IDENTIFICATION OF VIRUSES FROM PEANUT IN INDIA

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Abstract

Field surveys were conducted during the period 1977-1979 in the major peanut growing areas of India. Subsequently, six naturally occurring viruses on peanut were identified on the basis of symptomatology, host range, transmission, serological affinities, electron microscopy and a few other properties.

Bud necrosis disease of peanut was shown to be caused by tomato spotted wilt virus (TSWV). Thirty species in 7 plant families were susceptible to the virus. The virus was transmitted by thrips (*Scirtothrips dorsalis*). The haemagglutination test was adopted to detect the virus in crude extracts of peanut. Sap from infected plants contained spherical membrane bound particles 70-90 nm in diameter. The prevalence of TSWV in India and high incidence in peanut indicated that the virus is economically important.

A disease characterized by severely stunted plants with small dark green leaves was found in peanut, and it occurred in patches in the field. Seeds sown in soil collected from infected fields produced plants with typical disease symptoms. The disease was shown to be caused by a rod-shaped virus 24 nm in diameter with predominant particle lengths of 249 and 184 nm. The virus, named Indian peanut clump virus (IPCV), resembled peanut clump virus (PCV) reported from West Africa in symptomatology on peanut, particle morphology and soil-borne nature. However, it is not serologically related to the West African PCV isolate.

A virus causing mottle and interveinal depression of the peanut leaves was isolated. Sixteen species in 3 plant families were found to be susceptible to the virus. The virus was transmitted by 3 species of aphids in a nonpersistent manner and through seed of peanut. Purified virus preparations contained flexuous elongated rods about 750 nm in length. Thin sections of infected leaves showed cylindrical inclusions in the cytoplasm. The virus was serologically related to 2 isolates of peanut mottle virus (PMV) from U.S. The virus was identified as PMV on the basis of symptomatology, serological affinities and some other properties.

A disease characterized by mosaic on peanut leaves was observed. The causal virus was mechanically transmissible to 14 species in 5 plant families. It was transmitted by 2 species of aphid in a nonpersistent manner, but not through peanut seeds. The virus particles consisted of flexuous rods about 750 nm in length. Thin sections of infected peanut leaves showed cylindrical inclusions in the cytoplasm. No serological relationship was detected between this virus and 5 other potyviruses including PMV. The virus was identified as a new potyvirus, named peanut green mosaic virus.

Peanut plants showing stunting with necrosis of the leaves were observed, and a virus was isolated from the diseased plants. Twelve species in 3 plant families were found to be susceptible to the virus, and the virus was not transmitted by aphids and through peanut seeds. The virus particles consisted of slightly flexuous rods about

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610 nm in length. The virus was serologically related to cowpea mild mottle virus (CMMV) obtained from West Africa, and it was identified as a strain of CMMV.

A virus was isolated from diseased peanut plants showing severe stunting with chlorotic streaks on the leaves. The virus infected 32 species in 8 families. The virus particles were isometric and measured 45–50 nm in diameter. In thin sections, infected leaves contained spherical intracellular inclusions, with many virus-like particles embedded in a dense matrix. The virus particles and intracellular inclusions were similar to those induced by caulimoviruses. The virus seems to be a newly recognized one and the name of peanut chlorotic leaf streak virus was proposed.

1. Introduction

In India, the earliest record of a virus disease on peanut was peanut clump reported by Sundaraman (39) in 1927. The majority of the reports appeared from 1963 and onwards. Nariani and Dhingra (26) described a mosaic disease, and Bisht *et al.* (2) reported groundnut rosette. Sharma (37) reported three virus diseases of peanut, namely bunchy top, chlorosis and ring mottle. A bud necrosis or bud blight disease was reported by Reddy *et al.* (35) and by Chohan (5). A ring mosaic disease has been reported by Narayanasamy *et al.* (25). A sap transmissible peanut chlorotic spot virus was reported by Haragopal and Nayudu (13). All these diseases or viruses were considered to be new by each of the authors, although some of them were similar in symptomatology and transmission. Causal viruses have not been precisely characterized and their possible affinities with peanut viruses occurring in other countries have not been investigated. The majority of them were not transmitted by mechanical sap inoculations.

Since 1976, virus diseases of peanut occurring in India have been investigated in ICRISAT. Several papers have already been published from ICRISAT on the occurrence of various peanut virus diseases (12,16,17,29,30,32,33,38).

This paper describes the results of investigations which one of the authors was able to conduct during 1977–1979, as visiting scientist in Groundnut Pathology, at the International Crops Research Institute for Semi-Arid Tropics (ICRISAT). This visit to ICRISAT was sponsored by the Tropical Agriculture Research Center in Japan.

2. Materials and methods

Several peanut plants showing characteristic virus disease symptoms were collected during survey trips to different peanut growing areas in India. The lesions were propagated either by grafting or by mechanical sap inoculation, onto healthy peanut plants in a screenhouse and the plants were sprayed with insecticides at weekly intervals.

Mechanical inoculations were performed on Carborundum-dusted leaves using a cotton pad dipped in sap extracts prepared in 0.05 M phosphate buffer, pH 7.0.

For host range studies, five or more plants of each species or cultivar were inoculated mechanically. Plants were observed for at least one month for symptom development, then they were back-inoculated onto assay plants to determine the presence of the virus.

In aphid transmission tests *Aphis craccivora, A. gossypii* and *Myzus persicae* reared on healthy peanut, cotton and cabbage respectively, were used. Aphids were starved for 1 h and allowed an acquisition access period of 2–3 min on infected leaves. Later

they were transferred to healthy plants and allowed an inoculation access period of 1 day on host.

Seed transmission tests were carried out on seeds collected from diseased plants. Thereafter the seeds were sown in a screenhouse in sterile soil. Seedlings in which virus infection was suspected were tested by infectivity assays on diagnostic hosts.

Stability of the virus in crude sap was determined on crude infected leaf extracts diluted to 1:10 in 0.05 M phosphate buffer, pH 7.0. To determine the thermal inactivation point 1 ml of sap was heated in a water bath for 10 min at various temperatures.

Five different peanut viruses were clarified employing organic solvent treatments. With the exception of one virus, polyethylene glycol (PEG) precipitation was used for precipitating the viruses. Subsequently the viruses were purified by concentrating in sucrose density gradients. The type of buffer required, sucrose concentrations used and the interval required for centrifugation were determined for each virus. The virus zones, following gradient centrifugation in sucrose solutions were drawn by using a bent needle.

