

# Classification of Pome and Stone Fruit Viruses, Viroids, and Phytoplasmas

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## Introduction

In the current literature, more than 50 different viruses, 6 viroids, and more than 11 different phytoplasmas are recorded from pome and stone fruit species. Several viruses and phytoplasmas, however, are either of little economic importance, are poorly characterized, or are synonymous with previously described species. Thus, the tables below list only relevant viruses and viroids which, with few exceptions, are recorded in the 8th Report of the International Committee on Taxonomy of Viruses (ICTV) (Fauquet et al., 2005) as definitive or tentative species of known taxa. However, with respect to the 8th Report of ICTV, the classification of some of these taxa and virus species has changed recently following the establishment of two novel orders (*Picornavirales* and *Tymovirales*), two new families (*Secoviridae*, *Betaflexiviridae*) and a subfamily (*Comovirinae*, former family *Comoviridae*) approved in 2009 by the ICTV (E. Carstens, *personal communication*).

Since 1991 (Martelli, 1992; Mayo and Brunt, 2001) plant viruses and viroids are classified in the traditional taxonomic system (family-genus-species), according to the revised version of the "International Code of Virus Classification and Nomenclature" (Mayo and Horzinek, 1998). This code states that the ICTV does not address virus classification issues below the level of species. Viral and viroidal species are identified on the basis of a number of properties including structural, biological, serological, cytopathological, epidemiological, and molecular traits such as genome structure and organization and level of similarity or identity (expressed as percent) in the nucleotide and amino acid sequences. This latter trait is an important demarcation criterion for establishing whether two given viruses are different species or strains of the same species. There are no set rules for establishing the minimum level of genome sequence relatedness for species identification for it varies with the genus. Phylogenetic relationships of species within a genus and among genera within a family are determined on the relatedness of the whole genome sequence (viroids) or of the sequence of some key proteins such as coat protein, movement protein, and RNA-dependent RNA polymerase (viruses). Names of families, genera, and definitive (i.e., formal) viral and viroidal species are written in italic script, e.g., *Tombusviridae* (family), *Tombusvirus* (genus), and *Tomato bushy stunt virus* (species). Names of tentative species are written in roman

script, e.g., Maize necrotic streak virus. Abbreviations of virus names end with a "V" (e.g., *Tomato bushy stunt virus*, TBSV), abbreviations of viroid names end with a "Vd" (e.g., *Potato spindle tuber viroid*, PSTVd) (Mayo and Brunt, 2001).

Fruit tree viruses currently are classified into 12 different genera: *Ampelovirus*, *Capillovirus*, *Cheravirus*, *Foveavirus*, *Idaeovirus*, *Ilarvirus*, *Ourmiavirus*, *Potyvirus*, *Nepovirus*, *Sadwavirus*, *Tombusvirus*, and *Trichovirus*, belonging to six different families: *Bromoviridae*, *Closteroviridae*, *Secoviridae* (order *Picornavirales*), *Betaflexiviridae* (order *Tymovirales*), *Potyviridae*, and *Tombusviridae*.

Fruit tree viroids currently are classified into three different genera: *Hostuviroid*, *Apscaviroid*, and *Pelamoviroid*, belonging to two families, *Pospiviroidae* and *Avsunviroidae*.

Phytoplasma classification and nomenclature, in particular, differ from the above because of the incomplete characterization of these organisms. The name phytoplasma was adopted in 1992 (International Committee on Systematic Bacteriology, 1993) to describe non-helical, non-culturable, wall-less plant pathogenic prokaryotes inhabiting plants and insects. Previously, phytoplasmas were termed mycoplasma-like organisms (Doi et al., 1967) or MLO in resemblance to the wall-less human or animal pathogens and saprophytic members of the genus *Mycoplasma*. The traditional names given to the pathogens, e.g., apple proliferation or pear decline, describing the plant host and the most prominent disease symptom, are still in use for practical reasons. However, a comprehensive classification scheme for these organisms was lacking until molecular tools were employed. In particular, sequence analysis of the highly conserved 16S rRNA gene permitted identification of a given phytoplasma and revolutionized phytoplasma taxonomy. Restriction fragment length polymorphism (RFLP) analysis of the 16S rRNA gene proved to be practical and useful for a rapid classification (Lee et al., 1993; Schneider et al., 1993). According to this classification system, 17 RFLP groups and more than 40 subgroups have been defined (IRPCM Phytoplasma/Spiroplasma Working Team, 2004; Arocha et al., 2005). In a recent computer survey, more than 800 16S rRNA database entries were subjected to in-silico RFLP analysis, following which the number of 16S rRNA RFLP groups increased to 28 (Wei et al., 2007). Further analysis of conserved genes like the *tuf* gene or ribosomal protein genes corroborated and



