From 'topic description' to 'technical terms of reference'
&
Nomination of persons and institutes in charge

Wageningen, 7th December 2007

1 Introduction

The objective of this exercise is to establish the basis for the competitive call on the virtual pot (VP) topics and to proceed with the project initiation for the non-competitive (NC) topics.
For both VP and NC topics it is necessary to nominate a topic coordinator of the funder consortium. Although most of the necessary information for each topic is already available, it needs to be standardised and further defined in terms of scientific content and coordination responsibilities.

In this document, we request the VP topic coordinators, as volunteered in Ghent, to transform the 'topic descriptions' into a written form that will be used as the scientific terms of reference in the call documents. For your information, the VP topic descriptions are provided again in Annex 1.

As regard the NC topic, we can go a step further. The topic coordinator should identify a project leader, who then proceeds with elaborating a project description. Some volunteers for the important role of topic coordinator were put forward at the NMG meeting, however, we would like to gauge the interest of all participating institutes before making the final selection. For your information, the NC topic descriptions are provided again in Annex 1

The list of Euphresco Partners participating in each topic can be found in Annex 2.

In this document all text in red indicates a task that we ask you to execute.

2 Tasks for the VP topic coordinator

The topic descriptions should be further prepared for use in the call documents. For that it is necessary that they are transformed into basic terms of reference and checked to ensure these can be clearly understood by research providers. They will be very similar to the topic descriptions and most of the text can be 'cut and pasted' into the format for the project descriptions.
However, now we will need the additional information of the explicit support by all funding Partners interested in each topic. Not all topic descriptions were explicitly supported.

Note: in contrast to the NC mechanism, the project coordinator is selected by the bidding research consortia, and is therefore not known until the bidding process is complete.

The topic coordinators are:

EUPH02: *Erwinia amylovora* / On-site and laboratory diagnostics
Topic Coordinator: Sylvia Blümel (AT AGES)

EUPH04: *Potato spindle tuber viroid* (PSTVd) and other viroids / Studies informing risk assessment and risk management
Topic Coordinator: Steen Lykke Nielsen (DK DFFAB)

EUPH07: Grapevine flavescence dorée phytoplasma
Topic Coordinator: Hans Dreyer (CH FOAG)

EUPH13: *Ambrosia artemisiifolia* / Management and control methods / Risk management
Topic Coordinator: Katrin Kaminski (DE BBA)

EUPH18F: 'Kiln drying' / Efficacy testing
Topic Coordinator: Katrin Kaminski (DE BBA)

1. **Task for Topic Coordinator: verify that the intended research is unique and is not being carried out elsewhere. (we know that this check should have been carried out before, and probably it has been carried out by some of you)**

2. **Task for Topic Coordinator: Fill in the following outline using, adapting, amending the text in the topic descriptions. This will give the scientific base to be used in the call documents.**

Scientific Terms of Reference to be used in the call text

A. Project title
B. Background
C. Research need
D. Expected benefits

Keep in mind that the text in this 'scientific terms of reference' should allow both for novel approaches to addressing the research needs, as well as guaranteeing that competing researcher consortia have an equivalent understanding of the expected project.

3. **Task for Topic Coordinator: make certain that all Euphresco Partners who have shown interest in the topic agree on this rewritten text.**

4. **Task for Topic Coordinator: after agreement, arrange for these same Partners to sign a Letter of Intent (LoI) or Memorandum of Understanding (MoU), in which they confirm their commitment to the execution of the project by allowing one or more of their national research providers to participate in an international consortium. A draft proposal for this LoI or MoU (which will be detailed by Susanne/Manuela and made available later) can be found in Annex 3.**

5. **Task for all to whom it concerns: comment, if needed, on the draft proposal for this LoI / MoU.**

6. **Task for all, but oriented by the topic coordinator: as agreed in the Ghent meeting, steps 1 to 4 should be completed before Christmas (December 25th 2007), and reported to WP3.**

3 Tasks for participants in the NC mechanism

Under the non-competitive mechanism all four projects are ring tests or proficiency tests. Here it is necessary to distinguish:

1) a topic coordinator on the part of the funders, whose principal role is to guarantee the life line between the research project and the EUPRESCO Partners

2) project coordinator on the part of the participating institutes/labs (in contradiction to the VP mechanism, here we can already proceed with nominating such institutes and officials)
It seems more or less logical that 'topic coordinator' and 'project coordinator' are from the same country, but does not need to be so.

7. **Task to all Euphresco Partners participating in the NC mechanism: please inform WP3 if you would like to coordinate the NC topic.**

8. **Task to all participants in the NC mechanism: please inform WP3 whether one of your national institutes is interested in coordinating the project. Let us know before 21st December: Which institute you propose to take the lead and carry out all coordination? Please provide the name and contact details of a potential project coordinator.**

   Alan Inman suggested Neil Boonham (or colleague) at CSL as a potential project coordinator of

the virus (EUPH12) project; Maurice Moens (ILVO BE) and Paul van den Boogert (PPS NL) volunteered to coordinate the *Globodera* (EUPH06) project.

9. Task to all participants in the NC mechanism: please provide WP3 with the name of the participating institute and a contact person with whom the project coordinator can liaise. again, please let us know before 21st December.

