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RESPONSE OF WINTER OILSEED RAPE (*Brassica napus* L.) ON SOIL APPLIED HUMUS PREPARATION AND FOLIAR POTASSIUM FERTILIZER

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Abstract. The study aimed to evaluate the effect of soil application of the humic preparation Humistar and/or foliar application of the potassium fertilizer Drakar on biometric features, overwintering and state of nutrition, as well as the quantity and quality of winter oilseed rape yield. This study was based on a single-factorial field experiment conducted over 2006-2009 in Chrząstowo, on mesic Typic Hapludalf soil of a granulometric composition of light loam, in a randomized block design, in four replications. The effect of the following was evaluated: Humistar (12% humic acids and 3% fulvic acids), applied pre-sowing into soil, at a rate of 40 $dm^3 \cdot ha^{-1}$ (T1); Drakar (25.7% K and 3% N), applied in autumn after formation of 6 leaves (2 $dm^3 \cdot ha^{-1}$) and in spring, at the beginning of falling petals (4 $l \cdot ha^{-1}$) (T2); combined application of Humistar and Drakar (T3); and the control (T4). In this study has been shown that Drakar had a positive effect on seed yield of oilseed rape. The use of this fertilizer increased the chlorophyll index in the autumn and improved overwintering in relations to the control only when the earlier application of Humistar was made. Further studies are necessary to evaluate the effect of these preparations in weak soils with low content of organic matter and nutrients.

Key words: leaf greenness index, oil content, overwintering, seed yield

INTRODUCTION

In recent years, the use of biostimulants in sustainable agriculture has been growing [Nardi *et al*. 2002]. The global market for biostimulants is projected to increase 12% per

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year [Calvo *et al*. 2014]. Humic substances constitute an important group of biostimulants. The biostimulant effect of humic substances is characterized by structural and physiological changes related to nutrient uptake, assimilation and distribution. They can induce shifts in plant metabolism related to abiotic stress tolerance [Canellas et al. 2015]. It has been proved that they affect the activity of enzymes [Lotfi *et al.* 2015], exhibit stimulatory effects on plant cell growth and development [Muscolo *et al*. 2013] and stimulate roots growth and macronutrient uptake [Nardi *et al*. 2002, Verlinden *et al.* 2009, Billard *et al*. 2014]. Humic substances appear to display an hormone-like activity [Nardi *et al*. 2002, Trevisan *et al*. 2010]. Their impact on plant growth depends on the source, concentration and molecular weight of humic fractions [Muscolo 2013, Nardi 2002]. In plants from the family Brassicaceae it has been indicated that the application of humic substances increased the total marketable yield and head diameter of broccoli as well as quality parameters [Selim and Ali Mosa 2012]. Other studies have shown that the application of leonardite, which is readily available and high in humic acid, had a positive significant effect on dry matter yield of oilseed rape [Akinremi *et al*. 2000].

Supply in macroelements, including potassium, has also an essential influence on oilseed rape growth. At the flowering stage oilseed rape takes up between 140 and 300 kg $K \cdot ha^{-1}$ [Szczepaniak 2015]. Potassium balance affects taking up by oilseed rape other elements, which modifies plant growth and yield. The influence of fertilization with K was proved to affect an increase in dry matter accumulation in plants, as well as the LAI index [Yasari and Patwardhan 2006]. Cheema *et al.* [2012] report that increasing rates of K enhanced leaf area index and crop growth. Many works have indicated that potassium balance has an impact on taking up other nutrients. In conditions of K deficiency, there was observed a reduced uptake of N [Gaj 2011, Szczepaniak 2015] and P [Wang *et al*. 2015]. Cooperation of K and S fertilization in affecting seed yield was also indicated [Govahi and Saffari 2006]. In the study by Wang *et al*. [2015] K deficiency decreased individual growth and seed yield by 14,4%. An increase in seed yield under the influence of soil application of potassium was also indicated in the studies by Govahi and Saffari [2006], Amanullah and Malhi [2011b], Cheema *et al.* [2012] In contrast, the studies by Yasari and Patwardhan [2006] as well as Quezada *et al*. [2010] no response of yield was proved. According to Szczepaniak [2015], the response of seed yield depends on the content of K in vegetative parts of oilseed rape during the seed-filling period. Rates of applied potassium should be adapted to the quantity of yield and soil abundance of potassium [Gaj 2011]. However, even at the average K content in soil, plants may be insufficiently nourished with this element [Gaj 2011]. One of the causes might be exhaustion of resources of this element from the rhizosphere [Shi *et al*. 2004], which can prove the usefulness of its foliar application.

