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INFLUENCE OF SOOTY BLOTCH AND FLYSPECK (SBFS) FUNGI ON APPLE FRUITS DURING STORAGE

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Abstract. Sooty blotch and flyspeck is a fungal disease of complex etiology. SBFS fungi blemish the apple fruits surface reducing their market value. The aim of the study was to investigate the effect of fungi causing sooty blotch and flyspeck on apple during fruit storage. The experiments were conducted in 2007–2010. Fruits were collected from the orchard in Warsaw-Wilanów. Mass reduction in heavily and slightly infected or healthy apples per gram fresh weight of fruit was compared. Changes in quality of apples (wilting, wrinkling) were also observed during storage. Over a dozen to several dozen percent in reduction of the weight of heavily infested stored fruit as compared to a loss of fruit weight only slightly affected or healthy was found. It was also observed a wilting and wrinkling of the skin of heavily infested fruit.

Key words: *Microcyclosporella* sp., quality of apples, weight loss

INTRODUCTION

Sooty blotch is an apple disease caused by a complex of fungi [Grabowski 2004, Batzer et al. 2005, Frank et al. 2010, Ivanović et al. 2010]. The SBFS fungi colonize the epicuticular wax layer of apple fruit [Belding 2000]. Symptoms of the disease appear as sooty blotches with an indefinite outline on the surface of fruits. The term "sooty blotch" designates fungi that form dark mycelial mat with or without sclerotium-like bodies. Mycelial mats with sclerotium-like bodies varied in colour, density and margin type. Colour of sooty blotch mycelial mats with no sclerotium-like bodies ranged from pale olive to black [Batzer et al. 2005]. The "flyspeck" denotes colonies which develop clusters of shiny, black, round to oval, sclerotium like bodies and have no visible mycelial mat [Batzer et al. 2008]. SBFS fungi may utilize exuded nutrients present on the apple surface [Wrona and Gleason 2005]. The disease was initially attributed to the single fungus *Dothidea pomigena* Schw. [Baines and Gardner 1932]. The name of sooty

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blotch fungi was subsequently changed until Colby established a new genus *Gloeodes* and named the fungus causing sooty blotch *Gloeodes pomigena* (Schw.) Colby [Colby 1920 cited by Williamson and Sutton 2000]

This fungus was cited in the literature as the only causal agent of sooty blotch until 1998 when Johanson et al. [1997] showed that sooty blotch in the United States is a disease of complex etiology caused by three different fungi: *Peltaster fructicola* Eric M. Johnson, T. B. Sutton et Hodges, *Leptodontidium elatius* (Mangenot) de Hoog and *Geastrumia polystigmatis* Batista & M.L. Farr [Williamson and Sutton 2000]. In the past ten years the sooty blotch disease complex has been further expanded to include more than 30 species based on morphological characters and molecular tools [Batzer et al. 2005, Mirzwa-Mróz 2008, Frank et al. 2010, Ivanović et al. 2010].

In Poland the sooty blotch symptoms for the first time were observed in 1934 year by Knothe on apples from Lublin province. In 1937 *Gloeodes pomigena* fungus was found in Warsaw on marketing apples. Diseased fruits were collected from various regions of Poland [Siemaszko 1937]. Next studies conducted in southern Poland also showed that this disease is caused by different fungi. The most important of these include: *Tripospermum myrti* (Lind) S. Hughes, *Aureobasidium pullulans* (de Bary et Löwenthal) G. Arnaud, *Cladosporium cladosporioides* (Fres.) de Vries and *Tripospermum camelopardus* Ingold, Dann & P.J. McDougall [Grabowski 2004, 2007, Wrona and Grabowski 2004]. According to Grabowski [2007] 1.31 percent of all causal agents in southern Poland are the fungi of the genus *Miclocyclosporella* (formerly assigned as *Pseudocercosporella* sp. [Frank et al. 2010] while in Serbia and the USA they constitute almost 78% of all causal agents of sooty blotch [Ivanović et al. 2010].

The aim of the study was to examine the impact of SBFS fungi on apple during storage. Loss per unit weight of heavily affected apples in relation to the loss per unit weight of slightly affected fruits was examine in details. So far this kind of research was not conducted.

