

Effects of biologically active substances used in soybean seed treatment on oil, protein and fibre content of harvested seeds

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ABSTRACT

Procházka P., Štranc P., Pazderů K., Štranc J., Vostřel J. (2017): Effects of biologically active substances used in soybean seed treatment on oil, protein and fibre content of harvested seeds. *Plant Soil Environ.*, 63: 564–568.

In 4-year trials, soybean seeds were treated with the following biologically active substances: Lignohumate B (a mixture of humic acids and fulvic acids); Lexin (a mixture of humic acids and fulvic acids enriched with auxins); brassinosteroid (a synthetic analogue of natural epibrassinolide 24) and so-called 'Complex seed treatment' (a mixture of a saturated solution of sucrose, Lexin, the fungicide Maxim XL 035 FS and an adjuvant on the basis of pinolene). After harvesting soybean seeds from the individual treatments, they were analysed for oil, protein and fibre contents. The results show that the most effective method was the 'Complex seed treatment' which, compared to the untreated variant, significantly increased not only the yield but also the oil content of the seeds.

Keywords: *Glycine max*; biochemical composition; production; seed dressing

Soybeans are one of the world's leading protein components in compound feeds. For humans, soya is also important as an oil source, because soya is the second most important oilseed crop after oil palm. It contains a significant amount of Omega 6 and Omega 3 fatty acids, which are optimal in dietary terms. Compared to rapeseed oil, soybean oil does not contain erucic acid, which is very positive for human and animal health (Mousavi-Avval et al. 2011, Ramedani et al. 2011). One way to increase the production potential of soybean and thus the production of quality soybean oil is to prepare the biologically active seed prior to its sowing (Procházka et al. 2015). Seed treatment is a biological, chemical and physical (mechanical) process used to mitigate the negative effects of various external or internal influences. It improves its germination and vigour and thus promotes the formation of a healthy plant with increased production potential (Khanzada et al. 2002, Egli

et al. 2005). The process of seed treatment can be combined with the inoculation of legumes. It can be therefore said that seed improvement is one of the very cheap and highly effective methods of plant protection and stimulation of growth (Procházka et al. 2016).

Various growth regulators, enzymes, substances associated with plant bioenergy or even photosynthetic pigments forming protein complexes that participate in the conversion of energy of electromagnetic radiation into energy of chemical bonds can be considered as biologically active substances (Dřimalová 2005). A number of biologically active substances also have a beneficial effect on seed germination and subsequent growth of soybean plants. According to some authors, biologically active substances based on a mixture of synthetic auxins, humic acids and fulvic acids have been very beneficial. Similar performance was shown in many experiments using synthetic

analogues of some brassinosteroids that, among other things, positively interact with auxins. For example, gibberellins or carbohydrates can be included amongst the biologically active substances with anti-stress effects that act primarily on the cellular level (Kohout 2001, Chen et al. 2004, Anuradha and Rao 2007).

MATERIAL AND METHODS

The experiment was established to determine the effect of soybean seed treatment with biologically active substances on the formation of yield elements, yield and qualitative composition of the produced seeds (oiliness, protein content and amount of fibre). The following biologically active substances were used in the experiment:

Lignohumate B is a mixture of humic acids produced in the process of organic transformation from waste wood with the ratio of humic and fulvic acids 1:1 (Procházka et al. 2015).

Lexin is a concentrated solution of humic acids, fulvic acids and auxins supporting plant cell division and elongation. An improving influence to creation and growth of roots and increase of yield was observed (Procházka et al. 2016).

Brassinosteroids are a relatively new group of steroid phytohormones from the terpenic family. They were found in oil seed rape (*Brassica napus* L.) pollen in the USA in 1970 (Nováková et al. 2014). Substance No. 4154 (brassinosteroid), synthetic analogue of natural 24 – epibrassinolide (2 α ,3 α ,17 β -trihydroxy-5 α -androstan-6-on), was used in the experiment.

A complex treatment considered a mixture of saturated solution of saccharose, Lexin, fungicide Maxim XL 035 FS and surfactant agent pinolene (Agrovital).