This study aimed to evaluate the effect of soil application of the humic preparation Humistar and/or foliar application of the potassium fertilizer Drakar on biometric features, overwintering and state of nutrition, as well as the quantity and quality of winter oilseed rape yield.

In the research hypothesis it was assumed that the application of humic substances and/or foliar potassium fertilization will have a favourable effect on the quantity and quality of winter oilseed rape, by stimulation of physiological processes and growth and taking up nutrients.

MATERIAL AND METHODS

The study was based on a strict, one-factorial field experiment with winter oilseed rape, located at Chrząstowo (53°09′ N; 17 °35′ E), conducted in the randomized block design in four replications. The experiment compared the effects of application of the humic preparation Humistar and/or liquid foliar fertilizer Drakar in comparison with the control (without applications). The experiment was conducted over three growing seasons: 2006-2007, 2007-2008 and 2008-2009, on soil classified as mesic Typic Hapludalfs [Soil Survey Staff, 2010], soil quality class IIIa, of granulometric composition of light loam. This soil is characterized by a high content of available potassium and a very high content of available phosphorus, average content of organic carbon and a neutral reaction (Table 1). The winter oilseed rape cultivar Winner was sown at 22-24 August, at a density of 70 seeds·m^{-2}, on plots with an area of 12 m^2. Pre-sowing mineral fertilization was applied to soil, at rates taking into consideration the soil abundance: 20-33 kg·ha^{-1} P, 75-100 kg·ha^{-1} K, 16-25 kg·ha^{-1} N and also Humistar was applied in a dose of 40 l·ha^{-1} dissolved in water 250 l·ha^{-1}. Humistar is an extract made from leonardite (highly oxygenated form of brown coal from Canada) [Verlinden *et al*. 2010]. The foliar fertilizer Drakar (25.7% K and 3% N) was applied twice: in autumn, after formation of 6 leaves (3-4 October), in an amount of 2 l·ha^{-1} and in spring, at the beginning of falling petals (BBCH 65), at a rate of 4 l·ha^{-1}. Moreover, in spring there were applied as follows: ammonium nitrate (07-08 March – 135 kg·ha^{-1} N and 29--30 March – 50 kg·ha^{-1} N) and ammonium sulfate (19-20 March – 21 kg·ha^{-1} N + 24 kg·ha^{-1} S). Weeds were controlled in autumn using the following active substances: metazachlor 999 g + quinmerac 249 g·ha^{-1} and quizalofop-p-ethyl 50 g·ha^{-1}. To reduce the incidence of fungal diseases, metconazole 60 g·ha^{-1} was applied in autumn, and in spring – tebuconazole 187.5 g·ha^{-1} (beginning of April) and picoxystrobin 250 g·ha^{-1} (middle of May). Insecticides of various active groups were applied against pests: one application in autumn and 3-5 in spring. Desiccants were used at the end of maturation, and 7-12 days later harvest was performed with the single-stage method.

Table 1. Chemical properties of the soil (0-30 cm)

Year	pH in 1M KCl	C_{org} g·kg^{-1}	$N_{tot} – N_{og}$ g·kg^{-1}	P mg·kg^{-1}	K mg·kg^{-1}
2006	6.60	7.87	0.87	126.4	167.5
2007	6.54	8.11	0.92	151.2	180.0
2008	6.98	8.10	0.90	240.7	207.5
Mean	6.71	8.03	0.90	172.8	185.0

