Distribution and Pathogenic Peculiarities of Fungi of the *Alternaria* Genus On Vegetable Crops In Lithuania

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Abstract

In 1999–2001 the investigation of *Alternaria* species on vegetable crops: cabbage, cucumber, tomato and carrot was carried out and fungi species were isolated from vegetables and substrata used for plant cultivation. Isolated micromycetes: *Alternaria alternata, Alternaria tenuissima*, *Alternaria brassicae*, *Alternaria brassicicola*, *Alternaria cucurbitae*, *Alternaria dauci*, *Alternaria radicina* and *Alternaria solani* differed in pathogenic peculiarities, frequency of occurrence and their various reaction to fungicides (a.s. azoxystrobin, dichlofluanid, Cooper hydroxide, mancozeb, propamocarb hydrochloride), plant activator Bion (benzothadiazole) and antagonistic microorganisms (*Trichoderma harzianum, Streptomyces griseoviridis*).

Keywords: *Alternaria* spp.; alternaria leaf blight; cabbage seed plant; carrot; cucumber; tomato; *Trichoderma harzianum*; *Streptomyces griseoviridis*; pesticides; frequency of occurrence

INTRODUCTION

Diseases caused by fungi of the *Alternaria* Nees: Fr. genus are widely spread on many species of vegetables in Lithuania. They affect leaves, stems, flowers, roots and tubers. *Alternaria* diseases usually appear as leaf spots, leaf blights, stem, fruit, tuber and root rots and damping off of seedlings. Fungi of the *Alternaria* genus cause 20–100% losses in vegetable yields depending on various environmental conditions in Lithuania (DOBROVOLSKIENE 1993; GRIGALIUNAITE 1994).

Diseases, caused by *Alternaria* spp. are controlled primarily through the use of resistant varieties, disease-free or treated seeds, crop rotation, removal and burning of plant debris and chemical sprays with pesticides such as iprodione, chlorothalonil, maneb, captafol, mancozeb, propiconazole, penconazole (MUINO & PARRA 1992).

As the application of fungicides often produces a harmful effect on ecosystems, searching for microorganisms-antagonists to plant pathogens and introducing them into agroecosystems is one of the solutions in modern plant protection. The discovery of fungal antagonists has led to new challenges in research, development and registration of biocontrol products in a market where chemical pesticides dominate (AVIS *et al*. 2001; SURVILIENE 2002).

The purpose of this study was to investigate micromycetes occurrence frequency of the *Alternaria* genus and establish impact of some chemical and biological plant protection means on vegetable crops.

MATERIALS AND METHODS

In 2000–2001 the experiment was carried out at the Lithuanian Institute of Horticulture. Micromycetes were isolated and examined according to mycological and phytopathological investigation methods (KIRALY *et al*. 1970) and identified according to the descriptors of cultural and morphological characters (ELLIS 1971; LUGAUSKAS 1997).

Occurrence frequency of micromycetes species was established according to the formula: A = B/C $\times \cdot 100$ (%), where A – occurrence frequency of a micromycete specie; B – the number of samples, in which the specie has been detected; C – total number of investigated samples. Species, whose frequency of occurrence (A in %) was lower than 30%, were

ascribed to accidental species, higher than 30% – to typical species, higher than 50% – to dominating species (LUGAUSKAS 1988).

The impact of different plant protection means on the development of *Alternaria* spp. was estimated by using systemic fungicides: Amistar 250 SC (active substance azoxystrobin) at the rate of 0.8–1.0 l/ha and Previcur 607 SL (propamocarb hydrochloride) – 0.4 l/m^3, contact fungicides: Dithane 75 DG (mancozeb) – 2–3 kg/ha, Euparen 50 WP (tolylfluanid) – 1.5 kg/ha and Champion 50 WP (Copper hydroxide) – 2.5 kg/ha, plant activator Bion 50 WG (benzothadiazole) – 0.05 kg/ha and antagonistic microorganisms (*Trichoderma harzianum, Streptomyces griseoviridis*).

