Control of Dieback, Caused by *Eutypa lata*, in Red Currant (*Ribes rubrum*) and Gooseberry (*Ribes uva-crispa*) in the Netherlands

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Abstract

Over decades, growers in the Netherlands have problems with a disease that causes dying branches and stem cankers in red currant. For many years it was assumed that this disease was related to fungi such as *Nectria cinnabarina*, *Phomopsis* spp. and the insect *Synanthedon tipuliformis*. However, recently it was found by Applied Plant Research and the Plant Protection Service that the causal organism is the fungus *Eutypa lata*. The disease is considered of major economic importance, especially as red currant growing is rapidly expanding in the Netherlands. *E. lata* was identified with three detection methods (visual, plating and DNA). Symptoms of *E. lata* do not usually appear until currant plants are at least three to four years old. These cankers are always associated with old pruning wounds. Eventually, the entire branch is killed. High disease incidences and annual losses of 10% to 30% of the productive branches are reported. In some cases entire fields have to be replanted. *E. lata* is well known as one of the most destructive diseases of grapevines (*Vitis vinifera*). The importance of this disease in currant growing was not known. Research is focusing on the evaluation of control measures; e.g. chemical and biological control treatment of pruning wounds, and disease management such as sanitation practices. Also, the epidemiology of *E. lata* is studied. High densities of ascospores of *E. lata* were found in a spore trap placed in a red currant field in the Netherlands. In the subsequent field survey fruiting structures (stromata) and ascospores were found on dead infected red currant wood.

INTRODUCTION

Since decades, growers in the Netherlands encounter problems with a disease that causes dying branches and stem cankers in red currant (*Ribes rubrum*) and gooseberry (*R. uva-crispa*). For many years it was assumed that dieback in red currant was related to fungi such as *Nectria cinnabarina*, *Phomopsis* spp. or the insect *Synanthedon tipuliformis*. Control measures were therefore always focused on these (alleged) causal agents. However, control strategies against these diseases and pests did not result in reduced dieback incidences in commercial red currant plantations.

Symptoms of dieback do normally not appear until plants are at least four years old. The first symptom is usually a sudden wilt of a branch during mid-summer. Leaves die and berries develop very poorly but remain attached to the branch, or even fail to develop. Examination of the base of the dead branch will reveal a canker surrounding an old pruning wound. Dieback branch symptoms are mostly accompanied by a canker, which often appears V-shaped in a cross-section of the perennial wood (Fig. 1). Cankers have a definite margin between the living and dead wood, and are also characterized by typical splitting and cracking of bark. These symptoms refer to the classical symptoms of Eutypa dieback, as has been described by Carter (1991).

Eutypa lata is a pathogen of woody plants worldwide and occurs on at least 88 species of woody dicotyledons in 52 genera including *Prunus* spp. (peaches, plums,

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cherries), *Malus* spp. (apples), *Pyrus* spp. (pears) and *Juglans* spp. (walnuts) (Carter, 1991). It is known as one of the most destructive diseases of *Vitis vinifera* (grapevine) and responsible for significant economic damage in grape production (Munkvold et al., 1994; Wicks and Hall, 1997). The pathogen is disseminated by ascospores, which infect (pruning) wounds. The fungus invades the vascular system of the trunk and shoots, eventually leading to a characteristic dark and wedge-shape necrosis of woody tissues. Due to a slow disease progress, fungal infection is hardly noticeable during the first years. After an incubation period of three or more years the host is slowly killed (Carter, 1991). The anamorph and teleomorph states of the fungus are produced on dead wood.

Disease diagnosis of *E. lata* is commonly done by isolating the fungus from wood cankers on Potato Dextrose Agar (PDA) medium. On this medium *E. lata* grows as a white cottony mycelium which can produce pycnidia with conidia of the anamorph (*Libertella* sp.). Perithecia are not produced in culture and some isolates even fail to produce pycnidia in culture, making it difficult to distinguish *E. lata* from other diatrypaceous fungi.

Recently, *E. lata* was identified as the causal agent of dieback of red currant and gooseberry in the Netherlands (Wenneker et al., 2011). Research is focusing on the epidemiology of *E. lata* in the Netherlands and the evaluation of control measures; e.g. chemical and biological control treatment of pruning wounds, and disease management such as sanitation practices.

Field Surveys in the Netherlands

Fifteen commercial red currant and gooseberry plantations in the Netherlands were inspected carefully for the presence of plants showing dieback symptoms in 2006-2007. Infected branches with visible symptoms (i.e. cankers) were transported to the laboratory where isolations were made. Dead branches and dead stems were also inspected for the presence of the sexual stage (perithecia and ascospores) of the fungus. In a gooseberry plantation the disease incidence was recorded by evaluating individual plants.

E. lata was isolated from diseased plants sampled on 7 commercial red currant plantations and 4 gooseberry plantations in the Netherlands. Disease incidences were difficult to estimate, as affected plants are usually removed by the growers. In certain older (6-10 year) red currant plantings all plants were visibly affected, indicated by the removed main stems, possibly due to Eutypa dieback. A careful inspection of a 6-year old gooseberry plantation revealed that 130 out of 244 (=53%) plants ('Pax') were affected, most likely due to Eutypa dieback.

