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Effect of Mineral Fertilization and Seed Inoculation with Microbial Preparation on Seed and Protein Yield of Pea (*Pisum sativum* L.)

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Abstract: The aim of this study was to determine the effects of different NPK rates and N application methods and seed inoculation with a microbial preparation on selected elements of plant growth and the productivity parameters seed yield, protein content in seeds and the yield of protein. The research hypothesis suggested that seed inoculation and a split rate of N application with an optimal supply of plants with PK could improve the nutritional status and increase the efficiency of nutrient use in peas. The studies included two factors: the application of NPK at doses of $N_0P_0K_0$ (control), $N_{15}P_{15}K_{15}$ (pre-sowing), $N_{15}P_{30}K_{30}$ + N_{15} (pre-sowing + N_{15} at BBCH 22–23), $N_{30}P_{30}K_{30}$ (pre-sowing), $N_{30}P_{45}K_{45}$ + N_{15} (pre-sowing + N_{15} at BBCH 22–23) and $N_{45}P_{45}K_{45}$ (pre-sowing), and seed inoculation with the microbial preparation Rhizogumin. The results of the study showed significant effects of seed inoculation and mineral fertilization on pea plant growth and the productivity parameters seed yield, protein content and protein yield. It was concluded that among the studied combinations, seed inoculation and the application of mineral fertilizers with fractional nitrogen fertilization with $N_{30}P_{45}K_{45}$ + N_{15} were the most effective. This combination significantly increased seed yield, protein content and protein yield compared to the control treatment (by 26.2%, 11.1% and 43,5%, respectively).

Keywords: legumes; Rhizogumin; NPK; protein content in seeds

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1. Introduction

The problem of deficiency in plant protein resources for the production of feed and high-quality food, as well as combating the so-called “hidden hunger” caused by insufficient micronutrient intake, is becoming increasingly serious. Stabilizing the production of legumes, among which peas occupy a prominent place, can play an important role in solving this problem. The unquestionable advantage of this species in the food system is its high nutritional value, due to the presence of a large amount of protein with health-promoting properties [1], minerals, vitamins, carbohydrates and polyphenols, which exhibit a variety of biological activities [2]. The regular consumption of pea seeds can provide an adequate protein resource for people living in economically unstable regions, and also have additional health benefits for improving their health [3]. Therefore, it seems that interest in this crop as a valuable source of protein will be increased [4]. As a protein resource with balanced amino acid composition, peas are also used in the production of high-quality feed for livestock and poultry [4].

Peas are the most widely cultivated crop in the world, and the main producers of commercial peas are Russia, Canada, China, India and the United States [5]. The world’s pea acreage is about 7.04 million hectares, and pea grain production is at 10.0 million tons

[6]. The average yield of peas is about 1.5 t ha^{-1}, with an increase in the average value of this indicator in the most developed countries to 4.0–5.0 t ha^{-1} [7].

One of the problems in pea cultivation is the uneven supply of plants with nutrients such as nitrogen (N), which plays an important role in the development of the vegetative parts of plants, primarily leaves, which, in the process of photosynthesis, produce assimilates for the formation of plant biomass. Plants utilize N both from the biological fixation process as well as from the soil, taking priority to the first one, that occurs in an organically reduced form and is more available for metabolic processes [8]. Due to the rising interest in using biological methods for improving soil fertility and thus increasing the productivity of plants, seed inoculation with biological preparations based on nitrogen-fixing microorganisms has received particular attention in legume cultivation in recent years [9]. The inoculation of legume seeds with *Rhizobium* spp. not only improves nitrogen-fixing conditions, but also helps stimulate plant growth, increases their resistance to biotic stress factors [10] and has a positive effect on plant morphological traits, seed yield and the quality of seeds [11].

Pea plants are usually able to fully cover their N requirements from biological fixation, but during the initial stages of growth and development, before establishing symbiosis with rhizobia, and during nodule formation, they may require some mineral N from the soil [12,13]. Thus, the application of a starter dose of mineral N can ensure proper nutrition for plants before effective symbiosis begins, therefore enabling them to create an assimilation surface and produce biomass. Previous studies on peas have shown that nodule formation on roots begins at least eleven days after germination [14]. The applied dose of N and the timing of its application are important in this case [15].

Phosphorus (P) is required for the proper functioning of plants. An adequate P supply allows legumes to develop the root system well and provide optimal conditions for the symbiotic N fixation process to take place [16]. The physiological role of P is also to increase the photosynthetic efficiency of plants, which, in turn, has a positive effect on the morphological characteristics and size of plants' structural elements (e.g., the number of branches) and yield (e.g., the number of seeds per plant), thus increasing the overall seed yield of peas [17].

Potassium (K) plays an important role in the uptake of N and P by plants, stimulating vegetative growth and increasing the efficiency of photosynthesis. This is reflected in an increase in the proportion of solar energy absorbed by plants and an increase in the formation of assimilates under conditions of good K supply. The uptake of minerals by plants is interrelated. For example, K uptake by the root system depends on N availability [18]. On the other hand, the efficiency of N and P uptake by plants is strongly related to K availability in the soil [19]. A balanced plant nutrient supply can therefore improve conditions for the formation of pea productivity and seed quality, with the effectiveness of mineral fertilization varying according to the soil and climatic conditions of the growing area and the weather conditions prevailing during the growing season [20].

The high variability in the influence of environmental factors on the uptake of mineral nutrients by plants, as well as economic and environmental prerequisites, require the definition of the optimal level of plant nutrient supply to ensure the sustainable production of pea seeds. Therefore, resolving this issue is extremely important now [21].

The aim of this study was to determine the effect of different doses and methods of NPK fertilization and seed inoculation with the microbial preparation Rhizogumin on selected elements of plant structure, seed yield and elements of its structure and protein content in pea seeds and protein yield. The working hypothesis was that optimization of the dose of mineral fertilizers and seed inoculation with the microbial preparation could improve the nutritional status of the plants and increase their productivity. A split nitrogen rate could improve the efficiency of nutrient use by the plants and have a beneficial effect on pea seed yield and quality.

2. Materials and Methods

2.1. Experimental Conditions and Treatments

The field experiment was conducted in 2015–2017 on the territory of the Poltava State Agricultural Experimental Station of the Institute of Swine Production and Agro-Industrial Production in Ukraine (49.55° N and 34.78° E). The average altitude of the site is 175 m above sea level. The experiment was established on heavy clay soil (typical chernozem soil), with the following content in a 0–25 cm layer of humus: 5.15%, mineral elements: alkaline-hydrolyzed nitrogen—162 mg kg^{-1} soil; available phosphorus—150 mg kg^{-1} soil; available potassium—208 mg kg^{-1} soil; pH_{KCl}—5.8.

Weather conditions had a strong influence on pea productivity. The growing seasons in the years of the study varied in terms of weather conditions. They tended to have higher average daily air temperatures compared to the multi-year average and a rather uneven rainfall distribution (Figure 1). The most favorable season for pea growth and development was 2016, when 417 mm of rain fell between April and August, which is 1.7 times more than the multi-year average (247.5 mm). In 2017, rainfall totals were close to the multi-year average (219.3 mm), but the distribution was quite uneven, so conditions for growing peas were less favorable than in 2016. The worst conditions prevailed in 2015, when the amount of rainfall from May to August was 132.1 mm, which, combined with high air temperatures, contributed to a decrease in pea yield levels (Figure 2).

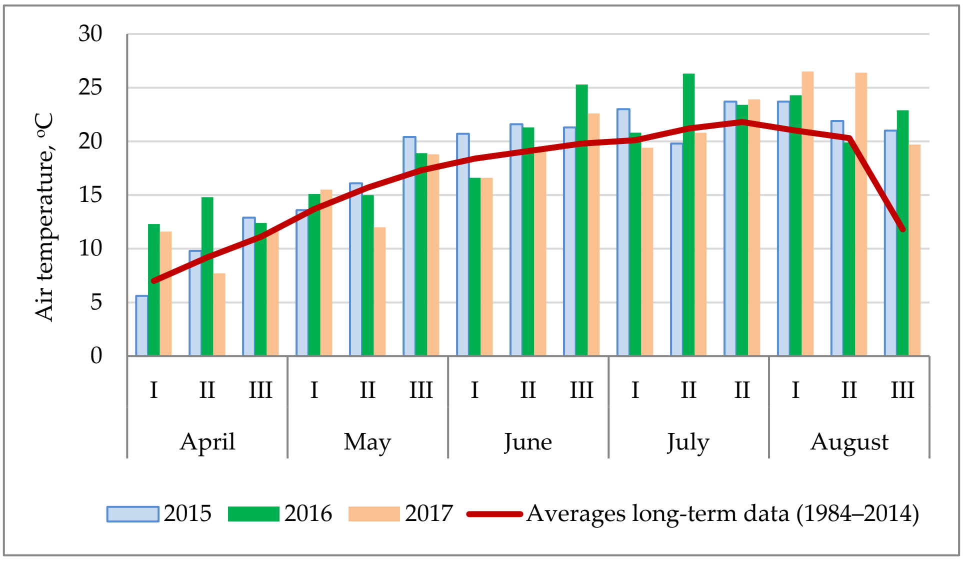

Figure 1. Average air temperature per decade (I, II, III) during the growing seasons of 2015–2017, according to the meteorological post of the Poltava State Agricultural Experimental Station of the Institute of Swine Production and Agro-Industrial Production in Ukraine.