Ring interface, agar gel diffusion and haemagglutination techniques were used for determining the serological relationships. In the ring interface and agar gel diffusion tests, 0.01 M phosphate buffer (pH 7.0) containing 0.85% NaCl was used for diluting both the antigen and the antiserum. In agar gel diffusion tests, 0.8% agar was prepared in 0.01 M phosphate buffer (pH 7.0) containing 0.85% NaCl and 0.05% sodium azide. For testing elongated viruses, 0.5% 3,5 diiodosalicylic acid (lithium salt) was incorporated into agar. Procedure for the haemagglutination test was similar to that described by Rajeahwari *et al.* (29).

For electron microscopy, the purified virus was negatively stained with 2% phosphotungstate, pH 7.2. For thin sectioning, small pieces of infected leaves were fixed for 2 h at 4°C in 3.5% glutaraldehyde, prepared in 0.01 M phosphate buffer (pH 7.0), and then post-fixed for 2h at 4°C in 1% osmium tetroxide prepared in 0.01 M phosphate buffer. The fixed smaples were dehydrated in graded acetone series and embedded in Epon 812. Sectioning was performed with an ultramicrotome using glass knives. Thin sections were stained in uranyl acetate and lead citrate, and observed under an electron microscope.

Details of some of the specific procedures, as applied to each individual virus, will be described in the results.

3. Results

1) Tomato spotted wilt virus

(1) Virus isolation

Extracts from young leaves of graft-infected plants were inoculated onto cowpea, and the virus isolated from a single local lesion was subsequently maintained in peanuts by mechanical sap inoculation.

(2) Host range and symptomatology

Thirty three plant species in 8 families were mechanically inoculated with the virus. Thirty plant species were susceptible to this virus (Table 1). Symptoms on selected hosts were as follows:

Arachis hypogaea cv. TMV 2. The leaf symptoms appeared 10 to 15 days after inoculation. Symptoms initially consisted of distinct chlorotic ringspots or chlorotic

Table 1. Host range of tomato spotted wilt virus (TSWV)

Plant species (cultivars)	Symptoms	
	Inoculated leaves	Upper leaves
Arachis hypogaea (TMV 2)	(cs)	N, M
Cajanus cajan	cs	0
Canavallia ensiformis	cs, ns	N, N
Crotolaria junecea	ns	0
Cyamopsis tetragonoloba	ns	M, N
Dolichos uniflorus	cs	M, N
Glycine max (Bragg)	ns	M, N
Phaseolus lunatus (Henderson Bush)	ns	M, N
P. vulgaris (Local)	ns	M, N
(Topcrop)	ns	M, N
(Bountiful)	ns	M, N
Pisum sativum (Bonnville)	ns	M, N
Vigna mungo	ns	N, M
V. radiata	ns	N, M
V. unguiculata (C-152)	cs, ns	(N, M)
(Early Ramshorn)	cs, ns	N, M
Datura stramonium	cs	M, N
Lycopersicon esculentum	cs	M, N
Nicotiana benthamiana	ns	N
N. clevelandii	ns	N
N. clevelandii × *N. glutinosa*	ns	N
N. glutinosa	ns	N, M
N. rustica	cs	0
N. tabacum (Xanthi nc)	cs	0
Petunia hydrida	ns	0
Physalis floridana	ns	N, M
Beta vulgaris	cs	0
Chenopodium amaranticolor	cs	0
C. quinoa	cs	0
Gomphrena globosa	ns	0
Cucumis sativus	cs	0
Vinca rosa	cs	M, N
Zinnia elegans	cs	M, N

Remarks: cs; chlorotic spot, ns; necrotic spot, M; mosaic, N; necrosis, 0; no symptom ( ); sometimes appeared.

Nonsusceptible plant species: *Brassica oleracea, B. rapa, Raphanus sativus.*

speckling on the quadrifoliate leaves immediately below the terminal bud. Sometimes concentric chlorotic ringspots, chlorotic ringspots with a few necrotic spots, or chlorotic leaf spots with green islands were seen on the leaves. The fully expanded quadrifoliate leaves immediately below the terminal bud often became flaccid. Later, necrosis was observed on the terminal bud which sometimes involved the stems and the petioles. New leaves were smaller than normal and showed a wide range of symptoms including distortion, mosaic mottling and general chlorosis. Internodes were reduced in length and short axillary shoots with distorted and mottled leaves developed. Infected plants were stunted and bushy, especially when the infection occurred in the seedling stage. The kernels from infected plants were shrivelled and the testae were discolored and mottled.

Vigna unguiculata cvs. C-152 and Early Ramshorn. The virus produced concentric chlorotic and necrotic lesions on the primary leaves of cowpea cv. C-152, 4 to 5 days

after inoculation. The new trifoliate leaves sometimes showed systemic infection as chlorotic and necrotic spots. Cowpea cv. Early Ramshorn showed chlorotic lesions on the inoculated leaves and the lesions later turned necrotic. Necrosis usually spread to newly produced leaves, sometimes leading to the death of the plant.

Lycopersicon esculentum cvs. Pusa Ruby and Perfection. On inoculated plants, yellow specks were observed followed by concentric necrotic rings and bronzing. With in 2-3 weeks, plants developed apical necrosis and leaf distortion.

Petunia hybrida. Necrotic local lesions were observed on inoculated leaves in 2 to 3 days without systemic infection.

Chenopodium quinoa. Local necrotic lesions without systemic infection were observed.

(3) Transmission

Adults of *Scritothrips dorsalis* collected from peanut fields with bud necrosis successfully transmitted the virus to 14 of 65 plants. Similarly, thrips larvae that had acquired the virus from infected leaves were allowed inoculation access of 12 to 15 days after becoming adults. Thirty one of 164 exposed peanut plants were infected.

Of nearly 6,000 seeds collected from the infected plants, 1,800 seeds produced normal plants, 540 seeds produced malformed plants and the remainder failed to germinate. None of the plants developed bud necrosis symptoms, and assays on cowpea from selected malformed and stunted plants gave negative results.

(4) Stability in crude sap

The infectivity dilution end point of the virus ranged between 10^{-2} and 10^{-3}, and the thermal inactivation point ranged between 45 and 50°C. Leaf extracts retained their infectivity for 4 h but not 5 h at 30°C.

(5) Selorogy

In two experiments, tomato spotted wilt virus (TSWV) antiserum received from Dr. Van Regenmortel gave haemagglutination titers of 1/3,200 with extracts from the infected leaves and of 1/200 to 1/400 with healthy leaf extracts. In three experiments, TSWV antiserum received from Dr. Gooding gave a titer of 1/20,480 to 1/32,000 with the infected leaf extracts and of 1/500 with healthy leaf extracts.

Cells sensitized with normal rabbit serum, peanut mottle virus antiserum and peanut stunt virus antiserum failed to react with any of the dilutions of extracts from healthy or infected leaves.

