

## Effect of seed inoculation and foliar fertilizing on structure of soybean yield and yield structure in Western Polissya of Ukraine

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**Abstract.** Growing soybeans requires adjustment of micronutrient nutrition on poorly fertile soils. Foliar fertilization can overcome the deficiency of micronutrients in plants in the most important period. Three factorial field experiment to study foliar fertilization with complex micronutrients, seed inoculation by *Bradyrhizobium japonicum* in two soybean varieties (Kassidy and ES Mentor) was conducted. Foliar fertilization with Quantum oil and WUXAL Oilseed significantly increased certain elements in soybean yield structure. Fertilizer WUXAL Oilseed with a higher concentration of Mo, Mn and B more effectively increased the number of pods and seeds from the plant and formed a higher yield in soybeans. Foliar fertilization with WUXAL Oilseed and Quantum oil increased seed yield to 3.00 t ha<sup>-1</sup> and 2.94 t ha<sup>-1</sup>, respectively in regard to variant without fertilizing, where yield was 2.71 t ha<sup>-1</sup>. Variety Kassidy had greater number of pods and seeds, seed weight in comparison to foliar fertilizing ES Mentor. Seed inoculation gives a stable increase in yield under different foliar fertilizations in varieties Kassidy and ES Mentor.

**Key words:** *Bradyrhizobium japonicum*, micronutrients, variety.

### INTRODUCTION

Symbiosis of nodule bacteria and the soybean root system is an important element in increasing productivity. Nodule bacteria *Bradyrhizobium japonicum* can persist in the soil for a long time, but inoculation of seeds is guaranteed to form enough nodes (Shcherbakova et al., 2018). Inoculation is an important source of nitrogen for soybean plants, but nodule bacteria can provoke a deficiency of micronutrients for soybean plant (Pacovsky, 1986). Deficiency of certain micronutrients can adversely affect the process of bud formation, flowering and soybean formation. Application of micronutrients before flowering is an effective way to compensate soil deficit of these elements in (Yasari & Vahedi, 2012).

Foliar fertilizing with  $H_3BO_3$  solution is inefficient, so application of available forms of complex micronutrients is more justified (Reinbott & Blevins, 1995). Complex fertilizers for foliar fertilization contain available forms of Mn, Mo, B and Zn (Hänsch & Mendel, 2009). Modern soybean genotypes are greater branched, so the B role in pod and seed formation is growing (Devi et al., 2012).

Positive effect of foliar Mo-fertilizing on yield is maximal in the flowering phase and decreases with late application (Campo et al., 2009). Soybean plants have varietal sensitivity to Mn, and foliar fertilization is more effective than soil application (Loecker et al., 2010).

Establishing the varietal sensitivity of soybean plant to foliar fertilizers with different concentrations of micronutrients allows to obtain stable soybean yields. Foliar fertilizing of inoculated and non-inoculated soybean plants can affect formation of elements of yield structure in different ways, what was tested on two soybean genotypes in a field conditions.

## MATERIALS AND METHODS

Field experiment was carried out in 2017–2019 in Kovel district Volyn region, Ukraine (51° 14' N; 24° 57' E) conditions of Ukrainian Western Polissya. Physical and chemical characteristics of sod-podzolic soil are: humus content - 1.2%;  $pH_{KCl}$  - 7.2, available nitrogen - 72.8 mg  $kg^{-1}$ ;  $P_2O_5$  in acetic acid extract - 26.0 mg  $kg^{-1}$ ; exchangeable potassium - 54.0 mg  $kg^{-1}$ ; B - 0.16 mg  $kg^{-1}$ ; Mo - 0.03 mg  $kg^{-1}$ ; Mn - 2.5 mg  $kg^{-1}$  of soil; soil density 1.58 g  $cm^{-3}$ . Soil characteristics were established by state institution 'Soil protective institute of Ukraine' (Volyn department).

### Climate conditions

Field experiment was conducted in 3 vegetation seasons (2017–2019). The average daily temperature of 1<sup>st</sup> vegetation season was 17.9 °C with maximum noticed in August (20 °C); precipitations sum was 311.4 mm. Second year (2018) of research was warmer than previous year. Average daily temperature was 19.2 °C with maximum in August (20.5 °C); precipitations sum was 429.2 mm. The last year (2019) of research was characterized by following parameters: daily average temperature was 18.6 °C with maximum obtained in June (21.8 °C), precipitations sum was 453.3 mm. All meteorological information collected from WMO #33173.

