

1. Content of the 'Topic Description' document

1.1. Topic area

E: Pest/vector characterization: genetics, biochemistry, etc.

1.2. Links to the Euphresco Strategic Research Agenda

The topic addresses the following objective(s) of the 2017-2022 Euphresco Strategic Research Agenda:

- ☒ Objective 2017-I-2.2: to contribute to databases for plant pests identification and diagnostics
- ☒ Objective 2017-R-5.4: to test and validate the use NGS (e.g. whole genome sequencing, metagenomics, deep sequencing, typing by sequencing) for routine diagnostics

1.3. Topic title

mtDNA characterisation of potato wart disease outbreaks.

1.4. Description of the problem the research should solve

Potato wart disease is caused by the quarantine soil-borne fungus *Synchytrium endobioticum*. This obligate biotrophic fungus can persist in infested fields for more than 30 years, even in absence of the host. Production of seed and ware potatoes is prohibited in contaminated fields, while the use in the safety zone of potato varieties resistant to the pathotype(s) present in the contaminated fields and strict phytosanitary measures are currently the only options for disease management (Council Directive 69/464/EEC). However, the pathogen develops new pathotypes which overcome varietal resistance. In Europe, the number of reported new outbreaks of potato wart is relatively low, but chances that new pathotypes and/or new outbreaks will arise are much greater than the probability of a new introduction from South America (reviewed by Obidiegwu *et al.*, 2014). This underlines the importance of a thorough molecular characterization of outbreak sites reported in Europe.

Research on potato wart disease is often complicated by the availability of material. Material related to outbreaks is sometimes stored as compost or as extracted resting spores. Vitality can be problematic, so that the material is no longer available through traditional propagation techniques. Recently, the annotated nuclear genome and mitogenomic sequence of *S. endobioticum* have become available (Van de Vossen *et al.*, 2018; 2019), introducing the possibility of track-and-trace the pathogen. In addition to the sequence of the pathotype 1(D1) reference strain MB42, genomic sequences of about 30 additional isolates are available. A preliminary analysis of the genome sequences shows that it is possible to molecularly trace the isolates on the basis of mitochondrial sequences. When non-vital collection material is also included in the analysis, this could place documented outbreaks in a historical perspective.

In this project, we aim to collect *S. endobioticum* material from a wide variety of sources (compost, resting spores, and herbarium specimens), including non-vital material. We want to generate at least 100 mtDNA genome sequences and build a reference database which can be used for track-and-trace. Apart from generating genotypic data, metadata regarding origin, the potato variety from which the fungal material was obtained, pathotype identity and pathotyping method including scoring results, and soil type will be collected.

In addition, the diversity among fungal populations with regard to the recently identified *AvrSen1* gene and its variants will be assessed. The *AvrSen1* gene product is recognised by the *Sen1* resistance gene which is underlying pathotype 1(D1) resistance. The identification of *AvrSen1* variants in *S. endobioticum* isolates is an important step towards a molecular pathotyping system that is built on Avr-R gene interactions.

References



- Obidiegwu *et al.*, 2014. Managing potato wart: a review of present research status and future perspective. Theor Appl Genet. 2014; 127(4): 763–780
- Van de Vossenbergh *et al.*, 2018. The linear mitochondrial genome of the quarantine chytrid *Synchytrium endobioticum*; insights into the evolution and recent history of an obligate biotrophic plant pathogen. BMC Evolutionary Biology 18:136
- Van de Vossenbergh *et al.*, 2019a. Comparative genomics of chytrid fungi reveal insights into the obligate biotrophic and pathogenic lifestyle of *Synchytrium endobioticum*. Scientific Reports 9: 8672
- Van de Vossenbergh *et al.*, 2019b. The *Synchytrium endobioticum* AvrSen1 triggers a Hypersensitive Response in Sen1 potatoes while natural variants evade detection. BioRxiv <https://doi.org/10.1101/646984>

1.5. Description of the expected results

Accessible database with mtDNA genome sequences of *S. endobioticum* for track and trace purposes. Protocols to determine the complete mitochondrial genome sequences of new samples *S. endobioticum* in a similar way.

1.6. Beneficiaries of this research product

National Plant Protection Organisations; National and EU policy makers; farmers, potato industry and other stakeholders; EPPO.

