Influence of changed ecological conditions on occurrence of London plane (*Platanus* × *hispanica* Münchh.) anthracnose

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Abstract

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During 2004–2007, reappearances of anthracnose on *Platanus* × *hispanica* caused by the microscopical fungus *Apiognomonia veneta* were recorded. Causal agent of the disease was isolated from symptomatic leaves and twigs, with characteristic spots and lesions, sampled from affected host trees growing in urban environment at the selected locality (Nitra). Subsequent identification and morphological description of fungal isolates was made by microscopical differentiation, according to the fungi identification key. Our study has confirmed interannual changes in the disease severity and influence of temperature in period decisive for occurrence and progress of the disease. Cool springs are more promoting severe plane anthracnose outbreaks. Repeated annual removal of twigs and leaves results in weakening of plane trees.

Key words

anthracnose, *Apiognomonia venet*a, *Discula platani*, ecological conditions, *Platanus* × *hispanica*, urban environment

Introduction

At the moment, plane trees belong to woody plants seriusly attacked by the microscopic parasitic fungus *Apiognomonia veneta* (Sacc. & Speg.) Höhn. [syn. *Gnomonia platani* Kleb., *Gn. veneta* (Sacc. & Speg.) Kleb., *Gn. errabunda* (Rob.) Auersw. (Arx, 1970)] that cause the disease known under the name anthracnose. This fungus, an intercellulare parasite is reported to be the causal organism of plane-tree anthracnose disease (Peace, 1962). Anthracnose was first recorded on *Platanus occidentalis* L. in Britain in 1815 (Neely, 1976).

The asexual stage of *A. veneta* is *Gloeosporium platani* (Mont.) Oud. [syn. *Gl. nervisequum* (Fckl.) Sacc.] or *Discula platani* (Peck.) Sacc. (Klebahn, 1905). *Gl. platani* was previously recorded in New Zealand by Brien (1939) on *Platanus orientalis* L. and by Hitchcock (1977) on *Platanus* × *hispanica* Münchh. (syn. *P.* × *acerifolia* (Aiton) Willd, *P.* × *hybrida* Brot.) and *Platanus occidentalis* L. Dingley (1969) reported that *Gl. platani* is host-specific, occurring only on leaves of *Platanus* spp. and that it is common throughout New Zealand. By the end of the 19th century, it had been reported in France, Germany and the USA (Milne and Hudson, 1987). The disease is widespread in North and South America, Europe, Asia and Australia (Jurc, 2006). In Slovakia only sporadic information are available on occurrence, spread and harmfullnes of this fungal pathogen (Juhásová, 1988; Juhásová and Hrubík, 1998; Juhásová and Ivanová, 1999).

The plane tree in Slovakia has primarily been used as an ornamental tree in large urban parks, many streets, and it is regularly used in many other forms of landscaping where a large tree can be used.

It has many advantages as an ornamental tree, including large size and longevity where these are required. Other advantages include a moderate shade that is sufficient to allow grass or other plants to grow below it, tolerance of pollution, tolerance of city conditions, tolerance of difficult soil conditions, drought and also tolerance to pruning, bad pruning included.

Planes are native to regions with warm summers. Summer droughts do not seem to be a problem for established trees though they probably slow down their growth. Plane trees do not do so well in regions with cooler summers. The planes survive strong, hot summers well and, unlike other species in the same areas, show no desiccated shoot tips due to high temperatures. This is partly owing to the effects of plane tree anthracnose that is much worse in damper and cooler climatic conditions and can overwhelm some trees. The tree prefers neutral or slightly acid soils, and growth is less vigorous or slower in chalk soils.

Although anthracnose is most noticeable on sycamores and London planes, it also can be found on oaks, maples, ashes, some elm species and walnuts. In Slovakia, the frequent and serious disease is anthracnose on London plane trees.

