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[translator's notes are italicised and in brackets]

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Institute of Soil Science and Plant Cultivation

State Research Institute

DEPARTMENT OF PLANT NUTRITION AND FERTILISATION

Report on tests called

Assessment of agricultural suitability of Total Humus biostimulator

conducted in 2015 under contract

no. 414 - 2/15

with

Eko World Matylda Mazur -Zaskórska company

Puławy, November 2015

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NIP [*Tax Identification Number*]: 716-000-42-81

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TEST PURPOSE AND METHOD

The purpose of the test was to determine the suitability of Total Humus plant growth stimulator in field cultivation of agricultural plants.

The tests were performed in 2015, during field experiments in the Experimental Department of UING-PIB in Grabów nad Wisłą.

The samples of Total Humus product for test purposes were taken by the sample-taker from the Regional Chemical and Agricultural Station in Gorzów Wielkopolski (Sample no. 2 Taking Report dated 13.01.2015).

In the experiments the effect of the stimulator compared to the control plant. The experiments were conducted in 4 repetitions, using a long stripes method. In all plants the same agrotechnology and fertilisation was applied, except for the spray application of biostimulator.

The product was tested on winter rape and maize.

Experiment on winter rape

The experiment involved the following combinations:

A -- Control plant - not sprayed

B -- Spray application of Total Humus 200 ml/ha

C -- Spray application of Total Humus 400 ml/ha

D -- Spray application of Total Humus 800 ml/ha

In plants B, C and D the product was applied in line with the manufacturer's recommendations, at three dates.

Field size at setup: 60 m².

Field size at harvest: 30 m².

Soil characteristics:

Type: Lessive soil based on light clay - class III a Agricultural suitability parameter: good wheat soil complex.

Drained field?: Yes

Moisture: correct

Soil culture: medium

Soil tilth: good

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Agrotechnology:

Sowing of rape, variety Pamela 4,1 kg /ha - 29.08.2014. Seeds dressed in CN.

Applied fertilisation and chemical plant protection products

Plant designation	Type of fertiliser of chemical	% of pure ingredien	Organic fertilisation in t/ha	Type and dosage in kg/ha		Date of applying
				pure ingredient	active substance	
Whole	Polifoska 6	30	K20	84.0	-	29. 08. 14
experiment	Polifoska 6	20	P205	56.0	-	29. 08. 14
	Polifoska 6	6	N	17.0	-	29. 08. 14
	Ammonium nitrate	34	N	102.0	-	18. 03. 15
	Ammonium nitrate	34	N	51.0	-	14. 04. 15
	Butisan Star	-	-	-	3.0l/ha	25. 09. 14
	Agil	-	-	-	0.5l/ha	25. 09. 14
	Horizon	-	-	-	0.7l/ha	28. 10. 14
	Durban EC				0.6L/HA	23. 04. 15
	Konker 416				1.5l/ha	05. 05. 15
200ml	TOTAL HUMUS	-	-	-	200ml/ha	13. 04. 15.
400ml	TOTAL HUMUS	-	-	-	400ml/ha	13. 04. 15.
800ml	TOTAL HUMUS	-	-	-	800ml/ha	13. 04. 15.
200ml	TOTAL HUMUS	-	-	-	200ml/ha	08. 05. 15.
400ml	TOTAL HUMUS	-	-	-	400ml/ha	08. 05. 15.
800ml	TOTAL HUMUS	-	-	-	800ml/ha	08. 05. 15.
200ml	TOTAL HUMUS	-	-	-	200ml/ha	18. 05. 15.
400ml	TOTAL HUMUS	-	-	-	400ml/ha	18. 05. 15.
800ml	TOTAL HUMUS	-	-	-	800ml/ha	18. 05. 15.

Pre-sowing cultivation and treatments during vegetation.

28/08/2014	Field worked with KBT disk harrow
28.08.14	Pre-sow ploughing with a reversible plough
29/08/2014	Broadcasting of potassium and phosphorous fertilisers using an MX disk type spreader
29. 08. 14	Seedbed preparation using a passive cultivation unit. Sowing winter rape of Pamela variety at 4.0kg/ha
25. 09. 14	Spray application of mixture of herbicides, Butisan Star, on rape, at 3.0l/ha +

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	Agil 0.5l/ha in 300 litres of water
28. 10. 14	Spray application of fungicide, Horizon, on rape at 0.7l/ha in 250 litres of water
18. 03. 15	Broadcast of nitrogen in the form of ammonium nitrate - 300kg/ha
14. 04. 15	Broadcast of dose II of nitrogen in the form of ammonium nitrate - 150kg/ha
23, 04. 15	Spray application of Durban EC on rape with insecticide against <i>Meligethes aeneus</i> at 0.6l/ha in 300 litres of water
05. 05. 15	Spray application of fungicide, Konker 416, on rape at 1.5l/ha in 250 litres of water
13. 04. 15	Spray application of Total Humus on rape, in line with instruction for experiment
08. 05. 15	Spray application of Total Humus on rape, in line with instruction for experiment
18. 05. 15	Spray application of Total Humus on rape, in line with instruction for experiment
27. 07. 15	Harvesting of rape

Phenological phases of plants:

code	Phase	date
00	Dry seed	29.08.2014
03	Start of seed swelling	30.08.2014
05	Rootlet formation	01.09.2014
07	Germ length - 1/2 of seed length	04.09.2014
11	Cotyledones above soil surface	06.09.2014
13	Development of cotyledones	10.09.2014
15	First leaf stage	12.09.2014
17	Second leaf stage	16.09.2014
19	Third leaf stage	28.09.2014
21	Fourth leaf stage	10.10.2014
22	Fifth leaf stage	17.10.2014
23	Sixth leaf stage	27.10.2014
24	Seventh leaf stage	31.10.2014
25	Eighth leaf stage	15.03.2015
26	Ninth-eleventh leaf stage	20.03.2015
31	Distance from base - 5 cm	02.04.2015
33	of leaves from top - 10 cm	09.04.2015
35	shoot growth over: -15 cm	13.04.2015
37	-20 cm	16.04.2015
39	-25 cm	20.04.2015
51	Bud formation start (buds wrapped in leaves)	23.04.2015
53	Inflorescence diameter up to 1 cm	25.04.2015
55	Largest bud diameter 2mm	27.04.2015
57	Inflorescence elongation	29.04.2015
61	Blooming of first flower	29.04.2015