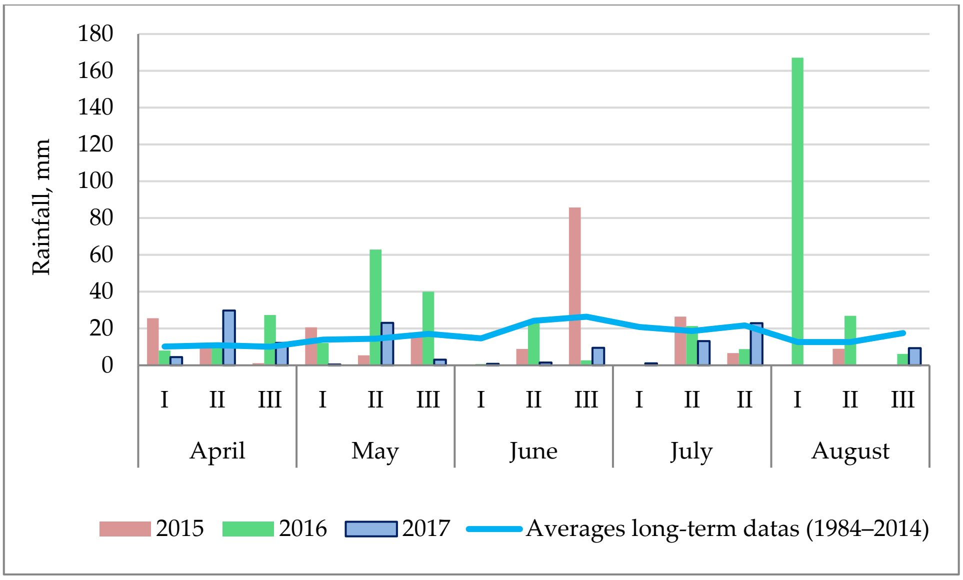

Figure 2. Sum of rainfall per decade (I, II, III) during the growing seasons of 2015–2017, according to the meteorological post of the Poltava State Agricultural Experimental Station of the Institute of Swine Production and Agro-Industrial Production in Ukraine.

The pea cultivar Tsarevych was tested in the experiment. The variety is medium–early with a growing season of 71–75 days. It is resistant to lodging and shedding, and moderately resistant to ascochitosis, fusarium and bacteriosis.

The factors of the field study were: A—mineral fertilization: $N_0P_0K_0$ (control treatment), $N_{15}P_{15}K_{15}$, $N_{15}P_{30}K_{30} + N_{15}$, $N_{30}P_{30}K_{30}$, $N_{30}P_{45}K_{45} + N_{15}$, $N_{45}P_{45}K_{45}$; B—inoculation with microbial preparation Rhizogumin: R1 (inoculation), R0 (no inoculation). The scheme of plots placement in the study was randomized (split-plot) in four replicates.

The experiment was conducted in a randomized complete bloc design in a split-plot arrangement with three replications. The sowing area of the plot was 100 m^2 (20 × 5 m) and the harvesting area was 80 m^2 (20 × 4 m). There were 0.5 m wide protective strips on both sides of each plot. Before sowing, mineral nitrogen in the form of ammonium nitrate (34.5% N), phosphorus in the form of amofos (12.0% N, 52.0% P_2O_5) and potassium in the form of potassium chloride (60% K_2Cl) were applied. In addition, in the third and fifth treatments, at the BBCH 22–23 plant stage, a second dose of nitrogen (N_{15}) in the form of ammonium nitrate (34.5% N) was applied to some objects, according to the experimental scheme. The inoculation of pea seeds was carried out using the commercial microbial preparation Rhizogumin, which contains a suspension of nodule bacteria based on the strain *Rhizobium leguminosarum* and biologically active substances (auxins, cytokinins, amino acids, humic acids), trace elements in chelated form and macronutrients at initial concentrations. Seed treatment was carried out at 2.0 L t^{-1} seeds.

The forecrop for peas in the rotation in all years of the study was maize grown for grain. Peas were sown on 15 April 2015, 11 April 2016 and 18 April 2017, at a rate of 1.2 million germinating seeds per hectare, with a row spacing of 15 cm. All other agronomic treatments in pea cultivation were applied according to agronomic principles. Peas were harvested at 15% moisture content in seeds by a breeding combine Sampo 500 (Sampo Rosenlew Ltd., Pori, Finland).

2.2. Leaf Area and Aboveground Dry Mass

The cutting method was used to determine leaf area. To determine this indicator at the flowering stage of peas (BBCH 71–74), 10 plants were selected from each plot. The leaves from each plant were collected and weighed to two decimal places, and incisions were made in the leaf area with a special key of a certain diameter. The leaf area per plant was determined by a formula using the values of the weight and area of the incision, as well as the total weight of the leaves per plant:

$$S = \frac{P * S1 * n}{P1} \tag{1}$$

where S is the total leaf area, cm^2; S1 is the area of one section, cm^2; P is the total leaf weight, g; n is the number of slices; and P1 is the weight of the sections, g.

Once the leaf area of each plant was determined, the average leaf area for each site was calculated. To do this, the average leaf area per plant was taken and multiplied by the number of plants per 1 m^2. To obtain the values of leaf area per ha, the result was multiplied by 10,000 [22]. To determine the dry weight of the plants, the selected test samples were oven-dried at 105 °C to a constant weight.

2.3. Seed Yield, Yield Components and Protein Content in Seeds

The elements of yield structure (total number of pods and seeds per plant) were determined on the basis of 15 plants taken from each plot before harvest [23]. Harvesting was carried out from each plot separately at the stage of full maturity of pea seeds (BBCH 89). During harvesting, samples were taken to determine the seed moisture and 1000-seed weight.

The Kjeldahl method was used to determine the protein content of the seeds [24,25]. The amount of total nitrogen in the raw material was multiplied by a nitrogen-to-protein conversion factor of 6.25 [26].

2.4. Statistical Analyses and Data Processing and Analysis

All data were statistically analyzed using the method of analysis of variance (ANOVA) for the two-factor split-plot experiment. Due to the similar results obtained in each year of the study, a pooled analysis was performed, and the results are presented as averages over the 3 years. The averages were compared using the Tukey test at a significance level of 0.05. Standard statistical procedures were used for calculating simple correlation coefficients.

3. Results

3.1. Leaf Area

The introduction of mineral fertilizers contributed to a significant increase in pea leaf area, on average by 22.7–60.3% (depending on the dose and method of N fertilization), while the variants with seed inoculation resulted in an increase in the value of the studied trait by an average 4.67% (Table 1). The effect of the interaction of factors was expressed as a significant increase in the leaf area of peas compared to the control. The values of this parameter were highest in the variant R1 + $N_{30}P_{45}K_{45}$ +N_{15} (Figure 3).

The application of the full pre-sowing nitrogen dose (N_{45}) against the full PK fertilization ($P_{45}K_{45}$) was less effective than the split N dose, as evidenced by a significant decrease in leaf area, both in the variant with seed inoculation and the variant without it. In turn, a decrease in the level of mineral supply led to a reduction in the size of the pea leaf area. Among the tested fertilization doses, the lowest values of this parameter were observed in the variant R0 + $N_{15}P_{15}K_{15}$.

Table 1. Effect of mineral fertilization and seed inoculation with the microbial preparation Rhizogumin on the average values of selected elements of plant growth and the productivity parameters protein content in seeds and seed and protein yields of peas (average 2015–2017).

