

Influence of irrigation in different phenophases on the chemical composition of soybean grains

Mariya Sabeva¹, Alexander Matev^{1*}, Katya Uzundzhaliyeva¹, Stanislav Stamatov¹, Zlatina Ur¹, Bilyana Harizanova-Petrova², Radost Petrova² and Nikolay Minev²

¹ *Agricultural Academy, Institute of Plant Genetic Resources “K. Malkov”, 4122 Sadovo, Bulgaria*

² *Agricultural University, 4004 Plovdiv, Bulgaria*

*Corresponding author: sa6_m@abv.bg

Abstract

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The present study aims to determine the influence of single irrigations during the different phenophases of the vegetation period on components of the chemical composition of soybean seeds. Data from a long-term field experiment conducted in the Agricultural University-Plovdiv on alluvial-meadow soil were used. The variants of the experiment are: 1) no irrigation, 2) only the 1st watering, 3) only the 2nd watering, 4) only the 3rd watering, 5) optimum irrigation with three waterings given at pre-irrigation soil moisture 80% FC for the layer 0-60 cm. The results show that the crude protein content is highest when irrigated only during the flowering period. Single irrigations have little effect on the crude fat content of soybean seeds. Watering during flowering increases it between 0.3 and 0.7%. A single watering during the pod formation period (R3-R4) is ineffective in very dry years. It is weakly effective in medium dry years, and its effect increases in more favourable years, increasing the fat content by about 1%. Watering during the period of bean formation provides 64-68% of the maximum yield of fat, and the results for the other phenophases are similar.

Keywords: soybean; irrigation; protein; fat; water deficit

Introduction

Soybean is an important agricultural crop, according to Malikov & Vasilchenko (2014), and Peshkova et al. (2016), the seeds contain 37-42% protein and 19-22% fat, as well as many vitamins. This is a prerequisite for increasing soybean production worldwide, looking for opportunities for an economically justified increase in seed yield and lowering the cost price of production, including protein and fat. One of the ways to achieve this aim is correct irrigation, as regarding its influence on the chemical composition of soybeans, scientific information is contradictory. The results in the specialized scientific literature show in general that when maintaining higher soil moisture, the protein content of the seeds decreases

and that of fat increases, and the yield of both components increases due to an increase in seed yield (Kirnak et al., 2010; Tolokonnikov et al., 2018; Candoğan & Yazgan, 2016; Ghassemi-Golezani & Farshbaf-Jafari, 2012; Morsy et al., 2018). For example, Balakai (2000) found that even in wet years irrigation can reduce the protein content by 0.5-2.2%, but in the same time it increases the yield by 1.5-2.5 times. Seren (2013) found a more significant positive impact of irrigation during dry years. Nematov (2017) reported that for the conditions of the Ferghana Basin in Uzbekistan, the application of optimal irrigation provides 42-48 kg/da protein yield. When applying a biologically optimal irrigation regime, Kresović et al. (2017) reported significantly higher yield values (protein 109.2 kg/da and fat 56.3 kg/da). Conducting an experiment

applying a different irrigation regime to soybeans, Restuccia et al. (1992) found that irrigation water efficiency was significantly higher for fat than for protein, especially when irrigated with higher rates. Some studies show that irrigation increases the protein content and decreases the fat content of soybean seeds (Kresović et al., 2017a).

A limited number of studies are aimed at establishing the influence of the irrigation regime during the different phenophases of the vegetation period of soybean on the content of protein and fat in the seeds and their yield.

Crude protein

Based on the results of a field experiment, Kirnak et al. (2010) proved that the optimization of soil moisture provides an increase in the crude protein content of soybean seeds by more than 4% compared to that under non-irrigated conditions. Ashraf et al. (2013) are of the opposite opinion, finding in their study that maximum protein content is reached under non-irrigated conditions (up to 41.2%). The values are high (over 40%) without watering during the two phenophases and only one watering is applied. Mahmoud & El Far (1994) reported an effect of irrigation on crude protein percentage, stressing on the importance of irrigation carried out until pod filling begins. A significant increase in protein yield (49-57%) was also reported by Panchenko (2003), which was the result of the increase in pre-irrigation soil moisture from 70 to 80% of FC only during the period of flowering and bean formation.

