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#### EFFECT OF SOYBEAN SEED PRIMING ON GERMINATION ENERGY

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**Abstract:** Six varieties were chosen from three different maturity groups: Galina and NS Princeza (0), Sava and NS Apolo (I), and Rubin and NS Zita (II) in order to examine the effect of priming on germination energy depending on soybean seed age. The seeds were produced in the period from 2012 to 2014, i.e., one- to three-year-old seeds were used. The following treatments were used for priming: distilled water - hydropriming (H<sub>2</sub>O), ascorbic acid 250 mg/l (ASA), folic acid 15 mg/l (Fol.), hydrogen peroxide 1% ( $H_2O_2$ ), and control – non-treated seeds (C). Results showed that the effects of priming depended on the variety, seed age, as well as on the applied treatment. Some varieties reacted to priming very well, while others had an inhibited reaction, so germination energy was significantly reduced. Rubin variety had the most favourable reaction, whereby all treatments of this variety led to an increase of quality, with an average increase of 2-8%. The application of primers on one-year-old soybean seeds had a weaker effect than on two- and three-year-old seeds. However, one-year-old seeds had minor oscillations in quality due to priming. The application of priming on one-year-old seeds can lead to an increase or decrease of germination energy by 3%, while two- and threeyear-old seeds reached 7%. The results showed that seed priming in ascorbic acid had a positive effect, while the application of other primers did not affect the value of germination energy, nor did it lead to a decrease of its value. Ascorbic acid activity led to the largest increase in the quality of three-year-old soybean seeds by 2.83%, while the increase in the quality of two- and three-year-old seeds was 1.87%. Therefore, we can conclude that there is no universal use of one single primer, as it might not be suitable for each particular variety and can ultimately result in the seed quality decrease.

Key words: germination energy, priming, seed aging, soybean.

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#### Introduction

The maximum potential of germination and vigour of soybean seed is accomplished in the physiological maturity phase, while during aging, this value decreases (Balešević-Tubić and Miladinović, 2014). The aging process is accelerated due to unfavourable conditions of storage and under the effect of high temperature and relative air humidity (Dolatabadiand et al., 2008). During aging, unsaturated fatty acids, components of the lipid, turn into free radicals under the effect of enzymes and atmospheric oxygen. The changes which occur affect permeability of the cell membranes, which leads to loss of seed vitality (Vimala and Pratap, 2014). Aging has a harmful effect on enzymes which are essential for nutrition of the embryo and obtaining normal seedlings (Iqbal et al., 2002). The application of a particular solution for seed priming can encourage biological activity and hence increase germination capacity (Schopfer et al., 2001). Priming leads to the partial hydration of seed up to the moment when germination processes are initiated, but are not finished (Basra et al., 2005). This treatment increases seed performances, and enables quicker and more uniform germination and emergence (Berhanu and Gebremedhn, 2013). Furthermore, it ensures an optimal flow of molecular and biological processes during germination, stimulates activation of different enzymes, mobilises protein reserves and prepares cells for division (Soleimanzadeh, 2013). Useful effects of priming are related to nucleic acid repair and development, increased protein synthesis, and repair of cell membranes and mitochondria (McDonald, 2000). Likewise, seed antioxidant mechanism is recovered (Siri et al., 2013).

Priming has a practical agronomic significance, primarily in unfavourable environmental conditions, such as a high content of salt in the soil (Foti et al., 2008), low and high temperature (Wahid and Shabbir, 2005), etc. The application of this measure successfully improves seed germination of diverse plants, especially vegetables and grasses (Arif et al., 2007). Useful effects of immersion have also been determined in other field crops, such as sugar beet (Sadeghian and Yavari, 2004), barley (Abdulrahmani et al., 2007), and rape seed (Ghassemi-Golezani et al., 2010).

There is little information in the literature about the effect of priming on germination energy of naturally aged seeds. Therefore, the aim of this research was to examine how soybean seeds of a particular variety and age react to priming, as well as to determine which primer gives the best results.

## **Materials and Methods**

The experiment was carried out in 2015 at the Institute of Field and Vegetable Crops in Novi Sad. Testing was conducted on six soybean varieties from three maturity groups: Galina and NS Princeza (0), Sava and NS Apolo (I), and Rubin and NS Zita (II). One variety from each group was chosen, which had already been widely produced, while the other variety was new in the assortment. Three-, twoand one-year-old seeds were produced in the experimental fields of PSS Ruma in 2012, 2013, and 2014, with variations in annual agro-meteorological conditions. The seeds were stored in a commercial seed storing facility, in which storage conditions depended on environmental factors.

The following treatments were used for priming:

1. Distilled water – hydropriming  $(H_2O)$ ;

2. Ascorbic acid 250 mg/l (ASA);

3. Folic acid 15 mg/l (Fol);

4. Hydrogen peroxide 1%  $(H_2O_2)$ ;

5. Control – non-treated seeds (C).

After six hours of priming, seeds were dried until they reached 11% of moisture content. After that, 4x50 seeds were taken from each variant and tested using standard laboratory methods at 25 °C in the period of five days (ISTA, 2009).

