

EPPO Datasheet: *Grapevine flavescence dorée phytoplasma*

Last updated: 2022-12-09

IDENTITY

Preferred name: *Grapevine flavescence dorée phytoplasma*

Taxonomic position: Bacteria: Tenericutes: Mollicutes:

Acholeplasmatales: Acholeplasmataceae

Other scientific names: '*Candidatus Phytoplasma vitis*' Firrao et al., *Phytoplasma vitis* Firrao et al.

Common names: bacco 22A disease, flavescence dorée of grapevine, flavescence dorée phytoplasma

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EPPO Categorization: A2 list

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EU Categorization: A2 Quarantine pest (Annex II B)

EPPO Code: PHYP64



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Notes on taxonomy and nomenclature

Grapevine flavescence dorée phytoplasma is a member of the genus '*Candidatus Phytoplasma*', a group of pleiomorphic, non-culturable bacteria, lacking cell wall known to be phloem limited and transmitted by insect vectors (IRPCM phytoplasma/Spiroplasma, 2004). *Grapevine flavescence dorée phytoplasma* sensu stricto refers to the phytoplasma strains of subgroups 16SrV-C and 16SrV-D that can be transmitted from vine to vine by the Delcocephalinae leafhopper *Scaphoideus titanus* Ball (Caudwell, 1990, Martini *et al.*, 1999) and constitute three genetic groups according to the sequence of their gene map (Arnaud *et al.*, 2007). *Grapevine flavescence dorée phytoplasma* has not yet been described as a '*Candidatus Phytoplasma*' species because the inclusion of a specific nucleotide signature in its 16S rDNA sequence, in common between the two taxonomic subgroups 16SrV-C and 16SrV-D, could not be fulfilled (Malembic-Maher *et al.*, 2011). *Grapevine flavescence dorée phytoplasma* sensu-stricto has recently been assigned to Vectotypes II and III that show transmissibility through *S. titanus* and are therefore epidemic into the vineyards, whereas this is not the case for phytoplasma strains of Vectotype I (Malembic-Maher *et al.*, 2020).

HOSTS

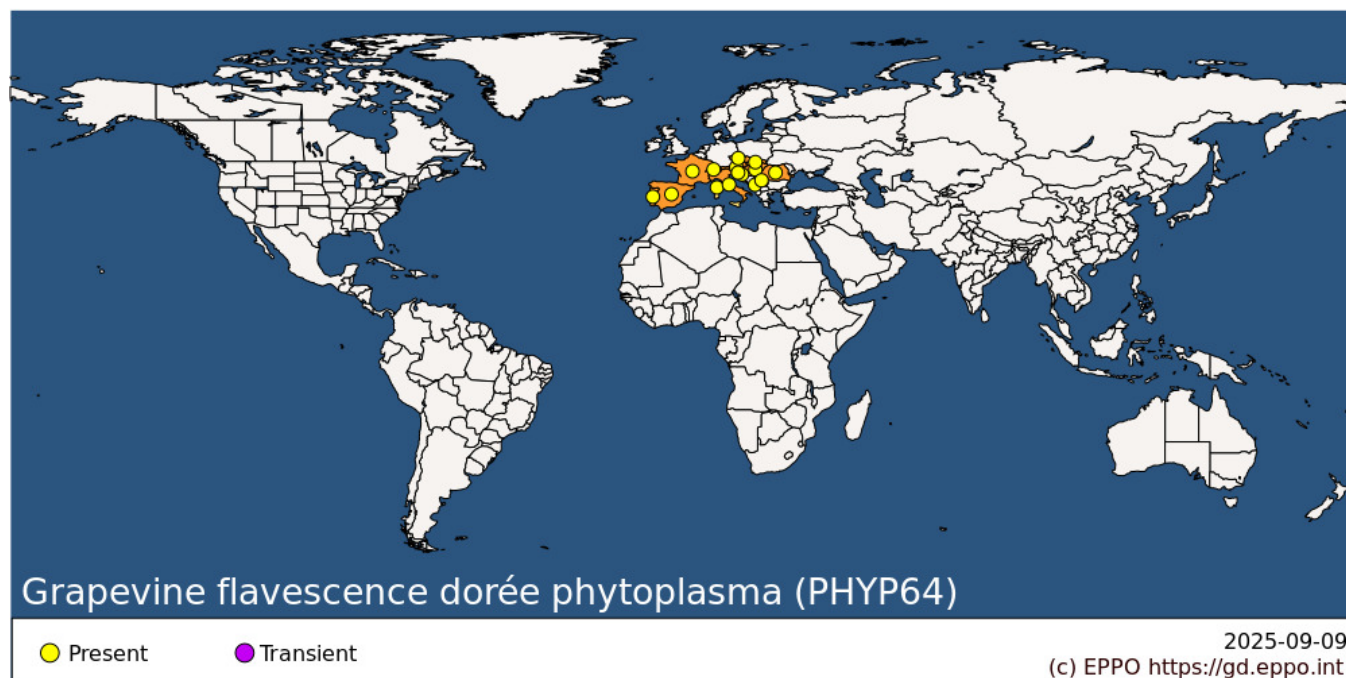
Grapevine flavescence dorée phytoplasma's natural host range includes *Vitis vinifera* and other *Vitis* species and hybrids used as rootstocks (Eveillard *et al.*, 2016), *Alnus glutinosa* and *Alnus incana* (Angelini *et al.*, 2001; Mehle *et al.*, 2011; Radonjic *et al.*, 2013), *Clematis vitalba* (Angelini *et al.*, 2004), *Ailanthus altissima* (Filippin *et al.*, 2011), *Corylus avellana* and *Salix* sp. (Casati *et al.*, 2017; Mehle *et al.*, 2019). Non-natural hosts such as *Vicia faba* and *Glebionis carinata* (synonym *Chrysanthemum carinatum*) have been experimentally inoculated for scientific purposes (Caudwell *et al.*, 1970).

