ACTA MYCOLOGICA
Vol. 48 (2): 227–236
2013
DOI: 10.5586/am.2013.024

*Dedicated to Professor Maria Ławrynowicz on the occasion of the 45th anniversary of her scientific activity*

# Diversity of fungi colonizing and damaging leaves of pontic azalea *Azalea pontica*

MARIA KOWALIK

Department of Plant Protection, University of Agriculture in Kraków
29 Listopada 54, PL-31-425 Kraków, m.kowalik@ogr.ur.krakow.pl

Kowalik M.: *Diversity of fungi colonizing and damaging leaves of pontic azalea Azalea pontica.* Acta Mycol. 48 (2): 227–236, 2013.

The research aimed at verification of fungi species colonizing phyllosphere of pontic azalea *Azalea pontica* L. and at comparison of the fungi species composition: – in the natural stand in the Kołacznia nature reserve, – in arboretum collections at Bolestraszyce and Rogów. 600 fragments of healthy, infected and fallen leaves of pontic azalea were collected for mycological analyses. The species forming the largest number of colonies identified from the healthy leaves were: *A. alternata*, *Ph. cyclaminis*, *E. nigrum*, *Ph. medicaginis* and *B. cinerea,* from infected leaves*: A. alternata*, *E. nigrum, Ph. cyclaminis*, *S. fimicola*, *T. viride* and *A. phaeospermum,* whereas: *E. nigrum*, *A. alternata*, *S. fimicola*, *Ph. cyclaminis and B. cinerea* were isolated from the fallen leaves, which indicates that a majority of fungi persistently colonize the leaves during vegetation period and damage them, which leads to defoliation. Colonization of pontic azalea phyllosphere in arboreta by more numerous fungi colonies and species than under conditions of natural sites evidences their increased pressure in the arboreta environment.

**Key words:** pontic azalea, arboretum, reserve, health status, leaves

## INTRODUCTION

Pontic azalea *Azalea pontica* L., i.e. yellow rhododendron [*Rhododendron luteum* Sweet, syn. *R. flavum* G. Don (Anioł-Kwiatkowska 2003)] is a critically endangered species included in The Polish Red Book of Plants (Kaźmierczakowa, Zarzycki 2001). Its only natural stand in Poland is located in the Kołacznia nature reserve at Wola Zarzycka near Leżajsk. The seeds collected on the natural site were used to cultivate the specimens which are growing in the collection of Arboretum and Department of Physiography in Bolestraszyce and in the Arboretum of the Warsaw University of Life Sciences (SGGW) in Rogów (Piórecki, Dubiel 2009; Piórecki, Zarzycki 2010).

Health status of azalea shrubs is affected by cultivation conditions resulting from the specific site requirements and disease agents. Identification of the organisms colonizing phyllosphere of pontic azalea, including pathogens causing health disturbances, is a comparative study comprising plants under conditions of natural sites and in arboreta. Identification of the population and species diversity of fungi in pontic azalea phyllosphere makes possible following the results of colonization and damaging of leaves.

The aim of the research was verification of fungi species colonizing the phyllosphere of *Azalea pontica* and comparison of fungi species composition on the natural site and in the arboreta.

## MATERIALS AND METHODS

Observations of the of pontic azalea health status were conducted in the third decade of May, July, August and September 2011 on a site in the Kołacznia nature reserve, on the collection of the Arboretum in Bolestraszyce (SE Poland) and on the collection of Arboretum in Rogów (Central Poland). On each date 10 healthy leaves (green, without obvious disease symptoms), infected leaves (showing necrotic symptoms) and fallen leaves were collected from the three sites. A total of 1800 leaf fragments were collected for mycological analyses. The leaf fragments were disinfected in 70% ethanol. Isolation and cultivation of mycobiota were conducted according to standard methods applied in mycology (Kowalik 2008).

For taxonomic identification of the mycobiota the following keys were used: Guba (1961); Domsch et al. (1980); Sutton (1980); Ellis, Ellis (1987) and Rifai (1987). The basis of classification was the system of Kirk et al. (2008) and the authors' epithets by fungal species names were verified according to Index Fungorum (2012).

On the basis of fungi specification considering the share of individual species in the total fungi community, they were classified to the group of dominants (constituting >5% of the entire community), influents (1-5%) and accessory fungi (<1%). Similarity coefficient (Sőrensen index) was calculated for the analysed sites, comprising the number of fungi species (Kowalik 1993).

## RESULTS

Altogether 2120 fungi colonies belonging to 64 species were isolated from the plant material: healthy, infected and fallen leaves of *Azalea pontica* collected on the natural site in the Kołacznia nature reserve, on the collection of the Arboretum in Bolestraszyce and on the collection of Arboretum in Rogów.