refined this classification system (Lee et al., 1998; Marcone et al., 2000). It became obvious that phytoplasmas causing similar symptoms in plants could be different pathogens and were not necessarily closely related as considered before. Phylogenetic analysis revealed that phytoplasmas are a coherent monophyletic group within the class *Mollicutes* with the nearest relative amongst the achleplasmas (Lim and Sears, 1989).

Since phytoplasmas are still non-culturable, the traditional binomial nomenclature according to the International Code of Nomenclature of Bacteria is not applicable. However, for the unambiguous description of non-culturable bacteria, the category *Candidatus* was proposed (Murray and Schleifer, 1994) and implemented (Murray and Stackebrandt, 1995), forming the basis for a preliminary phytoplasma species description (Firrao et al., 2005). As a minimal requirement for a *Candidatus* description, the 16S rDNA (>1,000 bases) sequence and a unique signature sequence of this gene must be provided. To restrict proliferating *Candidatus* species descriptions, it was agreed that phytoplasmas sharing more than 97.5% sequence similarity are considered to belong to the same species, unless other data (e.g., serology, vector transmission, host range) support a separate description. At the time of submitting the manuscript, 26 *Candidatus* Phytoplasma species assigned to the family *Acholeplasmataceae* have been described validly.

### Virus List

In the tables below there are 35 virus species listed. Of those, five, i.e., *Apple chlorotic leafspot virus* (ACLSV), *Apple mosaic virus* (ApMV), *Apple stem grooving virus* (ASGV), *Cherry rasp leaf virus* (CRLV), and *Tomato ringspot virus* (ToRSV) are pathogens on pome and stone fruit species. ACLSV, ApMV, and ASGV have a natural host range restricted

to fruit trees, whereas ToRSV and a number of other members of the genera *Nepovirus*, *Sadwavirus*, and *Tombusvirus* have a wider natural host range, comprising both herbaceous and woody crops.

### Viroid List

Of the six viroids listed, two, i.e., *Hop stunt viroid* (HSVd) and *Peach latent mosaic viroid* (PLMVd) infect pome and stone fruit species, whereas *Apple scar skin viroid* (ASSVd), *Apple dimple viroid* (ADVd), *Apple fruit crinkle viroid* (AFCVd), and *Pear blister canker viroid* (PBCVd) have a host range restricted to pome fruit trees.

### Phytoplasma List

Of the 11 phytoplasmas in the list, some were reported to occur in non-rosaceous plant families. For example, *Candidatus* Phytoplasma asteris, the agent of apple sessile leaf, belongs to the aster yellows phytoplasma group, which has the largest natural host range known. Other pome and stone fruit phytoplasmas have also been found in alternative hosts, but whether those findings reflect single infection events or whether those plants represent reservoirs remain unclear. Some pome and stone fruit diseases tentatively associated with phytoplasmas, namely Apple decline, Apple chat fruit, Apple rubbery wood, Cherry decline, Cherry rosette, Cherry albino, and Cherry blossom anomaly are not included in the list provided here. Although these diseases are graft transmissible, their causal agents remain to be identified. Detailed studies on Apple rubbery wood isolates including dsRNA extractions, different nested PCR protocols with phytoplasma group specific primers, and rPAGE failed to identify an RW associated causal agent (Menzel, 2003).