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62	Single flowers blooming on the main shoot	30.04.2015
63	Many flowers blooming on the main shoot, older flower start losing petals	04.05.2015
64	Full blossom, number of buds and primordia of siliques is similar	18.05.2015
65	End of full blossom, less than 5% undeveloped	20.05.2015
69	End of blossom	22.05.2015
71	First siliques with seeds on main shoot are of normal size	20.05.2015
75	Seeds in siliques in the lower part of the main shoot are developed	25.05.2015
79	Almost all siliques on the main shoot have developed seeds	27.05.2015
81	Larger siliques on main and lateral shoots have normal size seeds	01.06.2015
83	All siliques, except for top ones, are developed	10.06.2015
85	Seed coat of earlier set seeds is half-black	22.06.2015
87	Most seeds are half-black, the seed may be cut	05.07.2015
89	Seeds are hard and dark, siliques are partly dry	15.07.2015
91	Full maturity	22.07.2015

Weather during vegetation

Month	IX	X	XI	XII	I	II	III	IV	V	VI	VII	VIII	IX
Precipitation in mm	15.9	28.5	25.7	36.3	40.3	15.1	63.2	34.8	107.0	30.3	51.7	6.2	93.9
Average temperature in °C	17.9	9.8	4.7	0.5	1.0	0.5	5.0	8.1	12.7	16.9	19.7	22.1	15.0

Results of experiment

During rape vegetation the plant development indicators were observed and measured. At two dates (12.05.2015 and 18.05.2015) 5 plants were randomly selected from the objects of experiment. Shoots and roots were measured. Dry weight of plants was determined. Plant material samples were subject to chemical analysis and the content of primary nutrients (NPKMg) was determined. Chlorophyll content, which is an indirect measure of nitrogen-related nutritional state of the plant, was measured in vivo (SPAD).

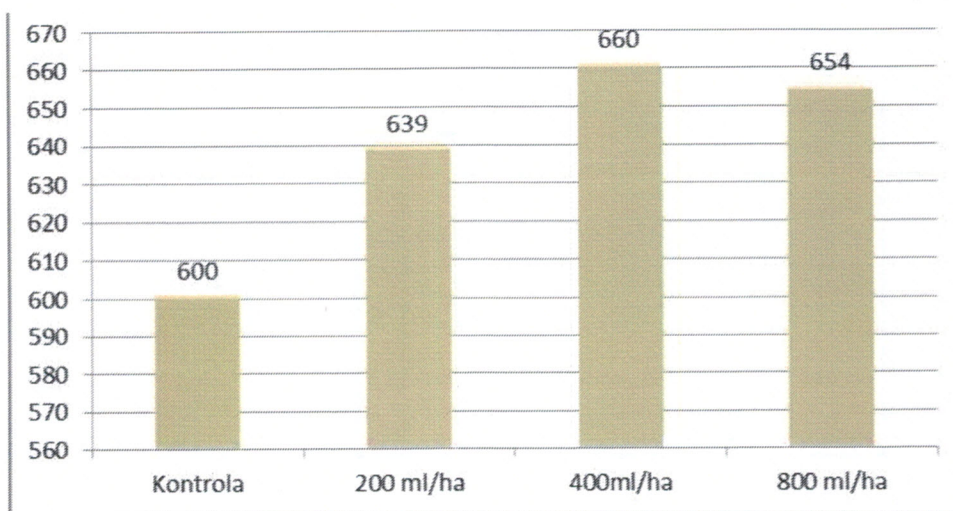


Fig. 1 Indirect indicator of chlorophyll content in rape leaves (average of 3 measurements)

Application of Total Humus resulted in changed in the SPAD indicator (fig. 1). The value of this indicator was higher in all plants sprayed with the product, when compared to the control batch. The highest value, which meant the best nitrogen nutrition of plants, was observed in the object that received 400 ml/ha. The application of a larger dose of the product resulted in lower indicator against the highest value.

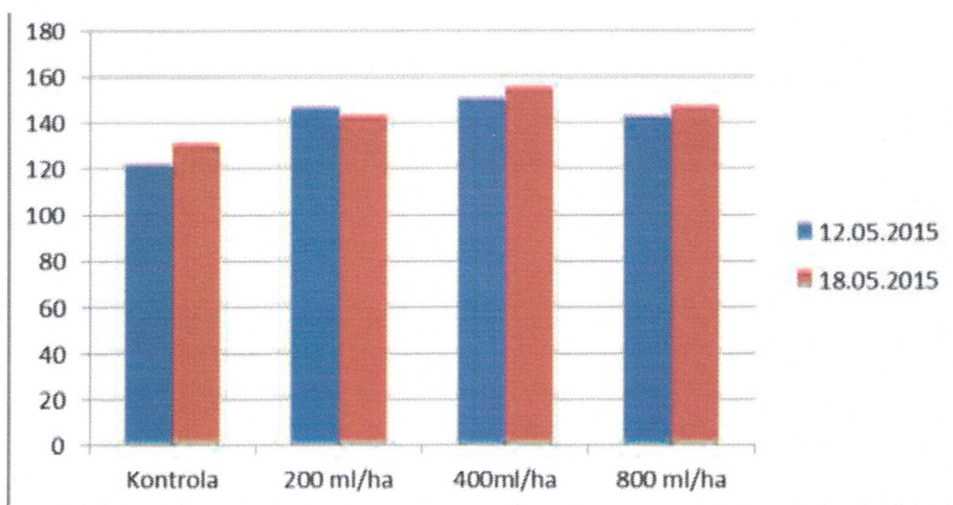


Fig. 2 Rape plant height at two next dates

The spray resulted in increase in rape plant height (fig. 2).

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In all batches treated with biostimulator, the plants were higher than in the control batch. In the batch treated with the highest dosage, the plants were, however, lower than in the case of batch sprayed with 400 ml/ha.

