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FUNGICIDES AND BIOTECHNICAL AGENTS IN THE PROTECTION OF HEATHERS AGAINST *Pestalotiopsis sydowiana* AND THEIR INFLUENCE ON PLANT GROWTH

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ABSTRACT

In the protection of heathers against shoot dieback caused by *Pestalotiopsis sydowiana*, the highest percentage effectiveness of 92.6% to 100% was found on plants sprayed 12 times every 7 days with the active substances: iprodione, pyraclostrobin + boscalid and trifloxystrobin and with the biotechnical agents containing: microcrystalline chitosan and extract from grapefruit seed and pulp, with an efficacy of 76.9% to 100%. The active substances like chlorothalonil, fluopyram + trifloxystrobin, iprodione, pyraclostrobin + boscalid, as well as microcrystalline chitosan and extract from grapefruit seed and pulp also stimulated plant growth. Heather plants sprayed with these agents and also those sprayed with cyprodinil + fludioxonil, trifloxystrobin and potassium carbonate + monopotassium phosphate were found to produce the highest number of new shoots. An increase in the fresh weight of the aboveground parts of the heather plants of more than 55.7% compared with the control plants was found after spraying the plants with the active substances such as: fluopyram + trifloxystrobin, iprodione, pyraclostrobin + boscalid, cyprodinil + fludioxonil, trifloxystrobin and with the biotechnological agents containing microcrystalline chitosan extract from grapefruit seed and pulp, and potassium carbonate + monopotassium phosphate. Similarly, the highest average dry weight of the aboveground parts was found after the application of these agents.

Key words: heather, fungicides, biotechnical agents, protection, *Pestalotiopsis sydowiana*

INTRODUCTION

Heathers are very often used as ornamental plants. In floristry, heathers are used to make wreaths and dried winter bouquets. Various varieties are often used to decorate urban green areas, allotments, home gardens, or they are planted in containers on balconies and terraces, and to decorate graves.

In recent years, producers of nursery-grown heather plants have increasingly more often reported to the National Institute of Horticultural Research in Skierniewice symptoms of shoot dieback and root and stem-base rot. *Pestalotiopsis sydowiana*, previously described in the literature as the cause of the disease, has most often been isolated from the diseased tissues [Hopkins 1996, McQuilken and Hopkins 2004, Ramlein-Starosta 2004]. Literature data indicate that *Pestalotiopsis* isolates obtained from the foliage, stem-base and roots of diseased container-grown ericaceous crops (*Calluna*, Erica, Pieris and *Rhododendron*) collected from UK nurseries were identified as *Pestalotiopsis sydowiana*

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(Bresad) B.C. Sutton on the basis of conidia morphology. Inoculum sources of the pathogen included diseased stock plants, crop debris, nursery soils, used growing media, pots and floor covering and dust collected from greenhouse walkways. Isolates were not host-specific and infected other species of ericaceous plants, with typical symptoms including browning of foliage, stems and roots and the presence of black or greenish-black acervuli on diseased tissue [McQuilken and Hopkins 2004]. When the disease symptoms are severe, nursery-grown heathers lose their decorative value and are often not suitable for trade. Scientific literature lacks information on the effectiveness of fungicides or biotechnical agents in the protection of heathers against *P.sydowiana*, or the data are very limited and do not apply to the fungicides and biotechnical agents that have appeared on the market in recent years. Modern environmentally sound plant disease management constantly requires new low-toxic, anti-polluting antifungal agents that differ from the fungicides currently developed in their mode of action and chemical properties [Kim and Hwang 2003]. Due to the minor importance of heather production in the overall agricultural output, the interest of chemical companies in producing new fungicides for this species is low. The result is a limited number of approved active substances of fungicides for the protection of heathers against foliar pathogens, thus creating the risk of too frequent use of agents with the same mode of action and the development of pathogen resistance to the active substances used. The existing literature data on the effectiveness of fungicides in the protection of various plant species against fungi of the genus *Pestalotiopsis* indicate the possibility of using for this purpose agents such as carbendazim, chlorothalonil, mancozeb, prochloraz and iprodione in the protection of heathers [Hopkins 1996, McQuilken and Hopkins 2004], chitosan in the protection of guava [El-Argawy 2015], copper oxychloride, mancozeb, methyl thiophanate in the protection of som [Ray et al. 2016] and thiram in the protection of strawberry [Essa et al. 2018]. The research on the protection of various species of ornamental plants against foliar pathogens, conducted for more than a dozen years, has shown the possibility of using not only fungicides but also growth stimulants, foliar fertilizers, and plant extracts that can limit the development of disease symptoms [Jeliazkova et al. 2012, Salamone et al. 2009, Wojdyła 2016].