(6) Electron microscopy

Spherical menbrane-bound virus particles 70-90 nm in diameter were observed in the cytoplasm of infected peanut and tomato leaves. Some of the particles were present in the cisternae of the endoplasmic reticulum which often were present in membranous bags.

(7) Identification

Bud necrosis disease was shown to be caused by tomato spotted wilt virus on the basis of serological, and electron microscopic characteristics, thrips transmission, low thermal inactivation point, host range and symptoms.

Tomato spotted wilt virus had earlier been reported to occur in different strains (15). The strain carrying bud necrosis of peanut differed from the others in host range. For example, it failed to infect systemically *Nicotiana tabacum* and *N. rustica.*

2) Indian peanut clump virus

(1) Virus isolation

A peanut disease characterized by severe stunting of plants was observed in the Punjab State. The disease occurred in paths in the field and was restricted to crops raised in sandy and sandy loam soils. Peanut plants showing characteristic symptoms were collected during surveys in the Punjab State and virus cultures were established by graft-inoculation to healthy peanuts. Mechanical inoculations were performed from symptom-bearing leaves of peanut to *Chenopodium quinoa*. A single lesion isolate was obtained form *C. quinoa* and maintained hereafter in *Nicotiana clevelandii* × *N. glutinosa* (*N.* hybrid) by mechanical inoculation.

(2) Host range and symptoms

Thirty two plant species in 8 families were inoculated with the virus mechanically. Recovery assays were performed on *Phaseolus vulgaris* cv. Local. The results indicated that 17 plant species belonging to 3 families were infected by the virus (Table 2). Symptoms on selected hosts are given below:

Arachis hypogaea cv. TMV-2. Typical symptoms were observed on newly emerged 2-3 week old seedlings. Plants were stunted and the new quadrifoliate leaves showed mosaic mottling with chlorotic rings. Subsequently, the infected plants were conspicuous because of severe stunting and dark green leaves. Flowering occurred on infected plants but the pegs did not develop normal size pods. Later infected plants produced small pods. Roots were dark colored and the outer layers could be peeled off exposing a pink colored inner layer. With the exception of the root symptoms, all the

Table 2. Host range of Indian peanut clump virus (IPCV)

Plant species (cultivars)	Symptoms	
	Inoculated leaves	Upper leaves
Arachis hypogaea (TMV 2)	0	M
Canavallia ensiformis	ns	0
Cassia obtusifolia	nl	0
C. occidentalis	0	M
Crotalaria juncea	0	M
Cyamopsis tetragonoloba	cs	0
Phaseolus vulgaris (Local)	nl	N
(Topcrop)	vn	N
(Kintoki)	nl	N
(Bountiful)	nl	N
Vicia faba	nl	0
Vigna radiata	ns	0
V. unguiculata (C-152)	cs	0
(Early Ramshorn)	cs	0
Capsicum annuum	0	m
Nicotiana benthamiana	0	M
N. clevelandii	0	M
N. clevelandii × *N. glutinosa*	0	M
Petunia hybrida	0	M
Chenopodium amaranticolor	cs	0
C. quinoa	cs	(cs)

Remarks: cs; chlorotic spot, ns; necrotic spot, nl; necrotic lesion, vn; vein necrosis, M; mosaic, m; faint mottling, N; necrosis, 0; no symptom, ( ); sometimes appeared.

Nonsusceptible plants: *Cajanus cajan, Glycine max* (Bragg, Norin No.4, Norin No.2), *Phaseolus vulgaris* (Kentucky Wonder, Pito U.I. No.72, Idaho Refugee) *Vigna mungo, V. angularis* (Odate No.1), *Pisum sativum* (Bonneville), *Datura stramonium, Nicotiana glutinosa, N. rustica, N. tabacum* (White Burley), *Sesamum indica, Zinnia elegans, Cucumis sativus, Tetragonia expansa, Brassica oleracea, Raphanus sativus.*

typical symptoms of the disease were reproduced on peanuts infected with this virus mechanically.

Phaseolus vulgaris cvs. Local, Topcrop, Bountiful, Kintoki. The virus produced veinal necrosis on inoculated leaves a week after inoculation. Occasionally when the infection became systemic similar symptoms were observed.

Canavallia ensiformis. In 4 to 5 days after inoculation, the virus produced circular necrotic local lesions. The lesions tended to spread and were surrounded by a large yellow halo around the spot and the infection resulted in systemic mottle.

Vigna mungo. Within 2 to 3 days after inoculation the virus produced necrotic lesions and systemic infection was observed.

(3) Transmission

The virus was transmitted by mechanical inoculation, and not by aphid (*Aphis craccivora*). Limited tests on seed transmission indicated that the virus is not transmitted through peanut seeds.

Of the 160 peanut seeds from healthy plants sown in soil collected at depths of 10 to 25 cm, 72 produced seedlings with typical symptoms and in addition extracts from them produced typical symptoms on French bean cv. Local.

(4) Stability in crude sap

The infectivity in crude sap of peanut leaves was assayed on French bean cv. Local. The thermal inactivation point ranged between 60 and 65°C, and the dilution end point between 10^{-3} to 10^{-4}. The virus remained infective for over 20 days at room temperature (25–30°C).

(5) Purification

The virus was purified from *Nicotiana* hybrid (*N. clevelandii* × *N. glutinosa*). Infected leaves were homogenized in 0.1 M phosphate buffer, pH 8.0, containing 0.2% 2-mercaptoethanol, and the extract clarified with 10% chloroform. Virus was precipitated by adding 6% PEG and 0.2 M NaCl, resuspended in 0.01 M borate phosphate buffer, pH 8.3 and further purified by 2 cycles of density gradient centrifugation in sucrose solutions. Purified virus retained over 50% of the infectivity present in clarified extracts. Ultraviolet absorption spectrum of the purified virus was typical of that of a nucleoprotein, with minimum and maximum absorbances at 245 and 260 nm, respectively. The A 260/280 ratio varied from 1.21 to 1.24.

(6) Serology

One ml of 1 mg virus was emulsified with an equal volume of Freund's incomplete adjuvant and injected intramuscularly at weekly intervals into rabbits. Serum was collected one week after the fourth injection and titered by the precipitin ring test using purified virus (150 μg/ml). The titer was 1/600 as determined by the precipitin ring test. In the microprecipitin tests, none of the dilutions of this virus antiserum gave a positive reaction with healthy leaf extracts of *N.* hybrid but a titer of 1/64 was obtained with infected leaf extracts. Antisera to the two West African peanut clump virus isolates, tobacco rattle virus and pea early browning virus did not react with healthy or infected *N.* hybrid leaf extracts.