Crude fats

According to Ghassemi-Golezani & Lotfi (2013), drought during any part of the reproductive period negatively affects fat synthesis, leading to further yield reduction. Detailed information on the influence of phenophases is presented by Ashraf et al. (2013). According to the authors, the phenophase of pod filling (R5-R6) is important, clearly showing the influence of the irrigation regime on its duration. The irrigation increases it, allowing fat accumulation over a longer period. For the same reason, crude fat content is lowest under non-irrigated conditions. Ali et al. (2009) found that irrigation, especially during the period of flowering and pod formation, lowered the crude fat content of soybean seeds.

The present study aims to determine the influence of single irrigations during the different phenophases of the vegetation period on components of the chemical composition of soybean seeds.

Material and Methods

To study the influence of single irrigations on the content and yield of the main chemical components, multi-year

data were used, including years with different character, with drought during different phenophases of the soybean growing season: very dry (with drought during the mass flowering-pod filling period – R2-R5), medium dry (with drought during the mass flowering and pod filling – phenophases R2 and R5-R6) and medium (with drought during the pod formation-filling period – R3-R6). The quantity and distribution of the rainfall during all experimental years required three irrigations to be carried out for obtaining an optimal irrigation regime. Thus, it is possible to irrigate only during one of the phenophases and cancel the remaining two irrigations and apply them during the other phenophases of plant. In this way, the individual influence of each watering on the content and yield of the investigated chemical components can be established, depending on the phenophase in which it was carried out. The experiment was conducted at the Agricultural University – Plovdiv, on alluvial-meadow soil. The variants of the experiment are: 1) no irrigation, 2) only 1st watering, 3) only 2nd watering, 4) only 3rd watering, 5) optimum irrigation with three waterings given at pre-irrigation soil moisture 80% FC for the layer 0-60 cm. The experiment was carried out in 4 repetitions, with the size of the trial plots 30 m², and the harvest plots – 10 m². The Biser variety (medium late) was used. The irrigation rate for all variants is 50 mm. The annual irrigation rate for variant 5 is 150 mm and for variants 2, 3, and 4 it is 50 mm. Irrigation was gravity type, using short closed furrows. In all variants of the experiment, the content and yield of the studied chemical components in the soybean seeds were determined as follows:

The crude protein content was determined by determining the total nitrogen by mineralizing the plant samples with concentrated sulphuric acid and selenium catalyst by the method of Kjeldahl (1883). The mineralized sample was then distilled using a Parnassus Wagner apparatus, and the total nitrogen values obtained were multiplied by a coefficient of 6.25.

The crude fat content was determined by the residual method of Soxhlet (1879) by extraction with petroleum ether.

The crude fiber was determined by the method of Hengerger and Stomann, by treating the plant material with solutions of acids and bases with a certain concentration.

The crude ash was determined by moderate incineration of the samples at a temperature of 550-600 °C.

Nitrogen-free extracts are obtained by the formula:

$$NFE = 100 - (\text{crude protein} + \text{crude fat} + \text{crude fiber} + \text{crude ash})$$

The results for the grain yields, protein, fat and carbohydrates were statistically processed using the software product ANOVA1, to establish the proof of the differences between

the treatments of the experiment. Yield is presented in units of kg/da (1 kg/da = 10 kg/ha).

