Powdery Mildew on Spinach (*Spinacia oleracea* L.)

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SUMMARY

Symptoms resembling powdery mildew appeared on spinach crops during April and May in 2007. Infected plants have suppressed growth, smaller and degenerated young leaves. The affected leaves become yellowish and wilt in a short time. High temperatures and draught conditions cause drying out of the affected plants. The symptoms are similar to physiological degeneration but are found on single plants or on small groups of plants. When plants are carefully examined, fine, exogenic mycelium is found on the leave blades. The mycelium can be clearly seen close to the main veins where it becomes denser and forms mycelium patches. On the leaf and flower stalks and plant stems the mycelium is white and fine at the beginning, and later becomes grayish.

Under microscope analysis ectophytic mycelia of exogenic origin and short chains of spores are observed. On short conidiophores, chains with two types of conidia are formed: macro conidia that are one-celled, colorless, thin-walled, elliptical to cylindrical, sized 24.5-28.4 x 17.5 μ m; and micro conidia – ovoid to elliptical, sized 10.4-14.1 x 7.7 μ m. Teleomorphs are found in groups or as a single structure mainly close to the leaf veins. They are roundish and have appendages with uncinate-circinate to helicoids apex, sized - 87.5-150 μ m. Cleistothecia have 4-5 to 8 asci (68.0 x 38.0 μ m), with 4 to 8 elliptical ascospores, sized 15.5-22.0 x 11.0-17.5 μ m. The causal agent of powdery mildew on spinach found in Bulgaria has been identified as *Uncinula* spp. (*Sawadaea* spp.), *Euoidium* type anamorph, subspecies *spinaciae*.

Keywords: Euoidium; Uncinula; Powdery mildew; Spinach

INTRODUCTION

Fungal plant diseases known as powdery mildews have been reported for a long time in the literature (Linnaeus, 1753, quoted by Belanger et al., 2002). The classical researches of this group of diseases started after publishing the fundamental works of Leveille (1851) on the taxonomy of powdery mildews (Braun et al., 2002). Evolutionary, this group of fungi invades new ecological niché and host plants (Hirata, 1966; Gorlenko, 1983; Amano, 1986). Their systematic structure is constantly complicating. New genera and species are appearing in the Family Erysiphaceae (Braun et al., 2002). This phenomenon can be explained with the adaptation to the constantly changing ecological conditions. For example, genus Leveillula originates from the genus Erysiphe, as the development of these fungi is connected with adapting to hot and dry conditions. In connection with the environment the evolution is seen not only in the teleomorph but also in the anamorph-conidia stage. The latter (anamorph) has clearly expressed species specificity (Braun, 1980). The structure of the conidia apparatus depends on the climate, where different fungal species develop. The single conidium coming through the stomata, are formed in fungal pathogens, living in areas with dry and hot climate (Gorlenko, 1983). The ecological conditions where plant species grow can probably explain the appearance of new phytopathogens from different genera of the family Erysiphaceae, adapted to live parasitically on new host plants.

During the first half of 20th century more than 200 fungal species of family *Erysiphaceae* were reported. They parasitize more than 1500 different plants – cultivated and wild, field, fruit and wood species (Atanasoff, 1934). Fakirova (1991) described 104 species and 4 varieties that belong to 10 genera of the family *Erysiphaceae*. 95 species out of 104, and 4 varieties are registered in Bulgaria, where 28 pathogens were reported for the first time in the country (Fakirova, 1991). The pathogenic fungi develop on 432 higher plants, and 201 of them are new host plants in Bulgaria. Probably, the great plant diversity in Bulgaria, favorable ecological conditions and high variability of the pathogens cause peculiar "geographical" specialization (Vasjagina et al., 1961).

The aim of the study was to describe the symptoms and to identify the causal agent of the disease.

