

## EFFECT OF COMPETITION AND WATER STRESS ON MORPHOLOGICAL TRAITS OF SPRING BARLEY GROWN WITH RED CLOVER

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**Abstract.** Based on a 3-year pot experiment carried out according to an additive design, the effect of competitive effect of red clover and water deficit was studied on morphological traits of spring barley (length of the root, shoot and ear, total and productive tillering, leaf number and grain number per ear). The experimental factors included: level of substrate moisture and sowing method. Two levels of moisture were used: the one which met plants' requirements, and one reduced by 50%. The plants were sown in a pure stand (18 plants of spring barley per pot) and in a mixed stand (18 plants of spring barley with 8 plants of red clover per pot). Biometric measurements were conducted in five development stages determined by the growth rhythm of barley in pure sowing with a higher water dose, i.e. in the stages of (BBCH scale): leaf formation (10-13), tillering (22-25), shooting (33-37), ear formation (52-55) and maturation (87-91). It was indicated that common cultivation of spring barley with red clover had no effect on the analyzed morphological traits of this cereal throughout the whole growing season. On the other hand, water deficit in the substrate resulted in a reduction in cereal growth in tillering-maturation stages, in the formation of a lower number of leaves (tillering-shooting) and in a poorer total and productive tillering. Water stress also caused formation of shorter ears with a lower number of grains filling them. Competitive effect of red clover amplified with water deficit resulted in the reduction in the number of productive shoots in barley. No effect of experimental factors was found on the root length in the initial growth stages (leaf formation-tillering).

**Key words:** development stages, *Hordeum vulgare*, interaction between plants, water deficit

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## INTRODUCTION

The beneficial effect of catch crops on physical, chemical and biological properties of soil, and on the reduction in weed infestation was confirmed in many studies [Andrzejewska 1999, Hauggaard-Nielsen *et al.* 2001, Jensen 1991, Thomsen and Christensen 2004]. Companion crops also reduce infestation with diseases [Moszczyńska and Płąkowska 2005] and contribute to an increase in productive capacities of successively grown plants [Płaza and Soszyński 2010]. Apart from their positive effect, companion crops while using the same environmental resources, become competitive for the cover crop, which may reduce its growth and yield [Sobkowicz and Lejman 2011, Wanic *et al.* 2006]. One of the most important environmental resources determining plant productivity is water. Plants' response to water deficit is complex. It includes adaptation changes and harmful effects of water stress, as a result of which there occurs disturbance in fundamental life processes [Singer *et al.* 2003]. One of the effects of these changes is reduction or inhibition of growth. Taking into consideration the above premises, the research was undertaken, whose aim was evaluation of the effect of competitive effect of red clover on morphological traits of spring barley in particular development stages under conditions of water stress. An alternative hypothesis was tested, which assumed a negative effect of competitive effect and reduced access to water supplies in soil on morphological traits of spring barley. Null hypothesis assumed lack of the effect of a competitive plant and diverse soil moisture on these parameters of the cereal.

## MATERIAL AND METHODS

The research was based on a pot experiment carried out in 3 cycles (of the duration of: 102, 97 and 98 days, respectively) and 4 replications in a greenhouse laboratory of the University of Warmia and Mazury in Olsztyn. The subject of analysis was spring barley (hull-less cultivar Rastik).

The experimental factors included:

- 1) level of the substrate's irrigation: higher one meeting the requirements of plants – H, and the lower one – dose reduced by 50% – L,
- 2) sowing type: pure – P and mixed – M (with clover).

The pots were filled with 8 kg of substrate, which was made up of leached brown soil, formed from heavy loam. It was characterized by a slightly acidic reaction (pH in 1 M KCl from 5.6 to 6.2), by the content of organic substance within the range from 1.84-2.52%, and by a high content of phosphorus (9.24-11.61 mg·100 g<sup>-1</sup> soil) and magnesium (8.80-9.11 mg), and an average content of potassium (12.87-14.53 mg). The soil material was fortified with mineral fertilizers at a rate of a pure component (g·pot<sup>-1</sup>): P – 0.2 g (monopotassium phosphate), K – 0.45 g (potassium sulphate) and N – 0.5 g under barley, 0.3 g – under mixture.

The experiment was set up according to an additive design [Semere and Froud-Williams 2001, Sobkowicz 2001], in which the number of plants in a mixture is the total of their density in a pure stand. According to the above design, the following density was provided in pots: in pure stand – 18 plants of spring barley, and in mixed sowing – 18 plants of spring barley along with 8 plants of red clover.

