Kinga MATYSIAK, Marta DUBAS, Roman KIERZEK, Sylwia KACZMAREK
Institute of Plant Protection – National Research Institute
ul. Władysława Węgorka 20, 60-310 Poznań, Poland
e-mail: ior.poznan.kinga@gmail.com

INFLUENCE OF SEAWEED EXTRACT (*ECKLONIA MAXIMA* L.) APPLIED WITH TEBUCONAZOLE ON TWO CULTIVARS OF WINTER RAPE

Summary

Field experiments were carried out in the years 2008-2010 in the Institute of Plant Protection – National Research Institute in Poznan (Poland). The experiments were established on two population cultivars of winter oilseed rape: 'Californium' and 'Monolit', in 4 replications, using randomized block design. The aim of the study was to assess the influence of tebuconazole (Brasifun 250 EC) and marine algae extract (Kelpak SL) applied separately or in the mixture on plants of two winter oilseed rape cultivars. Kelpak SL was used once or twice, in different growth stages of the crop. A mixture of the both examined preparations was applied only in one growth stage of winter oilseed rape. The effect of tebuconazole, marine algae extract and the mixture of the both substances on winter rape plants was much more determined by weather conditions of plant vegetation than by the cultivar factor. The mixture of tebuconazole and algae, similarly as algae alone, did not influence plant height. Only tebuconazole applied separately showed a growth retardant effect on winter oilseed rape. In the mixture the retardant effect of tebuconazole was inhibited by the addition of algae extract. All the experimental combinations were characterized by a greater leaf chlorophyll content. The examined substances applied separately or in a mixture considerably increased crop yielding.
***Key words**: biostimulant, triazole, seaweeds, mixture, cultivars, chlorophyll, yield*

OCENA DZIAŁANIA MIESZANINY EKSTRAKTU Z ALG (*ECKLONIA MAXIMA* L.) Z TEBUCONAZOLEM W ODMIANACH RZEPAKU OZIMEGO

Streszczenie

Ścisłe doświadczenia polowe prowadzono w latach 2008-2010 w Instytucie Ochrony Roślin – Państwowym Instytucie Badawczym w Poznaniu (Polska). Doświadczenia założono w dwóch odmianach populacyjnych rzepaku ozimego: Californium i Monolit, w 4 powtórzeniach, metodą bloków losowanych. Celem badań była ocena możliwości łącznego stosowania biostymulatora, będącego wyciągiem z alg (Kelpak SL) z popularnie stosowanym fungicydem – tebukonazolem (Brasifun 250 EC). Kelpak SL stosowano jednorazowo lub dwukrotnie, w różnych fazach rozwojowych rośliny uprawnej. Mieszaninę obu badanych preparatów aplikowano tylko w jednej fazie rozwojowej rzepaku. Działanie tebukonazolu, ekstraktu z alg morskich i mieszaniny obu substancji na rośliny rzepaku ozimego było w większym stopniu determinowane warunkami pogodowymi towarzyszącymi wegetacji roślin aniżeli czynnikiem odmianowym. Mieszanina tebukonazolu z algami, podobnie jak i same algi nie wpływały na wysokość roślin. Działanie skracające rzepak wykazał tylko tebukonazol aplikowany oddzielnie. W mieszaninie retardacyjne działanie tebukonazolu było hamowane przez dodatek wyciągu z alg. Wszystkie badane kombinacje doświadczalne charakteryzowały się większą zawartością chlorofilu w liściach. Badane substancje stosowane oddzielnie lub w mieszaninie znacznie zwiększały plonowanie rzepaku.
***Słowa kluczowe**: biostymulator, triazole, algi morskie, mieszanina, odmiana, chlorofil, plon*

1. Introduction

In the last few years importance of preparations containing marine algae in biostimulation of cultivated plants has been growing. Algae of the greatest activity regulating plant growth and development include brown algae, especially such species as: *Ascophyllum nodosum*, *Laminaria hyperborea, Laminaria digitata, Fucus vesiculosus, Durvillea potatorum* and ***Ecklonia maxima***. Phytohormones contained in brown algae (auxins, cytokinins, polyphenols, e.g. alginic acid) facilitate the processes of plant adaptation to stress conditions (mainly through stimulation of the root system) (Verkleij 1992). Phytohormones contained in algae help plants adapt to stress conditions mainly by stimulation of the root system and maintaining constant cell hydration. It has been demonstrated that algae effect greatly depends on used dose, frequency of application and species of cultivated plant. Literature data on the effect of algae extracts are not consistent. Many researches on the effectiveness of natural preparations are contradictory and at the same time difficult to verify due to scarce number of experiments. One of algae-based preparations which has gained a little more interest in worldwide scientific research is Kelpak. A positive effect of this biostimulator has been used in wine and citrus growing, orchard cultivation, ornamental plant cultivation and to a lesser degree also in agricultural farming. The current researches on preparation Kelpak are focused on determination of the optimal dosage and time of application. It has been demonstrated that foliar application of Kelpak enables to obtain higher yield of better quality (Ferreira and Lourens, 2002; Verkleij, 1992; Zodape, 2001).