### Experiment design

The experimental design was Randomized Complete Block with four replications. Its included 3 factors. Factor A was soybean cultivars: ES Mentor (early maturity, thousand seed weight 195–215 g, semi-determinate) and Kassidy (mid-early maturity, thousand seed weight 165–185 g, indeterminate). Factor B was seed inoculation ('Legume Fix', *Bradyrhizobium japonicum* 532c,  $2 \times 10^9$  colony-forming units per 1 g) and variant without inoculation. Factor C was foliar fertilization by 'Quantum Oil' ( $P_2O_5$  - 5%,  $K_2O$  - 9%,  $SO_3$  - 3%, B - 0.6%, Zn - 1.2%, Cu - 1.2%, Mn - 1.2%, Mo - 0.015%, Ni - 0.01%, Co - 0.003%); WUXAL Oilseed (B - 6%, Mn - 5%, Mo - 0.25%, S - 3%); and variant without foliar fertilizing.

The size of elementary plot was 50 m<sup>2</sup> (25 m<sup>2</sup> was used for harvesting). Previous crop is winter wheat. Soybean was sown with 12.5-cm inter-row spacing with rate

65 seeds per square meter. Sowing time depended on soil temperature in 5 cm depth (sowing began at 10–12 °C). Seed was inoculated in the same day before sowing by 'Legume Fix' © (*Bradyrhizobium japonicum 532c*) with rate 2.5 kg t<sup>-1</sup> seed. Fertilizing system included 100 kg ha<sup>-1</sup> Azofoska (N<sub>16</sub>P<sub>16</sub>K<sub>16</sub>) before ploughing, 150 kg ha<sup>-1</sup> ammonium nitrate (N<sub>52</sub>) and 110 kg ha<sup>-1</sup> ammonium sulphate(N<sub>23</sub>) before sowing. Foliar fertilizers were applied due recommendations: WUXAL oilseed was applied in BBCH 60-66 with rate 2 L ha<sup>-1</sup>; Quantum Oil was applied in two terms - 2 L ha<sup>-1</sup> in BBCH 50-59 and 1 L ha<sup>-1</sup> in BBCH 71-73. Plant protection included application herbicide Bazagran (bentazone 480 g L<sup>-1</sup>) in BBCH 11-13(soybean) and Haruma (Quizalofop-p-ethyl, 125 g L<sup>-1</sup>) in BBCH 12-14 (cereal weeds).

### **Sampling**

30 plants from each variant were analyzed to establish elements of yield structure of soybean. The number of pods per plants, seeds per plants, seed weight per plant and thousand seed weight for each variant were measured. Soybean yield was established by harvesting and seed yield, seed weight per plant and thousand seed weight was calculated to 14% moisture.

### **Statistical analysis**

*Fisher LSD* was conducted for establishing significant difference between variants in yield structure (pods and seeds per plant, seed weight and thousand seed weight). *Analysis of variance* and *Tukey HSD* were conducted by Statistica 13.3 for year and average seed yield. Means of yield structure were presented with standard error (SE).