The aim of the study was to determine the effectiveness of active substances included in 13 fungicides with different modes of action and substances of 4 biotechnical agents in limiting the development of heather shoot dieback (*Pestalotiopsis sydowiana* (Bresadole) B.C. Sutton).

MATERIAL AND METHODS

The following active substances (fungicides) were used in the study: 250 g l^{-1} azoxystrobin (Amistar 250 SC), 50% kresoxim methyl (Discus 500 WG), 100 g l^{-1} tetraconazole (Domark 100 EC) 500 g l^{-1} chlorothalonil (Gwarant 500 SC), 250 g l^{-1} tebuconazole (Horizon 250 EW), 250 g l^{-1} fluopyram + 250 g l^{-1} trifloxystrobin (Luna Sensation 500 SC), 50% tebuconazole + 25% trifloxystrobin (Nativo 75 WG), 500 g l^{-1} iprodione (Rovral Aquaflo 500 SC), 250 g l^{-1} difenoconazole (Score 250 EC), 6.7% pyraclostrobin + 26.7% boscalid (Signum 33 WG), 37.5% cyprodinil + 25% fludioxonil (Switch 62.5 WG), 100 g l^{-1} penconazole (Topas 100 EC), 50% trifloxystrobin (Zato 50 WG) and also substances included in biotechnical products: ammonium phosphite + microelements B, Cu, Fe, Mn, Mo, Zn (Actifos), 20 g l^{-1} microcrystalline chitosan (BetaChikol), 33% extract from grapefruit seed and pulp (Biosept Active), 48% potassium carbonate + 48% monopotassium phosphate (Solfan PK) for spraying heather plants grown in pots placed on windowsills in a greenhouse.

In the laboratory, the isolates P17, P34 and P45 of *P. sydowiana* fungus, isolated earlier from diseased heather shoots, were inoculated onto a potato-dextrose medium (PDA, Merck) in 90 mm diameter Petri dishes and then kept in a thermostat at 24°C. After 3 weeks, the spores were scraped with a scalpel into water to prepare a suspension (1×10^5 spores in 1 ml). The experiments were carried out in 2019 on heather plants of the cultivar Allegro susceptible to dieback, planted in 1 dm^3 containers placed on windowsills in the greenhouse. The plants were first sprayed with the spore suspension. After this treatment, the windowsills were covered with polyethylene film to ensure 100% air humidity. After 1 week, the plants were successively sprayed 12 times every 7 days with the test agents

at the concentrations given in Tables 1 and 2. The working liquid was prepared by mixing the test agents with water at a temperature of about 20°C and pH 7. The control plants were sprayed with water, whereas the remaining plants were sprayed with the test agents at various concentrations, applying 100 ml of working liquid per 1 m^2. The plants were sprayed in the morning (8–9 a.m.) with an Apor pneumatic laboratory sprayer with a tank capacity of 1.5 dm^3 and a liquid pressure of 0.2 MPa, adapted to spraying such plot areas. During the treatment, the spray nozzle was held at a height of 30 cm above the plants and so operated as to cover them thoroughly with the liquid. During the experiments, the air humidity in the greenhouse was maintained at about 70% and the temperature ranged from 20 to 25°C. The plants were watered by directing a stream of water directly onto the substrate in the pots or onto the capillary mat on which the containers were positioned. Three days after the 4th, 8th and 12th spray treatment, observations of the severity of disease symptoms were made according to a 6-point scale, where: 0 – no symptoms, 1 – up to 5% of shoot surface was necrotic, 2 – from 5% to 10%, 3 – from 10% to 20%, 4 – from 20% to 50%, 5 – more than 50% of shoot surface was necrotic. Observations of the possible phytotoxicity of the tested agents or their mixtures were carried out 3 days after each spraying, according to an 8-point scale: 0 – 0% damaged or deformed leaf and shoot surface, 1 – 0.1% to 1% damaged leaf and shoot surface, 2 – 1.1% to 6%, 3 – 6.1% to 15%, 4 – 15.1% to 30%, 5 – 30.1% to 50%, 6 – 50.1% to 80%, 7 – more than 80% damaged or deformed surface of leaves and shoots. In addition, observations were made for signs of yellowing or stunted growth.