A survey in heavily affected plantations revealed fruiting structures (stroma), asci and ascospores on dead infected red currant wood ('Rovada'). This is the first record of sexual fruiting bodies observed in red currant. Perithecia were about 0.5 mm in diameter, and irregularly distributed in one layer (honeycomb-like appearance). Asci were 30-60×5-8 µm, very numerous, spindle-shaped, and eight-spored. Ascospores were allantoid, subhyaline, and 7-10×2 µm. Based on these morphological features, the fungus was identified as *Eutypa* sp. (*Ascomycetes*, *Diatrypaceae*). Plating of ascosporic suspensions on PDA resulted in the growth of mycelium that eventually produced pycnidia and conidia and were included in a phylogenetic analysis (Wenneker et al., 2011).

This phylogenetic analysis showed that there is very little variability in the ITS sequences of the *E. lata* isolates from several cultivated and wild host species. These results also indicate that *E. lata* isolates obtained from different countries in Europe show very little variability in the ITS sequences. Therefore, we expect that cross infections between different host plants can easily occur, especially with highly susceptible species as grapevine and, apparently currants. This implies also that native – non-currant – hosts may act as inoculum reservoirs for currant and grapevine plantations in the Netherlands.

In our research *E. lata* was isolated from most commercially used red currant cultivars, e.g. 'Junifer', 'Rovada' and 'Roodneus', and gooseberry cultivars, e.g. 'Achilles', 'May Duke' and 'Pax'. This indicates that most red currant and gooseberry cultivars are susceptible for *E. lata* infections. Differences in disease severity are more likely related to management practices, i.e. pruning intensity or strategy, than to cultivars resistance or tolerance to *E. lata*. In grapevine cultivars differences have been found in susceptibility to *E. lata* and subsequent symptom development (Péros and Berger, 1994). However, no grape cultivar is known to be immune to infection.

From our observations we conclude that in (older) red currants and gooseberry plantations disease incidences are also very high. Questionnaires and interviews with growers, extension officers and own surveys revealed that Eutypa dieback in red currant is present in all growing regions of the Netherlands.

The costs of the Eutypa dieback in red currants and gooseberries are due to decreased yields, increased management and reduced longevity of the plants. More and more Dutch growers tend to replace old stocks for new material within 5 to 6 years, not awaiting severe disease development. To improve management strategies (e.g. sanitary measures) in order to control Eutypa dieback of currants in Dutch or European plantations it is necessary to identify potential inoculum sources in native and cultivated plant species. Epidemiology and control measures are currently investigated in the Netherlands. Also the spread of the disease via propagation material (nurseries) cannot be excluded, as high disease incidences were observed in young plantations in a region without old currant or grapevine plantations in the neighborhood.

In certain older (6-10 year) plantings virtually all plants were affected by dieback, indicated by the removed main stems. The cultivation of red currant is still growing in the Netherlands. Current growing changed in the past decades from small scale to intensive commercial growing. Modern cultivation practices in red currant growing, i.e., intensive pruning, will increase infection risks and hence disease incidence. In modern growing, red currant plants consist of two main branches, which will produce for 10-15 years on 2-year old shoots. Approximately, 6 shoots per main branch are maintained. After fruit bearing these shoots are pruned, and 6 new shoots are selected on the main branch. All other new (one-year) shoots are removed as well. This implies an annual creation of 6 larger pruning wounds and 15-20 smaller pruning wounds per branch. The large numbers of pruning wounds are probably created in a sensitive period for infection, i.e., ascospore discharge and suitable conditions for infections (Munkvold and Marois, 1995).

Managing Eutypa Dieback

1. Wound Treatment. The preferred method of control against *E. lata* infections would be prevention of infection by treatment of large wounds with fungicides or wound sealants (Moller and Kasimatis, 1978; Moller and Kasimatis, 1981, Sosnowski et al., 2008, 2009). However, effective (benzamidazole) fungicides are not registered as wound protectans in the Netherlands. At the moment, research in grapevine on alternative chemicals for wound protection, and biological control by treating pruning wounds is in progress (Sosnowski et al., 2004; John et al., 2005). Based on the information about grapevine protection research is started in the Netherlands to protect pruning wounds of currants against *E. lata* infections.

In our research the effect of chemical wound treatment is evaluated by Topsin-M (a.i. thiophanate methyl) application. The effect is tested using two application methods: (i) spray application and (ii) applied by hand to fresh pruning wounds. Research is currently under way to evaluate and to optimize application with spray equipment to provide a practical and efficient method of controlling Eutypa dieback in currant plantations. First results indicate that both application methods are very effective in controlling Eutypa dieback.

2. Pruning Shear. The application of wound protectans (fungicides, pastes and paints) by hand is very labor intensive and costly. Simultaneous application and pruning would reduce labor costs by decreasing the amount of time needed to complete both tasks. For that reason we are currently testing and modifying a Felco hand pruner to apply formulation upon cutting currant vines. First results showed that the modified hand pruner was effective in delivering an emulsion (=good coverage) to currant pruning wounds simultaneously with vine cutting. Further research will be carried out regarding the biological efficacy of the treatment.

3. Sanitation. Removal of dead wood from currant plantations may reduce inoculums levels. However, this alone is not sufficient for managing dieback. Spores can be blown up to 50 km in the wind and there are many alternative hosts, which may also act as inoculums sources (Carter, 1991).

4. Pruning. The risk of infection will be reduced if certain points are taken into consideration, such as: (i) pruning is avoided in wet weather when ascospores are released, (ii) pruning is performed in late season when wound healing is fast and the sap is flowing, (iii) the number and size of pruning wounds is kept to a minimum, (iv) wound protection is applied immediately to all larger pruning wounds.

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Figures

Fig. 1. Typical V-shaped area in a cross section of a canker due to Eutypa dieback in perennial red currant wood.