Results and Discussion

Influence of single irrigations during different phenophases on crude protein content and yield

The results of the influence of single irrigations during the different phenophases on the crude protein content of soybean seeds are presented in Table 1. The change in the content of this chemical component, compared to that of non-irrigated soybeans is presented. According to the results of scientific studies, the later watering (closer to the pod-filling phase), should lead to a lower content of crude protein. This is logical, since the earlier single irrigation creates conditions for soil drying at a later stage of the vegetation, and this is a factor positively affecting the crude protein content. However, such a scenario can only be observed in drier years. All this is also confirmed by the conditions of the present experiment. It should be noted that the crude protein content increases with each of the single irrigations compared to optimally irrigated soybeans. In the mid-dry and dry experimental years, the effect of later irrigations is clearly visible. Thus, for example, the content is highest when irrigated during flowering (42.05 and 39.24%, respectively) due to the subsequent significant water deficit, especially during the period of pod filling. Watering applied only during the period of bean formation compensates

for a relatively weak drought from the previous period and greatly improves the conditions during the pod-filling period. Therefore, the protein content decreases to 40.92 and 38.98%, respectively. A single watering during the pod filling phenophase had a direct effect on crude protein content, reducing it to levels comparable to those under optimal irrigation (40.88 and 36.56%, respectively). These results clearly show that the level of soil water availability during the pod-filling period is critical for the crude protein content of the pods. In more meteorologically favorable years (in terms of precipitation), this influence was not observed.

In terms of yield, it is significantly less than that obtained by giving two or three irrigations, and as can be seen from the data in the table, it is often not significantly different from the yield under non-irrigated conditions, and yield losses relative to the optimum irrigated soybean are significant and exceed 30%. The losses are smallest when irrigation is made during the bean formation period, which is due to the positive effect it has on seed yield. Next is the variant with irrigation only during the mass flowering, and with irrigation only in the pod-filling phenophase, the losses are the most significant (50-60%). In average years in terms of precipitation, there are relatively favorable conditions until the beginning of the pod-filling period, therefore the irrigation given in R5 contributes to limiting losses, and the later one (in R6) reduces them to 25%. All the mentioned differences compared to the optimal variant are statistically proven.

Table 1. Crude protein content and yield at single irrigation in different phenophases of the vegetation period

Variant	CP %	Y CP kg/da	To dry		Proof	To optimum		Proof	GD kg/da	
			± kg/da	%		± kg/da	%			
Medium dry years with drought in R2 and R5-R6										
1	without irrigation (dry)	42.52	55.5	St.	100.0	St.	-83.7	39.9	C	5% = 11.3 1% = 15.1 0.1% = 20.0
2	single irrigation at R2	42.05	77.7	22.2	140.0	C	-61.5	55.8	C	
3	single irrigation at R4	40.92	93.5	38.0	168.4	C	-45.8	67.1	C	
4	single irrigation at R5	40.88	69.6	14.1	125.5	A	-69.6	50.0	C	
5	optimum (all three irrigations)	40.79	139.2	83.7	250.9	C	St.	100.0	St.	
Dry years with drought in R2-R5										
1	without irrigation (dry)	42.22	46.0	St.	100.0	St.	-75.4	37.9	C	5% = 12.1 1% = 16.2 0.1% = 21.5
2	single irrigation at R2	39.24	53.3	7.3	115.8	n.s.	-68.1	43.9	C	
3	single irrigation at R4	38.98	76.0	30.0	165.2	C	-45.4	62.6	C	
4	single irrigation at R5	36.56	50.2	4.2	109.2	n.s.	-71.2	41.4	C	
5	optimum (all three irrigations)	38.20	121.4	75.4	263.8	C	St.	100.0	St.	
Middle years with drought in R3-R6										
1	without irrigation (dry)	41.18	88.5	St.	100.0	St.	-51.0	63.5	C	5% = 9.8 1% = 13.1 0.1% = 17.4
2	single irrigation at R4	37.55	86.8	-1.8	98.0	n.s.	-52.7	62.2	C	
3	single irrigation at R5	40.49	95.7	7.2	108.1	n.s.	-43.8	68.6	C	
4	single irrigation at R6	42.22	104.7	16.2	118.3	B	-34.8	75.1	C	
5	optimum (all three irrigations)	38.55	139.5	51.0	157.6	C	St.	100.0	St.	

