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PATHOGENICITY OF *Fusarium oxysporum*, *Fusarium avenaceum* AND *Sclerotinia sclerotiorum* AND THEIR EFFECT ON PHOTOSYNTHETIC ACTIVITY OF CHRYSANTHEMUM PLANTS

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Abstract. The aims of the investigation were to identify the fungi causing spots, necrosis or death of plants and the evaluation of pathogenicity of some of them to chrysanthemum plants. During the mycological investigations the fragments of plant roots, stems and leaves were placed on a mineral medium. Over 6080 colonies were obtained. *Alternaria alternata*, *Fusarium* spp. and *Sclerotinia sclerotiorum* were isolated most frequently. The largest numbers of colonies and species of fungi were isolated from cv. 'Snowdon'. In the second experiment, the pathogenicity of *F. oxysporum*, *F. avenaceum* and *S. sclerotiorum* populations was investigated. Disease index for inoculated plants was compared to chlorophyll fluorescence parameters. For most of isolates of higher pathogenicity, the disease index was correlated with the reduction of plants photosynthetic activity. However, in some cases the damage to the photosystem was more severe than external disease symptoms indicated, suggesting that chlorophyll fluorescence measurements might be helpful in early evaluation of disease severity.

Key words: pathogenic fungi, cultivars susceptibility, chlorophyll fluorescence

INTRODUCTION

Chrysanthemum (*Chrysanthemum grandiflorum* Ramat./Kitam.) is known as a ornamental plant all over the world and one of the most important ornamental plants in Poland. Biotic and abiotic stresses are contributive to the difference, usually large, between potential and attainable crop yield [Boyer 1982]. Among these stresses, fungal diseases are critical for plants grown in the field and under cover [Sadras et al. 2000, Berger et al. 2004, Wagner 2004]. Practices such as the use of resistant cultivar, crop

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rotation, grafting, soil replacement, fumigation, adaptation of soilless culture have been suggested. The effectiveness of these management practices is curtailed by the monocropping, with relatively high profit crops in commercial production [Yu 2001, Patkowska 2014].

Fusarium oxysporum and *Sclerotinia sclerotiorum* are important soil pathogens of chrysanthemum grown under covers. *Fusarium avenaceum* seems to be also dangerous [Kopacki and Wagner 2006]. The health status of chrysanthemum is very important for producers. Even slight disease symptoms decrease the value of chrysanthemum as ornamental plant. The investigations of the susceptibility of cultivars are essential as the producers need to know which cultivars are less susceptible to pathogens.

The severity of fusariosis does not always manifest itself in the form of necrosis or wilting, especially at early stages of plant growth, but can affect the process of photosynthesis [Pospieszny and Struszczyk 2003]. Chlorophyll fluorescence is an indicator of photosynthetic performance of plants [Maxwell and Johnson 2002, Wagner et al. 2007, Sawicka and Michałek 2008]. The use of chlorophyll fluorescence to monitor photosynthetic performance in plants is now widespread [Baker 2008, Maràczi et al. 2011]. The method is useful in field detection of plant disease [Moshou et al. 2005], in the tests of susceptibility of plant cultivars to insects [Gantner and Michałek 2010] and of effects of pesticides on the photosynthetic apparatus [Dias et al. 2014]. This review shows how fluorescence parameters can be used to evaluate changes in photosystem II (PSII) photochemistry and the possibility of correlations between pathogenic abilities of tested isolates and the photosynthetic capacity of infested plants. Such investigations on chrysanthemum plants have not been yet conducted in Poland.

MATERIALS AND METHODS

Plant material. The investigations were conducted in 2004–2006 in Lublin region. The objects of study were three cultivars of chrysanthemum plants (*Chrysanthemum grandiflorum* Ramat./Kitam.): 'Casablanca', 'Royalys' and 'Snowdon'. Mineral fertilization was used in accordance to fertilization recommendations for chrysanthemum plants. The plants were planted in the pots in June and placed in foil tunnels. Pesticides were applied according to Zalecenia Ochrony Roślin.