Table 1. Scheme of pre-sowing seed treatment

Treatment	Dose per 20 kg of seed
Lignohumate B	25.7 mL, water
Lexin	6.5 mL, water
Brassinosteroid	2.2 mL substance 4154, water
	saturated solution of saccharose
	6.5 mL Lexin
'Complex treatment'	10 mL Agrovital
	20 mL Maxim XL 035 FS
Untreated control	200 mL water

Total volume of all solutions was 200 mL

Field trials were carried out during the growing season from 2012 to 2015 with a very early soybean cv. Merlin (000+). In order to maintain the uniformity of the methodology, seeds were treated each time immediately prior to sowing, according to the scheme shown in Table 1. The sowing rate was determined on the recommendation of a seed company that was 68 seeds per square meter for the cv. Merlin. In all cases (in all variants), the seed was inoculated with Nitraxon+.

The experiment was designed as long plots, with three replications (1000 m² each) at the Studeněves area, Czech Republic. Weather details of experimental years and locality are presented in Table 2.

The pre-crops of soybean were spring barley, winter wheat, spring barley and winter wheat, in that order from 2012 to 2015, respectively. For all experimental variants, the same growing technology was used:

- Stubble breaking with disc harrow directly after pre-crop harvest;
- Chisel ploughing to 30 cm;
- NPK 15 fertilizing (15.0% N, 6.6% P, 12.5% K), dose 200 kg/ha before sowing in spring;

Table 2. Characterization of the experimental location

Year	Sowing date	Harvest date	Altitude (a.s.l. m)	Average annual temperature (°C)	Annual sum of precipitation (mm)	pH	P	K	Mg	Ca
							(ppm)			
2012	19.4.	16.9.	302	8.70	653	6.8	69	311	178	3933
2013	23.4.	10.10.	306	8.20	684	7.1	107	295	165	3487
2014	21.4.	21.10.	314	9.80	587	7.3	103	260	174	4250
2015	22.4.	13.10.	325	9.80	491	6.8	68	344	213	4250

All soil blocks are loam, arenic cambisol.

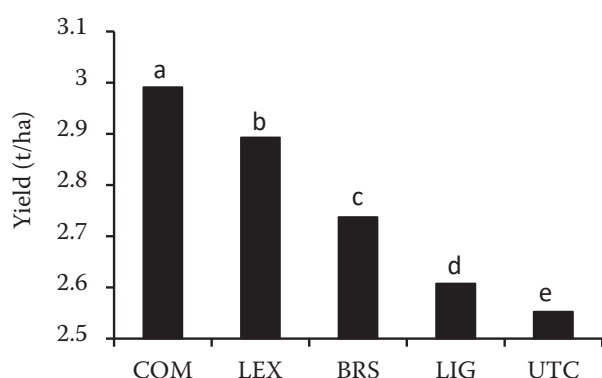


Figure 1. Average yields in soybean seeds by variants (2012–2015). Different letters are statistically significant. COM – complex treatment; LEX – Lexin; BRS – brassinosteroids; LIG – lignohumate; UTC – untreated control

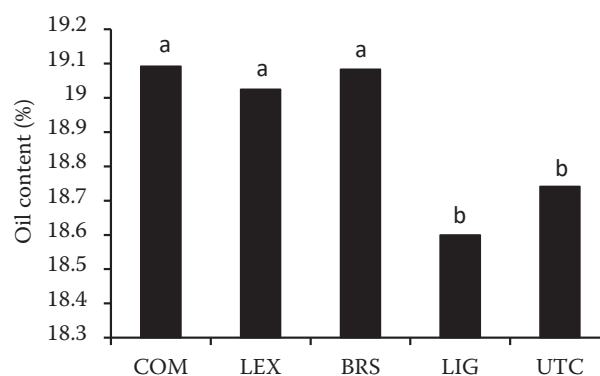


Figure 2. Oil content in soybean seeds by variants (average 2012–2015). Different letters are statistically significant. COM – complex treatment; LEX – Lexin; BRS – brassinosteroids; LIG – lignohumate; UTC – untreated control

- Pre-sowing tillage – 2 × cultivator to the depth of 6 cm;
- Seed treatment and sowing;
- Pre-emergent herbicide treatment;
- Harvest.

After the soybean harvest, the seeds were analysed for oil, protein and fibre contents using a NIR spectrophotometer (OmegAnalyzer G Bruins Instruments, Puchheim, Germany).