(7) Electron microscopy.

Infected peanut and *N.* hybrid leaves were cut into small pieces and fixed with 3% glutaraldehyde prepared in 0.1 M phosphate buffer, pH 7.3, for 1 h. Fixed tissues were rinsed in phosphate buffer and post-fixed in 2% osmium tetroxide for 3 h in a cold room. Dehydration was accomplished in a graded series of acetone and the pieces were embedded in Epon 812. In thin sections of infected tissues, masses of virus-like particles were observed arranged side by side in layers with the angle of their long

axis in adjacent layers alternating to give a "herring-bone" pattern.

Rod-shaped particles were observed at very low concentrations in leaf dip preparations of field-infected peanut and mechanically infected peanut, *N.* hybrid and *N. clevelandii.* Virus particles stained by uranyl acetate had a diameter of 24 nm and lengths of 249 and 184 nm. When stained by potassium phosphotungstate, the particles had a diameter of 20 nm and lengths of 239 nm and 169 nm. Several particles with lengths between 60 and 100 nm were observed but they were fewer in number after fixation with 1% glutaraldehyde suggesing that they were derived from the two main kinds of particles.

(8) Identification.

The virus resembled peanut clump virus (PCV) reported from West Africa by Thouvenel *et. al.* (40) in that both viruses which produced similar symptoms on peanut were soil-borne and both viruses resembled each other morphologically. However, in serological tests, West African PCV isolates and Indian PCV were unrelated to one another and to any of the tobraviruses tests. Furthermore, West African PCV and Indian PCV showed differences in host range; Indian PCV produced systemic veinal necrosis on *Phaseolus vulgaris* and PCV from West Africa showed mosaic symptoms. Unlike the Indian PCV, West African PCV failed to infect *Nicotiana tabacum* (41). The large number of broken particles present in unfixed samples, compared to PCV from West Africa might indicate a greater instability of the Indian virus. Since the virus from India was serologically distinct and differed in host range, it is referred to as Indian peanut clump virus. (IPCV).

3) Peanut Mottle Virus

(1) Virus isolation.

A virus disease characterized by mottling of the leaves and interveinal depression was observed in the Punjab, Maharashtra and Gujarat States. Several diseased plants were collected in farmer's fields in the Punjab State. Extracts from diseased leaves were mechanically inoculated onto peanut and the virus was subsequently maintained in a screenhouse in peanut (cv. TMV-2) and soybean (cv. Bragg) by mechanical inoculation.

(2) Host range and symptoms.

Thirty six species in 8 families were inoculated with the virus mechanically. All the infectivity assays were performed on French bean cv. Topcrop. The results indicated that 16 plant species in 3 families were susceptible to this virus (Table 3). Symptoms on diagnostic hosts are given below:

Arachis hypogaea cv. TMV-2. The newly formed leaves showed veinclearing 10 to 15 days after inoculation. Subsequently, the leaves showed mild mottling, upward curling of the leaf margins and interveinal depressions. As the leaf aged the symptoms became masked, but occasionally mottling consisting of dark green islands was visible. Though the symptoms are mild, the virus causes yield losses certainly.

Nearly 200 peanut germplasm lines and *Arachis* spp. were inoculated mechanically in a screenhouse. All of them were found to be susceptible to the virus. A few germplasm lines showed very mild symptoms and a low rate of infection, but none were immune to the virus. Four wild *Arachis* species were grafted with infected peanut cv. TMV 2. One month after grafting, recovery assays were performed on *Phaseolus vulgaris* cv. Topcrop. Results indicated that *A. correntia* and *A. glabrata* were not infected, whereas *A. chacoenses* and *A. stenocarpa* were infected systemically.

Table 3. Host range of peanut mottle virus (PMV)

Plant species (cultivars)	Symptoms	
	Inoculated leaves	Upper leaves
Arachis hypogaea	0	M
Cassia obtusifolia	ns	N
C. occidentalis	cs	M
Cajanus cajan	ns	N
Canavallia ensiformis	cs	M
Cyamopsis tetragonoloba	0	M
Glycine max (Bragg)	cs	M
Phaseolus lunatus (Henderson Bush)	0	Lat
P. vulgaris (Local)	cs	0
(Topcrop)	ns	0
(Kintoki)	cs	M, N
Pisum sativum (Bonneville)	cs	M
Vicia faba	0	M
Vigna anqularis (Odate No.1)	0	M
V. unguiculata (Early Ramshorn)	cs	m
Nicotiana benthamiana	0	m
N. clevelandii	0	m
Sesamum indica	cs	m

Remarks: cs; chlorotic spot, ns; necrotic spot, M; mosaic, m; faint mottling, Lat; latent infection, N; necrosis, 0; no symptom.

Nonsusceptible plant species: *Crotolaria juncea, Vigna mungo, V. sesquipedalis, Glycine max* (Norin No.2), *Datura stramonium, Lycopersicon esculentum, Nicotiana glutinosa, N. rustica, N. tabacum, Petunia hybrida, Physalis floridana, Beta vulgaris, Chenopodium amaranticolor, C. quinoa, Brassica oleracea, B. rapa, Raphanus sativus, Gomphrena globosa, Vinca rosea, Cucumis sativus, Zinnia elegans.*

Glycine max cv. Bragg. The newly unfolded leaves showed veinclearing 7 to 12 days after inoculation, followed by systemic mottling, crinkling and puckering.

Pisum sativum cv. Bonneville. Chlorotic spots and veinclearing were produced 7 to 10 days after inocualtion followed by distinct systemic chlorotic mottling.

Phaseolus vulgaris cv. Topcrop. Inoculated leaves developed reddish brown necrotic lesions 5 to 7 days after inoculation, which tended to spread along the veins and produce veinal necrosis. No systemic symptoms were observed. This cultivar is a good diagnosic and assay host.

Nicotiana benthamiana. Faint veinclearing and mottling were produced systemically 7 to 10 days after inoculation. Occasionally the upper leaves showed curling and interveinal depression.

(3) Transmission.

After short acquisition probes *Myzus persicae* and *Aphis craccivora* transmitted the virus from diseased peanut plants to healthy ones. Of the peanut seed collected from infected plants, 310 germinated and 4 plants were infected, giving a seed transmission frequency of 1.3%.

(4) Stability in crude sap.

The infectivity of this virus in crude sap of pea leaves was estimated by the number of local lesions produced on inoculated leaves of French bean cv. Topcrop. The thermal inactivation point of the virus ranged between 55 and 60°C, the dilution end point between 10^{-3} and 10^{-4}, and longevity in vitro (25°C) ranged between 2 and 3 days.

