







# MAPPING VIRUSES ASSOCIATED WITH CENTURY OLD "PSOROSIS GROUP" DISEASES OF CITRUS

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Psorosis, concave gum and impietratura are three century old transmissible citrus diseases one time considered as members of the "psorosis complex", because of a chlorotic flecking reaction induced on young leaves, associated to some specific symptoms.

**Citrus psorosis** (CPs) is a graft transmissible "scaly bark disease" associated to similar leaf flecking, caused by *Citrus psorosis ophiovirus*. **Concave gum** (CG) is a severe disease of trunk and limbs, associated to a Coguvirus. **Impietratura** (CI), characterized by specific symptoms on fruits, has been associated to a second coguvirus, CiVA. Both CG and CI induce psorosis-like symptoms in young leaves and affect xylem vessels of trees.

This study aims to investigate

the relation (if any) among the agents of this group of diseases.

if chlorotic flecking of young leaves, similar to that one observed for psorosis, is specifically associated with any them.

Materials and Methods	Citrus psorosis			
	Citrus Psorosis virus ( <i>Citrus psorosis ophiovirus, Ophiovirus, Aspiviridae</i> )			
Old sweet orange orchards of Sicily	Bark scaling of trunks and limbs, were detected in eight old or regrafted sweet orange trees.			



 and Calabria were surveyed to find symptoms of CG, CI and CPs.

Budsticks from 29 trees were grafted
on vigorous sour orange seedlings in greenhouse.

Field samples were tested by duplex RT-PCR (CCGaV+CiVA) [1] and real-time RT-PCR (CPsV) [2].

Twelve healthy trees were used as control. Additional indexing for other viruses and viroids

both by indicator plants and molecular tests is in progress.

## **Concave gum-blind pocket**

Transient chlorotic flecking of young leaves was observed in greenhouse propagations.



**Three additional** samples with symptoms of **scaly bark** and **concavities** resulted infected by CPsV, CCGaV and CiVA (**2**) and by CPsV and CCGaV (**1**). All **eight** trees were tested positive to CPsV, **3** of them were also infected by CCGaV and CiVA, and **1** only by CCGaV.

ID	Symptoms	Scion/stock	CPsV	CCaGV	CiVA
SL 1/21	scaly bark	Valencia/Sour O	+	+	+
SL 6/21	scaly bark	T. Gallo/Sour O	+	+	+
SL 17/21	scaly bark	T. Gallo VCR/Sour O	+	+	+
SL 5/21	scaly bark	T. Gallo/Sour O	+	+	-
Bov A.D.1	scaly bark	Navelina/Sour O	+	-	-
SL 9/21	scaly bark	T. Gallo/Sour O	+	-	-
SL 12/21	scaly bark	Navelina/Sour O	+	-	-
Valencia 1	scaly bark	Valencia/Sour O	+	-	-

#### Impietratura

### Citrus Concave Gum associated Virus (Citrus coguvirus, Coguvirus, Phenuiviridae)





**Six** out of 11 trees showing broad concavities on the limbs and trunk and/or extremely depressed concavities were positive to CCGaV.

**Three** of them were coinfected by CPsV and CiVA, one by CiVA.

ID	Symptoms	Scion/stock	CPsV	CCaGV	CiVA
CGA2	CG	Navelina/Sour O	+	+	+
SL 3/21	CG	T. Gallo/Sour O	+	+	+
SL 10/21	CG	Navelina/Sour O	+	+	+
SL 4/21	CG	T. Gallo/Sour O	+	-	+
SL 7/21	CG	T. Gallo/Sour O	-	+	+
SL 2/21	CG	T. Gallo/Sour O	+	-	-
SL 11/21	CG	Navelina/Sour O	+	-	-
SL 15/21	CG	T. Sciara/Sour O	+	-	-
PSTS1	Gumming	Navelina/Sour O	-	-	+
SL 18/21	CG	Tacle/Sour O	-	+	-
Winola 1	CG	Winola/L Volk	-	+	-

Citrus Virus A (Coguvirus eburi, Coguvirus, Phenuiviridae)



Only **seven** trees showing reduction in fruit size, skin hardening, and gumming in the albedo were found. Some fruits showed gum.