A higher dose of water was determined based on the trial experiment, in which soil moisture was measured, as well as evaporation, transpiration and water content in plants. In the growing season, the doses were diversified depending on the stage of plant's development and soil moisture content. In the successive cycles, a higher dose of water was in total: 13400, 12300 and 10900 cm<sup>3</sup> per each pot.

Plant material for the research was collected from particular pots on 5 dates, determined by the developmental rhythm of spring barley in pure sowing with a higher water dose, i.e. in the stages of (according to BBCH scale): leaf formation (10-13), tillering (22-25), shooting (33-37), ear formation (52-55) and maturation (87-91). Next, measurements of the following were taken: shoot length, leaf number per plant, ear length and the number of grains formed in them, as well as root length (taking the measurement of the longest root) in the stages of leaf formation and tillering. In further development stages, it was impossible to separate roots of the two species. In full maturity stage, the total and productive tillering was determined in barley.

The results were elaborated statistically with the use of analysis of variance for the completely randomized design, using test Duncan's test for comparisons between plots with P = 0.05. The results, being means from the 3 research cycles, are presented in the tables.

## RESULTS

Shoot length in spring barley was not modified by the sowing type throughout the whole growing season (Table 1). On the other hand, reduced water dose significantly reduced plant growth from the stage of tillering to harvest, and these differences compared to the plants with a higher water dose were from 12.1 to 16 cm.

Table 1. Shoot length in spring barley, cm  
Tabela 1. Długość pędów jęczmienia jarego, cm

Variation source (factors) Źródło zmienności (czynniki)	Factor level Poziom czynnika*	Development stages of spring barley Fazy rozwojowe jęczmienia jarego				
		leaf formation rozwój liści	tillering krzewienie	shooting strzelanie w zdźbło	ear formation kłoszenie	maturity dojrzałość
Sowing type Rodzaj siewu	P	10.6 a	35.4 a	46.2 a	53.1 a	52.9 a
	M	10.6 a	33.1 a	45.8 a	50.9 a	52.0 a
Water supply Zaopatrzenie w wodę	H	10.2 a	40.3 a	54.0 a	59.4 a	58.8 a
	L	11.0 a	28.2 b	38.0 b	44.5 b	46.1 b
Interaction between factors Współdziałanie czynników	P H	11.0 a	40.9 a	53.3 a	60.6 a	59.2 a
	P L	10.2 a	29.9 b	39.1 b	45.5 b	46.5 b
	M H	11.0 a	39.6 a	54.6 a	58.2 a	58.2 a
	M L	10.2 a	26.6 b	37.0 b	43.5 b	45.6 b

\* P – pure crop – siew jednogatunkowy, M – mix crop – siew mieszany, H – higher water dose – wyższa dawka wody, L – lower water dose – niższa dawka wody

a, b – homogenous groups: means designated with the same letter do not differ significantly with P = 0.05 – grupy jednorodne: średnie oznaczone tą samą literą nie różnią się istotnie przy P = 0,05

Sowing type had no effect on the total and productive tillering in barley, while lower water supply compared with a higher one resulted in a significantly poorer tillering and

developing a lower number of generative organs in the plant (Table 2). Compared with other plots, the competitive effect of red clover amplified with water deficit caused a significant reduction in the number of ear-bearing shoots in barley.

Table 2. Total tillering (shoot number·plant<sup>-1</sup>) and productive tillering (ear number·plant<sup>-1</sup>) in spring barley in the full maturity stage

Tabela 2. Krzewienie ogólne (liczba pędów·roślina<sup>-1</sup>) i produkcyjne (liczba kłosów·roślina<sup>-1</sup>) jęczmienia jarego w fazie dojrzałości pełnej

Variation source (factors) Źródło zmienności (czynniki)	Factor level Poziom czynnika	Total tillering Krzewienie ogólne	Productive tillering Krzewienie produkcyjne
Sowing type Rodzaj siewu	P	1.8 a	1.2 a
Rodzaj siewu	M	1.7 a	1.0 a
Water supply Zaopatrzenie w wodę	H	2.0 a	1.3 a
Zaopatrzenie w wodę	L	1.5 b	0.9 b
	P H	2.1 a	1.4 a
Interaction between factors Współdziałanie czynników	P L	1.5 b	1.0 ab
	M H	2.0 a	1.2 ab
	M L	1.5 b	0.8 b

for explanation see Table 1 – objaśnienia pod tabelą 1

Also, proximity of clover did not change the leaf number per spring barley plant in all development stages (Table 3). A significant influence of water stress was observed in the stages of tillering-shooting. The cereal developed fewer leaves then, compared with the cereal from plots with a higher water dose (by 2.1-2.9 in pure sowing and 2.4-4.9 in mixed sowing).