(5) Purification.

Infected pea leaves exhibiting systemic mosaic symptoms were homogenized in a blender with cold 0.1 M phosphate buffer, pH 8.0, containing 0.01 M sodium diethyldithiocarbamate (DIECA) and 0.2% 2-mercaptoethanol. The sap was filtered through a cheesecloth and shaken with 10% chloroform for 10 min. The emulsion was broken by centrifugation at 3,000g for 15 min. Polyethylene glycol (PEG) and NaCl were added to the clarified extract to reach final concentrations of 4% and 0.2 M, respectively. The mixture was incubated at 4 C for 90 min before centrifugation at 5,000g for 10 min. The pellets were resuspended in 0.01 M borte-phosphate buffer, pH 8.3, containing 0.2 M urea. Twenty-five ml of the virus preparation was then layered on a 13 ml column of 30% sucrose prepared in borate-phosphate buffer containing 4% PEG and 0.2 M NaCl. Following centrifugation at 24,000 rpm for 90 min in a Beckman SW 27 rotor, the pellets were resuspended in borate-phosphate buffer and subjected to rate-zonal density gradient centrifugation. The gradient columns were prepared by layering, respectively, 6, 9, 9 and 9 ml of 100, 200, 300 and 400 g/l of sucrose in borate-phosphate buffer. The virus preparation was layered on each gradient column and centrifuged at 24,000 rpm for 2 h in a SW 27 rotor. A diffuse light scattering zone at 6.4—7.0 cm and a single clear zone at 5.6—6.0 cm from the bottom of the tube were evident. Virus particles were detected in the lower zone.

The UV absorption spectrum of the purified virus had a shoulder at 290 nm. The A260/280 ratio was 1.20 and the A260/245 ratio 1.11.

(6) Serology.

Antiserum for this virus was prepared in rabbits by intramuscular and intravenous injections of purified virus. The titer was 1/128 as tested by ring interface test. The antiserum was compared by haemagglutination test with two samples of peanut mottle virus antisera obtained from Dr. Kuhn and Dr. Hebert. Extracts from infected peanut, soybean and pea plants, and purified virus preparations gave positive reactions with all the antisera. Extracts from healthy plants which were tested with all the antisera gave a negative reaction. The serological dilution end points of the virus as determined in haemagglutination test, from infected peanut, soybean and pea plants were 1/800, 1/1,600 and 1/6,400, respectively.

(7) Electron microscopy.

The purified virus preparations showed flexuous filamentous rod shaped virus particles with a modal length of 749± 10 nm. The width of the particles was 13±1 nm.

Ultrathin sections from infected leaves of peanut revealed the presence of inclusion bodies resembling pinwheels similar to those reported previously (6).

(8) Identification.

On the basis of symptoms on peanut, host range, transmission, serological affinity, morphology of the virus particles and cytoplasmic inclusions, the virus isolated from peanut in India was identified as peanut mottle virus (PMV) (22). PMV belongs to the potyvirus group and has been reported from almost all the peanut growing countries.

Several strains of PMV were reported on the basis of the symptoms. The virus in India resembled the mild strain of PMV reported by Paguio and Kuhn (28).

4) Peanut green mosaic virus

(1) Virus isolation

A disease characterized by mosaic on peanut leaves was observed in Andhra Pradesh State. The virus was first established in a screenhouse by graft-inoculation of

healthy peanut seedlings (cv. TMV 2). The virus was subsequently mechanically transmitted to peanut plants and to French bean cv. Local. The virus isolated from a single local lesion produced on French bean, was maintained in peanut plants.

(2) Host range and symptoms

Thirty nine species in 8 families were inoculated mechanically and recovery assays were performed on French bean cv. Local. The results indicated that 14 plant species in 5 families were susceptible to the virus (Tabel 4).

Symptoms on selected hosts are given below:

Arachis hypogaea cv. TMV 2. Mechanically inoculated peanut plants showed chlorotic spots and veinclearing on young, systemically infected quadrifoliate leaves 10 to 15 days after inoculation. Later the leaves showed severe mosaic appearing as dark green islands surrounded by chlorotic areas. As infected plants aged the symptoms on leaves became less obvious. The plants were stunted.

Phaseolus vulgaris. Chlorotic lesions developed on inoculated leaves of cvs. Kintoki, Bountiful and Local, 6 to 8 days after inoculation. The lesions became necrotic and some spread along the veins. In cv. Kintoki, systemic veinal necrosis was observed.

Petunia hybrida. Indistinct chlorotic spots appeared on the inoculated leaves 5 to 7 days after inoculation. In systemic infection, vein clearing was followed by a mild chlorotic mottling.

Nicotiana benthamiana. Inoculated plants showed pronounced mottling of the leaves followed by malformation and distortion of systemically infected leaves.

Chenopodium amaranticolor. The virus produced chlorotic lesions within 5-7 days after inoculation.

(3) Transmission

Aphis craccivora, A. gossypii and *Myzus persicae* colonies which were maintained on

Table 4. Host range of peanut green mosaic virus (PGMV)

Plant species (cultivars)	Symptoms	
	Inoculated leaves	Upper leaves
Arachis hypogaea (TMV 2)	0	M
Cassia occidentalis	0	M
Cyamopsis tetragonoloba	cs	vc
Phaseolus lunatus (Henderson Bush)	0	Lat
P. vulgaris (Local)	cs	0
(Kintoki)	nl	N
Vigna unguiculata (Early Ramshorn)	cs	0
Nicotiana benthamiana	0	m
N. clevelandii	0	m
N. clevelandii × *N. glutinosa*	0	vc
Petunia hybrida	0	m
Chenopodium amaranticolor	cs	0
C. quinoa	cs	0
Sesamum indica	0	m
Tetragonia expansa	cs	0

Remarks: cs; chlorotic spot, nl; necrotic lesion, vc; veinclearing, M; mosaic, m; faint mottling, Lat; latent infection, N; necrosis, 0; no symptom.

