

1. Content of the 'Topic Description' document

1.1. Topic area

A: Diagnostics, field detection, surveillance

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-R-2.2: to expand knowledge on transmission of disease and pathogens for healthy planting material
- Objective 2017-R-4.1: to validate risk-based sampling methodologies for phytosanitary inspections
- Objective 2017-R-5.1: to understand the biological significance of a positive molecular diagnosis
- Objective 2017-R-6.1: to test and validate methods for *in situ* detection and identification of pests

1.3. Topic title

Validation of molecular diagnostic methods for the detection of *Tomato brown rugose fruit virus* (ToBRFV) in seeds of tomatoes, chilies and eggplants.

1.4. Description of the problem the research should solve

Tomato (*Solanum lycopersicum* L.) is considered one of the most important vegetables worldwide (Gómez *et al.*, 2000), such as China, India, Turkey, United States of America, Egypt, Iran, Italy, Spain and Mexico. Trellised tomato plants grown in protected structures, greenhouses, net-houses are highly exposed to infections by mechanically transmitted viruses or viroids primarily by the prevalent genera of the *Tobamoviruses*, *Potexviruses* and *Pospiviroids*. Commercial tomato varieties resistant to several *Tobamoviruses* are available, but recently *Tomato brown rugose fruit virus* (ToBRFV) was shown to overcome Tm2.2 resistance gene (Luria *et al.*, 2017). *Tomato brown rugose fruit virus* has been reported to be present in Jordan¹, Germany², Israel³, Italy⁴, Mexico⁵, Palestine⁶, Turkey⁷ and the United States⁸.

Several molecular tests for the diagnosis of *Tomato brown rugose fruit virus* have been developed but harmonized protocols are missing. The Euphresco project shall aim at validating a number of PCR and real-time PCR tests for the detection and identification of the virus on seeds or propagative material of chili pepper and pepper (*Capsicum annuum*), tomato (*Solanum lycopersicum*) and eggplant (*Solanum melongena*). The tests that may be considered for the Test Performance Study are described in the following publications:

- Alkowni R, Alabdallah O, and Fadda Z (2019). Molecular identification of tomato brown rugose fruit virus in tomato in Palestine. Journal of Plant Pathology. <https://doi.org/10.1007/s42161-019-00240-7>

¹ <https://link.springer.com/article/10.1007/s00705-015-2677-7>

² <https://gd.eppo.int/reporting/article-6442>

³ <https://doi.org/10.1371/journal.pone.0170429>

⁴ <https://apsjournals.apsnet.org/doi/10.1094/PDIS-12-18-2254-PDN> and <https://gd.eppo.int/reporting/article-6443>

⁵ <https://www.pestalerts.org/oprDetail.cfm?oprID=765>

⁶ <https://doi.org/10.1007/s42161-019-00240-7>

⁷ <https://gd.eppo.int/reporting/article-6553>

⁸ <https://apsjournals.apsnet.org/doi/10.1094/PDIS-11-18-1959-PDN>

- Davino S *et al.*, (submitted). Developing of real time reverse transcription polymerase chain reaction for rapid detection of Tomato brown rugose fruit virus and its comparison with other techniques
- Li Y, Tan G, Lan P, Zhang A, Liu Y, Li R, and Li F (2018). Detection of tobamoviruses by RT-PCR using a novel pair of degenerate primers. *J. Virol. Methods* 259: 122-128. doi: 10.1016/j.jviromet.2018.06.012
- Luria N, Smith E, Reingold V, Bekelman I, Lapidot M, Levin I, *et al.* (2017). A New Israeli Tobamovirus Isolate Infects Tomato Plants Harboring Tm-22 Resistance Genes. *PLoS ONE* 12 (1): e0170429. doi:10.1371/journal.pone.0170429
- Rodríguez-Mendoza J, García-Ávila CJ, López-Buenfil JA, Araujo-Ruiz K, Quezada-Salinas A, Cambrón-Crisantos JM and Ochoa-Martínez DL (2019). Identification from *Tomato brown rugose fruit virus* by RT-PCR from a coding region of replicase (RdRP) (Identificación de *Tomato brown rugose fruit virus* por RT-PCR de una región codificante de la replicasa (RdRP)). DOI: 10.18781/R.MEX.FIT.1902-6
- Detection of infectious *Tomato brown rugose fruit virus* (ToBRFV) in tomato and pepper seed (ISHI-Veg) <https://www.worldseed.org/wp-content/uploads/2019/05/Pepper-ToBRFV-2019-1.2.pdf>
- First report of *Tomato brown rugose fruit virus* infecting greenhouse tomato in the United States. <https://doi.org/10.1094/PDIS-11-18-1959-PDN>
- Occurrence of Tomato brown rugose fruit virus Infecting Tomato Crops in Mexico. <https://doi.org/10.1094/PDIS-11-18-1974-PDN>