Triazole fungicides are commonly used for oilseed rape protection against diseases. Some fungicides of the group display not only fungicidal properties but also properties of plant growth and development regulators (Coules *et al*., 2002; Luster and Miller, 1993). From the biochemical point of view, properties of these triazoles result from their double effect on a plant: inhibition of gibberellin biosynthesis in a plant (retardant properties) and inhibition of sterole biosynthesis (fungicide properties). Substances exhibiting

such double effect include e.g. paclobutrazol, uniconazol and a newer generation of triazoles – metconazole and tebuconazole. The last scientific reports demonstrate that due to their properties, some of triazole preparations are applicable as substances influencing a oilseed rape growth and they are able to modify canopy structure as well (Berry and Spink, 2009; Henneken, 2000; Rademacher, 2000). It was demonstrated that the above-mentioned triazoles increase plant antioxidant potential by inhibition of gibberellin biosynthesis (Kraus *et al.*, 1995). The potential is expressed among other things as reduction in transpiration during drought by an increase in the amount of cuticular wax on leaves (Flecher *et al.*, 2000). In addition, they stimulate or inhibit the growth of roots, have a positive effect on the synthesis of chlorophyll and carotenoids (plants treated with triazoles have a typical dark green color), increase the activity of cytokinins, protect against frost and high temperature (thermotolerance) (Kraus *et al.*, 1995).

The aim of the study was to assess the influence of tebuconazole and marine algae extract applied separately or in the mixture on plants of two winter oilseed rape cultivars. The usefulness of the research presented in the study was largely based on the practical aspect because both triazole fungicides and biostimulating algae-based preparations can be used on winter oilseed rape plants in the same time (developmental stages). There are no reports in scientific literature on the possibility of joint use of algae extracts and synthetic plant protection products, including also fungicides. From the practical point of view, it seems logical to not undertake research on the use of algae extracts with herbicides (mitigation of herbicide effect by weed enhancement), however, in the case of a fungicide acting as a plant growth and development regulator, such research may be not only of scientific importance but also may be significant for agricultural practice. In this study only the plant growth regulation effect of both substances was estimated.

2. Materials and methods

Field experiments were carried out in the years 2008-2010 in the Experimental Station Winna Gora (52° 12′ 0″ N 17° 27′ 0″ E) belonged to the Institute of Plant Protection – National Research Institute in Poznan. The experiments were established in randomized block design in 4 replications with two popular in Poland cultivars of winter oilseed rape: 'Monolit' and 'Californium'. The surface area of plots was 16.5 m^2 and the width of interrow spacing – 25 cm. In 2008 the cultivated plants were sown on 26 August, and in 2009 on 27 August in the amount of 60 seeds/m^2. In the season 2008/2009 the forecrop was a mixture of cereals (spring wheat, spring barley and oat), and in the season 2009/2010 – winter wheat. The experiments were conducted in grey-brown podzolic soil, IVa class, of pH 5.8-5.9 and a content of organic matter of 0.82-0.86 %, depending on year of research. Mineral fertilization was used: N 185 kg·ha^{-1}, P 105 kg·ha^{-1} and K 105 kg·ha^{-1}. Plant protection was used in the whole experiment according to recommendations for the winter oilseed rape.

Experimental treatments included:

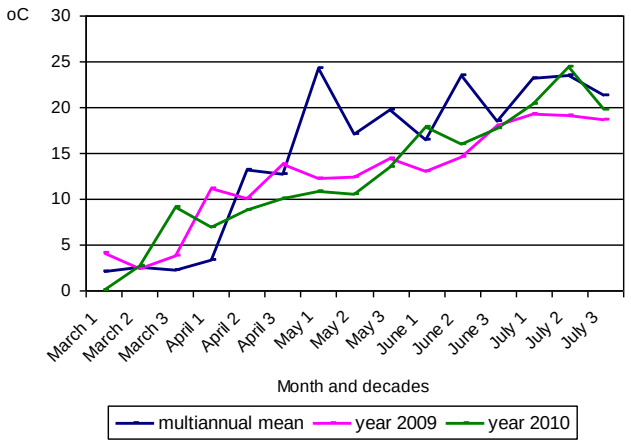
1. Control
2. Kelpak SL (extract of marine algae *Ecklonia maxima*), applied at a dose of 2.0 dm^3·ha^{-1} in the spring time (BBCH 31)
3. Kelpak SL, applied twice: at a dose of 1.5 dm^3·ha^{-1} in the spring time (BBCH 31) and at a dose of 1.5 $dm^3 \cdot ha^{-1}$ (BBCH 32)
4. Mixture of preparations Kelpak SL and Brasifun 250 EC (tebuconazole), applied at a dose of 2.0 dm^3·ha^{-1} + 1.0 dm^3·ha^{-1} in the spring time (BBCH 31)
5. Mixture of preparations Kelpak SL and Brasifun 250 EC (tebuconazole), applied at a dose of 1.5 dm^3·ha^{-1} + 1.0 dm^3·ha^{-1} in the spring time (BBCH 31)
6. Brasifun 250 EC (tebuconazole), applied at a dose of 1.0 dm^3·ha^{-1} in the spring time (BBCH 31).