RESULTS AND DISCUSSION

Samples of leaves, stems, seeds and substratum for mycological analysis were collected from different fields and greenhouses in Lithuania. Species of *Alternaria* genus from cucumber were isolated as follows: *Alternaria cucumerina* (Ellis & Everh.) Elliott, from cabbage – *Alternaria brassicae* (Berk.) Sacc. and *Alternaria brassicicola* Schw., from tomato – *Alternaria solani* Ellis et Martin, from carrot – *Alternaria dauci* (Kuehn) Groves et Skolko and *Alternaria radicina* M., D et E., from onion – *A. porri* (Ellis) Cif. Species *Alternaria alternata* (Fr.) Keissler was isolated from many of afore mentioned vegetables. Fungus *Alternaria tenuissima* (Kunze: Pers.) Wilt. was isolated from greenhouse substrates. All these species were pathogenic on all observed vegetable crops. Occurrence frequency (A) of micromycetes was different. The highest disease ratings were recorded on white cabbage seed plants. Species of *Alternaria* were seldom found in rhizosphere of cabbage seed plant (A < 30%), but *A. alternata* and *A. brassicae* were dominating (A > 50%), *A. brassicicola* – typical micromycetes (A > 30%) on the phyloplan. Pathogen *A. cucumerina* dominated (A > 50%) in injured cucumber phyloplan. Micromycetes *A. alternata* and *A. cucurbitae* were found in samples rather often of healthy and injured cucumber rhizosphere. Species *A. solani*, *A. alternata* were isolated from tomato and were ascribed to typical ones (A < 30%). Pathogens *A. radicina* dominated (A > 50%) in rhizosphere, *A. dauci* – in phyloplan of carrots. *A. alternata* was seldom found both in phyloplan and rhizosphere. Fungi *A. porri* dominated (A > 50%) usually as a secondary invader after appearance of onion downy mildew, *Peronospora destructor* (Berk.) Caspary.

The effect of fungicides on *Alternaria* spp. was investigated in order to find effective protection means for growing of cucumber and cabbage seed plants.

The first symptoms of alternaria leaf blight in white cabbage seed plants appeared in the middle of June. The black spots were generally dark brown to black, numerous and enlarging, and usually developing in concentric rings, which give the spots target-like appearance. Lower, senescent leaves were attacked the first, disease progressed upwards and made affected leaves turn yellowish and senescent. In this case were tested plant protection activator Bion, fungicides: Amistar, Euparen, Champion. All pesticides gave adequate control of alternaria leaf blight on white cabbage seed plants (Figure 1).

Application of pesticides increased the yield of cabbage seeds (0.17–0.19 t/ha). The biological efficiency of the fungicides against fungi of the *Alternaria* genus was 18.7–90.6%. Fungicide Amistar was most effective. It reduced disease prevalence (90.8%) and intensity (88.8%).

Alternaria leaf blights, caused by *A. alternata* and *A. cucumerina* occur on field cucumber usually at

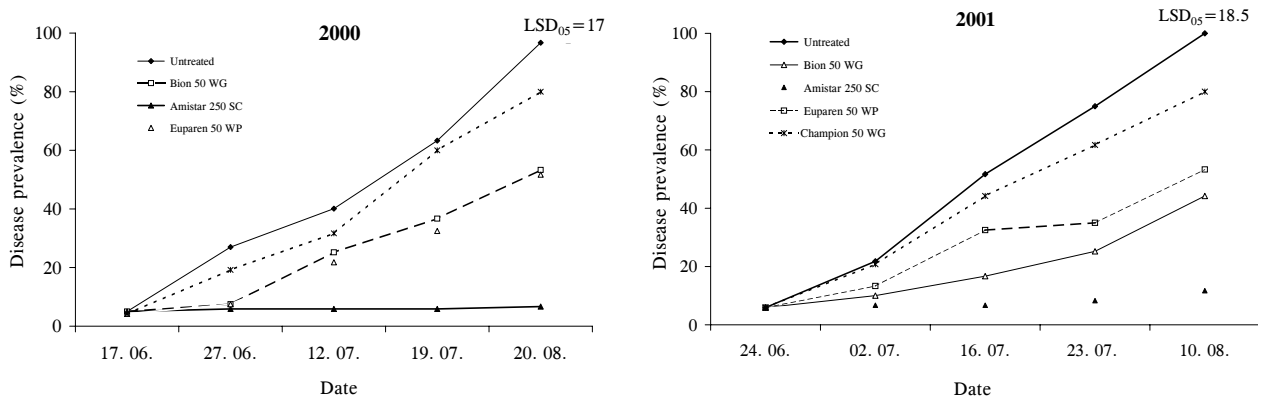

Figure 1. Effect of pesticides on development of alternaria leaf blight on white cabbage seed plants (2000–2001)

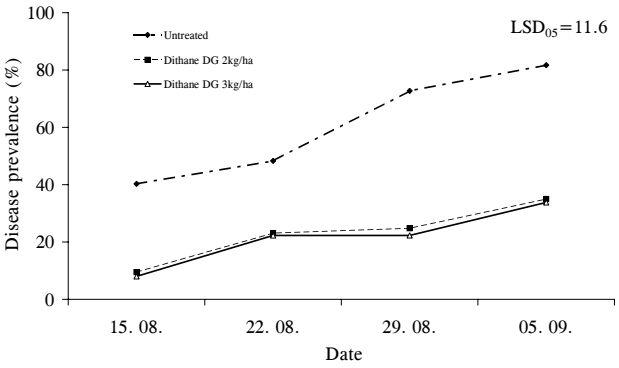

Figure 2. Development of alternaria leaf blight in field cucumber (2000–2001)

