soybean leaves under the influence of biopreparations. The control variant was characterised by the lowest Chl *a* content (median – 2.2 mg/g) and a narrow range of variation (1.73-2.47 mg/g). Mono-inoculation with the mycorrhizal preparation (variant 2) resulted in a moderate increase in Chl *a* concentration (median – 2.5 mg/g), accompanied by a broader fluctuation range. A marked increase in Chl *a* content was observed with the combined application of mycorrhiza and the rhizobial preparation (variant 3) and with the phytohormonal preparation (variant 4), where the median values reached 2.8 and 2.9 mg/g, respectively. The highest accumulation of Chl *a* was recorded in the triple combination treatment (variant 5), where the median reached 3.2 mg/g and the upper limit exceeded 3.51 mg/g – 26.7% higher compared with variant 2. It is worth noting that the integrated application of the biopreparations not only contributed to an increase in average values but also resulted in a broader variation range. This indicates enhanced adaptive plasticity of the plant photosynthetic apparatus under combined biological treatment and a greater capacity to respond dynamically to environmental factors.

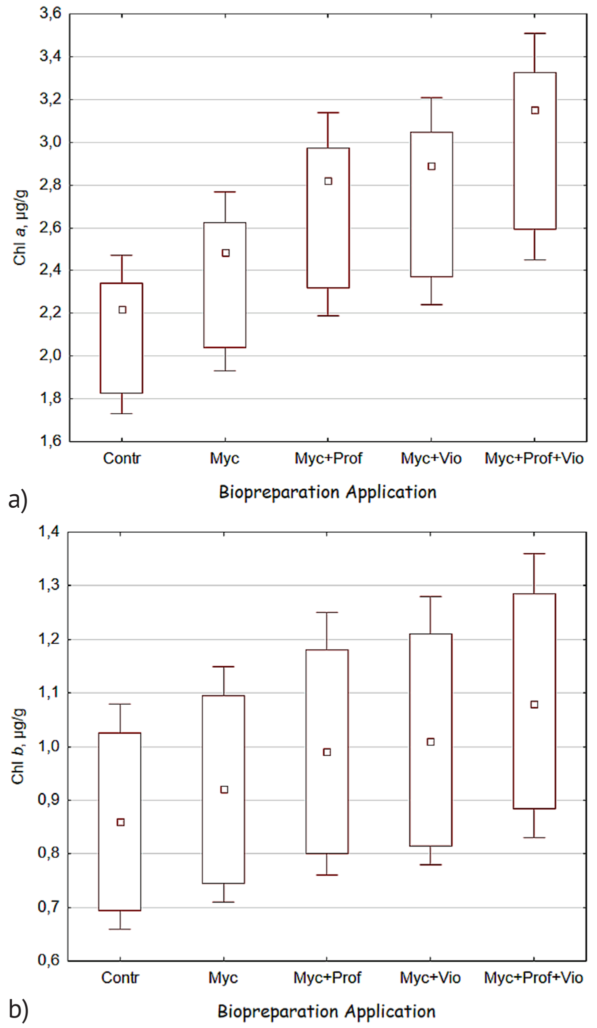

Figure 3. *Effect of biopreparation application on the content of Chl a (a) and Chl b (b) in soybean plants*
Notes: *1) treatment variants: Contr – control (1); Myc – Mycofriend (2); Myc+Prof – Mycofriend with Profix (3); Myc+Vio – Mycofriend with Violar (4); Myc+Prof+Vio – Mycofriend with Profix and Violar (5); 2) the content of Chl a and Chl b in soybean leaves was measured at the flowering stage*
Source: *developed by the authors*

Analysis of Figure 3(b), which illustrates the Chl *b* content in soybean leaves under various biopreparation combinations, reveals a distinct pattern of increasing pigment concentration corresponding to the complexity of the biogenic composition. The control variant exhibited the lowest Chl *b* content (median – 0.86 mg/g) with variability ranging from 0.66 to 1.08 mg/g. Monoinoculation with the mycorrhizal preparation (variant 2) led to a moderate increase in Chl *b* concentration (median – 0.92 mg/g). Increasing the complexity of the treatment by combining mycorrhiza with the rhizobial inoculant (variant 3) or with the phytohormonal preparation (variant 4) resulted in a further rise in pigment activity (medians – 0.99 and 1.01 mg/g, respectively), accompanied by a broader range of variation. The highest accumulation of Chl *b* was observed in the triple biopreparation combination (variant 5), where the median reached 1.08 mg/g and maximum values reached 1.36 mg/g – 18.3% higher compared with variant 2. Notably, this variant also displayed an expanded interquartile range, indicating increased variability in plant response and a possible differentiation in the expression of the synergistic effect under microecological heterogeneity.