Table 3. Leaf number in spring barley, leaf number·plant<sup>-1</sup>

Tabela 3. Liczba liści jęczmienia jarego, szt.·roślina<sup>-1</sup>

Variation source (factors) Źródło zmienności (czynniki)	Factor level Poziom czynnika	Development stages of spring barley Fazy rozwojowe jęczmienia jarego				
		leaf formation rozwój liści	tillering krzewienie	shooting strzelanie w zdźbło	ear formation kłoszenie	maturity dojrzałość
Sowing type Rodzaj siewu	P	3.1 a	7.1 a	8.0 a	7.0 a	6.5 a
Rodzaj siewu	M	3.2 a	6.8 a	8.7 a	6.7 a	6.5 a
Water supply Zaopatrzenie w wodę	H	3.3 a	8.1 a	10.3 a	6.7 a	6.5 a
Zaopatrzenie w wodę	L	3.0 a	5.8 b	6.4 b	7.0 a	6.5 a
Interaction between factors	P H	3.3 a	8.1 a	9.5 a	6.8 a	6.7 a
	P L	2.9 a	6.0 b	6.6 b	7.2 a	6.3 a
Współdziałanie czynników	M H	3.4 a	8.0 a	11.2 a	6.6 a	6.3 a
	M L	3.1 a	5.6 b	6.3 b	6.8 a	6.6 a

for explanation see Table 1 – objaśnienia pod tabelą 1

The study results indicate a lack of the effect of cultivating barley with red clover on ear length, unlike the use of water doses (Table 4). Deficit of this environmental resource, both in the stage of tillering and maturity, resulted in the formation of significantly shorter generative organs by 2 and 1.6 cm, respectively. The effect of applying two stresses was comparable with the effect of each of them separately.

Table 4. Ear length in spring barley, cm  
 Tabela 4. Długość kłosa jęczmienia jarego, cm

Variation source (factors) Źródło zmienności (czynniki)	Factor level Poziom czynnika	Development stages of spring barley Fazy rozwojowe jęczmienia jarego	
		tillering – kłoszenie	maturity – dojrzałość
Sowing type Rodzaj siewu	P M	5.9 a 6.2 a	5.8 a 5.6 a
Water supply Zaopatrzenie w wodę	H L	7.0 a 5.0 b	6.5 a 4.9 b
	P H P L Współdziałanie czynników	6.8 a 4.9 b 7.3 a	6.5 a 5.0 b 6.4 a
	M H M L	5.1 b	4.7 b

for explanation see Table 1 – objaśnienia pod tabelą 1

Also, no competitive effect was found on the grain number per barley ear (Table 5). However, water stress caused formation of a significantly lower number of grains compared with plants growing in an environment with a higher water supply in the tillering stage by 2, and at the end of vegetation by 2.8.

Table 5. Grain number per ear of spring barley  
 Tabela 5. Liczba ziaren w kłosie jęczmienia jarego

Variation source (factors) Źródło zmienności (czynniki)	Factor level Poziom czynnika	Development stages of spring barley Fazy rozwojowe jęczmienia jarego	
		tillering – kłoszenie	maturity – dojrzałość
Sowing type Rodzaj siewu	P M	8.2 a 9.0 a	6.7 a 6.1 a
Water supply Zaopatrzenie w wodę	H L	9.6 a 7.6 b	7.8 a 5.0 b
	P H P L Współdziałanie czynników	9.1 a 6.9 b 9.8 a	7.9 a 5.5 b 7.6 a
	M H M L	7.9 ab	4.5 b

for explanation see Table 1 – objaśnienia pod tabelą 1

Statistical analysis indicated that the experimental factors did not vary the root length in spring barley both in the stage of leaf formation or in the tillering stage (Table 6). Also, no significant effect was observed of their interaction on the growth of the underground parts of the cereal.