The root crown diameter was measured prior to the end of autumn growth (middle of November), on successive plants in a row with the length corresponding to an area of 1 m^2. The evaluation of overwintering was conducted based on the difference in plant density before winter and in spring, at the same place on each plot, on the row length corresponding to 1 m^2. The seed yield from each plot was weighed several days after threshing, its humidity was determined (with the drying method), and then it was counted over the fixed humidity 8%. Thousand seed weight was determined after 4-5 months from harvest, based on 200 seeds from each plot. The leaf greenness index (SPAD) was evaluated using the Chlorophyll Meter SPAD-502. It measures the difference in light absorption with the wavelength of 650 nm, (the maximal light absorption by chlorophyll a and b), and 940 nm (light kept by the leaf tissues). The quotient of those differences is displayed as SPAD (*Soil-Plant Analyses Development*) units and is called the leaf greenness index. Measurements were made prior to the end of autumn growth, at the beginning of November, (BBCH 19), and in the middle of April, (BBCH 55), on 30 fully formed youngest leaves on each plot. The oil content was determined using the Dionex ASE150 apparatus for fast extraction. The seeds, ground and sieved through laboratory screen with 1mm mesh, were dried to the fixed weight at 105ºC, and then cooled and placed in the desiccator. Extraction was conducted in samples of 0.5 g at 135ºC using changing high pressure of inert gas – nitrogen 5.0 and solvents hexane: acetone (4:1). The time of extraction was 40 minutes. After the extraction the cell was cooled in the desiccator for at least 24 h, and then weighed together with the sample. The loss of sample weight related to the weight of the initial sample determined the proportion of oil in the initial sample. Weighing was performed to an accuracy of 0.001 g. Chemical analyses were conducted with the use of devices bought as part of the project 'Realization of the 2nd Stage of the Regional Innovation Centre' co-funded from the European Regional Development Fund as a part of the Regional Operational Programme for the Kuyavian-Pomeranian Voivodeship for 2007--2013.

The obtained results were subjected to the statistical analysis. The analysis of variance of single experiments in the years and the synthesis from the years in the mixed model were made using the statistical program Analysis of variance for orthogonal experiments by the University of Technology and Life Sciences in Bydgoszcz. Significance of differences for the results were assessed with Tukey's test, assuming the significance level P = 0.05.

RESULTS AND DISCUSSION

The weather conditions in the region of conducting the field experiments varied during particular growing seasons of winter oilseed rape (Table 2). The season 2006--2007 was the warmest. In that season the period from sowing to winter rest was the longest. After twenty very warm days of January (the average ten-day air temperature 6.0 and 5.7^{o}C, respectively) there was a cold snap, with temperature decreasing to –13.1ºC near the ground, without any snow cover. In such conditions the plants were particularly susceptible to the action of low temperatures and their overwintering in that season was the weakest in the three-year period of the study (Table 3).

The total precipitation during the autumn growth from September to November was similar in successive years of the study, 2006-2008. However, in the period from spring start of growth to technical maturity, the least precipitation was measured in 2008.

Table 2. Temperature and precipitation from sowing to harvest of winter oilseed rape

Month	Years							
	2006-2007	2007-2008	2008-2009	1980-2009	2006-2007	2007-2008	2008-2009	1980-2009
	Temperature, °C				Precipitation, mm			
August	17.4	18.1	17.7	18.6	164.0	29.4	101	58.4
September	15.9	12.7	12.7	13.3	55.6	39.5	27.3	44.7
October	10.3	7.2	8.5	8.7	8.5	22.6	57.9	34.7
November	5.5	1.7	4.3	2.64	32.0	27.3	22.1	31.8
December	3.9	0.83	0.4	-0.1	32.6	35.9	33.5	38.5
January	3.4	0.7	-3.4	-1.5	73.0	57.7	14.5	31.1
February	-0.8	3.2	-1.2	-0.7	32.8	12.1	27.4	25.2
March	5.5	3.1	2.7	2.7	55.2	53.5	43	35.3
April	9.0	8.0	8.0	8.3	16.6	40.0	0.9	28.7
May	14.3	13.7	12.5	13.9	83.5	13.8	77.7	48.4
June	18.2	17.3	14.9	16.7	112.0	19.6	107	68.9
July	18.0	18.9	18.8	19.1	88.9	65	96.8	71.6
Mean/Total	10.1	8.8	8.0	8.5	754.4	416.4	609.1	517.3