During vegetation period (first decade of August) and flowering period (first decade of October) selected diseased chrysanthemum plants were sampled from foil tunnels.

Mycological analysis. The plants (leaves, stems and roots) were analysed in laboratory at the University of Life Sciences in Lublin. Plant material was precleaned, rinsed with tap water for 20 minutes and then disinfected with 50% ethyl alcohol and 0.1% sublimate for 1 minute. Disinfected plant material was rinsed 3 times in sterilized water. Next, 3 mm fragments of plants were placed on mineral medium in Petri dishes as described by Kopacki and Wagner [2006]. For each diseased plant, 30 dishes with plant material, (10 plant fragments of leaves, stems and roots per each dish) were prepared and incubated in the thermostat at 20–22°C for 7 days in darkness. The

obtained fungal colonies were transferred to potato dextrose medium (PDA, Difco) and identified according to the available monographs.

Analysis of pathogenicity and photosynthesis parameters. Laboratory tests of *F. oxysporum*, *F. avenaceum* and *S. sclerotiorum* isolates were carried out using the Mańka method described by Wagner [1997]. The cuttings of cv. 'Snowdon', sterilized in 50% ethanol and rinsed in sterile water, were planted in 11 cm pots filled with the substrate with or without the fungi. As inoculum, the isolates 10, 22, 160, 250, 471 of *Fusarium oxysporum*, 25, 31, 221, 401, 498 of *Fusarium avenaceum* and 140, 241, 15, 16, 162 of *Sclerotinia sclerotiorum* were selected for the tests. The isolates were obtained from diseased chrysanthemum plants in 2004–2006. Chrysanthemum plants were grown in the growth chamber at 22–23°C with 12 hours photoperiod for 7 weeks. The plants were evaluated for the degree of infection after two and five weeks, using 5-grade scale: 0 – no symptoms, 1 – yellowing of bottom leaves, 2 – yellow or necrotic spots on all leaves, 3 – wilting, 4 – death. The data were processed by the McKinney's formula [Kopacki and Wagner 2004], which generates a numeric disease index (DI) of the severity of the attack: $DI = (\Sigma vn) / (NV) \cdot 100$, where v represents the numeric value of the class, n is the number of plants assigned to the class, N is the total number of the plants in the replication and V is the numeric value of the highest class.

The parameters of fluorescence (Fo-minimum fluorescence yield in dark-adapter state, Fm-maximum fluorescence yield in dark-adapter state, Fv / Fm – quantum yield of PSII photochemistry in the dark adapted state (equivalent to [Fm – Fo] / Fm)) were measured in two periods with the PAM fluorometer (H. Waltz GmbH, Germany). For this test, the live leaves in the same position on plants were selected. Results were analyzed statistically with Tukey test. To test the correlation between the parameters and disease index the regression analysis was used [Frątczak et al. 2005].

Koch's postulate was met by re-isolating the fungi from diseased plants.

RESULTS

Mycological analysis. During the experiments at the stages of growth and flowering necrotic spots, leaves yellows, stem and root rot and tracheomycosis were observed. In 2004–2006 the percentage of diseased plants ranged from 5 to 40%. The highest percentage of infected plants was noticed in autumn (tab. 1).

Table. 1. Percentage of infected plants of chrysanthemums growing under cover

Cultivar	2004		2005		2006		Mean
	S	A	S	A	S	A	
Cassablanca	10	20	10	40	30	40	25
Royalys	5	10	5	5	10	30	11
Snowdon	10	20	10	30	10	40	20