The obtained results were statistically processed by the analysis of variance of a trifactorial split-split-plot experiment (A – variety, B – seed age, C – priming). Data were processed using the analytics software package Statistica 8, while results were sorted on the basis of the Duncan's test for the significance threshold of 5% (Hadživuković, 1991).

### **Results and Discussion**

The obtained results showed that the effect of seed priming, as a pre-sowing treatment, depended on variety, seed age, and the applied treatment. Some varieties reacted very well to this pre-sowing practice, while others had an inhibited reaction, thus significantly reducing germination energy. The variety Rubin had the most favourable reaction, whereby all treatments led to the increase of quality, by an average of 2–8%. The application of this pre-sowing practice led to significant differences in germination energy, even within the same maturity group. The smallest difference was determined within the 0 maturity group. Varieties Galina and NS Princeza reacted well to the use of all primers, except to distilled water, i.e., hydropriming. On average, the largest effect was accomplished with ascorbic acid which increased germination energy by 6%, i.e., by 4%. The varieties of the I maturity group, Sava and NS Apolo, mostly reacted to germination energy. A decrease of germination energy by 4–8% was observed in the variety Sava, while NS Apolo reacted only to the application of ascorbic acid (Table 1).

The greatest difference in reaction was determined within the II maturity group. While Rubin had the most favourable reaction to priming, particularly to the application of ascorbic acid, and both hydrogen peroxide and folic acid as well, this practice led to a decrease in quality of NS Zita by 5–7%. Examining a wheat sample, Dezfuli (2008) determined that the effect of priming on seed germination mostly depended on the genotype. Miladinov et al. (2014) determined different responses to priming by the seed of the same genotype but from different soybean lots.

Variety	Seed age	Treatment (C)					Mean	
(A)	(B)	H <sub>2</sub> O	ASA	Fol	$H_2O_2$	С	(A*B)	
Galina (0)	Three-year-old	21	28	28	29	22	27*	
	Two-year-old	65	77	75	70	70	72	
	One-year-old	95	98	96	96	94	96	
Mean (A*C)		60	68*	66*	65*	62		
NS Princeza (0)	Three-year-old	15	21	20	21	16	19*	
	Two-year-old	64	72	69	66	66	68	
	One-year-old	92	95	93	92	92	93	
Mean (A*C)		57	62*	61*	60	58		
Sava (I)	Three-year-old	23	27	22	21	30	23*	
	Two-year-old	78	72	68	70	78	72*	
	One-year-old	88	90	88	89	92	89	
Mean (A*C)		63*	63*	59*	60*	67		
	Three-year-old	23	37	34	24	32	30	
NS Apolo (I)	Two-year-old	71	81	81	69	78	76	
	One-year-old	91	94	93	92	91	93	
Mean (A*C)		62*	71*	69	62*	67		
Rubin (II)	Three-year-old	25	31	28	30	24	29*	
	Two-year-old	77	86	82	81	76	82*	
	One-year-old	93	95	92	93	90	93*	
Mean (A*C)		65	71*	67*	68*	63		
NS Zita (II)	Three-year-old	29	35	30	32	38	32*	
	Two-year-old	74	70	70	74	79	72*	
	One-year-old	86	88	87	87	90	87	
Mean (A*C)		63*	64*	62*	64*	69		
Mean(C)		62	67*	64	63	64	_	
*significant difference.								
LSD <sub>0.05</sub> Treatment 2.54		Variety x Seed age 2.87			Variety x Treatment 2.83			

Table 1. The priming effect on soybean seed germination energy (%).

The largest effect was accomplished with the use of ascorbic acid by 3%. Ascorbic acid is also an important metabolite included in many cell processes. First of all, it participates in cell division (De Gara et al., 2003). The exogenous application of ascorbic acid may affect a set of different processes in plants including seed germination, since it increases its absorption in different tissues, and participates in biosynthesis of other hormones like gibberellic acid and ethylene,

which are necessary in the germination process (Arrigoni and Detullio, 2000). Stasolla and Yeung (2001) determined the use of large quantities of ascorbic acid in the early stages of seed germination.

The reaction of a variety to the application of this pre-sowing treatment also depends on seed age. The use of primers on one-year-old soybean seeds had a weaker effect than on the two-or three-year-old seeds. On the other hand, fewer oscillations in the quality of one-year-old seeds were caused by priming. The application of primers can increase or decrease germination energy in one-year-old seeds by 3%, while this value reaches up to the 7% increase as seen in two- and three-year-old seeds, depending on the variety.

Three-year-old seed varieties Galina and NS Princeza achieved the 5% and 3% increase of germination energy, while there was no significant increase in the quality of two- and one-year-old seeds. The varieties from the I maturity group had an inhibited reaction to this practice in terms of seed quality, regardless of its age. The largest 7% decrease in the quality was noted in the three-year-old seeds of the variety Sava. The variety Rubin reacted positively in all cases, but germination energy in one-year-old seeds had a lower increase than in two- and three-year-old seeds. Unlike the variety Rubin, the application of this practice on the variety NS Zita resulted in a decrease of the quality by 3 to 7%. Improvement of seed quality depends on the level of deterioration, and the increase of germination energy can be achieved by priming seed into a particular solution, but only to a very limited extent (Wattanakulpakin et al., 2012).

