The BioChon story: deployment of *Chondrostereum purpureum* to suppress stump sprouting in hardwoods.

MEINDERT D. DE JONG

*Laboratory of Theoretical Production Ecology, Wageningen University, P.O. Box 430, 6700 AK Wageningen, The Netherlands. E-mail: TPEJONG@RCL.WAU.NL*

The biological control of stump sprouting in American bird cherry (*Prunus serotina*) and poplar (*Populus euramericana*) by applying a mycelium suspension of *Chondrostereum purpureum* is outlined. This fungus is being commercially deployed as a mycoherbicide under the brand name BioChon in order to prevent stump sprouting in hardwoods. It acts as a wood decay promoter. Effectivity of BioChon applied to control stump sprouting in *P. serotina* generally results in a kill of about 95% two years after treatment, and it appears to be as effective on poplar.

Research on *C. purpureum* is also being carried out in Canada to control unwanted hardwoods in silviculture, in Switzerland for vegetation management, and in New Zealand to control imported woody weeds in pastures. Infection danger to non-target plants due to use of this pathogen as a mycoherbicide seems to be negligible in most cases.

Worldwide marketing of the fungus is discussed.

**Keywords:** mycoherbicide, *Chondrostereum purpureum*, biological weed control, *Populus euramericana* cv. 'Robusta', *Prunus serotina*.

American bird cherry, also called black cherry (*Prunus serotina*), is a tree that was introduced to Europe from the United States and Canada. In The Netherlands it grows as a shrub on sandy soils in the needle (conifer) forests. Because it has become an exotic weed in many forests it has been given the nickname of “Bospest” (Forest Pest) in The Netherlands. Foresters who want to rejuvenate their tree stands, or recover their original vegetation, must use something to combat the American bird cherry. Simple cutting does not solve the problem because after some time the tree sprouts, often resulting in more branches than before.

**Silver leaf disease**

Silver leaf, a common disease in fruit trees like plum and cherry, is caused by a basidiomycete named the purple crust fungus, *Chondrostereum purpureum* (Pers.: Fr.) Pouzar. Research at the DLO-Institute of Agrobiological and Soil Fertility in Wageningen demonstrated that *C. purpureum* can be at least as effective in controlling stump sprouting as the usual chemical control agents (Scheepens & Hoogerbrugge, 1989). After a tree has been cut, the fungus can be put directly onto the wound surface, from which it enters and continues growing inside the wood of the tree (Fig 1).

When the fungus develops in the root system of the tree, silver leaf disease appears (Fig 2). The leaves become dull, losing their characteristic sheen, and the equally characteristic lead-grey colour appears. The affected trees and shrubs will eventually die because the disease prevents water being transported through the vascular system. Silver leaf is associated particularly with fruit trees such as plum and cherry, and to a lesser extent with apple and pear. During summer in older plum and cherry orchards a few trees with silver leaf may often be seen, even from a distance.

**Experiences with purple crust**

*C. purpureum* does not infect trees except through fresh open wounds and, because the native wood species and the European bird cherry (*Prunus padus*) are not pruned, the chance of these trees becoming diseased and dying off as a result of silver leaf is very small. The fungus is a naturally occurring organism and mostly does not lead to silver leaf disease.
Rather, it is a saprophyte living on dead wood. However, when we inoculated fresh wounds of sweet cherry (*Prunus avium*) and sloe (*Prunus spinosa*), silver leaf appeared in these trees as well. Apparently the organism is able to live either as a saprophyte, or as a parasite on living hosts.

In autumn the fungus is recognisable by its fruiting bodies that form on the wood substrate. It gets its name 'purple crust' from the shape and colour of the basidiocarps, and is often found on stumps and dead wood remains of poplar trees. In the Dutch province of Flevoland there are often several thousand basidiocarps on heaps of poplar wood piles and logs. These release many spores that are dispersed by wind and can infect fresh substrates.

The fungus can also develop large numbers of basidiocarps on stumps of sweet cherry (*Prunus avium*) (Fig 3) and alder (*Alnus glutinosa*, Figs 4, 5).

**Sudden death**

The good experimental results with control of American bird cherry have given rise to research to determine whether stump sprouting of other hardwood trees could also be stopped by this fungus. In the poplar forests in the recently (1966) reclaimed polder of Flevoland, experiments have been conducted to test *C. purpureum* against poplars. Stumps of recently cut poplars (*Populus euramericana* cv. 'Robusta' syn. cv. 'Zeeland'; which is the male clone of the hybrid *P. deltoides* $\times$ *P. nigra*) were inoculated with BioChon (brand name of the mycoherbicide based on *C. purpureum*) (Fig 6). This is the most often planted poplar cultivar in Flevoland and is one of the fastest growing poplar cultivars. It is a vigorously regenerating species owing to its abundant stump sprouting.