1.5. Description of the expected results

The proposal aims to compare and validate detection/identification molecular tests. Results will support the development of a validated Diagnostic Protocol that could facilitate the trade of seeds.

1.6. Beneficiaries of this research product

The beneficiaries of this protocol are the tomato, chili (pepper) and eggplant growers, the seed industry, the national plant protection organizations and policy makers.

1.7. Research funders and research contribution/ distribution

Funding organisation	Research activity and researchers involved
1. Centro Nacional de Referencia Fitosanitaria del SENASICA, México Clemente de Jesus García Ávila clemente.garcia@senasica.gob.mx	-Project coordination with partner 2 NVWA; -The group will be focus on the next crops: Solanum lycopersicum, Solanum melongena and Capicum annum, that are affected by Tomato brown rugose fruit virus; -Provision of material for the TPS; Contact person: Clemente de Jesus García Ávila E-mail address: clemente.garcia@senasica.gob.mx
2. National Plant Protection Organization, the Netherlands Martijn Schenk M.Schenk1@nvwa.nl	-Project coordination with partner 1 Senasica; -Participation in the TPS; Contact person: Annelien Roenhorst

	E-mail address: j.w.roenhorst@nvwa.nl
3. Federal Ministry for Sustainability and Tourism, Austria Sylvia Blümel sylvia.bluemel@ages.at	-Participation in the TPS; Contact person: Sabine Grausgruber Groeger E-mail address: sabine.grausgruber-groeger@ages.at
4. Flanders Research Institute for Agriculture, Fisheries and food, Belgium Kris de Jonghe Kris.DeJonghe@ilvo.vlaanderen.be	-Participation in the TPS; -National survey in host crops; -If available, reference material will be shared; Contact person: Kris de Jonghe E-mail address: Kris.DeJonghe@ilvo.vlaanderen.be
5. Bulgarian Food Safety Agency, Bulgaria Ani Becheva a.besheva@bfsa.bg	-Participation in the TPS; Contact person: Dora Panavotona E-mail address: d.panayotova@bfsa.bg
6. French Agency for Food, Environmental and Occupational Health and Safety, France Geraldine Anthoine geraldine.anthoine@anses.fr	-Participation in the TPS; Contact person: Pascal GENTIT E-mail address: pascal.gentit@anses.fr
7. Federal Ministry of Food and Agriculture, Germany Bettina Beerbaum Bettina.Beerbaum@bmel.bund.de Silke Steinmöller silke.steinmoeller@julius-kuehn.de	-Participation in the TPS; -Provision of material for the TPS; Contact person: Heiko Ziebell E-mail address: heiko.ziebell@julius-kuehn.de
8. Ministry of Agriculture, Plant Biosecurity Plant Protection and Inspection Yael Meller Harel YaelM@moag.gov.il	-Participation in the TPS; -Provision of material for the TPS; Contact person: Mouhammad Zeidan E-mail address: mouhammad.zeidan7@gmail.com
9. Council for Agronomic Research and the analysis of Bioeconomy, Italy Luca Riccioni luca.riccioni@crea.gov.it Laura Tomassoli laura.tomassoli@crea.gov.it	-Participation in the TPS; -Provision of material for the TPS; Contact person: Laura Tomassoli E-mail: laura.tomassoli@crea.gov.it
10. All Russian Plant Quarantine center, Russian Federation Natalia Sherokolava natalia_sh@mail.ru	-Participation in the TPS; -Testing of different seeds and plants by PCR and ELISA; Contact person: Yury Shneyder