Analysis of Figure 4(a), which presents the dynamics of soybean leaf surface area under various biological treatment combinations, reveals clear patterns of variation in this parameter. The control variant exhibited the smallest assimilative surface area (median – 23.4 thousand m^2/ha; range – 18,826.0 thousand m^2/ha), indicating relatively high variability in the experimental data. Application of the mycorrhizal preparation (variant 2) significantly stimulated leaf development (median – 26.0 thousand m^2/ha), exceeding the control by 11.1%, thereby confirming the positive effect of arbuscular mycorrhiza on the formation of the photosynthetic apparatus. Combining mycorrhiza with rhizobial bacteria (variant 3) led to a further increase in leaf surface area (median – 27.6 thousand m^2/ha), while a similar trend was observed in the variant combined with the phytohormonal preparation (variant 4), where the median reached 27.8 thousand m^2/ha. The strongest stimulatory effect was recorded in the triple combination treatment (variant 5), with a median of 28.9 thousand m^2/ha and maximum values reaching 33 thousand m^2/ha, representing a 14.0% increase compared with variant 2.

Notably, the increase in the number of biological agents in the treatment combinations is accompanied by both a rise in average values and an increase in the variability of the experimental indicator. This suggests a complex interaction between the components of the biopreparations and a heterogeneous response among plants of the same cultivar. At the same time, variants 3 and 5 show a tendency towards a narrowing of the interquartile range, indicating greater stability and predictability of the effect when these combinations are used. Additionally, a general trend towards an expansion of the minimum-maximum range (whiskers) of the box plots is observed with increasing treatment complexity, further highlighting the positive influence of bioagents on the parameters of the soybean photosynthetic apparatus.

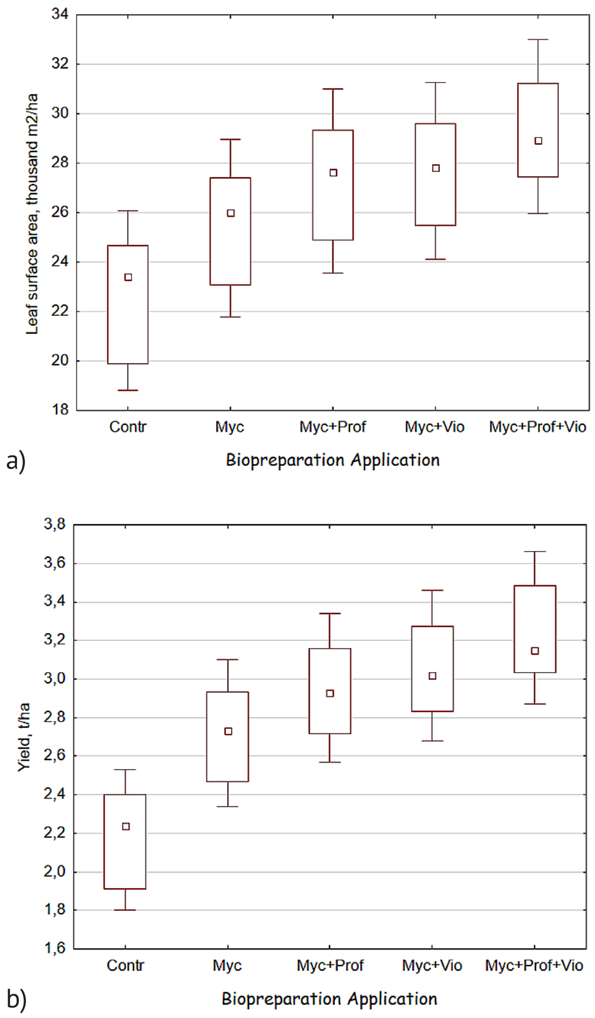

Figure 4. *Effect of biopreparation application on leaf surface area (a) and yield (b) of soybean plants*
Notes: *1) treatment variants: Contr – control (1); Myc – Mycofriend (2); Myc+Prof – Mycofriend with Profix (3); Myc+Vio – Mycofriend with Violar (4); Myc+Prof+Vio – Mycofriend with Profix and Violar (5). 2) the content of the leaf surface area was measured at the flowering stage*
Source: *developed by the authors*