After WP3 has made an inventory of the partners interested in leading the NC project, the participating institutions should coordinate amongst themselves, and produce a project document, for which instructions will be provided at a later date. Basically this project document would need to address issues including, but not necessarily limited to:

- who will produce the samples to be ring tested?
  [post scriptum: CSL volunteered to arrange the Rsol sample preparation and BBA the Cms samples for the Cms/Rs (EUPH03) pilot project; however a coordinator for the whole project would be needed.]
- invite non-Euphresco countries to participate.
- establish a project title (based on the topic description).
- define exactly what will be ring tested, including materials, methods, statistical procedures, quality criteria or certified protocols. (based on the topic description)
- design a budget for the project and for each collaborating institute (some financial contribution is essential for some coordinating meeting(s), and for production, purchase and postage and handling of materials)
- seek agreement in the form a letter of intent, committing all participating institutes to the success of this project.

Annex 1. Topic descriptions

EUPH02_Fireblight - Erwinia amylovora

1.1. Topic Area

Erwinia – diagnostics, including on-site detection

1.2. Project Title

Development and validation of innovative diagnostic tools for the detection of fire blight

1.3. Brief and concise description of the problem this research is going to solve

Erwinia amylovora, causal agent of fire blight, is a quarantine bacterium in Europe.

Rapid and on-site diagnosis, is critical for optimal implementation of phytosanitary control measures.

Different diagnostic screening tests for a rapid diagnosis of *E. amylovora* are commercially available, and new methods have recently been published. These have not yet been validated for phytosanitary applications and require ring-testing for validation. Validation is going to be performed including protocols recently revised by the European Plant Protection Organization (EPPO/OEPP) and it will support the preparation of the new protocol for the Food and Agriculture Organization of the United Nations (FAO/IPPC). Usually these tests are designed for application in lab settings and they may not be adequate for on-site diagnostics. Thus, innovative detection methods should be developed specifically suitable for on-site diagnosis.

Determination of asymptomatic infection is newly mandated by EU regulations, but currently no appropriate protocols are available. Sampling for asymptomatic infections is particularly difficult, since testing of a high number of randomised samples is necessary and the isolation of latent bacteria from woody tissues is challenging. Sampling protocols and tools are currently lacking and will be developed in this project.

The long-term control of fire blight requires eradication of inoculum reservoirs. Methods to source-track *E. amylovora* will be developed considering sampling protocols and molecular identification to the strain level.

1.4. Exact description of the research product this project will yield

New diagnostic tests (e.g., commercial PCR methods immunological tests) have been developed since the last European validation trials. The application of these tests for laboratory and on-site use requires validation in ring-trials. Innovative tests suited for on-site applications need to be developed in case the existing tools are insufficient. Recent advances with other diseases using immunological and on-site PCR tools will be used as models for developing similar tools for fire blight. Ring-trials with new tests will validate for application for:

i. detection of *E. amylovora* in asymptomatic plants on-site or in laboratories

ii. detection of *E. amylovora* in symptomatic plants on-site or in laboratories

1.5. Beneficiaries of this research product

1. Quarantine laboratories of several countries facing fire blight epidemics, (and countries which may face fire blight occurrence in the near future) as

i. Newly available methods are reviewed with respect to their specificity, sensitivity and performance. Innovative methods are developed as needed for on-site applications.

ii. the use of validated methods improves the quality of the laboratory

iii. the validation of methods in ring-tests is a major requirement for use in accredited laboratories

iv. the availability of validated screening test would allow larger number of samples in the detection of *E. amylovora* in asymptomatic plants

v. these methods are quicker and therefore labour costs are reduced (Note: this implies that the methods should be developed to be rapid as well as reliable and cheap)

2. Research laboratories, since the availability of a reliable screening method would allow accurate epidemiological studies to improve fire blight control measures

3. Plant protection service, since results are quickly available and decisions on phytosanitary measures can be reached earlier (especially in situations of imminent danger, e.g. in protected zones with a sudden disease outbreak)

4. Fruit growers and nurseries, since the availability of a reliable screening method would allow an economically feasible testing of plants intended for planting and would facilitate the development of certification schemes.

EUPH03_Brownrot and Ringrot - Clavibacter and Ralstonia

1.1. Topic Area

Ringtest on diagnostic methods

1.2. Project Title

Ringtest on diagnostic methods, including real-time PCR, for *Clavibacter michiganensis* ssp. s*epedonicus* (potato ring rot) and *Ralstonia solanacearum* (potato brown rot)

1.3. Brief and concise description of the problem this research is going to solve

The method of confirmation of *Clavibacter michiganensis* ssp. *sepedonicus* given in the revised directives RL 93/85/EWG and 28/57/EC is binding for all EU member states. In the directives RL 2006/56/EG *Ralstonia solanacearum* and 2006/63/EC the detection limits and the use of positive controls of defined concentrations of bacteria is fixed for the first time. The methods were developed within an EU project and tested in different laboratories in a ringtest, but without specifications for the detection limits.

Two laboratory tests are intended for the first investigation for verification of suspicions of infestation, PCR/FISH and IF-test. Both tests are considered equal related to the detection limit of 10^3 to 10^4 bacteria cells per ml resuspended potato pellet. This needs to be confirmed.

