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INFLUENCE OF WATER EXTRACTS FROM CORNFLOWER ON GERMINATION AND GROWTH OF CEREALS SEEDLINGS

Summary

The aim of research was to determine the effect of different concentrations of water extracts of cornflower on germination capacity and early growth of winter wheat (Triticum aestivum L) and rye (Secale cereale L.) seedlings. The experiment was conducted in laboratory conditions with the use of modified test of germination and early growth of seedlings-PHYTOTOXKIT^{TM}. Investigation was carried out in 3 independent experimental series, in three replications for each of them. Experimental series took place in two-week intervals. In the experiments there was evaluated germination capacity of cereals grains on the basis of the quantity of normally germinating seeds after five days from their sowing. Additionally there was measured the length and weight of roots and shoots. Water extracts from Centaurea cyanus fresh leaves did significantly inhibit roots growth of winter wheat and rye, irrespective of extract concentration. However, hypocotyls proved to be more tolerant to the examined extracts. Inhibitory effect of cornflower extracts on length and weight of shoots was observed only after applying the highest concentration. Research results reported in this work prove that water extracts from C. cyanus fresh leaves can have a negative, allelopathic effect on germination of T. aestivum and S. cereale.
***Key words**: water extract, cornflower, winter wheat, rye, germination*

WPŁYW WODNYCH WYCIĄGÓW Z CHABRA BŁAWATKA NA KIEŁKOWANIE ORAZ WZROST SIEWEK ZBÓŻ

Streszczenie

Celem badań było określenie wpływu różnych stężeń wodnych wyciągów z chabra bławatka (C. cyanus L.) na zdolność kiełkowania i początkowy wzrost roślin pszenicy ozimej T. aestivum L. i żyta Secale cereale L. Doświadczenie przeprowadzono w warunkach laboratoryjnych z wykorzystaniem zmodyfikowanego testu kiełkowania i wczesnego wzrostu roślin - PHYTOTOXKIT^{TM}. Badania wykonano w 3 niezależnych seriach, po 3 powtórzenia w serii. Serie wykonano w 2 tygodniowych odstępach. W doświadczeniu oceniano zdolność kiełkowania ziarniaków zbóż na podstawie ilości normalnie kiełkujących nasion po upływie 5 dni od momentu wysiania ziarniaków. Dodatkowo wykonano pomiary długości korzeni zarodkowych oraz pędów. Następnie izolowano korzenie i pędy od ziarniaków i określano ich świeżą masę. Wodne wyciągi ze świeżych liści chabra istotnie hamowały wzrost korzeni zarówno pszenicy ozimej jak i żyta, niezależnie od zastosowanego stężenia ekstraktu. Pędy natomiast wykazały większą tolerancję na badane ekstrakty. Inhibicyjny wpływ wyciągów z chabra na ich długość i masę był widoczny dopiero po zastosowaniu najwyższego stężenia. Wyniki prezentowane w pracy dowodzą, że wodne wyciągi ze świeżych liści C. cyanus mogą wykazywać ujemne oddziaływanie allelopatyczne na kiełkowanie T. aestivum i S. cereale.
***Słowa kluczowe:** wodne wyciągi, chaber bławatek, pszenica ozima, żyto, kiełkowanie*

1. Introduction

Secretion to the environment of biologically active compounds (allelochemicals) is one of the most important mechanisms in the world of plants. They can influence other plant species through chemical compounds, which through inhibitory or stimulatory effect, modify the process of seed germination, as well as plant growth and development [11, 14]. Leaves, then roots are the organs of plants richest in allelochemicals, while the lowest content of allelochemicals characterises seeds. Biological activity of these substances is governed, first of all, by phenolic compounds, alkaloids and volatile substances [2, 5]. The phenomenon of alleopathy is vital as far as agriculture is concerned [1, 7]. Research, conducted in a number of research facilities all over the world, confirmed that crops and weeds introduce chemical compounds to the environment, which can be toxic both for themselves and for other species [18, 21]. It has been proved that the influence of weeds on crops is connected not only with competition involving light, water and mineral components, but also with the fact that weeds secrete substances able to inhibit crops growth and development [3].