The analysis of the box plot diagram presented in Figure 4(b) illustrates the effect of different biopreparation combinations on soybean yield. The control variant is characterised by the lowest productivity, with a median of approximately 2.2 t/ha and a wide range of variation (from 1.80 to 2.53 t/ha), indicating unstable yield performance in untreated plants. Application of the mycorrhizal preparation (variant 2) results in a significant yield increase, with the median reaching 2.7 t/ha – 21.9% higher than the control – highlighting the crucial role of arbuscular mycorrhiza in enhancing crop productivity. The interquartile range remains moderate, but there is a general shift of the values towards higher levels. The binary combination of mycorrhiza with rhizobial bacteria (variant 3) leads to a further increase in yield, with a median of approximately 2.9 t/ha – 7.3% higher than variant 2 – and an extension of the upper range to 3.34 t/ha. This confirms the synergistic effect of the symbiotic partnership between mycorrhizal fungi and nitrogen-fixing bacteria. The interquartile range is relatively wide, indicating some variability in plant response to this treatment.

The combination of mycorrhiza with the phytohormonal preparation (variant 4) shows an even more pronounced positive effect on soybean productivity, with a median value of approximately 3.0 t/ha – 10.6% higher than variant 2. The interquartile range narrows slightly compared to variant 3, suggesting a more uniform plant response. The highest yield values are recorded with the combined application of all three components (variant 5), where the median reaches approximately 3.2 t/ha, representing a 17.2% increase over variant 2. The upper quartile reaches 3.66 t/ha – the highest value among all treatment variants.

A clear progressive trend can be observed in the increase of both median values and minimum and maximum yield indicators with each additional component of the biopreparations. This reflects the synergistic effect of their combined application on crop productivity. The minimum values (lower extremes) also show consistent growth – from 1.80 t/ha in the control to 2.87 t/ha in variant 5 – confirming the stability of the positive effect of biopreparations even under less favourable conditions. The maximum values (upper extremes) rise from 2.53 t/ha in the control to 3.66 t/ha in variant 5, indicating a high potential for yield improvement when biopreparations are applied in combination.

By integrating data from Tables 1-3 and Figure 3, it can be concluded that the increase in yield results from the overall positive influence of the biopreparations on the physiological condition of soybean plants – particularly the increase in Chl *a* and Chl *b* content, expansion of leaf surface area, enhanced proline synthesis, and reduction in oxidative stress (as indicated by MDA content). The box plot in Figure 5(a) illustrates the changes in proline levels in soybean plants under the influence of different biopreparation combinations. In the control variant, the proline content is relatively low, with a median of 5.4 mg/g and a moderate range of variation (~5.0 to 7.1 mg/g), indicating some variability within the control group. The application of the mycorrhizal preparation (variant 2) contributes to an increase in proline content, with a median of approximately 6.0 mg/g, which is 20.0% higher than the control. The interquartile range is slightly narrower than in the control, suggesting reduced variability.