For isolation of *Clavibacter michiganensis* ssp. *sepedonicus* the semi-selective medium MTNA and the biotest are available, but sensitivity and reproducibility are not jet verified.

1.4. Exact description of the research product this project will yield

(a) *Clavibacter michiganensis* ssp. *sepedonicus* :This project carries out a ring test consisting of four parts:

1. Test of the sensitivity, specificity and reproducibility of the results of PCR/FISH/IF tests.
2. The EPPO protocol lists also a real time PCR as screening test. The equivalent of the method with PCR/FISH/IF must be tested. Real time PCR has not been fully validated and ring tested and therefore this is required before it can be introduced as part of the official scheme. Comparisons could be made with the existing methods (conventional PCR method, FISH and IF). The most appropriate gold standard would be IF since screening required 1 serological method (IF) and one molecular method to be positive. There is therefore a need for equivalent sensitivity/specificity.
3. Comparison of the different extraction techniques from potato pellets in connection with tests according to 1. and 2.

Standardized material must be provided to all participants in order to achieve comparable results in the different laboratories and to avoid systematically errors. The standardized material comprises positive samples (10^3 – 10^4 bacteria cells per ml and 10^6 bacteria cells per ml) which are suitable for isolation. The material must be converted in a manner that guaranties the survival of the bacteria. Furthermore, the production of natural infested tuber material is required which could be used for the infestation of healthy potato heel end cores and the on-site production of diseased potatoes with 200 heel end cores in total. Could also use real positive samples diluted into negative samples to obtain the required pathogen concentrations. Antisera, standard primer sets and dNTPs are requisite.

The participating laboratories obtain the samples anonymous with hints for the subsequent use. They get 10 samples at the most, whereof four samples are in the range of the detection limit and the other samples are negative. Strict guidelines for storage of the samples, timeframe of tests, specification of the methods and the interpretation of the tests are prescribed.

(b) ***Ralstonia solanacearum:*** This project carries out a ringtest consisting of five parts:

1. Production of potato extracts according to the standard EU method. This could be done by pooling extracts which have already tested negative in national surveys or by sampling a large number of healthy potatoes.
2. Diluting extracts from infected potatoes (either naturally or artificially infected) in healthy potato extract to obtain the pathogen at the required concentrations 5x10^3 and 5x10^6 cells per ml. Prepare extracts with ring rot and brown rot pathogens. Prepare 5 ring rot samples and 5 brown rot samples. (Ring rot samples will serve as brown rot negative samples and vice versa).
3. Verify pathogen populations by IF. Aliquot and freeze dry samples for dispatch as blind samples.
4. Rehydrate extracts in laboratory and test using IF and real-time PCR protocols. Optionally use other laboratory tests (e.g. FISH, conventional PCR). Use Pastrik DNA extraction method (as in EU directive is only an option) and optionally use other commonly used extraction methods for comparison.
5. Quantify pathogen populations in each sample and return results for centralised analysis on pre-prepared data sheets.

1.5. Beneficiaries of this research product

The project offers validation and standardisation of the diagnostic methods used by national plant protection services and other relevant laboratories. The project would provide data for potential inclusion of real-time PCR methods in EC directives and would therefore support implementation of EC policy.

1.6. Any other relevant information

WP3/4 and/or Network Management Group and/or the funders interested in this topic will need to consider how to fund the organisation of the ring test, including the provision of ring test materials. Several possibilities exist for this, but consideration should be given to establishing a system that might be suitable for all other current and future ring tests.
Consideration should also be given to participation of ring testers in countries who are not involved in EUPHRESCO. Contact with the European Association of Phytobacteriologists might facilitate wider participation of ring testers in non-EUPHRESCO countries. additional ring testing participants under, potentially, their existing resources.
Further details of the ringtests need to be defined and agreed on by the research consortium.

EUPH04_PSTVd and other viroids

1.1. Topic Area

Epidemiology and diagnostics in support of risk assessment and risk management.

1.2. Project Title

Contributing elements to pest risk analysis for PSTVd and other viroids in ornamental and wild plants – occurrence in different host plants, investigation on potential pathways and harmonisation of testing methods

1.3. Brief and concise description of the problem this research is going to solve

As a consequence of findings of PSTVd in plants of *Solanum jasminoides* and *Brugmansia* spp. in several Member States of the EU in 2006 the Commission Decision 2007/410/EC provides for emergency measures to prevent the further spread of PSTVd with these plants. Recent experience gives rise to the assumption that PSTVd and other pospiviroids that infect potato and/or tomato may also be present in other ornamental plant species (e. g. *Solanum rantonettii*, *S. muricatum*, *Petunia* spp.). Currently there is no knowledge about possible occurrence of these pospiviroids in wild grown plant species of Solanaceae. In relation to risk assessment for potato and tomato crops it is essential to know the actual status of PSTVd and these other pospiviroids of potential relevance for these important crops; this requires good diagnostic tools and an understanding of some basic biology and pathways for introduction and spread.
In order to realistically judge the potential risk of viroid transmission from ornamental or wild grown host plants to potato or tomato crops the existence and likelihood of different pathways (like mechanical transmission during cultural practices, aphid transmission, transmission with pollinating insects, seed transmission) has to be evaluated.
Diagnosis of viroids is not a trivial task. A variety of methods has been published, including return-PAGE, nucleic acid hybridization, reverse transcription-PCR, real-time PCR. Each of these methods has advantages and disadvantages in respect of sensitivity, specificity, reliability, suitability for different host plants or host tissues, speed and labour requirements. Until now the EPPO protocol includes only the diagnosis of PSTVd in potato leaves. Approved protocols for the diagnosis in other host plants (e. g. tomatoes, solanaceous ornamentals) and other tissues (e. g. seeds) are required. For surveillance purposes a set of reliable and robust methods applicable under different laboratory conditions should be chosen, compared and if necessary further improved. A ring-test will be carried out.