There were 568 colonies comprising 39 species isolated from healthy green leaves (Tab. 1), 657 colonies and 40 species from infected leaves (with necrotic symptoms) (Tab. 2) and 895 colonies and 52 species from fallen leaves (Tab. 3).

The largest number of fungi colonies and species existed on healthy azalea leaves in the Kołacznia nature reserve, whereas the least number in the arboretum in Bolestraszyce. These leaves were in the first place colonized by: *Alternaria alternata*,

*Phialophora cyclaminis*, *Epicoccum nigrum*, *Phoma medicaginis* and *Botrytis cinerea.* These fungi were classified to the dominant group. *Cladosporium sphaerospermum*, *Trichoderma viride*, *Pestalotiopsis sydowiana*, *Isaria fumorosea*, *Mortierella alpina*, *M. parvispora*, *Khuskia oryzae*, *Trichoderma koningii*, *Sordaria fimicola*, *Mammaria echinobotryoides*, *Penicillim expansum*, *P. waksmanii*, *Truncatella truncata* and other were classified to the influent group, whereas the other 10 species were identified as accessory fungi (Tab. 1).

Table 1
Fungi colonizing healthy leaves of pontic azalea *Azalea pontica*

| Fungus | Kołacznia reserve | Bolestraszyce arboretum | Rogów arboretum | Total | Percentage [%] |
|---|---|---|---|---|---|
| *Alternaria alternata* (Fr.) Keissl. | 8 | 28 | 23 | 59 | 10.39 |
| *Apiospora montagnei* Sacc. | 12 | | | 12 | 2.11 |
| *Arthrinium euphorbiae* M.B. Ellis | 9 | | | 9 | 1.58 |
| *Arthrinium phaeospermum* (Corda) M.B. Ellis | | 3 | | 3 | 0.53 |
| *Aspergillus versicolor* (Vuill.) Tirab. | 1 | | | 1 | 0.18 |
| *Aureobasidium pullulans* (de Bary) G. Arnaud | 1 | | 7 | 8 | 1.41 |
| *Botrytis cinerea* Pers | 31 | | 1 | 32 | 5.63 |
| *Chaetomim globosum* Kunze | 2 | | | 2 | 0.35 |
| *Cladosporium cladosporioides* (Fresen.) G.A. de Vries | 1 | 1 | 9 | 11 | 1.94 |
| *Cladosporium herbarum* (Pers.) Link | | 5 | 4 | 9 | 1.58 |
| *Cladosporium sphaerospermum* Penz. | 8 | 5 | 11 | 24 | 4.23 |
| *Coleophoma rhododendri* Syd. | | 1 | | 1 | 0.18 |
| *Davidiella macrocarpa* Crous, K. Schub. & U. Braun | 9 | | 3 | 12 | 2.11 |
| *Epicoccum nigrum* Link | 6 | 27 | 11 | 44 | 7.75 |
| *Giberella pulicaris* (Fr.) Sacc. | | 2 | | 2 | 0.35 |
| *Ilionectria radicicola* (Gerlach & L. Nilson) P. Chaverri & Salgado | | 1 | | 1 | 0.18 |
| *Isaria fumorosea* Wize | 3 | 7 | 12 | 22 | 3.87 |
| *Khuskia oryzae* H.J. Hadson | 7 | | 12 | 19 | 3.34 |
| *Mammaria echinobotryoides* Ces. | 14 | | | 14 | 2.46 |
| *Mortierella alpina* Peyronel | 14 | | 6 | 20 | 3.52 |
| *Mortierella parvispora* Linnem. | | 14 | 7 | 21 | 3.70 |
| *Mucor hiemalis* f. *hiemalis* Wehmer | 1 | | 1 | 2 | 0.35 |
| *Parahoma chrysanthemicola* (Hollós) Gruyter, Aveskamp & Verkley | 1 | 1 | | 2 | 0.35 |
| *Penicillium expansum* Link | | | 13 | 13 | 2.29 |
| *Penicillium waksmanii* K.M. Zalessky | 5 | 8 | | 13 | 2.29 |
| *Pestalotiopsis sydowiana* (Bres.) B. Sutton | | 20 | 3 | 23 | 4.05 |
| *Phialophora cyclaminis* J.F.H. Beyma | 35 | 13 | 1 | 49 | 8.63 |
| *Phoma eupyrena* Sacc. | 2 | | 4 | 6 | 1.06 |
| *Phoma exigua* (Desm.) Gruyter, Aveskamp & Verkley | 1 | 1 | | 2 | 0.35 |
| *Phoma medicaginis* Malbr. & Roum. | 11 | 26 | | 37 | 6.51 |
| *Sordaria fimicola* (Roberge ex Desm.) Ces. & De Not | 11 | | | 11 | 1.94 |
| *Thanatephorus cucumeris* (A.B.Frank) Donk | | 3 | | 3 | 0.53 |
| *Trichoderma koningii* Oudem. | | | 17 | 17 | 2.99 |
| *Trichoderma pseudokoningii* Rifai | | | 11 | 11 | 1.94 |
| *Trichoderma viride* Pers. | 9 | 4 | 14 | 27 | 4.75 |
| *Truncatella truncata* (Lev.) Steyaert | 4 | | 8 | 12 | 2.11 |
| *Umbelopsis isabellina* (Oudem.) W. Gams | | | 9 | 9 | 1.58 |
| *Umbelopsis ramanniana* (Möller) W. Gams | | 2 | | 2 | 0.35 |
| *Umbelopsis vinacea* (Dixon-Stew.) Arx | | 2 | 1 | 3 | 0.53 |
| **Total** | **206** | **174** | **188** | **568** | **100.00** |