Treatment	LA	FPM	AGDM	NP	NS	TSW	SY	PC	YP
Average (A)									
R0	36.4 a	39.2 a	9.67 a	4.83 a	13.6 a	245.5 a	3.22 a	20.68 a	0.668 a
R1	38.1 b	45.1 b	11.17 b	5.27 b	14.7 b	251.4 b	3.37 b	21.19 b	0.716 b
Average (B)									
Control	27.7 a	34.1 a	8.05 a	4.33 a	12.5 a	235.8 a	2.97 a	19.83 a	0.589 a
$N_{15}P_{15}K_{15}$	34.0 b	39.5 b	9.74 b	4.86 b	13.5 b	242.0 b	3.14 b	20.65 b	0.649 b
$N_{15}P_{30}K_{30} + N_{15}$	37.2 c	44.7 d	10.44 c	5.00 c	14.1 c	246.3 c	3.30 c	21.09 d	0.696 c
$N_{30}P_{30}K_{30}$	38.0 c	43.2 c	10.59 c	5.19 d	14.5 d	252.5 d	3.28 c	20.92 c	0.687 c
$N_{30}P_{45}K_{45} + N_{15}$	44.4 e	46.9 e	12.23 e	5.53 f	15.3 f	258.5 f	3.59 e	21.82 f	0.783 e
$N_{45}P_{45}K_{45}$	42.1 d	44.6 d	11.49 d	5.39 e	14.9 e	255.5 e	3.50 d	21.31 e	0.747 d
Significance (*p* value)									
Inoculation (A)	***	***	***	***	***	***	***	***	***
NPK (B)	***	***	***	***	***	***	***	***	***
$HSD_{0.05}$									
Inoculation (A)	0.7615	0.9049	0.3569	0.0388	0.2759	2.4535	0.0541	0.0439	0.0051
NPK (B)	0.4709	0.3487	0.1375	0.0150	0.1063	0.9455	0.0208	0.0169	0.0020
SE									
Inoculation (A)	0.100	0.119	0.047	0.005	0.036	0.323	0.007	0.005	0.0006
NPK (B)	0.174	0.206	0.081	0.008	0.063	0.561	0.012	0.010	0.001

A: inoculation, B: fertilization, LA: leaf area (thousand m^2 ha^{-1}), FPM: fresh plant mass (g $plant^{-1}$), AGDM: aboveground dry mass of plant (g $plant^{-1}$), NP: number of pods per plant (psc $plant^{-1}$), NS: number of seeds per plant (psc plant $^{-1}$), TSW: 1000-seed weight (g), SY: seed yield (t ha^{-1}), PC: protein content (%), YP: yield of protein (t ha^{-1}). The differences between data indicated by the same letter are not statistically significant. Significance (*p* value) *** $p \leq 0.001$.

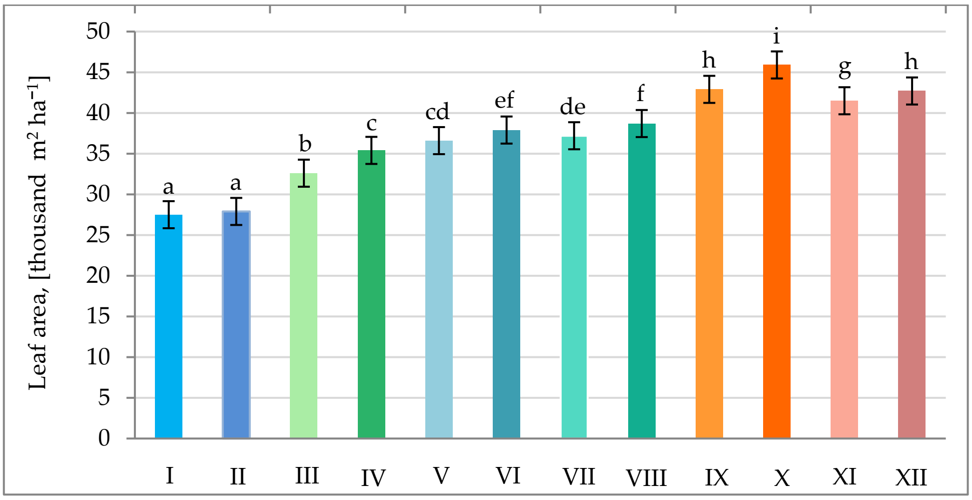

Figure 3. Leaf area of peas in various combinations of mineral fertilization and seed inoculation with the microbial preparation Rhizogumin. I—control object (R0 + $N_0P_0K_0$); II—R1 + $N_0P_0K_0$; III—R0 + $N_{15}P_{15}K_{15}$; IV—R1 + $N_{15}P_{15}K_{15}$; V—R0 + $N_{15}P_{30}K_{30}$ + N_{15}; VI—R1 + $N_{15}P_{30}K_{30}$ + N_{15}; VII—R0 + $N_{30}P_{30}K_{30}$; VIII—R1 + $N_{30}P_{30}K_{30}$; IX—R0 + $N_{30}P_{45}K_{45}$ + N_{15}; X—R1 + $N_{30}P_{45}K_{45}$ + N_{15}; XI—R0 + $N_{45}P_{45}K_{45}$; XII—R1 + $N_{45}P_{45}K_{45}$; R1—with seed inoculation, R0—without seed inoculation. The differences between data indicated by the same letter are not statistically significant ($p \leq 0.05$); ±SE.

3.2. Fresh Mass of Plant

A significant increase in the fresh mass of the aboveground part of plants in response to seed inoculation and mineral fertilization was shown, whereas mineral fertilization had a greater influence on the value of this parameter. With an increase in fertilizer application rate, plant biomass increased by an average of 15.8–37.5% compared to $N_0P_0K_0$, while seed inoculation increased the value of this parameter by an average of 15.0% (Table 1).

A significant interaction of the studied factors and its influence on the processes of accumulation of aboveground fresh mass by plants was demonstrated (Figure 4). This was most evident in the variants R1 + $N_{30}P_{45}K_{45}$ + N_{15} and R1 + $N_{45}P_{45}K_{45}$, where on average, plant biomass in the study years exceeded the control variant by 52.1 and 48.2%, respectively. The reduction in plant mineral supply led to a decrease in the intensity of the growth of the aboveground fresh mass of pea plants.

In the variant R1 + $N_{30}P_{30}K_{30}$, the fresh mass of plants increased by 42.9% compared to the control, while $N_{15}P_{15}K_{15}$ application in combination with seed inoculation provided a 30.1% increase in the value of this parameter. In the variants without seed inoculation, a similar trend of change was observed. The most effective application was $N_{30}P_{45}K_{45}$ + N_{15}. The fresh mass of plants in this variant exceeded the control by 35.0%. The growth of the aboveground part of the plants was least intensive following application of the variant R0 + $N_{15}P_{15}K_{15}$. In addition, it was shown that dividing the N rate was more effective than applying the whole N rate before sowing, but only on R0 treatments.

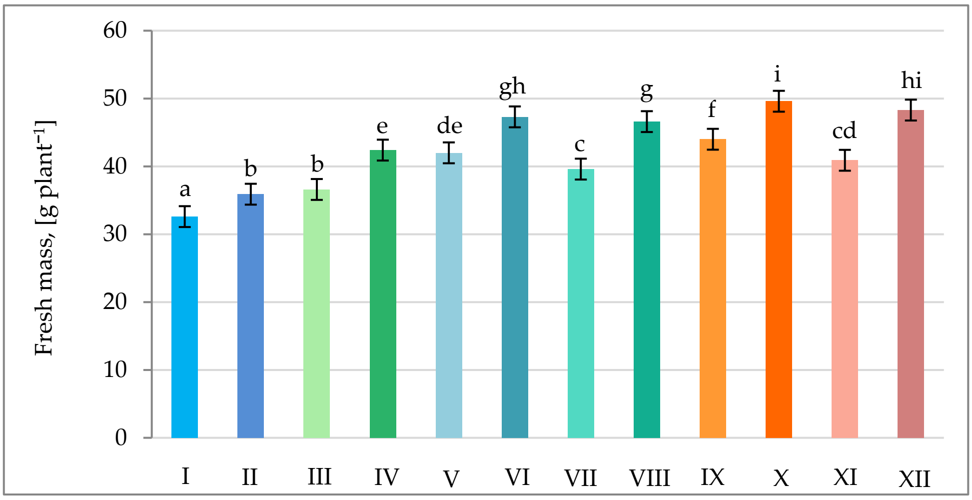

Figure 4. Fresh mass of pea plants in various combinations of mineral fertilization and seed inoculation with the microbial preparation Rhizogumin. I—control object (R0 + $N_0P_0K_0$); II—R1 + $N_0P_0K_0$; III—R0 + $N_{15}P_{15}K_{15}$; IV—R1 + $N_{15}P_{15}K_{15}$; V—R0 + $N_{15}P_{30}K_{30}$ + N_{15}; VI—R1 + $N_{15}P_{30}K_{30}$ + N_{15}; VII—R0 + $N_{30}P_{30}K_{30}$; VIII—R1 + $N_{30}P_{30}K_{30}$; IX—R0 + $N_{30}P_{45}K_{45}$ + N_{15}; X—R1 + $N_{30}P_{45}K_{45}$ + N_{15}; XI—R0 + $N_{45}P_{45}K_{45}$; XII—R1 + $N_{45}P_{45}K_{45}$; R1—with seed inoculation, R0—without seed inoculation. The differences between data indicated by the same letter are not statistically significant ($p \leq 0.05$); ±SE.

3.3. Aboveground Dry Mass of Plant

Mineral fertilization and seed inoculation had a significant positive effect on plant dry matter accumulation. The application of mineral fertilizers provided an increase in the dry mass of the aboveground parts of pea plants of 21.0–51.9% compared to the control, while seed inoculation increased the value of this parameter by 15.5% (Table 1).

The results of this study showed a significant interaction of the tested experimental factors and its effect on plants' dry aboveground mass (Figure 5). This was most pronounced in the variant R1 + $N_{30}P_{45}K_{45}$ + N_{15}, where on average, for 3 years, the dry aboveground mass of pea plants was higher by 71.3% compared to the $N_0P_0K_0$ application. The application of $N_{45}P_{45}K_{45}$ was significantly less effective, but also provided a large increase in the values of this parameter (by 60.7%) compared to the control. A similar trend of change was also observed in the variants without seed inoculation.