S – summer, A – autumn

Table. 2. Fungi colonizing roots of chrysanthemum cultivated under cover

Year	Fungus species	Cultivar						Total (%)
		Cassabl.		Royalys		Snowdon		
		S	A	S	A	S	A	
2004	*Alternaria alternata* (Fr.) Keiss.	28	30	31	48	22	83	242 (30)
	Botrytis cinerea Pers. ex Fries.	–	15	–	10	13	31	69 (8)
	Cylindrocarpon destructans (Zins.) Sch.	–	4	5	14	10	34	67 (8)
	Fusarium avenaceum (Corda ex Fries) Sacc.	2	6	7	34	12	45	106 (13)
	Fusarium culmorum (Smith) Sacc.	–	7	4	12	–	23	46(6)
	Fusarium equiseti (Corda) Sacc.	–	16	–	–	2	2	20 (2)
	Fusarium oxysporum Schlecht.	5	14	6	17	19	34	95 (12)
	Fusarium solani Mar. Sacc.	2	2	–	–	6	12	22 (3)
	Penicillium expansum Link ex S.F. Gray	9	11	3	6	12	32	73 (9)
	Thanatheporus cucumeris Kühn	–	13	4	2	–	15	34 (4)
	Trichoderma harzianum Rifai	–	4	–	–	–	3	7 (1)
	Trichoderma koningii Oud.	4	11	–	–	13	7	35 (4)
	Total	50	133	60	143	109	321	816
2005	*Alternaria alternata* (Fr.) Keiss.	13	29	12	41	22	31	148 (24)
	Botrytis cinerea Pers. ex Fries.	–	11	–	–	4	34	49(8)
	Cylindrocarpon destructans (Zins.) Sch.	–	–	5	23	–	8	36(6)
	Epicoccum nigrum Link	3	23	–	–	–	21	47(8)
	Fusarium avenaceum (Corda ex Fries) Sacc.	3	2	4	12	8	22	51(8)
	Fusarium culmorum (Smith) Sacc.	3	14	–	–	–	–	17(3)
	Fusarium equiseti (Corda) Sacc.	–	–	2	2	–	–	4 (1)
	Fusarium oxysporum Schlecht.	6	6	2	14	6	31	65 (11)
	Mucor mucedo Mich. Ex St.-Am.	–	–	7	12	–	–	19 (3)
	Penicillium expansum Link ex S.F. Gray	6	3	–	–	4	12	25 (4)
	Sclerotinia slerotiorum (Lib.) de By	–	–	–	12	2	32	46 (8)
	Trichoderma harzianum Rifai	–	2	11	5	6	21	45 (7)
	Trichoderma koningii Oud.	3	–	–	–	4	14	21 (3)
	Verticillium dahliae Kleb.	–	–	–	–	14	22	36 (6)
	Total	37	90	43	121	70	248	609
2006	*Alternaria alternata* (Fr.) Keiss.	5	37	11	26	22	41	142 (16)
	Botrytis cinerea Pers. ex Fries.	–	12	6	18	–	32	68 (8)
	Chaetomium globosum Kunze ex Steud.	–	14	–	–	19	–	33 (4)
	Epicoccum nigrum Link	6	–	4	5	–	11	26 (3)
	Fusarium avenaceum (Corda ex Fries) Sacc.	2	17	9	26	8	42	104 (11)
	Fusarium equiseti (Corda) Sacc.	–	9	5	–	11	14	39 (4)
	Fusarium oxysporum Schlecht.	11	26	21	41	10	43	152 (17)
	Fusarium solani (Mart.) Sacc.	–	–	7	6	–	10	23 (3)
	Penicillium expansum Link ex S.F.Gray	2	17	–	–	9	33	61 (7)
	Penicillium nigricans (Bain.) Thom	–	–	7	5	–	12	24 (3)
	Sclerotinia sclerotiorum (Lib.) de By.	–	23	–	14	16	31	84 (9)
	Thanatheporus cucumeris Kühn	6	27	–	–	–	16	49 (5)
	Trichoderma koningii Oud.	–	–	24	–	–	19	43 (4)
	Verticillium dahliae Kleb.	–	–	–	–	15	38	53 (6)
	Total	32	182	94	141	110	342	901