1.7. Research funders and research contribution/ distribution

Funding organisation	Research activity and researchers involved
<p>1. Netherlands Food and Consumer Products Safety Authority, the Netherlands</p> <p>Martijn Schenk m.schenk1@nvwa.nl</p>	<p>-Project coordination; -Make available collection material (>100 stored compost samples);</p> <p>Contact person: Bart van de Vossenbergh E-mail: b.t.l.h.vandevossenbergh@nvwa.nl</p> <p>-Isolate spores from collection material; - Perform DNA/RNA extraction, run a quality check on the DNA, analyse the samples using Illumina HiSeq, and add sequence data to the database;</p> <p>Contact person: Theo van der Lee E-mail: theo.vanderlee@wur.nl</p>
<p>2. Bulgarian Food Safety Agency, Bulgaria</p> <p>Ani Becheva a.besheva@bfsa.bg</p>	<p>-Make available material from Bulgarian isolates (at least 5 compost samples);</p> <p>Contact person: Lidia Dimitrova E-mail: clkr_samokov@mail.bg</p>
<p>3. Canadian Food Inspection Agency, Canada</p> <p>Philip Macdonald philip.macdonald@canada.ca</p>	<p>-Provide DNA and/or RNA extracts and /or sequence data from Canadian isolates; -Participation in ring tests, if applicable;</p> <p>Contact person: Donna Smith E-mail: Donna.Smith2@canada.ca</p>



	<p>Contact person: Marie-Claude Gagnon E-mail: marie-claude.gagnon@canada.ca</p> <p>Contact person: Guillaume Bilodeau E-mail: guillaume.bilodeau@canada.ca</p>
<p>4. Aarhus University, Denmark</p> <p>Mogens Nicolaisen mn@agro.au.dk</p>	<p>-Provide material of <i>S. endobioticum</i>;</p> <p>Contact person: Mette Vestergård Madsen E-mail: mvestergard@agro.au.dk</p>
<p>5. Julius Kühn Institute, Germany</p> <p>Steinmüller, Silke silke.steinmoeller@julius-kuehn.de</p>	<p>-Contribution to be detailed;</p> <p>Contact person: Kerstin Flath E-mail: Kerstin.flath@julius-kuehn.de</p>
<p>6. Benaki Phytopathological Institute, Greece</p> <p>Irene Vloutoglou i.vloutoglou@bpi.gr</p>	<p>-Make available <i>S. endobioticum</i> vital material (compost with resting spores) of pathotype 18(T1) originating in two contaminated fields in Greece together with metadata on soil type, potato varieties on which the pathotype was originally detected, and method used for pathotype identification;</p> <p>-As the Greek pathotype 18(T1) has already been sequenced by Vossenberget al. (2018), we could potentially participate in the validation of the protocol to determine the complete mitochondrial genome sequences of new samples of <i>S. endobioticum</i> originating in areas other than Greece;</p> <p>Contact person: Dimosthenis Kizis E-mail: d.kizis@bpi.gr</p> <p>Contact person: Irene Vloutoglou E-mail: i.vloutoglou@bpi.gr</p>
<p>7. Department of Agriculture, Food and Marine, Ireland</p> <p>Maria Destefanis Maria.Destefanis@agriculture.gov.ie</p>	<p>-Provide material of <i>S. endobioticum</i>;</p> <p>-Participation in the ring tests, if applicable;</p> <p>Contact person: Bourke, Andy E-mail: Andy.Bourke@agriculture.gov.ie</p>
<p>8. All Russian Plant Quarantine Center, Russian Federation, Russia</p> <p>Natalia Sherokolova natalia_sh@mail.ru</p>	<p>-Collection and analysis of samples from the outbreaks;</p> <p>-Participation in the ring tests, if applicable;</p> <p>Contact person: Maria Kopina E-mail: kopinamaria645@gmail.com</p> <p>Contact person: Julia Tsvetkova E-mail: yutska@mail.ru</p> <p>Contact person: Dmitry Shukhin E-mail: dmitriq.shukhin@gmail.com</p>
<p>9. Swedish Board of Agriculture, Sweden</p>	<p>-To provide data/information on Swedish isolates;</p>