Table 3. Diameter of the root crown and overwintering of winter oilseed rape

Characteristics	Treatment	Year			Mean
		2006	2007	2008	
Diameter of the root crown, mm	Humistar	9.37	7.73	7.35	8.15
	Humistar + Drakar	9.33	8.44	7.81	8.53
	Drakar	9.27	8.33	7.91	8.50
	Control	8.47	7.46	7.43	7.79
	Mean	9.06	7.75	7.63	5.31
	$LSD_{0.05}$	0.555	0.867	ns	0.579
		2007	2008	2009	Mean
Overwintering, %	Humistar	84.5	95.4	94.0	91.3
	Humistar + Drakar	90.0	96.7	95.1	93.9
	Drakar	89.6	97.1	93.1	93.3
	Control	84.0	95.4	92.3	90.6
	Mean	87.1	94.3	92.8	93.5
	$LSD_{0.05}$	4.643	ns	ns	3.245

ns – non-significant differences

In evaluation of the oilseed rape rosette condition prior to winter it was found that the root crown diameter was the highest in 2006, probably due to the warmest and longest period of autumn growth and assimilate storage (Table 3). The effect of the humic preparation on this biometric trait of oilseed rape was relatively small. Only in 2006 the application of Humistar resulted in an increase in the root crown diameter. Moreover, in the treatment where Humistar and Drakar were applied, the value of this trait was higher than in the control, but there was no difference as compared with the application of Humistar or Drakar only. In our study, in turn, a strong effect of the foliar application of the potassium fertilizer Drakar was observed. In the years 2006 and 2007, as well as on average in the long-term period, in treatments with Drakar the root crown had a higher diameter than in the control. This could result from a better plant supply in nutrients, which could stimulate accumulation of assimilates in root crowns. It is known from the literature data that both potassium and humic substances had a favourable effect on growth, physiological processes and nutrient uptake [Billard *et al*. 2014, Cheema et al. 2012, Govahi and Saffari 2006, Szczepaniak 2015, Verlinden *et al*. 2009, Wang *et al*. 2015, Yasari and Patwardhan 2006]. Overwintering of winter oilseed rape depends on the diameter of the root crown and the amount of assimilates stored in it [Velicka *et al.* 2006]. In the present study, the response of the root crown diameter to the application of the studied preparations was partly similar to the response of overwintering. In 2007, where the highest losses during the winter period were noted, there was found a favourable effect of the fertilizer Drakar, or its combined application with the preparation Humistar, on overwintering (Table 3). Also in the long-term period, oilseed rape treated with the foliar fertilizer Drakar overwintered slightly better, but a significant difference in relation to the control occurred only in the case where Humistar was also used. Favourable effect of potassium can be explained by its higher availability and uptake after foliar application of this element. The study by Velicka *et al*. [2005] indicates an interrelation between rapeseed overwintering and potassium content in the terminal bud.

The non-destructive method of measurement of the chlorophyll index (SPAD value) can be used to evaluate the plant nutritional status [Koeslin-Findeklee *et al*. 2015]. The present study proved relatively small but favourable effects of combined application of Humistar and Drakar on the value of this index (Table 4). The chlorophyll index measured in autumn in 2008, and also on average in the long-term period, was higher in the treatment with Humistar + Drakar than in the control. The response of chlorophyll index in spring in 2007 was similar. The results obtained in that year were similar to the response of the seed yield (Table 5). Also the study by Fanaei *et al*. [2009] indicates an increase in the SPAD value under the influence of potassium fertilization, as well as the relationship between the seed yield and SPAD values). Also other researchers indicated that oilseed rape plants treated with humic acids had more chlorophyll and higher total chlorophyll contents [Lofti *et al.* 2015].