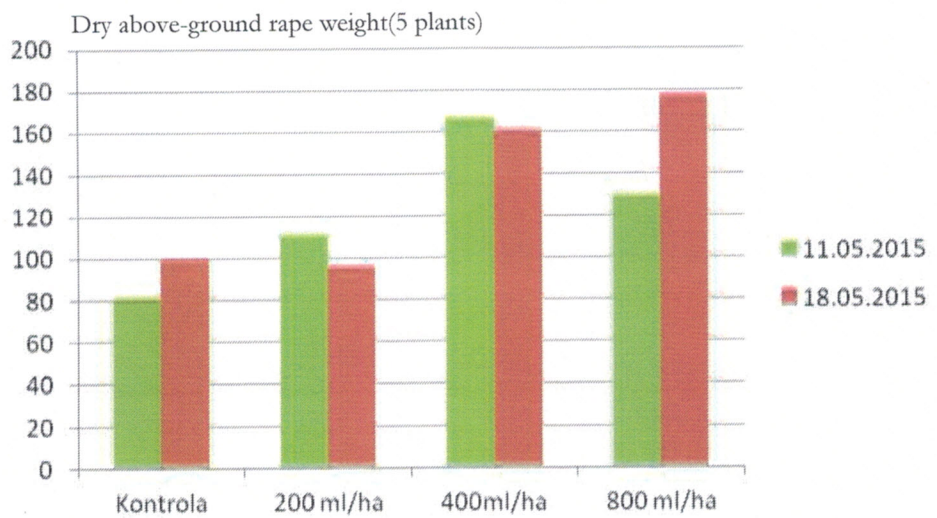
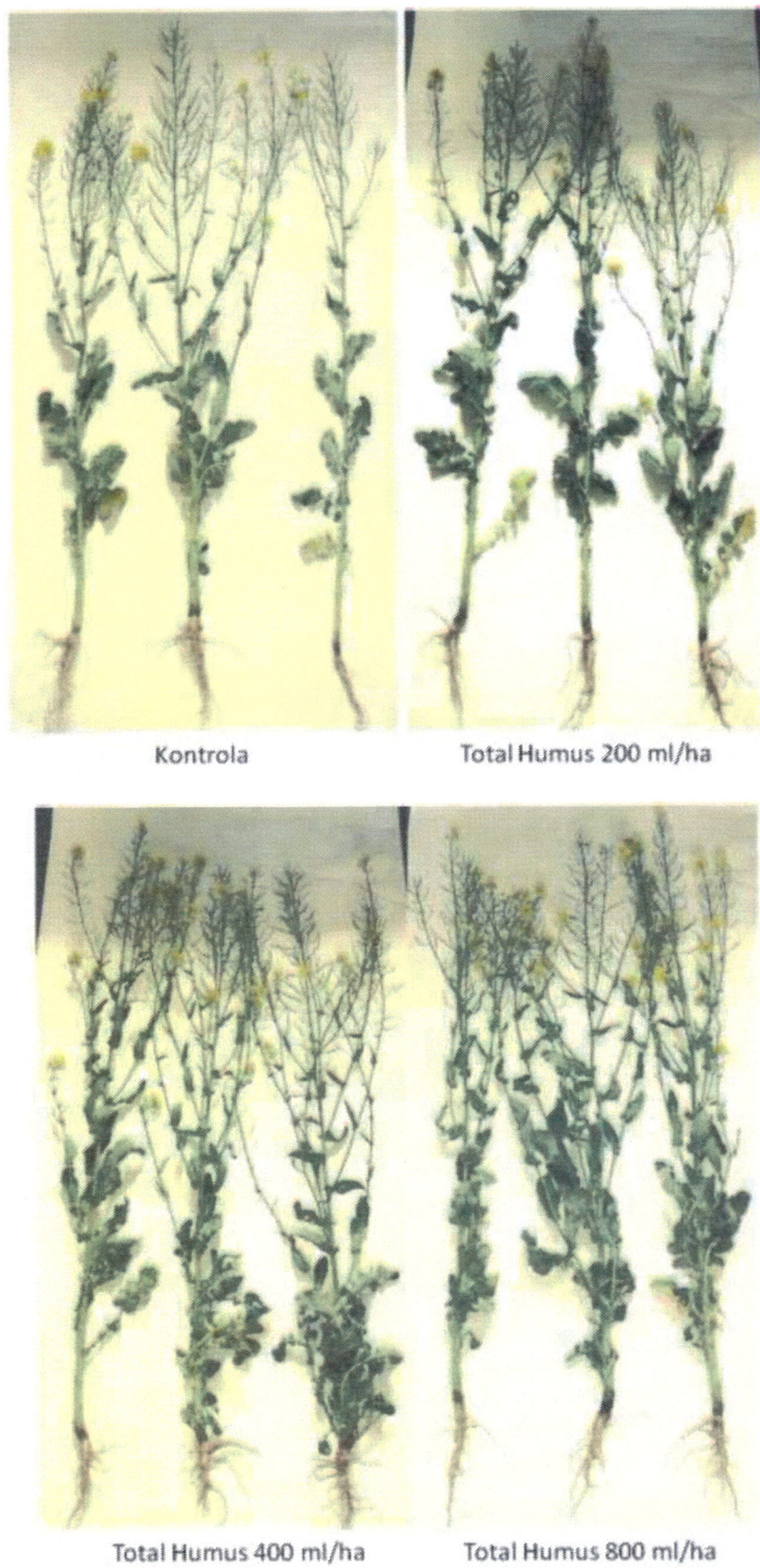


Fig. 3 Biostimulator's impact on rape biomass

Distinctive increase in maize dry weight against the control batch at both measurement dates were observed in batch treated with 400 ml/ha (fig. 3) (pict. 1). In the batch treated with a higher dosage of the product, the increase in weight occurred later, but it was not significant compared to the batch treated with 400 ml.