**Four** samples showing clear gum under the calyx and in the vascular bundles of the central axis were positive to CiVA.

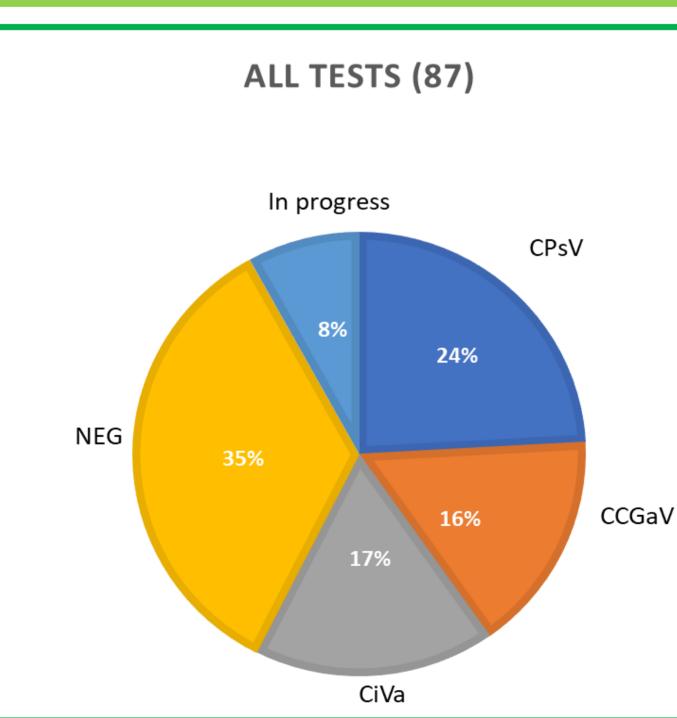
ID	Symptoms	Scion/stock	CPsV	CCaGV	CiVA
IMP-VLG2	Albedo & columella gumming	Tarocco/Sour O	+	+	+

Four trees resulted positive to CPsV or CiVA but not to CCGaV.Chlorotic flecking and oak leaf were observed in greenhouse.

One of them was associated to CCGaV and CPsV, and **two** samples with CPsV. One sample was infected only by CiVA.

IMP-SL	Imp	Moro/Sour O	+	-	+
VAN-GL	Imp	Vaniglia/Sour O	+	-	+
IMP 18/1	Imp	Sanguinello/Citr. T	-	-	+
SL 21/21	Imp	Navelina/Citr. C	-	-	-
SL 22/21	Imp	Navelina/Sour O	-	-	-
SL 23/21	Imp	Navelina/Sour O	-	-	-

#### **Results and Discussion**



- > As expected, 22 out of 29 sampled trees were single or multiple infected.
- No one was coinfected by CCGaV and CiVA, as observed in Greece [3], while in Spain [4] and in Italy few samples were coinfected by CCGaV and CiVA [5], [1].
- > Transient leaf symptoms were irregularly detected, with some slight or substantial difference.
- The results lead to conclude that more efforts are needed to clarify the relations between phenotypic expression and virus infection of these diseases.
- Biological indexing on a large host range plants and fulfillment of Koch postulates, associated to high throughput sequencing will be required.
- Samples found infected only by CCGaV (2) and by CiVA (1) will be helpful to start.

References: [1] Minutolo et al., J. Plant Pathol. (2020) 102:655–661; [2] Loconsole et al., Eur. J. Plant Pathol. (2010) 128:251-259); [3] Beris et al., Phytopathology (2021); [4] Velàzquez et al., J. Citrus Pathol. (2019) 6(1), 11-12; [5] Navarro et al., Front. Microbiol. (2018) 9:2340.



PSR Sicilia 2014–2020, action 16–Cooperation, sub-action 16.1.