Table 4. Chlorophyll index of winter oilseed rape (SPAD)

Date of measurement	Treatment	Year			Mean
		2006	2007	2008	
November	Humistar	43.4	39.2	42.5	41.7
	Humistar + Drakar	43.4	39.3	44.6	42.4
	Drakar	42.0	40.0	43.9	42.0
	Control	41.6	38.5	42.1	40.7
	Mean	42.6	39.3	43.3	41.7
	$LSD_{0.05}$	ns	ns	1.92	1.33
April		2007	2008	2009	Mean
	Humistar	55.2	52.5	43.9	50.5
	Humistar + Drakar	57.8	52.4	44.5	51.5
	Drakar	57.1	52.9	43.0	51.0
	Control	55.0	52.8	43.2	50.3
	Mean	56.3	52.6	43.1	32.7
	$LSD_{0.05}$	2.38	ns	ns	ns

ns – non-significant differences

In the present study, a very high average seed yield was obtained, amounting to 5.89 t·ha^{-1} (Table 5). This was two times higher than the average national yield in the analysed period [FAOSTAT 2016]. The highest seed yield was obtained in 2009, where the plants overwintered well (Table 2) and formed plump seeds (the highest thousand seed weight) (Table 5). The lowest yield was obtained in 2008, which was caused by limited rainfalls in May and June, i.e. at the stage of flowering and forming siliques. According to Gaj [2011], high yields of oilseed rape are determined by a favourable arrangement of meteorological conditions at critical stages of taking up nutrients. In the present study, a tendency to higher yield of winter oilseed rape was observed after the application of tested preparations Humistar and/or Drakar in all the years. The seed yield on average in the long-term period was significantly higher than in the control after foliar application of the potassium fertilizer Drakar, as well as in the treatment where both preparations were used. The effect of combined application of both preparations was significant also in 2007, where an increase in the seed yield was obtained by 490 kg·ha^{-1} (9%) as compared with the control. Many previous studies indicate positive effects of application of biostimulants containing humic substances in cultivated crops [Nardi *et al*. 2002, Verlinden *et al.* 2009, Trevisan *et al*. 2010, Muscolo *et al*. 2013, Billard *et al*. 2014, Canellas et al. 2015, Lotfi *et al.* 2015]. The few studies of oilseed rape show a favourable effect of the application of such substances (as leonardite) on dry matter yields and nutrients concentration and uptake [Akinremi *et al.* 2000]. The literature data also indicate that the application of potassium in oilseed rape might but does not have to cause an increase in seed yield [Govahi and Saffari 2006, Yasari and Patwardhan 2006, Quezada *et al*. 2010, Amanullah and Malhi 2011b, Cheema *et al.* 2012]. In our study the application of the potassium fertilizer Drakar brought a significant increase in the seed yield. Gaj [2011] claims that even at the average K content in soil oilseed rape may be insufficiently nourished with this element. A high potassium uptake probably caused exhaustion of resources of this element from the rhizosphere [Shi *et al*. 2004]. It was indicated that the species of the family Brassicaeae giving the highest yields, including oilseed rape, show the highest depletion rate of available K in the rhizosphere. There is also known a significant relationship between plant supply in potassium and the uptake of other nutrients by oilseed rape [Gaj 2011, Szczepaniak 2015, Wang *et al*. 2015].

No significant response of thousand seed weight to the used preparations was found in any growing season (Table 5). Only a tendency to increase TSW after foliar application of the potassium fertilizer Drakar was observed, mostly in the season 2006--2007. In other studies the response of TSW in oilseed rape to fertilization was very distinct (an increase by 0.2 to 0.7 g), but in that experiment both rates of K and N were increased [Borovko 2008]. In the present study, thousand seed weight and oil content in the seeds were the lowest in the season 2006-2007. The effect of applied preparations on oil content in the seeds was small. Only in the season 2007-2008, the use of the fertilizer Drakar alone resulted in an increase in oil content in the seeds of the studied winter oilseed rape cultivar. A favourable effect of potassium fertilization on the oil content in oilseed rape seeds was also indicated by Ammanullah and Malhi [2011a]. Yasari and Patwardhon [2006] in turn did not prove such a response. In the study by Borovko [2008], increasing N and K rates decreased oil content but increased the protein content within seeds.