Decreasing the level of mineral fertilization had a negative effect on the intensity of dry matter accumulation by pea plants. In the R0 + $N_{30}P_{30}K_{30}$ variant, the aboveground dry mass of plants increased by 26.3% compared to the control, while the same dose of mineral application in connection with seed inoculation provided a 50.8% increase in the values of this parameter. On the other hand, in the R0 + $N_{15}P_{15}K_{15}$ variant, the increase in the aboveground dry mass of plants compared to the control was the smallest (16.6%), while the sowing of inoculated seeds and application of the same mineral fertilizer dose contributed to a significant increase in plant dry mass accumulation (by 38.4% on average).

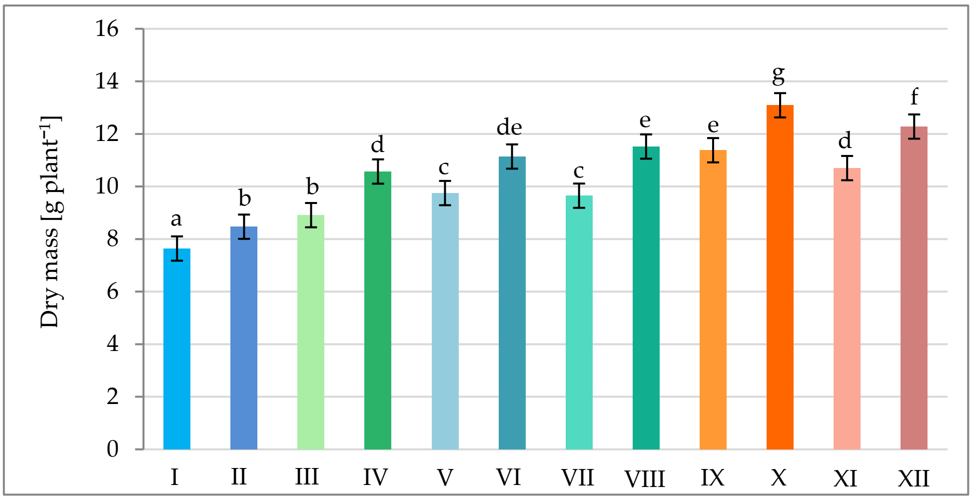

Figure 5. Dry mass of aboveground parts of pea plants in various combinations of mineral fertilization and seed inoculation with the microbial preparation Rhizogumin. I—control object (R0 + $N_0P_0K_0$); II—R1 + $N_0P_0K_0$; III—R0 + $N_{15}P_{15}K_{15}$; IV—R1 + $N_{15}P_{15}K_{15}$; V—R0 + $N_{15}P_{30}K_{30}$ + N_{15}; VI—R1 + $N_{15}P_{30}K_{30}$ + N_{15}; VII—R0 + $N_{30}P_{30}K_{30}$; VIII—R1 + $N_{30}P_{30}K_{30}$; IX—R0 + $N_{30}P_{45}K_{45}$ + N_{15}; X—R1 + $N_{30}P_{45}K_{45}$ + N_{15}; XI—R0 + $N_{45}P_{45}K_{45}$; XII—R1 + $N_{45}P_{45}K_{45}$; R1—with seed inoculation, R0—without seed inoculation. The differences between data indicated by the same letter are not statistically significant ($p \leq 0.05$); ±SE.

3.4. Number of Pods per Plant

This study showed a significant positive effect of mineral fertilizer, microbial preparation and their interaction on the number of pods formed on the plants. As the dose of mineral fertilizers increased, the number of pods per plant increased by an average of 12.2–27.7%, depending on the dose and method of N application, compared to the $N_0P_0K_0$ treatment. In contrast, microbial seed inoculation increased the number of pods per plant by 9.1% (Table 1).

There was also a significant interaction of the experimental factors and their effect on pod abundance. The highest number of pods was noted in the variant R1 + $N_{30}P_{45}K_{45}$ + N_{15} (Figure 6). In the variant R1 + $N_{45}P_{45}K_{45}$, the number of pods formed on pea plants was significantly lower than at the split nitrogen dose. In turn, the fewest pods were formed by plants in the R0 + $N_{15}P_{15}K_{15}$ variant, but even with the lowest dose of mineral fertilizers, the number of pods was 15.2% higher than with the control treatment.

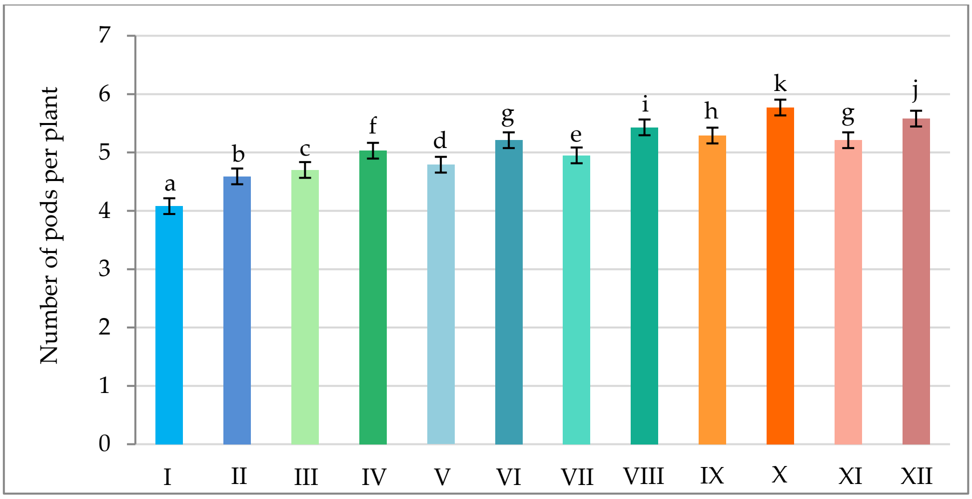

Figure 6. Number of pods per plant of pea in various combinations of mineral fertilization and seed inoculation with the microbial preparation Rhizogumin. I—control object (R0 + $N_0P_0K_0$); II—R1 + $N_0P_0K_0$; III—R0 + $N_{15}P_{15}K_{15}$; IV—R1 + $N_{15}P_{15}K_{15}$; V—R0 + $N_{15}P_{30}K_{30}$ + N_{15}; VI—R1 + $N_{15}P_{30}K_{30}$ + N_{15}; VII—R0 + $N_{30}P_{30}K_{30}$; VIII—R1 + $N_{30}P_{30}K_{30}$; IX—R0 + $N_{30}P_{45}K_{45}$ + N_{15}; X—R1 + $N_{30}P_{45}K_{45}$ + N_{15}; XI—R0 + $N_{45}P_{45}K_{45}$; XII—R1 + $N_{45}P_{45}K_{45}$; R1—with seed inoculation, R0—without seed inoculation. The differences between data indicated by the same letter are not statistically significant ($p \leq 0.05$); ±SE.

3.5. Number of Seeds per Plant

The number of seeds per pea plant differed significantly according to microbial preparation, mineral fertilizer dose and their interaction effect. The mineral fertilizer dose had the greatest effect on the number of seeds. Increasing the level of mineral fertilization contributed to a significant rise in the number of seeds per plant. The increase in the value of this indicator compared to the control treatment was 8.0–22.4%. The inoculation of seeds with a microbial preparation also had a positive effect on the number of seeds per plant. The average increase in the value of this indicator in the variants with inoculation was 8.1% (Table 1).

This study also showed an interaction effect (Figure 7). On average, over the years, the highest number of seeds per pea plant was observed in the variant R1 + $N_{30}P_{45}K_{45}$ + N_{15}, while a single application of N at a dose of 45 kg ha^{-1} did not significantly affect the value of this indicator. Also, in the treatment without seed inoculation, the application of N_{45} fertilization did not lead to a difference in the number of seeds per plant. The difference in the values of the described parameter in these variants was within the limits of statistical error. In contrast, the effect was significant at the lower nitrogen dose (N_{30}), independent of seed inoculation. The smallest number of seeds was formed by plants with the variant R0 + $N_{15}P_{15}K_{15}$, where the value of this parameter exceeded the control by 8.3%.