Influence of the applied agents on heather growth, number of shoots per plant and plant diameter. In the first experiment, the height of the plants, the number of shoots per plant and the diameter of the rosette of the aboveground part were measured after 8 and 12 treatments.

Influence of the applied agents on fresh and dry weight of aboveground parts and roots. After completion of the first experiment, the aboveground part of the plant was separated from the underground part with a scalpel and weighed to determine the fresh weight for all the plants in each replication. Dividing this by the number of plants in the replication, the average fresh weight of a single plant was obtained. Then, the aboveground parts from individual replications were placed in envelopes and kept at 70°C for 24 hours in forced-air incubators. After removal from the incubator, the plant material was re-weighed with and without the envelopes. The final dry weight of the aboveground parts was divided by the number of plants in the replication (5) to obtain the results on a per plant basis. A similar procedure was applied to the underground part, where, after removing the root system, the substrate was first mechanically removed and then the remnants of it were shaken off in water. Before weighing, the root system was dried on filter paper.

Statistical analysis. The experiment was set up in a completely random block design in 4 replications, with 5 plants each. The results were statistically analyzed by a one-way ANOVA. The significance of differences between means was assessed using Duncana multiple range test at $p \leq 0.05$. The percentage reduction in the extent of shoot dieback symptoms was calculated in relation to the control (unprotected plants), using the simplified Abbott's formula [Abbott 1925].

RESULTS

In the first experiment, after the heather plants had been sprayed 4 times, the recorded degree of infection of the control plants was 1.2 (Tab. 1). No disease symptoms were found on the plants sprayed with: iprodione, pyraclostrobin + boscalid, and trifloxystrobin. The remaining active substances showed an efficacy of 66.7% (azoxystrobin) to 95.8% (cyprodinil + fludioxonil). In the case of the substances included in biotechnical agents, the percentage effectiveness ranged from 16.7% (potassium carbonate + monopotassium phosphate) to 87.5% (ammonium phosphite + microelements, microcrystalline chitosan). After spraying the heather plants 8 times, the degree of infection of the control plants was 2.0 (Tab. 1). Among the tested, active substances the highest efficacy, from 92.5% to 97.5%, was shown for fluopyram + trifloxystrobin, iprodione, and trifloxystrobin. Among the biotechnical agents, the highest effectiveness was shown for microcrystalline chitosan and grapefruit extract. After 12 treatments, the degree of infection of the control plants was 2.7 (Tab. 1). Of the fungicides tested, the highest efficacy, from 87% to 98.6%, was found for the active

Table 1. Percentage effectiveness of fungicides used to protect 'Allegro' heather plants against *Pestalotiopsis sydowiana* (Experiment 1)

Treatment	Conc. [%]	Percentage effectiveness after weeks		
		4	8	12
Control – healthy	–	100a	100a	100a
Control – infected	–	0.0g	0.0h	0.00i
fungicides				
Amistar 250 SC – azoxystrobin	0.1	66.7e	75.0d–f	72.2fg
Discus 500 WG – kresoxim methyl	0.03	87.5a–c	80.0de	79.6d–f
Domark 100 EC – tetraconazole	0.05	79.2c–e	70.0f	72.2fg
Gwarant 500 SC – chlorotalonil	0.2	83.3b–d	82.5cd	87.0b–e
Horizon 250 EW – tebuconazole	0.075	79.2c–e	67.5f	62.2gh
Luna Sensation 500 SC – fluopyram + trifloxystrobin	0.08	91.7a–c	92.5ab	79.6d–f
Nativo 75 WG – tebuconazole + trifloxystrobin	0.3	87.5a–c	72.5ef	75.9ef
Rovral Aquaflo 500 SC – iprodione	0.2	100a	97.5ab	98.1ab
Score 250 EC – difenoconazole	0.05	91.7a–c	82.5cd	85.2c–e
Signum 33 WG – pyraclostrobin + boscalid	0.15	100a	90.0bc	92.6a–c
Switch 62,5 WG – cyprodinil + fludipxonil	0.1	95.8ab	90.0bc	87.0b–e
Topas 100 EC – penconazole	0.05	79.2c–e	72.5ef	70.4fg
Zato 50 WG – trifloxystrobin	0.015	100a	95.0ab	98.6a–c
biotechnical agents				
Actifos – phosphite + microelements	0.6	87.5a–c	50.0g	53.7h
BetaChikol – microcrystalline chitosan	1	87.5a–c	90.0bc	87.0b–e
Biosept Active – grapefruit extract	0.1	70.8de	82.5cd	87.0b–e
Solfan PK – potassium carbonate + monopotassium phosphate	0.5	16.7f	50.0g	62.9gh