Table 5. Seed yield, thousand seed weight (TSW) and oil content in seed of winter oilseed rape

Characteristics	Treatment	Year			Mean
		2007	2008	2009	
Seed yield, $t \cdot ha^{-1}$	Humistar	5.60	5.33	6.76	5.90
	Humistar + Drakar	5.92	5.21	6.84	5.99
	Drakar	5.77	5.21	6.90	5.96
	Control	5.43	5.04	6.72	5.73
	Mean	5.68	5.20	6.80	5.89
	$LSD_{0.05}$	0.372	ns	ns	0.230
TSW, g	Humistar	4.45	4.83	5.09	4.79
	Humistar + Drakar	4.49	4.74	5.13	4.79
	Drakar	4.57	4.90	5.14	4.87
	Control	4.32	4.92	5.02	4.75
	Mean	4.46	4.85	5.09	4.80
	$LSD_{0.05}$	ns	ns	ns	ns
Oil content, %	Humistar	43.7	45.4	45.5	44.8
	Humistar + Drakar	43.7	44.7	45.2	44.5
	Drakar	43.5	45.5	45.3	44.8
	Control	43.6	44.9	45.4	44.6
	Mean	43.6	45.1	45.3	44.7
	$LSD_{0.05}$	ns	0.576	ns	ns

ns – non-significant differences

CONCLUSIONS

In soil and weather conditions that enable obtaining seed yields of winter oilseed rape of 5.2-6.8 $t \cdot ha^{-1}$, favourable effects, such as an increase in the seed yield, and in some years also in the oil content in seeds, can be achieved by two-time (2 $l \cdot ha^{-1}$in autumn + 4 $l \cdot ha^{-1}$ in spring) foliar application of the potassium fertilizer Drakar. The use of this fertilizer with an additional pre-sowing application of the humic preparation Humistar (in a dose of 40 $l \cdot ha^{-1}$) increases the chlorophyll index in the autumn and improves overwintering in relations to the control, but it does not bring an increase in yield in comparison with the application of the same fertilizer. Further studies are necessary to evaluate the effect of these preparations in soils with low content of nutrients and humus substances.

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REAKCJA RZEPAKU OZIMEGO (*Brassica napus* L.) NA DOGLEBOWĄ APLIKACJĘ PREPARATU HUMUSOWEGO I DOLISTNE NAWOŻENIE POTASEM

Streszczenie. Celem badań była ocena wpływu preparatu humusowego Humistar i/lub dolistnego nawozu potasowego Drakar na cechy biometryczne, przezimowanie oraz stan odżywienia oraz wielkość i jakość plonu rzepaku ozimego. Podstawą badań było jednoczynnikowe doświadczenie polowe prowadzone w latach 2006-2009 w Chrząstowie, na glebie brunatnej właściwej, o uziarnieniu gliny lekkiej, w układzie losowanych bloków, w czterech powtórzeniach. Oceniano następujące kombinacje: Humistar (12% kwasy huminowe i 3% kwasy fulwowe) aplikowany przedsiewnie, doglebowo, w dawce 40 $dm^3 \cdot ha^{-1}$; Drakar (25.7% K i 3% N) stosowany w fazie 6 liści (2 $dm^3 \cdot ha^{-1}$) oraz na początku opadania płatków (4 $l \cdot ha^{-1}$); aplikację obu preparatów (Humistar + Drakar) oraz obiekt kontrolny. Wykazano, że aplikacja preparatu Drakar miała korzystny wpływ na plon nasion rzepaku. Zastosowanie tego nawozu zwiększyło indeks chlorofilu w jesieni oraz poprawiło przezimowanie w stosunku do obiektu kontrolnego tylko w przypadku wcześniejszej aplikacji preparatu Humistar. Potrzebne są dalsze badania oceny działania tych preparatów na glebach słabych, o niskiej zawartości materii organicznej i składników pokarmowych.

Słowa kluczowe: indeks zieloności liścia, plon nasion, przezimowanie, zawartość tłuszczu

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