Host list: *Ailanthus altissima*, *Alnus glutinosa*, *Alnus incana*, *Clematis vitalba*, *Corylus avellana*, *Salix* sp., *Vitis acerifolia*, *Vitis amurensis*, *Vitis berlandieri*, *Vitis coignetiae*, *Vitis hybrids*, *Vitis labrusca*, *Vitis pentagona*, *Vitis riparia*, *Vitis rupestris*, *Vitis vinifera* subsp. *sylvestris*, *Vitis vinifera*, *Vitis x champinii*, *Vitis x doaniana*

GEOGRAPHICAL DISTRIBUTION

Grapevine flavescence dorée phytoplasma only occurs in Europe. Its distribution is reported in eight of the main grape-growing EU Member States (Austria, Croatia, France, Hungary, Italy, Portugal, Slovenia and Spain) as well as in Switzerland and in Serbia (Jeger *et al.*, 2016). Localized cases have been reported in Germany (Jarausch *et al.*, 2021).

Since its introduction in Europe, probably in South-Western France, the vector of *Grapevine flavescence dorée phytoplasma sensu stricto: S. titanus*, native to North America (Papura *et al.*, 2012), has spread to 18 European countries (Austria, Bosnia-Herzegovina, Bulgaria, Croatia, Czech Republic, France, Hungary, Italy, Moldova, Montenegro, Portugal, Spain, Romania, Serbia, Slovakia Slovenia, Switzerland, Ukraine) making its distribution wider than that of *grapevine flavescence dorée phytoplasma* (Jeger *et al.*, 2016). *S. titanus*'s life cycle requires *Vitis* spp.; therefore, its presence is strictly connected to that of grapevines.



EPPO Region: Austria, Croatia, Czechia, France (mainland, Corse), Hungary, Italy (mainland), Montenegro, Portugal (mainland), Romania, Serbia, Slovakia, Slovenia, Spain (mainland), Switzerland

BIOLOGY

In the infected plants, *grapevine flavescence dorée phytoplasma* is located and multiplies within phloem sieve tubes (Caudwell *et al.*, 1971). In winter it survives in the roots and canes from where it moves to the upper parts of the plants during spring. *S. titanus* is the most efficient vector of *grapevine flavescence dorée phytoplasma* (Mori *et al.*, 2002; Schvester *et al.*, 1963) (see below for details of alternative vectors). At the initial stage of its life-cycle, *grapevine flavescence dorée phytoplasma* is acquired from phloem sap by the insects. It passes through the insect's alimentary canal and multiplies in the midgut. Then it colonises the haemolymph before entering and multiplying in the salivary gland. Finally, it is delivered to another host plant through saliva (Lefol *et al.*, 1993; Lefol *et al.*, 1994). There is a temperature-dependent latency period of 10 to 45 days between the phytoplasma's acquisition by *S. titanus* and its ability to transmit it to another host plant. The transmission mode is qualified as persistent and propagative, because after phytoplasma acquisition and latency the vector remains infectious for life.

