

Resistance to tobacco streak virus in groundnut, *Arachis hypogaea* L.

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Abstract

Peanut stem necrosis disease (PSND), caused by tobacco streak virus (TSV), is a new disease in groundnut, which appeared in 2000 in Anantapur district of Andhra Pradesh in India. Eleven peanut bud necrosis disease (PBND) field resistant genotypes of groundnut, *Arachis hypogaea* L. along with JL 24 (susceptible control) were screened in glasshouse for resistance to TSV. Plants were inoculated mechanically at 14 days after sowing (DAS) with two virus concentrations, 1:10 and 1:100. Five genotypes showed tolerance to TSV as compared to JL 24. These genotypes along with JL 24 were again screened in the glasshouse with larger plant population with two virus concentrations (1:10 and 1:100) and at two plant ages (14 DAS and 21 DAS) under Split-Split Plot Design. Among the six genotypes screened, ICGV 99029 (29.4%), ICGV 01276 (34.2%), ICGV 92267 (35.0%), and ICGV 00068 (37.4%) recorded less TSV infection than JL 24 (68.6%). In addition to their tolerance to TSV, these genotypes also possess tolerance to PBND, rust and late leaf spot. These would make good parents in a multiple disease resistance breeding program in groundnut.

Key words: Peanut stem necrosis disease, peanut bud necrosis disease, resistance sources, sap inoculation, peanut

Introduction

Peanut stem necrosis disease (PSND) of groundnut, caused by tobacco streak virus (TSV), occurred in epidemic proportion in the 2000 rainy season in Anantapur district of Andhra Pradesh. The disease affected nearly 0.23 m ha of the total 0.7 m ha, resulting in an estimated loss of Rs. 3 billion (Prasada Rao *et al.*, 2003b). The typical symptoms of PSND are necrosis of terminal portion of the stem (leaflets and terminal bud), which spreads rapidly to the entire stem resulting in complete death of the plant (Reddy *et al.*, 2002). The virus infects several crops and weed species. It is carried through pollen. When the vector thrips (*Frankliniella schultzei* (Trybom), *Megalurothrips usitatus* (Bagnall) and *Scirtothrips dorsalis* Hood.) with infected pollen on their bodies land on groundnut plants, the pollen dislodge. During thrips

feeding both damaged leaf tissue and pollen come in contact and the virus is transmitted. In the case of groundnut, *Parthenium hysterophorus* appears to play a significant role in natural spread of PSND (Prasada Rao *et al.*, 2003a).

To date, there are no satisfactory measures that can effectively control this disease in the field. However, disease management is possible following an integrated approach involving border cropping, intercropping, weed control, seed treatment with imidacloprid, and host plant resistance (Prasada Rao *et al.*, 2003b). Host plant resistance, if available, provides an effective, economical and environment friendly option to control the disease. In an earlier glasshouse screening (mechanical inoculation using sap from virus-infected plant at 1:10 virus concentration), all 150 released varieties of groundnut in India were susceptible. Similarly, all 51 wild *Arachis* accessions screened were susceptible to TSV and showed disease symptoms, except for one accession of *A. chacoense* (ICG 4983), which did not show symptoms in spite of TSV infection (Prasada Rao, unpublished data). The main aim of the present investigation was to identify TSV tolerant groundnut genotypes among the bud necrosis disease (PBND) resistant and thrips resistant groundnut genotypes. As both PBND and PSND are prevalent in peninsular India, a combined resistance to both the diseases is required.

Materials and methods

Eleven PBND and thrips tolerant groundnut genotypes, ICGV 99029, TCGS 647, ICGV 86590, ICGV 00068, ICGV 01270, ICGV 00005, ICGV 00064, ICGV 01276, ICGV 92267, ICGS 37, and ICGV 87486, identified at ICRISAT and elsewhere, and one susceptible control JL 24 were screened for resistance to TSV in glasshouse at ICRISAT Center during April-May 2003. This experiment was conducted in a randomized complete block design. Groundnut seedlings were raised in 20-cm plastic pots containing sterilized sand:soil mixture (2:1) in two replications. Each replication consisted of two pots/cultivar and each pot contained five plants. The plants were sap inoculated at 14 DAS following the procedure described by Prasada Rao *et al.*, 2003b. In addition to the groundnut genotypes, with each inoculation (1:10 and 1:100 virus concentrations) five control plants of cowpea were also inoculated. The inoculated cowpea plants provided

