501452

REPCO

Replacement of Copper Fungicides in Organic Production of Grapevine and Apple in Europe

Specific Targeted Research Project

Priority 8.1 Policy-oriented research

Publishable Executive Summary
Period 1

Period covered: from 1 Nov 2003 to 31 Oct 2004 Date of preparation: 10 Dec 2004

Start date of project: 1 Nov 2003 Duration: 1 Nov 2003 – 31 Oct 2007

Project coordinator: Dr. J. Köhl
Project coordinator: Plant Research International

Publishable Executive Summary

Summary description of project objectives

The objective of REPCO is to contribute to the replacement of copper fungicides in organic agriculture by new measures for control of downy mildew (*Plasmopara viticola*) in grapevine and scab (*Venturia inaequalis*) in apple. Both major European organic crops strongly depend on copper fungicides. Permitted amounts will be reduced stepwise during the following years (Council Regulation (EEC) 2092/91, Annex II) to avoid environmental risks. In European countries where copper fungicides are already out of use, production of organic apples suffers severe economical problems because of insufficient scab control.

Potentiators of resistance, organically based fungicides and biocontrol agents will be screened and evaluated in grapevine and apple. The risk of pathogen evolution during use of novel control measures will be estimated to allow the development of sustainable strategies. Effects of crop management practices in organic agriculture on overwintering of *Venturia inaequalis* will be assessed. Novel disease control measures and knowledge will be integrated into organic management systems. 'Pipeline' products already under development elsewhere will be included and where necessary optimised in their use.

Implementation by end-users and industries qualified for commercialisation of project findings will be strongly emphasised. SME partners will ensure a strong link between end-users and research. At the end of the project several compounds and biocontrol agents will be delivered to qualified industries for development of products for use in organic agriculture. Additionally, knowledge of integrated use of control measures will be delivered to organic growers.

The project will thus strongly support EU policies to replace the use of copper fungicides in organic agriculture in the nearby future.

Contractors involved

1. Plant Research International B.V. (PRI), Bornsesteeg 65, PO Box 16, 6700 AA Wageningen, The Netherlands; Project leader: Dr Jürgen Köhl; Tel. +31.317.476017; Fax +31.317.475701; E-mail: jurgen.kohl@wur.nl
2. Staatliches Weinbauinstitut (WBI), Merzhauser Strasse 119, D-79100 Freiburg im Breisgau, Germany; Project leader: Dr. Hanns-Heinz Kassemeyer; Tel. +49.761.4016530; Fax +49.761.4016570; E-mail: Heinz.Kassemeyer@wbi.bwl.de
3. Research Institute of Organic Agriculture (FiBL), Ackerstrasse, 5070 Frick, Switzerland; Project leader: Dr. Lucius Tamm; Tel. +41.62.8657238, Fax +41.62.8657273; E-mail lucius.tamm@fibl.ch
4. Swiss Federal Institute of Technology Zürich (ETHZ), Universitätstrasse 2, 8092 Zürich, Switzerland; Project leader: Dr. Cesare Gessler; Tel. +41.1.6323871; Fax +41.1.6321572; E-mail cesare.gessler@ipw.agrl.ethz.ch
5. Istituto Agrario di San Michele all'Adige (IASMA), Mach 13, 8010 S. Michele all"Adige, Italy; Project leader: Dr. Ilaria Pertot; Tel. +39.0461.615222; Fax +39.0461.650872; E-mail Ilaria.pertot@ismaa.it
6. Groupe de Recherche en Agriculture Biologique (GRAB), Chemin de la castelette, Site Agroparc P.O., Box 1222, 84911 Avignon, Cedex 9, France; Project leader: Cyril Bertrand; Tel. +33.4.90840170; Fax +33.4.90840037; E-mail direction.grab@freesbee.fr
7. Applied Plant Research (PPO), P.O. Box 200, 6670 AE Zetten; Lingewal 16, 6668 LA Randwijk, The Netherlands; Project leader: Dr. Bart Heijne; Tel. +31.488.473718; Fax +31.488.473717; E-mail bart.heijne@wur.nl
8. The Royal Veterinary and Agricultural University (KVL) Bülowsvej 17 DK-1870 Frederiksberg C, Denmark; Project leader: Dr. John Hockenhull; Tel. +45.35283308; Fax +45.35283310; E-mail johoc@kvl.dk