Applications were carried out with a plot sprayer of the volume of 4 dm^3. During the applications 200 dm^3 of water per ha of the pressure of 3 bar were used. Temperature during the applications was 13°C for the BBCH 31 stage and 17°C for the applications at the BBCH 32 stage in 2009, and 15°C and 19°C in 2010, respectively.

In the experiments in the sample of 25 plants from each plot, plant height, number of siliques per plant and number of seeds per silique were assessed. Chlorophyll content in leaves was measured 14 days after the last treatment with the use of the SPAD method. Assessments of chlorophyll content were made on the basis of 30 measurements of one experimental plot. The surface area of a SPAD measurement on a leaf was 2 mm x 3 mm. Thousand seed weight was calculated for 4 samples of oilseed rape, 250 seeds each, of each experimental combination. Also yield, seed moisture content and seed protein and fat content were assessed. Winter oilseed rape harvesting was conducted with a Wintersteiger combine of the whole plot surface area and yield was expressed as tons per ha at humidity of 12 %. Seed qualitative analysis was carried out with an Inframatic 8100 analyzer.The results were statistically analyzed with the use of analysis of variance, significance of differences was assessed with Tukey's test and significance level was 0.05. Statistical calculations were performed using FR – ANALWAR – 4.3 software.

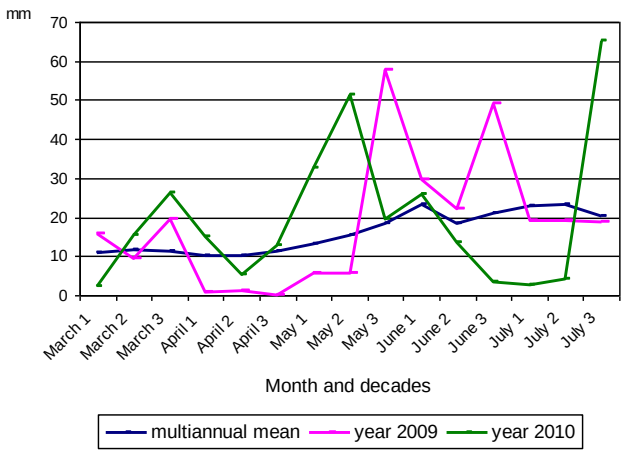
2.1. Meteorological conditions

Field experiments were run under slightly different meteorological conditions every year of the experiment (Figure 1 and 2).



Source: Own work

Figure 1. Air temperature during spring vegetation of winter oilseed rape



Source: Own work

Figure 2. Precipitation during the spring vegetation of winter oilseed rape

The year 2009 was characterized by higher temperatures during spring vegetation of the crop in comparison with the year 2010, however, precipitation was considerably lower. In the beginning of spring vegetation of oilseed rape (late March, April) in 2009, temperature was similar to the mean of the multiannual period, while in 2010 temperature of this period considerably exceeded the multiannual mean. In the both years of the research, comparable temperatures, yet lower than the multiannual mean, were observed in May and June. A considerable precipitation shortage occurred in 2009 in April until mid-May. In 2010 the amount of precipitation in April did not differ significantly from the multiannual mean, but in early- and mid-May high rainfall was observed, nearly three times exceeding the multiannual mean value for this period.

3. Results

Statistical analysis of the results showed that in the both oilseed rape cultivars the lowest plants were observed in the experimental subjects which received tebuconazole (Table 1). In cultivar 'Monolit' in the first year of the research tebuconazole shortened the plants by 8 % in comparison with the control and by 6-7 % when compared to algae preparation. In the next year of the research, the obtained plants were shorter by 10 % after application of tebuconazole when compared to the control, and by 9 % shorter than the plants after application of the algae preparation in two developmental stages and by 12 % shorter than the plants receiving one-time application of Kelpak (2.0 $dm^3 \cdot ha^{-1}$). In the case of cultivar 'Californium', tebuconazole shortened the plants by 9-10 % (depending on year) in comparison with the control, and by 11-12 % when compared to preparation Kelpak SL. In the both examined cultivars, the use of the mixture of tebuconazole and algae extract did not result in a significant decrease in plant height, however, a tendency was observed for the plants to shorten. None of the oilseed rape cultivars responded with changes in plant height to application of algae extract.