Table 3. Fungi colonizing stems of chrysanthemum cultivated under cover

Year	Fungus species	Cultivar						Total (%)
		Cassabl.		Royalys		Snowdon		
		S	A	S	A	S	A	
2004	*Alternaria alternata* (Fr.) Keiss.	22	46	31	32	28	68	227(24)
	Alternaria chrysanthemi Simm. & Crossier	–	–	–	–	12	37	49(5)
	Botrytis cinerea Pers. ex Fries.	–	4	–	10	–	21	35 (4)
	Bipolaris setariae (Saw.) Shoemaker	3	11	–	–	–	12	26 (3)
	Cladosporium cladosporioides (Fres.)	–	31	–	–	–	7	38 (4)
	Fusarium avenaceum (Corda ex Fries) Sacc.	–	14	–	13	17	26	70 (7)
	Fusarium culmorum (Smith) Sacc.	5	23	–	–	–	–	28(3)
	Fusarium equiseti (Corda) Sacc.	2	4	–	–	11	17	34 (4)
	Fusarium oxysporum Schlecht.	5	14	3	4	16	29	71 (8)
	Fusarium sporotrichioides Scherb.	3	–	11	–	–	–	14 (2)
	Penicillium verrucosum Dierckx	–	–	–	17	–	35	52 (6)
	Phoma chrysanthemicola Hollos	–	13	–	–	–	–	13 (1)
	Rhizoctonia solani Kühn	6	11	2	2	7	21	49 (5)
	Sclerotinia slerotiorum (Lib.) de By	–	23	–	–	15	36	74 (8)
	Trichoderma harzianum Rifai	12	8	21	29	16	33	119(13)
	Truncatella angustata (Pers. ex Lk) Hughes	–	14	–	–	–	19	33 (3)
	Total	58	216	68	107	122	361	932
2005	*Alternaria alternata* (Fr.) Keiss.	27	17	11	12	18	39	124(20)
	Botrytis cinerea Pers. ex Fries.	–	12	–	18	7	7	44 (7)
	Epicoccum nigrum Link	2	–	–	13	–	–	15 (2)
	Fusarium avenaceum (Cordaex Fries) Sacc.	3	–	19	13	19	21	75 (12)
	Fusarium culmorum (Smith) Sacc.	2	–	–	19	–	–	21 (3)
	Fusarium equiseti (Corda) Sacc.	–	12	–	–	–	–	12 (2)
	Fusarium oxysporum Schlecht.	11	18	–	14	9	27	79 (12)
	Mucor mucedo Mich. ex St.-Am.	–	7	–	–	–	21	28 (4)
	Penicillium expansum Link ex S.F. Gray	–	–	23	–	–	13	36 (6)
	Sclerotinia slerotiorum (Lib.) de By	–	17	–	29	14	28	88 (14)
	Trichoderma harzianum Rifai	–	22	13	11	–	19	65 (10)
	Trichoderma koningii Oud.	14	–	–	23	13	–	50 (8)
	Total	59	105	66	152	80	175	637
2006	*Alternaria alternata* (Fr.) Keiss.	27	34	11	17	29	33	151(23)
	Botrytis cinerea Pers. ex Fries.	–	12	–	23	2	31	68 (10)
	Cylindrocarpon destructans (Zins.) Sch.	2	2	5	13	–	–	22 (3)
	Epicoccum nigrum Link	4	3	–	–	11	8	26 (4)
	Fusarium avenaceum (Corda ex Fries) Sacc.	2	3	3	17	8	19	52 (8)
	Fusarium culmorum (Smith) Sacc.	8	5	–	–	–	14	27 (4)
	Fusarium oxysporum Schlecht.	7	12	–	14	12	21	66 (10)
	Fusarium solani (Mart.) Sacc. .	–	–	–	–	–	4	4 (1)
	Penicillium nigricans (Bain.) Thom	3	3	5	–	–	8	19 (3)
	Phoma egzigua Desm.	5	11	–	–	–	–	16 (2)
	Thanatheporus cucumeris Kühn	3	7	–	–	5	4	19 (3)
	Sclerotinia slerotiorum (Lib.) de By	9	13	–	11	17	39	89 (13)
	Trichoderma koningii Oud.	–	–	15	7	19	24	65 (10)
	Verticillium dahliae Kleb.	–	–	–	–	15	28	43 (6)
	Total	70	105	39	102	118	233	667

