

## Occurrence and distribution of viral diseases on vanilla (*Vanilla planifolia* Andrews) in India

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Received 28 June 2004; Revised 12 August 2004; Accepted 08 September 2004

### Abstract

Sixty-five vanilla (*Vanilla planifolia*) plantations in 28 locations of Karnataka and Kerala were surveyed for the occurrence and distribution of viral diseases. Two viral diseases namely, mosaic and stem necrosis with an average incidence ranging from 0% to 5% and 0% to 10%, respectively, in various locations were noticed. The distribution of the diseases was random and not restricted to any particular region or locality. Electron microscopy of leaf dip preparations of the diseased plants showed three kinds of flexuous particles resembling *Potexvirus*, *Potyvirus* and *Closterovirus*, and an isometric particle.

**Key words:** survey, vanilla, *Vanilla planifolia*, viral diseases.

Vanilla (*Vanilla planifolia* Andrews), a native of Mexico, was introduced to India as early as 1835 (Sudharshan 2003). But large scale commercial cultivation of the crop in the country started in 1990s with the rise in prices due to increased demand for natural vanillin. The crop is now cultivated on a large scale in Karnataka, Kerala and Tamil Nadu in 2545 ha with a production of about 92 t (Anonymous 2003). However, diseases caused by fungi and viruses have become important constraints in vanilla production in the country. Though information on fungal diseases affecting vanilla is available (Thomas *et al.* 2003), little information is available on the viral diseases of the crop. Bhai *et al.* (2003)

reported the occurrence of a mosaic virus disease from Idukki District of Kerala based on surveys conducted in nine vanilla plantations in the District. Recently, the occurrence of mosaic and leaf curl diseases was reported in Chikmagalur, Hassan and Shimoga districts of Karnataka (Sudharshan *et al.* 2003). In this paper we report the occurrence, incidence and distribution of viral diseases in vanilla plantations at Idukki, Kozhikode and Wayanad districts of Kerala, and Dakshina Kannada, Udupi and Uttara Kannada districts of Karnataka.

Sixty-five vanilla plantations in 28 locations of Karnataka and Kerala were surveyed during August to November 2003 for the inci-

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dence and distribution of viral diseases. A standard pro forma was used for collection of data from the plantations which included, terrain, age of the crop, standard, soil type, cropping system, weed flora, agro-practices followed, etc. For recording the incidence of viral diseases, 100 vanilla vines selected randomly within each plantation were observed for mosaic and stem necrosis and their mean incidence for each location was determined. The diseased vines showing distinct symptoms were collected and maintained under insect proof conditions. Representative samples from these were then subjected to leaf dip electron microscopy at the Division of Plant Pathology, Indian Agricultural Research Institute, New Delhi. For this, the sap extracted from the diseased samples was placed on the carbon coated grid, negatively stained with 2% uranyl acetate (pH 4.5) and examined under JEOL-100-CF-II transmission electron microscope.

The average incidence (calculated for each location) of mosaic disease ranged from 0% to 5% while that of stem necrosis from 0% to 10% (Table 1). Maximum incidence of mosaic disease (5%) was recorded at Adimali (Idukki) while that of stem necrosis disease (10%) was observed at Nellipoyil (Kozhikode). Ankehalli (Hassan), Gadikai (Uttara Kannada), Arakkulam, Myaladampura (Idukki) and Muttill (Wyanad) were free from both mosaic and stem necrosis diseases. In Adyanadka, Peeramogaru (Dakshina Kannada), Idu (Udupi), Adimali, Ayyappancoil, Vandiperiyar (Idukki), Kodenchery, Nellipoyil and Thiruvambadi (Kozhikode) both mosaic and stem necrosis diseases were observed. Mosaic disease alone was recorded in Sediypapu (Dakshina Kannada), Neernahalli (Uttara Kannada), Kudavathoor (Idukki) and Koovapoyil (Kozhikode). Stem necrosis alone was recorded from Shibara (Dakshina Kannada), Koppa, Kumbrakottige, Malenahalli (Uttara Kannada), Muttom, Santhapara (Idukki), Madakkimala, Panyaram, Raroth and Vythiri (Wayanad). Both the diseases were noticed in all ages of

the crop. Arecanut, cashew, *Gliricidia sp.*, *Erythrina sp.* and unidentified forest trees were the live standards used in the areas surveyed and vanilla was grown under natural shade in these plantations. Two plantations (out of 65 plantations surveyed) were established with artificial shade using nylon net and granite pillars as standards. Both mosaic and stem necrosis diseases were found in vanilla irrespective of the type of standards used. The survey clearly indicated that the distribution of both the diseases was random in the plantations surveyed and not restricted to any particular region or locality. This indicates that the disease spread is mainly through the planting material.