The nomenclature of the pathogen causing anthracnose of *Platanus* has been the subject of a considerable confusion. According to Goidànich (1964), the causal agent of anthracnose of the plane tree is the fungus *Gnomonia veneta* (Sacc. et Speg.) Kleb., whose anamorphs are *Discula platani* (Peck.) Sacc., *Gloeosporium nervisequum* (Fkl.) Sacc., *Sporonema platani* Baüml., and *Microstoma platani* Eddelb. et Eng. Before Goidànich, Viennot–Bourgin (1949) cited the teleomorphs *Gnomonia* (*Apiognomonia*) *veneta* (Sacc. et Speg.) Kleb. and *Laestadia veneta* Sacc. et Speg., and the anamorphs *Gloeosporium nervisequum* (Fkl.) Sacc., *Gloeosporium platani* (Mont.) Oud., *Discula platani* (Peck.) Sacc., *Sporonema platani* Baüml. and *Microstoma platani* Eddelb. et Eng. Barr (1978) accepted that the *Platanus* and *Quercus* anthracnose fungi are morphologically and pathologically distinct and used the name *Apiognomonia veneta* (Sacc. et Speg.) Höhn. for the *Platanus* pathogen. Later, Sutton (1980) named this fungus *Apiognomonia errabunda* (Rob.) Höhn. and its anamorphs *Discula umbrinella* (Berk. et Br.) Sutton, *Gloeosporium umbrinellum* (Berk. et Br.) Petrak, and *Myxosporium platanicola* Ell. et Ev. Sutton explained that *A. errabunda* comprises the two species *Gnomonia platani* Kleb. and *G. quercina* Kleb. The sexual stage is included in the class Ascomycetes, family Gnomoniaceae (Tello et al., 2000).

Anthracnose is the common name given to a group of fungal pathogens which cause dark, usually sunken lesions. The disease cause tan to brown or black lesions on the leaves, branches, twigs, stems, flowers and fruits. Dieback of plane branches and necrotic lesions near the veins are typical symptoms. The most conspicuous symptom of the disease in early spring is dying twigs and new shoots. Small black fruiting structures of the fungus break through the dead bark of blighted, one-year-old shoots. Repeated removal of young twigs results in abnormal branching and gives the tree a ragged appearance. After the bud break, planes show scorching and wilting new shoots and leaves. Later, fully expanded leaves develop elongated tan to brown lesions parallel to the midrib and veins. This should not be confused with summer scorch of plane which causes burnt leaf margins. Infected leaves scorch and shed. In exceptionally cool, wet springs, plane trees leaf out and then can defoliate heavily.

Apiognomonia veneta (syn. *Apiognomonia errabunda*, *Gnomonia veneta*, *G. platani*) is a regular problem but it rarely causes more than minor damage to most trees. Leaves are disfigured, and shoots and twigs sometimes go away. Park ground can be covered with a thin layer of infected plane leaves or leaf litter from early June onwards. The disease is promoted by damp weather during shoot extension in spring and early summer. It is, or it used to be reduced by pollution in cities. The fungus overwinters in fallen leaves, and also on the tree bark. Anthracnose has different effects on various species and cultivars. *Platanus orientalis* is little affected or it is not affected at all, *P. occidentalis* is very seriously affected and can die (Swift, 2001).

Several cultural practices can reduce the severity of anthracnose. Removing dead leaves in autumn helps limit the amount of fungal inoculum present for infection of new leaves in the following spring. Fallen branches and leaves should be removed to reduce the severity of disease. They would produce fungus spores in the following spring if not destroyed (Tello et al., 2000, 2005; Svihra, 2003; Pscheidt, 2006). Proper tree spacing and placement promoting good air circulation reduces the number of hours when leaf surfaces remain wet, and decreases the likelihood of fungal infection.

In spring after the leaf emergence, it is profitable to fertilize trees suffering from repeated attacks of anthracnose, as evidenced by twig dieback and lack of vigor. Nitrogen fertilization may increase the tree‘s tolerance or resistance to anthracnose, but care should be taken to avoid overfertilization (Berry, 1985). Dry winters weaken trees, and increase the effects of diseases. To reduce this problem, the trees should be watered once a month during snowless winters. It is necessary to water when air temperatures are above the freezing point, early enough to allow the water to soak in the ground before nightfall (Swift, 2001; Svihra, 2003; Pscheidt, 2006).