Means in columns, marked with the same letter, do not differ significantly (5%) according to Duncan's test

substances: iprodione, pyraclostrobin + boscalid and trifloxystrobin. Among the tested biotechnical agents, microcrystalline chitosan and grapefruit extract showed the highest effectiveness of 87%.

In the second experiment, after spraying the heather plants 4 times, the recorded degree of infection of the control plants was 0.75 (Tab. 2). There were no disease symptoms on the plants sprayed with: fluopyram + trifloxystrobin, iprodione, pyraclostrobin + boscalid, penconazole, trifloxystrobin and the biotechnical agent – microcrystalline chitosan. After 8 treatments, the degree of infection of the control plants was 1.2 (Tab. 2). As was the case after 4 sprayings, the same active substances contained in fungicides and the same substance included in biotechnical agent completely protected the heather plants against infection by the pathogen after the treatments had been carried out 8 times. After 12 treatments, the degree of infection of the control plants was 1.3 (Tab. 2). No disease symptoms were found on the heather plants protected with the active substances such as: iprodione, pyraclostrobin + boscalid, trifloxystrobin and the biotechnical substances microcrystalline chitosan. No phytotoxicity symptoms were found on the heather plants after using the agents tested.

Influence of the applied agents on heather growth, number of shoots per plant and plant diameter. After applying the fungicides 8 times, the height of the control plants was 66.45 mm (Tab. 3). Significantly taller plants were found after spraying with the fungicides contained: chlorothalonil, iprodion and pyraclostrobin + boscalid and with the microcrystalline chitosan – plant growth stimulant. Significantly shorter plants were found after using the tebuconazole and the fertilizer based on ammonium phosphite + microelements. After 12 applications of the agents, the height of the control plants was 72.45 mm (Tab. 3). Significantly taller plants were found when sprayed with the chlorothalonil, fluopyram + trifloxystrobin and pyraclostrobin + boscalid. On the other hand, significantly lower plants were found after using the tebuconazole.

After 8 treatments with the agents, the infected plants were found to have an average of 11.3 shoots per plant (Tab. 3). Significantly more shoots were found on the plants sprayed with the biotechnical agents contained microcrystalline chitosan and grapefruit extract. On the other hand, the lowest number of shoots, from 6.6 to 7.45, was counted on the plants sprayed with tebuconazole and the fertilizer contained ammonium phosphite + microelements. After 12 applications of the agents, 12 shoots on average were found on the infected plants (Tab. 3). Significantly more shoots per plant, ranging from 17.9 to 33.25, were found after the application of: chlorothalonil, difenoconazole, fluopyram + trifloxystrobin, iprodione, pyraclostrobin + boscalid, cyprodinil + fludioxonil and trifloxystrobin, and after applying the biotechnical agents contained microcrystalline chitosan, grapefruit extract and potassium carbonate + monopotassium phosphate. On the other hand, the lowest number of shoots, from 8.1 to 8.35, was counted on the plants sprayed with the fungicide included tebuconazole and the fertilizer based on ammonium phosphite + microelements.

