

Plant viruses and viroids released from the NIAS Genebank Project, Japan

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INTRODUCTION

The National Institute of Agrobiological Sciences (NIAS), Japan, is implementing the NIAS Genebank Project for the conservation and promotion of agrobiological genetic resources, which include plants, microorganisms, animals and DNA (excluding lumber and fisheries), to contribute to the development and utilization of agriculture and agricultural products (Nagai *et al.*, 2005, 2006; Sato *et al.*, 2004a, 2004b; Takeya *et al.*, 2011). The collection of microorganism genetic resources in the project mainly consists of fungi and bacteria in terms of number, but also includes plant viruses and viroids. Plant viruses are composed of DNA or RNA, normally wrapped in coats of proteins. Viroids, the molecules of which are less than one-tenth the size of the smallest viruses, are a few hundred nucleobases of highly complementary, circular, single-stranded RNA without the coats of proteins that are typical for viruses. Both plant viruses and viroids are pathogens causing plant diseases, and are economically important because many of them sometimes inflict catastrophic damages on the production of crops, vegetables, flowers and so on in agriculture. Since 1985, the project has also promoted research and preservation of plant viruses and viroids as agrobiological genetic resources. This is the largest such collection in Japan, and has established a reputation for originality and preciousness as well as scale (Anonymous, 2010, 2011). Here, we introduce plant viruses and viroids released from the NIAS Genebank Project.

CLASSIFICATION OF ISOLATES

Plant viruses and viroids in the NIAS Genebank Project have been classified based on their experimental characterization in comparison with information such as reports of the International Committee

on Taxonomy of Viruses (ICTV), a committee of the Virology Division of the International Union of Microbiology Societies (IUMS), with the task of developing, refining and maintaining a universal virus taxonomy (Carstens, 2010; Carstens & Ball, 2009; Fauquet *et al.*, 2005). As of February 2012, all of the isolates of plant viruses and viroids released from the project are from Japan, totaling 252 and 16, respectively, including ICTV-unassigned species of Butterbur mosaic virus, Amazon lily mild mottle virus, Citrus vein enation virus, Broad bean wilt virus, Tobacco necrosis virus and Citrus viroid original source (Table 1).

Five and two of the viral isolates are the double-stranded DNA virus of *Cauliflower mosaic virus* belonging to *Caulimovirus* of *Caulimoviridae* and the negative sense single-stranded RNA virus of *Tomato spotted wilt virus* belonging to *Tospovirus* of *Bunyaviridae*, respectively. The other 245 viral isolates are the positive sense single-stranded RNA virus, and are divided into 67 species belonging to 25 genera of at least 10 families as follows: *Alphaflexiviridae*, *Betaflexiviridae*, *Bromoviridae*, *Closteroviridae*, *Luteoviridae*, *Potyviridae*, *Secoviridae*, *Tombusviridae*, *Tymoviridae* and *Virgaviridae*. The genera of *Benyvirus* and *Sobemovirus* have not yet been categorized into families by ICTV. The viroid isolates, which are the unencapsidated single-stranded RNA viroid, are divided into 9 species belonging to 4 genera of *Pospiviroidae*.

The viral families with the largest number of genera are *Bromoviridae* (4 genera) and *Secoviridae* (4 genera). Those of species and isolates are *Potyviridae* (*Potyvirus* only/16 species/58 isolates) and *Virgaviridae* (3 genera/9 species/62 isolates), respectively. The viral genus with the largest number of species is *Potyvirus* of *Potyviridae*, and that of isolates is *Tobamovirus* (7 species/58 isolates) of *Virgaviridae*. The viral species with the largest

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Table 1 Classification of plant viruses and viroids released from the project (February 2012)