In the first year of the research, a significant increase in SPAD value, describing leaf chlorophyll content in plants which received the mixture of algae and tebuconazole and the combination with fungicide alone, was observed in cultivar 'Monolit' (Table 1). The increase in chlorophyll content in those combinations amounted to 7-9 % for the mixture of preparations and 15 % for the fungicide alone. The algae preparation used alone did not change the amount of chlorophyll in leaves of the cultivated plant. Slightly different results for the cultivar were obtained in the subsequent year of the research. All the experimental subjects exhibited an increase in the trait, and the most beneficial effect (an increase in chlorophyll content by 17 %) was actually obtained for the algae extract, applied once on oilseed rape plants. Application of the mixture of algae and tebuconazole resulted in an increase in the content of the chlorophyll by 12-14 %, and application of the fungicide alone – by 15 %, in comparison with the control. In the case of cultivar 'Californium' in the first year of the research, plants of all the experimental combinations were characterized by a higher chlorophyll level in leaves when compared to the control. Algae applied separately or at a greater dose in the mixture with fungicide increased chlorophyll content in leaves by 19-21 %, while tebuconazole alone and its mixture with algae (a smaller dose) increased chlorophyll content by 16 % in comparison with the control. Significant differences were not observed between the analyzed subjects for this trait. In the subsequent year of the research on this cultivar, a significant increase in chlorophyll content by 6-7 % was obtained only in the combinations where tebuconazol was used alone or in the mixture.

All treatments of both cultivars were characterised by significant increase of number of siliques per plant. In the both experimental years the best effect on this trait was observed after application of seaweed extract with tebuconazole. The preparation mixtures increased number of siliques per plant by over 20 % for cultivar 'Monolit' and 15 % for cultivar 'Californium' (Table 2). Changes in the number of seeds per silique of cultivar 'Monolit' were obtained only after application of the mixture of algae preparation and tebuconazole (a higher dose of the algae-based preparation; 2.0 + 1.0 $dm^3 \cdot ha^{-1}$) (Table 2). In the subject oilseed rape siliques contained about 30 % more seeds than the control. However, such a result was obtained only in one year of the research (2008/2009). The seeds of the subject were also characterized by greater weight. Thousand seed weight was higher by 30 % than that of the control (Table. 3). A significant increase in thousand seed weight was obtained also in the plants treated with the mixture of the algae preparation, applied at a lower dose with tebuconazole. In cultivar 'Californium' in the both years of the research, the mixture of the algae preparation and fungicide (irrespective of the dose of the algae preparation) increased the number of seeds per silique – in the first year by 33 % and in the second year by 24 % (Table 2).

An increase in seed yield was obtained in the both years of the research for the both winter oilseed rape cultivars in subjects treated with the preparations separately or in the mixture (Table 3). In the case of cultivar 'Monolit', the increase in yield in 2008/2009 amounted to 16-26 %, and in 2009/2010 – 7-14 %, when compared to the control. In the first year of the research, the fungicide displayed the most beneficial effect on the trait, while in the second year it was its mixture with algae, applied at a higher dose. However, statistical analysis did not show differences in yield between the plants treated with algae alone and those treated with the mixture of algae and fungicide. For cultivar 'Californium' in the first year of the research the increase in yield was 16-23 %, and in the second year – 18-22 %, when

compared to the control. In 2008/2009 the lowest yielding – 16 %, was obtained after one-time application of preparation Kelpak SL. No statistically significant differences were observed between the other experimental combinations. Also in 2009/2010 no significant differences were found between the treated subjects.

Tebuconazole used together with algae increased protein content in oilseed rape seeds (Table 4). The changes were observed only in the first research year in the both cultivars. In that research combination, 11-13 % more protein was noted in cultivar 'Monolit' than in the seeds of the control plants, while in the case of cultivar 'Californium' the increase in the parameter was 6-8 %. The examined preparations did not affect seed fat content.

Table 1. Influence of seaweed extract Kelpak SL and tebuconazole (Brasifun 250 EC) on plant height and chlorophyll content in the winter oilseed rape leaves

Treatment and time of application in BBCH scale	Dose dm· ha^{-1}	Plant height (cm)			Chlorophyll content in leaves (SPAD)		
Winter oilseed rape cv 'Monolit'		Year		Average of years	Year		Average of years
		2008/ 2009	2009/ 2010		2008/ 2009	2009/ 2010	
Control	–	141	147	144	585	705	645
Kelpak BBCH 31	2,0	139	150	145	573	827	700
Kelpak BBCH 31/BBCH 32	1,5/1,5	140	145	143	598	761	680
Kelpak + Brasifun BBCH31	2,0 + 1,0	134	138	136	604	790	697
Kelpak + Brasifun BBCH 31	1,5 + 1,0	136	136	136	637	802	720
Brasifun BBCH 31	1,0	130	132	131	672	813	743
	$LSD_{0,05}$	8.3	11.2	10.7	51.1	48.6	50.1
Winter oilseed rape cv 'Californium'							
Control	–	150	149	150	529	712	621
Kelpak BBCH 31	2,0	153	146	150	628	696	662
Kelpak BBCH 31/BBCH 32	1,5/1,5	155	150	153	639	705	672
Kelpak + Brasifun BBCH31	2,0 + 1,0	142	141	142	630	760	695
Kelpak + Brasifun BBCH 31	1,5 + 1,0	144	139	142	616	758	687
Brasifun BBCH 31	1,0	136	134	135	613	758	686
	$LSD_{0,05}$	11.1	9.4	12.4	32.7	44.7	42.7