The symptoms of mosaic disease were more prominent on younger leaves. Mosaic was the common symptom observed in most of the plantations surveyed. Various kinds of mosaic such as mild mottle, mild mosaic and mild chlorotic mottle streak (seen when the leaf is held against light) were observed. In a few cases such mosaic was also associated with leaf distortion and wavy margins. The size of the leaves was reduced, and in advanced stages, became brittle and severely crinkled with twisting of leaf lamina and growing point of stem region (Fig. 1a). In a few other cases, mosaic disease consisted of chlorotic to yellow green patches with distorted leaf margins. Stem necrosis disease was characterized by the presence of brown necrotic patches of varying length (1 mm up to 15 cm) on the stem (Fig. 1b). The necrotic region was very dry and brittle with a shriveled appearance and gave a cracking sound when broken. When the diseased stem region was cut open, an array of dead tissues were seen. In advanced stages, necrosis led to death of tissues at the affected region and thus affecting transportation of water and other nutrients to rest of the area eventually leading to death of the vine.

Electron microscopy of leaf dip preparations of the diseased plants showed four kinds of particles. Three kinds of flexuous particles resembled *Potexvirus* (particle length of about 500 nm), *Potyvirus* (particle length of about

Table 1. Distribution and incidence of viral diseases on vanilla in Karnataka and Kerala

State/District	Location Village	No. of gardens surveyed	Disease noticed	Mean incidence (%)
<b>Karnataka</b>				
Dakshina Kannada	Adyanadka	3	Mosaic	0.50
			Necrosis	0.75
	Peeramogaru	2	Mosaic	0.15
			Necrosis	0.25
Sediyapu	2	Mosaic	0.40	
		Necrosis	0	
Shibara	1	Mosaic	0	
		Necrosis	1.00	
Hassan	Ankehalli	1	Mosaic	0
Udupi	Idu	3	Necrosis	0
			Mosaic	3.00
Uttara Kannada	Gadikai	2	Mosaic	2.00
			Necrosis	0
	Koppa	1	Mosaic	0
			Necrosis	2.00
	Kumbarakottige	1	Mosaic	0
			Necrosis	1.00
	Malenahalli	2	Mosaic	0
			Necrosis	1.00
Neernahalli	2	Mosaic	1.00	
		Necrosis	0	
<b>Kerala</b>				
Idukki	Adimali	1	Mosaic	5.00
			Necrosis	1.00
	Arakkulam	2	Mosaic	0
			Necrosis	0
	Ayyappancoil	6	Mosaic	1.20
			Necrosis	1.40
	Kudavathoor	2	Mosaic	2.00
			Necrosis	0
	Muttom	1	Mosaic	0
			Necrosis	4
	Myladumpara	1	Mosaic	0
			Necrosis	0
	Santhapara	1	Mosaic	0
			Necrosis	4.00
Vandiperiyar	1	Mosaic	3.00	
		Necrosis	4.00	
Kozhikode	Kodenchery	3	Mosaic	0.40
			Necrosis	2.70
	Koovapoyil	1	Mosaic	2.00
			Necrosis	0
	Nellipoyil	11	Mosaic	0.60
Necrosis			10.00	
Thiruvambadi	6	Mosaic	2.20	
		Necrosis	1.00	
Wayanad	Madakkimala	2	Mosaic	0
			Necrosis	3.00
	Muttill	1	Mosaic	0
			Necrosis	0
	Panyaram	3	Mosaic	0
			Necrosis	2.00
	Raroth	1	Mosaic	0
			Necrosis	1.00
Vythiri	2	Mosaic	0	
		Necrosis	1.00	