Viruses	NI	(continued)	NI
— Viruses —			
Double-stranded DNA		<i>Secoviridae</i>	
<i>Caulimoviridae</i>		<i>Comovirus</i>	
<i>Caulimovirus</i>		<i>Radish mosaic virus</i>	1
<i>Cauliflower mosaic virus</i>	5	<i>Squash mosaic virus</i>	1
Negative sense single-stranded RNA		<i>Fabavirus</i>	
<i>Bunyaviridae</i>		Broad bean wilt virus ^{UN}	1
<i>Tospovirus</i>		<i>Broad bean wilt virus 2</i>	6
<i>Tomato spotted wilt virus</i>	2	<i>Gentian mosaic virus</i>	1
Positive sense single-stranded RNA		<i>Nepovirus</i>	
<i>Alphaflexiviridae</i>		<i>Arabis mosaic virus</i>	2
<i>Potexvirus</i>		<i>Cycas necrotic stunt virus</i>	3
<i>Alstroemeria virus X</i>	1	<i>Mulberry ringspot virus</i>	2
<i>Asparagus virus 3</i>	1	<i>Tobacco ringspot virus</i>	1
<i>Narcissus mosaic virus</i>	1	<i>Tomato ringspot virus</i>	1
<i>Nerine virus X</i>	1	<i>Sadwavirus</i>	
<i>Potato virus X</i>	4	<i>Satsuma dwarf virus</i>	1
<i>Betaflexiviridae</i>		<i>Tombusviridae</i>	
<i>Capillovirus</i>		<i>Carmovirus</i>	
<i>Apple stem grooving virus</i>	1	<i>Carnation mottle virus</i>	1
<i>Carlavirus</i>		<i>Melon necrotic spot virus</i>	7
Butterbur mosaic virus ^{UN}	1	<i>Necrovirus</i>	
<i>Potato virus M</i>	1	<i>Olive latent virus 1</i>	2
<i>Potato virus S</i>	1	<i>Olive mild mosaic virus</i>	1
<i>Vitivirus</i>		Tobacco necrosis virus ^{UN}	1
<i>Grapevine virus A</i>	3	<i>Tombusvirus</i>	
<i>Grapevine virus B</i>	2	<i>Grapevine Algerian latent virus</i>	1
<i>Bromoviridae</i>		<i>Tymoviridae</i>	
<i>Alfavirus</i>		<i>Maculavirus</i>	
<i>Alfalfa mosaic virus</i>	3	<i>Grapevine fleck virus</i>	1
<i>Bromovirus</i>		<i>Virgaviridae</i>	
Amazon lily mild mottle virus ^{UN}	1	<i>Hordeivirus</i>	
<i>Cucumovirus</i>		<i>Barley stripe mosaic virus</i>	2
<i>Cucumber mosaic virus</i>	39	<i>Tobamovirus</i>	