Table 2
Fungi colonizing infected leaves of pontic azalea *Azalea pontica*

| Fungus | Kołacznia reserve | Bolestraszyce arboretum | Rogów arboretum | Total | Percentage [%] |
|---|---|---|---|---|---|
| *Alternaria alternata* (Fr.) Keissl. | 21 | 85 | 56 | 162 | 24.65 |
| *Arthrinium euphorbiae* M.B. Ellis | 2 | | | 2 | 0.30 |
| *Arthrinium phaeospermum* (Corda) M.B. Ellis | 16 | 11 | 7 | 34 | 5.17 |
| *Arthrinium sphaerospermum* Fuckel | 2 | | | 2 | 0.30 |
| *Aureobasidium pullulans* (de Bary) G. Arnaud | 1 | | 3 | 4 | 0.60 |
| *Botrytis cinerea* Pers. | 11 | | 7 | 18 | 2.74 |
| *Chaetomim globosum* Kunze | 2 | | 12 | 14 | 2.13 |
| *Cladosporium cladosporioides* (Fresen) G.A. de Vries | 2 | 8 | 3 | 13 | 1.98 |
| *Cladosporium herbarum* (Pers.) Link | | 4 | 6 | 10 | 1.52 |
| *Cladosporium sphaerospermum* Penz. | 1 | 15 | 8 | 24 | 3.65 |
| *Davidiella macrocarpa* Crous, K. Schub. & U. Baun | | | 2 | 2 | 0.30 |
| *Epicoccum nigrum* Link | 14 | 32 | 47 | 93 | 14.16 |
| *Fusarium culmorum* (W.G. Sm.) Sacc. | | 2 | 1 | 3 | 0.46 |
| *Fusarium oxysporum* Schltdl. | 2 | | 3 | 5 | 0.77 |
| *Fusarium poae* (Peck) Wollenw. | | | 4 | 4 | 0.60 |
| *Giberella pulicaris* (Fr.) Sacc. | | | 7 | 7 | 1.07 |
| *Ilionectria radicicola* (Gerlach & L. Nilson) P. Chaverri & Salgado | | | 4 | 4 | 0.60 |
| *Khuskia oryzae* H.J. Hads. | 2 | | | 2 | 0.30 |
| *Mammaria echinobotryoides* Ces. | 6 | 1 | | 7 | 1.07 |
| *Mortierella alpina* Peyronel | 6 | | 7 | 13 | 1.98 |
| *Mortierella parvispora* Linnem. | | 7 | 3 | 10 | 1.52 |
| *Mucor hiemalis* f. *hiemalis* Wehmer | | | 1 | 1 | 0.15 |
| *Paraconiothyrium minitans* (W.A. Campb.) Verkley | | 4 | | 4 | 0.60 |
| *Paraphoma chrysanthemicola* (Hollós) Gruyter, Aveskamp & Verkley | | 3 | | 3 | 0.46 |
| *Paraphoma fimeti* (Brunaud) Gruyter, Aveskamp & Verkley | | 5 | | 5 | 0.77 |
| *Penicillium expansum* Link | | 1 | | 1 | 0.15 |
| *Penicillium waksmanii* K.M. Zalessky | 9 | 7 | | 16 | 2.44 |
| *Pestalotiopsis sydowiana* (Bres) B. Sutton | | | 17 | 17 | 2.59 |
| *Phialophora cyclaminis* J.F.H. Beyma | 16 | 18 | 16 | 50 | 7.61 |
| *Phoma exigua* (Desm.) Aveskamp, Gruyter & Verkley | 1 | | 4 | 5 | 0.77 |
| *Phoma medicaginis* Malbr. & Roum. | 7 | | | 7 | 1.07 |
| *Phoma putaminum* Speg. | 4 | | | 4 | 0.60 |
| *Pleurostomophora richardsiae* (Nannf.) L. Mostert, W. Gams & Crous | | 1 | 4 | 5 | 0.77 |
| *Rhizopus stolonifer* (Ehrenb.) Vuill. | | 6 | | 6 | 0.92 |
| *Sordaria fimicola* (Roberge ex Desm.) Ces & De Not | 23 | 8 | 11 | 42 | 6.40 |
| *Talaromyces wortmannii* C.R. Benj. | 2 | | | 2 | 0.30 |
| *Thanatephorus cucumeris* (A.B. Frank) Donk | | 7 | | 7 | 1.07 |
| *Trichoderma viride* Pers. | 29 | 4 | 2 | 35 | 5.33 |
| *Umbelopsis isabellina* (Oudem.) W. Gams | 1 | 8 | 1 | 10 | 1.52 |
| *Umbelopsis ramanniana* (Möller) W. Gams | 4 | | | 4 | 0.60 |
| **Total** | **184** | **237** | **236** | **657** | **100.00** |