Source: Own work

Table 2. Influence of seaweed extract Kelpak SL and tebuconazole (Brasifun 250 EC) on number of seeds per silique and number of siliques per plant of winter oilseed rape

Treatment and time of application in BBCH scale	Dose dm· ha^{-1}	Number of seeds per silique			Number of siliques per plant		
Winter oilseed rape cv 'Monolit'		Year		Average of years	Year		Average of years
		2008/ 2009	2009/ 2010		2008/ 2009	2009/ 2010	
Control	–	16	20	18	121	117	119
Kelpak BBCH 31	2,0	17	22	20	135	132	137
Kelpak BBCH 31/BBCH 32	1,5/1,5	18	22	20	143	137	140
Kelpak + Brasifun BBCH31	2,0 + 1,0	21	24	23	146	141	138
Kelpak + Brasifun BBCH 31	1,5 + 1,0	16	24	20	149	142	146
Brasifun BBCH 31	1,0	15	23	19	139	131	135
	$LSD_{0,05}$	4.1	ns	ns	12.1	14.4	8.6
Winter oilseed rape cv 'Californium'							
Control	–	18	21	20	112	129	121
Kelpak BBCH 31	2,0	21	23	22	124	145	137
Kelpak BBCH 31/BBCH 32	1,5/1,5	19	25	22	125	139	132
Kelpak + Brasifun BBCH31	2,0 + 1,0	24	26	25	129	149	141
Kelpak + Brasifun BBCH 31	1,5 + 1,0	24	26	25	132	151	139
Brasifun BBCH 31	1,0	20	25	23	126	145	139
	$LSD_{0,05}$	4.5	5.0	4.5	11.8	9.7	13.6

ns – not significant differences

Source: Own work

Table 3. Influence of seaweed extract Kelpak SL and tebuconazole (Brasifun 250 EC) on weight of 1000 seeds and seed yield of winter oilseed rape

Treatment and time of application in BBCH scale	Dose dm· ha^{-1}	Weight of 1000 seeds			Yield t·ha^{-1}		
Winter oilseed rape cv 'Monolit'		Year		Average of years	Year		Average of years
		2008/ 2009	2009/ 2010		2008/ 2009	2009/ 2010	
Control	–	4.54	5.91	5.23	4.08	3.66	3.87
Kelpak BBCH 31	2,0	4.81	5.81	5.31	4.96	4.02	4.49
Kelpak BBCH 31/32	1,5/1,5	4.47	5.83	5.15	5.00	3.98	4.49
Kelpak + Brasifun BBCH31	2,0+1,0	5.09	5.81	5.45	4.93	4.18	4.56
Kelpak + Brasifun BBCH 31	1,5+1,0	4.91	5.84	5.38	4.74	3.93	4.34
Brasifun BBCH 31	1,0	4.84	5.78	5.31	5.13	4.08	4.61
	$LSD_{0,05}$	0.367	ns	ns	0.536	0.214	0.29
Winter oilseed rape cv 'Californium'							
Control	–	4.93	5.91	5.42	3.67	3.54	3.61
Kelpak BBCH 31	2,0	5.11	5.81	5.46	4.26	4.24	4.25
Kelpak BBCH 31/32	1,5/1,5	5.17	5.72	5.45	4.39	4.21	4.30
Kelpak + Brasifun BBCH31	2,0+1,0	5.08	5.91	5.50	4.49	4.32	4.41
Kelpak + Brasifun BBCH 31	1,5+1,0	5.00	5.83	5.42	4.43	4.27	4.35
Brasifun BBCH 31	1,0	5.19	5.92	5.56	4.53	4.19	4.36
	$LSD_{0,05}$	ns	ns	ns	0.505	0.297	0.363

ns – not significant differences

Source: Own work

Table 4. Influence of seaweed extract Kelpak SL and tebuconazole (Brasifun 250 EC) on protein and fat content in seeds of winter oilseed rape

Treatment and time of application in BBCH scale	Dose dm· ha^{-1}	Protein content (%)			Fat content (%)		
Winter oilseed rape cv 'Monolit'		Year		Average of years	Year		Average of years
		2008/ 2009	2009/ 2010		2008/ 2009	2009/ 2010	
Control	–	14.9	19.6	17.3	50.5	47.2	48.9
Kelpak BBCH 31	2,0	14.1	19.6	16.9	51.2	47.3	49.3
Kelpak BBCH 31/32	1,5/1,5	15.7	19.4	17.6	50.2	47.4	48.8
Kelpak + Brasifun BBCH31	2,0+1,0	16.8	19.7	18.3	49.5	47.3	48.4
Kelpak + Brasifun BBCH 31	1,5+1,0	16.5	19.3	17.9	50.9	47.7	49.3
Brasifun BBCH 31	1,0	14.5	19.8	17.2	50.8	47.3	49.1
	$LSD_{0,05}$	1.41	ns	ns	ns	ns	ns
Winter oilseed rape cv. 'Californium'							
Control	–	19.6	19.8	19.7	45.8	45.6	45.7
Kelpak BBCH 31	2,0	19.1	20.1	19.6	45.0	45.4	45.2
Kelpak BBCH 31/32	1,5/1,5	20.5	19.2	19.9	44.3	46.2	45.3
Kelpak + Brasifun BBCH31	2,0+1,0	21.1	19.7	20.4	45.1	45.6	45.4
Kelpak + Brasifun BBCH 31	1,5+1,0	20.8	20.0	20.4	45.6	45.4	45.5
Brasifun BBCH 31	1,0	20.6	20.3	20.5	45.1	45.0	45.1
	$LSD_{0,05}$	1.11	ns	ns	ns	ns	ns