1.4. Exact description of the research product this project will yield

The project will complement the official survey according to article 3(1) of Decision 2007/410/EC carried out by the Member States on a systematic and harmonised basis. A rough judgement will be possible whether PSTVd (and other viroids) already might be present in wild plants, thus increasing the risk for potato or tomato crops.
Transmission experiments carried out under 'worst case' conditions will indicate whether there are potential pathways for viroids spreading from ornamental or wild plants to potato and tomato crops. Results will give a first indication whether such events will result in stable establishment in these crops. The relevance and probability of the different pathways will be assessed.
Diagnostic methods (e. g. for tomato seeds) will be improved and proposed to the EPPO. A set of ring-tested methods for detection and identification of different viroids in different host plants will be available for use by the Plant Health authorities responsible for surveillance and control of plant pests.

1.5. Beneficiaries of this research product

The results will provide important elements for pest risk analysis. The project will underpin future policy decisions (e.g. in the Standing Committee on Plant Health) in relation to improving phytosanitary regulations aiming at prevention of the spread of viroids in important agricultural and horticultural crops. Plant Health Services of the Member States who are in charge of surveying and testing crops for the presence or continued absence of viroids will benefit from the harmonisation of diagnostic methods within the EU.

1.6. Any other relevant information

For the project part dealing with ring tests it could be useful to include laboratories (at their own expense) other than the research consortium and perhaps also from other countries than EUPHRESCO participants.

EUPH05 Pantoea stewartii ssp. stewartii

1.1. Topic Area

Diagnostic ring tests

1.2. Project Title

Ring test on diagnostic methods for *Erwinia stewartii* ssp. *stewartii* (*Pantoea stewartii* ssp. *stewartii*)

1.3. Brief and concise description of the problem this research is going to solve

For the EU member states there is no harmonized diagnostic method for *Erwinia stewartii* ssp. *stewartii* (*Pantoea stewartii* ssp. *stewartii*). The EPPO protocol PM 7/60 (1) lists several tests for the first investigation and the following isolation/screening and identification. These tests haven't been tested in ring tests so far. For the preliminary examination IF and ELISA and conventional PCR are proposed. The EPPO protocol contains no information on sensitivity, specificity and reproducibility. In addition, naturally infested seeds were not used to development these protocols. If real-time assays are available, these will also be considered for inclusion in the ring test.

1.4. Exact description of the research product this project will yield

This project envisages a ring test consisting of four parts, though a more detailed project specification will be defined and agreed by the ring testing consortium in line with the overall objectives outlined by the EUPHRESCO participating Partners/funders:

1. Determination of the sensitivity, specificity and reproducibility of the different methods.
2. Comparison of different media for the isolation of the bacteria, and comparison of isolation with other tests, using artificially contaminated seed.
3. Test of the sensitivity, specificity and reproducibility of the results of methods used so far (IF and ELISA) and the PCR (with previously released primers) with naturally infected seed, or with artificially contaminated samples if naturally infected seed is not available.
4. A key issue will be sample size in relation to levels and probability of detection: comparison of methods using replicates of 400 seeds will be done.

Standardized material must be provided to all participants in order to achieve comparable results in the different laboratories and to avoid systematically errors. The standardized material comprises positive samples (10^3 – 10^4 bacteria cells per ml and 10^6 bacteria cells per ml) which are suitable for isolation. The material must be converted in a manner that guaranties the survival of the bacteria. Furthermore, natural infested seed, antisera, standard primer sets and dNTPs are required.

The participating laboratories obtain the samples anonymous with hints for the subsequent use. They get 10 samples at the most, whereof four samples are in the range of the detection limit, one sample is naturally infested and the other samples are negative. Strict guidelines for storage of the samples, timeframe of tests, specification of the methods and the interpretation of the tests are prescribed.

1.5. Beneficiaries of this research product

The project offers validation and standardisation of the diagnostic methods used by national plant protection services and other relevant laboratories.

1.6. Any other relevant information

WP3/4 and/or Network Management Group and/or the funders interested in this topic will need to consider how to fund the organisation of the ring test, including the provision of ring test materials. Several possibilities exist for this, but consideration should be given to establishing a system that might be suitable for all other current and future ring tests.

Consideration should also be given to participation of ring testers in countries who are not involved in EUPHRESCO, especially Observers. Contact with the European Association of Phytobacteriologists might facilitate wider participation of ring testers in non-EUPHRESCO countries. As this Project is currently assigned to the non-competitive mechanism, this should be able to accommodate additional ring testing participants under, potentially, their existing resources.