Table 3
Fungi colonizing fallen leaves of pontic azalea *Azalea pontica*

| Fungus | Kołacznia reserve | Bolestraszyce arboretum | Rogów arboretum | Total | Percentage [%] |
|---|---|---|---|---|---|
| *Actinomucor elegans* (Eidam) C.R. Benj. et Hesselt | 3 | | | 3 | 0.34 |
| *Alternaria alternata* (Fr.) Keissl. | 21 | 47 | 51 | 119 | 13.29 |
| *Apiospora montagnei* Sacc. | | 2 | | 2 | 0.22 |
| *Arthrinium euphorbiae* M.B. Ellis | | 1 | | 1 | 0.11 |
| *Arthrinium phaeospermum* (Corda) M.B. Ellis | | | 1 | 1 | 0.11 |
| *Arthrinium sphaerospermum* Fuckel | | 1 | | 1 | 0.11 |
| *Aspergillus ustus* (Bainier) Thom et Church | 2 | | | 2 | 0.22 |
| *Botrytis cinerea* Pers. | | 32 | 28 | 60 | 6.70 |
| *Cadophora malorum* (Kidd et Beaumont) W. Gams | 5 | | | 5 | 0.56 |
| *Calonectria morganii* Crous, Alfenas & M.J. Wingf. | 1 | 1 | | 2 | 0.22 |
| *Chaetomim globosum* Kunze | | | 6 | 6 | 0.67 |
| *Cladosporium cladosporioides* (Fresen.) G.A. de Vries | 2 | 4 | 3 | 9 | 1.01 |
| *Cladosporium sphaerospermum* Penz. | | | 7 | 7 | 0.78 |
| *Epicoccum nigrum* Link | 72 | 72 | 67 | 211 | 23.57 |
| *Fusarium culmorum* (W.G. Sm.) Sacc. | | 2 | 3 | 5 | 0.56 |
| *Fusarium flocciferum* Corda | | | 2 | 2 | 0.22 |
| *Fusarium oxysporum* Schltdl. | | | 4 | 4 | 0.45 |
| *Giberella pulicaris* (Fr.) Sacc. | | | 2 | 2 | 0.22 |
| *Giberella tricincta* El-Gohll, McRitchie, Schoult. et Ridings | | 5 | | 5 | 0.56 |
| *Humicola fuscoatra* var. *fuscoatra* Traaen | | | 4 | 4 | 0.45 |
| *Humicola grisea* Traaen | 7 | | 3 | 10 | 1.12 |
| *Ilionectria radicicola* (Gerlach & L. Nilson) P. Chaverri & Salgado | | 2 | 5 | 7 | 0.78 |
| *Isaria fumosorosea* Wize | | | 2 | 2 | 0.22 |
| *Mammaria echinobotryoides* Ces. | 5 | | | 5 | 0.56 |
| *Mortierella alpina* Peyronel | 2 | | 16 | 18 | 2.01 |
| *Mucor hiemalis* f. *hiemalis* Wehmer | | 5 | 7 | 12 | 1.34 |
| *Oidiodendron griseum* Robak | | | 1 | 1 | 0.11 |
| *Oidiodendron tenuissimum* (Peck) S. Huges | | 1 | 4 | 5 | 0.56 |
| *Paraphoma chrysanthemicola* (Hollós) Gruyter, Aveskamp, Gruyter & Verkley | 1 | 2 | | 3 | 0.34 |
| *Penicillium citrinum* Thom | 1 | 2 | | 3 | 0.34 |
| *Penicillium expansum* Link | | | 3 | 3 | 0.34 |
| *Pestalotiopsis sydowiana* (Bres.) B. Sutton | 1 | | 7 | 8 | 0.89 |
| *Phialophora cyclaminis* J.F.M. Beyma | 42 | 14 | 11 | 67 | 7.49 |
| *Phoma destructiva* Plowr. | 1 | | | 1 | 0.11 |
| *Phoma eupyrena* Sacc. | 9 | | | 9 | 1.01 |
| *Phoma exigua* (Desm.) Aveskamp, Gruyter & Verkley | 4 | 28 | | 32 | 3.58 |
| *Phoma herbarum* Westend. | | | 11 | 11 | 1.23 |
| *Phoma medicaginis* Malbr. & Roum. | 18 | 16 | | 34 | 3.80 |
| *Phoma putaminum* Speg. | 1 | | 3 | 4 | 0.45 |
| *Pleospora azalea* (Voglino) Priest | | | 13 | 13 | 1.45 |
| *Pleurostomophora richardsiae* (Nannf.) L. Mostert, W. Gams & Crous | | | 1 | 1 | 0.11 |
| *Rhizopus stolonifer* (Ehrenb.) Vuill. | | 16 | 11 | 27 | 3.02 |
| *Scopulariopsis brumptii* Salv.-Duval | | 6 | | 6 | 0.67 |
| *Scopulariopsis chartarum* (G. Sm.) F.J. Morton et G.Sm. | | 11 | | 11 | 1.23 |
| *Sordaria fimicola* (Roberge ex Desm.) Ces. et De Not | 30 | 28 | 22 | 80 | 8.94 |
| *Talaromyces wortmannii* C.R. Benj. | | | 3 | 3 | 0.34 |
| *Trichoderma koningii* Oudem. | | | 6 | 6 | 0.67 |
| *Trichoderma pseudokoningii* Rifai | | | 2 | 2 | 0.22 |
| *Trichoderma viride* Pers. | | 9 | 5 | 14 | 1.56 |
| *Truncatella truncata* (Lév.) Steyaert | | 3 | 15 | 18 | 2.01 |
| *Umbelopsis isabellina* (Oudem.) W. Gams | 12 | 5 | 7 | 24 | 2.68 |
| *Umbelopsis ramanniana* (Möller) W. Gams | 4 | | | 4 | 0.45 |
| **Total** | **244** | **315** | **336** | **895** | **100.00** |