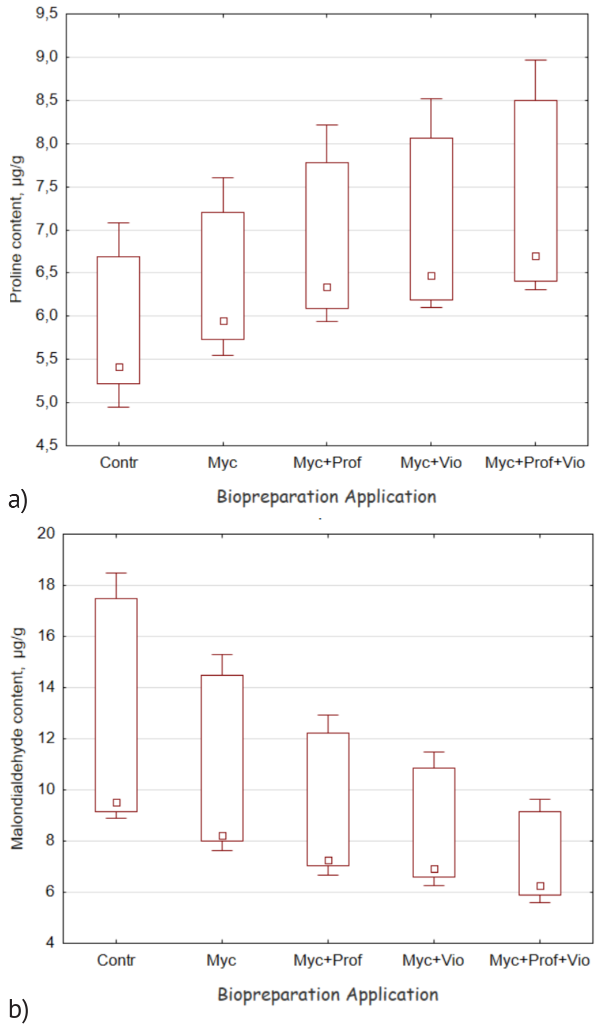

Figure 5. *Effect of biopreparation application on proline (a) and MDA (b) content in soybean plants*

Notes: *treatment variants: Contr – control (1); Myc – Mycofriend (2); Myc+Prof – Mycofriend with Profix (3); Myc+Vio – Mycofriend with Violar (4); Myc+Prof+Vio – Mycofriend with Profix and Violar (5)*

Source: *developed by the authors*

The combined application of mycorrhiza with rhizobial bacteria (variant 3) results in a further increase in proline concentration, with a median of approximately 6.3 mg/g and an extended variation range reaching up to 8.2 mg/g. This indicates the activation of adaptive mechanisms in plants, along with considerable variability in responses. Even more pronounced proline accumulation is observed when mycorrhiza is used in combination with the phytohormonal preparation (variant 4), where the median value reaches 6.5 mg/g – 8.3% higher than in variant 2. At the same time, the interquartile range narrows compared to variant 3, suggesting a more uniform physiological response among plants. The highest proline accumulation is recorded in variant 5, where all three agents – mycorrhizal preparation, rhizobial bacteria, and phytohormonal supplement – are applied together. The median reaches approximately 6.9 mg/g, which is 15.0% higher than in variant 2 and 27.8% higher compared to the control.

The upper quartile rises to 8.6 mg/g, while the lower extreme increases to approximately 6.0 mg/g, indicating not only an overall enhancement of the adaptive response but also its stability across all treated plants. The overall trend of increased proline content in the variants with biopreparations confirms that the response to hydrothermal stress largely depends on the degree of involvement of symbiotic interactions and the bioregulatory activity of the applied agents. The accumulation of proline, as one of the key osmoprotectants and stabilisers of cellular metabolism, is regarded as an indicator of enhanced plant tolerance to adverse environmental conditions, which may also indirectly influence yield.

The analysis of the box plot diagram in Figure 5(b) reveals clear patterns in the changes in MDA content in soybean plants under the influence of various biopreparation combinations. The control variant is characterised by the highest MDA levels, with a median of approximately 9.5 mg/g and a wide range of variation (8.89-18.47 mg/g), indicating intense lipid peroxidation and the development of oxidative stress in untreated plants. The application of the mycorrhizal preparation (variant 2) significantly reduces MDA content (median ~8.2 mg/g, 13.7% lower than the control) and decreases indicator variability, pointing to the activation of the antioxidant defence system under the influence of mycorrhizal fungi. Although the interquartile range (7.63-15.3 mg/g) remains fairly wide, it is less pronounced compared to the control. The binary combination of mycorrhiza and rhizobial bacteria (variant 3) results in a further reduction in MDA content to a median of approximately 7.3 mg/g, demonstrating the enhanced protective effect of the symbiotic partnership between the two microbial groups. The interquartile range narrows (6.66-12.93 mg/g).

An even more pronounced reduction in MDA levels is achieved with the combination of mycorrhiza and the phytohormonal preparation (variant 4), where the median value reaches approximately 6.9 mg/g – 15.9% lower than in variant 2. This effect is attributed to the stimulation of endogenous antioxidant defence mechanisms by phytohormones. The interquartile range is narrower still, at 6.25-11.48 mg/g. The lowest MDA content is recorded in the triple combination of biopreparations (variant 5), with a median value of approximately 6.3 mg/g – 23.2% lower than in variant 2. This variant also exhibits the narrowest interquartile range (5.59-9.65 mg/g), indicating a stable anti-stress effect resulting from the integrated application of biopreparations. MDA is one of the primary products of lipid peroxidation and is widely used as a marker of oxidative stress in plant tissues. A reduction in its content reflects an increase in antioxidant potential and overall plant stress tolerance. The data obtained confirm that the combined use of biopreparations significantly enhances the ability of plants to withstand stress factors, particularly when mycorrhiza, rhizobial bacteria, and phytohormonal preparations are applied together.