Fungicide treatments are not generally recommended, but in urban trees it can sometimes be used. The sprays should be applied at the moment the buds begin to swell. During rainy springs, additional applications are needed at 7 to 14 days intervals until conditions for this disease are not favourable any longer. Thorough coverage and proper timing of the sprays are essential for an adequate control. Early sprays are critical for control. The second set of leaves is to be protected with fungicide sprays if cool, moist conditions take place. Fungicides registered for the control of plane anthracnose include chlorothalonil, thiabendazole, thiophanate-methyl, mancozeb and copper fungicides (cupric hydroxide) (Swift, 2001; Svihra, 2003; Nameth and Chatfield, 2007).

Treatment with Arbotect Macro-Infusion, ensures fungicidal protection from the inside of the tree as it coats small twigs and branches where the disease grows. One treatment is sufficient for 3 years (Leininger et al., 1999; Stipes, 2000; Swift, 2001). According to Pscheidt (2006), for spring application, Fungisol is recommended only.

The aim of this study was to assess the occurrence and spreading of the anthracnose fungus *Discula platani* (Peck) Sacc. [teleomorph *Apiognomonia veneta* (Sacc. & Speg.) Höhn.] on London plane (*Platanus* × *hispanica* Münchh.) planted in urban environment in relation to changing ecological conditions over the period 2004–2007.

Material and methods

Material, isolation and cultivation

The fungus *Apiognomonia veneta* on *Platanus* × *hispanica* was isolated from symptomatic leaf and twigs with characteristic spots and lesions sampled during the growing season (from April to August 2004–2006 and April to May 2007) from affected host trees growing in urban environment (street plantings) at the selected locality (Nitra). The material was sampled from lower parts of tree crowns. The age of the evaluated trees was from 20 to 150 years.

The samples were surface-sterilized in 70% ethanol, then immersed for 15–20 minutes in sodium hypochlorite (1% available chlorine), rinsed in sterile distilled water (2–3 times) and dried carefully with filter paper. After surface sterilization, the tissue samples were cut to small pieces (2–3 mm), placed on 1% malt extract agar (MA) (10 g l^{-1} Difco agar, 7 g l^{-1} malt extract) and subsequently incubated in Petri dishes at 24–25 ºC, in the dark. Pure fungal cultures were obtained after multiple purification.

Identification procedure

Subsequent identification of *Apiognomonia veneta* isolates was made by microscopical differentiation (Clinical microscope BX41, Olympus) according to the "Simplified Fungi Identification Key" by Woodward (2001) and morphological description by Arx (1957, 1970), Hitchcock and Cole (1978), Sutton (1980) and Tello et al. (2000).

Results and discussion

The causal agent of plane tree anthracnose is a stem colonising fungus *Apiognomonia veneta* (the asexual stage *Discula platani*, syn. *Gloeosporium platani*) that causes defoliation, twig dieback and ragged appearance in the attacked trees. The seriousness of this disease is dependent on the health state of the infected tree. Healthy trees can resist many years of defoliation, while trees in stressed conditions can experience severe dieback and decline.

Microscopical identification of *Gloeosporium* Desm. et Mont. isolates according to the "Simplified Fungi Identification Key" by Woodward (2001) confirmed the presence of conidia produced in an acervulus, spores that rupture through the host tissue at maturity. The conidia are one-celled, hyaline (colourless) or cream, or pink-coloured in mass, ovoid, cylindrical or slightly curved in shape.

Arx (1957) accepted *Discula quercina* as the correct name of the anamorphosis for *A. errabunda*, but unfortunately this is later homonym of *D. quercina*. According to this author, the next available epithet for this fungus is *Gloeosporium umbrinellum* as a valid synonym for *Discula umbrinella*, anamorph for *A. errabunda*. According to Arx (1970), it is difficult to find a name for the conidial state that is valid with accordance with the code of nomenclature.