The results of the field trial were processed by the General Linear Model (GLM ANOVA) using the SAS statistical program, version 9.4 (Carry, USA). Differences between the mean values were evaluated by the Tukey's *HSD* (honestly significant difference) test at the level of significance $P = 0.05$.

RESULTS AND DISCUSSION

The results show that soybean seed treatment with biologically active substances has increased its yield (Figure 1).

The highest average seed yield was given in the COM variant, namely 2.99 t/ha. Similar results were achieved in the LEX treatment (2.89 t/ha). All differences between treatments were significant against control (Table 3). The highest seed yield was given in 2013 for all variants. It was caused by a sufficient amount of precipitation and optimal temperatures at the time of seed production in the pods.

Lexin, as mentioned above, is a mixture of humic acids, fulvic acids and auxins, and the synergy of these components, especially in the early growth phases, promotes faster division and growth of cells, tissues and vascular bundles, and thus the growth and formation of the plant as a whole. The positive effect of Lexin and brassinosteroids was also shown at experiments by Štranc et al. (2008) or Adamčík et al. (2016). The positive influence of auxins on the division and growth of plant cells is also mentioned by Procházka et al. (2015). Brassinosteroids often act in synergy with auxins (especially IAA), which is also the cause of the positive results of the variant treated with

Table 3. Results of the statistic evaluation (average of the years 2012–2015)

The parameter monitored	COM	LEX	BRS	LIG	UTC	<i>HSD</i>
Yield of seeds	2.99 ^a	2.89 ^b	2.74 ^c	2.61 ^d	2.55 ^e	0.0545
Oil content	19.09 ^a	19.03 ^a	19.08 ^a	18.60 ^b	18.74 ^b	0.2413
Protein in seed	33.35 ^{ab}	33.01 ^{bc}	32.49 ^c	33.83 ^a	33.44 ^{ab}	0.685
Fibre content	4.95 ^{ab}	4.93 ^{ab}	4.96 ^a	4.90 ^{bc}	4.85 ^c	0.0543

Means with the same letters are not statistically significant. *HSD* – honestly significant difference; UTC – untreated control; LIG – lignohumate B; BRS – brassinosteroid; LEX – lexin; COM – complex treatment

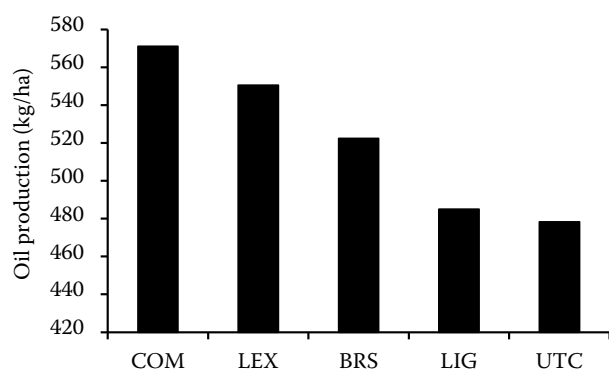


Figure 3. Oil production per hectare by variants (average 2012–2015). COM – complex treatment; LEX – Lexin; BRS – brassinosteroids; LIG – lignohumate; UTC – untreated control

brassinosteroid (a synthetic analogue of natural 24-epibrassinolide). A similar effect of brassinosteroids and auxins as found in this study is also reported by Gomes (2011) and others.

Figure 2 shows the oiliness of the seeds in each variant. The results show that soya seed treatment with biologically active substances (containing especially phytohormones) significantly increased the oil content of the produced seeds. The highest average oil content in the seeds was provided in the variant 'Complex treatment' (19.09%). Very similar results were achieved in the treatments with brassinosteroid (19.08%) and Lexin (19.03%), all significantly different from the untreated control (Table 3).

