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TECHNICAL DATA SHEET

Flavecence Dorée: how to manage the disease with more precision ?

In the regions already infected by Flavescence Dorée disease

Network for the exchange and transfer of innovative knowledge between European wine growing regions

This project has received funding from the European Union's Horizon 2020 research and innovation programme under **grant agreement No 652601**

A more precise FD management

Introduction

Flavescence dorée (FD) is the most important and destructive phytoplasma disease of grapevines. FD induce severe impacts, including reduced vitality of vines, yield reductions and reduced wine quality. Without control measures, the disease spreads rapidly, affecting up to the totality of vines in a few years. Despite mandatory control in Europe for this disease, it is still spreading and need permanent monitoring to detect new infected areas.

A good management of FD go through a combination of methods working both on the vector, *Scaphoideus titanus* and on the disease, once grapevine are contaminated by the phytoplasma. In infected regions, management of the vector and of the disease go hand in hand. The conscientious application of the methods described in this datasheet (vector, vineyard and wild vines management and when needed, hot water treatment) are essential to control FD in a territory and to limit its spread.

Vector management

Timing for spraying, a key factor

Application time is the key of success in managing *Scaphoideus titanus* population in vineyard. The leafhopper transmits FD phytoplasma from one grapevine to another very quickly. In order to slow disease's propagation, a good **control of the vector** and **vineyard monitoring** at large scale are necessary.

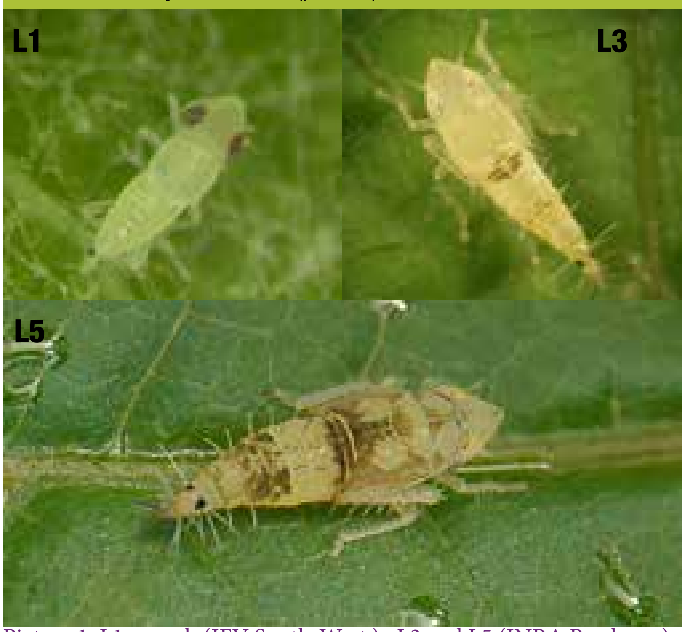
In order to apply insecticides against the leafhopper at the good timing, several methods exist to determine the best application date:

1- **Emerging cages**: these cages are a very effective tool to determine when *Scaphoideus titanus* first hatching appears. Two years old wood, preferred for egg laying, from a plot where *Scaphoideus titanus* presence was detected in the previous year are kept in a cage and hatching moment is monitored every day. A sticky trap is put inside the cage in order to capture nymphs as soon as they hatch. **When date of first hatching is determined the effective date for first insecticides treatment is positioned one month later**.

2- **Modelling**: decision support systems exists and are able to predict in vineyard (with its local conditions) pests and diseases, including *Scaphoideus titanus*. One model currently in development can predict when each stage of *Scaphoideus titanus* nymphs will appear according to specific climatic conditions (this implies to have a weather station close to the plot). Model is based on observations made the years before and historical and daily temperatures. In order to provide a more accurate possible real observation data on stages need to be informed.

3- **Monitoring of nymphal development stages** on leaves is requested to choose the right time for the first treatment, but cannot usually be made by winegrowers and requires trained technicians. Data on nymphs repartition can allow to divide a region into macroclimatic areas, monitor the insect and suggest the **best time for spraying** in each area, and even suggesting a different timing depending on the kind of product used. **Visual control** need to be done on **leaf underside** and on grapevine basal shoots (suckers) and basal leaves from mid-May to August in the early morning avoiding to move excessively the vegetation.