S. titanus is monophagous on grapevine. It has one generation a year; eggs laid in the bark of >2-year-old grapevine wood from the end of July are the overwintering stage. There are five larval instars, which develop from mid-May to mid-July. Adults appear from July until the beginning of September (Chuche & Thiery, 2014). The larval stages and adults are capable of acquiring the phytoplasma, but males are more efficient than females in transmitting the disease. There is no evidence of adult-to-egg transmission (Schvester *et al.*, 1969).

Transmission is also possible through grafting (Boudon-Padieu, 2002). There are no reports on transmission by root grafting. *Grapevine flavescence dorée phytoplasma*, like all phytoplasmas, is not known to be transmissible by mechanical inoculation.

Phytoplasmas of the taxonomic group 16SrV-C are widespread in alders (*A. glutinosa* and *A. incana*) all over Europe (Atanasova *et al.*, 2014; Holz *et al.*, 2016; Malembic-Maher *et al.*, 2020; Mehle *et al.*, 2011; Radonjic *et al.*,

2013). Some of the phytoplasma strains that are transmitted from alder to alder by the Macropsinae leafhopper *Oncopsis alni* and occasionally to grapevine are referred to as Palatinate Grapevine Yellows (PGY) and are not compatible with *S. titanus*. Other phytoplasma strains transmitted from alder to alder and occasionally to grapevine by the Deltocephalinae leafhopper *Allygus mixtus* and *Orientus ishidae* can be acquired and be transmitted by *S. titanus* (Maixner & Reinert, 1999; Maixner *et al.*, 2000; Malembic-Maher *et al.*, 2020). In Italy and Serbia, particular *grapevine flavescence dorée phytoplasma* strains are present in wild clematis (*C. vitalba*) from which they can be transmitted to grapevine by the planthopper *Dictyophara europaea* (synonym: *Eiptera europaea*) (Filippin *et al.*, 2009). The frequency of phytoplasma transmission from alders and clematis to grapevine remains to be determined but this cannot provoke a *grapevine flavescence dorée phytoplasma* outbreak in the absence of the leafhopper *S. titanus*.

DETECTION AND IDENTIFICATION

Symptoms

Symptoms are used as indicators of the presence of *grapevine flavescence dorée phytoplasma* but the symptoms of other grapevine yellows are identical, and therefore molecular tests are required for identification. Symptoms of *grapevine flavescence dorée phytoplasma* generally become clearly apparent during summer. Either a group of shoots is affected on each grapevine, or the whole vine may show symptoms (Belli *et al.*, 1973; Caudwell, 1964).

On shoots

The shoots of susceptible grapevines usually fail to lignify and during winter, the non-lignified branches blacken and die. If infected later in the season, lignification is interrupted.

On leaves

The leaves show colour aberrations and downward-rolled margins. In white-fruited cultivars there is a yellowing of the portion of the lamina exposed to the sun that confers a metallic lustre to the leaf surface. Red-fruited cultivars develop a similar pattern of colour changes of the leaves, but the discolorations are reddish. The central portion of the discoloured areas becomes necrotic and dries out.

On fruit

Fruit setting is reduced on grapevines infected early in the season, and the inflorescences dry out and fall off. In later infections, bunches become brown and shrivelled, and the peduncles dry out.

Morphology

Grapevine flavescence dorée phytoplasma can be visualized by electron microscopy in the phloem of infected grapevines as wall-less rounded pleiomorphic bodies with an average diameter ranging from 200 to 800 µm (Caudwell *et al.*, 1971).