Morphological description of the examined fungus is in accordance with description by Sutton (1980). The author, providing microscopical characteristic of *Discula* describes immersed, branched, septate, hyaline or pale brown mycelium, hyaline, aseptate, smooth, thin-walled, straight or slightly curved ellipsoid or clavate conidia with obtuse apex and truncated base. Conidiophores are hyaline, septate and branched only at the base, straight or curved to irregular, tapered towards the apices, formed from the upper cells of the conidiomata. The author described the conidial stage of the fungus as an acervular conidiomata (up to 150 µm in diameter), which is epidermal, separate, or confluent and with irregular dehiscence.

Tello et al. (2000) detected some differences in the conidiomata, depending on whether it is formed on the leaves or on the twigs or shoots, and being more open when formed on twigs or shoots. The conidiophores are hyaline, septate, and branched at the base only. The conidia are hyaline, aseptate, smooth, thin-walled, and straight or slightly curved; the apex is obtuse; the base is more or less truncate, ellipsoid, or clavate, with a size of 9.5 to 12 × 3.5 to 5 µm.

According to HITCHCOCK and COLE (1978), the conidia are irregular in shape elliptical, obpyriform or ovate to oblong, unicellular and hyaline, measuring 9–14 µm. Freshly prepared specimens show that these spores are biguttulate, ie contain two oil globules. *In vivo* conidia in our observation were oval to ellipsoid, hyaline, unicellular with hard wall, 10 × 4 µm in size on average, containing two oil drops (Fig. 1).

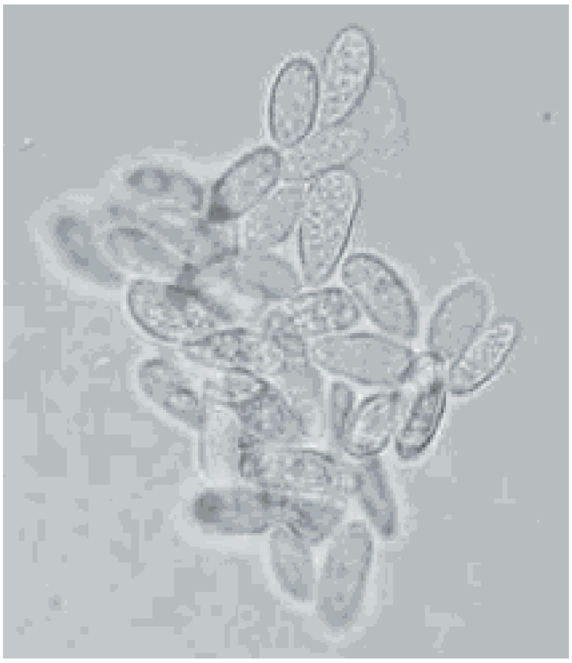

Fig. 1. Oval to ellipsoid conidia of *Discula platani* produced *in vivo*

According to HITCHCOCK and COLE (1978), isolation from diseased tissue is achieved by planting infected host tissue or spores produced in acervuli onto potato dextrose agar. In the culture, two types of hyphae can be distinguished measuring 1–2.5 µm and 3–6 µm, respectively. Conidia produced in the culture are slightly larger than those produced *in vivo* ie 10–15 × 4–6 µm and are produced terminally, singly or in basipetal succession from phialides (terminal portion of a hypha, from apex of which thin-walled conidia are abstracted) on the mycelium. Microconidia or secondary conidia not produced *in vivo* were formed in culture measuring approximately 5 × 1 µm, being pyriform in shape and are also produced from phialides. The mycelium is intercellular, in the leaves (VIENNOT–BOURGIN, 1949), immersed, branched, septate, and hyaline or pale brown (SUTTON, 1980). The results correlate with our observations, where mycelium after 4-days cultivation on MA medium was yellowy or tan (Fig. 2), later tawny in colour. According to HITCHCOCK and COLE (1978) the fungus produces a circular colony on potato dextrose agar consisting of zones of elevated mycelium alternating with regions of mycelium adpressed to the agar surface. The raised regions have exudate droplets within the aerial hyphae and hard dark structures, resembling microsclerotia measuring 100–200 µm in diameter.