Pict. 1. Impact of Total Humus biostimulator on the size of above-ground rape weight

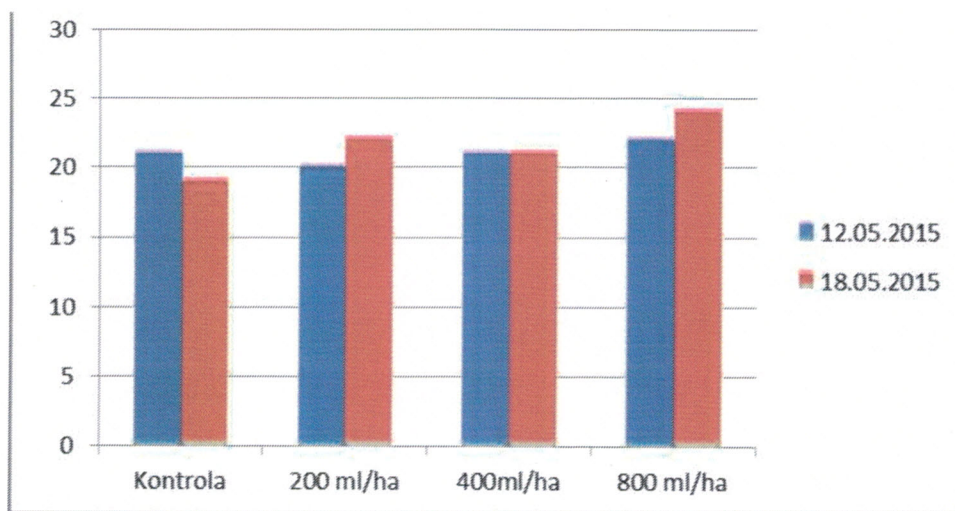


Fig. 4 Rape plant root length at two next dates

The impact of biostimulator on the length of rape roots was not clear cut (fig. 4), however, an elongation of the root system could be observed in the batch treated with the highest dosage of product. On the other hand, the root system weight increased visibly under the influence of the product (fig. 5) (pict. 1).

Dry rape root weight(5 plants) 12.05.2015

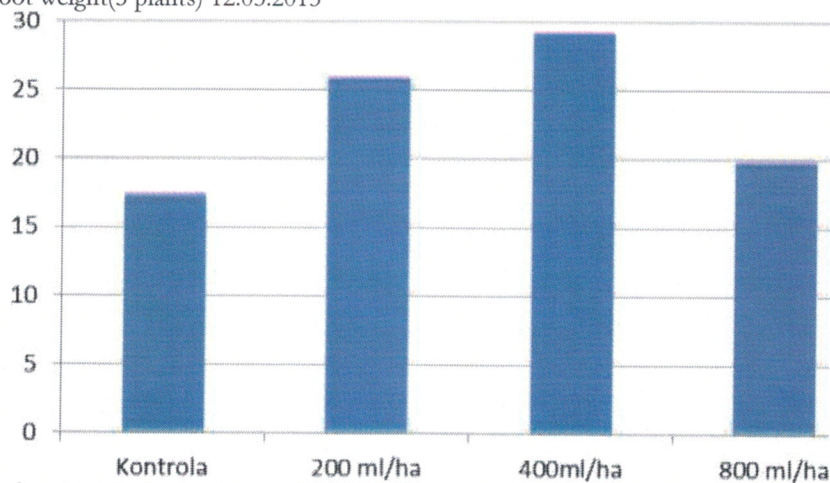
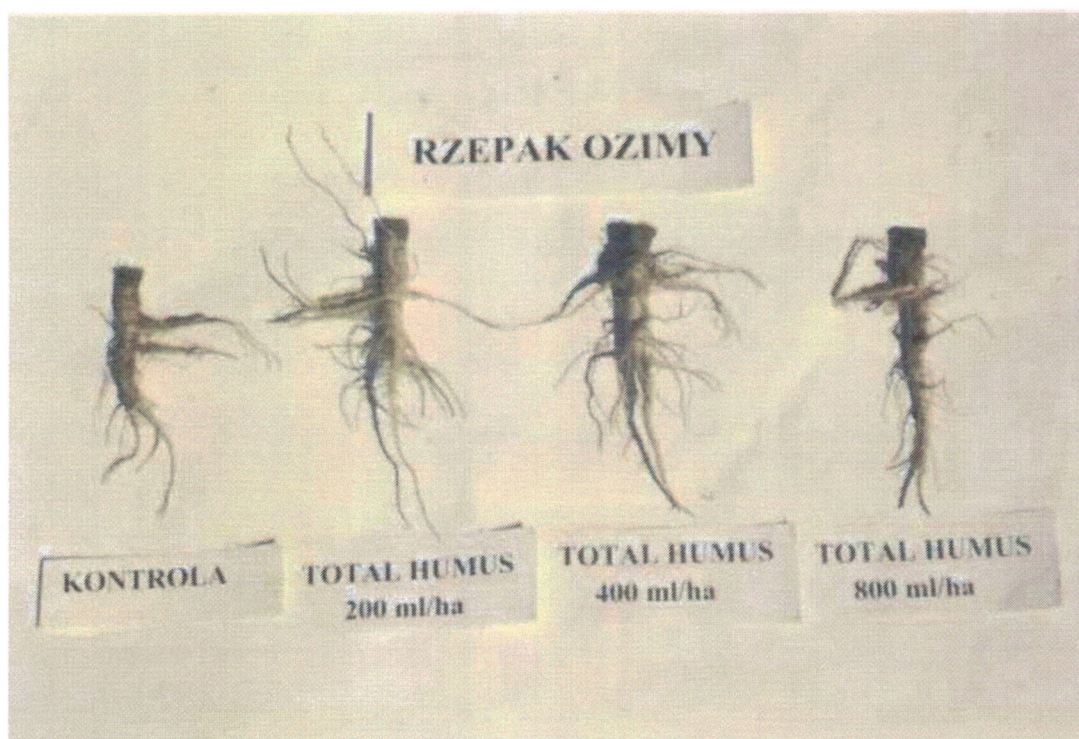


Fig. 5 Dry weight of roots of 5 plants taken on 12.05.2015





Pict. 1. Impact of Total Humus on the development of rape root system

The application of biostimulator at 200 and 400 ml/ha resulted in increased contents of nutrients, in particular nitrogen, in the above-ground parts of plants (table 1). The result of this, and of increase in rape weight under the influence of biostimulator at 200 and 400 ml/ha, the absorption of nutrients increased (table 1): nitrogen (N) by 90%, phosphorus (P) by 69%, potassium (K) by 77% and magnesium (Mg) by 73%. The application of 800 ml/ha slightly boosted the absorption of PKMg, but the uptake of nitrogen dropped.