Detection and inspection methods

Visual examination of *flavescence dorée* requires skilled inspectors as it is complicated by many factors: the cultivar-dependent symptom expression (particularly when mild) and the possible risk of misidentification. Detecting symptoms is even more difficult in abandoned vineyards and in wild *Vitis* spp. plants, which are often difficult to gain access to. Furthermore, the absence of symptoms on infected rootstocks affects both feasibility and effectiveness of the surveillance for the detection of all infected plants in nurseries (Jeger *et al.*, 2016). The susceptibility of the plants to *grapevine flavescence dorée phytoplasma* infection is also genotype-dependent: 28 *Vitis* genotypes were compared including grapevine cultivars, rootstocks and wild species, proving that even wild *Vitis* rootstock plants may be highly infected with *grapevine flavescence dorée phytoplasma* in their natural environment (Eveillard *et al.*, 2016). General guidelines for inspection of *Vitis* plants for planting are available in the phytosanitary procedure PM 3/85 *Inspection of places of production – Vitis plants for planting* (EPPO, 2018).

According to EPPO diagnostic protocol PM 7/079, sampling should be performed from July to October: A total of 20 symptomatic leaves per plant should be sampled, pooling a maximum of five plants together (EPPO, 2016b).

Current detection methods rely on the use of polymerase chain reaction (PCR) (Chabirand *et al.*, 2017; EPPO, 2016b). For tracing pathways for spread, various genetic typing methods are commonly used (Arnaud *et al.*, 2007; Botti & Bertaccini, 2007; Martini *et al.*, 2002; Rossi *et al.*, 2019). The distribution of the eleven phytoplasma genotypes associated with outbreaks of flavescence dorée varies depending on the geographical regions (Casati *et al.*, 2017; Jeger *et al.*, 2016; Krstic *et al.*, 2022; Malembic-Maher *et al.*, 2020; Mehle *et al.*, 2011; Plavec *et al.*, 2019; Rossi *et al.*, 2019).

Identification of epidemic vectotypes II and III can be achieved by partial or complete sequencing of vmp genes encoding surface adhesins involved in vector colonization (Malembic-Maher *et al.*, 2020; Rossi *et al.*, 2019).

PATHWAYS FOR MOVEMENT

Two pathways are identified for the long-distance spread of *grapevine flavescence dorée phytoplasma*: infected propagative material being moved; and infested vectors flying from adjacent spatial units, transported on plants for planting or hitchhiking in vehicles. Local spread is also possible by transfer from infected alder or clematis to vineyards by dispersion via alternative vectors. The relative contribution of these various mechanisms to the spread of *grapevine flavescence dorée phytoplasma* was estimated upon historical reconstruction of *grapevine flavescence dorée phytoplasma* spread in the European regions. Propagation events through propagated material and dispersion through vector were estimated as contributing 37% and 57% respectively, while local transfer from wild plants via alternative vectors was estimated at 6% (Jeger *et al.*, 2016). Within vineyards, natural spread of the disease is likely to occur over up to a few hundred metres owing to the flight behaviour of the *S. titanus* vector (Lessio *et al.*, 2014).

Grapevine flavescence dorée phytoplasma cannot be transmitted mechanically (e.g. with pruning scissors), but it can be transmitted, with very low efficiency, by grafting (Osler *et al.*, 2002).

PEST SIGNIFICANCE

Economic impact

In infested grapevine areas where the disease is epidemic, *grapevine flavescence dorée phytoplasma* has a dramatic impact on yield. When allowed to spread uncontrolled, epidemic flavescence dorée had catastrophic consequences. Between 1949 and 1954 in Armagnac and Chalosse (France), all Baco 22A grapevines became infected. The quality of the wine produced from infected grapevines is also affected owing to a lower sugar content and higher acidity of the grapes. The estimated yield loss under current management practices is estimated 0.5-1% of the EU wine and grape production (Jeger *et al.*, 2016).

In addition to the high cost of repeated insecticide treatments for vine growers, non-intentional impacts such as negative effects on pollinators or interference with integrated pest management targeting other pests may occur (EFSA, 2014).