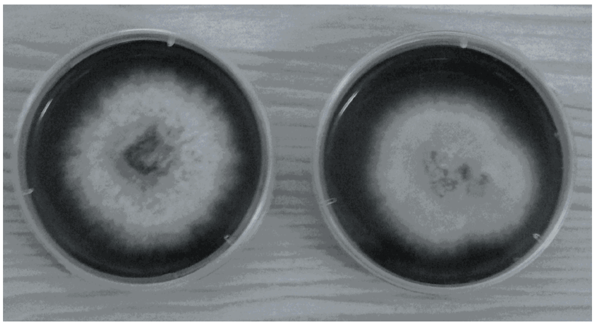

Fig. 2. Whitish to yellowy culture of *Discula platani* after 4-days cultivation on 2% malt extract agar

The sexual stage is characterized by globose, immerse, short-necked (130 to 400 µm) perithecia. Asci (40 to 55 × 9 to 13 µm) carry eight hyaline, two-celled, ovoid ascospores (12 to 16 × 4 to 6 µm) (VIENNOT-BOURGIN, 1949). Perithecia are immersed, non stromatic, globose or slighty flattened at base, 200–280 µm in diameter when mature. Apex of ascocarp is elongated into a protruding neck 80–125 µm long. Ascocarp wall is 20–40 µm thick, dark brown with a slightly roughened wall, double layered with the outer layer composed of thick walled, pigmented cells. Asci are subcylindrical or clavate, 40–62 × 12–15 µm unitunicate, apically thickened, inoperculate with a bright refractive ring surrounding the pore and containing eight spores. Ascospores are biseriate, hyaline, 10–20 × 4–6 µm, usually straight or slightly curved, arcuate to fusiform – elliptical, two celled with a septum 2–4 µm from the base. The upper larger cell from which the germ tube usually emerges has dense granular cytoplasmatic contents in fresh specimens, with one or two guttules (HITCHCOCK and COLE, 1978).

According to LEININGER et al. (1999), colourless ascospores, which have two unequal cells, are released in spring from black perithecia on fallen leaves. Conidia are produced in small cups on dead leaves in spring and in small (0.5 to 0.9 mm) black pycnidia in the bark of twigs.

The anthracnose fungi attack plane trees early in the spring, causing a rapid wilt of newly emerging leaves. This rapid wilting is frequently misidentified as frost damage. Larger, more mature leaves develop a brown growth along the main veins (Fig. 3), often in V-shaped patterns (TERRELL, 2004), (Fig. 4). Infected leaves often curl and eventually fall, littering the ground.

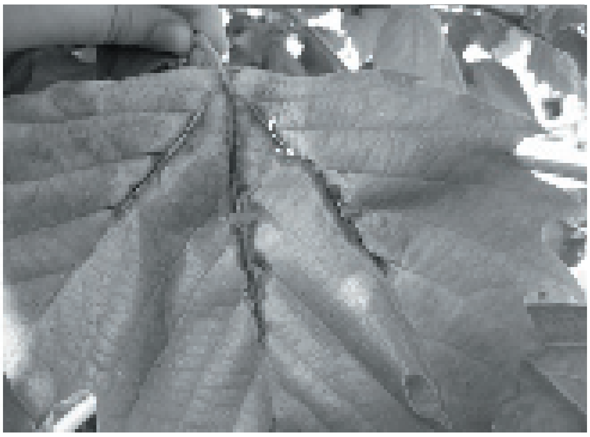

Fig. 3. Leaf blight of *Platanus* × *hispanica* with necrotic lesions bordering the veins

Fig. 4. Anthracnose symptoms on mature leaves often in V-shaped patterns

Anthracnose infection occurs at budbreak and during leaf expansion. Besides defoliation, symptoms include a brown leaf blotch associated with the leaf veins. The disease moves from the leaf to the twig where it produces a canker and causes dieback (Fig. 5). Repeated annual killing of twigs results in clusters of old dead twigs and live branches, "witches' brooms" (Pilotti et al., 2002; Terrell, 2004; Nameth and Chatfield, 2007). On heavily infected trees dieback can be extensive leading to the gradual decline (Fig. 6) and death of the tree.