ns – not significant differences

Source: Own work

4. Discussion

The retardant effect of triazole fungicides acting as growth regulators (metoconazole and tebuconazole) is partially confirmed in literature data. Although the plant shortening degree obtained in the present research (on average by 8-10 %), resulting from application of triazole fungicide, is within the range of values provided by literature, not all authors agree with the statement that triazole fungicides have a height-reducing effect under every conditions. Cieślicki and Toboła (2007) and Cieślicki and Toboła (2008) state that rape plant shortening as an effect of triazole application may amount to ca. 5-6 %. A higher percentage of plant shortening (even by 17%) was obtained by Bączkowska *et al.*, (2006). A decrease in winter oilseed rape height resulting from application of triazoles was obtained also by Berry and Spink (2009), Gundula *et al.* (1990), Mączyńska *et al.*, (2007) and Weber and Karolewski (2001). On the other hand, Cieślicki and Muśnicki (2006) state that tebuconazole applied in the spring does not affect oilseed rape height. Similarly, Cieślicki and Toboła (2006) demonstrated in their research that triazole fungicide effective as a plant growth and development regulator (metoconazole) does not exhibit retardant activity.

The research presented in this study demonstrates the fact that the retardant effect of tebuconazole is inhibited by influence of the algae extract, however, the tendency for plants to shorten was maintained. Algae applied separately did not affect the trait. Similarly, there are reports in scientific literature on lack of influence of algae on plant height (De Waele *et al.*, 1988; Matysiak *et al.*, 2012) or, more frequently, reports on increased plant height as a biostimulant effect of algae preparations (Blunden, 1991; Matysiak *et al.*, 2010).

Literature reports describing influence of triazole fungicides on plant structure and physiological processes taking place in a plant are unanimous that triazoles have a considerable influence on the intensity of photosynthesis and the content of photosynthetic pigments. It is largely related to increased size of chloroplasts in plant cells and greater chlorophyll concentration (Fletcher *et al.*, 2000; Sopher *et al.*, 1999). Beneficial influence of tebuconazole on this trait in winter oilseed rape plants has been confirmed among others by Matysiak *et al.*, (2010), who obtained an increase in chlorophyll (18-20 %) comparable to the results obtained in the present study (15-16 %). In the study by Matysiak *et al*., (2010), tebuconazole was applied in the same developmental stage as in the present study. Also algae extracts show a beneficial effect on chlorophyll content in plant cells, although it seems that their influence on this trait is more determined by agrotechnological and environmental conditions than it is in the case of other plant growth and development regulators, including triazoles. Some researchers state that chlorophyll content in leaves as an effect of algae is very low (Venkataraman et al., 1997), however, there are also studies showing a highly beneficial influence of algae on this trait (Blunden *et al*., 1996). The results obtained in the present study demonstrate that both tebuconazole and algae extracts, applied separately or in the mixture, considerably increase chlorophyll level in rape leaves, however, the results for this trait do not prove clearly the fact that the mixture of fungicide and algae exhibits a more beneficial effect than separate application. Additionally, in the case of chlorophyll content in leaves, meteorological conditions of plant vegetation and the cultivar factor seem to be of key importance, which is confirmed by the results of the present study.

In the conducted experiments an increase in the number of seeds per silique occurred only after application of the mixture of the examined preparations. No changes were observed for the trait neither in the case of separate application of algae nor after application of triazole fungicide. Lack of differences in the number of seeds per silique after application of triazole fungicide has been reported also by Cieślicki and Muśnicki (2006), Matysiak *et al*., (2010) and Toboła *et al*., (2008), whereas Cieślicki and Toboła (2007) have found an adverse influence of tebuconazole on that trait. However, a beneficial effect of triazole preparations on silique formation has been reported by Berry and Spink (2009). In scientific literature it has been demonstrated that algae extracts significantly increase thousand seed weight of oilseed rape (Pietryga and Matysiak 2002). However, in the present study an advantageous effect on thousand seed weight of winter rape was obtained only as a result of application of algae and tebuconazole only in one of the examined cultivars ('Monolit') in one research year. Influence of tebuconazole on thousand seed weight of oilseed rape has been described e.g. in a paper by Mączyńska and Krzyzińska (2000), who obtained an increase in thousand seed weight of 3-4% after the use of triazole fungicide. In the present research, the mixture of algae and tebuconazole caused an increase in thousand seed weight by 8-12 %. Cieślicki and Toboła (2007) have not observed any differences in thousand seed weight of winter rape after application of tebuconazole. Similarly, no influence of metoconazole has been demonstrated in a study by Pits *et al*., (2008).