There are 5 successive nymphs stages from hatching to adult, nymphs are identifiable thanks to **two symmetrical black points** in dorsolateral position at abdomen posterior end (nymphs can be confused with *Phlogottetix Cyclops* who has also two black points on the second-last segment of abdomen, see «mistakable species»). These points are visible to the naked eye from L2 (pict. 1).



Picture 1: L1 nymph (IFV South-West,), L3 and L5 (INRA Bordeaux)

The nymph, when disturbed, show a typical behaviour: it tends to **jump away**.
This behaviour can be used to discriminate *S.titanus* nymphs to other leafhopper juvenile forms that should be present at the same time on the grapevine leaves, such as *Empoasca vitis* (when disturbed move laterally on the leaf surface) and *Zygina rhamni* (when disturbed move along a straight line on the leaf surface).

4- **Monitoring adult with traps: yellow sticky traps** can be set up in vine-plot and can be monitored by growers themselves if properly instructed, or by specialists. A good tool to find again the traps for people who control many of them is to use GPS localized shots. Traps will help to decide when apply adulticide treatment. Adult flights have been often observed from the beginning of July to later than expected (October).

The late control of vector leafhopper population is important to decide the opportunity of an additional insecticide against adults.
Sticky traps are yellow because *Scaphoideus titanus* adults are more attracted by this colour. (pict. 2) Sticky traps need to be set up when L4 to L5 nymphs are observed on the plot. Sticky traps can be used on plots with a strong FD history in order to monitor the flight and on plots free from FD to prevent infection. On the plot with a strong *S.tianus* populations history one sticky trap can usefully help to know when adults are flying, for a plot free from FD vector, several traps should be hanged in the plot, with a focus on the edges (Fig.1).
Traps need to be **hanged inside the foliage**, the as close as possible to vegetation, where leafhoppers are present, usually they are placed on the height of 1,5 m between two wires (pict.3). Number of traps per plot is usually different according to the size of the plot and number of surrounding vineyard or areas with wild *Vitis* species. Usually, 5-6 sticky traps/ha are placed and checked every 7 days.
Another method to monitor adults is the beating method. It consist of the use of an entomological umbrella made of white cloth which is positioned under the vine canopy to collect *S.titanus* individuals that fall in after vine shaking (then they are counted). Efficiency of this method is limited but can present better efficiency in the first two-three days of adult appearance because they are less mobile. After these days the adults tend to fly away flying up to few kilometres. So, it is more probably that they are attracted from the yellow colour of traps. Another alternative method is suckering of leafhopper with D-Vac.

Scaphoideus titanus adult size range from 4,8 to 5,8 mm, has brown colour and stripes on the head (from 1 to 3 according gender).

Picture 2: *Scaphoideus titanus* adult on a sticky trap (IPTPO)

Picture 3: Sticky trap (IFV South-West)

Figure 1: Monitoring of S. titanus with yellow sticky traps in an area with several vineyard plots and wild Vitis species – potential number and distribution of traps (white arrow: potential movement of S. titanus adult; red dots: distribution of yellow sticky traps.