Severe dieback of plane trees following shoot and twig infection by *A. veneta* is favoured by mild winters followed by late spring frosts.

Anthracnose fungi overwinter in leaf debris on the ground and/or dead areas of the bark on the tree, called cankers. In early spring, spores of the fungus are produced in fruiting structures and are dispersed by splashing rain. The expanding leaf buds, shoots or in some cases young leaves are infected by the spores.

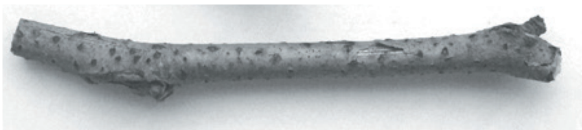

Fig. 5. Cankers on twigs and small branches of affected plane trees

Fig. 6. Infection of expanding shoots and leaves results in gradual withering of affected plane tree

Acervules are formed on the leaves with ovoid to elliptical unicellular conidia. Next spring, the perithecia with ascospores developing on fallen leaves transfer the primary infection to young leaves.

Disease severity changes from year to year and it is dependent on temperature. Cool springs are more conductive to severe plane anthracnose outbreaks. The infection process is favored by relatively cool temperatures and prolonged periods of leaf wetness. Therefore, the disease tends to be more severe during wet, cool springs. After infection, the anthracnose fungus colonizes leaf tissue and begins to produce new fruiting structures and spores capable of reinfecting expanding leaf tissue. Disease development may continue throughout the spring into early summer if favorable weather persists. These diseases tend to be less of a problem during hot, dry summer weather. As temperatures increase, the disease becomes less active and the trees releaf.

The intensity of this disease is generally determined by the weather conditions during the last two weeks of April and the first week of May. If temperatures are not higher than 10 °C and moderate to heavy rain occurs, defoliation to plane will be noticeable varying from light to heavy. This will mean that planes will not develop full foliage until June. Weather determines the severity of anthracnose. Frequent rains and cool temperatures promote the disease. If the average temperature during the two-week period following the emergence of the first leaves is below 12 °C, the shoot-blight phase of the disease will be serious. Disease intensity decreases as the average temperature increases from 12 °C to 15 °C. Little or no anthracnose will occur if average temperatures during this vulnerable stage are above 15 °C (Hitchcock and Cole, 1980; Berry, 1985; Leininger et al., 1999; Swift, 2001; Jurc, 2006).

In general, mild winter and the following late frosts with longer cool and wet weather in spring during sprout budding and prolongation initiate plane anthracnose disease caused by the fungus *Apiognomonia veneta*. Recurring infection year after year weakenes woody plants gradually, so these are more predisposed to other fungal diseases and damage by insect pests. Reccurence of infection caused by *Apiognomonia veneta* was observed at many localities in Slovakia during 2004–2006. These observations correspond to the data of SHMÚ (2004–2007). In comparison with the long-term average values (1959–1980), the average monthly air temperatures in the years 2004–2007 (Table 1) in the observed area were higher, but in the months decisive for origin of infection (period from 17 April to 7 May) in the evaluated years 2004–2007, the temperatures ranged between 10.0 and 12.5 °C. As for the summer period, an increasing trend in temperature was recorded for locality Nitra and the whole of Slovakia.

Table 1. Average air temperature (°C) in observed period 2004–2007 in locality Nitra and in Slovakia

		Locality	
		Nitra	Slovakia
2004	17 April–7 May	14.219	11.849
	Summer	20.091	17.534
2005	17 April–7 May	12.595	10.657
	Summer	20.031	17.980
2006	17 April–7 May	14.109	12.506
	Summer	21.141	18.918
2007	17 April–7 May	12.690	10.925
	Summer	–	–

Average rainfall for locality Nitra and for the whole of Slovakia in the observed summer periods 2004–2007 varied considerably (Table 2). In observed period of the years 2004 and 2005, the value was around 149 mm, in 2006 above 160 mm, but in 2007 value somewhat over 63 mm was recorded. Since significant progress of infection caused by the fungus *Apiognomonia veneta* can be noticed at temperatures lower than 12–13 °C and at higher rainfall in the growing season (above 350 mm), the progress of infection didn‘t induce general withering and tree dieback, but only certain weakening of the trees. The temperatures in the observed period 2007 were below 12 °C, very low rainfall (63.28 mm in whole area of Slovakia), however, was recorded, therefore progress of infection was not noticed.