Table 1. Nutrient concentration and uptake by rape (30.06.2015)

Batch	Nutrient content in plant dry weight (%)				Nutrient uptake (g/m ²)			
	N	P	K	Mg	N	P	K	Mg
Control	1.7	0.44	3.2	0.14	1.67	0.433	3.15	0.138
200 ml/ha	1.9	0.46	3.3	0.16	1.79	0.434	3.11	0.151
400ml/ha	2.0	0.46	3.5	0.15	3.18	0.732	5.57	0.239
800 ml/ha	1.7	0.42	3.3	0.14	2.99	0.740	5.81	0.247

The better development of root system and improved nutrition of plants caused by the applied product resulted in higher yield of rape seeds. The yield obtained in the entire experiment were high, from 4.3 t/ha in the control batch to 5.4 t/ha in the batch treated with 400 ml/ha. The yield growth was statistically significant. The yield obtained from the batch treated with the highest dosage of product was slightly lower than the optimum variant, but higher than from the control batch.

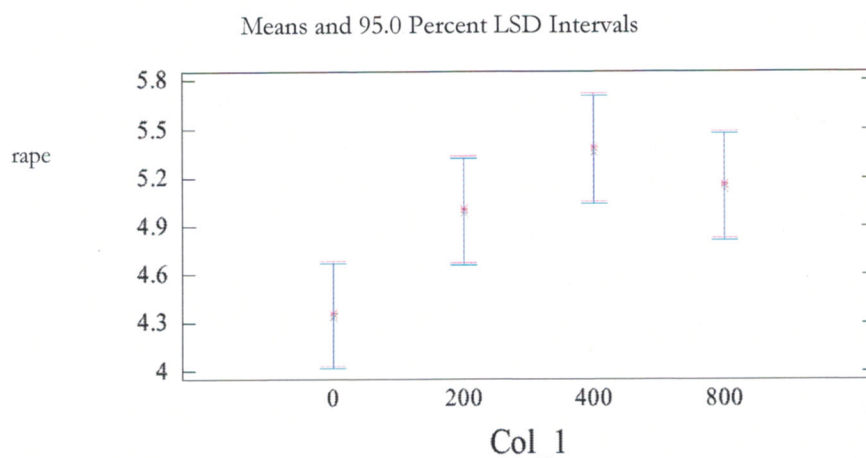


Fig. Average rape seed yield per batch and confidence half-intervals

The test results confirmed the effectiveness of the Total Biohumus product in the cultivation of rape.

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Experiment with maize

The experiment involved the following combinations:

A -- Control plant - not sprayed

B -- Spray application of Total Humus 200 ml/ha

C -- Spray application of Total Humus 400 ml/ha

D -- Spray application of Total Humus 800 ml/ha

In plants B, C and D the product was applied in line with the manufacturer's recommendations, at two dates.

2

Field size at setup: 60 m².

2

Field size at harvest: 28 m².

Soil characteristics:

Type: Lessive soil based on light clay - class III a

Agricultural suitability parameter: good wheat soil complex

Drained field?: Yes

Moisture: correct

Soil culture: medium

Soil tilth: good

Agrotechnology:

Sowing of maize LG 30-260 29.04.2015

Applied fertilisation and chemical plant protection products

Plant designation	Type of fertiliser or chemical	% pure ingredien	Organic fertilisation in t/ha	Type and dosage in kg/ha		Date of applying
				pure ingredient	active substance	
Whole	Polifoska 6	30	K20	84.0	-	28. 04. 15
experiment	Polifoska 6	20	P205	56.0	-	28. 04. 15
	Polifoska 6	6	N	17.0	-	28. 04. 15
	Urea	46	N	92.0	-	30. 04. 15
	Polidap	46	P205	23.0	-	30. 04. 15
	Polidap	18	N	9.0	-	30. 04. 15
	Ammonium nitrate	34	N	40.0	-	26. 06. 15
Whole	Majster 310 WG	-	-	-	150g/ha	19. 05. 15
Whole	TOTAL HUMUS	-	-	-	400ml/ha	29. 04.15
200ml	TOTAL HUMUS	-	-	-	200ml/ha	03. 06. 15
400ml	TOTAL HUMUS	-	-	-	400ml/ha	03. 06. 15
800ml	TOTAL HUMUS	-	-	-	800ml/ha	03. 06. 15

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200ml	TOTAL HUMUS	-	-	-	200ml/ha	16. 06. 15
400ml	TOTAL HUMUS	-	-	-	400ml/ha	16. 06. 15
800ml	TOTAL HUMUS				800ml/ha	16. 06. 15

Cultivation from the last harvest and treatments during vegetation.

22.11.14	Winter ploughing with a reversible plough at the depth of 23-25 cm.
28.04.15	Broadcasting of potassium and phosphorous fertilisers using an MX disk type spreader
28. 04.15	Field worked with an active cultivation unit
29. 04. 15	Seedbed preparation using a passive cultivation unit. Sowing of maize variety LG 30-260 at spacing of 70/13.5 cm, using Planter 2 single-seed drill
19. 05. 15	Spray application of Majster 310 WG herbicide on maize at 150g/ha + 250 litres of water
29. 04. 15	Spray application of Total Humus on maize at 400ml/ha without control batch
03. 06. 15	Spray application of Total Humus on maize , in line with instruction for experiment
26. 06. 15	Sowing of nitrogen fertilisers (ammonium nitrate) at 40kg/ha
16. 06. 15	Spray application of Total Humus on maize , in line with instruction for experiment
07. 09. 15	Harvesting silage maize

Phenological phases of plants:

code	Phase	Maize
01	Dry grain	2015.04.29
03	Start of seed swelling	2015.05.02.
05	Rootlet formation	2015.05.08.
07	Coleoptile emerge from grain	2015.05.10.
09	Coleoptile of 2.5 cm in length	2015.05.12.
11	Coleoptile above soil surface	2015.05.16
13	Start of first leaf formation	2015.05.18.
15	First leaf developed	2015.05.20
17	Visible top of the second leaf	2015.05.22.
19	Second leaf developed	2015.05.26
21	Third leaf developed	2015.05.29.
22	Fourth leaf developed	2015.06.14.
23	Fifth leaf developed	2015.06.26.
24	Sixth leaf developed	2015.07.10.
25	Seventh leaf developed	2015.07.15.