Results showed that immersing seeds into ascorbic acid had a positive effect, while the application of other primers did not have any effects on the germination energy value, nor did it lead to the decrease of its value (Table 2).

Sand and (D)	Treatment (C)								
Seed age (B)	$H_2O$	ASA	Fol	$H_2O_2$	С	- Mean (B)			
Three-year-old	22.67*	29.83*	27.00	26.17	27.00	26.53*			
Two-year-old	71.50*	76.33*	74.17	71.67*	74.50	73.63*			
One-year-old	90.83	93.33*	91.50	91.50	91.50	91.73*			
*significant difference	2.								
$LSD_{0.05}$ Seed age 10.37			Seed age x Treatment 2.33						

Table 2. The effect of treatment and interaction with seed age on soybean seed germination energy (%).

Ascorbic acid had the largest effect on the increase of the quality of threeyear-old soybean seeds by 2.83%, while the increase of two- and three-year-old seed quality was the same -1.87%. The application of this practice on onion showed a higher increase of germination in older seeds (Patil and Manjare, 2013), while the best results were achieved in treatments of fresh seeds of vetch and sunflower (Karta et al., 2011). It was determined that the application of ascorbic acid and hydrogen peroxide on aged sunflower seeds can increase germination even by 12.23% (Dolatabadian et al., 2008). The same authors determined that the application of ascorbic acid on safflower seeds did not lead to positive results, while the application of hydrogen peroxide resulted in the increase of seed quality, since hydrogen peroxide acidifies the inhibitor which is found in seed coat and thus increases germination energy. The application of hydrogen peroxide on wheat had a stimulative effect on seed germination, since it increases the activity of peroxidase enzyme (Liheng et al., 2009).

### Conclusion

The results showed that the effect of this pre-sowing practice depended on variety, seed age, and the applied treatment. Some varieties reacted very well to priming, while others had an inhibited reaction, so the seed quality was significantly reduced. Hence, it was observed that the application of one solution on one variety affected the increase in the quality of some parameters, while it had an inhibitory effect on the quality of others. Therefore, we can conclude that there is no universal use of one single primer, as it might not be suitable for each particular variety and can ultimately result in the seed quality decrease.

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## UTICAJ POTAPANJA SEMENA SOJE NA ENERGIJU KLIJANJA

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#### Rezime

Da bi se ispitao efekat potapanja na energiju klijanja starog semena soje, za analizu je odabrano šest sorti iz tri različite grupe zrenja: Galina i NS Princeza (0 grupa), Sava i NS Apolo (I grupa), Rubin i NS Zita (II grupa zrenja). Seme je proizvedeno u periodu od 2012. do 2014. godine, odnosno seme starosti tri, dve i jednu godinu. Za potapanje su korišćeni sledeći tretmani: destilovana voda (H<sub>2</sub>O), askorbinska kiselina 250 mg/l (ASA), folna kiselina 15 mg/l (Fol.), vodonikperoksid 1% (H<sub>2</sub>O<sub>2</sub>) i kontrola – netretirano seme (C). Rezultati su pokazali da efekat potapanja zavisi od sorte, starosti semena, ali i tretmana koji se koristi. Neke sorte veoma dobro reaguju na potapanje, dok kod drugih dolazi do inhibitornog delovanja, pa se energija klijanja semena značajno smanjuje. Sorta Rubin je najbolje reagovala i jedino su kod ove sorte svi tretmani doveli do povećanja kvaliteta, u proseku od 2% do 8%. Primena prajmera kod jednogodišnjeg semena soje ostvarila je slabiji efekat nego kod dvogodišnjeg i trogodišnjeg semena. Međutim, kod jednogodišnjeg semena manje su oscilacije u kvalitetu usled potapanja. Kod jednogodišnjeg semena dejstvom prajmera energija klijanja može da se poveća odnosno smanji za 3%, dok se kod dvogodišnjeg i trogodišnjeg ta vrednost kreće do 7%. Rezultati pokazuju da je potapanje semena u askorbinsku kiselinu ostvarilo pozitivan efekat, dok primena ostalih prajmera nije uticala na vrednost energije klijanja ili je dovela do pada njene vrednosti. U proseku, dejstvom askorbinske kiseline najviše je došlo do povećanja kvaliteta trogodišnjeg semena soje - 2,83%, dok je povećanje dvogodišnjeg i trogodišnjeg bilo isto i iznosilo je 1.87%. Zbog toga se ne može govoriti o univerzalnoj primeni samo jednog prajmera, jer se može dogoditi da on ne odgovara određenoj sorti, ali i starosti semena, pa može doći do pada kvaliteta semena.

Ključne reči: energija klijanja, potapanje, starenje semena, soja.

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