9. Danish Institute of Agricultural Sciences (DIAS), Kirstinebjergvej 10 P.O. Box 102 DK-5792 Aarslev, Denmark; Project leader: Dr. Hanne Lindhard Pedersen; Tel. +45.63904343; Fax +45.63904396; E-mail: hanne.lindhard@agrisci.dk
10. ECOVIN: Bundesverband Ökologischer Weinbau, Poststrasse 17, D-79423 Heitersheim, Germany Project leader: Dipl. Ing. Paulin Köpfer; Tel. +49.7634.552818; Fax +49.7634.552818; E-mail ecovin@t-online.de
11. Bio Fruit Advies (BioFruitAdvies), Dorpsstraat 32, 4111 KT Zoelmond, The Netherlands; Project leader: Marc Trapman; Tel. +31.345.502627; Fax +31.345.501721; E-mail m.trapman@wxs.nl
12. Prophyta Biologischer Pflanzenschutz GmbH (Prophyta), Inselstrasse 12, D-23999 Malchow/Poel, Germany; Project leader: Dr. Stefan Brückner; Tel. +49.384.252324; Fax +49.384.252323; E-mail sbrueckner@prophyta.com

Work performed and results achieved so far

Development of potentiators of resistance and organically based fungicides

Objectives during the reporting period were to collect candidate compounds for control of *Plasmopara viticola* (Pv) and *Venturia inaequalis* (Vi), to conduct a preliminary risk assessment for each compound and to select potentiators of resistance and organically based fungicides for Pv control in grapevine and Vi control in apple in a first series of screening experiments.

A list of more than 100 potential candidate materials for control of Vi and Pv was prepared. The list contains plant extracts and oils, products from micro-organisms, salts and other materials. Preliminary assessments of the materials covering availability and costs (economic feasibility), acceptability for organic growing and human and eco-toxicology were carried out.

A first series of agents for Pv control was tested on grapevine leaf discs (WBI). Seven compounds were indicated for further supporting tests so far. FiBL screened several compounds against Pv in indoors experiments and in a field experiment. KVL carried out a preliminary screening of over 20 materials using an in vitro assay designed to detect inhibition of Vi spore germination and a plant assay based on symptom development in apple seedlings artificially inoculated with Vi. Several promising materials showing significant reduction of Vi were identified. Screening work continues.

Development of novel biocontrol agents

Objectives during the reporting period were to build up collections of micro-organisms isolated from grapevine or apple leaves as candidates for antagonist screenings and to conduct a first selection round.

A protocol for a preliminary assessment of production costs and possible risks for candidate micro-organisms has been developed. More than 100 epi- or endophytic micro-organisms isolated from apple leaves were assessed (PRI). Twenty five candidates were applied in the orchard just before leaf fall for screening against Vi ascospore production. Possible hyperparasites were isolated from Vi infected leaf areas (>200 leaf samples; >400 isolates). Prophyta investigated the mass production of various fungal isolates to assess the economics of production of candidate antagonists. The economically sound production of various isolates seems to be feasible.

Components for Integrated Management Systems

Objectives during the reporting period were to (1) quantify the possible selection pressure of potentiators of resistance and organically based fungicides on Pv under controlled conditions; (2) to set up broad and group-specific PCR-DGGE systems applicable for green / senescent apple leaves and to quantify microbial populations in apple leaves after various treatments; and (3) to set up experiments on decomposition of apple leaves by earth worms.

ETHZ studied the change in ratio of genotypes (e.g. disappearance/dominance) under controlled conditions by inoculating mixes of sporangia suspensions from different genotypes of Pi. DNA was extracted from each generated lesion and analysis was performed with the SSR markers ISA, CES

and GOB. Preliminary test for the adjusting of the methodology and test if the available molecular markers are sensitive enough to recover selection pressure were conducted at Fibl in Frick. Sampling and epidemiological observations in an experimental parcel in Cugnasco (Ticino, Switzerland) were performed.
PRI adapted protocols for DNA extraction, PCR and DGGE-tools for analysing fungal and bacterial communities in and on apple leaves. Applying these molecular tools will include evaluation of the not yet cultured or unculturable micro-organisms which are expected to be present in apple leaves. Effects of cultural practises on micro-organisms naturally present on or in green and senescing leaves and their interaction with Vi ascospore production can now be evaluated. Samples from field experiments aimed at stimulation of leaf decomposition were obtained. Total microbial populations were determined by plating endo- and epiphytic bacteria and fungi. None of the treatments stimulated microbial colonisation on or in leaves. Several treatments reduced the size of microbial populations.
PPO carried out a literature review on (1) methods to monitor earthworms, (2) on methods to increase the palatability of apple leaves to earthworms and (3) on application of such measures in the field. A monitoring of earthworms was started in plots treated with different types of organic amendment with the aim to find methods that stimulate leaf consumption by earthworms. Substantial numbers of earthworms, including the major leaf eating earthworm *Lubricus rubellus*, were found. Information resulting from the literature review was used to set up a cultivation of the earthworm species, *Lumbricus terrestris* and *L. rubellus*, with the aim to test methods to improve the palatability of apple leaves. Feeding experiments are ongoing.