Control

In practice, it is not possible to cure a plant that is infected by *grapevine flavescence dorée phytoplasma*; however, several control methods are currently applied in the EU. Insecticide application against the vector, *S. titanus*, is compulsory where both the vector and *grapevine flavescence dorée phytoplasma* are present; the number of applications in commercial vineyards varies from one to three per year. These target nymphs and adults, and can be more numerous in nurseries. The presence of the vector is currently monitored by hanging yellow sticky traps in the vineyards and direct counting of nymphs under the grapevine to support decisions on insecticide application and timing. Roguing of infected plants is compulsory and, when infected plants exceed 20 % to 30 % of the plots, all the plants in the plot have to be removed. Abandoned plots and wild *Vitis* rootstock plants in the areas surrounding vineyards should be removed as they represent a reservoir of both *grapevine flavescence dorée phytoplasma* and its vector. Hot-water treatment (45 min at 50°C) of the dormant rootstocks and scions or grafted cuttings is widely

applied and is known to be effective in killing both *grapevine flavescence dorée phytoplasma* and vector eggs (Caudwell *et al.*, 1997; Caudwell *et al.*, 1992; Linder *et al.*, 2010; Mannini & Marzachi, 2007). EPPO Standard PM 10/18 describes the conditions under which such a treatment should be performed (EPPO, 2012). The production of certified grapevine varieties and rootstocks should also be performed under conditions (e.g. maintenance, testing etc.) preventing *grapevine flavescence dorée phytoplasma* infections, as recommended in EPPO Standard PM 4/8 (EPPO, 2008).

When an outbreak is reported, risk areas can be defined around *grapevine flavescence dorée phytoplasma* infected vines for surveillance, bearing in mind that their size depends on the spread capacity of the vectors and the availability of host plants around these locations. For a detection survey, a risk area of about 50 m wide around the possible risk locations can be defined. Around the infested zone, a buffer zone of at least 1.3 km wide was recommended by EFSA (EFSA, 2020). The areas where the vector is present can also be prioritized for *grapevine flavescence dorée phytoplasma* detection surveys, and different levels of risk can be defined based on vector density in the vineyards. To prevent the development of large outbreaks, the EFSA Panel of experts has proposed that one-third of vineyards should be surveyed on a yearly basis in addition to the already surveyed buffer zones around outbreaks (EFSA, 2020).

Increased surveillance and mapping of major spots of the wild *Vitis* spp. vegetation, their removal in the surrounding of the vineyards and before establishing new plantations are currently poorly applied but would result in a higher effectiveness in *grapevine flavescence dorée phytoplasma* control (Jeger *et al.*, 2016).

Phytosanitary risk

There is limited information on *grapevine flavescence dorée phytoplasma*'s response to temperature, and it is likely that this phytoplasma would be able to infect grapevines wherever they grow. Although *grapevine flavescence dorée phytoplasma* has been reported in several of the main grape and wine producers in Europe, it has sometimes still a relatively restricted distribution in these countries. The reports of *grapevine flavescence dorée phytoplasma* in Burgundy, France and in Southern Italy, together with the increasingly southerly distribution of the *grapevine flavescence dorée phytoplasma*-specific vector, *S. titanus*, indicate that the bacterium could establish in new important viticultural areas in the EU, namely in the Mediterranean region (EFSA, 2014)

Being a principal pathway for the long-distance spread of *grapevine flavescence dorée phytoplasma* (Jeger *et al.*, 2016), the movement (including trade) of *Vitis* plant propagating material is at risk when originating from countries where *grapevine flavescence dorée phytoplasma* is already present. The presence of American vines left from the rootstocks of abandoned vines and other *Vitis* spp. growing in the wild provides a reservoir for both the pathogen and the vector with movement of *S. titanus* from untreated to treated vineyards (Pavan *et al.*, 2012; Ripamonti *et al.*, 2020).

PHYTOSANITARY MEASURES

Appropriate phytosanitary measures to import *Vitis* plants for planting (other than seeds) into the EPPO region could require that these plants are produced in a pest free area or in a pest free place/site of production for grapevine flavescence dorée phytoplasma established according to EPPO Standard PM 5/8 *Guidelines on the phytosanitary measure 'Plants grown under physical isolation'* (EPPO, 2016a). The physical isolation should ensure that both the phytoplasma and the vector are prevented from entering the place/site of production. The production under a pest-free place of production with a buffer zone, and treatments at appropriate times throughout the growing season to prevent vector infestation may also be considered. A number of EPPO countries already ban the import of *Vitis* plants for planting (other than seeds) (e.g. EU countries: Annex VI, points 10 of Regulation 2019/2072 (EU, 2019). Host plants for planting could also be imported using post-entry quarantine (in the framework of a bilateral agreement).

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