After applying the agents 8 times, the diameter of the aboveground part of the infected plants was 49.85 mm (Tab. 3). The largest diameter of the leaf rosette, above 58 mm, was found on the heather plants sprayed with chlorothalonil and pyraclostrobin + boscalid, and with the microcrystalline chitosan and grapefruit extract. After 12 applications of the agents, the diameter of the aboveground part of the infected plants was 51.6 mm (Tab. 3). Significantly the largest diameter of the leaf rosette, above 71 mm, was found on the heather plants sprayed with the fungicide contained pyraclostrobin + boscalid. The smallest diameter of 42.08 mm was found after spraying the plants with ammonium phosphite + microelements.

Influence of the applied agents on fresh and dry weight of aboveground parts and roots. After applying the agents 12 times, the average weight of the aboveground part of the infected plants was 2.03 g (Tab. 4). Significantly higher fresh weight of aboveground parts of plants, ranging from 3.16 g to 4.66 g, was found after spraying the heather plants with the fungicides based on: fluopyram + trifloxystrobin, iprodione, pyraclostrobin + boscalid, cyprodinil + fludioxonil, trifloxystrobin and with the microcrystalline chitosan, grapefruit extract and potassium carbonate + monopotassium phosphate. The lowest fresh weight of the aboveground parts was found in the plants treated with the fertilizer contained ammonium phosphite + microelements. The average dry weight of the aboveground parts of the infectedplants was 0.68 g (Tab. 4).

Table 2. Percentage effectiveness of fungicides used to protect 'Allegro' heather plants against *Pestalotiopsis sydowiana* (Experiment 2)

Treatment	Conc. [%]	Percentage effectiveness after weeks		
		4	8	12
Control – healthy	–	100a	100a	100a
Control – infected	–	0.0h	0.0g	0.0g
fungicides				
Amistar 250 SC – azoxystrobin	0.1	66.7c–e	58.3de	57.7de
Discus 500 WG – kresoxim methyl	0.03	80.0a–d	70.8b–e	65.4c–e
Domark 100 EC – tetraconazole	0.05	73.3b–e	75.0b–d	66.9c–e
Gwarant 500 SC – chlorotalonil	0.2	33.3a–b	79.2bc	80.8bc
Horizon 250 EW – tebuconazole	0.075	33.3g	58.3de	53.8e
Luna Sensation 500 SC – fluopyram + trifloxystrobin	0.08	100a	100a	96.2ab
Nativo 75 WG– tebuconazole + trifloxystrobin	0.3	53.3e–g	54.2e	57.7de
Rovral Aquaflo 500 SC – iprodione	0.2	100a	100a	100a
Score 250 EC – difenoconazole	0.05	66.7c–e	62.5c–e	61.5c–e
Signum 33 WG – pyraclostrobin + boscalid	0.15	100a	100a	100a
Switch 62,5 WG – cyprodinil + fludipxonil	0.1	66.7c–e	75.0b–d	73.1c–e
Topas 100 EC – penconazole	0.05	100a	100a	96.2ab
Zato 50 WG – trifloxystrobin	0.015	100a	100a	100a
biotechnical agents				
Actifos – phosphite + microelements	0.6	60.0d–f	20.8f	23.1f
BetaChikol – microcrystalline chitosan	1	100a	100a	100a
Biosept Active – grapefruit extract	0.1	86.7a–c	83.3b	76.9cd
Solfan PK – potassium carbonate + monopotassium phosphate	0.5	86.7a–c	75.0b–d	57.7de

Means in columns, marked with the same letter, do not differ significantly (5%) according to Duncan's test

Table 3. Influence of the fungicides and biotechnological agents used to protect heather plants against *Pestalotiopsis sydowiana* on plant growth