Integration of control measures
Field experiments were carried out in Italy and France in grapevine on Pv control combining natural compounds. In Italy (IASMA), all treatments, if compared with the untreated control, gave a certain control of the disease, but only treatments with copper based compounds (copper hydroxide and copper pepidate) and one compound not containing copper resulted in a commercially satisfying control of the disease. Other compounds tested may only be acceptable in low disease pressure seasons in organic vineyards. Several resistance inducers were totally ineffective in controlling the disease in the field.
In the two field experiments of GRAB (France), climatic conditions were not favourable for downy mildew development and average severity never exceeded 10 % in the non-treated control. Copper (reference) was more effective than other products tested. In the experiment at Montélimar, two compounds added to the lower dose of copper were more effective than the same dose of copper alone.
Two experiments were carried out by PPO to measure apple leaf decomposition and ascospore production of Vi. Treatments were applied in autumn by spraying just before leaf fall or by dipping. Leaf degradation was rather poor. Only urea (reference) enhanced leaf degradation. The numbers of ascospores in the remaining leaf tissue were very variable. No differences between treatments could be demonstrated except for urea.
During spring and summer, apple scab was controlled in an experiment according to EPPO-guidelines with natural compounds. Natural compounds were sprayed according the RimPro scab warning system from start of bud break until the mid of June. Severity and incidence was measured on the leaves and the fruit. Phytotoxicitiy and russeting was assessed. Schedules of natural compounds combined with sulphur were compared with the standard biological fungicides copper hydroxide and sulphur alone. Some of the treatments gave a level of control similar to copper in spite of a high infection pressure. However, some of the good-performing schedules produced substantial amount of russeting.

Expected end results

Novel potentiators of resistance, novel organically based fungicides and novel biocontrol agents for promising control strategies will be systematically screened and developed for use to control *Plasmopara viticola* in grapevine and *Venturia inaequalis* in apple. All screened and developed compounds and organisms will fulfil the requirements of Council Regulation (EEC) No 2092/91.

Novel integrated management systems will be evaluated, including exploitation of 'pipeline' products for disease control, such as already known, but not registered, potentiators of resistance, organically based fungicides and biocontrol agents.

The sustainability of novel control measures in respect to evolution of the pathogen population will be monitored.

Intentions for use and impact

The policy of the European Commission aims at the promotion of sustainable, quality-based agriculture production systems and the implementation of Council Regulation (EEC) No 2092/91 Appendix II Part B-IV on (reduced) use of copper fungicides. Derogation of the use of copper fungicides because of their specific environmental risks especially to soils, as scheduled for the period after 2006 (Council Regulation (EEC) No 2092/91), is threatening organic grapevine and apple production if alternatives for copper fungicides will not become available. REPCO will strongly support EU policy of promotion of sustainable, quality-based agriculture production systems by developing new organically based fungicides and potentiators of resistance, new biocontrol agents and new integrated management systems for disease control of *Plasmopara viticola* in organic grapevine and *Venturia inaequalis* in organically grown apple.

Scientific information obtained in the project will be published in research reports, scientific papers and contributions to international workshops and congresses. Partners ECOVIN and BioFruitAdvies form a platform to reach organic growers as the end-users of the developed technology. The regularly updated Website (www.rep-co.nl) contains information on partners, workpackages and project progress.

Results relevant for commercial exploitation will be scientific and technological knowledge on the use of potentiators of resistance, organically based fungicides and biocontrol agents as well as filed data relevant for registration on field experiments, production and formulation of such compounds and organisms. Commercial exploitation of results is foreseen by industrial partners.

Before publication of any information on knowledge and technology produced in the project, it will be evaluated whether specific details crucial for patent filing will not be made public before being protected by a patent. This will guarantee optimal exploitation of the results in respect to future commercialisation by interested industries.

Knowledge produced by the project which can directly be introduced in organic agriculture without the need of commercial product development by industrial partners, such as integration of existing control measures in management systems, will be communicated to extension services and end-users thereby facilitating a fast implementation in organic agriculture.