Table 2. Average rainfall (mm) in observed period in the years 2004–2007 in locality Nitra and Slovakia

		Locality	
		Nitra	Slovakia
2004	17 April–7 May	0.66	147.57
	Summer	2.38	475.52
2005	17 April–7 May	2.93	148.27
	Summer	1.57	579.51
2006	17 April–7 May	1.44	160.28
	Summer	1.82	477.39
2007	17 April–7 May	1.24	63.28
	Summer	–	–

Average temperatures recorded at locality Nitra in all observed critical periods were above 12 °C. Values of temperatures varied from 12.60 to 14.22 °C, and they didn‘t exceed 15 °C representing the value for remission of infection. Rainfall in the period of years 2004–2007 varied noticeably. In 2007 it reached the value of 1.24 mm. This fact in association with temperature 12.69 °C had not a favourable influence on infection progress.

According to Surry and Flückiger (1991), who compared the leaf infections on *P.* × *hispanica* during two years, while leaf necroses were widespread after the 1987 infection, they appeared only sparsely in 1988. This striking contrast might have been caused by the differing climatic conditions. In Basel, the mean temperature in May 1987 was 10.8 °C, compared to 15.0 °C one year later, and the severity of *Apiognomonia* leaf blight is correlated with low temperatures in spring.

According to Swart et al. (1990) all symptoms (bud blight, twig blight, shoot blight) except leaf blight disappeared during the summer months in each year. This observation is consistent with the view of Neely (1976) that anthracnose infection of *Platanus* occurs mainly in early spring. This view corresponds to observations in our conditions. It is further supported by the fact that severity of shoot-blight is determined by the mean daily temperature during the 2-wk period immediately following the emergence of the first leaves in spring – the optimum temperature for disease development ranging 10–13 °C (Neely and Himelick, 1963). Comparing the results obtained in our four-year observations of leaf infection of *P.* × *hispanica*, we also came to similar conclusions. Whereas in spring periods 2004–2006 cool and wet weather was prevailing, dry spring 2007 (precipitation only 63.28 mm) coming after a mild winter caused that no infection on woody plants appeared.

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Vplyv zmenených ekologických podmienok prostredia na výskyt antraknózy platana javorolistého (*Platanus* × *hispanica* Münchh.)

Súhrn

V rokoch 2004–2007 sme na *Platanus* × *hispanica* Münchh. zaznamenali opakovaný výskyt antraknózy spôsobený hubou *Apiognomonia veneta* (Sacc. & Speg.) Höhn. Pôvodca ochorenia, huba *Apiognomonia veneta* bola izolovaná zo symptomatických listov a konárov s charakteristickými škvrnami a léziami z hostiteľských drevín rastúcich v mestskom prostredí (lokalita Nitra). Mikroskopickým vyšetrením boli získané izoláty huby následne identifikované a morfologicky popísané použitím určovacích identifikačných kľúčov. Práca ďalej poukazuje na trend vývoja ochorenia meniaci sa z roka na rok v závislosti na podmienkach prostredia, pričom je možné potvrdiť vplyv teplotných podmienok v období rozhodujúcom pre vznik a ďalší postup infekcie. Každoročne opakované infekcie, ktoré spôsobujú poškodenie listov a konárov platanov vedú k postupnému oslabovaniu a zníženej rezistencii napadnutých drevín, a tak k ich zvýšenej predispozícii na vznik iných hubových chorôb, prípadne na poškodenie hmyzími škodcami.

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