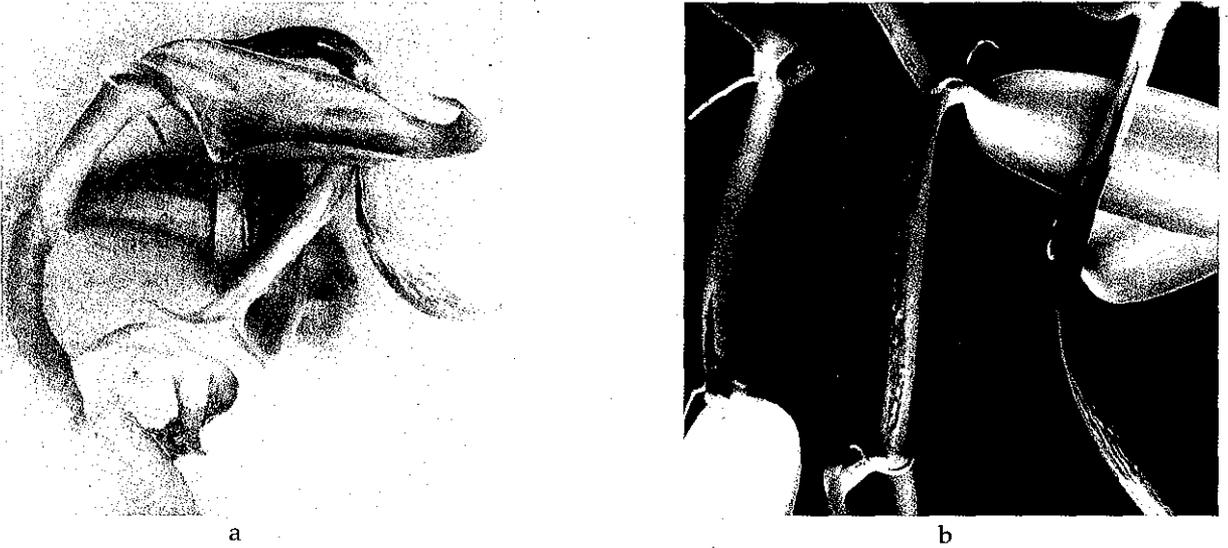


Fig. 1. Symptoms of viral diseases on vanilla (a) mosaic affected plant showing mosaic, severe crinkling, twisting of leaves and shoot (b) stem necrosis affected plant showing necrosis of varying length at the stem region.