MATERIAL AND METHODS

The experiments were carried out during 2007–2010 in orchard at Warszawa-Wilanów (52°9'26"N, 21°6'19"E) (Mazovia province). The trees in the plot were spaced 3m by 1m. Each tree came from seed obtained in selections of Vf scab resistance program. Hand crossings were performed in the years 1995 to 1997 and seedlings were selected for scab resistance then grafted on M9 rootstock. Some tree prunings were done occasionally and none fungicides were applied. Insecticides against apple blossom weevil (*Anthonomus pomorum*) were used in some years. Apple fruits were harvested from actually fruiting trees in the first decade of October. Immediately after harvest in laboratory assessments of sooty blotch disease was done. The surface of the fruits covering by SBFS fungi was estimated using a key drown according to the following scale: 0 – no visible symptoms; 1 – up to 1%; 2 – from 1% to 5%; 3 – from 5% to 20%; 4 – from 20% to 50%; 5 over 50%. Calculations of fruit surface cover by SBFS mycelium were caried out using mean percentage value of each scale grade expressed in decimal fraction. For calculations the mean of disease level taken from two sides of each fruit was used. Apples were also weighed fortnightly for about four months. Fruits were stored in

a camera with temperature 10–15°C. Additionally in the year 2007 cold store was also used. In this store high humidity (85–90% RH) and temperature 2–4°C were kept. Once a week roting fruits were removed. Comparison of means were done using t-test in Statgraphics Plus 4.1 program. Relative fruit weight decrease $mg \cdot g^{-1}$ was calcutated using Microsoft Excel version 5.1.2600.

RESULTS AND DISCUSSION

In our studies on the surface of cuticle of apple fruits fungal colonies were olive-green to black in colour and ranged in shape from nearly circular to rather large amorphous spots. Sometimes blotches coalesce to cover much of the fruit surface. Most of the spots present on apples were with sclerotium-like bodies similar to described by Batzer et al. [2005] in the United States.

In this experiment blotches on apples surface rapidly darkened in store but their enlargement was not observed. It is in contrary to results of Williamson and Sutton [2000], where mycelium development in store was noted. It might be explained by differences in etiology of sooty blotch. The disease is not a problem in Polish orchards with full chemical control of apple scab. Although may be dangerous in ecological orchards and on scab resistant cultivars [Grabowski 1999, Pitera 2000].

However, in countries with favorable conditions for disease development, apples with SBFS symptoms are unacceptable to consumers because the mycelium blemish the fruits. In the United States the market value of the crop coming from orchard even with full SBFS chemical protection could be reduced by 5–10% [Williamson and Sutton 2000, Batzer et al. 2002].

Another kind of sooty blotch disease harm is wilting and wrinkling of skin of affected apples compared with healthy fruits during storage which was observed in this experiment (fig. 1).

Similar phenomenon was observed only by Godec [pers. comm. cited by Frank et al. 2010] in Slovenia. In the literature there is a lack of information concerning the impact of fungi on fruit storage.

In this work all apple fruits lost gradually their weight both in cold and cool camera storage (tab. 1)

The decrease was the highest in the first weeks of storage. Fruits heavily covered with sooty blotch mycelium (from about 13% to about 18% of skin surface) showed bigger relative fruit weight loss than fruits with slight cover or lack of the fungi. In each year statistically significant differences in relative weight loss between heavily affected fruits and fruits with none or small mycelium cover were observed. In cool camera the difference ranged from about 15% in the season 2007/2008 to over 28% in 2009/2010. In profesional storage conditions (cold camera) the difference reached only 13.6%. The biggest difference between relative weight loss of slightly and heavily affected apples was observed in 2009/2010. The smallest difference between relative weight loss of slightly and heavily affected apples was observed in 2007/2008 in cold storage. In seasons 2007/2008 and 2008/2009 the differences were intermedium. Storage of fruits at higher temperatures (cool camera) better reveal the impact of the disease on weight loss.