MATERIAL AND METHODS

The research was carried out during 2007-2010 at the Department of Phytopathology, at the Agricultural University of Plovdiv. Pathogen identification was done based on the disease symptoms and on morphological characteristics of teleomorph and anamorph "which in high degree describe the species specificity (specific characters) of the pathogen" (Braun, 1980; Gorlenko, 1983; Fakirova, 1991; Braun et al., 2002). The description of symptoms was done on the naturally infected plants, from cotyledon stage to seed formation and ripening, and on artificially inoculated spinach plants.

Following treatments were examined in a way to discover the first appearance of symptoms, to follow the development of powdery mildew, as well as the formation of the pathogens structures (anamorph and teleomorph):

- Field experiments in 2008 and 2009: spinach seeds were sown in areas (two replicates, 25m² each) infected with powdery mildew during the previous year (2007 or 2008), with extra introduced inoculum from cut powdery mildew infected spinach leaves, flowering stalks and stems.
- Micro experiments in laboratory (2008-2010): spinach seeds were sown in river sand and sterile peat mixture, 1:1 (company Diko-2004) in plastic containers, sized 50 x 30 x 20 cm, in four replicates. The substrate was inoculated by mixing with cut spinach leaves, flowering stalks and stems infected by powdery mildew.
- Control plants: Spinach seeds were sown in a sterile peat and sand mixture, 1:1, without inoculum introduction in containers (in field and in laboratory).

RESULTS AND DISCUSSION

During April-May 2007 in the spinach (Spinacia oleracea L.) crops plants with suppressed growth, smaller and degenerated young leaves were found. In a very short time the affected leaves become yellowish and wilt, and when high temperatures and draught occur in the following period, the plants dry out. The reported symptoms resemble physiological degeneration, but differ from them because they do not affect the whole crop. After careful examination, fine, exogenic, cobweb mycelium is found on the leaf blades (Figure 1). The mycelium can be clearly seen close to main veins where it becomes thicker and forms mycelium patches. On the leaf and flower stalks, and the plant stems the mycelium is white, glossy and fine at the beginning, and later becomes gray. The teleomorphs of the pathogen are formed on the infected organs.

Under the microscope ectophytic mycelia with exogenic origin conidiophores is discovered. On the hypha, on the short conidiophores, chains of two types of conidia are formed: macro - and micro-conidia.



Figure 1. Exogenic, cobweb mycelium of powdery mildew on the upper leaf surface

With the development of the disease, mainly close to the leaf veins, the pathogens cleistothecia are formed. They are found as single structures or in groups, and have filiform appendages hooked at the end.

A new type of powdery mildew on spinach caused by *Oidiopsis haplophylli* was reported in the literature



Figure 2. Micro conidia of Euoidium (Sawadaea)

(Park et al., 2008). This fungus is related to *Leveillu-la* spp., whose representatives have conidiophores of endogenic origin with one conidium on the top (type *Pseudoidium*) (Fakirova, 1991; Braun et al., 2002). For this species teleomorph has not been discovered.

The research shows that the observed symptoms in Bulgaria differ from the ones described in the literature (Park et al., 2008). There are also differences in morphological characteristics of the anamorph stage. The pathogen in our country forms short conidiophores with micro- and macro- conidia in chains, which belong to type *Euoidium* (genus *Sawadaea*) (Braun, 1981; Braun et al., 2002) (Figures 2, 3).



Figure 3. Macro conidia of Euoidium (Sawadaea)

The macro conidia have an elliptical to cylindrical form, sized 24.5-28.4 x 17.5 μ m, while the micro conidia are oval to elliptical, 10.4-14.1 x 7.7 μ m.

The teleomorphs are roundish, dark brown with straight appendages hooked at the end. They are formed under unfavorable conditions – dry weather and high temperatures (dry and hot weather). They contain 4-5-8 asci (68.0 x 38.0 μ m), with 4-5 to 6-8 elliptical ascospores, 15.5-22.0 x 11.0-17.5 μ m.