Table 4
Similarity coefficient for fungi communities isolated from leaves of pontic azalea *Azalea pontica* on three sites

| Leaves | Similarity coefficient [%] | | |
|---|---|---|---|
| | Kołacznia - Bolestraszyce | Kołacznia - Rogów | Bolestraszyce - Rogów |
| Healthy | 39.13 | 45.83 | 50.00 |
| Infected | 48.88 | 48.97 | 56.52 |
| Fallen | 44.44 | 28.57 | 31.03 |
| **Total** | **47.79** | **40.79** | **44.59** |

Infected leaves, showing symptoms of necrosis collected in arboreta were colonized by a comparable number of fungi colonies, while leaves in Bolestraszyce and on the natural site in Kołacznia by a similar number of fungi species (Tab. 2). Dominant colonies: *A. alternata*, *E. nigrum*, *Ph. cyclaminis*, *S. fimicola*, *T. viride* and *Arthrinium sphaerospermum* were isolated from these leaves in a prevailing number. 14 species which were classified to the influents included: *C. sphaerospermum*, *B. cinerea*, *Chaetomium globosum*, *P. sydowiana*, *M. alpina* and *P. waksmanii.*

The least numerous fungi colonies and species existed on the fallen leaves gathered in the Kołacznia reserve, whereas the largest number was found in the Rogów arboretum (Tab. 3). *Epicoccum nigrum*, *A. alternata*, *S. fimicola*, *Ph. cyclaminis* i *B. cinerea* colonies dominated in fungi community isolated from the fallen azalea leaves. In the influent group the most numerous were: *Ph. medicaginis*, *Ph. exigua* and *Rhizopus stolonifer*.

A total of 634 colonies comprising 37 species were identified on the leaves in three analyzed sites in the Kołacznia reserve, 726 colonies and 43 species in the Bolestraszyce arboretum and 28 species within 760 colonies in the arboretum in Rogów.