Scaphoideus titanus

Scaphoideus titanus **nymph (L1) (IFV South-West)**

Scaphoideus titanus **adult (INRA Bordeaux)**

Mistakeable species

Phlogotettix cyclops **nymph (INRA Bordeaux)**

Phlogotettix cyclops **adult (IPTPO)**

Empoasca vitis **nymph (IFV South-West)**

Emposasca vitis **adult (IFV South-West)**

Dictyophara europaea **(IPTPO)**

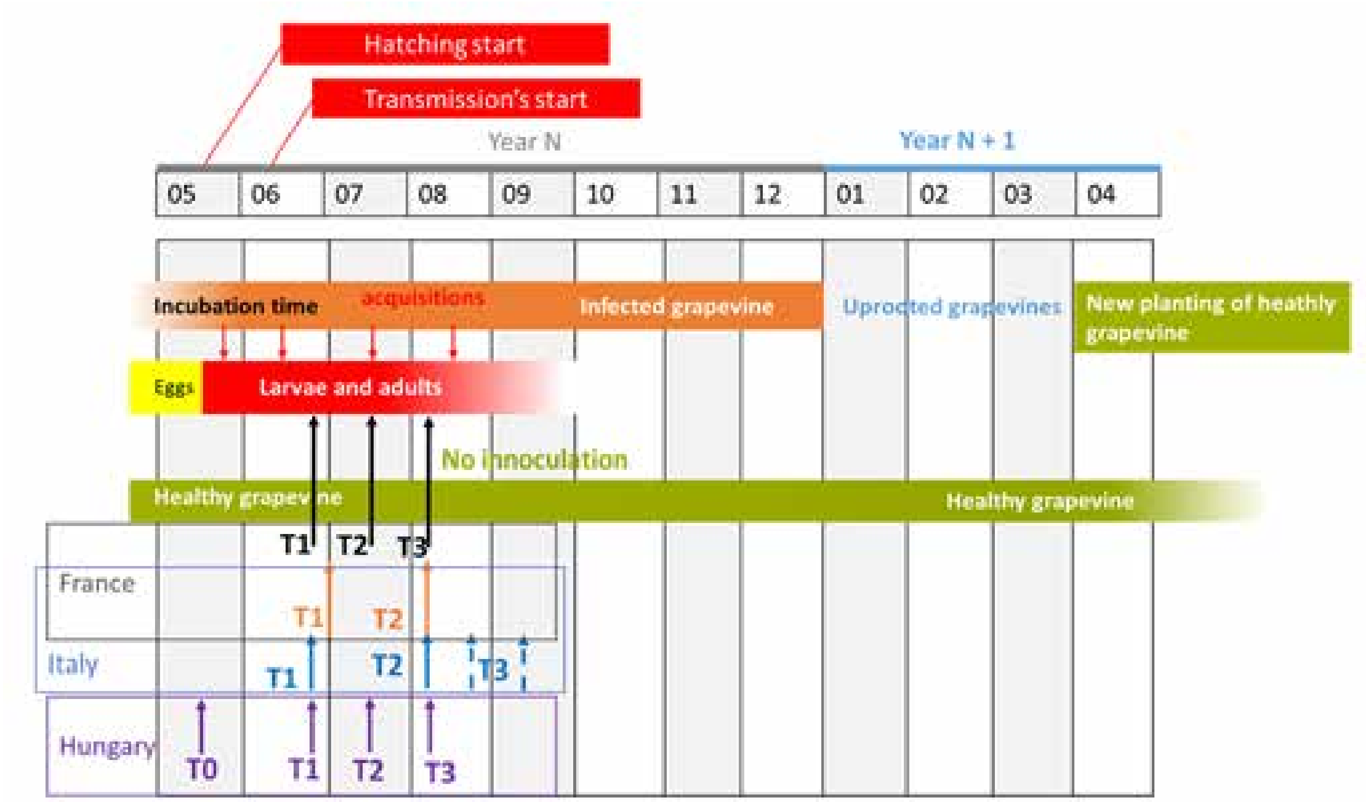
Oncopsis alni **(INRA Bordeaux)**

Hyalesthes obsoletus (hemiptera-databases.org)

Treatment strategies and application

According to the region and even to the country, several treatment strategies exist. One example is given in figure 2. **The first treatment is the most important,** eggs hatching need to be monitored (with one of the methods described above) in order to know when positioning first treatment. The first treatment need to be positioned **one month after hatching** because when nymphs first feed on an infected grapevine there is an **incubation time of one month for the nymphs to be infectious and transmit the phytoplasma again**.

Insecticide treatment application need to cover all grapevine leaf area (fruiting canes and basal suckers), from the basis to the top. Indeed, *Scaphoideus titanus* nymphs are often located close to the first leaves of the trunk and on the suckers, mainly because eggs were laid under 2 years-old bark. Prior to treatment application it is recommended to remove basal shoots and spray need to cover trunk and shoots.
Application need to be done respecting product's recommendation and at the proper timing.