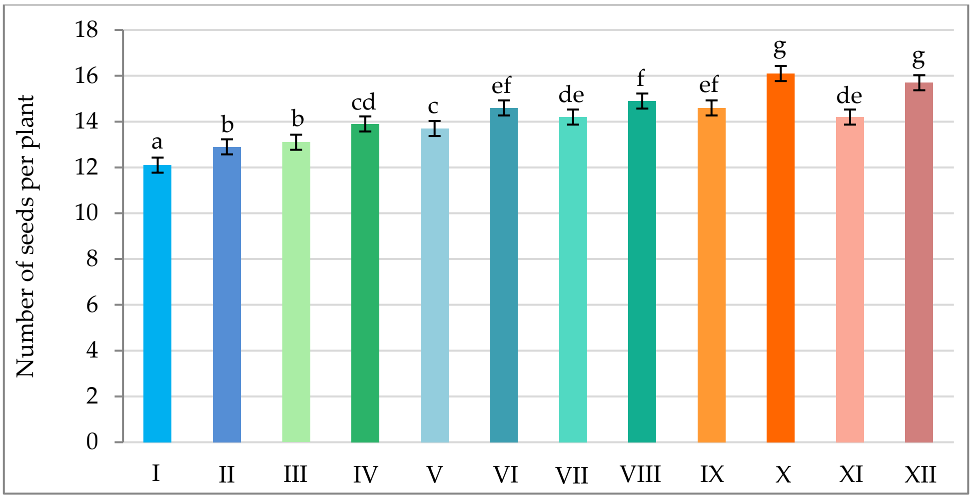

Figure 7. Number of seeds per plant of pea in various combinations of mineral fertilization and seed inoculation with the microbial preparation Rhizogumin. I—control object (R0 + $N_0P_0K_0$); II—R1 + $N_0P_0K_0$; III—R0 + $N_{15}P_{15}K_{15}$; IV—R1 + $N_{15}P_{15}K_{15}$; V—R0 + $N_{15}P_{30}K_{30}$ + N_{15}; VI—R1 + $N_{15}P_{30}K_{30}$ + N_{15}; VII—R0 + $N_{30}P_{30}K_{30}$; VIII—R1 + $N_{30}P_{30}K_{30}$; IX—R0 + $N_{30}P_{45}K_{45}$ + N_{15}; X—R1 + $N_{30}P_{45}K_{45}$ + N_{15}; XI—R0 + $N_{45}P_{45}K_{45}$; XII—R1 + $N_{45}P_{45}K_{45}$; R1—with seed inoculation, R0—without seed inoculation. The differences between data indicated by the same letter are not statistically significant ($p \leq 0.05$); ±SE.

3.6. Thousand-Seed Weight

Mineral fertilization, seed inoculation and their combination significantly influenced the weight of 1000 seeds of pea. Of the studied factors, mineral fertilization had the greatest effect on the value of this indicator. In the variants of mineral fertilization, the weight of 1000 seeds of pea increased by 2.62–9.62% compared to the control, while seed inoculation provided a 2.4% increase in the value of this parameter.

The interaction effect of the tested experimental factors was demonstrated by a significant increase in the weight of 1000 pea seeds compared to the control treatment (Figure 8). This study showed a significant effect of the combination of seed inoculation and increased mineral fertilization doses on the weight of 1000 seeds. The value of this parameter was highest in the variants R1 + $N_{30}P_{45}K_{45}$ + N_{15} and R1 + $N_{45}P_{45}K_{45}$ (increase by 12.1 and 10.8%, respectively). As plant nutrient supply decreased, the weight of 1000 seeds decreased. Of the studied levels of mineral fertilization, the least effect was obtained by R0 + $N_{15}P_{15}K_{15}$ application. In this variant, the increase in 1000-seed weight compared to the control was 3.0%. The application of the microbial preparation Rhizogumin was more effective when the dose of mineral fertilizer was increased.

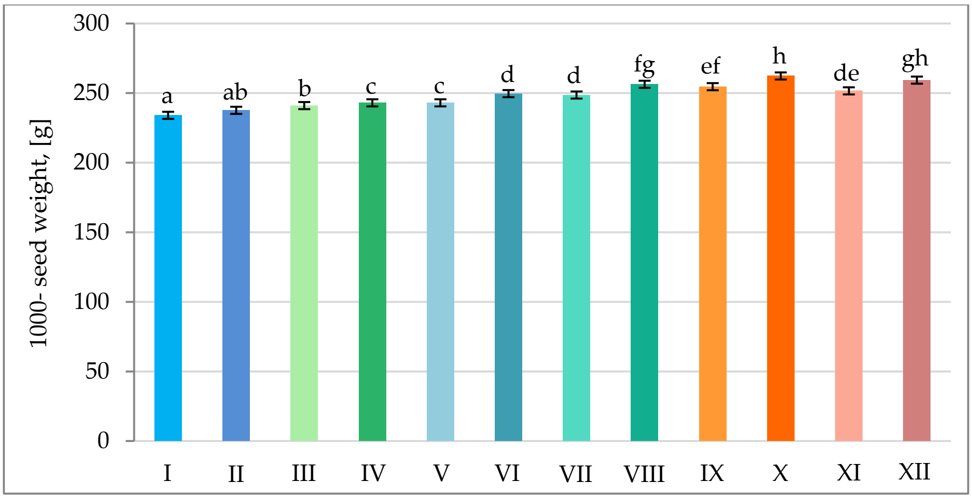

Figure 8. Thousand-seed weight of peas in various combinations of mineral fertilization and seed inoculation with the microbial preparation Rhizogumin. I—control object (R0 + $N_0P_0K_0$); II—R1 + $N_0P_0K_0$; III—R0 + $N_{15}P_{15}K_{15}$; IV—R1 + $N_{15}P_{15}K_{15}$; V—R0 + $N_{15}P_{30}K_{30}$ + N_{15}; VI—R1 + $N_{15}P_{30}K_{30}$ + N_{15}; VII—R0 + $N_{30}P_{30}K_{30}$; VIII—R1 + $N_{30}P_{30}K_{30}$; IX—R0 + $N_{30}P_{45}K_{45}$ + N_{15}; X—R1 + $N_{30}P_{45}K_{45}$ + N_{15}; XI—R0 + $N_{45}P_{45}K_{45}$; XII—R1 + $N_{45}P_{45}K_{45}$; R1—with seed inoculation, R0—without seed inoculation. The differences between data indicated by the same letter are not statistically significant ($p \leq 0.05$); ±SE.

3.7. Seed Yield

The results of this research showed a significant positive effect of the interaction of the studied factors on the yield of pea seeds. Its highest values were obtained in 2016 with the most favorable moisture conditions for the growth and development of pea plants. The least favorable period for the formation of pea productivity was the growing season of 2015. The yield of pea seeds in this year compared to 2016 decreased by 68.3–79.6%. The decrease in the values of this indicator in 2017 compared to the favorable year of 2016 was 44.6–78.2% (Table A1, Appendix A).

On average, during the 3 years of the study, there was a significant positive effect of the studied factors (seed inoculation and fertilization) on pea seed yield. In general, the value of this parameter varied from 2.90 to 3.66 t ha^{-1}. On average, during the 3 years of the study, the increase in pea seed yield under the influence of mineral fertilization ranged from 5.7 to 20.9%. Seed inoculation contributed to an increase in the value of this parameter by 4.7% compared to the control (Table 1).

Statistical analysis of the experimental results showed a significant interaction between seed inoculation and mineral fertilization during pea productivity (Figure 9). A significant increase in seed yield was obtained by improving the mineral supply to the plants. On average, over the 3 years of the study, the highest values (3.66 t ha^{-1}) were obtained in the variant R1 + $N_{30}P_{45}K_{45}$ + N_{15}. In the variant of combining seed inoculation and $N_{45}P_{45}K_{45}$ application, the values of this parameter decreased slightly (the differences were not statistically confirmed). With the combined application of R1 + $N_{30}P_{30}K_{30}$, no significant changes in seed yield depending on the method of nitrogen supply were observed. The increase in seed yield compared to the control in the variant R1 + $N_{15}P_{15}K_{15}$ was 11.0%.

When sowing pure seeds (without inoculation), pea yield was slightly lower at all levels of mineral fertilization, while an increase in the dose of mineral fertilizers led to a significant increase in yield. At the same time, the separate application of nitrogen proved to be more effective.

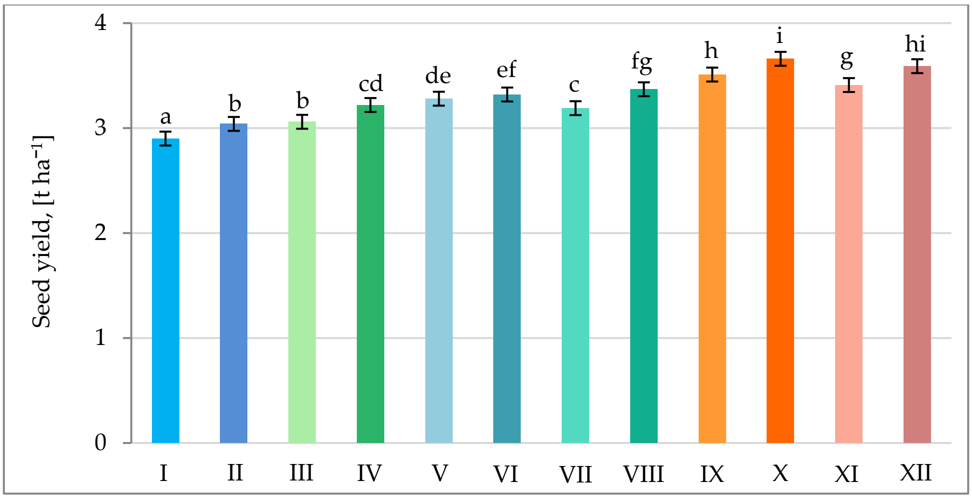

Figure 9. Seed yield of pea in various combinations of mineral fertilization and seed inoculation with the microbial preparation Rhizogumin (average for 2015–2017). I—control object (R0 + $N_0P_0K_0$); II—R1 + $N_0P_0K_0$; III—R0 + $N_{15}P_{15}K_{15}$; IV—R1 + $N_{15}P_{15}K_{15}$; V—R0 + $N_{15}P_{30}K_{30}$ + N_{15}; VI—R1 + $N_{15}P_{30}K_{30}$ + N_{15}; VII—R0 + $N_{30}P_{30}K_{30}$; VIII—R1 + $N_{30}P_{30}K_{30}$; IX—R0 + $N_{30}P_{45}K_{45}$ + N_{15}; X—R1 + $N_{30}P_{45}K_{45}$ + N_{15}; XI—R0 + $N_{45}P_{45}K_{45}$; XII—R1 + $N_{45}P_{45}K_{45}$; R1—with seed inoculation, R0—without seed inoculation. The differences between data indicated by the same letter are not statistically significant ($p \leq 0.05$); ±SE.