26	8-11 leaves developed	2015.08.01.
27	12+ leaves developed	2015.08.11.
31	First node palpable	2015.07.25.
32	First node visible	2015.07.28.
33	Second node palpable	2015.08.02.
34	Second node visible	2015.08.04.
35	Third node visible	2015.08.08.
36	Fourth node visible	2015.08.12.
51	Start of tasseling	2015.08.14.
53	Visible tassel top	2015.08.16.
55	Full tasseling	2015.08.18.
59	End of tasseling	2015.08.25.
61	Start of pollen shedding	2015.08.18.
65	Half of tassels shed pollen	2015.08.21.
67	End of pollen shedding	2015.08.26.
71	Visible cob	2015.08.14.
73	Pistils comes of the husks	2015.08.16.
75	Pistils fully out	2015.08.18.
77	Start of pistils wilting	2015.08.20.
79	Pistils fully wilted	2015.08.24
81	Grain setting	2015.08.26.
82	Milk-ripe stage	2015.08.30.
83	Dough stage	2015.09.06
84	Yellow maturity	2015.09.07.
85	Physiological maturity (35 - 40 % in 1g)	

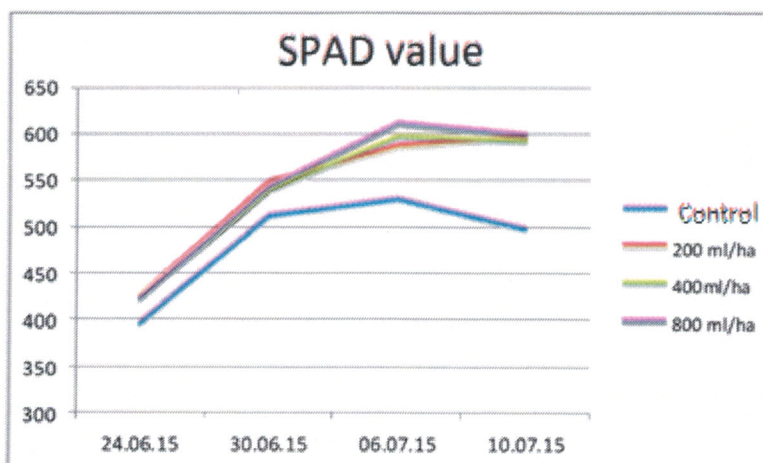
Weather conditions

Month	IX	X	XI	XII	I	II	III	IV	V	VI	VII	VIII	IX
Precipitation in mm	15.9	28.5	25.7	36.3	40.3	15.1	63.2	34.8	107.0	30.3	51.7	6.2	93.9
Average temperature in °C	17.9	9.8	4.7	0.5	1.0	0.5	5.0	8.1	12.7	16.9	19.7	22.1	15.0



Results of experiment

In 2015 the weather conditions for maize growth were extremely unfavourable due to draught. During rape vegetation the plant development indicators were observed and measured. At two dates (12.05.2015 and 18.05.2015) 5 plants were randomly selected from the objects of experiment.



Control batch 200 ml/ha 400ml/ha 800 ml/ha

Fig. 1. Impact of Total Humus biostimulator on SPAD indicator

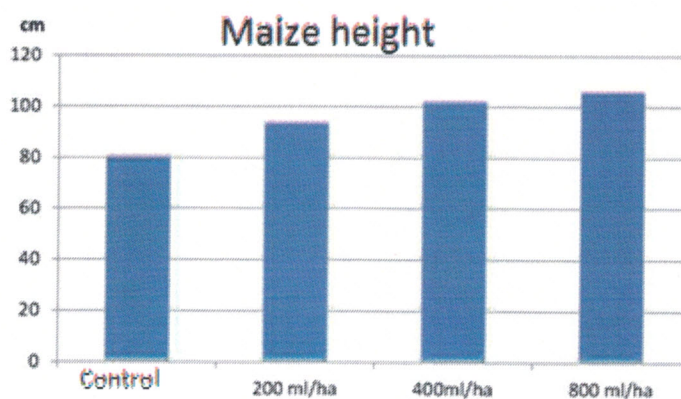


Fig. 2 Impact of Total Humus biostimulator on maize plant height

SPAD measurements at 4 dates indicate better nitrogen nutrition in batches sprayed with Total Humus compared with the control batch (fig. 1). Plant measurements made on 30.06.2015 demonstrated an increase in height (fig. 2) (pict. 1) and dry plant weight (fig. 3) under the influence of increasing product dosage.

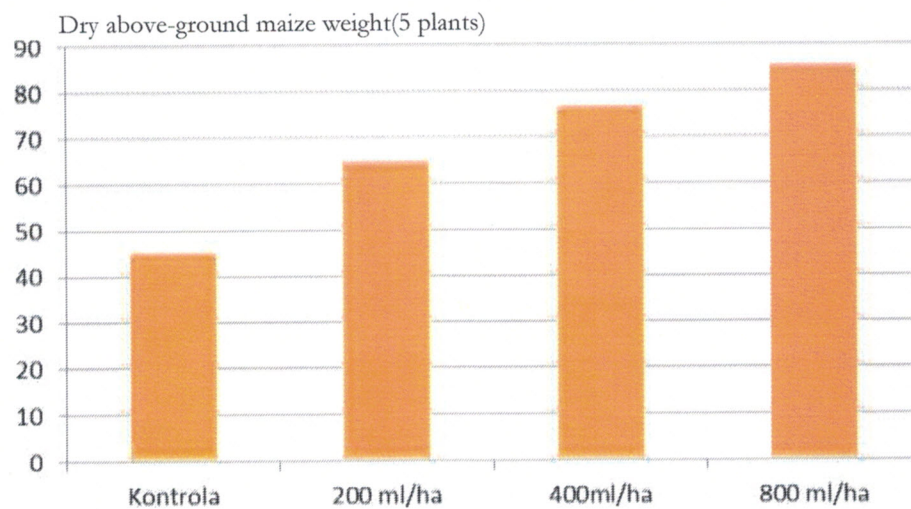
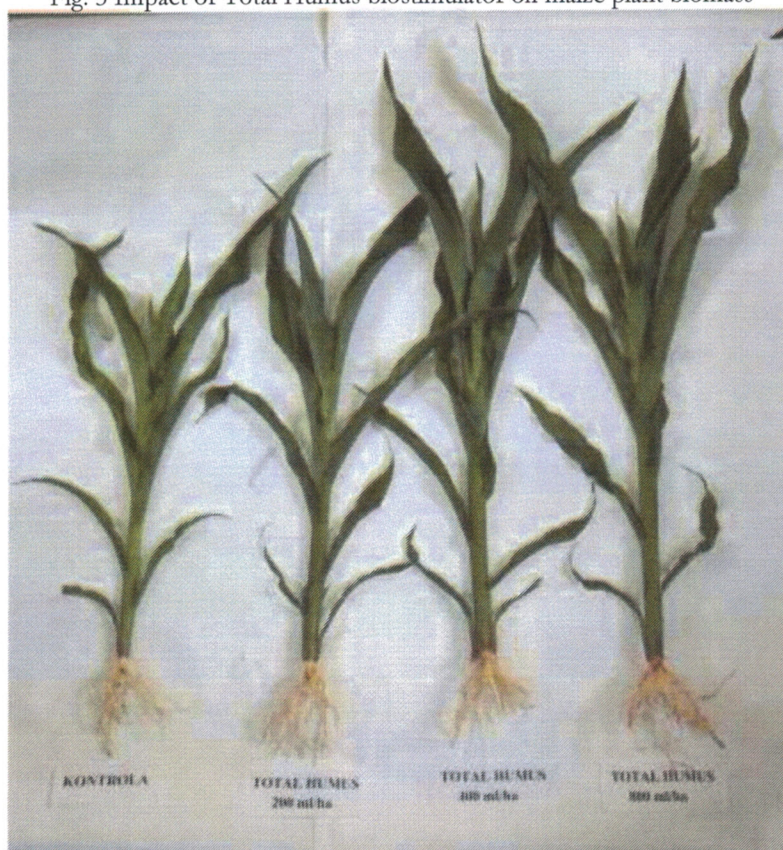