Oksana Dobrovolskaya oxana-d@yandex.ru	E-mail address: yury.shneyder@mail.ru
11. Ministry of Agriculture Forestry and Food, Slovenia Erika Oresek erika.oresek@gov.si	-Participation in the TPS; Contact person: Natasa Mehle E-mail address: natasa.mehle@nib.si
12. Eurofins Plant Pathology Laboratory, France Abdelnasser El Ghazouani AbdelnasserElGhazouani@eurofins.com	-Participation in the TPS; Contact person: Christopher Wattier E-mail address: ChristopherWattier@eurofins.com
13. The French Variety and Seed Study and Control Group, France Valérie Grimault valerie.grimault@geves.fr	-Participation in the TPS; Contact person: Valérie Grimault E-mail address: valerie.grimault@geves.fr
14. Hazera, Israel Hila Danino Hila.Danino@hazera.com	-Participation in the TPS; Contact person: Hila Danino E-mail address: Hila.Danino@hazera.com
15. University of Palermo, Italy Salvatore Davino salvatore.davino@unipa.it	-Participation in the TPS; -Provision of material for the TPS; Contact person: Salvatore Davino E-mail address: salvatore.davino@unipa.it
16. BASF, the Netherlands Aubrey Klerks aubrey.klerks@vegetableseeds.bASF.com	-Participation in the TPS; -Provision of material for the TPS; -Other contributions to be determined; Contact person: Joanne Peters E-mail address: joanne.peters@vegetableseeds.bASF.com
17. Enza Zaden, the Netherlands Gerbert Hiddink G.Hiddink@enzazaden.nl	-Participation in the TPS; -Provision of material for the TPS; Contact person: Gerbert Hiddink E-mail address: G.Hiddink@enzazaden.nl
18. Naktuinbouw, the Netherlands Marcel Toonen m.toonen@naktuinbouw.nl	-Participation in the TPS; -Validation according to EPPO PM 7/98 (2) of a detection protocol for ToBRFV based on the ISHL-Veg protocol (2); Contact person: Maaike Bruinsma E-mail address: m.bruinsma@naktuinbouw.nl
19. Spanish Plant Breeders Association, Spain Elena Saenz elena.saenz@anove.es	-Test development; -Participation in the TPS; -Provision of material for the TPS; Contact person: Leandro de León Guerra

E-mail address: Ideleon@anove.es

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

Information to define the profile of sought partners could be useful (but not mandatory): country/region (if there are preferences), skills/expertise required, etc.

1.9. Any other relevant information on content

The EU funded project VALITEST will organize a TPS in the spring 2020. Synergies shall be pursued between the two projects. Contact person: Géraldine Anthoine
geraldine.anthoine@anses.fr

2. Euphresco management aspects of the project

2.1. Indication of the topic budget

Funding organisation ^a	Mechanism ^b	Total Budget ^c
1. Senasica (MX)	NC	€
2. NVWA (NL)	NC	€
3. BMNT (AT)	NC	€
4. ILVO (BE)	NC	€
5. BFSA (BG)	NC	€
6. ANSES (FR)	NC	€
7. BMEL (DE)	NC	€
8. MOAG (IL)	NC	€
9. CREA (IT)	NC	€
10. VNIIKR (RU)	NC	€
11. MKGP (SI)	NC	€
12. Eurofins (FR)	NC	€
13. GEVES (FR)	NC	€
14. Hazera (IL)	NC	€
15. UNIPA (IT)	NC	€
16. BASF (NL)	NC	€
17. Enza Zaden (NL)	NC	€
18. Naktuinbouw (NL)	NC	€
19. ANOVE (ES)	NC	€
total		€

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

18 months. An early start of the project is suitable, i.e. by October 2019.

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

^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.