To validate the observed trends and determine the functional relationships between the indicators, a correlation analysis was conducted to provide a deeper insight into the mechanisms of biopreparation action. A strong positive correlation between chlorophyll content (*a* and *b*) and yield ($R^2 = 0.897$, $r = 0.947$, $p < 0.05$) confirms the crucial role of photosynthetic activity in determining plant productivity. A high positive correlation between leaf surface area and chlorophyll content ($R^2 = 0.956$, $r = 0.978$, $p < 0.05$) indicates the interdependence of both quantitative and qualitative characteristics of the photosynthetic apparatus.

In addition, a statistically significant positive correlation was identified between leaf surface area and yield ($R^2 = 0.949$, $r = 0.974$, $p < 0.05$), further confirming the pivotal role of a well-developed photosynthetic apparatus in ensuring high plant productivity. Under the influence of complex biopreparations, a notable increase in the correlation between photosynthetic parameters and yield was observed, indicating an optimisation of internal functional relationships within the plant. Statistical analysis of the experimental data confirmed a statistically significant negative correlation between MDA content and soybean yield ($R^2 = 0.661$; $r = -0.813$; $p < 0.05$), suggesting a considerable inhibitory effect of intensified lipid peroxidation on crop productivity. Moreover, significant negative correlations were found between MDA content and key photosynthetic indicators: Chl *a* ($r = -0.882$; $p < 0.05$), Chl *b* ($r = -0.832$; $p < 0.05$), and leaf surface area ($r = -0.878$; $p < 0.05$). All identified correlations are strong ($|r| > 0.7$) and statistically significant ($p < 0.05$), confirming a close relationship between the intensity of oxidative processes and the functional state of the photosynthetic apparatus.

The obtained results indicate that oxidative stress, as reflected by MDA accumulation, exerts a pronounced negative effect on the formation of structural components of the photosynthetic system and on chlorophyll biosynthesis, which in turn reduces plant productivity. This confirms the critical role of antioxidant protection in ensuring effective photosynthetic activity and in determining yield outcomes. The established correlations may be interpreted as evidence of the synergistic action of biopreparation components on key physiological processes, leading to the development of a balanced donor-acceptor system within the plant and, consequently, to an increase in its productive potential. Such functional relationships are of fundamental importance for understanding the mechanisms underlying crop productivity and may inform the development of strategies for optimising cultivation practices. Thus, under organic farming conditions, achieving high soybean yields remains particularly challenging – even under favourable soil conditions – due to the impact of hydrothermal stress.

To promote the development of resilient agroecosystems for leguminous crops, particularly soybeans, the application of seed treatments with inoculants, arbuscular mycorrhizal (AM) fungi, biostimulants, and hormonal agents is considered highly relevant. This is supported by the findings of M. Nadeem *et al.* (2019) and R. Zia *et al.* (2021). In the present study, pre-sowing application of biopreparations had a positive effect on the physiological and biochemical parameters of the plants, notably contributing to an increase in chlorophyll content, which stimulated the photochemical activity of chloroplasts and enhanced photosynthetic efficiency. Specifically, the use of Mycofriend, Profix, and Violar, in various combinations, resulted in an average increase in Chl *a* and Chl *b* contents by 12.2%-15.9% and 7.0%-10.9%, respectively, compared with the control. This rise in photosynthetic pigment levels was accompanied by an expansion of the leaf surface area by 12.6%-28.9%, reaching 25.53-29.24 thousand m^2/ha, along with a reduction in the adverse effects of hydrothermal stress by 2.1%-5.7% relative to the control. These findings are consistent with those of A. Radzykhovskyi (2019), where the combined application of the biopreparations Rizoline, Rizosave, and Mycofriend led to an increase in leaf area by 5.2%-8.5%. A strong positive correlation was found between chlorophyll content ($r = 0.947$), leaf surface area ($r = 0.974$), and yield level. The yield increase varied depending on the specific biological treatment, ranging from 24.2% to 47.5% (2.72-3.23 t/ha compared with 2.19 t/ha in the control). The relationship between seed treatment and increased soybean biological productivity is also supported by the articles of S. Zimmer *et al.* (2016), M. Leggett *et al.* (2017), and M. Serafin-Andrzejewska *et al.* (2024).