CP – crude protein; Y – yield

Influence of single irrigations during different phenophases on crude fat content and yield

Single irrigations have little effect on the crude fat content of soybean seeds. Compared to the non-irrigated variant, irrigation during flowering increases it by between 0.3 and 0.7% (Table 2). Single irrigation during bean formation (R3-R4) is ineffective in very dry years, and in terms of fat content, it is known that with increasing soil moisture availability, it increases, i.e. watering in a very dry year could not significantly change the values. The results in more favorable years confirm that thesis. The irrigation in medium-dry years has a positive effect, increasing the crude fat in the seeds by 0.6%, and in middle years by 1.0%. These changes are at first glance small but show the influence of the degree of drought and the specificity of this phenophase. Watering during the pod filling period has a positive effect on fat content, but only in drier years. It increases from 0.6% in medium dry years to 1.0% in dry years. These results prove that this phenophase is decisive and the level of soil moisture affects the fat content. Due to the absence of a significant and long-lasting water deficit until the beginning of the pod-filling, irrigation during phase R5 or R6 does not have a positive effect.

The additional yield resulting from single irrigations is significantly less than that obtained when two or three waterings are applied. Clear trends are also available here, but again they are more related to the effect of specific irrigations

on seed yield. Thus, for example, during the drier experimental years, the additional yield of crude fat is the largest (around and over 80%) when a single irrigation is carried out during the period of bean formation while the irrigation in the other two periods increases it by 30-40%. In more meteorologically favorable (middle) years, the advantage is again on the side of the irrigation in R4, but here the differences between the phenophases are much smaller, as for the R2 phenophase, they absent.

When compared with the optimal variant, logical results are observed, against the background of significant losses of seed yield. The relative fat yield varied within very narrow limits, notwithstanding the great difference in the character of the experimental years. The irrigation during bean formation provides 64-68% of the maximum fat yield. The values for the other two phenophases are close, which indicates approximately the same sensitivity regarding this indicator. This was observed in all experimental years.

Influence of single irrigations during different phenophases on the content and yield of nitrogen-free extracts (NFE)

Regarding the NFE content, there is a clear differentiation of the considered three sub-periods of the reproductive period, but in general, the irrigation carried out during any of them has a positive effect, expressed in an increase of their content in soybean seeds. The later irrigation leads to a

Table 2. Crude fat content and yield at single irrigation in different phenophases of the vegetation period

Variant	CF %	Y CF kg/da	To dry		Proof	To optimum		Proof	GD kg/da	
			± kg/da	%		± kg/da	%			
Medium dry years with drought in R2 and R5-R6										
1	without irrigation (dry)	20.21	26.4	St.	100.0	St.	-43.4	37.8	C	5% = 5.4 1% = 7.3 0.1% = 9.6
2	single irrigation at R2	20.50	37.9	11.5	143.7	C	-31.9	54.3	C	
3	single irrigation at R4	20.82	47.5	21.2	180.3	C	-22.2	68.1	C	
4	single irrigation at R5	20.83	35.5	9.1	134.5	B	-34.3	50.8	C	
5	optimum (all three irrigations)	20.45	69.8	43.4	264.6	C	St.	100.0	St.	
Dry years with drought in R2-R5										
1	without irrigation (dry)	21.48	23.4	St.	100.0	St.	-41.9	35.8	C	5% = 6.9 1% = 9.2 0.1% = 12.2
2	single irrigation at R2	22.19	30.1	6.7	128.7	n.s.	-35.2	46.1	C	
3	single irrigation at R4	21.43	41.8	18.4	178.5	C	-23.5	64.0	C	
4	single irrigation at R5	22.49	30.9	7.5	132.0	A	-34.4	47.3	C	
5	optimum (all three irrigations)	20.55	65.3	41.9	279.0	C	St.	100.0	St.	
Middle years with drought in R3-R6										
1	without irrigation (dry)	21.81	46.9	St.	100.0	St.	-32.6	59.0	C	5% = 5.4 1% = 7.3 0.1% = 9.6
2	single irrigation at R4	22.79	52.7	5.8	112.3	A	-26.8	66.3	C	
3	single irrigation at R5	21.02	49.7	2.8	106.0	n.s.	-29.8	62.5	C	
4	single irrigation at R6	21.46	53.2	6.3	113.5	A	-26.2	67.0	C	
5	optimum (all three irrigations)	21.96	79.5	32.6	169.5	C	St.	100.0	St.	