Fig. 1. Wilting effect of sooty blotch fungi on apple fruit in comparison with mycelium free skin of fruit

Rys. 1. Efekt więdnięcia owoców pokrytych grzybnią grzybów kompleksu brudnej plamistości w porównaniu z kontrolą

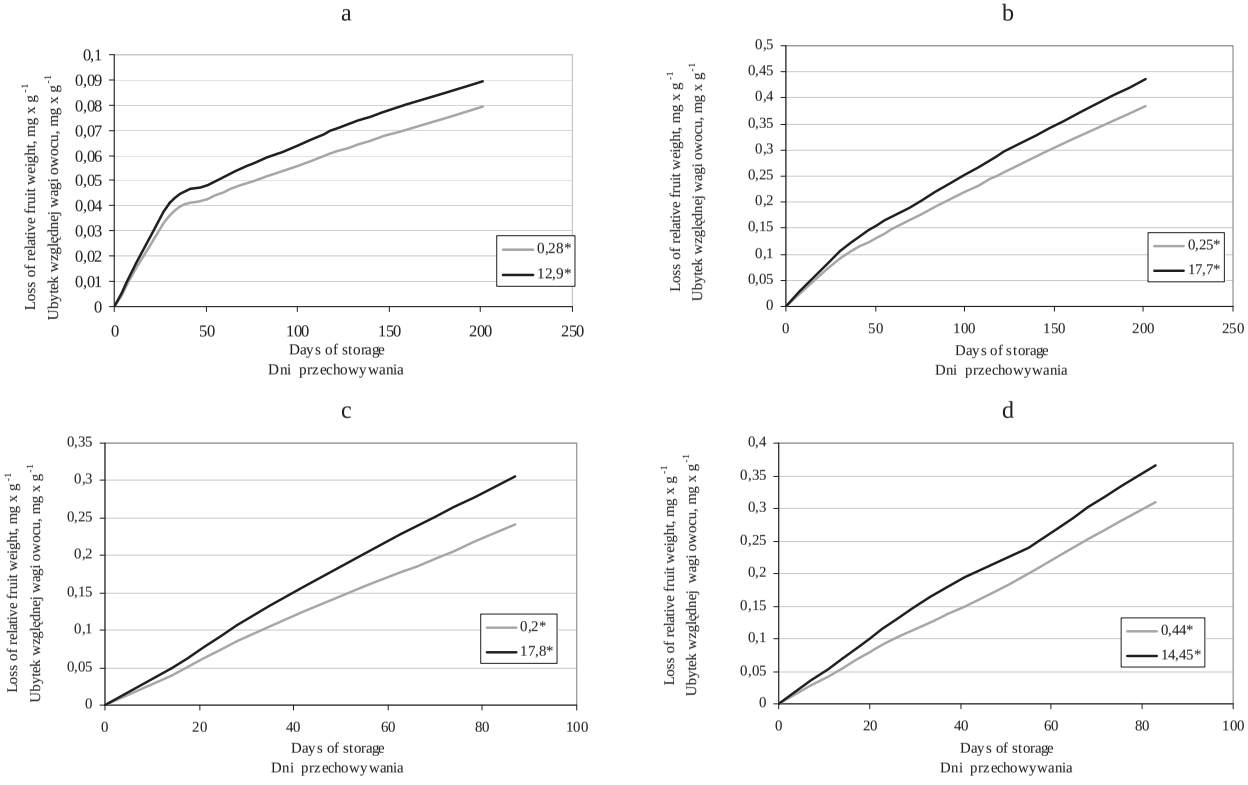
Table 1. Influence of sooty blotch on loss of apple fruit fresh weigh during storage

Tabela 1. Wpływ brudnej plamistości jabłek na zmniejszenie masy owoców w czasie przechowywania

Storage condition and year of observations Warunki przechowywania owoców i rok obserwacji	Cold camera Chłodnia		Cool camera Chłodne pomieszczenie					
Storage season Sezon przechowywania	2007/2008		2007/2008		2008/2009		2009/2010	
Number of fruits Liczba owoców	69	62	61	67	125	192	66	179
Percent of fruit surface covered by the mycelium Procent powierzchni owocu pokrytej przez grzybnię	0.28	12.9	0.25	17.7	0.2	17.8	0.44	14.45
Relative fruit weigh loss mg · g^{-1} Względna utrata wagi owoców mg · g^{-1}	36.1a*	41.0b	91.4a	105.2b	85.4a	107.2b	101.4a	129.9b