Table 4. Fungi colonizing leaves of chrysanthemum cultivated under cover

Year	Fungus species	Cultivar						Total (%)
		Cassabl.		Royalys		Snowdon		
		S	A	S	A	S	A	
2004	*Alternaria alternata* (Fr.) Keiss.	11	34	8	17	21	30	121(22)
	Alternaria chrysanthemi Simm. & Crossier	–	12	–	–	–	19	31(6)
	Botrytis cinerea Pers. ex Fries.	–	7	3	9	–	14	33(6)
	Epicoccum nigrum Link	5	2	–	–	6	11	24(4)
	Fusarium avenaceum (Cordaex Fries) Sacc.	–	12	–	8	4	18	42(8)
	Fusarium oxysporum Schlecht.	–	8	–	–	5	7	20(4)
	Gliocladium catenulatum Gillman & Abbot	12	23	–	19	7	11	72(13)
	Penicillium expansum Link ex S.F. Gray	–	19	14	6	4	13	56(10)
	Rhizoctonia solani Kühn	–	11	–	–	–	21	32(6)
	Septoria chrysanthemella Sacc.	–	28	–	–	–	14	42(7)
	Stemphyllium lycopersici (Enjoji) W. Yam.	–	19	–	–	11	–	30(5)
	Trichoderma koningii Oud.	–	–	11	26	–	14	51(9)
	Total	281	175	36	85	58	172	554
2005	*Alternaria alternata* (Fr.) Keiss.	4	25	–	11	–	31	71(16)
	Botrytis cinerea Pers. ex Fries.	–	13	2	8	–	12	35(8)
	Cladosporium cladosporioides (Fres.)	6	31	–	–	–	17	54(13)
	Fusarium avenaceum (Corda ex Fries) Sacc.	–	7	–	–	12	16	35(8)
	Fusarium equiseti (Corda) Sacc.	5	–	2	–	8	4	19(5)
	Fusarium oxysporum Schlecht.	2	6	–	–	5	18	31(7)
	Mucor mucedo Mich. Ex St.-Am.	4	6	–	16	21	12	59(14)
	Penicillium nigricans (Bain.) Thom	–	–	–	9	3	14	26(6)
	Sclerotinia slerotiorum (Lib.) de By		12	–	–	–	31	43(10)
	Trichoderma harzianum Rifai	6	11	3	–	3	–	23(5)
	Trichoderma koningii Oud.	8	–	–	–	–	27	35(8)
	Total	35	111	7	44	52	182	431
2006	*Alternaria alternata* (Fr.) Keiss.	10	23	4	31	12	34	114(21)
	Aspergillus niger van Tieghem	–	–	–	–	–	27	27(5)
	Botrytis cinerea Pers. ex Fries.	–	–	–	–	14	31	45(8)
	Chaetomium globosum Kunze ex Steud.	–	29	–	–	2	–	31(6)
	Epicoccum nigrum Link	3	7	7	–	12	–	29(6)
	Fusarium avenaceum (Corda ex Fries) Sacc.	–	–	10	8	13	22	53(10)
	Fusarium culmorum (Smith) Sacc.	7	5	–	–	–	12	24(4)
	Fusarium oxysporum Schlecht.	2	2		7	–	9	20(4)
	Penicillium expansum Link ex S.F. Gray	–	11	18	–	–	–	29(5)
	Penicillium verrucosum Dierckx	8	–	–		14	9	31(6)
	Septoria chrysanthemella Sacc.	14	25	–	11	–	23	73(13)
	Trichoderma koningii Oud.	6	5	–	24	12	19	66(12)
	Total	50	107	39	81	79	186	542