<i>Peanut stunt virus</i>	3	<i>Cucumber green mottle mosaic virus</i>	7
<i>Tomato aspermy virus</i>	2	<i>Kyuri green mottle mosaic virus</i>	3
<i>Apple mosaic virus</i>	1	<i>Odontoglossum ringspot virus</i>	1
<i>Asparagus virus 2</i>	1	<i>Pepper mild mottle virus</i>	4
<i>Closteroviridae</i>		<i>Tobacco mosaic virus</i>	15
<i>Ampelovirus</i>		<i>Tomato mosaic virus</i>	27
<i>Grapevine leafroll-associated virus 3</i>	6	<i>Youcai mosaic virus</i>	1
<i>Closterovirus</i>		<i>Tobravirus</i>	
<i>Citrus tristeza virus</i>	6	<i>Tobacco rattle virus</i>	2
<i>Luteoviridae</i>		Families are undefined.	
<i>Polerovirus</i>		<i>Benyvirus</i>	
Citrus vein enation virus ^{UN}	2	<i>Beet necrotic yellow vein virus</i>	1
<i>Potyviridae</i>		<i>Sobemovirus</i>	
<i>Potyvirus</i>		<i>Cocksfoot mottle virus</i>	6
<i>Alstroemeria mosaic virus</i>	5	<i>Ryegrass mottle virus</i>	1
<i>Amazon lily mosaic virus</i>	1	<i>Southern bean mosaic virus</i>	2
<i>Bean common mosaic virus</i>	4	— Viroids —	
<i>Bean yellow mosaic virus</i>	2	Single-stranded RNA	
<i>Carnation vein mottle virus</i>	1	<i>Pospiviroidae</i>	
<i>Clover yellow vein virus</i>	4	<i>Apscaviroid</i>	
<i>Dasheen mosaic virus</i>	1	<i>Apple fruit crinkle viroid</i>	1
<i>Konjac mosaic virus</i>	2	<i>Apple scar skin viroid</i>	4
<i>Lettuce mosaic virus</i>	1	<i>Citrus bent leaf viroid</i>	1
<i>Peanut mottle virus</i>	1	<i>Citrus viroid III</i>	1
<i>Potato virus A</i>	1	Citrus viroid original source ^{UN}	1
<i>Potato virus Y</i>	5	<i>Cocadviroid</i>	
<i>Soybean mosaic virus</i>	8	<i>Citrus viroid IV</i>	1
<i>Turnip mosaic virus</i>	6	<i>Hostuviroid</i>	
<i>Watermelon mosaic virus</i>	5	<i>Hop stunt viroid</i>	1
<i>Zucchini yellow mosaic virus</i>	11	<i>Pospiviroid</i>	
		<i>Chrysanthemum stunt viroid</i>	5
		<i>Citrus exocortis viroid</i>	1