Cornflower (*Centaurea cyanus* L.) is weed species densely infesting cereals crops in Poland. It occurs on friable soils, rich in nutrients. This plant is extremely competitive species for cereals. One plant produces even up to 1600 seeds which retain their germination capacity for 5–10 years. Besides, achenes contained in the soil in neighbourhood of cereals grains, germinate at the same time, thus competing from the very early stages of plant development for living space.

C. cyanus, apart from being a common weed in cereals, serves as raw material in cosmetic industry and it is a medicinal plant used in natural medicine [8]. Chemical composition of cornflower flowers includes different com-

pounds like: flavonoids, tannins, mineral salts, organic acids, as well as anthocyanins, providing flowers with characteristic sapphire color [4]. Research carried out by Sarker et al. [16] and Shoeb at al. [17] proved that cornflower seeds are the source of numerous alkaloids.

Allelopathic substances contained in cornflower plants can be released through their leaching from above – ground parts during rainfalls or during weed biomass decomposition after ploughing. Therefore, it is important to know if allelochemicals released from *C. cyanus* plants can affect crops and if their effect is of a stimulatory or inhibitory character.

The aim of was to determine the effect of different concentrations of water extracts of cornflower on germination capacity and early growth of winter wheat (*T. aestivum* L). and rye (*Secale cereale* L.) seedlings.

2. Material and methods

The experiment was conducted in laboratory conditions, in Department of Weed Science and Tillage Systems in Wroclaw, with the use of modified test of germination and early growth of seedlings-PHYTOTOXKIT^{TM} [15]. Investigation was carried out in 3 independent experimental series, in three replications for each of them. Experimental series took place in two-week intervals.

C. cyanus L. seeds were collected from the fields located in the region of Wroclaw. They were sown to plastic pots containing 2:1 mixture of peat and sand. Seedlings were culture in greenhouse conditions, at the temperature of 20-22°C and air humidity ranging of about 70%. In four - leaf stage (BBCH 14) the plants were cut in order to obtain the material for water extracts.

Out of the material prepared in that way there were weighed 5 , 12 and 25 g of fresh matter. Then the material was quenched with 100 ml of distilled water and mixed. The solution was left for 24 hours without access to light, at room temperature. After that time the extract was centrifuged and obtained solutions were applied in the experiment.

Tests plates were filled with calcined sand of a diameter 0,6-0,8 mm. The examined objects involved plates soaked with 25 ml: 5%, 12% and 25% water extract of *C. cyanus*. On control object sand was soaked with 25 25 ml of distilled water. Then the plates were covered with paper filter and test seeds of *T. aestivum* (of Bogatka cultivar) and *S. cereale* (of Walet cultivar), in the amount of 5 units./per one plate, were sown. Microbiotests, prepared according to the procedure mentioned above, were incubated in a vertical position for 5 days, at the temperature of 25°C and without access to light. After that period test plates image were recorded with the use of a digital camera. Details regarding test procedure were described in Standard Operating Procedure [15].

In the experiments there was evaluated germination capacity of cereals grains on the basis of the quantity of normally germinating seeds after five days from their sowing.

Additionally, the length of roots and shoots was measured using the program of image analysis "Image Tools". Then roots and shoots were isolated from grains to determine their fresh matter.

Result tables contain values from 3 replications and from 3 experimental series. Research results were subject to statistical analysis, applying ARM8 program and differences between means under different treatments were calculated by Least Significant Difference (*LSD*) test at 5% probability. The results regarding germination capacity were transformed with the use of arc sin x function.

3. Results

The examined concentrations of extracts from cornflower leaves had a diverse impact on early growth of winter wheat. Water extracts of 5% and 12% concentrations were characterised by a stimulatory effect on germination capacity of *T. aestivum* grains causing its increase by 8 and 12% respectively, as compared to control object treated with distilled water. The highest concentration (25%) brought about an inhibitory effect on germination capacity of grains (considerably decreasing it by 38% in comparison to control data). The examined extracts from cornflower plants significantly reduced the length of roots in relation to control object. After application of extract concentrations of 5% and 12% the length of wheat roots ranged from 60 to70 mm, while in control object it amounted to about 83 mm. The shortest roots (30 mm) were recorded after introduction of the highest extract concentration (25%). Water extracts from *C. cyanus* leaves, applied in concentrations of 5% and 12% did not result in any significant differences in roots weight, nor in the length and weight of shoots. Only the highest extract concentration (25%) caused significant decrease in values of the examined parameters, as compared to control object (Table 1).