Fig. 3 Impact of Total Humus biostimulator on maize plant biomass



Pict. 1. Impact of biostimulator on maize growth

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The measurements of maize roots length were not explicit (fig. 4) but the measurement of dry root mas showed a clear growth under the increasing dosage of product (fig. 5) (pict. 2).

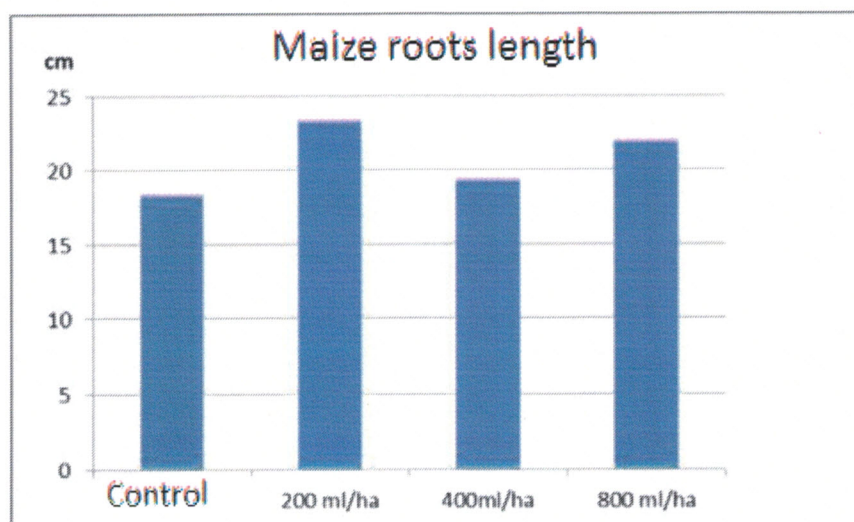


Fig. 4. Impact of Total Humus biostimulator on maize root length

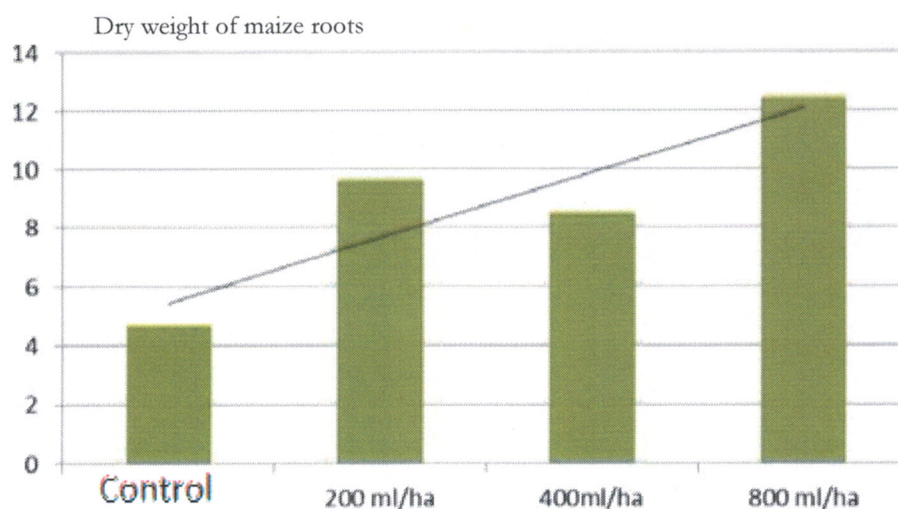
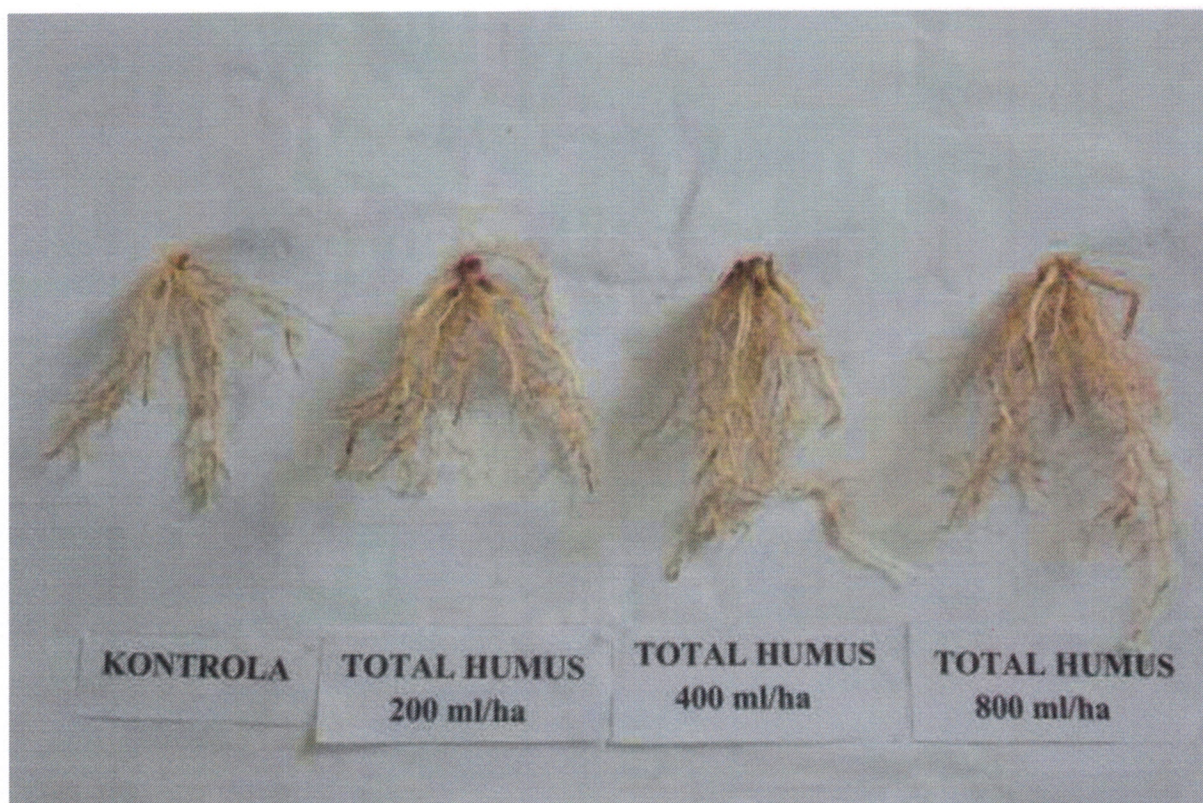