NI: Number of isolates. UN: ICTV-unassigned.

Table 2 List of plant viruses and viroids released from the project by isolation source plant family (February 2012)

— Viruses —	NI	(continued)	NI
Alstroemeriaceae		<i>Potyviridae</i>	
Positive sense single-stranded RNA		<i>Potyvirus</i>	
<i>Alphaflexiviridae</i>		<i>Lettuce mosaic virus</i>	1
<i>Potexvirus</i>		<i>Secoviridae</i>	
<i>Alstroemeria virus X</i>	1	<i>Nepovirus</i>	
<i>Bromoviridae</i>		<i>Arabis mosaic virus</i>	1
<i>Cucumovirus</i>			
<i>Cucumber mosaic virus</i>	4	Brassicaceae	
<i>Potyviridae</i>		Double-stranded DNA	
<i>Potyvirus</i>		<i>Caulimoviridae</i>	
<i>Alstroemeria mosaic virus</i>	5	<i>Caulimovirus</i>	
<i>Secoviridae</i>		<i>Cauliflower mosaic virus</i>	5
<i>Fabavirus</i>		Positive sense single-stranded RNA	
<i>Broad bean wilt virus 2</i>	1	<i>Bromoviridae</i>	
<i>Virgaviridae</i>		<i>Cucumovirus</i>	
<i>Tobamovirus</i>		<i>Cucumber mosaic virus</i>	2
<i>Youcai mosaic virus</i>	1	<i>Potyviridae</i>	
Amaryllidaceae		<i>Potyvirus</i>	
Positive sense single-stranded RNA		<i>Turnip mosaic virus</i>	6
<i>Alphaflexiviridae</i>		<i>Secoviridae</i>	
<i>Potexvirus</i>		<i>Comovirus</i>	
<i>Narcissus mosaic virus</i>	1	<i>Radish mosaic virus</i>	1
<i>Bromoviridae</i>		<i>Virgaviridae</i>	
<i>Bromovirus</i>		<i>Tobamovirus</i>	
Amazon lily mild mottle virus^{UN}	1	<i>Tobacco mosaic virus</i>	2
<i>Cucumovirus</i>			
<i>Cucumber mosaic virus</i>	2	Caryophyllaceae	
<i>Potyviridae</i>		Positive sense single-stranded RNA	
<i>Potyvirus</i>		<i>Potyviridae</i>	
<i>Amazon lily mosaic virus</i>	1	<i>Potyvirus</i>	
<i>Secoviridae</i>		<i>Carnation vein mottle virus</i>	1
<i>Nepovirus</i>		<i>Tombusviridae</i>	
<i>Arabis mosaic virus</i>	1	<i>Carmovirus</i>	
<i>Tomato ringspot virus</i>	1	<i>Carnation mottle virus</i>	1
<i>Virgaviridae</i>		<i>Bromoviridae</i>	
<i>Tobravirus</i>		<i>Cucumovirus</i>	
<i>Tobacco rattle virus</i>	1	<i>Cucumber mosaic virus</i>	1
Apiaceae		Undefined	
Positive sense single-stranded RNA		<i>Benyvirus</i>	
<i>Secoviridae</i>		<i>Beet necrotic yellow vein virus</i>	1
<i>Fabavirus</i>			
Broad bean wilt virus^{UN}	1	Cucurbitaceae	
Araceae		Positive sense single-stranded RNA	
Positive sense single-stranded RNA		<i>Bromoviridae</i>	
<i>Potyviridae</i>		<i>Cucumovirus</i>	
<i>Potyvirus</i>		<i>Cucumber mosaic virus</i>	2
<i>Dasheen mosaic virus</i>	1	<i>Potyviridae</i>	
<i>Konjac mosaic virus</i>	2	<i>Potyvirus</i>	
Asteraceae		<i>Watermelon mosaic virus</i>	4
Negative sense single-stranded RNA		<i>Zucchini yellow mosaic virus</i>	11
<i>Bunyaviridae</i>		<i>Secoviridae</i>	
<i>Tospovirus</i>		<i>Comovirus</i>	
<i>Tomato spotted wilt virus</i>	1	<i>Squash mosaic virus</i>	1
Positive sense single-stranded RNA		<i>Tombusviridae</i>	
<i>Betaflexiviridae</i>		<i>Carmovirus</i>	
<i>Carlavirus</i>		<i>Melon necrotic spot virus</i>	7
Butterbur mosaic virus^{UN}	1	<i>Virgaviridae</i>	
<i>Bromoviridae</i>		<i>Tobamovirus</i>	
<i>Cucumovirus</i>		<i>Cucumber green mottle mosaic virus</i>	7
<i>Cucumber mosaic virus</i>	2	<i>Kyuri green mottle mosaic virus</i>	3
<i>Tomato aspermy virus</i>	1	Cycadaceae	
		Positive sense single-stranded RNA	
		<i>Secoviridae</i>	
		<i>Nepovirus</i>	
		<i>Cycas necrotic stunt virus</i>	1