## The Pathogens and Their Classification

### Pome fruit viruses

Virus	Abbreviation	Family	Genus	Main hosts
<i>Apple chlorotic leafspot virus</i>	ACLSV	<i>Betaflexiviridae</i>	<i>Trichovirus</i>	Apple, pear, quince
<i>Apple latent spherical virus</i>	ALSV	<i>Secoviridae</i>	<i>Cheravirus</i>	Apple
<i>Apple mosaic virus</i>	ApMV	<i>Bromoviridae</i>	<i>Ilarvirus</i>	Apple
<i>Apple stem grooving virus</i>	ASGV	<i>Betaflexiviridae</i>	<i>Capillovirus</i>	Apple, pear, quince
<i>Apple stem pitting virus</i>	ASPV	<i>Betaflexiviridae</i>	<i>Foveavirus</i>	Apple, pear, quince
<i>Cherry rasp leaf virus</i>	CRLV	<i>Secoviridae</i>	<i>Cheravirus</i>	Apple
<i>Raspberry bushy dwarf virus</i>	RBDV	Unassigned	<i>Idaeovirus</i>	Quince
<i>Tomato ringspot virus</i>	ToRSV	<i>Secoviridae</i>	<i>Nepovirus</i>	Apple
<i>Tulare apple mosaic virus</i>	TAMV	<i>Bromoviridae</i>	<i>Ilarvirus</i>	Apple

### Pome fruit viroids

Viroid	Abbreviation	Family	Genus	Main hosts
<i>Hop stunt viroid</i>	HSVd	<i>Pospiviroidae</i>	<i>Hostuviroid</i>	Pear
<i>Apple scar skin viroid</i>	ASSVd	<i>Pospiviroidae</i>	<i>Apscaviroid</i>	Apple, pear
<i>Apple dimple fruit viroid</i>	ADFVd	<i>Pospiviroidae</i>	<i>Apscaviroid</i>	Apple
<i>Apple fruit crinkle viroid</i>	AFCVd	<i>Pospiviroidae</i>	<i>Apscaviroid</i>	Apple
<i>Pear blister canker viroid</i>	PBCVd	<i>Pospiviroidae</i>	<i>Apscaviroid</i>	Pear
<i>Peach latent mosaic viroid</i>	PLMVd	<i>Avsunviroidae</i>	<i>Pelamoviroid</i>	Pear, wild pear, quince

### Stone fruit viroids

Viroid	Abbreviation	Family	Genus	Main hosts
<i>Hop stunt viroid</i>	HSVd	<i>Pospiviroidae</i>	<i>Hostuviroid</i>	Peach, plum, apricot, almond
<i>Peach latent mosaic viroid</i>	PLMVd	<i>Avsunviroidae</i>	<i>Pelamoviroid</i>	Peach, apricot, cherry, plum, almond