Both the algae-based preparation, fungicide preparation and the mixture of the both substances favorably affected yielding of rape plants. The results were obtained in the both years of the research for the both rape cultivars. However, no significant differences were found between the treatments. The results obtained in the present study are confirmed by literature data. An increase in yield after triazole fungicide application has been demonstrated also by Bączkowska *et al*., (2006), Cieślicki and Toboła (2007); Mączyńska and Krzyzińska (2000), Sova *et al*., (1998) and Toboła *et al*., (2008) and a positive algae effect on oilseed rape yielding has been described among others by Matysiak (2012) and Pietryga and Matysiak (2002). However, all the named researchers are unanimous that influence of triazole fungicides regulating plant growth and development and influence of biostimulators containing algae on yielding is greatly determined by developmental stage of a cultivated plant at the moment of treatment and by weather conditions. Also influence of these preparations on yield quality depends on humidity and thermal conditions during the plant vegetation period. Changes in rape seed quality were revealed only in the combinations with the use of the mixture of algae and tebuconazole. Admittedly, the changes were observed for the both cultivars of the cultivated plant but only in one research year. In the year with a higher amount of precipitation, just after application of the preparations, protein content in seeds increased. However, the experiments did not show influence of the examined preparations on fat content in seeds. The lack of influence of triazole preparations on oil content in oilseed rape seeds has been demonstrated also by Baylis and Wright (1990). Scarce scientific reports on the influence of algae on qualitative parameters of yield of cultivated plants indicate that algae preparations may affect yield quality, and thus e.g. Matysiak *et al*., (2010) in their study have proved that algae may increase protein content in winter oilseed rape seeds.

5. Conclusions

The effect of tebuconazole, marine algae extract and the mixture of the both substances on plants of two winter rape cultivars depended mainly on weather conditions during the plant vegetation period. The mixture of tebuconazole and algae, similarly as algae alone, did not affect plant height. Only separately applied tebuconazole exhibited plant-shortening effect. A higher chlorophyll content in leaves was observed in all the combinations with the preparations than in the control. The trait largely depended on year. None of the examined cultivars exhibited differences in chlorophyll amount in leaves between the mixture of tebuconazole and algae and tebuconazole alone. Significant increase in the number of seeds per silique was obtained only for cultivar 'Californium' after using the mixture of tebuconazole and algae (irrespective of algae preparation dose). For the same subjects a considerable increase in thousand seed weight was obtained for cultivar 'Monolit'. In the first year of the research, an increase in protein content in seeds was noted in the both cultivars under the influence of the mixture of tebuconazole and algae. The examined substances used separately or as the mixture similarly increased rape yielding.

6. References

[1] Baylis, A.D. and Wright, T.J. 1990. The effects of lodging and a paclobutrazol chlormequat chloride mixture on the yield and