Strategy with 3 treatments
T1: One month after hatching
T2: End of remanance of T1 product, objective to cover end of hatching
T3: On adults
Or
T1: One month after hatching or prior L4-L5
T2: On adults
T3: On adults late summer or after harvest depending on Sc.t population

Strategy with 2 treatments (low population of Sc. t. and no infected grapevines):
T1: On nymphs
T2: On adults

Strategy with extra treatments (high population of Sc.t in the previous years)
T0: Extra treatment on L1 in middle of May, before flowering
T1: On nymphs
T2: End of remanance of T1 product, objective to cover end of hatching
T3: On adults

Figure 2: Insecticide treatment strategies. Adapted from « Jaunisses à phytoplasmes de la vigne, Groupe de Travail National Flavescence Dorée, 2006 »

Products

Conventional chemical control of FD vector, *Scaphoideus titanus*

Active Ingredient	Usable in nursery	Nbr max of treatment/year	Use authorised in vineyard in
Acetamiprid	Italy	3	Italy
Acrinathrin	France, Italy, Portugal	1-3	France, Italy, Portugal
Alpha-Cypermethrin	France, Italy, Portugal, Hungary	2-3	France, Italy, Portugal, Hungary
Alphamethrin	France		France
Azadirachtin	Croatia	2	Italy, Portugal, Coratia
Beta-Cylfuthrin	France, Italy	2-3	France, Italy, Hungary
Buprofezin	Italy	2-3	Italy
Chlorantraniliprole-Thiame-thoxam	France, Portugal, Hungary	1	France, Hungary, Italy, Portugal
Chlorpyrifos-ethyl	France, Italy	3	France, Italy
Chlorpyrifos-ethyl+-Cyper-methrin	France, Italy, Portugal, Hun-gary, Croatia	1-2	France, Portugal, Hungary, Croatia
Chlorpyrifos-methyl	France, Italy, Croatia, Hun-gary	2-3	France, Italy, Hungary, Croatia
Clorpyrifos-methyl+cyper-methrin	France, Hungary	1	France, Hungary
Cypermethrin	France, Italy, Portugal, Croatia	2-3	France, Italy, Portugal, Croatia
Deltamethrin	France, Italy, Portugal, Croa-tia, Spain	2-3	France, Italy, Portugal, Hun-gary, Croatia, Spain
Esfenvalerate	France, Italy, Croatia	2-3	France, Italy, Croatia
Etofenprox	France, Italy	1-3	France, Italy
Fenpyroximate	Portugal	1	Portugal
Gamma-cyhalothrin	France	3	France
Imidacloprid	Portugal	2	Portugal
Indoxacarb	Italy	1	Italy
Lambda-Cyhalothrin	France, Italy, Portugal, Hun-gary, Spain	1-3	France, Italy, Portugal, Hun-gary, Spain
Pyrethrum- Abamectin	Italy		Italy
Spinosyn	Hungary		Hungary
Spirotetramat	Hungary		Hungary
Tau-Fluvalinate	France, Italy, Hungary	1-3	France, Italy, Hungary
Thiamethoxam	France, Italy, Portugal, Hun-gary, Croatia	1-3	Portugal, Croatia
Zeta-Cypermethrin	France, Italy	1-3	France, Italy

Table 1: Active ingredients registered for the control of *Scaphoideus titanus* in Europe, both in vineyard and nursery

Control of FD vector in organic vineyards

Active ingredient	Authorized use in vineyard	Usable in nusery
Natural Pyrethrum	France, Hungary, Italy	Hungazry and Italy
Pyrethrum - Piperonyl Butoxide	Italy	Italy
Azadirachtin	Italy, Portugal	

Table 2: Active ingredients registered for *Scaphoideus titanus* control in organic viticulture

Pyrethrum-based products' characteritics

Name	Pyrethrum content	Dose/ha	Pyrethrum dose/ha	Regulatory status
Pyrévert	20 g/l	1,5 l/ha	30 g/ha	Use authorized in France and Italy against Flavescence Dorée leafhopper Use authorized in Hungary in emergency exemption between 1st July and 30th September on organic vineyard and nurseries. Maximum 3 application/year
PiretroNatura	40 g/l	0,75 l/ha	30 g/ha	Use authorized in Italy on grapevine against Flavescence Dorée leafhopper
Biopiren plus	18,6 g/l	150-200 mg/hl water		Use authorized in Italy
Name	**Pyrethrum content**	**Dose/ha**	**Azadiratchin dose/ha**	**Regulatory status**
Neemazal- T/S	10,6 g/l	2-3 l/ha	20-30g/ha	Use authorized in Italy one grapevine leafhoppers

Table 3: Pyrethrum-based products characteristics

In general, active substances reported in table 3 are more effective on juvenile forms of *S.titanus*. So, its use is recommended when the insect population is constituted mainly of juvenile forms. Moreover, since the persistence is not so high it is suggested to repeat treatment after one week. As light-sensitive, pyrethrum-based products are more effective along time if they are distributed in the evening or at early morning.