In 2004–2006 from roots, as the result of mycological analysis, 2326 fungal colonies belonging to 18 species were isolated. In 2004–2006 on stems, 2236 fungal colonies belonging to 22 species were isolated (tab. 2). From leaves only 1527 colonies (19 species) were isolated (tab. 4). More colonies were obtained in autumn than in summer. *Alternaria alternata*, *Fusarium* spp. and *Sclerotinia sclerotiorum* predominated among fungi which are regarded as pathogenic. The occurrence of individual species of

Fusarium depended on the season of vegetation as well as on the cultivar. *F. oxysporum* was isolated frequently from all plant organs of cv. 'Casablanca' in summer and in autumn. A different tendency was observed for *F. avenaceum*, the number of isolated colonies was higher in autumn. Similarly, *S. sclerotiorum* colonised frequently stems at the end of vegetation period (tab. 3). *A. alternata* occurred on cvs. 'Casablanca' and 'Snowdon' plants in both seasons but more frequently in autumn. Also *Cylindrocarpon obtusisporum*, *Rhizoctonia solani* and *Septoria chrysanthemella* were isolated frequently in autumn.

Fusarium spp. occurred less frequently on cv. 'Royalys' plants. *Fusarium oxysporum* and *F. avenaceum* were also isolated in each season. Other *Fusarium* spp. were obtained less often. More colonies of *S. sclerotiorum* were obtained in autumn than in summer. *A. alternata* was isolated in all seasons but more frequently in autumn. Also in autumn numerous colonies of *Bipolaris setariae* were obtained.

The lowest number of colonies was obtained from cv. 'Royalys' plants. Only *A. alternata* occurred frequently. Among *Fusarium* spp. only *F. oxysporum* was isolated frequently, especially in autumn. Other *Fusarium* spp. were obtained less frequently. Also *R. solani*, *B. cinerea* and *S. chrysanthemella* colonised the plants very rarely (tabs 2, 3, 4).

Analysis of pathogenicity and photosynthesis parameters. In one combination with *F. oxysporum* isolate the plants showed disease symptoms at late stages of growth. The affected plants were stunted, with yellow leaves. One isolate (Fo22) was pathogenic to chrysanthemum (disease index 4.8%) (tab. 5). Statistical analysis showed that most of fluorescence parameters of this isolate differ significantly (LSD 5%) from those of control plants (tab. 5).

Table 5. The photosynthetic activity of chrysanthemums growing with *F. oxysporum* isolates

Isolates	1st period			2nd period		
	Fo	Fm	Fv/Fm	Fo	Fm	Fv/Fm
Control	0.286a*	1.562a	0.739a	0.309a	1.762a	0.796a
Fo10	0.278a	1.240c	0.692c	0.279bc	1.401c	0.597b
Fo22	0.264ab	1.215c	0.680d	0.288b	1.360d	0.682ab
Fo160	0.248bc	1.400b	0.711b	0.220d	1.313e	0.700ab
Fo250	0.271ab	1.203c	0.678d	0.270c	1.425b	0.700ab
Fo471	0.233c	1.316bc	0.664e	0.283bc	1.319e	0.686ab
LSD (0.05)	0.0282	0.1142	0.0115	0.016	0.0115	0.1584

* – mean values in columns differ significantly ($P \leq 0.05$), if they are not marked the same letter

In most combinations with *F. avenaceum* isolates, the plants showed symptoms at early stages of growth. The affected plants were stunted with various degrees of necrosis on their stems. Statistical analysis showed that nearly all isolates were pathogenic to chrysanthemum. For four isolates (Fa25, Fa31, Fa221, Fa498) the disease index was below 50% (tab. 6). One isolate (Fa401) showed very high pathogenicity (disease index 86, 8% and 96, 6%). The results of pathogenicity tests were confirmed partly by fluorescence measurements. The values of minimal (Fo) and maximal (Fm) fluorescence yield were lower in all combinations with the pathogen than in the control.

The photochemical quantum yield of PSII was also reduced at both periods and differ significantly (LSD 5%) from that of control plants (tab. 6).