Water extracts from cornflower leaves of 5 and 12% concentration did not influence germination capacity of rye. The examined extract of the highest concentration caused inhibitory effect on germination capacity of tested cereals grains, decreasing it even by about 40% in comparison to control object.

Table 1. Effect of different concentrations of water extracts from leaf of cornflower on *T. aestivum* growth
Tab. 1. Wpływ różnych stężeń wodnych wyciągów z liści chabra bławatka na wzrost T. aestivum

Extract concentration (%)	Germination capacity (%)	Lenght of root (mm)	Weight of root (g)	Lenght of shootl (mm)	Weight of shoot (g)
00	81a	82.6a	0.0396a	46.1a	0.0545a
05	96a	69.1b	0.0414a	45.0a	0.0522a
12	91a	61.9b	0.0346a	44.3a	0.0536a
25	50b	30.5c	0.0183b	22.3b	0.0256b
LSD (0.05)	17.36t	10.47	0.00589	9.15	0.00871
SD	8.69t	5.26	0.00295	4.58	0.00436
CV	13.42t	8.63	8.81	11.61	9.37

t – Mean descriptions are reported in transformed data units, and are not de-transformed; LSD – Least Significant Difference; SD – Standard Deviation; CV – Coefficient of variation

Source: own work / Źródło: opracowanie własne

Table 2. Effect of different concentrations of water extracts from leaf of cornflower on *S. cereale* growth
Tab. 2. Wpływ różnych stężeń wodnych wyciągów z liści chabra bławatka na wzrost S. cereale

Extract concentration (%)	Germination capacity (%)	Lenght of root (mm)	Weight of root (g)	Lenght of shoot (mm)	Weight of shoot (g)
00	99a	74.9a	0.0443a	43.5a	0.0393a
05	100a	63.5b	0.0384a	41.1a	0.0414a
12	93a	46.1c	0.0299b	40.2a	0.0398a
25	57b	28.8d	0.0182c	29.5b	0.0273b
LSD (0.05)	13.87t	8.01	0.00770	4.01	0.00696
SD	6.94t	4.01	0.00386	2.01	0.00349
CV	2.79t	7.52	11.80	5.20	9.42

t – Mean descriptions are reported in transformed data units, and are not de-transformed; LSD – Least Significant Difference; SD – Standard Deviation; CV – Coefficient of variation

Source: own work / Źródło: opracowanie własne

Tested concentrations of extracts from cornflower leaves significantly differentiated the length of rye roots. After application of water extract of the lowest concentration (5%), shortening of roots to 63 mm was observed. The value for control object was 75 mm. The most considerable shortening of roots was recorded for application of 25% extract concentration (29 mm). Statistical analysis of roots weight did not show any significant differences in the values of that parameter between control object (0.0443 g) and the lowest (5%) extract concentration (0.0384 g). The increase in extract concentration resulted in significant decrease in rye roots weight. After application of 25% extract concentration roots weight amounted to 0.0182 g. The investigated extracts of 5% and 12% concentration did not significantly diversify the length and weight of shoots. Only application of the highest concentration (25%) caused significant inhibition of shoots growth (Table 2).

4. Discussion

Numerous research conducted all over the world proves that plants provide the environment with different kinds of allelopathic compounds, including phenols, alcaloids, fatty acids, terpenoids, flavonoids [2, 5]. They are able to influence other plants in a negative or a positive way [18, 21]. A number of allelopathic compounds are provided by leaves, roots, pollen, flowers, seeds and fruit [19, 20]. This is of a considerable importance from agricultural point of view. In the crop – weed system allelopathy is a phenomenon which can bring about changes in the composition of weed species, as well as disturbances in plants growth and yielding. Negative or positive effect of water extracts from weeds on crops depend on a weed species, concentration of water extracts and acceptor sensitivity [6, 9, 11]. Foreign literature lacks information about the effect of *C. cyanus* extracts on crops. In Poland this species often causes weed infestation of winter wheat and rye plantations. At the same time, it is a serious problem as this weed is resistant to some herbicide [12]. Cornflower plants, without weed control, can become a source of numerous allelochemicals introduced into the soil, can be of a negative or positive influence on crops. Therefore, it is essential to conduct research to determine allelopathic relations between cornflower and crops.