quality of oilseed rape. Ann. Appl. Biol., 116, 287-295.
[2] Bączkowska, E., Praczyk, T. and Stachecki, S. 2006. Effect of chloromequat and paclobutrazol on the growth and yield of winter rape. Prog. Plant Prot./Post. Ochr. Rośl., 46(2): 291-294.
[3] Berry, P.M. and Spink, J.H. 2009. Understanding the effect on a triazole with anti-gibberelin activity on the growth and yield of oilseed rape (Brassica napus). J. Agr. Sci., 147: 273-285.
[4] Blunden, G. 1991. Agricultural uses of seaweeds and seaweed products. In: European Seaweed Resources: Uses and Potential. Giury, M.D. and Blunden, G. eds.: 65-81.
[5] Blunden, G., Jenkins T. and Liu, Y.W. 1996. Enhanced chlorophyll levels in plants treated with seaweed extract. J. Appl. Phycol., 8: 535-543.
[6] Bruns, G., Kuchenbuch R., Jung J. 1990. Influence of a triazole plant growth regulator on root and shoot development and nitrogen utilisation of oilseed rape (Brassica napus L.). J. Agron. Crop Sci., 165 (4): 257-262.
[7] De Waele, D., McDonald, A.H. and De Waele, E. 1988. Influence of seaweed concentrate on the growth of maize and reproduction of Pratylenchus zeae (Nematoda). Nematologica, 34(1): 71-77.
[8] Cieślicki, W. and Toboła, P. 2007. Influence of growth regulators on plant habitus and yielding of spring oilseed rape. Prog. Plant Prot./ Post. Ochr. Rosl., 47(3): 60-63.
[9] Cieślicki, W. and Muśnicki, Cz. 2006. Influence of Horizon 250 EW on healthiness and growth of winter oilseed rape. Prog. Plant Prot./Post Ochr. Rosl., 46(2): 688-691.
[10] Coules, A.E., Lunn, G.D. and Rossal, S. 2002. Disease and canopy control in oilseed rape using triazole fungicides. The BCPC Conference: Pests and diseases, Vol 1 and 2. Proceedings of an International Conference, Brighton, UK, 18-21 November 2002: 617-622.
[11] Fletcher, R.A., Gilley, A., Sankhla, N. and Davis, T.D. 2000. Triazoles as plant growth regulators and stress protectants. Hort Rev., 24: 55-138.
[12] Luster, D.G. and Miller, P. A. 1993. Triazole plant growth regulator binding to native an detergent-solubilized plant microsomal cytochrome P450. Pestic. Biochem. Phys., 46: 27-39.
[13] Ferreira, M.I. and Lourens, A.F. 2002. The efficacy of liquid seaweed extract on the yield of canola plants. S. Afr. J. Plant and Soil, 19 (3): 159-161.
[14] Henneken, M., Föller, I., Edel, U. and Paul, V.H. 2000. First results on the effects of the new triazole fungicide Caramba® against oilseed rape diseases and plant growth in regard to application dates in 1998/99. IOBC/WPRS Bull., 23(6): 95-108.
[15] Kraus, T.E., Evans, R.A., Fletcher, R.A. and Pauls, K.P. 1995. Paclobutrazol enhances tolerance to increased levels of UV-B radiation in soybean (Glycine max) seedlings. Can. J. Bot., 73: 797-806.
[16] Matysiak, K., Kaczmarek, S. and Adamczewski, K. 2010. The influence of trinexapac-ethyl, chlorocholine chloride, metconazole and tebuconazole on plant morphology and yield of winter oilseed rape depending on time of application. Rośl. Oleiste – Oilseed Crops, 31(2): 363-374.
[17] Matysiak, K., Kaczmarek, S. and Leszczyńska, D. 2012. Influence of liquid seaweed extract of Ecklonia maxima on winter wheat cv Tonacja. J. Res. Appl. Agric Engng, 57(4): 44-48.
[18] Matysiak, K., Kaczmarek, S., Kierzek, R. and Kardasz, P. 2010. Effect of seaweeds extracts and humic and fulvic acids on the germination and early growth of winter oilseed rape (Brassica napus L.). J. Res. Appl. Agric. Engng., 55(4): 28-33.
[19] Mączyńska, A., Głazek, M. and Krzyzińska, B. 2007. New possibilities of growth regulation and protection against fungal diseases of winter oilseed rape using the preparation A 14049 (difenconazole and paclobutrazol). Prog. Plant Prot./Post Ochr. Rosl., 47(2): 198-202.
[20] Mączyńska, A. and Krzyzińska, B. 2000. Influence of Horizon 250 EW on health, development and yield of winter oilseed rape. Rosl. Oleiste – Oilseed Crops, 21(1): 105-112.
[21] Pietryga, J. and Matysiak, K. 2003. The biological evaluation of Kelpak growth regulator in winter rape. Prog. Plant Prot/Post. Ochr. Rosl., 43(2): 863-865.
[22] Pits, N., Kubacki, K. and Tys, J. 2008. Influence of plant growth regulators and desiccants on a yield and quality of winter oilseed rape. Int. Agrophysics, 22: 67-70.
[23] Rademacher, W. 2000. Growth retardants: Effects on gibberelin biosynthesis and other metabolic pathways. Annu. Rev. Plant. Physiol. Plant Mol. Biol., 51: 501-531.
[24] Sopher, C.R., Król, M., Huner, N.P.A., Moore, A.E. and Fletcher R.A. 1999. Chloroplastic changes associated with paclobutrazol-induced stress protection in maize seedlings. Can. J. Bot., 77: 279-290.
[25] Sova, A.V., Vasak, J. and Soukup, J. 1998. Warianty technologii uprawy rzepaku ozimego (Brassica napus L. napus). Rosl. Oleiste – Oilseed Crops, 19: 105-111.
[26] Toboła, P., Cieślicki, W. and Kępiński, M. 2008. Influence of Cerone 480 SL and Caramba 60 SL on plant habitus and lodging of spring oilseed rape. Prog. Plant Prot./Post Ochr. Rosl., 48(2): 687-690.
[27] Verkleij, F.N. 1992. Seaweed extracts in agriculture and horticulture: A review. Biol. Agric. Hortic., 8: 309-324.
[28] Venkataraman Kumar V., Mohan V.R. 1997.Effect of seaweed liquid fertiliser on black gram. Phykos. 36(2): 43-47.
[29] Weber, Z. and Karolewski, Z. 2001. Influence of some selected triazole fungicides on winter oilseed rape growth and their protection against pathogenic fungi. Prog. Plant Prot./Post. Ochr. Rosl., 41(2): 773-775.
[30] Zodape, S.T. 2001. Seaweeds as a biofertilizer. J.Sci. Ind. Res., 60 (537): 8-382.