

In the frame of Italian roles on phytosanitary aspects (D.L. n. 214 / August 19/2005) a national laboratory organization is defined as follows:



Laboratories distributed on the territory



•To establish official diagnostic protocols
•To transfer official protocols to regional and private labs
•To train personnel
•To organize proficiency tests

Armonization of all partners involved in official diagnosis

To establish official diagnostic protocols

Several official diagnostic protocols have been established, transferred to Plant Protection inspectors by training courses.

Most of them are routinely applied to run official controls

All of them have been validated

Interlaboratory tests among regional phytosanitary labs have been organized to test their reproducibility

The following reference protocols have been published

5 fungi 1 bacterium 2 phytoplasmas

Ceratocystis platani Giberella circinata Monilia fructicola Guignardia citricarpa Phytophthora ramorum Xanthomonas arboricola pv. pruni Candidatus Phytoplasma mali Candidatus Phytoplasma prunorum Pepino mosaic virus Plum pox virus Tomato infectious chlorosis virus Tomato chlorosis virus Grapevine viruses covered by phytosanitary roles (GLRaV 1, GLRaV 2, GLRaV 3, GVA, GVB, ArMV, GFLV, GFkV) Potato spindle tuber viroid

> 12 viruses 1 viroid

Under preparation

Soil fungi covered by certification protocols by macroarrays (Chondrostereum pupureum, Verticillium dahlie, V. albo-atrum, Armillariella mellea, Nectria galligena, Phytophthora cactorum)

Tilletia indica Guignardia citricarpa Phytophthora lateralis Phytophthora kaernoviae Chalara fraxinea Plasmopara halstedii

Xylella fastidiosa Xanthomonas vesicatoria Pseudomonas syringae pv. actinidiae Acidovorax citrulli Ca. Liberibacter solanacearum

Plum pox virus LAMP Grape vine Pinot gris virus Prune dwarf virus Prunus necrotic ringspot virus Peach latent mosaic viroid Tomato chlorotic dwarf viroid

Flavescence dorée *Candidatus* Phytoplasma pyri

Plasmopara halstedii

PCR and Real time PCR (EPPO protocol) Sunflower seeds 6 laboratories

Samples (seed DNA spiked with fungal DNA) Protocols Primers and probes Results elaboration

Xylella fastidiosa



ELISA, PCR, Reat time PCR, LAMP PCR (selected from bibliography) Liophylized plant extracts spiked with devitalized bacterial cells 18 italian labs

Pre test among 3 labs to confirm the stability of samples and the analytical sensitivity of each method





PPV

Lamp PCR (set up by IPAD LAB and validated by CRA-PAV) Fresh tissue of stone fruits 18 laboratory

> Pre test among 5 labs to confirm the analytical sensitivity of the method

Samples (fresh tissue) Protocol LAMP kit Results elaboration



Grapevine pinot gris virus RT-PCR, RT-Reat time PCR Leaves and woody material 17 italian labs



Pre test among 5 labs to select the best methods according to different parameters (Analitical sensitivity, specificity)

Samples (FED MACH San Michele all'Adige) Protocol Reagents Results elaboration



The Central laboratory must be accreditated

ACCREDIA is the national body

We are under way for accreditation. The inspection visit is planned next November

PSTVd Viruses

Erwinia amilovora Monilinia fructicola Guignardia citricarpa RT-PCR+RFLP, Real Time ELISA* (Flexible scope has been aknowledged) morfological, immunofluorescence, PCR PCR multiplex Real time PCR

Problems:

Personnel Costs Availability of quarantine structures Iabs, screen houses, glass houses Validation of protocols: Reference samples* Quality system of labs joining interlaboratory tests