Table 6. Root length in spring barley, cm  
 Tabela 6. Długość korzeni jęczmienia jarego, cm

Variation source (factors) Źródło zmienności (czynniki)	Factor level Poziom czynnika	Development stages of spring barley Fazy rozwojowe jęczmienia jarego	
		leaf formation – rozwój liści	tillering – krzewienie
Sowing type Rodzaj siewu	P M	5.1 a 4.2 a	18.4 a 19.8 a
Water supply Zaopatrzenie w wodę	H L	5.0 a 4.3 a	19.4 a 18.8 a
	P H P L Współdziałanie czynników M H M L	4.8 a 5.3 a 5.2 a 3.2 a	18.6 a 18.4 a 20.0 a 19.6 a

for explanation see Table 1 – objaśnienia pod tabelą 1

## DISCUSSION

In the analyzed experiment, proximity of red clover did not vary significantly the shoot length in spring barley. The result was confirmed in the studies of Kostrzewska *et al.* [2013] on the competitive interaction between spring barley and perennial ryegrass under conditions of water stress. A similar response of the discussed cereal to cultivating in mixture was observed by Sobkowicz [2001], in whose study, shoot length in barley cultivated in mixture with oats and in pure sowing was similar. Also, Rudnicki *et al.* [1996] in their studies on monoculture cultivation of spring cereal mixtures indicated that shoot length in barley growing together with wheat did not differ significantly from pure cultivation. However, in the experiments of Ignaczak and Andrzejewska [1997] as well as Kotecki *et al.* [1997], cereals cultivated in mixtures with leguminous plants were higher than in pure cultivation. Orzech [2013] found that undersown red clover stimulated barley to form longer shoots than in pure sowing. This may be connected with a dynamic barley growth and effective use of environmental resources at the beginning of the growing season, which is indicated by Lamb and Cahill [2006] as well as Lambers and Poorter [1992]. Results of the above mentioned authors are inconsistent with the studies of Michalska and Wanic [2008], Rudnicki [1994], Rudnicki and Wasilewski [1993] as well as Sobkowicz [2003], who indicated that cultivating barley in a mixture with another species reduced this cereal's height. In our studies, water deficit from the tillering stage caused shortening of barley shoots. Moreover, the effect of the water factor was stronger than the competitive effect of clover. It was the period of the highest barley requirement for water because of an intensive growth, whereas slowly growing clover plants were at that time yet poorly developed. Our results to a large extent correspond with those presented by Kostrzewska *et al.* [2013], who observed shortening of the shoot length in barley on plots with a reduced substrate moisture independent of the presence of competitive ryegrass. Also, Semere and Froud-Williams [2001] in their studies on the competition under conditions of water stress, indicated that deficit of this component reduced height of corn by 27%, of pea by 32% compared with plants cultivated under conditions of a higher substrate moisture. Also, Clary *et al.* [2004] observed significant differences in the height of *Rosmarinus officinalis* and *Avena fatua*, competing with each other, in

favor of grasses having access to greater water supplies. Strong negative effects of water stress on barley growth is also confirmed by Samarah *et al.* [2009].

Proximity of red clover had no effect on tillering in plants or on the leaf number in spring barley throughout the whole experiment. This is confirmed by the results of other experiments with barley cultivated in a mixture with Italian ryegrass [Kostrzewska *et al.* 2013] and red clover [Orzech 2013]. On the other hand, Płaza and Ceglarek [2004] indicated a beneficial effect of the undersown white clover and mixture of white clover with Italian ryegrass on the shoot number in barley. However, Michalska and Wanic [2008] observed a negative effect of barley cultivation in mixture with pea on the number of shoots and leaves in the cereal plant. Also, Rudnicki and Wasilewski [1993] as well as Sobkowicz [2001] indicated that barley cultivated in a mixture showed poorer tillering than in the pure sowing. In the conducted experiment, water deficit in the substrate reduced the total and productive tillering, and resulted in a reduction in the number of leaves in the stages tillering – shooting, which is confirmed in the studies of Kostrzewska *et al.* [2013].

In our research, no effect of clover on the ear length in spring barley was indicated or on the grain content of the ear. Analogical results were obtained by Orzech [2013]. On the other hand, Sobkowicz and Lejman [2011] observed a negative effect of Persian clover, while Michalska and Wanic [2008] as well as Rudnicki and Wasilewski [2000] of pea on the length of generative organs in barley. Lower grain yields in barley cultivated with undersown crops, including red clover, were found by Jaskulski [2004], Kankänen and Eriksson [2007] as well as Wanic *et al.* [2012]. The negative effect of water deficit on the ear length and the grain number per ear, which was observed in our studies, is also confirmed by Kostrzewska *et al.* [2013], Massoudifar *et al.* [2013] as well as Samarah *et al.* [2009]. According to Oosterhuis and Cartwright [1983], the effects of water deficit in soil in the stage of ear formation, are often irreversible and may be compensated in later growth stages. Water stress may also lead to flower death in the ear in the tillering stage [Morgan, 1980].