standard for virus infectivity. Among the 11 groundnut genotypes screened, promising genotypes were screened again with larger plant population and at two plant ages during August-September 2003, under Split-Split Plot Design. These tolerant genotypes were raised in 25-cm plastic pots in six replications. Each replication consisted of 3 pots/cultivar and each pot contained 5 plants. Plants were inoculated at 14 and 21 DAS using two virus concentrations 1:10 and 1:100. Observations on disease appearance were taken at weekly intervals and later all plants were tested by ELISA for the presence of the virus.

Virus culture: TSV was originally taken from groundnut and inoculated onto cowpea. Single lesion isolation was made from cowpea to cowpea. Following three successive single lesion transfers by mechanical sap inoculations, virus was multiplied on cowpea (Prasada Rao *et al.*, 2003b).

Mechanical sap inoculations: Cowpea infected leaves were harvested from the glasshouse grown plants. Two virus dilutions 1:10 (1 g of infected tissue : 9 ml of inoculation buffer) and 1:100 were made to inoculate groundnut plants. Inoculations were performed with the help of mortar and pestle after dusting the plants with carborundum.

Observations on disease appearance were recorded at weekly intervals on the inoculated plants. After 25 days of inoculation, leaf samples from all the plants were collected for ELISA tests for the presence of virus. From symptomatic plants, only young leaflets that showed early disease symptoms were tested, and from the nonsymptomatic plants, young leaflets from three branches were pooled and tested. Samples were processed by using direct antigen coating (DAC) ELISA method (Hobbs *et al.*, 1987).

As the data sets in both screenings satisfied the assumption of homogeneity of variance, they were analysed without transformation.

Results and discussion

In preliminary studies, conducted during October - November 2002, the virus failed to produce symptoms in groundnut (symptom expression in cowpea was delayed), probably due to prevailing low temperatures (minimum temperature was 8-9°C). Considering the influence of temperature on disease expression, the present experiments were conducted during April-September 2003 (minimum temperature 20-22°C), which resulted in a high level of TSV infection. Olorunju *et al.* (1995) also observed that warm to hot temperatures in May and July apparently caused groundnut rosette virus (GRV) to appear earlier and to be more severe than cooler temperatures in October and December in susceptible groundnut plants in Nigeria. Cowpea plants inoculated along with test plants showed 100% infection. Groundnut plants without the disease symptoms gave negative results in ELISA.

At a higher virus concentration (1:10), the disease incidence among the 11 genotypes ranged from 35 to 100%. ICGV 01276 recorded the lowest and ICGV 87846 the highest disease incidence. ICGV 99029 and ICGV 92267 had 55% disease incidence. In the remaining genotypes the disease incidence was 80% and above (Table 1). Inoculation with lower virus concentration (1:100) provided a more discernible disease picture among the genotypes. Two genotypes ICGV 92267 and ICGV 00068 did not show any disease symptoms or infection of TSV. The maximum disease incidence observed was 80% in ICGV 87846. ICGV 99029 and ICGV 01276 showed 15%, ICGS 37 showed 30% and ICGV 00005, ICGV 00064 and TCGS 647 showed 40% disease incidence (Table 1).

Table 1 Reaction of peanut bud necrosis disease field tolerant groundnut genotypes to tobacco streak virus (TSV) in glasshouse, ICRISAT Centre, April-May, 2003

Genotype	TSV infection (%)		Average TSV infection (%)
	Virus concentration		
	1:10	1:100	
ICGV 99029	55	15	35.0
TCGS 647	100	40	70.0
ICGV 86590	95	65	80.0
ICGV 00068	85	0	42.5
ICGV 01270	80	55	67.5
ICGV 00005	90	40	65.0
ICGV 00064	80	40	60.0
ICGV 01276	35	15	25.0
ICGV 92267	55	0	27.5
ICGS 37	80	30	55.0
ICGV 87846	100	80	90.0
Control : JL 24	95	75	82.5
CD (P=0.05)			34.2