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<p>10. Agriculture and Agri-Food Canada, Canada</p> <p>Hai Nguyen hai.nguyen2@canada.ca</p>	<p>-Contribution to be detailed;</p> <p>Contact person: Hai Nguyen E-mail: hai.nguyen2@canada.ca</p>
<p>11. Batumi Shota Rustaveli State University, Georgia</p> <p>Natela Tsiklashvili deputyrector-n@bsu.edu.ge</p>	<p>-Make available wart material from Georgian isolates;</p> <p>Contact person: Zoia Sikharulidze E-mail: zsikharulidze@ymail.com</p>
<p>12. Böhm-Nordkartoffel Agrarproduktion GmbH, Germany</p> <p>Rafal Zgadzaj rzgadzaj@bna-kartoffel.de</p>	<p>-Contribution to be detailed</p> <p>Contact person: Rafal Zgadzaj E-mail: rzgadzaj@bna-kartoffel.de</p>
<p>13. Leibniz University of Hannover, Germany</p> <p>Thomas Debener debener@genetik.uni-hannover.de</p>	<p>-To provide potato wart samples;</p> <p>Contact person: Thomas Debener E-mail: debener@genetik.uni-hannover.de</p> <p>Contact person: Dennis Reckwell E-mail: reckwell@genetik.uni-hannover.de</p>
<p>14. Animal and Plant Quarantine Agency, Korea</p> <p>Hyun Ju Kim hjkim0805@korea.kr</p>	<p>-To provide potato wart samples;</p> <p>Contact person: Hyun Ju Kim E-mail: hjkim0805@korea.kr</p>
<p>15. National Academy of Agrarian Sciences, Ukraine</p> <p>Liliya Janse liliya.janse@gmail.com</p>	<p>-Make available collection material from Ukraine (of different origin / pathotypes); -Isolate spores from the collection material, perform DNA/RNA extraction (if consumables are provided and these tasks for our team are foreseen);</p> <p>Contact person: Liliya Janse E-mail: liliya.janse@gmail.com</p>

1.8. Research project partnership outside Euphresco

Euphresco funding ensures a certain level of transnational collaboration among Euphresco member countries. It is possible, if the funding consortium is interested, to contact funding organisations or research groups outside the geographical area covered by Euphresco members. The Euphresco coordinator could advertise the research topic in order to have an enlarged collaboration. If funders are interested in this possibility, please check the case below:

☒ The funding consortium of the topic mentioned in section 1.2 requires that the topic is advertised outside the Euphresco network.

In particular countries with a reported presence of potato wart disease outside the Euphresco member countries are encouraged to participate in this project. These countries include, but

are not restricted to, Peru (believed region of origin), South Africa, India, China, and Bangladesh. In addition, Euphresco member countries with reported potato wart presence that are not included in this topic description as partners (yet) will be encouraged to participate and provide samples with associated metadata. These countries include, but are not limited to, Germany, Poland, Czech Republic, and New Zealand.

1.9. Any other relevant information on content

None.

2. Euphresco management aspects of the project

2.1. Indication of the topic budget

Funding organisation ^a	Mechanism ^b	Total Budget ^c
1. NVWA (NL)	NC	€
2. BFSA (BG)	NC	€
3. CFIA (CA)	NC	€
4. AU (DK)	NC	€
5. JKI (DE)	NC	€
6. BPI (GR)	NC	€
7. DAFM (IE)	NC	€
8. VNIKR (RU)	NC	€
9. JV (SE)	NC	€
10. AAFC (CA)	NC	€
11. BSU (GE)	NC	€
12. BNA	TBD	€
13. UNI-Hannover (DE)	NC	€
14. APQA (KR)	NC	€
15. NAAS (UA)	NC	€
total		€

2.2. Expected duration of the project (only for non-competitive topics)

24 months.

2.3. Identification of project coordinator

Has the research project coordinator been identified?

- ☒ Yes
☐ No

2.4. Any other relevant information on topic organisation and management

None.

^a First member is project coordinator. A minimum of two partners are necessary for each proposal. Add lines as needed.

^b Please indicate the preferred mechanism (e.g. real pot RP; virtual pot VP; non-competitive NC), or several mechanisms if there is flexibility.

^c Optional, as this amount can still change in the next phase. In-kind contribution should also be indicated in this column.