Nonsusceptible plant species: *Cajanus cajan, Canavallia ensiformis, Crotolaria juncea, Glycine max* (Bragg) *Phaseolus vulgaris* (Topcrop, Kentucky Wonder, Idaho Refugee, Pinto U. I. No. 72), *Pisum sativum* (Bonneville), *Vicia faba, Vigna angularis, V. mungo, V. radiata, V. unguiculata* (C-152), *Sesbania grandiflora, Capsicum annuum, Datura stramonium, Lycopersicon esculentum, Nicotiana glutinosa, N. rustica, N. tabacum* (Samsum NN), *Physalis floridana, Cucumis sativus, Brassica oleracea, B. rapa, Raphanus sativus, Beta vulgaris, Zinnia elegans.*

healthy pigeon pea, cotton and cauliflower plants, respectively, were starved for 1 h and then allowed to make a single acquisition probe of 30 sec to 3 min on detached young peanut leaves. At least 10 insects were then transferred to each healthy peanut test plant and after 14 h, killed by spraying with insecticides. All the inoculated plants showing symptoms were assayed on French bean. After a 30 sec acquisition access period both *A. gossypii* and *M. persicae* transmitted the virus to 26 of 50 peanut and 3 of 5 *N. benthamiana* plants. None of the 20 peanut plants and 3 *N. benthamiana* plants exposed to aphids from healthy colonies were infected. *A. craccivora* failed to transmit the virus.

Seeds from mechanically inoculated peanut (cv. TMV 2) with conspicuous symptoms were sown in sterile soil and the plants were maintained in a screenhouse for 45 days. All the three weeks old seedlings were tested for virus presence by inoculation onto French bean. Of the 480 seeds collected from infected plants, 460 germinated, none of them showed symptoms and no virus could be recovered.

(4) Stability in crude sap

The thermal inactivation point ranged between 55 and 60°C, the infectivity dilution end point between 10^{-3} and 10^{-4}, and the infectivity remained for 3 days at room temperature (25–30°C), but not for 4 days.

(5) Purification

The virus was purified from infected peanut leaves employing the procedure adapted for the purification of PMV.

A single light scattering zone about 2.5 cm from the bottom of the tube was observed following density gradient centrifugation. Samples from this zone diluted in 0.05 M phosphate buffer, pH 7.0, produced typical symptoms of the virus on pea and French bean. The ultraviolet absorption spectrum of the purified virus showed a shoulder at 290 nm, and the A260/280 ratio was 1.19. The A260/245 ratio was 1.26. Assuming an extinction coefficient of 3.0, nearly 1 mg of virus was obtained from 50 g of leaf.

(6) Serology

Antiserum against this virus was prepared in rabbits by intramuscular and intravenous injections of purified virus. The antiserum had a titer of 1/256 in ring interface test. In agar gel diffusion tests purified virus consistently gave a single precipitin line and undiluted antiserum did not react with healthy peanut leaf extracts. No positive reaction was observed when the purified virus was tested against the antisera of clover yellow vein, soybean mosaic, azuki bean mosaic, bean yellow mosaic and peanut mottle viruses.

In haemagglutination tests, extracts from healthy and PMV-infected peanut leaves gave no reaction but infected leaf extracts with this virus had a titer of 1/6,400 to 1/12,800. In addition, cells sensitized with PMV antiserum gave no reaction when tested with the virus infected leaf extracts and purified preparations.

(7) Electron microscopy

Purified virus preparations contained flexuous filamentous rods about 750 nm in length. Thin sections of infected peanut and *N. benthamiana* leaves contained cylindrical cytoplasmic inclusions.

(8) Identification

On the basis of the size and morphology of the virus particles, the presence of cylindrical inclusions in infected cells, aphid transmission and stability in crude sap, this virus was assigned to the potyvirus group. PMV was also shown to be a potyvirus occurring naturally in peanut (3, 14, 18, 22, 36). This virus differs from PMV in

infectivity to *Petunia hybrida, Chenopodium amaranticolor* and *C. quinoa* which PMV does not infect. The virus failed to infect French bean cv. Topcrop which is an important diagnostic host for PMV. In addition, this virus does not infect *Pisum sativum, Canavallia ensiformis, Glycine max, Phaseolus vulgaris* (cvs. Kentucky Wonder and Pinto U.I. No. 72) which are hosts of PMV. Turnip mosaic virus (TuMV) was also isolated from peanut plants (19). However the virus reported here unlike TuMV does not infect *Brassica oleracea, B. rapa, Raphanus sativus* and *Gomphrena globosa*. Recently, two additional potyviruses, groundnut eyespot virus (GEV) (8) and a virus causing peanut mild mottle disease (VPMM) (42) were isolated from naturally infected peanut. Host range of GEV differs markedly from that of this virus. GEV infects *Lycopersicon esculentum* and *Physalis floridana,* while the virus reported here does not infect them. The virus isolated in India also differed from VPMM in symptomatology on peanut and in host range. Recently, peanut mottle virus-ring type (10) has been reported on peanut and it seems to be similar to peanut stripe virus (7) which was also reported recently. Both viruses were serologically distinct from PMV and peanut stripe virus did not react with the virus reported here serologically.

Although more serological tests are needed to determine the relationship of the virus reported here with other potyviruses, it is considered to be distinct from previously described potyviruses. Consequently, the name of peanut green mosaic virus (PGMV) is proposed for this virus.

5) Cowpea mild mottle virus

(1) Virus isolation

A disease of peanut characterized by severe stunting, downward rolling and necrosis of leaves was observed in farmers' crops in the Maharashtra, Punjab, Uttar Pradesh, Tamil Nadu and Andhra Pradesh States. Although the disease was widely distributed, the incidence was usually below one percent. Diseased plants were established in a screenhouse by graft infection of healthy peanut cv. TMV 2. Then leaves with symptoms were extracted and mechanically inoculated to peanut and *Chenopodium quinoa* in which single lesion isolation was performed. A single lesion isolate was maintained in peanut plant, and used in this study as a virus source.

(2) Host range and symptoms

Twenty seven plant species in 6 families were inoculated with the virus mechanically. The recovery assays were performed on *Chenopodium quinoa.* The results indicated that 12 plnat species in 3 families were susceptible to the virus (Table 5). Symptoms on selected host plants are as follows:

Arachis hypogaea cv. TMV 2. On newly formed quadrifoliates, symptoms were observed 7 to 10 days after inoculation. Veinclearing, veinbanding and leaf rolling were observed in systemic infection. Severe initial symptoms were followed by veinal necrosis which extended along the petioles causing abscission of old leaves. Infection usually resulted in severe stunting.

Glycine max cv. Bragg. Necrotic lesions and veinal necrosis appeared on inoculated leaves. In systemic infection, severe mosaic and general leaf chlorosis, occasionally follwed by apical necrosis, were observed.

Vigna unguiculata cv. Early Ramshorn. Inoculated plants developed faint chlorotic blotches on inoculated leaves within 7-10 days, followed by systemic mottling or symptomless infection.

Chenopodium quinoa. The virus produced discrete chlorotic lesions on inoculated

leaves within 4-5 days after inoculation.

(3) Transmission

Aphis craccivora and *Myzus persicae* failed to transmit the virus in a non-persistent and persistent manner.