Treatment	Conc. [%]	Plant height [mm]		Number of shoots [per plant]		Plant diameter [mm]	
		observation after weeks					
		8	12	8	12	8	12
Control – healthy	–	80.60fg	87.10fg	30.45g	36.05h	66.38h	83.13h
Control – infected	–	66.45b–e	72.45b–f	11.30a–d	12.00bc	49.85c–g	51.60a–f
fungicides							
Amistar 250 SC – azoxystrobin	0.1	68.25b–e	73.25b–f	9.80a–d	11.30a–c	50.75c–g	51.03a–e
Discus 500 WG – kresoxim methyl	0.03	71.85b–g	80.00c–g	10.10a–d	11.60a–c	53.45d–g	59.00a–g
Domark 100 EC – tetraconazole	0.05	61.40bc	65.20a–c	10.10a–d	10.80a–c	46.50a–f	48.03a–d
Gwarant 500 SC – chlorotalonil	0.2	82.75g	90.15gh	12.95b–d	17.90de	58.13gh	67.25e–h
Horizon 250 EW tebuconazole	0.075	49.95a	53.65a	6.60a	8.35ab	45.05a–d	45.18a–c
Luna Sensation 500 SC – fluopyram + trifloxystrobin	0.08	74.65d–g	90.25gh	13.90c–e	21.50ef	55.78fg	63.50d–g
Nativo 75 WG tebuconazole + trifloxystrobin	0.3	67.35b–e	73.20b–f	7.90ab	13.70c	44.88a–d	52.63a–f
Rovral Aquaflo 500 SC – iprodione	0.2	81.00fg	87.60fg	14.55de	23.35fg	54.75e–g	64.55d–g
Score 250 EC – difenoconazole	0.05	70.00b–f	81.35c–g	13.25b–e	19.35ef	51.75d–g	59.18a–g
Signum 33 WG – pyraclostrobin + boscalid	0.15	80.50fg	90.50gh	15.15de	26.35g	58.50gh	71.04gh
Switch 62,5 WG – cyprodinil + fludipxonil	0.1	72.70c–g	78.85c–g	11.00a–d	19.50ef	51.75d–g	60.63b–g
Topas 100 EC – penconazole	0.05	62.85b–d	68.20a–d	8.80a–c	14.40cd	41.50a–c	49.33a–d
Zato 50 WG – trifloxystrobin	0.015	76.40e–g	85.05e–g	13.50c–e	26.45g	53.13d–g	67.95e–h
biotechnical agents							
Actifos – phosphite + microelements	0.6	50.65a	57.95ab	7.45a	8.10ab	39.50a–b	42.08a
BetaChikol – microcrystalline chitosan	1	94.23h	104.60h	20.30f	33.25h	59.50gh	58.00a–g
Biosept Active – grapefruit extract	0.1	73.50d–g	83.35d–g	18.20ef	21.05ef	58.88gh	68.75f–h
Solfan PK – potassium carbonate + monopotassium phosphate	0.5	75.50e–g	82.10d–g	13.90c–e	19.00ef	54.13d–g	61.13c–g

Means in columns, marked with the same letter, do not differ significantly (5%) according to Duncan's test

Table 4. Influence of the fungicides and biotechnological agents used to protect heather plants against *Pestalotiopsis sydowiana* on fresh and dry weight of plant parts

Treatment	Conc. [%]	Fresh weight of aboveground parts [g]	Dry weight of aboveground parts [g]	Fresh weight of roots [g]	Dry weight of roots [g]
Control – healthy	–	4.48i	1.41g	0.69e–g	0.25e–g
Control – infected	–	2.03c–e	0.68ab	0.55d–f	0.21b–f
fungicides					
Amistar 250 SC – azoxystrobin	0.1	2.01c–e	0.78a–d	0.38a–d	0.16a–e
Discus 500 WG – kresoxim methyl	0.03	2.22de	0.74a–c	0.42a–d	0.15a–d
Domark 100 EC – tetraconazole	0.05	1.77b–d	0.74a–c	0.26a–c	0.12ab
Gwarant 500 SC – chlorotalonil	0.2	2.39d–f	0.97d–f	0.39a–d	0.16a–e
Horizon 250 EW – tebuconazole	0.075	1.85c–e	0.76a–c	0.48b–e	0.19b–f
Luna Sensation 500 SC) – fluopyram + trifloxystrobin	0.08	3.33gh	1.02ef	0.54d–f	0.21b–f
Nativo 75 WG – tebuconazole + trifloxystrobin	0.3	2.54d–g	0.88b–e	0.69e–g	0.24c–f
Rovral Aquaflo 500 SC – iprodione	0.2	3.45h	1.09ef	0.36a–d	0.16a–e
Score 250 EC – difenoconazole	0.05	2.76e–h	0.92c–e	0.49c–e	0.21b–f
Signum 33 WG – pyraclostrobin + boscalid	0.15	3.16f–h	0.99d–f	0.36a–d	0.15a–d
Switch 62,5 WG cyprodinil + fludioxonil	0.1	3.38gh	1.08ef	1.18j	0.33g
Topas 100 EC – penconazole	0.05	2.41d–f	0.69ab	0.39a–d	0.14a–c
Zato 50 WG – trifloxystrobin	0.015	3.25f–h	1.03ef	0.78f–h	0.26d–g
biotechnical agents					
Actifos – phosphite + microelements	0.6	1.00ab	0.67ab	0.21ab	0.08a
BetaChikol – microcrystalline chitosan	1	4.66i	1.36g	0.97h–j	0.28fg
Biosept Active – grapefruit extract	0.1	3.23f–h	1.01ef	1.04ij	0.25d–g
Solfan PK – potassium carbonate + monopotassium phosphate	0.5	3.47h	1.14f	0.92g–i	0.27fg