Conditions of pyrethrum application

Natural pyrethrum is a delicate molecule, sensitive to high temperature and UV radiation. Half-life time is estimated to 10-12 minutes for a pyrethrum solution exposed to sun. Product has a "choc action" by contact and act on insect nerve-conduction. Application of natural pyrethrum can be done in association with copper or sulphur application (Sudvinbio, 2013). Natural pyrethrum is effective on L1 to L3 stages of S.titanus. Unfortunately, **efficiency of pyrethrum treatment is variable** and **serious monitoring of vector population** and observation of the plot need to be done regularly before and after treatment with pyrethrum-based chemical specialities.

Recommendations for application

- Apply at the end of the day (low light and low temperatures)
- Water pH < 6.8 with an optimum between 6.0 and 6.5
- Apply quickly after mixture preparation
- Use a new can or one aged less than 6 months
- Maximum 3 application during vegetative season

Management of Flavescence Dorée in organic vineyards

Organic viticulture is built on prevention and on the implementation of a self-regulating system that minimizes diseases and pest occurrence, increasing the functional biodiversity and, as a consequence, minimizing the spaces for pest and diseases breakouts. For example, flora has an incidence on *Scaphoideus titanus* populations, the vector can live **both on grapevine and on flora** between rows, with intensity of population varying with time (Trivellone et al, 2013). Nymphs can develop, among other, on *Trifolium repens* and *Ranunculus repens,* both common species used in vineyards (Trivellone et al, 2013). Moreover the presence of flowers contributes to increase the population and diversity of several predators such as spiders that are founded to reduce leafhopper population under cover cropped vineyards.

A more precise FD management

Biological control of *S.titanus* has been tried out either by releasing natural enemies from their native habitat or by increasing local natural enemies' populations but both strategies yielded poor results. Fitness and efficacy of some biocontrol agents were assessed in relation to grape pest and insects. For instance, the fungus *Lecanicillium lecanii* was used against S.titanus juvenile forms and proved to be virulent to the second instar nymph stage.

Where prevention is not sufficient to keep FD vector under control, european organic winegrowers use to spray natural products allowed under EC Reg. 889/08, namely natural pyrethrum and Azadirachtin according to the country. As explained above, they need to be applied with high precision and respecting certain specific requirements, otherwise their efficacy (especially pyrethrum) is too limited.

A care for bees and polinisators

It is important to remember to **minimize the risk for pollinators** while spraying insecticide on big territories at the same time. Spraying must not take place at vine flowering time and they should not be localized on flowers of vineyard's inter-rows. Soil should be tilled or grass mowed before spraying. Moreover spraying have to be performed in the evening after the sunset or in the early morning. In windy days spraying must be avoided in order to reduce drift risk. In Italy the spraying period vary from year to another depending to vine and vector phenological development. Generally the first treatment is performed after flowering and at fruit set starting, to avoid damaging to bees.
Be aware of Metcalfa or mealybug infestation, encouraging bees to visit the vineyard to pick up the honeydew. In this case it's better not to use a neonicotinoid for the second spray against adults; better to use it eventually for the first treatment against nymphs, to control Metcalfa as well.

Vineyard management

Monitoring vineyard

Vineyard sanitation is a key point in FD management. Monitoring vineyard is one of the main key element of FD management and must be done at scale of a territory and individual vineyard scale, often at communal scale in order to have a **collective implication in FD management**. In order to be effective, the better is monitoring organized and controlled by a dedicated organism but monitoring should be done also individually by each winegrower on his own vineyard with respect of monitoring protocols. Symptomatic grapevines (pict 4-5) need to be identified and uprooted Laboratory analysis can be done in case of doubt and is the only way to distinguish Flavescence Dorée from Bois Noir.