The consortium should especially try to involve researcher from Hungary who might perhaps be able to provide naturally infested seed.

Linkages with ISTA (International Seed Testing Association) might usefully be considered; and also potentially FAO-IPPC.

EUPH06_Globodera

1.1. Topic Area

Development and validation of diagnostic methods

1.2. Project Title

Development of a simple test for identification of the pathotypes of potato cyst nematode (*Globodera rostochiensis* and *G. pallida*)

1.3. Brief and concise description of the problem this research is going to solve

Potato cyst nematodes (PCN) *Globodera pallida* and *Globodera rostochiensis* cause serious damage to potato crops world-wide by restricting root growth and uptake of nutrients, leading to a loss in tuber yield. At present, PCN are thought to account for losses of more than 9% of the average potato crop yield in Europe (Turner, Row, 2006). That is the reason PCN to be included in A2 list and to be of a scope of specific Council Directive 69/465/EEC3 on the control of potato cyst eelworm.
Cultivar resistance plays an important role in managing PCN. Even though many potato cultivars that are totally resistant to *G. rostochiensis* exist, none of them are fully resistant to *G. pallida*. Moreover, the use of *G. rostochiensis* resistant cultivars has led to an increase in the prevalence of G. pallida (Cook, Star, 2006). Some of the populations are homozygous for virulence (e.g. Ro1 for *G. rostochiensis* and Pa1 for *G. pallida*), but most populations of *G. pallida* and *G. rostochiensis* are heterogeneous and give varying results. The application of the standard schemes of Kort et al. (1977) to establish the pathotype, unfortunately is not relevant here as other and more resistance sources are used.
The most usable PCR methods for species identification and taxonomy are often presented to be RAPD (Williams et al. 1990, Roosien et al. 1993, Folkertsma et al. 1994, Blok et al. 1997, Bendezu et al. 1998, Thiery et al. 1997 and Fullaondo et al. 1999), STS (Zouhar et al. 2000 and Skupinova et al. 2002) and RT PCR (Kenyon, 2006).
Establishment of validated set of protocols for PCN pathotype identification is important for the accuracy of monitoring and control management of the parasites in the EU area.

1.4. Exact description of the research product this project will yield

The main goal of the project is to estimate and validate all existing methods of molecular diagnostic of PCN pathotypes. All existing methods and applied protocols will be estimated and validated in ring test of all partner laboratories. Selected methods will be tested on single and mixed field PCN populations. Referent collection to differentiate pathotypes of *Globodera pallida* and *G. rostochiensis* will be established in order to facilitate future monitoring and management programmes in the EU area.

1.5. Beneficiaries of this research product

- Diagnostic laboratories and inspection services in member states. Monitoring programmes.

The monitoring to be carried out will be more reliable and will provide up-to-date information on the distribution of the species. The establishment of a referent collection will significantly facilitate correct identification and therefore the elaboration of programs for control of PCN.

- Producers cultivating potatoes.

In regions where PCN are not yet established, producers will have the opportunity to modify the conditions for cultivation so that no threat of establishment of the parasites is present; in regions where PCN are already established, it will be possible to undertake adequate measures for their control.

- Validation of diagnostic protocols

The project will provide opportunity to demonstrate how trans-national cooperation can facilitate the validation of diagnostic protocols

EUPH07_Grapevine flavescence dorée

1.1. Topic Area

Risk management

1.2. Project Title

Evaluation of the risk of spread of *Scaphoideus titanus*, the vector of Grapevine Flavescence dorée, with commercial grapevine propagation material

1.3. Brief and concise description of the problem this research is going to solve

Flavescence dorée (FD) is a yellows disease of grapevine, caused by phytoplasmas of the elm-yellows (16Sr-V) group. It is a quarantine disease in Europe. FD is graft transmissible and can be spread with grapevine propagation material. However, disease outbreaks depend on the presence of the vectoring leafhopper *Scaphoideus titanus*, a nearctic species that has been introduced to France presumably in the early 20th century. The vector is present around the 45th degree of latitude and spread already from France to Spain and Portugal, Italy and Slovenia. Recently, also Serbia and southern areas of Hungary, Austria and Switzerland have been colonized. *S. titanus* is currently extending its range to the north, presumably due to changing climate variables. However, studies of the genetic structure of European populations revealed dissemination with grapevine material as the most probable means of spread. This is possible, because this strictly ampelophagous species deposits its eggs in the bark of one and (mostly) two year old wood of grapevine, on which it fulfils its whole life cycle.

FD causes severe damage to viticulture in southern viticultural areas e.g. in France and Italy. In addition, efforts to minimize pesticide use are impeded by the regular insecticide treatments against the vector, which are required to disrupt the epidemic cycle. Preventing the further spread of *S. titanus* is therefore an important phytosanitary measure not only for countries that are completely free of this pest like Germany but also those where the distribution is still restricted. Additional information is required on the risk of introducing eggs of *S. titanus* with grapevine propagation material and the chance that viable populations could develop from those eggs in vector free areas.