## Stone fruit viruses

Virus	Abbreviation	Family	Genus	Main hosts
American plum line pattern virus	APLPV	Bromoviridae	Ilarvirus	Plum, peach, sweet cherry
Apple chlorotic leafspot virus	ACLSV	Betaflexiviridae	Trichovirus	Almond, apricot, cherry, peach, plum
Apple mosaic virus	ApMV	Bromoviridae	Ilarvirus	Almond, apricot, cherry, hazelnut, peach, plum
Apple stem grooving virus	ASGV	Betaflexiviridae	Capillovirus	<i>Prunus mume</i>
Apricot latent virus	ApLV	Betaflexiviridae	Foveavirus	Apricot, peach
Apricot pseudo chlorotic leafspot virus	APCLSV	Betaflexiviridae	Trichovirus	Apricot, peach
Arabidopsis mosaic virus	ArMV	Secoviridae	Nepovirus	Cherry, peach
Asian prunus virus 1, 2, 3	APV-1, APV-2, APV-3	Betaflexiviridae	Foveavirus	Peach, <i>Prunus mume</i>
Cherry green ring mottle virus	CGRMV	Betaflexiviridae	Foveavirus	Cherry, apricot
Cherry leafroll virus	CLRV	Secoviridae	Nepovirus	Cherry, walnut
Cherry mottle leaf virus	CMLV	Betaflexiviridae	Trichovirus	Almond, apricot, cherry, peach, plum
Cherry necrotic rusty mottle virus	CNRMV	Betaflexiviridae	Foveavirus	Apricot, cherry, peach
Cherry rasp leaf virus	CRLV	Secoviridae	Cheravirus	Cherry, peach, <i>Prunus mahaleb</i>
Cherry rosette virus	CRV	Secoviridae	Nepovirus	Cherry
Cherry virus A	CVA	Betaflexiviridae	Capillovirus	Apricot, cherry, peach, plum
Epirus cherry virus	EpCV	Unassigned	Ourmiavirus	Cherry
Little cherry virus 1	LChV-1	Closteroviridae	Unassigned to the family	Cherry
Little cherry virus 2	LChV-2	Closteroviridae	Ampelovirus	Cherry
Myrobalan latent ringspot virus	MLRSV	Secoviridae	Nepovirus	Myrobalan, peach
Peach chlorotic mottle virus	PcCMV	Betaflexiviridae	Foveavirus	Peach
Peach mosaic virus	PcMV	Betaflexiviridae	Trichovirus	Almond, apricot, peach plum
Peach rosette mosaic virus	PRMV	Secoviridae	Nepovirus	Peach
Petunia asteroid mosaic virus	PAMV	Tombusviridae	Tombivirus	Cherry
Plum bark necrosis and stem pitting-associated virus	PBNSPaV	Closteroviridae	Ampelovirus	Almond, apricot, cherry, peach, plum
Plum pox virus	PPV	Potyviridae	Potyvirus	Almond, apricot, cherry, peach, plum
Prune dwarf virus	PDV	Bromoviridae	Ilarvirus	Almond, apricot, cherry, peach, plum
Prunus necrotic ringspot virus	PNRSV	Bromoviridae	Ilarvirus	Almond, apricot, cherry, peach, plum
Raspberry ringspot virus	RpRSV	Secoviridae	Nepovirus	Cherry, plum
Stocky prune virus	StPV	Secoviridae	Cheravirus	Plum
Strawberry latent ringspot virus	SLRSV	Secoviridae	Sadwavirus	Apricot, cherry, peach
Tobacco ringspot virus	TRSV	Secoviridae	Nepovirus	Cherry
Tomato black ring virus	TBRV	Secoviridae	Nepovirus	Almond, cherry, peach
Tomato ringspot virus	ToRSV	Secoviridae	Nepovirus	Apricot, cherry, peach, plum
Tulare apple mosaic virus	TAMV	Bromoviridae	Ilarvirus	Apricot, hazelnut

## Pome fruit phytoplasmas

Common name of the disease	Abbreviation	Species name	Group affiliation	Main hosts
Apple proliferation	AP	<i>Candidatus</i> Phytoplasma mali	Apple proliferation (AP)	Apple
Apple sessile leaf	ApSL	<i>Ca.</i> Phytoplasma asteris	Aster yellows (AY)	Apple
Pear decline	PD	<i>Ca.</i> Phytoplasma pyri	AP	Pear, quince

## Stone fruit phytoplasmas

Common name of the disease	Abbreviation	Species name	Group affiliation	Main hosts
Almond witches broom = (Almond brooming)	AlmWB	<i>Candidatus</i> Phytoplasma phoenicium	Pigeon pea witches broom (PPWB)	Almond, possibly other <i>Prunus</i> species
Cherry lethal yellows	CLY	<i>Ca.</i> Phytoplasma ulmi	Elm yellows (EY)	Cherry
European stone fruit yellows = (apricot chlorotic leafroll, plum leptonecrosis, plum decline, peach yellows, peach decline, European peach yellows)	ESFY	<i>Ca.</i> Phytoplasma prunorum	AP	Almond, apricot, cherry, European plum, Japanese plum, peach
Peach red suture (related to peach yellows)		Undetermined	X-disease	Japanese plum, peach
Peach rosette		Undetermined	X-disease	Peach, Japanese plum
Peach yellow leafroll	PYLR	Undetermined	AP	Peach
Peach yellows = (Little peach)	PY	Undetermined	X-disease	Almond, apricot, peach, nectarine
X-Disease = (Cherry buckskin, Cherry Western X disease, Prunus X disease)	WX	<i>Ca.</i> Phytoplasma pruni	X-disease	Almond, cherry, French prune, Japanese plum, peach