3.8. Protein Content in Seeds

The results of this research showed that the protein content of pea seeds was significantly influenced by mineral fertilizers, seed inoculation and their interaction.

The value of protein content in pea grain varied during the years of research. The lowest values of this parameter were observed in 2016, with the highest level of moisture availability during the growing season of peas. Under the dry conditions of 2015 and 2017, an increase in protein content in pea grain was observed. Within each year of research, the value of protein content in pea grain increased as the supply of nitrogen nutrition to the plants improved. It should be noted that the separate application of nitrogen fertilizers contributed to an increase in protein content along with the full provision of plants with mineral nitrogen from the beginning of the growing season (Table A2, Appendix A).

On average, over 3 years of research, the most significant determinant of seed protein content was mineral fertilization. Depending on the amount of NPK, the values of this parameter exceeded the control variant by 4.1–10.0%. The increase in this parameter in the variants with seed inoculation compared to the control treatment was 2.5% (Table 1).

The interaction effect of the studied factors was expressed as a significant increase in protein content in pea seeds compared to the control (Figure 10). It was the highest in the variant of fertilization $N_{30}P_{45}K_{45}$ + N_{15}, where the increase in the value of this parameter was 13.7% when inoculated seeds were sown, and 10.5% when pure seeds were sown. With a single application of the dose $N_{45}P_{45}K_{45}$, the protein content of pea seeds decreased significantly compared to the split dose.

A similar trend can be observed in the variant of $N_{30}P_{30}K_{30}$ application, both with and without seed inoculation, indicating that splitting the nitrogen rate contributes to the protein content in pea seeds. The deterioration of the plant nutrition regime led to a decrease in the protein content of pea seeds. In variants combining seed inoculation and fertilization with $N_{15}P_{30}K_{30}$ +N_{15}, $N_{30}P_{30}K_{30}$ and $N_{15}P_{15}K_{15}$, the increases in protein content compared to the control were 9.4%, 8.1% and 7.1%, respectively. In the variants where the microbiological preparation was not used, the protein content in pea seeds was lower (7.2%, 6.7% and 5.0%, respectively) compared to the control treatment.

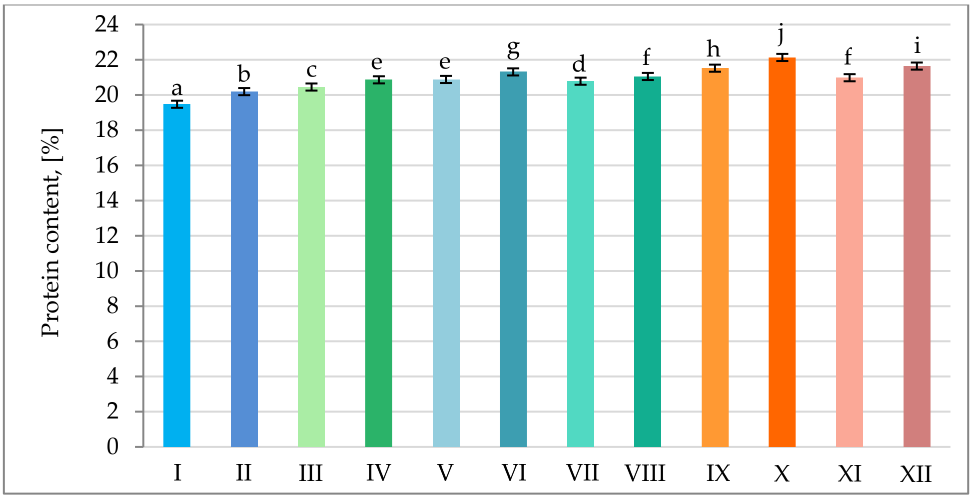

Figure 10. Average protein content in pea seeds in various combinations of mineral fertilization and seed inoculation with the microbial preparation Rhizogumin. I—control object (R0 + $N_0P_0K_0$); II—R1 + $N_0P_0K_0$; III—R0 + $N_{15}P_{15}K_{15}$; IV—R1 + $N_{15}P_{15}K_{15}$; V—R0 + $N_{15}P_{30}K_{30}$ + N_{15}; VI—R1 + $N_{15}P_{30}K_{30}$ + N_{15}; VII—R0 + $N_{30}P_{30}K_{30}$; VIII—R1 + $N_{30}P_{30}K_{30}$; IX—R0 + $N_{30}P_{45}K_{45}$ + N_{15}; X—R1 + $N_{30}P_{45}K_{45}$ + N_{15}; XI—R0 + $N_{45}P_{45}K_{45}$; XII—R1 + $N_{45}P_{45}K_{45}$; R1—with seed inoculation, R0—without seed inoculation. The differences between data indicated by the same letter are not statistically significant ($p \leq 0.05$); ±SE.

3.9. Protein Yield

Protein yield depended on mineral fertilization, the inoculant and their interaction. Despite a decrease in protein content in grain, the total protein yield per hectare in 2016 was offset by a significant increase in pea seed yields (Table A3, Appendix A). The three-year results of the experiment showed that the fertilizer factor had the greatest effect, increasing this index by an average of 10.2–32.9% compared to the control, and to a lesser extent, the pre-sowing seed inoculation (by an average of 7.1%) (Table 1).

Statistical analysis of the results confirmed the interaction of the tested factors (Figure 11). The combination of seed inoculation and the application of $N_{15}P_{30}K_{30}$ + N_{15}, in which protein yield per 1 ha exceeded the control by 43.7%, proved to be the most reasonable. The increase in protein yield per 1 ha in the variant R1 + $N_{45}P_{45}K_{45}$ compared to the control treatment was slightly lower (37.9%). The deterioration of plant mineral supply in both variants with and without seed inoculation led to a decrease in seed yield and protein content, resulting in a significant decrease in total protein yield per 1 ha compared to the maximum fertilizer application rate. It was lowest in the variant R0 + $N_{15}P_{15}K_{15}$, where the protein yield per 1 ha exceeded the control variant by 19.1% with the seed inoculated before sowing and by 10.9% without seed inoculation.

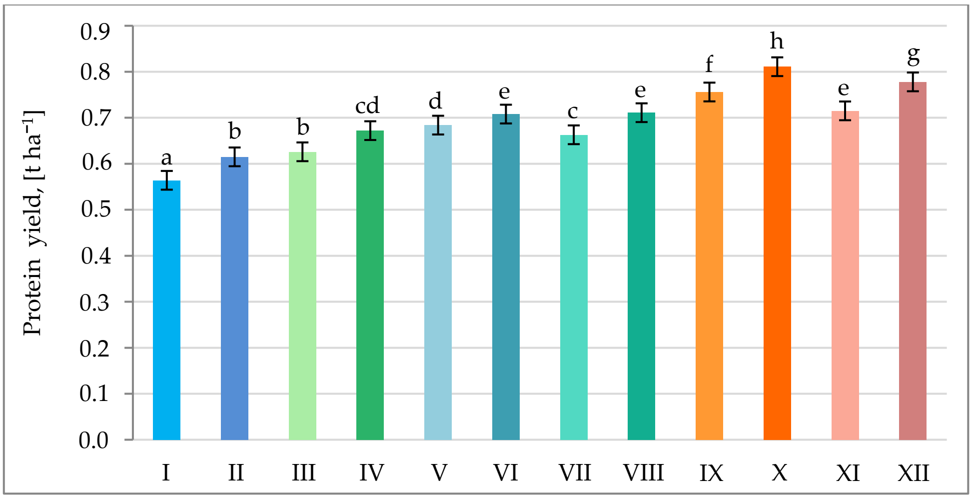

Figure 11. Average protein yield of pea in various combinations of mineral fertilization and seed inoculation with the microbial preparation Rhizogumin. I—control object (R0 + $N_0P_0K_0$); II—R1 + $N_0P_0K_0$; III—R0 + $N_{15}P_{15}K_{15}$; IV—R1 + $N_{15}P_{15}K_{15}$; V—R0 + $N_{15}P_{30}K_{30}$ + N_{15}; VI—R1 + $N_{15}P_{30}K_{30}$ + N_{15}; VII—R0 + $N_{30}P_{30}K_{30}$; VIII—R1 + $N_{30}P_{30}K_{30}$; IX—R0 + $N_{30}P_{45}K_{45}$ + N_{15}; X—R1 + $N_{30}P_{45}K_{45}$ + N_{15}; XI—R0 + $N_{45}P_{45}K_{45}$; XII—R1 + $N_{45}P_{45}K_{45}$; R1—with seed inoculation, R0—without seed inoculation. The differences between data indicated by the same letter are not statistically significant ($p \leq 0.05$); ±SE.