In one biocontrol trial near Lelystad ('Boswachterij Hollandse Ilout'), 112 poplar stumps (diameter of 400-600 mm) were inoculated with BioChon in autumn. In the subsequent spring a small number of the poplar stumps did not sprout and in the summer it was appearing to be less effective. However, a lot of stumps had not yet sprouted and subsequently, a month later, the new shoots on many sprouting stumps with wilted and died (de Jong & Scheepens, 1996). Later in autumn these stumps appeared to be dead, while 54 alders cut simultaneously in the same forest, but not inoculated, had only 15% mortality. Apparently in summer 'sudden death' of the poplar stumps had occurred (Fig 7). These affected stumps had black shoots with dead leaves giving the appearance of having been burnt by fire. In the end all the 112 inoculated poplar stumps appeared to be dead.

**Infection risk**

In order to investigate infection risk of the intended biocontrol method a few additional minor investigations were performed in addition to a rather theoretical risk analysis of spore dispersal (De Jong et al. 1990). Risk might be reduced by preventing fructification of *C. purpureum* and to investigate this several experiments were carried out. Addition to the inoculum of a saprophytic wood invading fungus (*Stereum hirsutum* or *Noctria cinabarinina*) did not reduce fructification of *C. purpureum*. In another experiment, fructification of some single-spore isolates occurred and, unexpectedly as measured by disease incidence and fructification, single-spore isolates were as pathogenic as multiple spore cultures (De Jong et al., 1998).
Fig 2 Silver leaf symptoms on an inoculated American bird cherry (Prunus serotina).

Fig 3 The purple crust fungus (Chondrostereum purpureum) on sweet cherry (Prunus avium).

Fig 4 Heavy infection of C. purpureum on a stump of alder (Alnus glutinosa).

Fig 5 C. purpureum developing on alder (Alnus glutinosa), showing the detail of many mature basidioceps (from de Jong et al., 1998).

Fig 6 BioChon, sold as a watery suspension of mycelium of Chondrostereum purpureum. (Koppert Biological Systems, P.O. Box 155, 2650 AD Berkel en Rodenrijs, The Netherlands)

Young poplar trees (Populus euramericana cv. 'Robusta', 2-4 m tall, 5-8 trees in a new plantation were selected) and alder trees (Alnus glutinosa) 3-4 m tall were inoculated with a 10% dilution of BioChon in order to test susceptibility for silver leaf disease. Freshly made wounds (2-5 per tree, diameter 2-8 cm) were inoculated with C. purpureum using a 10% dilution of BioChon. All trees remained healthy.
Fig 7 Sudden death of a stump of a poplar (*Populus euramericana* cv. 'Robusta') (From De Jong & Scheepens, 1996).

Fig 8 First biological control of *Prunus serotina* in practice; two years after treatment of stumps with BioChom.

(compared to non-inoculated trees) except for a few belated wound closures and a little callus formation on the wound surface. Therefore, freshly pruned trees of these cultivars in or near a biocontrol site are not exposed to high risk.
According to a risk analysis of spore dispersal, a safety zone of 0.5 km around biocontrol sites is taken as necessary, based upon the natural (other than bioherbicide-derived) levels of inoculum present. Later on the Dutch Plant Protection Service suggested covering very big stumps with litter.

**Worldwide perspective**

From research in Canada it appeared that *C. purpureum* offers a method to control the growth of deciduous hardwood species such as alder, birch, maple, and poplar. After pasting of stumps with *C. purpureum* parts of the cut stumps appeared to be dying slowly while the remainder of the stumps exhibited stunted and unhealthy growth of the regenerating shoots.

This is advantageous because under the power lines in the Canadian province of Quebec, trees must not be allowed to grow too tall. The electricity is transported from the northern power plants to the cities via an extensive network and the power companies want to prevent extensive tree growth. After the year 2000 no chemical control agents should be used for this purpose.

Similarly in British Columbia (the most western province of Canada) the vegetation below electrical cables must be kept below certain levels. Additionally there is a need to limit the growth of deciduous trees in extensive conifer plantations (de Jong et al., 1996). The 'biological silvicide' based on *C. purpureum* is to be marketed soon under the brand name of ECoclear.

In southern Switzerland, we have started the first experiments to control stump sprouting in the invasive *P. serotina* by *C. purpureum* (de Jong et al., 1998). In New Zealand, Graeme Bourdôt and a team at AgResearch have set up some trials for biocontrol of invasive broom and gorse in pastures.

**Marketing *C. purpureum* as a mycoherbicide**

In The Netherlands, Koppert Biological Systems, a Dutch company, is marketing BioChon, a product containing a mycelium suspension of *Chondrostereum purpureum* (Fig 8). In the Belgian region of Flanders, interest is also increasing in application of BioChon (Van Den Meerasschat et al., 2000).

BioChon is the first European mycoherbicide. It is, however, not yet officially registered and the registration procedure is proving to be very expensive and laborious. Hopefully, The Dutch Executive Board for Registration of Pesticides will make approval of biological control agents easier.

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**References**