The results of conducted experiments indicate stimulatory and inhibitory effect of water extracts from *C. cyanus* leaves on germination and early growth of winter wheat and rye. There was observed a slight stimulatory influence of extracts (applied in 5% and 12% concentrations) on germination of *T. aestivum* grains. Increased concentration value of extract (to 25%) brought about significant inhibition of wheat germination capacity as compared to control object. The influence of extract of cornflower on winter wheat was more evident in the stage of early growth, which be proved by obtained results regarding the length and weight of roots and shoots. Observation involved inhibitory effect of the examined extract on the length of wheat roots, irrespective of extract concentration. The remaining parameters did not show such a strong differentiation under the influence of different extract concentrations. The weight of rootlets, the length and weight of shoots underwent significant inhibition only after application of 25% extract concentration in comparison to control object.

Stimulatory effect of extracts from plants on grain germination and the height of *T. aestivum* plants were observed by Marwat and Khan [13]. Research by these authors proved that water extract from fresh leaves of *Prosopis juliflora* increased germination capacity and the height of winter wheat. Inhibitory influence of water extracts on germination and growth of *T. aestivum* seedlings were reported by Ankita and Chabbi [2]. The authors examined the influence of extracts from leaves, stems and roots of 5 plant species: *Phalaris minor* L., *Chenopodium mulare* L., *Sonchus oleracea, Cyanodon dactylon* L., and *Convolvulus arvensis* L on grain germination capacity, as well as the length and dry matter weight of *T. aestivum* seedlings. The most considerable inhibitory effect on germination and early growth of winter wheat resulted from application of extracts from leaves and stems of *Phalaris minor* L. while smaller effect was obtained for roots of that plant. Other research indicated stimulatory (at lower concentrations) and inhibitory (at higher concentrations) influence of water extracts from *Chenopidium album* leaves on *T. aestivum* growth and yielding [11].

Research results presented in this work prove that the effect of extracts from fresh leaves of *C. cyanus* on germination and early growth of *S. cereale* was less considerable. The grain germination capacity was differentiated only by the highest extract concentration (25%), which was expressed by significant inhibition of germination capacity. Similar result was obtained when treating *S. cereale* grains with water extract from *Glicine max* shoots. Twenty five per cent concentration of the examined extract caused inhibitory influence on rye germination [10]. Different results were reported by Kwiecińska-Poppe et al. [9], who proved that water extracts from fresh matter of *Galium aparine* and *Matricaria maritima* subsp. *inodora* decreased germination capacity of rye grains even at 2% extract concentration.

More significant allelopathic effect of extracts from fresh leaves of *C. cyanus* was visible in early growth of rye roots. A considerable inhibition of roots length was observed, irrespective of applied extracts concentration and roots weight - after application of 12% and 25% extract concentration. Inhibiting effect of the examined extract on shoots growth became visible only after the use of the highest extract concentration. Significant shortening of rye roots was observed also after application of 25% water extract of soybean shoots [10]. Research carried out by Kwiecińska-Poppe et al. [9], involving the effect of water extract from *Galium aparine* fresh matter on the length of the first leaf of rye, confirmed inhibiting effect of the examined extract (at its highest concentration 8%).

According to Khaldi et al. [7], plant roots were more sensitive to allelochemicals than shoots. Research results obtained by the authors and presented in this work allow to confirm the thesis.

5. Conclusions

1. Water extracts from *C. cyanus* fresh leaves significantly inhibited roots length of winter wheat and rye, irrespective of extract concentration.
2. Shoots proved to be more tolerant to the examined extracts.
3. Inhibitory effect of cornflower extracts on length and weight of shoots was observed only after application of the highest concentration.
4. The water extracts from *C. cyanus* fresh leaves can have a negative, allelopathic effect on germination of *T. aestivum* and *S. cereale*.

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