The proposed project consists of three parts:
Part I - Significance of rootstock and scion propagation material as carriers of *S. titanus* eggs
Part II - Potential role of propagation material for the spread of *S. titanus*
Part III - Risk of establishment of viable populations of *Scaphoideus titanus* after introduction

1.4. Exact description of the research product this project will yield

Information on the infestation of propagation material with *S. titanus* eggs
Information on the susceptibility of eggs to the propagation procedure
Estimation of the risk of spread of *S. titanus* with propagation material into vector free areas and its potential to establish in vine growing regions north of the 46th degree of latitude..

1.5. Beneficiaries of this research product

The project will reveal additional information about the risk of introduction of *S. titanus* for vector free areas. Data on the effects of the propagation process on *S. titanus* eggs and on the significance of waste wood for the dissemination of the vector will help nurseries to minimize the risks by adapting the propagation practice or appropriate treatment of the waste material.

EUPH12: Whitefly viruses

1.1. Topic Area

On-site and laboratory diagnostics

1.2. Project Title

Validation of diagnostics methods for the detection and identification of whitefly transmitted viruses on regulatory or quarantine concern to the EU

1.3. Brief and concise description of the problem this research is going to solve

The project would validate new real-time PCR diagnostic methods for a range of whitefly-transmitted viruses of concern to Europe, but currently most damaging to Mediterranean horticulture. These protocols would be ring tested against existing 'gold standard' methods and EPPO protocols produced or revised. Additionally, some on-site (in-field) methods would be validated, e.g. Lateral Flow Devices (LFD) for TYLCV and potentially also for CYSDV. Specific work might include:

- Validation by ring testing of real time PCR assays for TYLCV, CVYV, CYSDV, TiCV and ToCV. This might involve an initial training element/workshop.
- Depending on resources and time, adaptation of real-time PCR assays into on-site formats using, for example, the Smartcycler platform. Protocols could be validated by ring testing.
- Further laboratory-based validation, and new in-field validation in Mediterranean countries where the virus is commonly found, of a TYLCV LFD; this would involve comparative testing against lab-based methods, e.g. PCR.
- Potential laboratory-based and in-field validation of a prototype LFD for CYSDV, though this would be dependent on whether a prototype can be successfully in time for the pilot projects (from an existing Defra/CSL project); this would involve comparative testing against other lab-based methods, e.g. PCR.

1.4. Exact description of the research product this project will yield

Not only describe the end product but also intermediate products and deliverables.

1.5. Beneficiaries of this research product

- Diagnostic laboratories and inspection services in member states.
- Although these predominantly whitefly transmitted viruses are mainly of concern to Mediterranean horticulture, other European countries also have a concern with some of these viruses. Some are EC listed (TYLCV); others EPPO listed (EPPO A2 List: CYSDV, CVYV, ToCV, TYLCV; EPPO Alert List: TiCV) and several are on the EPPO Action list (e.g. ToCv, CYSDV, CVYV)
- The Project would offer a good opportunity to demonstrate how cooperation between countries can progress the validation of diagnostic protocols, e.g. in-field validation in countries where the pathogens occur.
- The Project would promote and validate a real-time PCR.

1.6. Any other relevant information

WP3/4 and/or Network Management Group and/or the funders interested in this topic will need to consider how to fund the organisation of the ring tests, including the provision of ring test materials. Consideration should also be given to participation of ring testers in countries who are not involved in EUPHRESCO.

EUPH16 Ambrosia

1.1. Topic Area

Management and control

1.2. Project Title

Optimising control of *Ambrosia artemisiifolia*

1.3. Brief and concise description of the problem this research is going to solve

The ongoing spread of Ambrosia artemisiifolia in Europe is threatening to increase its impacts on plant health and on human health, which together cost in the range of hundreds of millions of €. Some countries have already taken steps against this spread, others are about to follow. The EPPO is supporting countries by also preparing recommendations on how to control this plant. Whereas a lot of information on control of *A. artemisiifolia* is available, there are specific knowledge gaps. They concern specifically:

- the reaction of plants to mechanical damage (as by, e.g., mowing) in various developmental stages,
- the best choice of herbicides, their application timing and mode,
- the best way to suppress germination of Ambrosia seeds from the seed bank.
- herbicidal effects on the viability of seeds
- how to best disrupt the life cycle in one action
- combination of mechanical and chemical methods for control along road sides
- questions about composting of *A. artemisiifolia*

1.4. Exact description of the research product this project will yield

The project will produce a set of guidelines for the control of *A. artemisiifolia* based on a detailed scientific understanding of the plant's reaction to specific management practice.

1.5. Beneficiaries of this research product

Farmers, road services, gardeners (privates and professionals) and the broad public in European countries in which *A. artemisiifolia* is already present and widespread as well as those where the species is beginning to invade and spread. The benefit to farmers will consist of lower weed impacts of *A. artemisiifolia* in agricultural fields, the general public will experience less health impacts, when a national program of Ambrosia control can be based on a sound set of guidelines.

