Prospecting for organic fungicides and resistance inducers to control scab (*Venturia inaequalis*) in organic apple production

M. Bengtsson, H.J.L Jørgensen, E. Wulff, and J. Hockenhull.

***Abstract* – As a part of the REPCO project we are prospecting for plant extracts and other materials to control apple scab in order to find substitutes for copper fungicides in organic apple production. Two routine screening systems are used to evaluate the potential of materials to control apple scab; one testing the effect of materials on conidium germination on glass slides, and one testing the effect of materials on scab development on apple seedlings grown in growth chambers. Several interesting materials have been identified and a number have been selected for testing in experimental organic apple orchards. Studies of the mode of action of promising materials including induced resistance are on-going and work on improvement of timing and formulation is initiated at KVL.[1]**

Introduction

Apple scab (*Venturia inaequalis* (Cke.) Wint.) causes serious losses in quality and yield of organically as well as conventional grown apples. Only elemental sulphur is permitted for scab control in organic apple production in Denmark, while copper fungicides are used in organic production in many European countries. However, the use of copper fungicides in the EU is now being phased out (EU Council Regulation (EEC) No. 2092/91) and alternative fungicides for control of apple scab and other diseases in other crops are increasingly needed. The EU project REPCO (2003-2007) has the objective to contribute to the replacement of copper fungicides in European organic production of grapevine and apple with new control measures against downy mildew in grapevine and scab in apple. KVL contributes to the project in a work package on development of resistance inducers and organically based fungicides. This includes searching and screening of potential materials to control *V. inaequalis* in laboratory and growth chamber experiments and studies on mode of action and improvement of promising materials. With special focus on plant extracts we also look for materials working as resistance inducers (Jørgensen *et al*., 2004). Based on the results from the screening assays, promising materials are selected for orchard testing at DIAS, Denmark, and PPO, The Netherlands.

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Searching for materials

The search for candidate materials for apple scab control is a continuous process and a list (~100), including many plant extracts, has been compiled (REPCO, unpublished). Materials on this list have been found through literature searches, on the web, via personal contacts, preliminary results from the StopScab project (Bengtsson *et al.*, 2004; Bengtsson *et al*., *in press*) and through company contacts. Before working with a material it is first subjected to a preliminary database screening for toxicological risks, assessment of economic feasibility and acceptability for use in organic growing (carried out by FIBL, Switzerland). Among the plant extracts both crude extracts made from leaves, flowers, seeds, roots etc. and extracts prepared and formulated by companies are included. Primarily we try to select extracts made from plant species traditionally used for medicine, food additives or flavouring and some are made from residues from the food industry which may make a future registration easier and, especially in the latter category, cheaper to produce.

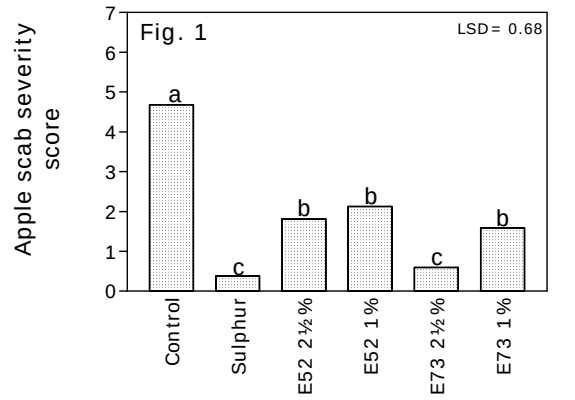
Screening systems

Two routine screening systems are used, one testing the effect of materials on conidium germination on glass slides, and one testing the effect of compounds on disease symptom expression on leaves of apple seedlings (Bengtsson *et al., in press*). Inoculum of *V.inaequalis* is produced artificially by the bottle wick method (Williams, 1978). Solutions of test materials are prepared according to protocols supplied by companies and other sources or, for new materials, by trial and error. Generally, hot or cold water extractions of crude plant materials and residues are first tested in standard concentrations, e.g. 2.5% and 5% (w/v). In the spore germination test conidia are mixed with the test solutions, incubated for 24 and 48 h and the percentage of germinated conidia assessed and compared to a water control. In the plant assay, apple seedlings (*Malus* x *domestica* cv. 'Golden Delicious') are inoculated with conidia and test solutions are applied either one day before or one day after inoculation. Sulphur and water are used as reference treatments. Two weeks after inoculation, disease symptoms are scored on the leaves according to a standardised scale.

RESULTS AND DISCUSSION

Screening of candidate materials

Over 100 plant extracts and other materials have been tested at KVL in the routine screening assays. Among the materials that inhibit conidial germination on glass slides many have either no or only a low effect in the plant assay. However, promising materials include several plant extracts from plant species belonging to different taxonomical groups and geographical origins. Two plant extracts, E52 and E73 (Nor-Natur Aps, Denmark) have consistently shown promising efficacies especially when used preventively (Fig. 1). The results shown in Fig. 2 illustrate the problems we face finding materials with good curative action. As can be seen, sulphur and plant extract E52 have only very low efficacies while plant extract E73 is more effective.



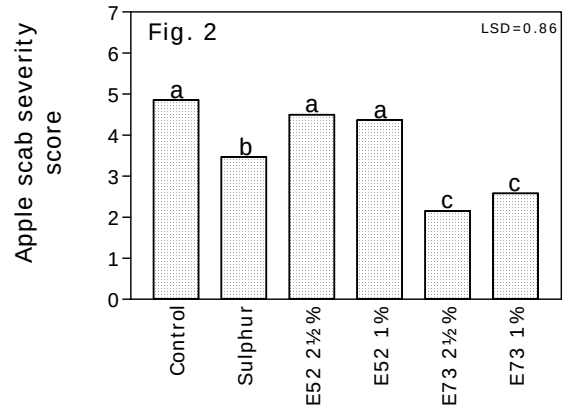


Figure 1 & 2. Effect of sulphur, plant extract E52 (2.5% and 1%, w/v) and plant extract E73 (2.5% & 1%, w/v) on scab symptoms on leaves of apple seedlings. Treatments applied one day before inoculation (preventive, Fig. 1.) or one day after inoculation (curative, Fig. 2). Disease assessment performed two weeks after inoculation. Treatment means with the same letter are not significantly different.

Mode of action of plant extracts

Most of the plant extracts with effect in the plant assay have a moderate to strong effect on conidium germination in the glass slide test, thereby indicating that their primarily modes of action are fungistatic or fungicidal. However, extracts from some plant species appear to act very differently as they do not inhibit spore germination on glass slides but reduce symptoms when applied to seedlings. Such plant extracts might be acting as resistance inducers, by triggering natural defence systems (Jørgensen *et al.*, 2004) in the apple seedlings. Recently, we made a histopathological study of the infection of *V. inaequalis* in seedling leaves pre-treated with a synthetic resistance inducer (acibenzolar-S-methyl, Bion WG 50™; Syngenta Crop Protection, Switzerland) (Bengtsson *et al., in press*) and plant extract E73 (Bengtsson *et al.*, submitted). This study indicated that induced resistance operated in apple leaves against *V.inaequalis* treated with Bion™ while the action of E73 was mainly fungicidal. Several other approaches for studying induced resistance have now been initiated on apple leaves treated with some of the promising plant extracts.

CONCLUSIONS

Several materials with promising control efficacies against apple scab have been identified using a condium germination test and apple seedling assay. Materials have either protective or curative effect or both. The most promising materials undergo testing in organic apple orchards. Studies on selected plant extracts, possibly working as resistance inducers, are ongoing at KVL and work on improvement of timing and formulation of selected materials is to be carried out in the near future.

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