Fig. 5 Impact of Total Humus on the weight of root system





Pict. 2. Impact of Total Humus on the development of root system

The analysis of the chemical composition of the plants showed a slight increase in nitrogen content in plants treated with biostimulator. The concentrations of other nutrients were similar to those of control plants (table 1).

Table 1. Concentrations of nutrients in plants and their uptake by maize (30.06.2015)

Batch	Nutrient content in plant dry weight (%)				Nutrient uptake (g/m ²)			
	N	P	K	Mg	N	P	K	Mg
Control	2.2	0.29	5.9	0.19	1.747	0.230	4.685	0.151
200 ml/ha	2.5	0.27	6.0	0.17	2.868	0.310	6.882	0.195
400ml/ha	2.6	0.28	6.0	0.19	3.539	0.381	8.166	0.259
800 ml/ha	2.4	0.27	5.7	0.18	3.655	0.411	8.681	0.274

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 NRTP/48/13
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Due to higher weight of plants in batches treated with the biostimulator, the uptake of nutrients was much higher than in the control batch. Nitrogen (N) uptake was up by as much as 109%, phosphorus (P) by 66%, potassium (K) by 74% and magnesium (Mg) by 72%.

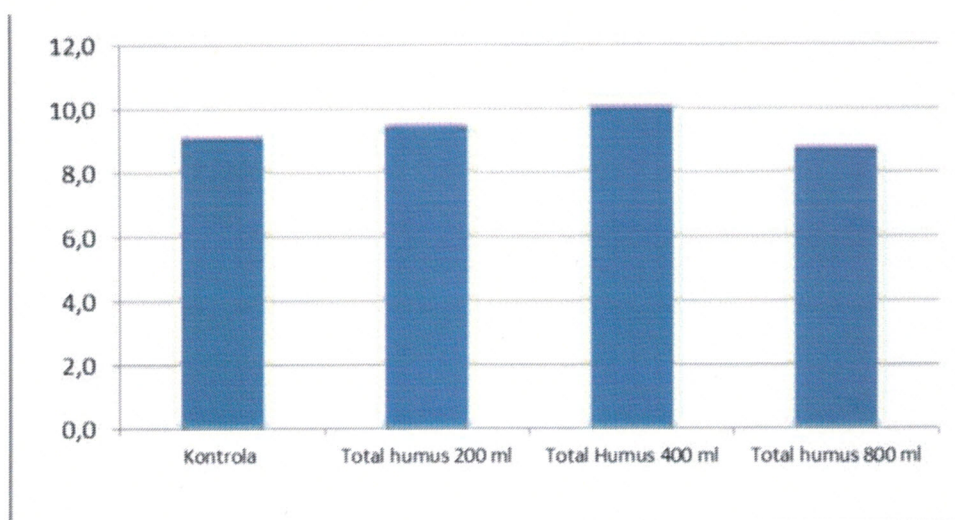


Fig. 6 Impact of Total Humus biostimulator on the yield of dry weight of silage maize

Maize yield in 2015 was low due to water shortage during the vegetation period and was between 9.1 t of dry weight from 1 ha in the control batch to 9.96 t of dry weight in the batch treated with Total Humus at 400 ml/ha. The maize yield was lowest from the batch treated with 800 ml/ha. The yield in the batch treated with 400 ml/ha was by almost 10% higher than in the control batch. The yield was significantly differentiated in individual repetition in the experiment, which results in absence of statistical significance. The variability between repetitions resulted surely from extreme weather conditions in the year of experiment.

CONCLUSIONS

1. The test have confirmed a favourable impact of Total Humus on plant growth, the development of root system, nutrients uptake and yield volume.
2. The best effects were obtained after the application of 400 ml/ha at 3 dates in the cultivation of rape and at 2 date for maize.
3. Due to specific conditions for maize vegetation in 2015, to confirm of exclude the advisability of using the dosage of 800 ml/ha in the cultivation of maize, it would be recommended to repeat the test in the next season.

4. The results confirm that Total Humus has a stimulating effect (due to a scarce amount of nutrients provided while spraying any nutritional effect is excluded).
5. Total Humus is suitable for use in field cultivation of agricultural plants.

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I, Agata Kordylewska, certified translator of the English language entered in the register of certified translators kept by the Minister of Justice under entry number TP/48/13, do hereby certify the above to be a true and faithful translation of the document presented to me in the Polish language.

Translation no. 128/2017
Poznań, 19 December 2017

Agata Kordylewska