Picture 4: Flavescence Dorée symptoms on cv. Syrah (IFV South-West) and cv. Cortese (Maurizio Gily)

Picture 5: Flavescence dorée dessiccation symptom on bunches (IFV South-West)and characteristic leaf roll (IPTPO)

Infected plants management

Uprooting or destruction of infected grapevines is ever suggested and mandatory in most of regions by national decree. During period of leafhopper presence (from May to August) as soon as an infected vine is detected it **must be uprooted. The sooner infected vine is uprooted, the better is it**, in this way *Scaphoideus titanus* can't feed on the infected vine, won't become infectious and won't participate to disease's propagation.
Uprooting need to be done rigorously avoiding any growth of suckers or rootstock. **Rootstocks are healthy carriers**, they can host FD phytoplasma but do not express any symptoms. Eradication of grapevine regrowth need to be done in the vine-plot but also outside the plot. If a quick uprooting is not easy to do during the growing season, it can be postponed after harvest, but the vine, or the symptomatic parts of the canopy, even under suspicious, must be cut away as soon as possible.
Leaving infected grapevines in the plot will increase considerably infection level in the coming years.

Disease evolution

- Enlargement of disease area from grapevine to grapevine around initial grapevine
- Fast increase of diseased grapevines : amount of diseased grapevine can be multiplied by 10 each year (figure 3)
- After inoculation, delay in symptoms expression of one year.

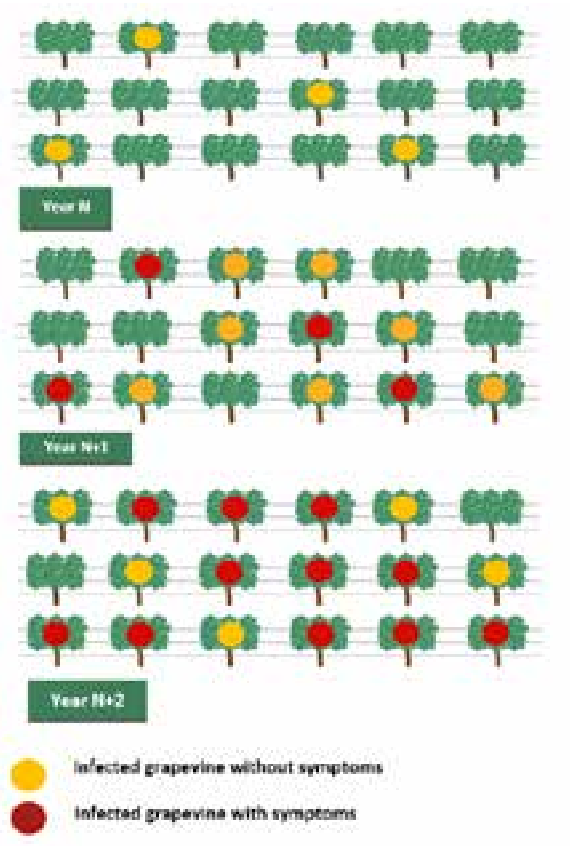


Figure 3: Flavescence Dorée spreading through the years

Wild vines management

Wild vines host both the disease and the vector.
The various crosses of wild vines (*Vitis rupestris, V. riparia, V. berlandieri* etc.) and the ones between them and European vines, used as rootstocks, are capable of hosting the FD phytoplasmas, although they generally do not exhibit the typical symptoms of cultivated vine, or don't show any symptoms at all: therefore they are healthy carriers. The vector *Scaphoideus titanus* feeds indifferently on all species of the genus *Vitis*, so it can **acquire the phytoplasma and transmit the infection from wild to cultivated vines**. In addition, these areas can serve as **refuges for the vector**, therefore reducing the effectiveness of their control.

The flight of *Scaphoideus titanus*

The adult insect vector is able to make quite long flights and capable of dispersing from wild to cultivated grapevine, (Alma, 2015; Lessio, 2014). However, the greater the distance of the «wild» outbreak from the vineyard, the lower is the probability of a migration into the vineyard itself. The migration of insects from outside towards inside a vineyard is also demonstrated by the greater frequency of the presence of symptoms at the border of the vineyard. Most of insects move into the vineyard up to 20-30 meters from hedgerows but in some cases it is possible that adults move up forward, wind allowing long distance movments (Steffek et al, 2007).