the beginning of August, and spread most rapidly in wet seasons or through excessive overhead irrigation. *Alternaria* spp. requires a water film on leaves to infect the plant (JARVIS 1992). Disease symptoms appeared on leaves of untreated control plants as small, water-soaked, circular, gray-tan spots on the upper surface enlarged in concentric rings; on the lower surface they were less distinct. The lesions were 1 cm wide and eventually coalesced. Untreated cucumbers practically died in three weeks since the appearance of first symptoms of alternaria leaf blights. Obviously, previously sprayings by fungicide Dithane at the rate of 2–3 kg/ha 4 times per season were efficient enough to control alternaria blights in field cucumbers (Figure 2). The prevalence of disease on the control cucumber leaves in the middle of August reached 40.3% and increased to 82% in three weeks. The last cucumber fruits in the control plants were picked on September 12, while in the trials with Dithane – on September 26. Application of fungicide reduced the prevalence of alternaria leaf blights from 52.1% to 80.15% and increased the yield of cucumber by 51–66%. The residues of active substance mancozeb were found in cucumbers (0.38 mg/kg fruit) only in the trial with Dithane at the rate of 3 kg/ha 3 days past application, but were absent after 5 days following after application (by colorimetric method).

From the non-treated greenhouse substrate *Alternaria* species were isolated most frequently as dominating ones (Table 1). Among soil-borne fungi, which cause root rot and damping-off *Alternaria* species are important (SURVILIENE 2001). It should be noted that *A. alternata* destroy lignin-cellulose complex and pectin in substrate, produce various enzymes and toxically materials (tenuazonin acid, tentoxin, altenuisol, altenusin, dehydroaltenusin, izocumarin, alternarid and other). For these peculiarities *A. alternata* adapt in different substrates easily (LUGAUSKAS 1997). The obtained data show that species *A. alternata* is sufficiently well adapted to changing chemical and microbiological conditions in the substrate. Only *T. harzianum* suppressed functions of *A. alternata* completely. Antagonistic *S. griseoviridis* species was aggressive against *A. alternata* partly; in this case *A. alternata* functioned as typical species (A > 30%). Systemic fungicide Previcur inhibited *A. alternata* spreading to accidental level (A < 30%). Fungus *Alternaria tenuissima* is cosmopolitan, extremely common and recorded on a very wide range of plants, usually as a secondary invader rather than parasite

Table 1. Reaction of *Alternaria* spp. to antagonistic microorganisms and fungicide in greenhouse substrate

Treatment	Rate (ml, g/m^3)	*Alternaria alternata (=A. tenuis)*	*Alternaria tenuissima*
Before treatment	–	+++, dominating species, A > 50%	+++, dominating species, A > 50%
Previcur 607 SL (Propamocarb hydrochloride)	400 ml	+, accidental species, A < 30%	–, not isolated
Streptomyces griseoviridis (min. 10^8 c.f.u./g)	5 g	++, typical species, A > 30%	–, not isolated
Trichoderma harzianum (min. 10^8 c.f.u./g)	7 g	–, not isolated	–, not isolated

(ELLIS 1971). The functional activity of *A. tenuissima* was depressed completely in all treatments.

References

AVIS T.J., HAMELIN R.C., BELANGER R.R. (2001): Approaches to molecular characterization of fungal biocontrol agents: some case studies. Can. J. Plant Pathol., **23**: 8–12.

DOBROVOLSKIENE A. (1993): The most important cabbage seed plants diseases and means to reduce their harm. Horticulture and vegetable growing. Sci. Works, **1**: 167–174.

ELLIS M.B. (1971): Dematiaceous Hyphomycetes. Kew, Surrey, England: 507.

GRIGALIUNAITE B. (1994): Fungi on vegetable seeds. Agric. Sci., **3**: 77–89.

JARVIS W.R. (1992): Cucumber Diseases. Agriculture Canada Publ.: 49.

KIRALY Z., KLEMENT Z., SOLYMOSY F., VOROS J. (1970): Methods in Plant Pathology. Academiai Kiadó, Budapest: 343.

LUGAUSKAS A. (1988): Micromycetes of Agricultural Soils in Lithuania. Vilnius, Mokslas (in Russian).

LUGAUSKAS A. (1997): Microbiological deteriorations of materials. Institute of Botany, Vilnius.

MUINO B.L., PARRA E. (1992): Efecto del iprodione sobre *Alternaria* spp. en condiciones de laboratorio. Protect. Plantas, **2**: 83–87.

SURVILIENE E. (2001): Phytosanitary state and urgent problems of greenhouse vegetable production in Lithuania. In: Proc. 9th Int. Conf. Horticulture, Lednice, 2: 392–397.

SURVILIENE E. (2002): Reaction of micromycetes to antagonistic organisms and fungicides in substrate. Biologija, **1**: 15–17.