Table 6. The photosynthetic activity of chrysanthemums growing with *F. avenaceum* isolates

Isolates	1st period			2nd period		
	Fo	Fm	Fv/Fm	Fo	Fm	Fv/Fm
Control	0.286a*	1.562a	0.739a	0.309a	1.762a	0.796a
Fa25	0.201c	0.988e	0.514d	0.284a	1.460c	0.688d
Fa31	0.199c	1.303b	0.566c	0.300a	1.565b	0.677e
Fa221	0.216b	1.250c	0.621b	0.300a	1.439d	0.701c
Fa401	0.113d	0.858f	0.420e	0.186a	1.250f	0.519f
Fa498	0.200c	1.213d	0.614b	0.317a	1.368e	0.713b
LSD (0.05)	0.0115	0.0115	0.0115	0.1584	0.016	0.0027

* – mean values in columns differ significantly ($P \leq 0.05$), if they are not marked the same letter

Table 7. The photosynthetic activity of chrysanthemums growing with *S. sclerotiorum* isolates

Isolates	1st period			2nd period		
	Fo	Fm	Fv/Fm	Fo	Fm	Fv/Fm
Control	0.286a*	1.562a	0.739a	0.309a	1.762a	0.796a
Ss15	0.167b	1.05b	0.546b	0.281c	1.360b	0.640b
Ss16	0.000e	0.000c	0.000e	0.000e	0.000c	0.000c
Ss140	0.115c	0.600b	0.514c	0.285b	1.360b	0.600b
Ss162	0.000e	0.000c	0.000e	0.000e	0.000c	0.000c
Ss241	0.102d	1.02b	0.504d	0.248d	1.300b	0.605b
LSD (0.05)	0.0022	0.4883	0.0022	0.0022	0.1131	0.1125

* – mean values in columns differ significantly ($P \leq 0.05$), if they are not marked the same letter

In all combinations with *S. sclerotiorum* isolates the plants showed disease symptoms at early stages of growth. The affected plants were stunted with yellows and various degrees of necrosis on stem and roots. Statistical analysis showed that all isolates were pathogenic to chrysanthemum. The disease index for all isolates was above 50% (tab. 7). All isolates showed very high pathogenicity and two isolates (Ss16, Ss162) caused damping-off of all plants after two weeks. The results of pathogenicity tests were confirmed partly by fluorescence measurements. The values of minimal (Fo) and maximal (Fm) fluorescence yield were lower in all combinations with the pathogen than in the control. The photochemical quantum yield of PSII was also reduced in both periods and differ significantly (LSD 5%) from those of control plants (tab. 7).

Table 8. The correlation between disease index and potential quantum yield PS II

Isolates	1st period		2nd period	
	DI (%)	Fv/Fm	DI (%)	Fv/Fm
Control	0	0.739	0	0.796
Fo10	0	0.692	0	0.597
Fo22	0	0.680	4.8	0.682
Fo160	0	0.711	0	0.700
Fo250	0	0.678	0	0.700
Fo471	0	0.664	0	0.686
Fa25	0	0.514	0	0.688
Fa31	40.2	0.566	41.8	0.677
Fa221	14.8	0.621	18.4	0.701
Fa401	86.8	0.420	96.6	0.519
Fa498	10.0	0.614	11.6	0.713
Ss15	79.8	0.546	83.2	0.640
Ss16	100.0	0	100.0	0
Ss140	65.0	0.514	65.0	0.600
Ss162	100.0	0	100.0	0
Ss241	65.0	0.504	73.2	0.605

Regression analysis showed that there was the correlation (r) between actual quantum yield and disease index: -0,723351 (tab. 8).

