Brussels, 20th July 2016

Recommendation of the Working Group on the Annexes of the Council Directive 2000/29/EC – Section II – Listing of Harmful Organisms as regards the future listing of *Candidatus* Phytoplasma mali *[Apple Proliferation Mycoplasm]* [1]

Current regulatory status

Candidatus Phytoplasma mali Seemüller & Schneider (CPm) is currently regulated in Annex I, Part A, Section II of the plant health Directive 2000/29/EC as 'Apple proliferation mycoplasm'.

Import requirements of plants of *Malus* Mill. are listed under Annex IVAI requiring an official statement (phyto certificate (Annex VBI) that (a) the plants originate in areas known to be free from CPm or (b) no symptoms of CPm disease have been observed on plants at the place of production, or on susceptible plants in its immediate vicinity, since the beginning of the last complete three cycles of vegetation and mother plants derive from an official certification scheme or other scheme of testing for CPm at least once within the last six complete cycles of vegetation.

Intra-EU movement of plants of *Malus* Mill. is subject to a plant passport (Annex IVAII and Annex VAI) specifying that a) the plants originate in areas known to be free from CPm or b) no symptoms of CPm disease have been observed on plants at the place of production, or on susceptible plants in its immediate vicinity, since the beginning of the last complete three cycles of vegetation and mother plants derive from an official certification scheme or other scheme of testing for CPm at least once within the last six complete cycles of vegetation.

Marketing directive 2008/90/EC regulates *Malus* propagating material and fruit plants intended for fruit production, plant health requirements are specified in Commission Directive 2014/98/EU.

Identity of the pest

Apple proliferation mycoplasma has recently been renamed *Ca.* Phytoplasma mali Seemüller & Schneider (CPm). It belongs to Apple proliferation group together with *Ca.* Phytoplasma pyri (Pear decline) and *Ca.* Phytoplasma prunorum (European stone fruit yellows)[2].

Phytoplasmas reside endocellularly within the plant phloem and insects, which feed from phloem. Naturally they are spread among plants by insect vectors, mainly leafhoppers (Hemiptera: Cicadellidae, Cixiidae, Delphacidae and Derbidae), and psyllids (Psyllidae). *C*Pm is phloem-obligate parasite of plants transmitted by sap-feeding psyllids *Cacopsylla picta* (the most important; syn. *C. costalis*) and *C. melanoneura*. Putative vector is *Fieberiella florii*. There are some other potential vectors, which were found positive, but transmission has not been confirmed*: C. pyri, C. peregrina, C. affinis, C. crataegi.*

CPm multiplies within the tissues of its insect vector and is transferred in the salivary secretions to new host plants during feeding. Phytoplasma epidemiology involves a tritrophic relationship between the pathogen and usually several plant hosts and vectors. CPm infects manly apple trees *Malus* spp., but it was confirmed also in *Prunus avium, P. armeniaca, and P. domestica.*

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[1] Scientific basis for the recommendation: Prima Phacie, 2012. Pest risk assessment for the European Community plant health: A comparative approach with case studies. External scientific report by group of authors: http://www.efsa.europa.eu/fr/supporting/doc/319e.pdf

[2] Firrao G., Andersen, M., Bertaccini, A., Boudon, E. et al., 2004. 'Candidatus Phytoplasma'a taxon for the wall-less, non-helical prokaryontes that colonize plant phloem and insects. International Journal of Systematic and Evolutionary Microbiology 54, 1243-1255.

CPm is a well-defined species, for which molecular detection assays are available (PCR, RFLP, Biplex nested PCR).

Two psyllids Cacopsylla picta and *C. melanoneura* are recognized vectors which transmit the phytoplasma in a persistent manner. In winter, the phytoplasma largely degenerates simultaneously with the degeneration of the phloem tissue in the aerial part of an infected tree but it survives in the roots where functional sieve tubes are present throughout the whole year. From the roots, the stem may be recolonized in the following spring.

Distribution of the pest

CPm is endemic in the European area. As an obligate symbiont of *Malus* plants it is already widespread in the EU. It is present in at least 17 of the 28 EU countries, including the largest apple production areas in Europe (https://gd.eppo.int/taxon/PHYPMA/distribution). It is not reported to occur in apples in Cyprus, Denmark, Estonia, Ireland, Latvia, Lithuania, Luxembourg, Portugal, Sweden, Luxembourg, and UK (in Malta apple is not produced). There is an insignificant trade of *Malus* plants from non-EU countries where Ca. Phytoplasma mali is present.

Potential for establishment and spread in the PRA area

The probability of CPm being associated with the planting material at origin strongly depends on the rate of occurrence of both the phytoplasma and its vectors in the country and even the region and the field where the plants are grown. The likelihood of detection and removal of infested lots depends on the climatic conditions for symptom expression and on the intensity of inspections and testing at the place of production. The inspection and testing regime in all EU countries are unknown.

Therefore, the likelihood of association with the pathway at origin is highly uncertain, and was judged moderate. With no post-harvest treatment available and plants not inspected/tested at the place of destination, this likelihood remains until the point of transfer.

Overall likelihood of entry and transfer via planting material from infected areas to uninfected area in the EU was assessed low. CPm has been a listed quarantine organism since 1992 and under current regulations the potential area occupied is not expected to change significantly over the next 20 years.

The main pathway for entry is via infected planting material used for apple production (bud- and graftwood, rootstocks and wholesale plants). The sources of initial inoculum for infection of the aerial part of host *Malus* trees during spring are infected rootstocks or vector psyllids.