Infected peanut plants produced few seeds. However, over 90% of 153 peanut seeds and 510 soybean seeds germinated and none showed typical symptoms of the disease. Randomly selected seedlings gave negative recovery assays.

(4) Stability in crude sap

The thermal inactivation point ranged between 75°C and 80°C (10 min), and the dilution end point was between 10^{-3} and 10^{-4}. The virus remained infective at room temperature (25-30°C) for 8 days but not 10 days.

(5) Purification

The virus was purified from infected soybean leaves. Extracts prepared in phosphate buffer containing DIECA and 2-mercaptoethanol, were clarified with carbon tetrachloride. Virus in clarified extracts was concentrated by high speed centrifugation. Further purification was achieved by sucrose density gradient centrifugation. Ultraviolet absorption spectrum of the purified virus was typical of that of a nucleoprotein, with minimum and maximum absorbances at 246 nm and 260 nm, respectively.

(6) Serology

Antiserum for the virus had been prepared by intramuscular and intravenous injections of purified virus into a rabbit. The titer of the antiserum was 1/500 by ring interface and 1/8,000 by haemagglutination tests. Antiserum for cowpea mild mottle virus (CMMV) was obtained from Dr. Brunt, and haemagglutination technique was used to detect the virus antigen crude peanut and soybean extracts. Both extracts gave a positive reaction with CMMV antiserum.

Table 5. Host range of cowpea mild mottle virus (CMMV)

Plant species (cultivars)	Symptoms	
	Inoculated leaves	Upper leaves
Arachis hypogaea (TMV 2)	0	M (N)
Cajanus cajan	cs, ns	0
Canavallia ensiformis	cs (ns)	M
Cyamopsis tetragonoloba	ns	0
Glycine max (Bragg)	nl	M
(Norin No.4)	cs	M
Pisum sativum (Bonneville)	0	vc
Vigna unguiculata (Early Ramshorn)	cs	m
Nicotiana benthamiana	0	m
N. clevelandii	0	m
Chenopodium amaranticolor	cs	0
C. quinoa	cs	0
Beta vulgaris	cs (ns)	0

Remarks: cs; chlorotic spot, ns; necrotic spot, nl; necrotic lesion, vc; veinclearing, M; mosaic, m; faint mottling, N; necrosis, 0; no symptom, ( ); sometimes appeared.

Nonsusceptible plant species: *Cassia obtusifolia, Crotolaria juncea, Phaseolus vulgaris* (Local, Topcrop), *Vicia faba, Vigna angularis* (Odate No.1), *V. mungo, V. radiata, Nicotiana glutinosa, N. rustica, N. tabacum, Petunia hybrida, Capsicum annuum, Gomphrena globosa, Cucumis sativus, Tetragonia expansa, Zinnia elegans.*

(7) Electron microscopy

Purified preparations revealed the presence of slightly flexuous rods of about 15 nm diameter with a modal length of 610 nm. In thin sections of infected peanut and soybean leaves, scattered virus particles, and sometimes feathery-like structures consisting of virus-like particles were observed in the cytoplasm.

(8) Identification

On the basis of serology, morphology of the virus particles, host range and symptomatology, the virus isolated from peanut was identified as a strain of cowpea mild mottle virus (CMMV). Host range and symptoms produced on susceptible hosts were similar to those of CMMV reported from West Africa (4), and it was serologically related to CMMV from W. Africa. However, unlike the W. Africa CMMV isolate, Indian CMMV infected *Pisum sativum,* showed a higher thermal inactivation point and was not seed-transmitted. Iwaki *et al.* (20) demonstrated that CMMV isolated from soybean in Thailand was transmitted by the whitefly, *Bemisia tabaci.* Indian CMMV was not transmitted by *Aphis craccivora* or *Myzus persicae,* but recently, it was shown to be transmitted by *Bemisia tabaci* in a non-persistent manner (24).

6) Peanut chlorotic leaf streak virus

(1) Virus isolation

A disease characterized by chlorotic spots, streaks and puckering of leaves followed by severe stunting was observed in the Andhra Pradesh, Maharashtra and Gujarat States. A sap transmissible virus was isolated from infected plants.

(2) Host range and symptoms

Thirty two plant species in 7 families were inoculated with the virus mechanically and recovery assays were performed on cowpea cv. C-152. The results indicated that 24 plant species in 5 families were infected with the virus (Table 6). Symptoms on selected plants are given below:

Arachis hypogaea cv. TMV 2. Plant inoculated at the second quadrifoliate stage required nearly 4 weeks to show the first signs of infection. Small chlorotic spots which were observed on young leaves, which gradually increased in size longitudinally, resulting in chlorotic streaks along the veins. As the leaves aged chlorotic streaks became masked and mild mottling was visible. The leaves were puckered and narrow. Infected plants were severely stunted and produced fewer flowers. The kernels were reduced in size. Mottling of the teste and shrivelling of kernels were not noticed.

Naturally infected plants were severely stunted and the upper leaves showed chlorotic streaks. Older leaves were dark green in color and puckered. Axillary shoot proliferation was not noticed. Flowering was noticed on infected plants and the kernels collected from a few infected plants were reduced in size.

Vigna unquiculata cv. C-152. The virus produced chlorotic lesions with a necrotic spot in the center and sometimes a necrotic ring surrounding them. In systemic infection, mild mottling was observed.

Cyamosis tetragonoloba. Concentric dark colored necrotic lesions were observed on inoculated leaves and veinclearing in systemic infection.

Datura stramonium. Concentric necrotic lesions on inoculated leaves, systemic infection with veinclearing, puckering and downward curling of leaves were observed.

Gomphrena globosa. Chlorotic and necrotic local lesions were observed on inoculated leaves.

Petunia hydrida. Systemic mosaic mottling and curling of leaves were observed.

Nicotiana rustica. Chlorotic lesions were observed on inoculated leaves followed by veinclearing. In systemic infection young leaves were puckered and reduced in size.

(3) Transmission

The virus was transmitted by mechanical sap inoculation and grafting. Several attempts to transmit the virus by *Aphis craccivora* in non-persistent and persistent manners were unsuccessful. Nearly 50 seeds, collected from artificially inoculated plants, failed to produce the disease.

(4) Stability in crude sap

Extracts from infected peanut leaves prepared in 0.05 M phosphate buffer were used. The virus had a thermal inactivation point ranging between 80 and 85°C for 10 min. Infectivity was recovered from extracts diluted to 10^{-3} but not 10^{-4}. The virus was infective after 3 days of exposure to 25°C, but not after 4 days.