Based on the above results, five genotypes, ICGV 99029, ICGV 00068, ICGV 01276, ICGV 92267 and ICGS 37, were identified as promising against TSV infection and screened further using larger plant population at two plant growth stages (14 DAS and 21 DAS). The results are summarized and presented in Table 2. At the higher virus concentration, the mean disease incidence (51.7%) was significantly more than the lower virus concentration (35.9%). Similarly, at young plant age the disease incidence (48.1%) was significantly more than the older plant age (39.4%). If virus infection occurs at later stages of plant growth, more plants are likely to escape the disease. At 1:10 virus concentration and plant age 14 DAS, the disease incidence ranged from 21% in ICGV 01276 to 59.8% in JL 24, whereas at the same virus concentration and plant age 21 DAS, the disease incidence ranged from 13.3% in ICGV 00068 to 57.7% in JL 24. At 1:100 virus concentration and plant age 14 DAS, the disease incidence ranged from 30% in ICGV 99029 to 86.6% in JL 24, whereas at the same virus concentration

and plant age 21 DAS, the disease incidence ranged from 21% in ICGV 00068 to 70.2% in JL 24. All the five genotypes screened further had significantly lower disease incidence than the control JL 24 (68.6%). Although, the disease incidence in ICGV 99029 was the lowest (29.4%), other genotypes, ICGV 01276 (34.2%), ICGV 92267

(35.0%), and ICGV 00068 (37.4%), did not differ significantly from it. In addition to bud necrosis disease (another virus disease caused by peanut bud necrosis virus), which is prevalent in peninsular India, these varieties also have tolerance to TSV/PSND. They can provide field protection against both the virus diseases.

Table 2 Reaction of promising groundnut genotypes for resistance to tobacco streak virus (TSV) in glasshouse, ICRISAT Centre, August-September, 2003

Genotype	Average TSV infection (%)												Overall mean
	P ₁			P ₂			VC ₁			VC ₂			
	VC ₁	VC ₂	Mean	VC ₁	VC ₂	Mean	P ₁	P ₂	Mean	P ₁	P ₂	Mean	
ICGV 92267	28.8	55.5	42.2	23.3	32.2	27.7	28.8	23.3	26.1	55.5	32.2	43.9	35.0
ICGS 37	43.3	68.8	56.1	53.5	66.7	60.1	43.3	53.5	48.4	68.8	66.7	67.8	58.1
ICGV 99029	32.0	30.0	31.0	18.8	36.8	27.8	32.0	18.8	25.4	30.0	36.8	33.4	29.4
ICGV 00068	47.5	67.7	57.6	13.3	21.0	17.2	47.5	13.3	30.4	67.7	21.0	44.4	37.4
ICGV 01276	21.0	36.5	28.8	31.3	47.8	39.6	21.0	31.3	26.2	36.5	47.8	42.2	34.2
Control: JL-24	59.8	86.7	73.3	57.7	70.2	64.0	59.8	57.7	58.8	86.7	70.2	78.5	68.6
Mean	48.1			39.4			35.9			51.7			
CD (P=0.01): Genotype (G) = 11.4; Plant age (P) = 6.2; Virus conc. (VC) = 7.2; G x P = 15.1; P x VC = NS; G x VC = NS; G x P x VC = NS													
NS = Non-significant; P ₁ = Plant age 1 (14 DAS); P ₂ = Plant age 2 (21 DAS) VC ₁ = Virus concentration 1 (1:100) VC ₂ = Virus concentration 2 (1:10)													

ICGV 99029 is also tolerant to rust (caused by *Puccinia arachidis* Speg.) and ICGV 00068 to both rust and late leaf spot (caused by *Phaeoisariopsis personata* Berk and Curt.) diseases (ICRISAT, unpublished data). ICGV 92267, besides being tolerant to rust and late leaf spot, is early maturing and is also tolerant of low temperature at germination (Upadhyaya et al., 2002). All these genotypes will make good parents in a multiple disease resistance breeding program in groundnut.

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