3.10. Correlation Relationships between Morphological Features of Plants, Yield Components and Seed Yield

The correlations between seed yield and the values of the morphological parameters of the plants and elements of their productivity were evaluated. The results show a high positive correlation between the yield and the studied vegetative growth parameters and productivity elements of pea plants. The analyses showed that seed yield correlates most strongly with the size of the leaf area (r = 0.961 ± 0.01), plant dry weight (r = 0.957 ± 0.01) and plant fresh weight (r = 0.857 ± 0.01) (Table 2).

This study found strong correlations between the number of pods formed on the plants and the leaf area (r = 0.923 ± 0.01), the fresh mass of plant (r = 0.938 ± 0.01), the aboveground dry mass of the plant (r = 0.976 ± 0, 01), the number of seeds formed on the plants (r = 0.975 ± 0.01) and seed yield (r = 0.945 ± 0.01) and also between the number of seeds per plant and leaf area size (r = 0.921 ± 0.01), the fresh mass of the plant (r = 0.945 ± 0.01), the plant dry aboveground weight (r = 0.967 ± 0.01), the number of pods formed on the plants (r = 0.975 ± 0.01) and seed yield (r = 0.945 ± 0.01).

There were also high correlations between seed protein content and plant fresh weight (r = 0.936 ± 0.01), plant dry weight (r = 0.957 ± 0.01), the number of pods formed on the plants (r = 0.956 ± 0.01), the number of seeds per plant (r = 0.955 ± 0.01) and the 1000-seed weight (r = 0.916 ± 0.01). In contrast, the protein yield per unit of area sown was largely determined by seed yield, as evidenced by the very high correlation between these values (r = 0.996 ± 0.01).

Table 2. Correlation relationships between seed yield of peas and morphological features of plants, yield components and protein yield depending on mineral fertilization, seed inoculation with the microbial preparation Rhizogumin and their combination.

Features	SY	PY	PC	TSW	NS	NP	AGDM	FPM	LA
SY	1.000								
PY	0.996 ***	1.000							
PC	0.956 ***	0.977 ***	1.000						
TSW	0.945 ***	0.946 ***	0.916 ***	1.000					
NS	0.945 ***	0.958 ***	0.955 ***	0.957 ***	1.000				
NP	0.945 ***	0.956 ***	0.956 ***	0.958 ***	0.975 ***	1.000			
AGDM	0.957 ***	0.967 ***	0.957 ***	0.946 ***	0.967 ***	0.976 ***	1.000		
FPM	0.902 ***	0.919 ***	0.936 ***	0.888 ***	0.945 ***	0.938 ***	0.957 ***	1.000	
LA	0.961 ***	0.962 ***	0.940 ***	0.939 ***	0.921 ***	0.923 ***	0.924 ***	0.857 ***	1.000

SY—seed yield, PY—protein yield, PC—protein content, TSW—1000-seed weight, NS—number of seeds, NP—number of pods, AGDM—aboveground dry mass of plant, FPM—fresh mass of plant, LA—leaf area; significance levels are represented as *** $p \leq 0.0001$.

4. Discussion

The results of this study showed a positive effect of mineral fertilizer application, pre-sowing seed inoculation with Rhizogumin and their combination on the formation of pea leaf area as a key photosynthetic organ. Increasing the dose of mineral fertilizers from $N_{15}P_{15}K_{15}$ and $N_{30}P_{30}K_{30}$ to $N_{45}P_{45}K_{45}$ resulted in an increase in the level of plant nutrient supply, leading to an increase in pea leaf area by 32.7% and 41.5%, respectively. During development, pea plants can obtain nitrogen from the soil and from the air through biological nitrogen fixation by the root nodule bacteria *Rhizobium* [27]. One method that can improve plant nitrogen nutrition due to the formation and functioning of symbiosis between legumes and Gram-negative nitrogen-fixing bacteria from the *Rhizobium* genus, thus increasing the biological availability of nutrients from the soil and intensity of plant growth and development, is seed inoculation with microbial preparations [28].

The positive effect of this treatment on pea plant growth was reported in our study, in which the application of Rhizogumin contributed to a significant increase in pea leaf area. A similar effect was also noted in the studies of Zając et al. [29], where the size of the leaf surface of peas in the variant of seed treatment with Nitragin increased in the phase of flowering by 7.6% compared to the control. In the study of Khiangte et al. [30], the inoculation of pea seeds with *Rhizobium* increased the number of leaves on the plants by 15.4% compared to the control.

The results of our study showed a significant positive effect of supplementary mineral nitrogen fertilization at the dose of N_{15} at growth stage (BBCH) 22–23 of pea plant growth and development. This can be explained by the positive synergistic effect of mineral nutrients on the initial development of the root system and aboveground plant parts, which promotes the growth of leaf blades and increases the total leaf area [31]. In turn, the increase in leaf area allows more photosynthetically active radiation to be absorbed and light energy to be used in the formation of plant biomass [32]. The highest aboveground weight values of pea plants were obtained in response to a combination of seed inoculation and $N_{45}P_{45}K_{45}$ application, with the split nitrogen application being more effective. This can be explained by the fact that the application of N_{30} before sowing ensured the initial growth of plant seedlings before the active symbiosis of legumes with nitrogen-fixing rhizobia started, while the application of N_{15} at pea growth stage BBCH 22–23 stimulated the growth and development of the vegetative part of the plants. This may be possible by introducing active, virulent, nitrogen-fixing strains of microorganisms into the rhizosphere [33]. The literature on this subject reports that adequate availability of phosphorus and potassium to plants can increase colonization of the pea rhizosphere by rhizobia [34], stimulate growth processes, increase photosynthetic efficiency, provide an adequate supply of synthesized carbon to organs that need it and increase the accumulation of aboveground biomass by plants [35]. This was confirmed in our study, where the highest plant dry weight was recorded in the variant combining seed inoculation and $N_{30}P_{45}K_{45}$ + N_{15} application. At the same time, an increase in the accumulation of organic dry matter by plants and an increase in the supply of organic compounds to generative organs during their formation promotes plant productivity [36].

Our study showed an increase in the number of pods formed per plant and the number of seeds per pod with an increase in the dose of mineral fertilizers. At the same time, the highest numbers of pods and seeds per plant were observed in variants combining the seed inoculation of the microbial preparation Rhizogumin and the $N_{45}P_{45}K_{45}$ application. A positive effect of mineral fertilization on elements of pea yield structure (number of pods and seeds per plant) was also reported by Badr and Fayed [37]. In a study by Bunker et al. [38], a combination of seed inoculation with *Rhizobium* and $N_{20}P_{40}$ application increased the number of pods per plant and seeds in pods by 57.2% and 34.2%, respectively, compared to the control variant. Also, Khatana et al. [39] found that the highest number of pods formed on black gram (*Vigna munga* L.) plants could be obtained with the combined application of NPK and seed inoculation with *Rhizobium*. The results of our study showed a significant positive effect of mineral fertilization on the weight of 1000 pea seeds. This may be due to the synergistic effect of the supplied nutrients, an increase in the photosynthetic activity of plants, the formation of more organic compounds and their more efficient use by plants in the process of seed formation and filling [40]. At the same time, additional plant nutrition with nitrogen during the growing season can enhance the growth of the vegetative part of the plant, extend the period of active functioning of the leaf surface and increase the level of nutrient supply to the seeds during the filling and maturation phase [41]. In general, our experiment showed the highest efficiency of the tested factors with the complex application of mineral fertilizers at a rate of $N_{30}P_{45}K_{45}$ +N_{15} and the microbial preparation Rhizogumin. The efficiency of the microbial application may be due to the plants' preferential use of fixing nitrogen compared to mineral nitrogen.

The increase in plant productivity had a positive effect on seed yield. Its value increased as the nutrient supply to the plants improved. In general, the combination of seed inoculation with the microbial preparation and the application of $N_{30}P_{45}K_{45}$ + N_{15} proved to be the most effective. This variant had the highest seed yield (3.66 t ha^{-1}). The results of our study are consistent with an earlier study by Janusauskaite [20], in which pea seed yield increased with increasing NPK dose. The positive effect of mineral fertilization on pea seed yields is also confirmed by the studies of Lalito et al., and Chandel et al. [42,43]. As noted by Sharma et al. [44], nitrogen is a highly mobile element, and therefore, increasing the dose of its application may be ineffective due to leaching losses. This position was also reflected in our study, where the split application of mineral nitrogen proved to be more effective.