Means in columns, marked with the same letter, do not differ significantly (5%) according to Duncan's test

Significantly higher dry weight of aboveground parts of plants, ranging from 0.92 g to 1.36 g, was found after spraying the heather plants with chlorothalonil, fluopyram + trifloxystrobin, iprodione, difenoconazole, pyraclostrobin + boscalid, cyprodinil + fludioxonil and trifloxystrobin and with the biotechnical agents containing microcrystalline chitosan, grapefruit extract, and potassium carbonate + monopotassium phosphate.

After 12 treatments with the agents, the average fresh weight of the root system of the infected plants was 0.55 g (Tab. 4). Statistically, significantly higher fresh weight of roots, ranging from 0.92 g to 1.18 g, was found after spraying the heather plants with cyprodinil + fludioxonil and microcrystalline chitosan, grapefruit extract and potassium carbonate + monopotassium phosphate. In contrast, the lowest fresh weight of roots, ranging from 0.21 g to 0.26 g, was recorded after spraying the heather plants with tetraconazole and the fertilizer contained ammonium phosphite + microelements. The average dry weight of the root system of the control plants was 0.21 g (Tab. 4). Significantly higher dry weight of the root system, 0.33 g, was recorded after spraying the heather plants with cyprodinil + fludioxonil, whereas the lowest dry weight of roots, amounting to 0.08 g, was found after spraying the heather plants with the fertilizer contained ammonium phosphite + microelements.

DISCUSSION

The European Union, aware of the dangers of excessive pesticide use, is implementing regulations that eliminate or restrict some chemicals. In addition to the positive aspect of protecting the environment and consumer health, withdrawing pesticides also has negative effects. The elimination of some chemical active substances used in plant protection may result in excessive use of commercially available products containing the same active substance. The lack of rotation of plant protection products with different modes of action can result in the resistance and spread of pests. It is therefore necessary to search for new substances in order to be able to produce horticultural and agricultural crops in an economically viable way.

In the present study, active substances contained in 13 fungicides and substances included in 4 biotechnical products were tested in the protection of heathers against *P. sydowiana*.

The high effectiveness of the fungicides used in the protection of heather plants is in agreement with the results of previous own research carried out on roses. In those studies, the active substances such as azoxystrobin, tetraconazole, tebuconazole, fluopyram + trifloxystrobin, tetraconazole + trifloxystrobin, difenoconazole, pyraclostrobin + boscalid, penconazole and trifloxystrobin used in rose protection showed high effectiveness in controlling powdery mildew, rust and black spot [Wojdyła 2018]. The obtained results regarding good effectiveness of chlorothalonil and iprodione used as a curative measure are in agreement with the experiments conducted on heathers [Hopkins 1996]. *In-vitro* studies, in turn, had shown high effectiveness of difenoconazole in inhibiting the growth of mycelium of *Pestalotia psydii*, the causal agent of dieback in guava (*Psydum guajava* L.) and in limiting the production of acervuli [Younis et al. 2004]. Similarly, in *in-vitro* studies on PDA (potato-dextrose agar), tiophanate-methyl (Topsin M) used at a concentration of 0.1% to 0.2% completely inhibited the growth of the mycelium of *Pestalotiopsis disseminata* that causes grey blight disease in the som plant (*Persea bombycina* Kost.) [Ray et al. 2016]. Earlier trials had shown that foliar applications of chlorothalonil or iprodione were effective in controlling this pathogen [Hopkins 1996, McQuilken et al. 1997]. Literature data explaining the mode of action of the tested fungicides from the group of strobilurins (azoxystrobin, kresoxim-methyl, pyraclostrobin, trifloxystrobin) indicate their inhibitory effect on mitochondrial respiration in fungal cells [Ammermann et al. 2000]. By comparison, triazoles (difenoconazole, penconazole, tebuconazole, tetraconazole) used in the experiments block the biosynthesis of sterols in fungal cells [Young et al. 2001]. In the case of chlorothalonil, its exact mode of action is unknown; however, it can be described as inactivation of glutathione-related enzymes involved in respiratory processes in fungal cells [Arvanites and Boerth 2001]. The mode of action of boscalid and fluopyram – SDHI fungicides (succinate dehydrogenase inhibitors) on pathogens is based on blocking the activity of succinic acid dehydrogenase [Wachowska et al. 2017]. The high effectiveness of chitosan in the protection of heathers against *P. sydowiana* shown in the present study also finds confirmation in the lite-