How to clean up ?

It is not necessary to deforest. The vine is a sun-loving plant, looking for sunlight and then its canopy grows mainly on the edge of forests, at the border of cultivated fields and roads. But if it is allowed to grow and develop big canopies, it goes to ripen fruit, and at that point can also be propagated by seed, through the birds. This should be avoided.

Cleaning in winter time	Cleaning in springtime
After the harvest, since late autumn, until vine budburst: cleaning of fallow stripes bordering the vineyard by paying particular attention to vines climbing on trees. The debris must be removed and burnt, this step is very important because they can contain Scaphoideus eggs and could sprout from the cuttings and multiply. Where possible, eradicate the roots of the vine with an excavator.	May-June: before treatment against the youth insects in the vineyard, destroying sprouts of survived American vines is required.

CAUTION: Do not destroy wild American vines in summer, because the adults may migrate from the wild to the vineyard (Fig. 4)

The procedure must be repeated in subsequent years, trying to eliminate all American vinestocks.

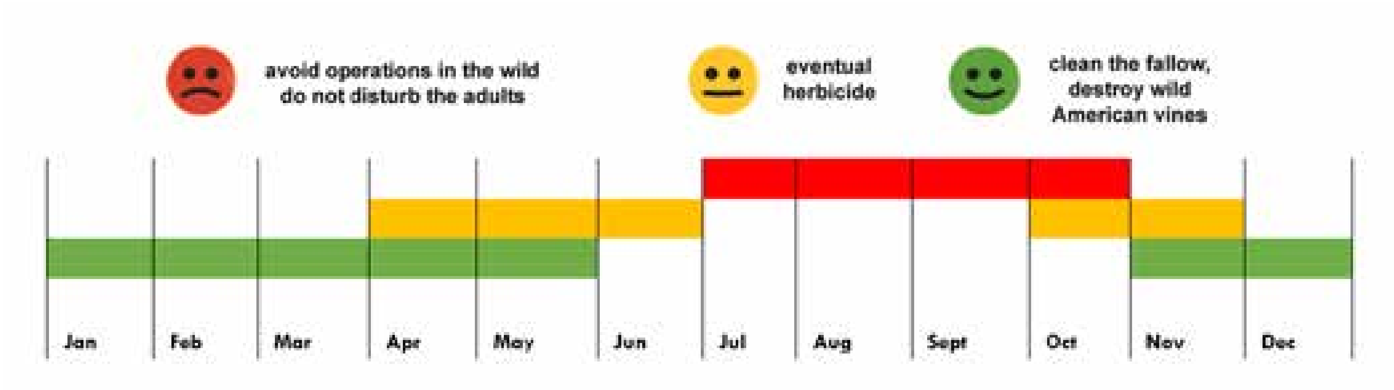


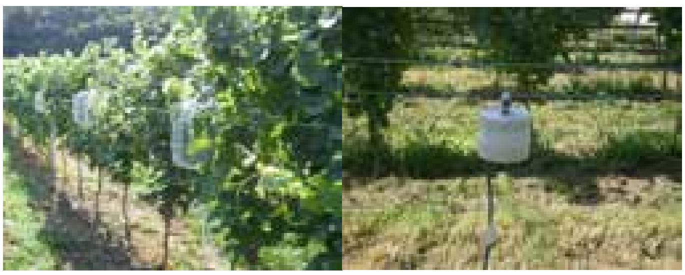
Figure 4: Wild American vines management (D. Eberle, 2015)

Innovative aspects in the management

Alternative methods to chemical management of Flavescence Dorée vector are searched by practitioners, mostly to be used in organic production and in the objective to decrease pesticides use and protect the environment.

Mating disruption

Prevent *Scaphoideus titanus* population to grow from one year to another by **disturbing its reproduction** is one innovative method in development. The method consists in disturbing signals emitted by males to attract females and inhibit their reproduction. Adults communicate with **vibrational signals**, males have a "call and fly" behaviour and emit a courtship song and females emits signals in responses to males (Mazzoni et al., 2009). A mechanism broadcast vibrational signals through a supporting wire and the playback of disruptive vibrational signals reduced the level of male calling and interrupted an established male-female duet that consequently resulted in a significantly reduced number of copulations (Mazzoni et al, 2009).