The results of our study showed a variation in seed yields depending on the weather conditions of the years of the experiment. The highest values were observed in the wet 2016, and the lowest were in 2015, which was characterized by insufficient moisture and the uneven distribution of precipitation during the growing season. In contrast to the grain yield, the protein content of the grain changed in the opposite direction, which is also confirmed by previous studies by Nikolopoulou et al., and Walter et al. [45,46]. One of the reasons for this effect is the dilution of nitrogen by non-protein compounds. Since the amount of nitrogen in grain is constant, high grain yields lead to a decrease in protein content in grain, and vice versa [47]. The results of our study showed that the improvement in plant nutrient supply through the combination of seed inoculation and NPK mineral fertilization led to an increase in seed protein content, and protein yield per 1 ha. This may be due to improved nitrogen delivery during remobilization from the well-developed vegetative part of the plant, as well as ongoing nitrogen uptake by the plant's root system [48]. The increase in seed protein content may also be due to improved plant nutrition with phosphorus and potassium, as phosphorus is a key component of nitrogen transformation and the regulation of enzymatic activity to increase legume–rhizobia symbiosis [49], while potassium stimulates protein synthesis processes [36]. At the same time, scientists have hypothesized that it is the nitrogen status of the soil that determines the intensity of protein accumulation in pea grain [50]. The results of our study confirm this hypothesis. In particular, the protein content in pea grain reached its maximum value in the variants of N_{45} application, and separate nitrogen application was more effective. The positive effect of increasing the level of mineral nitrogen supply to plants on the protein content of pea seeds is also evidenced by the research of Igbasan et al. [51]. The highest values of these indices were provided by the combined use of seed inoculation with Rhizogumin and $N_{30}P_{45}K_{45}$ + N_{15} application.

The results of this research showed that the value of pea seed yield was controlled to the greatest extent by the size of the leaf surface, which, in turn, is related to interception of solar radiation by the plant canopy and its conversion into chemical bonds of organic compounds in the process of photosynthesis. The latter are used by plants in the process of forming vegetative mass and reproductive organs [52]. In accordance with this, the results of our studies showed a high positive correlation between the size of the leaf surface and the dry and fresh weight of plants, the number of formed beans and grains on the plants and the weight of 1000 seeds. The calculation of simple correlation coefficients between seed yield and seed protein content showed a negative correlation between these two indicators in each year of research, but in the variants of mineral fertilizer application, on average, during the 3 years of research, seed protein content and pea seed yield had a close positive relationship. McLnen et al. [50] also showed a positive relationship between seed yield and seed protein content for different levels of nitrogen supply. However, the relationships between these values for varieties and rainfall during the pea growing season were negative.

5. Conclusions

This study showed a significant increase in the values of the analyzed productivity parameters, seed yield, seed protein content and total protein yield per 1 ha, after the application of the biological preparation Rhizogumin and different doses of mineral fertilizers. Of the factors tested, mineral fertilization had a more significant effect on plant development and yield than seed inoculation. An higher value of plant morphological traits and productivity elements was also observed in the face of improved plant nutrient supply with fractional nitrogen fertilization (pre-sowing and BBCH 22–23) than a single pre-sowing dose. The best combination of the factors was a combination of seed inoculation and mineral fertilization at the rate of $N_{30}P_{45}K_{45}$ + N_{15}. This combination made it possible to increase the leaf area (by 66.9%), fresh and dry weight of the plants (by 34.9 and 71.3%, respectively), the number of beans and the number of seeds per plant (by 41.4 and 33.0%, respectively), the weight of 1000 seeds (by 12.0%), seed yield, protein content and total protein yield per ha (by 26.2, 11.1 and 43.5%, respectively) compared to the control.

Given the growing deficit in high-protein food production and the interest in greening agriculture, it is advisable to continue research into improving pea cultivation technology through the use of biological products that will increase seed yield, improve quality traits and reduce the pressure of chemicals on the ecosystem.

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Appendix A

Table A1. Average values of seed yield of peas for the interaction of experimental factors (inoculation × mineral fertilization) in each year of the experiment.

Inoculation	Fertilization	Seed Yield ($t\ ha^{-1}$)		
		2015	2016	2017
No inoculation	Control	2.31 a	4.07 a	2.32 a
	$N_{15}P_{15}K_{15}$	2.42 b	4.24 bc	2.56 b
	$N_{15}P_{30}K_{30} + N_{15}$	2.55 c	4.50 de	2.71 cd
	$N_{30}P_{30}K_{30}$	2.50 b	4.46 de	2.62 bc
	$N_{30}P_{45}K_{45} + N_{15}$	2.73 ef	4.66 fg	3.15 g
	$N_{45}P_{45}K_{45}$	2.69 de	4.61 ef	2.93 f
Rhizogumin	Control	2.52 c	4.20 ab	2.38 a
	$N_{15}P_{15}K_{15}$	2.60 cd	4.38 cd	268 bcd
	$N_{15}P_{30}K_{30} + N_{15}$	2.68 de	4.58 ef	2.79 de
	$N_{30}P_{30}K_{30}$	2.71 ef	4.57 ef	2.85 ef
	$N_{30}P_{45}K_{45} + N_{15}$	2.83 g	4.83 h	3.34 h
	$N_{45}P_{45}K_{45}$	2.79 f	4.77 gh	3.23 gh
Significance (*p* value)		**	**	***
$HSD_{0.05}$		0.0895	0.1504	0.1190
SE		0.017	0.029	0.023

The differences between data indicated by the same letter are not statistically significant. Significance (*p* value): ** $p \leq 0.01$; *** $p \leq 0.001$.

Table A2. Average values of protein content in pea seeds for the interaction of experimental factors (inoculation × mineral fertilization) in each year of the experiment.

Inoculation	Fertilization	Protein Content in Pea Seeds, (%)		
		2015	2016	2017
No inoculation	Control	20.42 a	18.71 a	19.29 a
	$N_{15}P_{15}K_{15}$	21.19 b	20.04 c	20.13 c
	$N_{15}P_{30}K_{30} + N_{15}$	21.34 c	20.63 e	20.68 ef
	$N_{30}P_{30}K_{30}$	21.37 c	20.42 d	20.57 d
	$N_{30}P_{45}K_{45} + N_{15}$	21.83 d	21.26 g	21.46 i
	$N_{45}P_{45}K_{45}$	21.44 c	20.79 f	20.72 f
Rhizogumin	Control	21.10 b	19.66 b	19.82 b
	$N_{15}P_{15}K_{15}$	21.46 c	20.52 d	20.61 de
	$N_{15}P_{30}K_{30} + N_{15}$	22.09 e	20.89 f	20.94 h
	$N_{30}P_{30}K_{30}$	21.83 d	20.58 e	20.76 g
	$N_{30}P_{45}K_{45} + N_{15}$	22.50 f	21.97 h	21.93 k
	$N_{45}P_{45}K_{45}$	21.94 d	21.15 g	21.85 jk
Significance (*p* value)		***	***	***
$HSD_{0.05}$		0.1178	0.1092	0.0746

SE		0.023	0.021	0.014

The differences between data indicated by the same letter are not statistically significant. Significance: (p value) *** $p \leq 0.001$.

Table A3. Average values of protein yield for the interaction of experimental factors (inoculation × mineral fertilization) in each year of the experiment.

Inoculation	**Fertilization**	**Protein Yield, (t ha^{-1})**		
		2015	**2016**	**2017**
No inoculation	Control	0.472 a	0.762 a	0.447 a
	$N_{15}P_{15}K_{15}$	0.512 b	0.842 b	0.515 b
	$N_{15}P_{30}K_{30} + N_{15}$	0.544 cd	0.944 e	0.560 cd
	$N_{30}P_{30}K_{30}$	0.534 c	0.911 cd	0.539bc
	$N_{30}P_{45}K_{45} + N_{15}$	0.596 fg	0.991 fg	0.676 f
	$N_{45}P_{45}K_{45}$	0.576 ef	0.958 ef	0.607 e
Rhizogumin	Control	0.532 b	0.834 b	0.472 a
	$N_{15}P_{15}K_{15}$	0.558 de	0.898 c	0.552 c
	$N_{15}P_{30}K_{30} + N_{15}$	0.592 fg	0.941 de	0.584 de
	$N_{30}P_{30}K_{30}$	0.591 f	0.940 de	0.592 e
	$N_{30}P_{45}K_{45} + N_{15}$	0.636 h	1.061 h	0.732 g
	$N_{45}P_{45}K_{45}$	0.612 g	1.009 g	0.706 fg
Significance (p value)		*	***	***
$HSD_{0.05}$		0.0195	0.0314	0.0252
SE		0.003	0.006	0.004

The differences between data indicated by the same letter are not statistically significant. Significance: (p value) * $p \leq 0.05$; *** $p \leq 0.001$.

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