As confirmed by the study conducted by L.B. Martinez-Garcia *et al*. (2013), the effectiveness of mycorrhizal application under various agroecological conditions contributed to yield increases of 15%-40% for soybeans, 20%-70% for maize, 15%-30% for cereal crops, and up to 200% for vegetables. These effects are attributed to the genotypic characteristics of the crops, cultivation conditions, and the efficiency of symbiotic interactions. Similar findings were reported by A. Radzykhovskyi (2019), where the combined use of the biopreparations Rizoline, Rizosave, and Mycofriend resulted in an increase in soybean yield to 4.18-4.23 t/ha, representing a 25.5-27.0% improvement compared with the control. These data confirm the existence of synergy between inoculants and mycorrhizal agents, expressed through enhanced nutrient uptake and increased stress tolerance within agroecosystems.

According to N.A. Milošević *et al*. (2012), soil microorganisms improve plant drought resistance by producing polysaccharides that enhance soil structure, as well as biologically active substances (indoleacetic acid, deaminase, and proline), which support water regime regulation via fungal mycelium. M. Ashraf & M.R. Foolad (2007) also highlight the protective role of proline under water stress conditions. The study confirms an increase in proline concentration during the droughtaffected year of 2024, ranging from 25.7% to 32.4%, depending on the biopreparations applied. These findings are consistent with the results of X. Yang *et al*. (2021) demonstrated that, under drought conditions, the accumulation of osmotically active compounds is positively correlated with plant resilience. According to S.A. Mona *et al*. (2017), inoculation promotes proline accumulation under both normal and stress conditions.

Research by K. Bogati & M. Walczak (2022) indicates that arbuscular mycorrhizal (AM) fungi can reduce MDA content by 32% under moderate drought. The present results support this trend, showing a reduction in MDA levels across treatment variants by 15.4% to 47.5%, with the highest effect observed under triple treatment. The findings demonstrate that, under organic farming conditions and increasing hydrothermal stress, the most effective strategy is the combined application of biopreparations, including rhizobial bacteria, AM fungi, and biostimulants. Their synergistic interaction ensures the development of resilient soybean agrocenoses with high yield potential, as confirmed by experimental data and aligned with findings from other researchers.

CONCLUSIONS

The research findings indicate that the intensification of climate change and the increasing frequency of extreme weather events necessitate a revision of conventional agronomic practices, with the integration of mandatory anti-stress protection measures. Notably high effectiveness in enhancing the adaptive potential of soybeans was observed with the use of the combined formulation of Mycofriend, Profix, and Violar. The application of these biopreparations led to an average increase in Chl *a* and Chl *b* content in soybean leaves of 12.2%-42.2% and 7.0%-26.7%, respectively, compared to the control, with the greatest effect achieved under simultaneous application of all three treatments. Importantly, biopreparation treatment maintained elevated chlorophyll levels even under the hydrothermal stress conditions of 2024. A strong positive correlation was established between the content of Chl *a* and Chl *b* and both leaf surface area ($r = 0.978$) and yield ($r = 0.947$), demonstrating the interdependence between quantitative and qualitative parameters of the photosynthetic system and confirming the critical role of photosynthetic activity in determining plant productivity.

It has been demonstrated that the leaf surface area of soybean plants responds positively to biopreparation treatment, increasing by 12.6%-28.9% compared to the control, with the greatest effect observed under combined application of all biopreparations (29.24 thousand m^2/ha). The use of biopreparations significantly mitigated the effects of hydrothermal stress in 2024, as the reduction in leaf surface area compared to the favourable conditions of 2023 was as follows: control – 22.1%; Mycofriend – 20.0%; Mycofriend with Profix – 19.3%; Mycofriend with Violar – 18.1%; Mycofriend with Profix and Violar – 16.4%. These findings confirm the effectiveness of biopreparations in stimulating the development of the leaf apparatus even under hydrothermal stress, particularly when biopreparations of different origins are applied in combination. A notable positive effect of biopreparations on soybean yield was also recorded, with an average increase of 24.2%-47.5% compared to the control, reaching a maximum of 3.23 t/ha under the combined treatment. In 2024, under more stressful conditions for plant growth due to hydrothermal stress, particularly during the critical stages of soybean vegetation, yield reductions compared to 2023 also varied depending on the biopreparation treatment: control – 24.4%, Mycofriend – 20.0%, Mycofriend with Profix – 18.2%, Mycofriend with Violar – 17.9%, and Mycofriend with Profix and Violar – 17.1%.