## **RESULTS AND DISCUSSION**

### **Elements of soybean productivity**

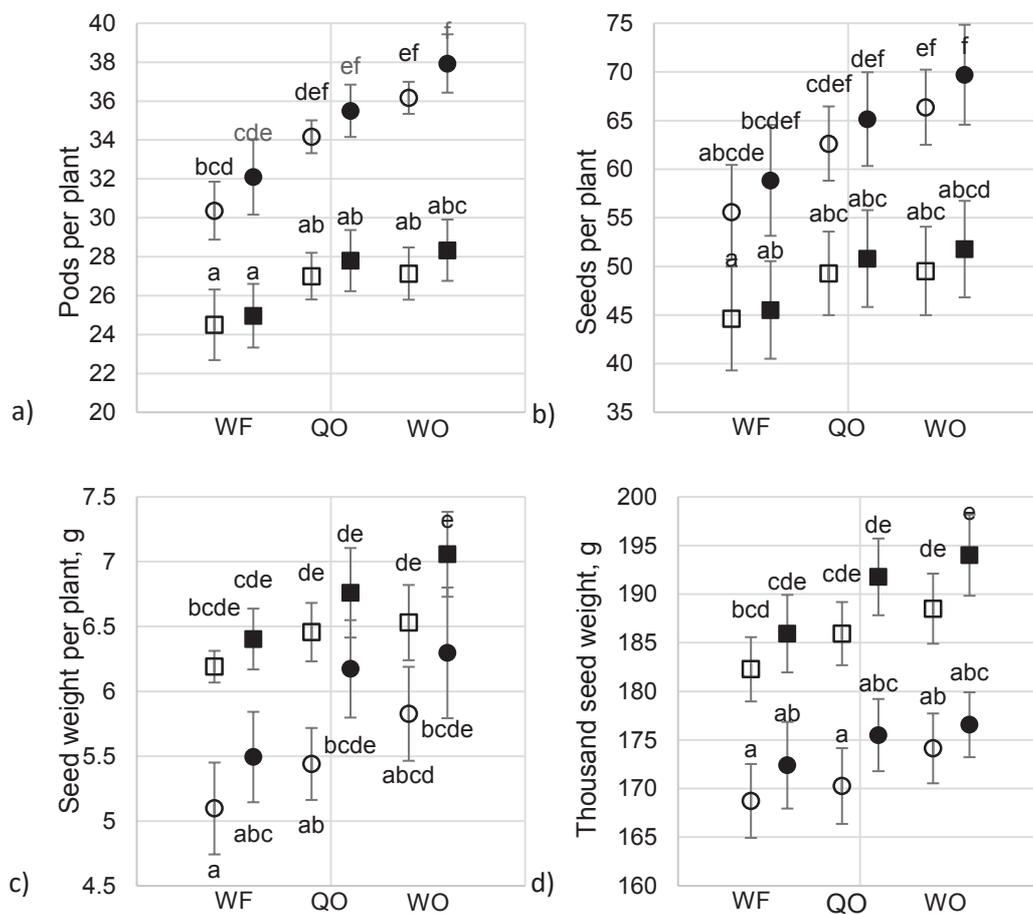
Varieties had a significant difference in terms of productivity. Foliar fertilizing and seed inoculation increased the average value of pods per plant and seed weight of each variety (Fig. 1), but some variants had no significant difference.

Variety Kassidy formed significantly more pods per plant than ES Mentor in all variants (Fig. 1, a). Seed inoculation had no significant effect 'number of pods per plant' in each cultivar, but number of pods increased by 4.8% at Kassidy and 3.1% at ES Mentor, compared to variants without inoculation. Foliar fertilizing significantly increased number of pods in the variety Kassidy in inoculated and non-inoculated variants.

Application of fertilizer with a higher B content (WUXAL Oilseed, B - 6%) also significantly increased the number of pods per plant compared with Quantum Oil (B - 0.6%) in the variants without seed inoculation. Increasing boron rate and time of application may influence on number of pods per plant. Foliar fertilizing with boron at a rate of 0.5 kg ha<sup>-1</sup> compared to variant without fertilizing significantly increased the number of pods, but a multiple increase in rate had less effect in this parameter (Devi et al., 2012). Applied boron rate in this research is low (0.018 kg ha<sup>-1</sup> in Quantum Oil, 0.12 kg ha<sup>-1</sup> in WUXAL Oilseed), so increasing the rate of boron by few times had a strong effect on the number of pods in variety ES Mentor. In other side, variety Kassidy had no significant difference between variants of different foliar fertilizing. Certain varieties may not respond to fertilization of boron by increasing the number of pods

(Rerkasem et al., 1993) or boron fertilization have a less effect in some varieties (Ross et al., 2006).

Seed inoculation has a less effect on number of pods per plant, but it can improve this parameter in combination with foliar fertilizing by micronutrients (Jarecki et al., 2016). From the other side, seed inoculation can have a significant effect on number of pods per plant, but it needs a specific condition (Afzal et al., 2010; Ntambo et al., 2017). The maximum efficiency of nodule bacteria occurs during flowering, while the boron content in the plant is significantly reduced at that moment (Yamagishi & Yamamoto, 1994). Foliar fertilizing with boron should compensate deficit and promote the normal development of pods.



**Figure 1.** Elements of soybean yield structure depend on foliar fertilization and seed inoculation (□ – Kassidy, non-inoculated; ■ – Kassidy, inoculated; ○ – ES Mentor, non-inoculated; ● – ES Mentor, inoculated; WF – without fertilizing; QO – Quantum Oil; WO – WUXAL Oilseed, different superscripts denote statistical significance at  $p \leq 0.05$  by Fisher's post-hoc test).