Potential for consequences in the PRA area

CPm can have considerable impact on fruit production. In areas with less suitable conditions and/or absence or low abundance of vectors impact on yield and quality would be limited. However, the importance of the eco-epidemiological perspective and the understanding of tripartite interactions among host plant, plant pathogen and herbivore vector that is crucial for the successful control of plant diseases as those, caused by *Ca.* Phytoplasma mali, *Ca.* Phytoplasma prunorum, *Ca.* Phytoplasma pyri.

In case CPm was removed from the plant health directive 2000/29/EC, the probability that the pest is found to be associated with planting material is similar to the current situation and likelihood of entry remains similar since the Marketing Directive will provide certain protection similar to the current measures. In a scenario with no official control at all, i.e. no measures applied in orchards and no certification system for *Malus* plants, then the number of outbreaks in the area of origin will increase and lead to the likelihood of association at origin increasing. However, the overall

likelihood of entry and transfer increases only moderately since conditions for establishment and disease expression still remain low.

Recommendation

The WG recommends re-classifying this organism as an RNQP with the possibility of Protected Zone status.

For Protected Zones, monitoring of *Malus* plants, through sampling and testing, particularly those asymptomatic, should be further considered, as plants can be infected and remain without symptoms for up to five years after infection.

The WG highlights as well the need of a proper a scientific evaluation of the risk reduction options and their feasibility as proposed by the Prima Phacie project (presented in the section below). Combination of risk reduction options could be applied depending on the type and category of plants for planting.

Background information on possible measures to be further developed

- Visual inspection – CPm is not so likely detected during visual inspection of consignments at import or export; visual inspections of nurseries is more reliable.

- Testing - needed for confirmation of visual symptoms and in certification schemes (mainly for pre-basic mother plants (nuclear stock); testing of nurseries plants will have to be performed at random, since it is quite laborious and costly).

- Control treatments - no effective chemical or biological treatment available; thermotherapy by hot air is possibility for obtaining pre-basic mother plants free of CPm; thermotherapy may not be practiced in mass production of plants for planting.

- Removal of infected plants - removal of infected trees contributes to control the phytoplasma, but some infected trees may be overlooked at early phase of infection; decision should be made on the removal of either all plants from a lot/field or a selected number of plants. Removal of infected plants is important in breaking of epidemiological cycle in tripartite interactions among host plant, plant pathogen and herbivore vector (diseased trees attract psyllids Cacopsylla picta by allomone ß-kariofilen – more effective acquisition and transmission)

- Replacing summer budding by late winter grafting, when CPm move from branches in the phloem of infested plants to overwinter in the roots of susceptible rootstocks, where functional sieve tubes are present throughout the year. Such grafting reduces the risk of APP transmission from infected mother plants to the grafted rootstocks.

- Use of resistant cultivars/rootstocks (potential for development, not yet available). Wide variation in susceptibility of different apple cultivars exists. High levels of resistance were obtained for rootstocks of M. sieboldii-derived progenies. However, trees on these resistant rootstocks are more vigorous and have lower yields than trees on the rootstock M9, which is generally used for commercial apple production.

- Complete physical isolation may be used to prevent new infections in *Malus* plants by the psyllid vectors (in insect-proof greenhouse or screenhouse). In this case plants must originate from mother plants that have been tested and found free of CPm, followed by visual inspections and retesting. Appropriate for production of pre-basic mother plants and

maybe also basic mother plants (the first generation of plants from the nuclear stock) under insect-proof conditions in areas where both CPm and its vectors occur. For further stages in the production of *Malus* plants for planting, physical isolation may be too costly.

- Optimising growing conditions (water and nutrient supply) in commercial orchards would reduce damage and contribute to management of CPm. The root system of young infected trees forms compact felt-like masses of short roots and root weight is substantially reduced. As a result uptake of water and nutrients is hindered.

- Vector control: In areas where the disease is present, both the summer stage and the immigrating winter stage must be controlled to reduce the spread of CPm. Because of vector's highest transmission efficiencies, especial attention should be given to re-migrating adult psyllid vectors in course of winter or the beginning of spring. However, the control of the winter form in early spring can be difficult in years when oviposition coincides with the period of blossom, when insecticides cannot be applied. Alternatively, control can be concentrated on preventing the reproduction of the vector in apple orchards, which consequently, also reduces the number of re-migrants. In addition, the control of psyllid vectors by insecticides may be hampered due to low population pressure and the lack of adequate pesticides.

- Use of certified plants: enable production of *Malus* trees with quite high but not absolute guarantees of CPm freedom. Official certification systems including adequate (re)testing, isolation of propagation material, eradication of infected trees and vector control should preferred over CAC material because of better guarantees for the health status regarding APP

  Council Directive 2008/90/EC of 29 September 2008 applies to the marketing of fruit plant propagation material and fruit plants, including plants of apple (*Malus* spp.). Article 3 specifies that propagating material may only be marketed as either pre-basic, basic, certified or CAC (Conformitas Agraria Communitatis) material (European-Council-Directive, 2008). Detailed harmonised plant health requirements are specified in Commission Directive 2014/98/EU.

- Pest free place of production / pest free area (= geographical isolation)

  1) Pest free areas could be implied in *Malus* producing areas within the European Union where CPm is absent, effective measures are in operation preventing the introduction of CPm with plants for planting, and the pest status is checked conclusively. In such areas pest-free places of production or pest-free production sites are superfluous.

  2) In *Malus* producing areas within the European Union where CPm and vectors are present, a role for pest-free places of production or pest-free production sites should be properly assessed about its feasibility.