DISCUSSION

Chlorophyll fluorescence reflects the effectiveness of the plant photosystem, which depends on several factors [Maxwell and Johnson 2000]. One of these is plant health status. The results proved that *F. oxysporum*, *F. avenaceum* and *S. sclerotiorum* had an effect on some parameters of chloroplyll fluorescence in chrysanthemums leaves. The decrease of fluorescence parameters, especially Fv/Fm, proved that the photosystem activity of chrysanthemum plants was reduced. The results of tests showed correlation between diseases index values and fluorescence parameters. The isolates showing the highest pathogenic abilities caused damage to plants equivalent to a 0 on the fluorometer scale. Other reports also prove the negative effects of fungal inoculation on chlorophyll fluorescence [Ye et al. 2004]. Most of the reports, however, describe the pathogens of leaves and other green parts of plants [Murray and Walters 1992, Pośpieszny and Struszczyk 2003]. Our results show that root pathogens can also cause dramatic changes in the plant photosystem.

Grzesiuk et al. [1999] informed that intensity of photosynthesis might be reduced even by 75%. The reduction of photosynthesis intensity is the result of the decrease of assimilative surface due to the destruction of green organs or necrotic spots. In such cases the chlorophyll content is reduced, the chloroplasts are degraded and the functions of stomata are disturbed.

Chlorophyll fluorescence parameters help to evaluate the photosynthesis activity of plants, especially under such stress as the infection of plant by pathogens [Mikos-Bielak and Michałek 1999, Nogues et al. 2002]. When roots or vessels are infected, the uptake of water is disturbed which has a negative effect on photosynthesis. All tested isolates

affected the photosynthetic intensity, even when the disease index was low or equal to zero.

The reduced fluorescence in plants without disease symptoms can be explained by their manner of colonization. In our experiment in the combinations with four isolates of *F. oxysporum* and one isolate of *F. avenaceum* the decrease in fluorescence was observed in plants without disease symptoms. Santos et al. [2002] investigating the effect of *Colletotrichum musae* and *F. moniliforme* on the photosynthetic efficiency on banana and maize, respectively, found that both fungi impaired the photosynthesis, even if the plants did not showed any disease symptoms. They showed that both fungi caused similar reductions in photosynthetic capacity. It was possible that the reduction in the maximum yield of photosynthesis in both plants was the result of toxins produced by the fungi.

Both their results and ours confirm that with the measurements of chlorophyll fluorescence it is possible to detect the infection before the appearance of disease symptoms.

CONCLUSIONS

1. Among the investigated cultivars the least susceptible seems to be 'Royalys'.
2. *Alternaria alternate*, *Sclerotinia aclerotiorum* and *Fusarium* spp. predominated among fungi isolated from diseased chrysanthemum plants.
3. *S. sclerotiorum* proved to be the most pathogenic to chrysanthemum plants.
4. The disease index and the reduction of photosynthetic activity of plants were strongly correlated for most of tested isolates.
5. With the measurement of chlorophyll parameters it is possible to detect the disease very early.

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PATOGENICZNOŚĆ *Fusarium oxysporum, Fusarium avenaceum* I *Sclerotinia sclerotiorum* I ICH WPŁYW NA AKTYWNOŚĆ FOTOSYNTETYCZNĄ ROŚLIN CHRYZANTEM

Streszczenie. W przeprowadzonych eksperymentach określano grzyby zasiedlające korzenie, podstawę pędu oraz liście trzech odmian chryzantem w uprawie pod osłonami. Następnie badano patogeniczność *Fusarium oxysporum, Fusarium avenaceum* i *Sclerotinia sclerotiorum* względem chryzantemy odmiany Snowdon. Określano także indeks chorobowy na porażonych roślinach i porównywano do parametrów fotosyntezy. Dla większości izolatów indeks chorobowy był skorelowany ze spadkiem fotosyntezy roślin, jednak w pewnych przypadkach ograniczenia wydajności fotosyntezy były silniejsze niż wynikało to z zewnętrznych objawów chorobowych. Sugeruje to, że pomiary fluorescencji chlorofilu mogą być pomocne dla szybkiej oceny rozwoju choroby.

Słowa kluczowe: grzyby patogeniczne, podatność odmian, fluorescencja chlorofilu

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