Similarity indices between individual sites of pontic azalea sites, computed for the communities of fungi species (Tab. 4), evidence the greatest number of common species (50-56%) occurring on infected and healthy leaves in the arboreta in Bolestraszyce and Rogów. The same high similarity index (over 48%) was computed for infected leaves collected in the Kołacznia reserve and in both arboreta. Fungi communities isolated from fallen leaves differed considerably by their species composition, the least number of common species (28%) occurred in the fungi community isolated from leaves in the Kołacznia reserve and Rogów arboretum.

## DISCUSSION

Azalea shrubs growing on natural sites, in collections of arboreta or botanical gardens, but also in city green areas or in household gardens are under pressure of various pathogenic organisms, most frequently fungi and fungus-like organisms (Kita, Mazurek 2003; Kowalik 2008; Kowalik et al. 2012). The importance of phyllosphere, in which a competition exists between pathogens and saprotrophs resulting in diseases leading to premature plant defoliation, is emphasized in discussion on plant health status.

Generally, fungi species colonizing healthy pontic azalea leaves were saprotrophs from the genera of *Apiospora*, *Arthrinium*, *Isaria*, *Khuskia*, *Mammaria*, *Mortierella*, *Trichoderma* and *Umbellopsis.* The share of necrotrophs, such as *A. alternata*, *E. nigrum* and *S. fimicola* in the total fungi community isolated from azalea leaves on the natural site in the Kołacznia reserve was small, whereas *S. fimicola* did not colonize azalea leaves in the arboreta at all. While comparing fungi communities isolated from pontic azalea leaves and evergreen rhododendron leaves (Kowalik 2009), it was noticed that also numerous saprotrophs from the *Acremonium*, *Aspergillus*, *Chaetomium* and *Humicola* genera colonized azalea leaf blades.

Asymptomatic colonization of pontic azalea by *B. cinerea* in the Kołacznia reserve in May during the vegetation period resulted in the occurrence of grey mould on the bushes with visible symptoms of necrosis on the edges of leaf blades.

The number of fungi in the community colonizing healthy laves of azalea growing on the natural site in the Kołacznia reserve was much higher than in the arboreta in Bolestraszyce or Rogów, however, the number of pathogen colonies in this community was low. A similar relationship was described by Kita and Mazurek (2003) who compared azalea health status in the arboretum in Wojsławice and Botanical Garden in Wrocław (SW Poland), however ascribing the effect of polluted air on reduction of pathogenic fungi seems unfounded.

Diversified similarity indices for fungi communities isolated from healthy leaves indicate a species diversity of fungi colonizing azalea leaves on natural sites and in arboreta.

*Alternaria alternata* and *E. nigrum* dominated on infected, necrotic azalea leaves collected in the arboreta in Bolestraszyce and Rogów, which confirms previous research results (Kowalik 2009; Kowalik et al. 2011). The same fungi, but also *Coelophoma empetri*, *Humicola fuscoatra*, *H. grisea*, *P. expansum*, *Penicillium verrucosum*, *P. sydowiana*, *Septoria azalea*, *S. fimicola*, *Umbelopsis isabellina*, as well as fungi from *Cylindrocarpon*, *Fusarium*, *Mortierella*, *Phialophora*, *Phoma*, *Trichoderma* and other genera were found on necrotic evergreen rhododendron and azalea leaves (Kowalik, Muras 2007; Kowalik 2008; Kowalik et al. 2010a; 2010b; 2012). The results obtained confirm the pathogenicity and aggressiveness of many of these fungi species, previously registered on rhododendron leaves. Leaf damage due to *Thanatephorus cucumeris,* a pathogen only sporadically isolated from rhododendron leaves (Kowalik et al. 2010b) is visible in the presented investigations.

High similarity indices for fungi communities isolated from infected leaves indicate that necrosis of azalea leaves on three analyzed sites was caused by a group of pathogenic species, including necrotrophs. Numerous investigations (Kita, Mazurek 2003; Kowalik, Muras 2007; Kowalik et al. 2010 a; 2011) documented the presence and role of necrotrophs in the process of causing necrotic blots, dieback and intensified, premature falling of leaves. It was demonstrated that toxic fungus *A. alternata* played the dominant role. Presented investigations draw attention to a low number of this species on healthy and infected leaves on the natural site in Kołacznia in comparison with the arboreta in Bolestraszyce and Rogów. This situation may be due to vicinity of other species from Ericaceae family – a reservoir of pathogenic fungi (Kowalik, Sagan 2005; Kowalik, Wandzel 2005).