Picture 6: Mechanical system for mating disruption (Lucchi et al, 2013)

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Push pull strategy

"Push and pull" strategies involve manipulating insect behavior via the combined use of **attractive and repulsive items**, including lures or plants capable of drawing pests into the area where they will be destroyed. The use of this technique against vector of phytoplasma disease has already shown promising results.

Orange oil application

Another alternative to chemical control of *Scaphoideus titanus* is the use of orange essential oil. This product is used by some winegrowers in Europe to control *S.titanus* populations but **complementary to chemical treatment** as **essential oil is not registered as an insecticide**. The active ingredient of orange oil is a terpene, D-limonene and identified as a natural insecticide. The D-limonene as the properties to **desiccate nymphs** and can be effective on the young stages of *Scaphoideus titanus*. Effectiveness of this product still need to be demonstrated.

Kaolin application

The kaolin spraying has **repellent function** towards the leafhoppers; but some studies also prove mortality of nymphs. It is mainly used in organic farming, where the only insecticide admitted and slightly effective is the pyrethrum. It is not an alternative but a possible integration. Kaolin is more effective on early instars than towards adults. Given the high cost of the product and it's proven, even if partial, effectiveness, the optimization of timing and doses should be further studied.

Picture 7: Kaolin application on leaves (ADVID)

Biocontrol agents

Among other biological control agents, the use of endosymbionts, such as Wolbachia, seems to be encouraging and also the symbiotic bacteria of genus *Cardinium*, which is responsible for impaired reproduction and behaviour of the vector (Chuche et al, 2017). This bacteria specie was found in natural populations of both *S.titanus* sexes with a high prevalence (94%) and in differents organs. Finally, *S.titanus* also can host symbiotic bacteria of the genus *Asaia*, transmitted vertically by female as well by male during mating, or during feeding. Some author have suggested using these symbionts to decrease the vector's capacity for *S.titanus.* Further studies should be performed.

Hot water treatment in nurseries

France, Italy, Portugal, Croatia and Hungary have a national decree imposing mandatory treatments on nurseries and according to the country they can be specific to some areas.
In some countries or regions there are specific rules regarding nurseries: mandatory hot water treatment, or in some case prohibition to manage nursery activity in FD outbreak regions.
Hot-water treatment is proposed to c**ure dormant wood material from phytoplasmas** and to supress surface parasite and pests. Pathogenic agent, the phytoplasma is heat-sensitive. **Time and temperature need to be such as supress phytoplasma without affecting plant development capacity**. The use of HWT should intervene to complement preventive measures as insecticides treatments, suppression of contaminated rootstocks and should supplement absence of chemical treatment against phytoplasmas. Propagating material (either cuttings or grafted plants) are immerged into a bath of water at **50°C during 45 minutes.** These parameters allow to eliminate phytoplasma and to have a partial effect on *Scaphoideus titanus* eggs (presents under bark) (Caudwell et al, 1997).

Picture 8: Hot Water Treatment installation (IFV South-West)

Conclusion

A good knowledge on the 3 pillars involved in the FD fight, **control of the vector, management of the disease in vineyard and nurseries and regulation** allow viticultural sector to be more efficient in FD management. Being a complex diseases, all parameters are important, and a **good knowledge of the vector, its feeding behaviour and the evolution mechanisms of the diseases are essential** to manage precisely the interventions allowing to fight the disease. Furthermore, **collaboration and commitment** between winegrowers, dedicated organisms, and other actors of FD management are indispensable in effective disease management.

More information on

www.winetwork-data.eu

Technical datasheets :

- **Guide of good practices in FD management**
- **Hot Water Treatment**

Video seminar: State of the art of scientific research on Flavescence Dorée (François-Michel Bernard, IFV)

Experiences of collective management of FD in France (François-Michel Bernard, IFV)

Source of information

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CONSULT WEBSITES:

Les jaunisses à phytoplasme de la vigne
Region Piemonte
Vineyard leafhoppers (Regione Sicilia)

Work realized in common by the facilitators agents of Winetwork project. Data came from practice, through the help of 219 interviews and from a review of scientific litterature.