The observed symptoms and morphological characteristics correspond to the described ones for the pathogens of the genus *Euoidium* (genus *Sawadaea*) with two spore types-macro- and micro- conidia. The species *Sawadaea bicornis* (syn. *Uncinula bicornis*) and *Sawadaea tulasnei* (syn. *Uncinula tulasnei*) are reported in the literature (Fakirova, 1991; Braun et al., 2002).

Based on the research results it can be accepted that spinach powdery mildew that appeared in Bulgaria is caused by *Uncinula* spp. (*Sawadaea* spp.), an anamorph type *Euoidium*, subspecies *spinaciae*. According to Mori et al. (2000) and Takamatsu et al. (1998) the genus *Sawadaea* differs from genera *Cystotheca* and *Podosphaera*, and is characterized with poliascal ascomata with clearly formed appendages, with uncinate-circinate apex. It also has a different type of a conidial stage, belonging to genus *Oidium* subgen. *Octagoidium*.

The observations on the development of the teleomorph stage show that cleistothecia formation develops mainly in mycelia patches, close to main veins. They are formed in large numbers on the flower stalks, too. Formed asci and ripen ascospores are found at the end of spinach vegetation period.

The results obtained in the field and micro experiments are also in agreement with these data. Under laboratory conditions the first symptoms are seen on cotyledon leaves, which become yellowish, necrotic and finally drop out. Typical symptoms are discovered at the stage of 2nd to 3rd true leaf. On a large scale powdery mildew mainly appears on young, still growing leaves, as well as on the flowering stems.

CONCLUSIONS

Based on the research results, the following conclusions can be made:

- Powdery mildew discovered on spinach (Spinacia oleracea L.) in Bulgaria is caused by Uncinula spp. (Sawadaea spp.), Euoidium type anamorph probably subspecies spinaciae.
- The pathogen is preserved in plant debris in the soil as a teleomorph and perhaps in seeds.

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Pepelnica na spanaću (Spinacia oleracea L.)

U oktobru i maju 2007. godine primećeni su simptomi na biljkama spanaća koji podsećaju na peplnicu. Inficirane biljke imaju slabiji porast, male i degenerisane mlade listove. Zahvaćeni listovi žute i za kratko vreme venu. Visoke temperature i suše dovode do sušenja obolelih biljaka. Simptomi su slični degenerativnim promenama fiziološkog porekla, ali se javljaju ili na pojedinačnim biljkama ili na manjim grupama biljaka. Prilikom pažljivog pregleda biljaka uočava se nežna, egzogena micelija na obodu lista. Micelija je jasno uočljiva u blizini glavnih lisnih nerava gde postaje gušća i formira spletove. Na lisnim i cvetnim drškama i stabljici micelija je u početku nežna i bela, a kasnije dobija sivkastu boju. Pod mikroskopom se uočava ektofitna micelija egzogenog porekla i spore u kratkim nizovima. Na kratkim konidioforama formiraju se nizovi dva tipa konidija: jednoćelijske, bezbojne, eliptične do cilindrične makrokonidije tankih zidova, veličine 24,5-28,4 x 17,5 μm; i mikrokonidije – jajaste do eliptične, veličine 10,4-14,1 x 7,7 µm. Teleomorfne strukture se pronalaze u grupama ili pojedinačno, uglavnom u blizini lisnih nerava. Okruglastog su oblika i imaju apendicese sa kukastim-prstenastim ili helikoidnim vrhom, veličine 87,5-150 µm. Kleistotecije imaju 4-5 do 8 askusa (68,0 x 38,0 μm) sa 4 do 8 elipsoidnih askospora, veličine 15,5-22,0 x 11,0-17,5 µm. Prouzrokovač pepelnice spanaća pronađen u Bugarskoj identifikovan je kao Uncinula spp. (Sawadaea spp.), bespolni stadijum tipa Euoidium, podvrsta spinaciae.

Ključne reči: Euoidium; Uncinula; pepelnica; spanać