From the economic point of view, it is very important that the COM treated soybean seed, compared to the untreated control, contributed to 19.4% increase in oil production per hectare (Figure 3). In absolute terms, this means an increase in oil

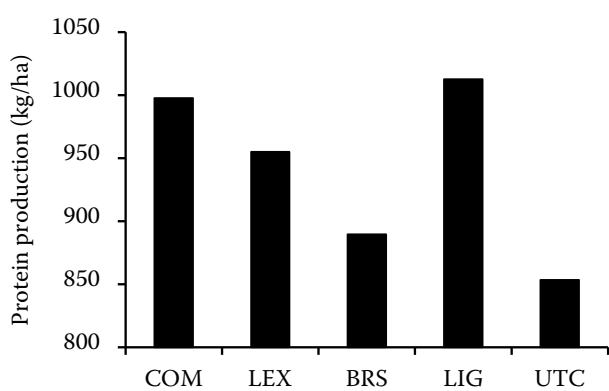


Figure 5. Protein production per hectare by variants (average 2012–2015). COM – complex treatment; LEX – Lexin; BRS – brassinosteroids; LIG – lignohumate; UTC – untreated control

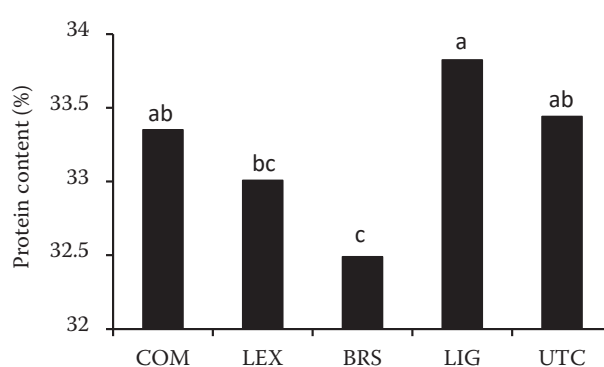


Figure 4. Protein content in soybean seeds by variants (average 2012–2015). Different letters are statistically significant. COM – complex treatment; LEX – Lexin; BRS – brassinosteroids; LIG – lignohumate; UTC – untreated control

production of 92.7 kg/ha, thus increasing its sales by 75.2 EUR per hectare at current prices (Rotterdam, May 2017). In the case of Lexin, oil production increased by 14.2% compared to the untreated control and thus an economic contribution of 53.7 EUR per hectare. Treatment with brassinosteroid also appears to be economically efficient as oil production increased by about 10% (brassinosteroid is not yet available in the Czech domestic market). For the sake of completeness, it should be noted that the approximate price of Lexin for soybean seed treatment is 2.1 EUR per hectare and in the case of 'Complex treatment' it is 8.6 EUR per hectare. The positive effect of Lexin and brassinosteroid on oil content also confirms the results of Štranc et al. (2008). Very good results of brassinosteroids utilization for the yield formation and production of substances usable for

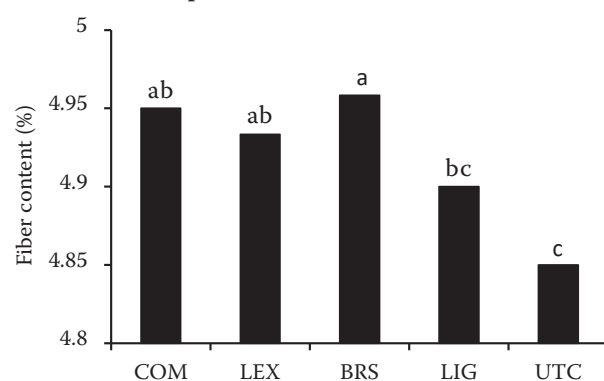


Figure 6. Fibre content in soybean seeds by variants (average 2012–2015). Different letters are statistically significant. COM – complex treatment; LEX – Lexin; BRS – brassinosteroids; LIG – lignohumate; UTC – untreated control

doi: 10.17221/702/2017-PSE

man were confirmed by Hradecká et al. (2009) in their experiment with sugar beet.

Soybean oil production is usually linked to protein content, as exemplified, for example, by Panthee et al. (2005). The established oiliness and protein content (Figures 2, 4) confirm the generally valid fact that the proportion of these substances in soybean is indirect (Figures 3 and 5).

From the values of Figure 6, it is obvious that the seed treatment with biologically active substances slightly increased the fibre content of the harvested seeds (statistically significant to untreated control except the LIG variant – Table 3).

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Received on November 8, 2017

Accepted on December 1, 2017

Published online on December 6, 2017