NI: Number of isolates. UN: ICTV-unassigned.

Table 2 Continued

		(continued)	
Fabaceae	NI	Plumbaginaceae	NI
Positive sense single-stranded RNA		Positive sense single-stranded RNA	
<i>Bromoviridae</i>		<i>Potyviridae</i>	
<i>Alfavirus</i>		<i>Potyvirus</i>	
<i>Alfalfa mosaic virus</i>	3	<i>Clover yellow vein virus</i>	1
<i>Cucumovirus</i>			
<i>Cucumber mosaic virus</i>	10		
<i>Peanut stunt virus</i>	3	Poaceae	
<i>Potyviridae</i>		Positive sense single-stranded RNA	
<i>Potyvirus</i>		<i>Virgaviridae</i>	
<i>Bean common mosaic virus</i>	4	<i>Hordeivirus</i>	
<i>Bean yellow mosaic virus</i>	1	<i>Barley stripe mosaic virus</i>	2
<i>Clover yellow vein virus</i>	3	Undefined	
<i>Peanut mottle virus</i>	1	<i>Sobemovirus</i>	
<i>Soybean mosaic virus</i>	8	<i>Cocksfoot mottle virus</i>	6
<i>Watermelon mosaic virus</i>	1	<i>Ryegrass mottle virus</i>	1
<i>Secoviridae</i>			
<i>Fabavirus</i>		Ranunculaceae	
<i>Broad bean wilt virus 2</i>	1	Positive sense single-stranded RNA	
<i>Nepovirus</i>		<i>Bromoviridae</i>	
<i>Cycas necrotic stunt virus</i>	1	<i>Cucumovirus</i>	
Undefined		<i>Cucumber mosaic virus</i>	4
<i>Sobemovirus</i>			
<i>Southern bean mosaic virus</i>	2	Rosaceae	
Gentianaceae		Positive sense single-stranded RNA	
Positive sense single-stranded RNA		<i>Betaflexiviridae</i>	
<i>Secoviridae</i>		<i>Capillovirus</i>	
<i>Fabavirus</i>		<i>Apple stem grooving virus</i>	1
<i>Broad bean wilt virus 2</i>	1	<i>Bromoviridae</i>	
<i>Gentian mosaic virus</i>	1	<i>Iilarvirus</i>	
Iridaceae		<i>Apple mosaic virus</i>	1
Positive sense single-stranded RNA		<i>Tombusviridae</i>	
<i>Bromoviridae</i>		<i>Necrovirus</i>	
<i>Cucumovirus</i>		<i>Olive latent virus 1</i>	1
<i>Cucumber mosaic virus</i>	2	<i>Olive mild mosaic virus</i>	1
<i>Potyviridae</i>		<i>Closteroviridae</i>	
<i>Potyvirus</i>		<i>Closterovirus</i>	
<i>Bean yellow mosaic virus</i>	1	<i>Citrus tristeza virus</i>	6
<i>Secoviridae</i>		<i>Luteoviridae</i>	
<i>Nepovirus</i>		<i>Polerovirus</i>	
<i>Cycas necrotic stunt virus</i>	1	Citrus vein enation virus^{UN}	2
<i>Tobacco ringspot virus</i>	1	<i>Secoviridae</i>	
Liliaceae		<i>Sadwavirus</i>	
Positive sense single-stranded RNA		<i>Satsuma dwarf virus</i>	1
<i>Alphaflexiviridae</i>			
<i>Potexvirus</i>		Solanaceae	
<i>Asparagus virus 3</i>	1	Negative sense single-stranded RNA	
<i>Nerine virus X</i>	1	<i>Bunyaviridae</i>	
<i>Bromoviridae</i>		<i>Tospovirus</i>	
<i>Iilarvirus</i>		<i>Tomato spotted wilt virus</i>	1
<i>Asparagus virus 2</i>	1	Positive sense single-stranded RNA	
<i>Tombusviridae</i>		<i>Alphaflexiviridae</i>	
<i>Necrovirus</i>		<i>Potexvirus</i>	
Tobacco necrosis virus^{UN}	1	<i>Potato virus X</i>	4
Moraceae		<i>Betaflexiviridae</i>	
Positive sense single-stranded RNA		<i>Carlavirus</i>	
<i>Secoviridae</i>		<i>Potato virus M</i>	1
<i>Nepovirus</i>		<i>Potato virus S</i>	1
<i>Mulberry ringspot virus</i>	2	<i>Bromoviridae</i>	
Orchidaceae		<i>Cucumovirus</i>	
Positive sense single-stranded RNA		<i>Cucumber mosaic virus</i>	10
<i>Virgaviridae</i>		<i>Tomato aspermy virus</i>	1
<i>Tobamovirus</i>		<i>Potyviridae</i>	
<i>Odontoglossum ringspot virus</i>	1	<i>Potyvirus</i>	
		<i>Potato virus A</i>	1
		<i>Potato virus Y</i>	5
		<i>Secoviridae</i>	
		<i>Fabavirus</i>	
		<i>Broad bean wilt virus 2</i>	3