*Letters a and b in each year of observation indicate statistically significant differences according to t-test at 95% confidence level

*Litery a i b w każdym roku obserwacji oznaczają statystycznie istotne różnice według testu-t przy poziomie istotności 95%



*Percent of fruit surface covered by the mycelium of SBFS fungi
*Procent powierzchni owocu pokrytej przez grzybnię sprawców brudnej plamistości jabłek

Fig. 2. Comparison of relative fresh weight loss in years: 2007 – a (cold camera), b – (cool camera), 2008 – c, 2009 – d
Rys. 2. Suma ubytków względnej wagi przechowywanych owoców w latach: 2007 – a (chłodnia), b – (chłodne pomieszczenie), 2008 – c, 2009 – d

The graphes (fig. 2a, b, c, d) show sum of relative weight loss during storage of fruits slightly covered by SBFS fungi and apples with much bigger mycelium presence. The loss grew up visibly in time in all casses but low affected fruits lost they relative weight slower.

According to available literature it is known that fungi of the SBFS complex grow superficially on the epicuticular wax of the fruit but penetration through the cuticle into the epidermal cells does not occur [Baines and Gardner 1932, Belding et al. 2000, Johanson et al. 1996, Siemaszko 1937]. However Groves [1933] has found such sooty blotch fungi that penetrated the cuticle or even beneth.

In this work similarly as in the USA, the Balkans, Germany and Southern Poland [Batzer 2005, Díaz Arias et al. 2010, Frank et al. 2010, Grabowski 2007, Ivanović et al. 2010] fungi from genera *Microcyclosporella* have been causal agents of sooty blotch in recent years. According to Mirzwa-Mróz [2008] and further works [unpublished data] it might be expected that isolates of *Microcyclosporella* sp. predominated in population of SBFS fungi at the Warsaw-Wilanów orchard. It was also found [publishing in preparation] that *Microcyclosporella* sp. may penetrate and stratify cuticule. This activity of *Microcyclosporella* sp. may also indicate on one of the causes of fruit wilting during storage.

CONCLUSIONS

1. Fruits with heavy cover of SBFS fungi faster loose their relative weight than healthy ones.

2. Statistically significant differences between the loss of relative weight of disasesed and healthy fruits were the biggest in the first weeks of apple storage

3. Heavy covered fruits with SBFS fungi leads to earlier shriveling and wrinkling of fruits skin.

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WPŁYW KOMPLEKSU GRZYBÓW POWODUJĄCYCH BRUDNĄ I KROPKOWANĄ PLAMISTOŚĆ JABŁEK NA PRZECHOWYWANIE OWOCÓW

Streszczenie. Brudna plamistość jabłek jest chorobą o złożonej etiologii. Grzyby powodujące tę chorobę rozwijają się na powierzchni skórki jabłek powodując oszpecenie owoców i obniżenie ich wartości handlowej. Celem pracy było zbadanie wpływu grzybów powodujących brudną plamistość jabłek na przechowywanie owoców. Doświadczenia przeprowadzono w latach 2007–2010. Owoce pochodziły z sadu w Wilanowie (dzielnica Warszawy. Porównywano zmniejszanie się masy silnie i słabo porażonych lub zdrowych

jabłek w przeliczeniu na gram świeżej masy owoców. Obserwowano również zmiany jakości jabłek (więdnięcie, marszczenie się) w trakcie przechowywania. Stwierdzono kilkunasto- do kilkudziesięcioprocentowe zmniejszanie się masy przechowywanych owoców silnie porażonych w porównaniu do utraty masy owoców słabo porażonych i zdrowych. Obserwowano również szybsze więdnięcie i marszczenie się skórki owoców silnie porażonych.

Słowa kluczowe: *Microcyclosporella* sp., szkodliwość, jakość jabłek

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