(5) Purification

The virus maintained in *Nicotiana rustica* consistently yielded high infectivity assays. Purification was performed on the material derived from *N. rustica.* Leaf extracts were prepared in 0.1 M phosphate buffer, pH 7.0, containing 0.5% 2-mercaptoethanol and 0.01 M Na-DIECA. Filtered sap was clarified with 10%

Table 6. Host range of peanut chlorotic leaf streak virus (PCLSV)

Plant species (cultivars)	Symptoms	
	Inoculated leaves	Upper leaves
Arachis hypogaea (TMV 2)	0	M
Canavallia ensiformis	cs	cs
Cyamopsis tetragonoloba	ns	M
Glycine max (Bragg)	0	M
Phaseolus lathyroides	ns	0
P. vulgaris (Local)	nl	N
(Topcrop)	cs	0
Pisum sativus (Bonneville)	0	M
Vicia faba	nl	0
Vigna angularis	0	M, N
V. mungo	0	Lat
V. radiata	0	M
V. unguiculata (Early Ramshorn)	cs, ns	M
(C-152)	cs, ns	M
Datura stramonium	cs	vc
Nicotiana benthamiana	cs	M
N. clevelandii	cs	M
N. clevelandii × *N. glutinosa*	cs	M
N. glutinosa	cs	M
N. rustica	cs	M
N. tabacum	cs	M
Petunia hybrida	0	M
Chenopodium amaranticolor	cs	0
C. quinoa	cs	0
Gomphrena globosa	ns	0
Zinnia elegans	0	M

Remarks: cs; chlorotic spot, ns; necrotic spot, nl; necrotic lesion, vc; veinclearing, M; mosaic, Lat; latent infection, N; necrosis, 0; no symptom.

Nonsusceptible plant species: *Cassia obtusifolia, Cajanus cajan, Crotolaria juncea, Solanum melongena, Cucumis sativus, Brassica oleracea, B. rapa, Raphanus sativus.*

chloroform, and one cycle of differential centrifugation, two cycles of sucrose density gradient centrifugation were used to purify the virus.

Ultraviolet absorption spectrum of the purified virus was typical of that of a nucleoprotein, with minimum and maximum absorbances at 245 nm and 260 nm respectively.

(6) Electron microscopy

Purified virus preparation was fixed in 1.5% glutaraldehyde and stained in 2% PTA, pH 7.0. The preparations revealed the presence of isometric particles 45–50 nm in diameter. Leaf dip preparations from infected peanut and *N. rustica* also revealed the presence of the same particles.

In thin sections, infected peanut and *N. rustica* leaves contained spherical or ellipsoidal intracellular inclusions, with many virus-like particles embedded in a dense matrix. The virus particles and intracellular inclusions were similar to those induced by caulimoviruses.

(7) Identification

The disease was shown to be caused by an isometric virus about 50 nm in diameter. The virus produced inclusion bodies in infected cells, resembling those associated with caulimovirus infections. This virus resembled a member of caulimovirus group in morphology as well as in possessing rather high thermal inactivation point.

Many viruses belonging to the caulimovirus group were reported. However, only one virus, soybean chlorotic mottle virus (SCMV) has been isolated from leguminous plant (21). The host range of SCMV is restricted to the Leguminosae. SCMV appears to different from the virus isolated in India which infected not only Leguminosae but also Solanaceae, Chenopodiaceae and Compositae plants.

Chemical characterization has not yet been performed, however, initial data (unpublished) indicate that the virus belongs to the caulimovirus group containing dsDNA. The virus seems to be a newly recognized one the name of peanut chlorotic leaf streak virus (PCLSV) is proposed.

4. Discussion

Several virus diseases of peanut have been reported from all over the world, and some are recognized as economically important. Peanut mottle virus (PMV) is the most widespread and can cause considerable yield losses (22). Other economically important virus diseases have a more restricted distribution. For instance, groundnut rosette is important in Africa, south of Sahara, peanut clump in West Africa (40), peanut mild mottle (42) and peanut stripe (7) in China, and peanut mottle virus-ring type (10) in Thailand.

In India, peanut mottle has not been found to be economically important. Although groundnut rosette has been reported by Bisht *et al.* (2) and by Raychaudhuri (31), we have not been able to locate rosette in our disease surveys in India.

The most important virus disease of peanut in India is "bud necrosis" caused by tomato spotted wilt virus. The disease is occurring in all the peanut growing areas in India, especially in the semi-arid regions where both the vector (thrips) and alternate hosts are abundant. Although insecticides could be used to reduce the thrips population, it is not economical. It is impractical to eliminate the alternate hosts in tropical regions because the virus has been shown to infect more than 200 species of plants (1). Nearly 7,000 germplasm lines of peanut have been screened and none showed any marked resistance to the virus. However, 2 cultivars consistently showed

a lower than average incidence of the disease under field conditions (11). Some of the *Arachis* species showed a high degree of resistance and transfer of this resistance to cultivated peanut appears to be extremely important.

Clump disease is widely distributed in India and has been found to cause economic losses to the peanut crop, especially in the Punjab State. The disease resembles clump reported from Upper Volta and Senegal (40), in symptoms, structure of the virus particles and transmission. Indian virus was shown to be serologically unrelated to the African virus. Recently the virus has been shown to be transmitted by the fungus, *Polymyxa graminis,* and appeared to belong to the new group of plant viruses "Furovirus" (34).

The occurrence of PGMV on peanut was first reported from Tirupati, Andhra Pradesh State, in 1971. This virus produced symptoms resembling those of PMV. However, the virus appears to be a new potyvirus and is not related to PMV serologically. In laboratory tests, peanut was infected with bean yellow mosaic, pea mosaic and passionfruit woodiness viruses in the potyvirus group (9). Although more serological tests are needed to determine the relationship of PGMV with other potyviruses. PGMV is considered to be a distinct and previously undescribed potyvirus.

Although CMMV is widely prevalent on cowpea in West Africa (4), soybeans in Thailand (20), its natural occurrence on peanut has not been reported. It is suspected that CMMV occurs on peanut in many tropical countries, because of the high natural incidence of the virus in Nigeria (unpublished). CMMV has been shown to be seed-transmitted in cowpea, soybean and French bean (4). Since *Bemisia tabaci* which transmits CMMV is abundant in the tropics, CMMV could become a very important virus in tropical countries.

PCLSV was widely distributed in India and it has the potential to cause severe losses in yield. The virus which appears to be a newly recorded virus resembling caulimoviruses in the structure of the virus particles, the type of inclusion bodies found in the cytoplasm, is possibly a dsDNA.

A few other virus diseases have been observed on peanut in India and they have not yet been characterized. The virus causing yellow spot disease appears to belong to the TSWV goup, and a yellow mosaic disease has recently been shown to be caused by a member of the geminiviruses.

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