Effects of the interactions of mixture components are expressed in the modification of the aboveground plant parts and roots. From our studies it follows that cultivation with papilionaceous plants did not affect significantly organ length of the underground parts of barley at the beginning of the growing season. In the studies of Michalska *et al.* [2008], proximity of pea stimulated root length in barley in the initial growth stages. On the other hand, Kostrzewska *et al.* [2013] in the analogical stage, found that barley cultivated with Italian ryegrass, developed significantly shorter roots than in pure sowing. Diverse levels of substrate moisture also had no effect on the discussed trait in barley, which is in line with the results of the above-mentioned studies of Kostrzewska *et al.* [2013].

## CONCLUSIONS

1. Cultivating spring barley with red clover had no effect on morphological traits of barley throughout the whole growing season.
2. The effects of reduced substrate moisture revealed in barley from the tillering stage in the form of a reduction in the shoot length (until the end of growing), in the leaf number (shooting stage) and in reduced tillering. After earing, grain number per ear decreased, as well as the ear length in the final development stage.

3. Overlapping of stresses caused by the competitive effect of red clover and water deficit, resulted in a reduction in the number of productive shoots in barley.

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## **WŁYWA ODDZIAŁYWAŃ KONKURENCYJNYCH I STRESU WODNEGO NA CECHY MORFOLOGICZNE JĘCZMIENIA JAREGO UPRAWIANEGO Z KONICZYNĄ CZELOWONĄ**

**Streszczenie.** Na podstawie 3-letniego doświadczenia wazonowego realizowanego według schematu addytywnego badano wpływ oddziaływań konkurencyjnych ze strony koniczyny czerwonej oraz deficytu wody na cechy morfologiczne jęczmienia jarego (długość korzenia, pędu i kłosa, krzewienie ogólne i produkcyjne, liczba liści i liczba ziaren w kłosie). Czynnikami doświadczenia były poziom nawadniania podłoża oraz sposób siewu. Zastosowano poziom nawadniania wyższy, odpowiadający zapotrzebowaniu roślin, i obniżony o 50%. Rośliny wysiewano w siewie jednogatunkowym (18 roślin jęczmienia jarego na wazon) i mieszanym (18 roślin jęczmienia jarego z 8 roślinami koniczyny czerwonej na wazon). Pomiarów biometrycznych dokonywano w pięciu fazach rozwojowych wyznaczonych przez rytm wzrostu jęczmienia w siewie czystym z wyższą dawką wody, tj. w fazach (skala BBCH): rozwoju liści (10-13), krzewienia (22-25), strzelania w źdźbło (33-37), kłoszenia (52-55) i dojrzewania (87-91). Wykazano, że wspólna uprawa jęczmienia jarego z koniczyną czerwoną pozostawała bez wpływu na analizowane cechy morfologiczne tego zboża przez cały okres wegetacji. Z kolei deficyt wody w podłożu skutkował ograniczeniem wzrostu zboża w okresie krzewienie –

dojrzewanie, wykształceniem mniejszej liczby liści (krzewienie – strzelanie w żdżbło) oraz słabszym krzewieniem ogólnym i produkcyjnym. Stres wodny spowodował również wytworzenie krótszych kłosów z mniejszą liczbą zawiązanych w nich ziaren. Konkurencyjne oddziaływanie koniczyny czerwonej spotęgowane deficytem wody skutkowało redukcją liczby pędów produkcyjnych jęczmienia. Nie stwierdzono wpływu czynników doświadczenia na długości korzeni w początkowych fazach wzrostu (rozwój liści – krzewienie).

**Slowa kluczowe:** deficit wody, fazy rozwojowe, *Hordeum vulgare*, oddziaływanie między roślinami

Accepted for print – Zaakceptowano do druku: 15.01.2015

For citation – Do cytowania:

Treder, K., Jastrzębska, M., Kostrzewska, M.K., Makowski, P., Wanic, M. (2014). Effect of competition and water stress on morphological traits of spring barley grown with red clover. *Acta Sci. Pol. Agricultura*, 14(1), 75-84.