CF – crude fat; Y – yield

stronger effect on NFE accumulation (Table 3). The amount of carbohydrates in soybean seeds depends mostly on the conditions under which the pod-filling period occurs. When only one watering is applied during the earlier phenophases, and if the drought during the period R5-R6 is greater than the permissible one, the accumulation of the maximum amount of BEV is not possible. A single irrigation during the same period does not completely restore the optimal soil moisture, but only partially, and thus creates the necessary conditions for the synthesis of the maximum amount of carbohydrates in the soybean seeds, which reaches 24.7-25.8%. Single irrigations during the earlier phenophases provide a high NFE content, but compared to the above values, it is lower, especially as a result of irrigation during the R3-R4 period, when, in addition to vegetative growth, the plants must ensure the formation and development of the maximum number of beans. This negative difference compared to the result achieved with irrigation through R5 reaches from 1 to over 4%.

Influence of single irrigations during the different phenophases on the content and yield of lysine

The data about the influence of single irrigations on the content and yield of lysine are presented in Figure 1. There is a trend of inversely proportional dependence between the degree of water availability and the lysine content, i.e. the supply of one irrigation lowers the value compared to non-irrigated variants and increases them compared to those – under

optimal irrigation (case of 3 irrigations). According to the data in Figure 1A, no clear trend was observed in relation with the different phenophases, except for the smaller variation of the data of the experimental years. Lysine yield strongly depends on seed yield, with the positive effect of single irrigations being small compared to the losses relative to the optimal variant. Thus, for example, the additional yield is proven only with the irrigation given during the period of bean formation. All losses, however, are proven statistically at the highest rank. They are also affected by the conditions of the year, and in dry years they are significant (1.5-1.7 kg/da), even with irrigation during bean formation. Conversely, in more favorable years, losses are in the range of 0.4-0.8 kg/da.

Effect of single irrigations during different phenophases on crude fiber and crude ash content and yield

Fiber content is relatively weakly affected by the irrigation regime, but there is a tendency for it to decrease as a result of later irrigations, and it is better expressed when comparing irrigation during pod formation and irrigation during pod filling. The soil moisture provided by these two irrigations has a different influence on the processes going on in the plant related to productivity, which may be one of the reasons for the indicated differences (Figure 2A).

In terms of the yield, a relatively low almost equal efficiency of the single irrigations is observed, which is largely related to the seeds yield (Figure 2B). The impact is com-

Table 3. NFE content and yield at single irrigation in different phenophases of the vegetation period

Variant	NFE %	Y NFE kg/da	To dry		Proof	To optimum		Proof	GD kg/da	
			± kg/da	%		± kg/da	%			
Medium dry years with drought in R2 and R5-R6										
1	without irrigation (dry)	20.21	26.4	St.	100.0	St.	-43.4	37.8	C	5% = 5.4 1% = 7.3 0.1% = 9.6
2	single irrigation at R2	23.54	43.5	13.3	144.1	C	-33.9	56.2	C	
3	single irrigation at R4	24.28	55.5	25.3	183.7	C	-22.0	71.6	C	
4	single irrigation at R5	25.02	42.6	12.4	141.2	C	-34.8	55.0	C	
5	optimum (all three irrigations)	20.45	69.8	43.4	264.6	C	St.	100.0	St.	
Dry years with drought in R2-R5										
1	without irrigation (dry)	21.48	23.4	St.	100.0	St.	-41.9	35.8	C	5% = 6.9 1% = 9.2 0.1% = 12.2
2	single irrigation at R2	22.43	30.5	7.7	133.7	A	-49.7	38.0	C	
3	single irrigation at R4	21.92	42.7	20.0	187.6	C	-37.4	53.3	C	
4	single irrigation at R5	24.73	34.0	11.2	149.1	B	-46.2	42.4	C	
5	optimum (all three irrigations)	20.55	65.3	41.9	279.0	C	St.	100.0	St.	
Middle years with drought in R3-R6										
1	without irrigation (dry)	21.81	46.9	St.	100.0	St.	-32.6	59.0	C	5% = 5.4 1% = 7.3 0.1% = 9.6
2	single irrigation at R4	21.17	48.9	4.2	109.5	n.s.	-34.4	58.7	C	
3	single irrigation at R5	25.80	61.0	16.3	136.5	C	-22.3	73.2	C	
4	single irrigation at R6	24.54	60.9	16.2	136.3	C	-22.4	73.1	C	
5	optimum (all three irrigations)	21.96	79.5	32.6	169.5	C	St.	100.0	St.	