EUPH18F Kiln drying

1.1. Topic Area

Management

1.2. Project Title

Risk Analysis and efficacy analysis of “kiln dried” (K.D.) as phytosanitary treatment prior to import laid down in Directive 2000/29/EC for wood and wooden product

1.3. Brief and concise description of the problem this research is going to solve

Several wood samples from packaging wood intercepted during import inspection and analysed at the BBA contained live *Bursaphelenchus* species as well as live *Monochamus* larvae which were able to develop to adult beetles. The moisture content was between 12 and 14% and so much lower than the

requirement of 20% or less after K.D. treatment according to Directive 2000/29/EC. It is suspected, that the KD requirement often used for wood in Annex IV A I may not be sufficiently effective under all circumstances.

1.4. Exact description of the research product this project will yield

The aim of the proposed project is to determine whether organisms of phytosanitary concern, particularly those mentioned in Directive 2000/29/EC, can survive the currently stated K.D. requirement and whether this is linked to particular life stages or strategies of pest organisms.

Approach: Laboratory testing within the proposed project will be carried out with naturally and artificially infested wood. Starting with a high moisture content, development of the associated organisms (at least 2 insect species, 1 nematode and 1 fungus) will be assessed during controlled lowering of the moisture content. A comparison between technical drying of the wood in a laboratory kiln and drying under outdoor conditions will be carried out. Different life stages of the test organisms will be investigated. The work will examine the two principal variables of decreased moisture content and artificially applied elevated temperature during the kiln-drying process separately and in combination. The context for inclusion of temperature is for comparison with the requirements for heat treatment e.g. as in the International Standard for Phytosanitary Measures (ISPM) No. 15 for wood packaging which includes Heat Treatment to a core temperature of 56^{o}C for 30 minutes.

1.5. Beneficiaries of this research product

Current doubts on the efficiency of applied methods for treatment of wood and wooden products prior to import require testing of the method in order to assure that efficient methods are applied. Introduction of new pest organisms via wood and wooden products in the EU could only be achieved by efficient treatments of wood and wooden products.

EUPH24 Aquatic invasive weeds

1.1. Topic Area

Invasive alien (non-native) aquatic weeds

1.2. Project Title

Development of methods for eradicating invasive alien (non-native) aquatic and riparian weeds relevant to the UK, the Netherlands and the wider EU.

1.3. Brief and concise description of the problem this research is going to solve

Within the European Commission, invasive alien weed species are the policy responsibility of The Commission's DG SANCO. Little research has been done on the management of invasive alien weeds, though there has been some focus on risk assessment. There is a need for the development of effective but environmentally sensitive methods for controlling/managing outbreaks of invasive weeds on environmental concern. Associated with this is the need for expertise to be both developed and shared at the European level. Control of aquatic non-native weeds presents one of the most challenging areas due to the highly invasive nature of many aquatic species, their ease of spread within and between waterways and their large economic, social and biodiversity impacts. Furthermore there are significant constraints imposed on the type of chemical approaches that can be used in water-courses. There are also problems with physical removal for some species since tiny fragments can regenerate new plants and exacerbate the spread. Currently, the species of aquatic weeds most concern and relevant to the UK/NL include species of *Azolla filiculoides*, *Cabomba caroliniana*, *Crassula helmsii*, *Egeria densa*, *Elodea nuttallii*, *Hydrocotyle ranunculoides*, *Lagarosiphon major*, *Ludwigia grandiflora, Myriophyllum aquaticum.* Proposed work might also extend to riparian invasive alien weeds.

1.4. Exact description of the research product this project will yield

The research will yield the following outputs:

- Generic and/or specific methods for the eradication/control of serious invasive, alien aquatic or riparian weeds of concern to the funding countries (UK/NL) and to the wider EU.
- Decision support systems to help managers: identify water systems that are at risk, e.g. based on water quality parameters; decide what control methods to use and how to make best use of different methods, including integrated approaches which combine appropriate chemical control, physical control, approved biological control agents, or other novel techniques.
- Methods which will help MS meet the future requirements of the EC Water Framework Directive (WFD) which requires EU waterways to achieve good ecological status by 2015.
- Sharing of expertise and development of increased EU capacity/capability in the area of invasive alien aquatic weed control.

1.5. Beneficiaries of this research product

The beneficiaries of the outputs will be:

- Government bodies (especially those that are tasked with implementing the WFD), in the funding countries (UK/NL) that have policy responsibility for aquatic/riparian invasive alien weed species.
- The wider EU (member states and DG SANCO) or EPPO region through increased sharing and information exchange on methods and an increased European capacity in this area.
- The scientific community and water/fisheries managers etc. working in this area, through collaborative research on shared needs and through exchange of expertise.

1.6. Any other relevant information

See main paper for how the real pot will operate in terms on the bidding process (2-stage process to encourage bids; and possible competition with a second topic), eligibility (contractors from funding countries, but sub-contracting to science providers outside these countries is allowed if there is a proven need), weighting of criteria (collaboration will be encouraged), project length (will consider projects longer than just 12-15 months, e.g. up to 18–24 months), etc.

EUPH25 Whole Genome Amplification (WGA)

1.1. Topic Area

Validation and harmonisation of methods supporting DNA banks and diagnostics

1.2. Project Title

Validation and harmonisation of whole genome amplification (WGA) techniques for quarantine pests/pathogens of plants relevant to the UK/NL and wider EU.