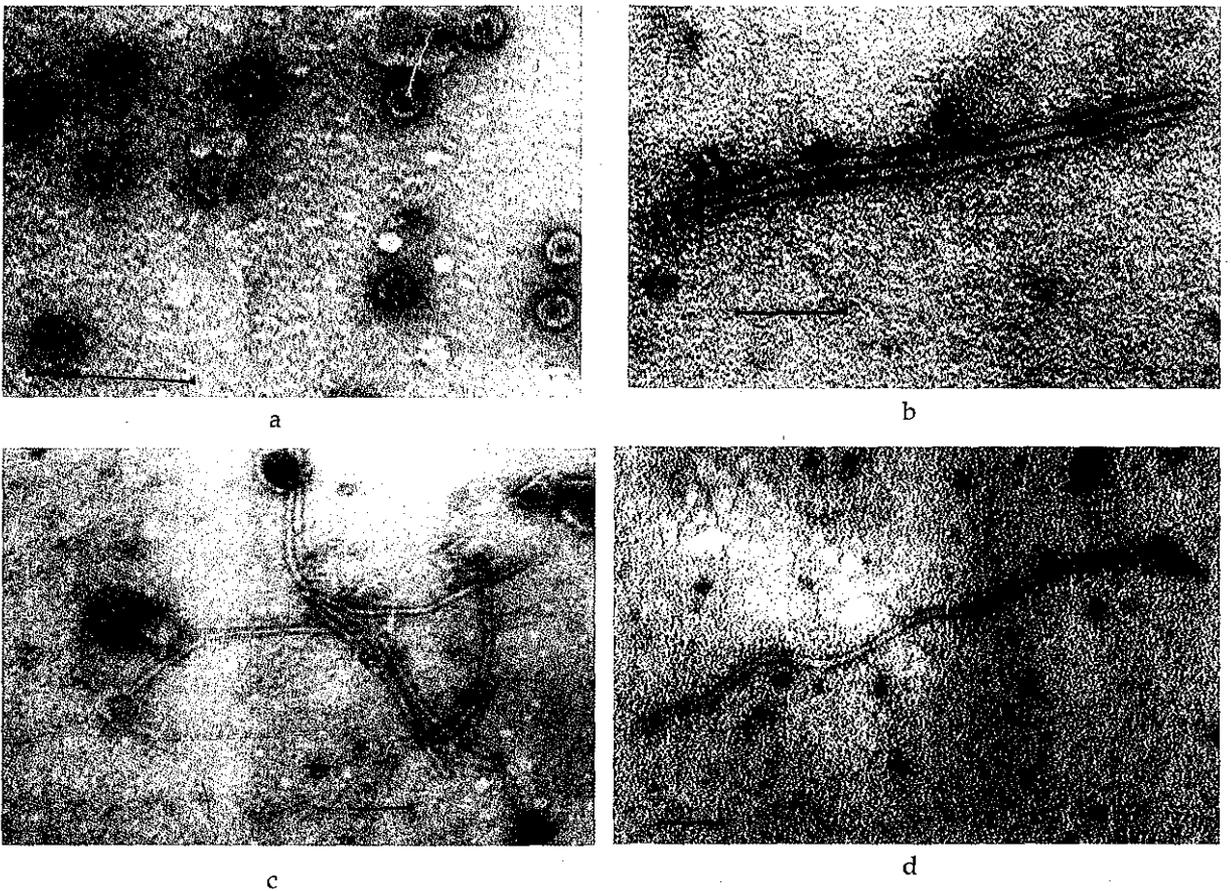


Fig. 2. Electron micrograph of virus particles from diseased vanilla vines (a) isometric particles (b) flexuous *Potexvirus* like particles (c) flexuous *Potyvirus* like particles and (d) flexuous *Closterovirus* like particle (bar represents 100 nm).

660 nm) and *Closterovirus* (particle length of about 1000 nm); and fourth was an isometric particle (about 28 nm) (Fig. 2). These particles were seen independently and also in combination in the diseased samples, thus making it difficult to assign a particular virus for a particular kind of symptom. All the four kinds of particles were seen in mosaic affected leaves, while the stem necrosis affected stem region predominantly showed flexuous particles belonging to *Potyvirus* genus.

Viruses are known to cause highly significant yield reduction of vanilla in many vanilla growing countries of the world (Pearson *et al.* 1991). Five different viruses belonging to three genera have been reported on vanilla from different parts of the world namely, *Cucumber mosaic virus* (CMV) (Wisler *et al.* 1987; Farreyrol *et al.* 2001), *Cymbidium mosaic virus* (CyMV) (Wisler *et al.* 1987; Benzet *et al.* 2000), *Odontoglossum ringspot virus* (ORSV) (Pearson *et al.* 1991; Wisler *et al.* 1987; Pearson *et al.* 1993), *Vanilla mosaic virus* (VMV) (Wisler *et al.* 1987; Zettler & Wisler 1990) and *Vanilla necrosis virus* (VNV) (Pearson & Pone 1988; Wang *et al.* 1993). Besides, two uncharacterized viruses belonging to *Potyvirus* and *Rhabdovirus* genera were reported (Pearson *et al.* 1991). While this study reports similar kinds of viruses with vanilla in India, presence of a *Closterovirus* like particle would be the first report but needs further confirmation. In India, viral disease problems have been noticed only recently and our study clearly indicates that these diseases are in their initial stages and large scale spread has not occurred. As vanilla is propagated through vegetative means, use of infected planting material (cutting) will lead to the spread of the disease. Hence it is essential to identify, characterize and develop rapid diagnostics against viruses for use in indexing and certification of planting material to check further spread of the viruses. Meanwhile empirical measures to destroy the infected plants must be enforced to control further spread of the viruses.

#### Acknowledgements

We thank the officials of Spices Board, Kochi,

for their help in the survey and Scientist in Charge, Virology Unit, Division of Plant Pathology, Indian Agricultural Research Institute, New Delhi, for electron microscopy work.

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