Number of seeds per plants had same trend as the number of pods, but it had a greater variation of the average mean (Fig. 1, b). Number of seeds per plant did not differ significantly from the studied factors in variety ES Mentor. Seed inoculation did not

have significant effect on seeds per plant in variety Kassidy, but foliar fertilizers was effective in this variety. Foliar fertilizing by WUXAL Oilseed significantly increased the number of seeds per plant compared to variant without fertilizers in inoculated and non-inoculated variants in both cultivars. Seed inoculation has a low effect on number of seeds per plant (Nyoki & Ndakidemi, 2018). Number of seed per plant has a large variation, but seed inoculation can improve number of seeds and decrease variation (Adeyeye et al., 2017).

Seed weight per plant of ES Mentor was much higher than its in variety Kassidy (Fig. 1, c). Foliar fertilization significantly increased seed weight in variety Kassidy by the treatment with WUXAL Oilseed. Fertilizing with Quantum oil had a same effect in inoculated variant in this variety, but efficiency was significantly lower in non-inoculated variant. Foliar fertilization increased seed weight per plant in variety ES Mentor, but this effect was without significant differences between variants. Seed inoculation increased the grain weight of the plant more than the number of beans, because it optimized the accumulation of dry matter, but did not affect the quantitative elements of yield structure. The same effect of seed inoculation is typical for other legumes, too (Shcherbakova et al., 2017).

Thousand seed weight varied the least depend on studied factors (Fig. 1, d). Variety ES Mentor formed larger seeds than Kassidy on average, which was manifested through seed weight per plant. Seed inoculation and foliar fertilizing had insignificant effect on this parameter, but they increase average means. Effect of foliar fertilizing by Mo has almost no effect on thousand seed weight, but affects its chemical content (Galindo et al., 2017). Inoculation has a low impact on thousand seed weight but improve efficiency of N accumulation in seed (Htwe et al., 2019).

### Grain yield

All studied factors significantly affected soybean yield (Table 1). The largest participation (85.7%) in yield variation was due to weather conditions (year). Seed inoculation and foliar fertilization was due to only 6.5 and 6.1% variation, respectively. Interaction ‘seed inoculation - foliar fertilizing’ and ‘seed inoculation - weather conditions (year)’ had the significant effects, and other interactions were statistically insignificant.

**Table 1.** ANOVA for soybean yield (average 2017–2019)

Effect	MS	<i>p</i>	Participance, %	Significance
Cultivar(A)	0.247	< 0.001	1.3	*
Seed inoculation(B)	1.262	< 0.001	6.5	*
Foliar fertilization (C)	1.167	< 0.001	6.1	*
Year (Y)	16.542	< 0.001	85.7	*
B x C	0.013	0.009	0.1	*
B x Y	0.039	< 0.001	0.2	*
Others	< 0.01	> 0.05	0.1	ns

Soybean yields varied over the years, but the effect of the studied factors was through. Yield of variety ES Mentor significantly exceeded variety Kassidy (Table 2). Difference in yield between inoculated and non-inoculated variants was greater than between varieties in 2017. Foliar fertilization also had a significant effect on yield

compared to variant without fertilization. There was no significant difference between the type fertilizers in 2017, but foliar fertilizing by WUXAL Oilseed significantly increased the yield of soybeans compared to variant without fertilization and Quantum Oil treated in 2018 and 2019. Application of WUXAL Oilseed insignificantly increased yield by 0.04 t ha<sup>-1</sup> more than Quantum Oil in 2017. Difference between the type fertilizers was significantly in years with high amount of precipitations, foliar application by WUXAL Oilseed additionally increased yield by 0.09 and 0.07 t ha<sup>-1</sup> compared to treatment with Quantum Oil in 2018 and 2019 accordingly.