It was found that proline accumulation in soybean leaves during the experimental period showed a clear dependence on the hydrothermal regime and the application of biopreparations. Their use resulted in an average increase in proline levels of 9.1%-25.3% compared to the control, and under hydrothermal stress conditions, by 7.6%-32.4% compared to 2023. This indicates the successful activation of the plants' defence mechanisms in response to biopreparations and confirms their ability to stimulate a more effective adaptive response to hydrothermal stress. It was established that the lowest concentrations of MDA were recorded following the application of biopreparations (7.14-10.3 mg/g), whereas the highest level was observed in the control (12.17 mg/g). The stressful conditions of 2024 caused a significant increase in MDA content compared to 2023, although the intensity varied depending on the treatment: control – 96.2%; Mycofriend – 88.7%; Mycofriend with Profix – 82.7%; Mycofriend with Violar – 72.4%; Mycofriend with Profix and Violar – 62.7%.

A statistically significant negative correlation was found between MDA content and soybean yield ($r = -0.81$), Chl *a* content ($r = -0.882$), Chl *b* content ($r = -0.832$), and leaf surface area ($r = -0.878$). Thus, the application of biopreparations – particularly the combination of all three components – contributes to reducing MDA accumulation and enhancing the antioxidant defence of plants, thereby supporting the efficiency of photosynthetic activity and the formation of high yields. Further research should focus on assessing the impact of biopreparations on the hormonal status of plants, which will provide a deeper understanding of the mechanisms underlying soybean stress tolerance and help identify the most effective combination of biological agents for minimising the effects of climate change on agroecosystem productivity under organic farming conditions.

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CONFLICT OF INTEREST

None.

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Вплив біопрепаратів на адаптивні властивості сої в органічному землеробстві в умовах гідротермічного стресу

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Анотація. В умовах органічного землеробства та гідротермічного стресу особливого значення набуває використання біологічних препаратів, зокрема арбускулярної мікоризи, ризобіальних бактерій і фітогормональних препаратів, як інструментів підвищення адаптивних властивостей сої. Їх застосування сприяє пом'якшенню негативного впливу абіотичних чинників і підтриманню продуктивності культури. Метою дослідження було провести оцінку адаптивного потенціалу сої в умовах органічного землеробства за дії мікоризних, бактеріальних і фітогормональних біопрепаратів за гідротермічного стресу. На основі польового дослідження в умовах Полтавської області досліджено вплив біопрепаратів різної природи на біохімічні, фізіолого-морфологічні, стрес-індикаторні показники та продуктивність сої сорту Хорол. Застосування біопрепаратів достовірно підвищувало біохімічні, фізіолого-морфологічні показники та продуктивність сої, що свідчить про покращення загального фізіологічного стану рослин. Найвищі концентрації хлорофілів *a* і *b* спостерігалися за умов трикомпонентної обробки (Мікофренд із Profix і Віолар) у 2023 році, де приріст Chl *a* становив у середньому за роки дослідження 42,3 %, а Chl *b* – 26,7 % порівняно з контролем. Асиміляційна поверхня листків досягала середнього значення 29,24 тис. м²/га, а врожайність – 3,23 т/га, що перевищувало контроль на 28,9 % і 49,0 % відповідно. У посушливому 2024 році завдяки адаптогенному застосуванню біопрепаратів відбулось зменшення втрат площі листкової поверхні в межах 16,4-20,0 % і врожайності – 17,1-20,0 %, тоді як на контролі втрати становили 22,1 % і 24,4 % відповідно. Використання біопрепаратів за гідротермічного стресу 2024 року призвело до зростання концентрації проліну (до 7,27 мг/г) та зниженням вмісту MDA (до 7,14 мг/г), що свідчить про зменшення оксидативного стресу й покращення осморегуляції завдяки синергії мікоризоутворення, азотфіксації та дії фітогормонів. Практичне значення отриманих результатів полягає у розкритті потенціалу комплексної біологічної обробки для підвищення врожайності та стресостійкості сої в умовах органічного землеробства й кліматичних викликів

Ключові слова: арбускулярні мікоризні гриби; ризобіальні бактерії; фітогормони; хлорофіл; площа листкової поверхні; врожайність; пролін; малоновий діальдегід