rature. El-Argawy [2015] demonstrated high effectiveness of 2.5% chitosan against *Pestalotiopsis* spp. causing scab on guava fruit (*Psidium guajava* L.). On the other hand, the biotechnical agents Actifos (phosphite + microelements), BetaChikol (microcrystalline chitosan), Biosept Active (grapefruit extract) and Solfan PK (potassium carbonate + monopotassium phosphate) proved to be highly useful in protecting roses against powdery mildew [Wojdyła 2016]. Literature data explaining their mode of action against pathogens have shown that phosphites in the fertilizer Actifos induce resistance in protected plants when applied at a lower concentration, and affect pathogens directly when used at a higher concentration [Smillie et al. 1989, Guest and Grant 1991]. Potassium carbonate, one of the components of the fertilizer Solfan PK, transforms into the bicarbonate form in an aqueous environment. Potassium bicarbonate and potassium phosphate (the second component of Solfan PK) have a direct effect on pathogens – they cause dehydration and severe deformation of the mycelium and spores [Reuveni et al. 1998, Wojdyła et al. 2010], while phosphates induce systemic resistance to bacteria, fungi and viruses [Mucharromah and Kuc 1991]. Chitosan, on the other hand, is not only an elicitor of plant resistance to pathogens, but can also actively inhibit their growth. Stössel and Leuba [1984] showed that chitosan can cause defects in the cell wall, vacuolization, and sometimes protoplasm decay. In plants, chitosan causes lignification of the cell wall, an increase in phytoalexin production, the synthesis of proteinase inhibitors, and stimulates the activity of hydrolytic enzymes (chitinase, chitosanase and ß-1,3-glucanase) [Benhamou and Nicole 1999]. Biosept Active contains endogenous flavonoids and glycosides as well as terpenes, coumarins and furanocoumarins, which show strong antifungal properties, inhibiting spore germination, the growth of infectious hyphae and mycelium development [Saniewska 2004].

The stimulation of plant growth and the formation of new shoots caused by the use of chitosan had been demonstrated by previous studies on carnation [Wojdyła and Orlikowski 1997]. On the other hand, the experiments have shown that the growth of heather plants is limited by fungicides containing triazoles. Triazole fungicides are known from the literature as plant growth-limiting agents [Wojdyła 2000]. The mode of action of triazole fungicides is to inhibit gibberellin biosynthesis, which leads to limitation of elongation growth of roots and shoots [Görtz et al. 2008]. The very diverse mode of direct and indirect action of the tested fungicides and biotechnical agents on pathogens may prove to be particularly important in controlling races of pathogens resistant to the fungicides used and find a wide application in integrated plant protection. Literature data indicate that the fungus *P. sydowiana* can be pathogenic for many plant species [McQuilken and Hopkins 2004]; therefore, the results of tests on heathers can also be used for other plant species.

CONCLUSIONS

1. Among the tested active substances, iprodione, pyraclostrobin + boscalid and trifloxystrobin showed the highest effectiveness in protecting heather.
2. Substances included in biotechnical products such as microcrystalline chitosan as well as extract from grapefruit seed and pulp, reduced the growth of *Pestalotiopsis* in about 80%.
3. Both biotechnical products and chemical active substances had beneficial effects on heather plant development.

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