NFE – nitrogen-free extracts; Y – yield

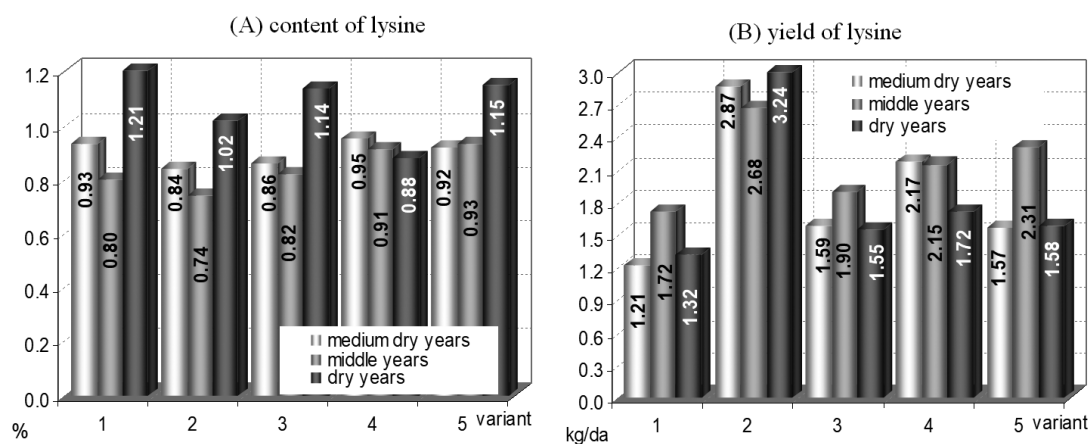


Fig. 1. Influence of single irrigations during the different phenophases on the content and yield of lysine

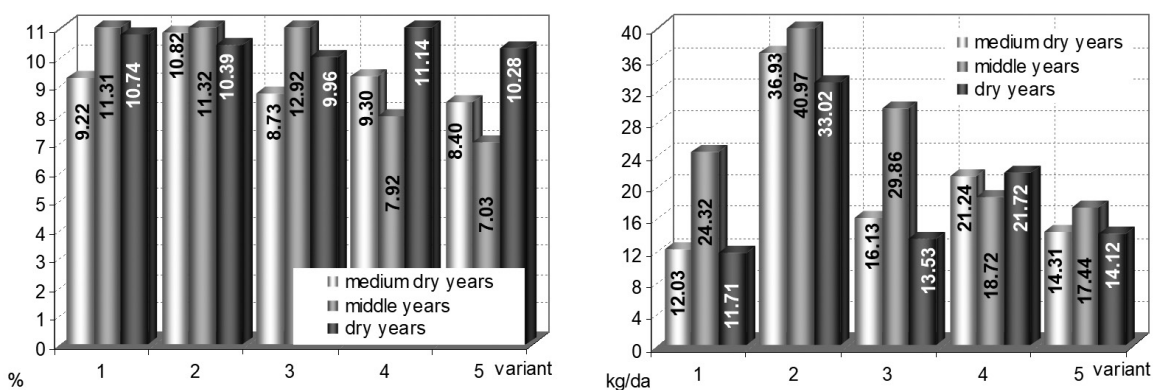


Fig. 2. Effect of single irrigations during different phenophases on crude fiber content and yield