**Table 2.** Grain yield of soybean, t ha<sup>-1</sup>

Cultivar (A)	Inoculation (B)	Foliar fertilizing (C)			Average		
		Without fertilizing	Quantum Oil	WUXAL Oilseed	A × B	A	B
2017							
ES Mentor	–	2.05	2.25	2.28	2.19 <sup>b</sup>	2.25 <sup>b</sup>	2.14 <sup>a</sup>
	+	2.17	2.39	2.37	2.31 <sup>c</sup>		2.27 <sup>b</sup>
Kassidy	–	1.92	2.15	2.21	2.09 <sup>a</sup>	2.17 <sup>a</sup>	
	+	2.05	2.30	2.36	2.24 <sup>b</sup>		
Average C		2.05 <sup>A</sup>	2.27 <sup>B</sup>	2.31 <sup>B</sup>			
2018							
ES Mentor	–	2.93	3.08	3.25	3.09 <sup>b</sup>	3.21 <sup>b</sup>	3.05 <sup>a</sup>
	+	3.11	3.43	3.45	3.33 <sup>d</sup>		3.29 <sup>b</sup>
Kassidy	–	2.81	3.05	3.17	3.01 <sup>a</sup>	3.13 <sup>a</sup>	
	+	3.07	3.32	3.37	3.25 <sup>c</sup>		
Average C		2.98 <sup>A</sup>	3.22 <sup>B</sup>	3.31 <sup>C</sup>			
2019							
ES Mentor	–	3.07	3.25	3.35	3.22 <sup>b</sup>	3.32 <sup>b</sup>	3.18 <sup>a</sup>
	+	3.22	3.48	3.52	3.41 <sup>d</sup>		3.37 <sup>b</sup>
Kassidy	–	2.95	3.18	3.27	3.13 <sup>a</sup>	3.23 <sup>a</sup>	
	+	3.14	3.39	3.45	3.33 <sup>c</sup>		
Average C		3.10 <sup>A</sup>	3.33 <sup>B</sup>	3.40 <sup>C</sup>			
Average 2017–2019							
ES Mentor	–	2.68	2.86	2.96	2.83 <sup>b</sup>	2.93 <sup>b</sup>	2.79 <sup>a</sup>
	+	2.83	3.10	3.11	3.02 <sup>d</sup>		2.98 <sup>b</sup>
Kassidy	–	2.56	2.79	2.88	2.75 <sup>a</sup>	2.84 <sup>a</sup>	
	+	2.75	3.00	3.06	2.94 <sup>c</sup>		
Average C		2.71 <sup>A</sup>	2.94 <sup>B</sup>	3.00 <sup>C</sup>			

Different superscripts (a, b, c ... in column and A, B, C in rows) denote statistical significance at  $p \leq 0.05$  by Tukey's post-hoc test.

Weather conditions were more favourable in 2018, so soybean formed a significantly higher yield. Inoculation increased the yield by 0.24 t ha<sup>-1</sup> on average, which almost reached the effect of foliar fertilizing.

Variations in yield across treatments and genotypes, had a significant difference in 2019, compared with other years but exceeded yield in 2018 by only 0.1 t ha<sup>-1</sup>. Difference between the varieties was similar to 2017 and 2018, but the efficiency of inoculation and foliar fertilizing decreased compare to 2018.

Inoculation increased yields by 0.19 t ha<sup>-1</sup> on average over 3 years. Low efficiency of inoculation may be due to large rate of nitrogen fertilizers (N<sub>75</sub>) applied before sowing. Formation the high yield of soybeans on poorly fertile soils is possible only with high rates of nitrogen fertilizers, and nitrogen fixation by nodule bacteria is only an additional source of nitrogen. Nodule bacteria may consume soil nitrogen instead of nitrogen fixation when applying nitrogen fertilizers, so the effect of inoculation on yield may be low or absent (Hungria et al., 2006).

Foliar fertilizing allowed significant increase of the soybean yield, and WUXAL Oilseed had a significantly higher effect than Quantum Oil treatment. Higher yield of fertilized varieties is due to the increase in the number of pods, seeds and, accordingly seed weight per plant. Foliar application with Mn and Zn microfertilizers can have a variety effect and responses to seed yield (Zolfaghari Gheshlaghi et al., 2019). Deficiency of certain micronutrients significantly reduce yields, even with a sufficient of others. Zn deficiency has a large impact on yield formation (Dimkpa et al., 2019). Application of micronutrient fertilizers has a positive effect on the formation of pods and seeds, and the rate of boron may play a key role in realizing yield potential (Sutradhar et al., 2017).

## CONCLUSIONS

Seed inoculation is an effective way to increase soybean yields. Inoculation had different effects on the formation of yield parameters, but its effect on seeds yield was the same in both varieties. Inoculation had a greater effect on the number of pods, seeds, and seed weight per plant in variety Kassidy. Foliar fertilizing by WUXAL Oilseed had a significant effect on the number of pods and seeds compared to variant without foliar fertilizing. Inoculation and foliar fertilizing increased some yield elements insignificantly, but they had a significant impact on yield in general. Application of Quantum Oil increased the yield of soybeans by 8.5%, but fertilizer with greater boron concentration (WUXAL Oilseed) increased yield on 10.7%. Inoculation increased yield on 6.8%. Fertilizers with high boron content allowed to form more pods and seeds, which led to an increase in seed weight and yield in both studied varieties.

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