Isolation of almost twice larger number of fungi colonies and species from prematurely fallen leaves, in comparison with healthy leaves, evidences an intensified

colonization and damaging of leaf blade by various fungi species. Species, which earlier caused leaf necroses, among others *E. nigrum*, *A. alternata*, *S. fimicola*, *Ph. cyclaminis* i *B. cinerea* played the main role. These fungi constituted almost 60% of the identified isolates. Equally big share of *A. alternata*, *E. nigrum*, *S. fimicola* and P. *expansum* was registered in previous papers by Kowalik et al. (2011, 2012). Presence of fungi from *Actinomucor*, *Mucor*, *Rhizopus*, *Trichoderma*, *Mortierella* and *Umbelopsis* genera on fallen leaves may be connected with the plant age, represented by senile specimens with rotten, moist shoots.

Presence of over a dozen fungi species of between once and thrice frequency is noticeable in the fungi communities isolated from fallen leaves, which may evidence an accidental colonizing of fallen leaves by the species living in soil.

Isolation of pathogenic *Fusarium*, *Giberella*, *Ilionectria*, *Phoma* fungi from infected and fallen pontic azalea leaves was corroborated in the literature of the subject (Kita, Mazurek 2003; Kowalik, Muras 2007; Kowalik 2008; Kowalik et al. 2010a; 2010b; 2011; 2012).

A comparison of *P. sydowiana* (syn. *Pestalotia sydowiana*) and *Truncatella truncata* (syn. *Pestalotia truncata*) in causing necrotic symptoms on the leaves of azalea and evergreen rhododendron leaves (Kowalik 2008, 2009; Kowalik et al. 2010a, 2010b, 2012) revealed that these fungi were more often colonizing healthy leaves of pontic azalea, than infected or fallen leaves.

The highest percentage differences between similarity indices for fallen azalea leaves evidence a considerable diversity of mycobiota colonizing this kind of leaves.

## CONCLUSIONS

1. Fungi communities existing in pontic azalea phyllosphere on the natural site in the Kołacznia nature reserve and on the collection of the Arboretum and Department of Physiography in Bolestraszyce, as well as on the Warsaw University of Life Sciences (SGGW) Arboretum collection in Rogów differed with the species composition and number of colonies.
2. The species whose colonies were isolated in largest numbers from healthy leaves of pontic azalea comprised: *A. alternata*, *Ph. cyclaminis*, *E. nigrum*, *Ph. medicaginis* and *B. cinerea*, from infected leaves: *A. alternata*, *E. nigrum*, *Ph. cyclaminis*, *S. fimicola*, *T. viride* and *A. phaeospermum,* and from fallen leaves: *E. nigrum*, *A. alternata*, *S. fimicola*, *Ph. cyclaminis* and *B. cinerea*, indicating that majority of them persistently colonized and damage leaves during vegetation period leading to their premature falling.
3. Colonization of pontic azalea phyllosphere in the arboreta at Bolestraszyce and Rogów by a much higher number of fungi species and colonies than in conditions of the natural site in Kołacznia evidences their intensified pressure in the arboreta environment.

REFERENCES

Anioł-Kwiatkowska J. 2003. Wielojęzyczny Słownik Florystyczny. Wydawnictwo Uniwersytetu Wrocławskiego.

Domsch K.H., Gams W., Anderson T.H. 1980. Compendium of Soil Fungi. Acad. Press. London, New York, Toronto, Sydney, San Francisco.

Ellis M.B., Ellis J.P. 1987. Microfungi on Land Plants. Croom Helm. London, Sydney.

Guba E.F. 1961. Monograph of Monochaetia and Pestalotia. Harvard Univ. Press. Cambridge, Mass.

Index Fungorum 2012. (www.index fungorum.org), access 15.12.2012.

Kaźmierczakowa R., Zarzycki K. 2001. Polska Czerwona Księga Roślin. Paprotniki i rośliny kwiatowe. Instytut Botaniki im. W. Szafera PAN, Kraków: 281-283.

Kirk P.M., Cannon P.F., Minter D.W., Stalpers J.A. 2008. Ainsworth & Bisby's Dictionary of the Fungi.10th Edition. CABI, Wallingford, UK.

Kita W., Mazurek J., 2003. Skład gatunkowy fyllosfery różanecznika w Ogrodzie Botanicznym we Wrocławiu i w Arboretum w Wojsławicach. Erica Polonica 14: 25-36.

Kowalik M. 1993. Grzyby gleby inicjalnej industroziemnej rekultywowanego w kierunku rolnym i leśnym zwałowiska Kopalni Siarki "Machów". Zesz. Nauk. AR w Krakowie, Rozpr. hab. 180: 1-78.