1.3. Brief and concise description of the problem this research is going to solve

Within the European Commission, quarantine plant pest species are the policy responsibility of The European Commission's DG SANCO. National diagnostic or National Reference Laboratories (NRL) play a key role in diagnosis of quarantine/regulated plant pests and diseases. Highly qualified reference DNA material is required to develop, validate, harmonize and standardize molecular diagnostics. Diagnostic laboratories serving national Plant Health Services often face limited DNA quantities, due to the absence or time-consuming *in vitro* cultivation methods of the target organisms. The use of cloned DNA fragments can be a solution to create an unlimited source of nucleic acid; however, these clones do not represent the complete genome and can only be used for assays amplifying the specific target. WGA is a way to increase the amount of DNA from small samples and, contrary to cloning, representing the complete genome for a diversity of down-stream processes, e.g.

molecular identification and detection. WGA is particularly useful for non-cultivable or biotrophic pathogens, or of single/limited individuals in collections, e.g. insects.

Only limited work on WGA and DNA storage has been done for quarantine plant pests/pathogens. There is a need to validate WGA kits for their performance to efficiently amplify whole genomes, in particular for organisms of low availability (NB. the project should also address WGA in standard/conserved specimens). Associated with this is the need for expertise to set markers for WGA and storage conditions for DNA samples.

1.4. Exact description of the research product this project will yield

The research will yield the following outputs:

- Published/publicly available generic, harmonised protocols for WGA on reference species and target groups or species of concern to the funding countries UK, NL and to the wider EU.
- Published/publicly available protocols (accepted and ring tested) for durable storage of amplified DNA samples which will help inspection services meet the quality standards imposed by authorized reference laboratories in the domain of Plant Health.
- Sharing of expertise and development of EU capacity/capability in the area of WGA and DNA storage supporting European diagnostic collaboration and possibly linked to EPPO Standards.

1.5. Beneficiaries of this research product

The beneficiaries of the output will be:

- The plant health services, or other government bodies (especially those that are tasked with implementing diagnostics), in the funding countries UK, NL that have policy responsibility for quarantine plant pests and diseases.
- The wider EU (member states and DG SANCO) or EPPO region through increased sharing and information exchange on methods and an increased European capacity in this area.
- The scientific community working in this area, through collaborative research on shared needs and through exchange of expertise.

1.6. Any other relevant information

See main paper for how the real pot will operate in terms on the bidding process (2-stage process to encourage bids; and possible competition with a second topic), eligibility (contractors from funding countries, but sub-contracting to science providers outside these countries is allowed if there is a proven need), weighting of criteria (collaboration will be encouraged), project length (will consider projects longer than just 12-15 months, e.g. up to 18–24 months), etc.

Annex 2 EUPHRESCO PARTNERS' PARTICIPATION IN PILOT TOPICS

Partner Number & Name	Non-competitive mechanism (NC)				Virtual Common Pot mechanism (VP)						Real Common Pot mechanism (RP)	
	Ringrot brownrot	Pantoea	Globodera	Whitefly Viruses	Fireblight	PSTVd	Grapevine FD	Ambrosia	Kiln drying	Total	Aquatic weeds	WGA
	EUPH03	EUPH05	EUPH06	EUPH12	EUPH02	EUPH04	EUPH07	EUPH16	EUPH18F	VP funds	EUPH24	EUPH25
1 UK-CSL	Y	Y		Y						0		
2 UK-DEFRA	Y	Y		Y		30-50				30-50	100^	
3 AT-BMLUFW					30**	30**	**	**	30**F	90		
4 AT-AGES		Y			30	**	**	**		30		
5 BE-ILVO	Y		Y	Y						0		
6 BE-FPS												
7 BG-NSPP	Y		Y	Y						0		
8 CY-ARI				Y		10				10		
9 CZ-NAAR										0		
10 DK-DFFAB						150		150		300		
11 FI-MMMFI	Y					NC				0		
12 FR-DGAL	Y	Y	Y	Y	25	5				30		
13 FR-INRA					10	10	10	10		40		
14 DE-BMELV						30		30	30	90		
15 DE-BBA	Y	Y								0		
16 IE-DAF			Y							0		
17 IT-MPAF			Y				30		60	90		
18 IT-CRA										0		
19 NL-PD	Y		Y	Y		30				30		
20 NL-LNV										0	150^	
21 SL-MAFF					25	25**	**	**	30	80		
22 ES-INIA			Y		30					30		
24 CH-FOAG					30		30	30		90		
24 TR-GDAR	Y	Y	Y	Y						0		
Number of Partners participating	9	6	7	8	7	11	6	7	4		2	
	15				14						2	
	20											
Budget	N/A				Totals per topic depend on final allocation						250	

Annex 3 Draft proposal LoI (or MoU) for Virtual Common Pot Funder Consortium

LoI (or MoU) for Virtual Common Pot Funder Consortium

Date:

[fill in date]

This is to confirm that

[fill in full name of funding partner (institute, partner number)]

is committed to participating as a funding partner in the first EUPHRESCO pilot calls. We will contribute a total of

€ [fill in amount]

to this call, divided over specific projects in the following way:

Project title	Confirmed budget, in €

[add lines as needed]

Yours,

[add signature of authorized person]

[add name and position of signing person]