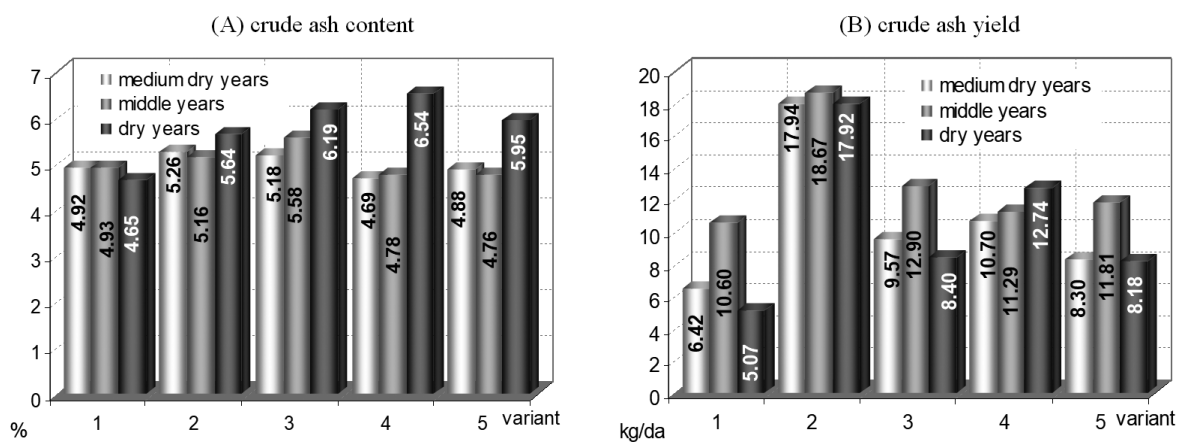


Fig. 3. Effect of single irrigations during different phenophases on crude ash content and yield

plex. Irrigation during pod formation provides a statistically significant higher yield of fibers, compared to those applied during the other two periods considered, which in turn lead to approximately the same results.

Figure 3 shows the content of crude ash in soybean seeds, established in the different variants of the experiment. According to data from Figure 3A, it increases more significantly as a result of the first watering, but no specific trend is observed related to the processes in the plant during the different phenophases, the water supply, and the ash content of the soybean seeds. The production of this component is also closely related to the production of seeds. In that connection, irrigation during the period of bean formation is most effective (Figure 3B).

Conclusions

The protein content is highest when irrigation during flowering is applied, due to the subsequent significant water deficit, including phases R5-R6. Watering applied only during the period of bean formation lowers water stress during R5-R6, and as a result, the protein content decreases. Irrigation only through R5-R6 has a direct effect on crude protein content, reducing it to levels comparable to those under optimal irrigation.

Single irrigations have little effect on the crude fat content of soybean seeds. Compared to the non-irrigated variant, irrigation during flowering increases it between 0.3 and 0.7%. Single irrigation during pod formation (R3-R4) is ineffective in very dry years. It is weakly effective in medium dry years, and its effect increases in more favorable years, increasing the fat content by about 1%. This irrigation provides 64-68% of the maximum fat yield, and the results of irrigation during the other phenophases are similar. Irrigation during the pod filling period has a positive effect on fat content, but only in drier years.

The single watering during R5-R6 increases the soil moisture, creating the necessary conditions for the synthesis of the maximum amount of carbohydrates in the soybean seeds (up to 24.7-25.8%). Single irrigations during the earlier phenophases provide a high NFE content, but compared to the above values, it is lower (from 1 to over 4%), especially for irrigation during the R3-R4 period. The reason is that, in addition to increased vegetative growth, it must ensure the formation and development of the maximum number of beans.

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