Kowalik M. 2008. Fungi and fungi-like Oomycetes isolated from affected leaves of rhododendron. Acta Mycol. 43 (1): 21-27.

Kowalik M. 2009. Bioróżnorodność grzybów występujących w fyllosferze różanecznika zimozielonego Rhododendron L. Zesz. Probl. Post. Nauk Roln. 539: 341-348.

Kowalik M., Kierpiec B., Bonio J., Żołna M. 2011. Fungi inhabiting spots and necroses on the leaves of azaleas (Rhododendron) in the Botanical Garden of the Jagiellonian University. Phytopathologia 62: 41-48.

Kowalik M., Kierpiec B., Żołna M. 2012. Fungi living at the fallen leaves of rhododendron and azalea (Rhododendron L.). Acta Sci. Pol., Hortorum Cultus 11 (2): 161-166.

Kowalik M., Muras P. 2007. Grzyby zasiedlające opadłe liście różanecznika. Rocz. AR w Poznaniu 383. Ogrodnictwo 41: 69-73.

Kowalik M., Muras P., Kierpiec B., Żołna M. 2010a. Zdrowotność liści różaneczników zawsze zielonych Rhododendron L. Zesz. Probl. Post. Nauk Roln. 551: 117-123.

Kowalik M., Muras P., Żołna M., Kierpiec B. 2010b. Grzyby wyosobnione z nekrotycznych plam na liściach różaneczników zawsze zielonych Rhododendron L. Zesz. Probl. Post. Nauk Roln. 554: 49-55.

Kowalik M., Sagan A. 2005. Fungi causing dying out heather in permanent plantings. Acta Mycol. 40 (2): 191-195.

Kowalik M., Wandzel A. 2005. Grzyby powodujące zamieranie sadzonek wrzosu. Acta Agrobotanica 58 (2): 237-242.

Piórecki J., Dubiel E. 2009. Volynian Polesia – main source of the Yellow Azalea (*Rhododendron luteum* Sweet) in European garden and parks. Rocznik Polskiego Towarzystwa Dendrologicznego 57: 29-32.

Piórecki J., Zarzycki K. 2010. Arboretum Bolestraszyce. Przewodnik. Bolestraszyce.

Rifai M. A. 1987. A revision of genus *Trichoderma*. Mycol. Papers 116: 1-56.

Sutton B. C. 1980. The Coelomycetes. CMI, Kew, Surrey.

# Różnorodność grzybów zasiedlających i uszkadzających liście azalii pontyjskiej *Azalea pontica*


## Streszczenie

Obserwacje stanu zdrowotnego azalii pontyjskiej *Azalea pontica* przeprowadzono w 2011 roku na stanowisku naturalnym w rezerwacie Kołacznia, w kolekcji Arboretum i Zakładu Fizjografii w Bolestraszycach oraz w kolekcji Arboretum SGGW w Rogowie. Celem badań była weryfikacja gatunków grzybów zasiedlających fyllosferę azalii pontyjskiej oraz porównanie składu gatunkowego grzybów na naturalnym stanowisku i w arboretach.

Analizie mykologicznej poddano liście zdrowe, porażone i opadłe. Stwierdzono, że zbiorowiska grzybów bytujących w fyllosferze azalii pontyjskiej na stanowisku naturalnym i w arboretach różniły się składem gatunkowym i liczbą kolonii. Gatunkami wyodrębnionymi w największej liczbie kolonii ze zdrowych liści azalii pontyjskiej były: *A. alternata*, *Ph. cyclaminis*, *E. nigrum*, *Ph. medicaginis* i *B. cinera*, z liści porażonych: *A. alternata*, *E. nigrum, Ph. cyclaminis*, *S. fimicola*, *T. viride* i *A. phaeospermum*, a z liści opadłych: *E. nigrum*, *A. alternata*, *S. fimicola*, *Ph. cyclaminis* i *B. cinerea*, co wskazuje, że większość z nich w okresie wegetacji stale zasiedla liście i uszkadza je, co prowadzi do ich przedwczesnego opadania. Zasiedlenie porażonych liści azalii w arboretach przez porównywalną liczbę kolonii i gatunków grzybów (w tym patogenów), znacznie większą niż w rezerwacie, może świadczyć o wpływie sąsiadujących roślin żywicielskich na ich stan zdrowotny. Kolonizacja fyllosfery azalii pontyjskiej w arboretach w Bolestraszycach i Rogowie przez dużo większą liczbę kolonii i gatunków grzybów, niż w warunkach naturalnego stanowiska w Kołacznii, świadczy o wzmożonej presji grzybów w środowiskach arboretów.