Table 2 Continued

<i>Tombusviridae</i>	NI	— Viroids —	NI
<i>Necrovirus</i>		Asteraceae	
<i>Olive latent virus 1</i>	1	Single-stranded RNA	
<i>Tombusvirus</i>		<i>Pospiviroidae</i>	
<i>Grapevine Algerian latent virus</i>	1	<i>Pospiviroid</i>	
<i>Virgaviridae</i>		<i>Chrysanthemum stunt viroid</i>	5
<i>Tobamovirus</i>			
<i>Pepper mild mottle virus</i>	4	Rosaceae	
<i>Tobacco mosaic virus</i>	9	Single-stranded RNA	
<i>Tomato mosaic virus</i>	27	<i>Pospiviroidae</i>	
<i>Tobravirus</i>		<i>Apscaviroid</i>	
<i>Tobacco rattle virus</i>	1	<i>Apple fruit crinkle viroid</i>	1
		<i>Apple scar skin viroid</i>	4
Vitaceae			
Positive sense single-stranded RNA		Rutaceae	
<i>Betaflexiviridae</i>		Single-stranded RNA	
<i>Vitivirus</i>		<i>Pospiviroidae</i>	
<i>Grapevine virus A</i>	3	<i>Apscaviroid</i>	
<i>Grapevine virus B</i>	2	<i>Citrus bent leaf viroid</i>	1
<i>Closteroviridae</i>		<i>Citrus viroid III</i>	1
<i>Ampelovirus</i>		Citrus viroid original source^{UN}	1
<i>Grapevine leafroll-associated virus 3</i>	6	<i>Cocadviroid</i>	
<i>Tymoviridae</i>		<i>Citrus viroid IV</i>	1
<i>Maculavirus</i>		<i>Hostuviroid</i>	
<i>Grapevine fleck virus</i>	1	<i>Hop stunt viroid</i>	1
		<i>Pospiviroid</i>	
Unclear		<i>Citrus exocortis viroid</i>	1
Positive sense single-stranded RNA			
<i>Virgaviridae</i>			
<i>Tobamovirus</i>			
<i>Tobacco mosaic virus</i>	4		

number of isolates is *Cucumber mosaic virus* (39 isolates) belonging to *Cucumovirus* of *Bromoviridae*. The viroid genus with the largest number of either species or isolates is *Apscaviroid* (5 species/8 isolates).

Although isolation sources of 4 isolates of *Tobacco mosaic virus* are unclear, the other respective viral isolates are from the plant family of Alstroemeriaceae, Amaryllidaceae, Apiaceae, Araceae, Asteraceae, Brassicaceae, Caryophyllaceae, Cucurbitaceae, Cycadaceae, Fabaceae, Gentianaceae, Iridaceae, Liliaceae, Moraceae, Orchidaceae, Plumbaginaceae, Poaceae, Ranunculaceae, Rosaceae, Solanaceae or Vitaceae (Table 2). Among the viral isolates, the isolates from Solanaceae are the largest and most varied by isolation source. The respective viroid isolates are from the plant family of Asteraceae, Rosaceae or Rutaceae. Among the viroid isolates, the isolates from Rutaceae are the largest and most varied by isolation source.

PROVISION OF ISOLATES WITH INFORMATION

As the NIAS historically has taken over the Genebank Project from the Ministry of Agriculture,

Forestry & Fisheries of Japan (MAFF), respective registration numbers of microorganism genetic resources in the project uniformly have the acronym, MAFF, e.g. MAFF104087. All of the viral and viroid isolates, which replicate only in compatible host plant cells, are being maintained by preserving host plant tissues infected with each isolate, according to the following two methods: 1) keeping in storage tanks controlled at -165°C with the vapor phase of liquid nitrogen, and 2) keeping in deep freezers at -80°C after vacuum drying of the tissues in glass ampules without freezing. For the respective isolates, detailed information such as hosts, collection localities, properties, providers to the project, relevant statutes and references as well as the MAFF accession numbers is freely web-accessible through search systems established on databases of the project (http://www.gene.affrc.go.jp/index_en.php), and the glass ampule preparations are usually forwarded to applicants (users) wishing to conduct research or education, based on the project's administrative regulations complying internal and international decisions related to genetic resources of plant viruses and viroids, e.g., the Plant Protection Act (Japan), the Foreign Exchange

& Foreign Trade Control Law (Japan), and the Convention on Biological Diversity (Takeya *et al.*, 2011; Tomioka, 2010; Tomioka *et al.*, 2009, 2010).

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