The occurrence of aphids and rosette in groundnut bait plots in Central Malawi, an area with monomodal rainfall and an extended dry season, was studied during the 1996 dry season. The preceding wet season saw high rosette incidence in this area. It was assumed that if alternative hosts for rosette existed in the region, they would act as a source of inoculum for transmitting rosette to groundnut bait plots sown in the dry season. We collaborated with vegetable growers in "dambo" areas (moist valleys with waterlogging) in a 30-km radius around Chitedze, Central Malawi. Thirty-seven plots in five villages, were sown with Malimba, a short-duration cultivar, during the last week of Aug 1996. Groundnut was sown at 30 × 30 cm in raised beds in an area of 100 m^2 per farm, in soil varying from heavy black clay to light sandy loams. The plots were irrigated with well water. Pesticides were not applied. Populations of *A. craccivora* and incidence of rosette were recorded until the end of dry season. Aphids were first noticed during the last week of Oct (about 2 months after sowing), in 29 % of the fields. The crop was in the flowering stage. However, the canopy was not fully covered. Aphid infestation ranged from 6 to 32%, and on each plant the size of colony did not exceed 50 individuals. Aphid populations disappeared 2 weeks later, and did not reappear. Rosette symptoms were not noticed. Crop growth was satisfactory in all the locations.

Results show that rosette did not occur during the 1996 dry season in Central Malawi despite the presence of the vector. Surveys in farmers' fields in the Chitedze area also did not reveal rosette symptoms on groundnut volunteers (Bottenberg and Subrahmanyam 1997). In addition, rosette was not noticed during two visits in Oct 1996 to a 6-ha irrigated vegetable farm with about 2 ha of groundnut near Salima, 100 km east from Chitedze (Bottenberg, unpublished data). These observations support our conclusion that rosette is not harbored by alternative hosts in Central Malawi during the dry season.

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Dry Season Survival of Groundnut Volunteer Plants and Ground Keepers (*Arachis hypogaea* L.) in Central Malawi

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One of the deficiencies in our understanding of the epidemiology of groundnut rosette, a major virus disease of groundnut in Africa, is where the virus complex survives during the dry season. The vector *Aphis craccivora* Koch has been recorded from a wide variety of alternative hosts during the dry season but none have been shown to carry any of the rosette viruses

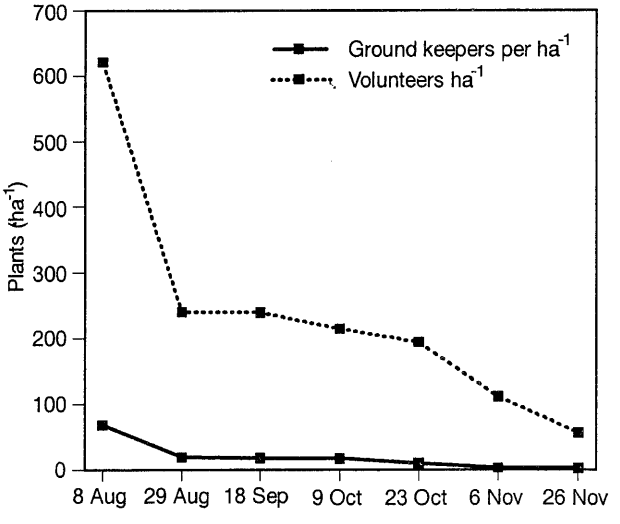

Figure 1. Densities of groundnut volunteers and ground keepers in farmers' fields (n = 21) in Central Malawi during the 1996 dry season.

components under natural conditions (Adams 1967, Reddy et al. 1985). Groundnut volunteer plants and "ground keepers" may survive during the dry season depending on the cultivar used and local climatic conditions. Volunteer plants originate from seeds of short-duration spanish/valencia cultivars that were left in the ground after harvest in soils with residual moisture. Ground keepers are plants or their parts not lifted during harvest, thus left in the ground. Such plants may be short- or long-duration types. Under favorable conditions, volunteers and ground keepers may carry aphid colonies and provide rosette inoculum (Evans 1954). Currently the importance of volunteers and ground keepers in Central Malawi in perpetuating the rosette inoculum is not known (Bock and Nigam 1988, Hildebrand et al. 1991). The objective of this study was to monitor the survival of ground keepers and volunteers in farmers' fields during the dry season to assess their importance in providing a source for the survival of aphids and in perpetuating the rosette disease inoculum.

During an initial survey in Aug 1996, 21 fields out of a random sample of 104 harvested groundnut fields within a 30-km radius around Chitedze, Central Malawi, were found to have ground keepers and groundnut volunteers. In each of these fields, the number of groundnut plants was counted in a centrally located sample area of 0.1 ha; the condition and growth stage of each plant and the presence of aphids and rosette was also noted. Assessments were made every 2–3 weeks until the end of the dry season.

Volunteer plants persisted throughout the dry season (Fig. 1). Densities decreased from 622 plants ha^{-1} in early Aug to 57 plants ha^{-1} by the end of Nov. Growth stage ranged from 2-leaf stage to flowering stage; many plants showed profuse branching because the central shoot was chewed off by goats and cows. Very little growth was noted during the period of observation. Ground keepers were less common. There were 69 plants ha^{-1} in early Aug which dropped to 3 plants ha^{-1} in late Nov. Ground keepers often had a withered and tattered appearance and consisted mostly of one or two branches with a partial root system. Plant mortality resulted from drought, browsing by animals (if the entire plant was destroyed), and physical removal by farmers during land preparation at the end of the dry season. There were a total of three localized, light showers (<5 mm) during Oct and Nov which may have provided sufficient moisture for germination of some remnant groundnut seed. However, volunteers that developed early in the dry season were always more vigorous than those that germinated at a later date, presumably because 'early' volunteers had a more extensive root system than 'late' volunteers and were therefore less sensitive to drought. In addition, removal of the primary shoot by browsing animals, reduced the plant size with lateral branches closely hugging the ground that may have further increased drought resistance of early volunteers. Land preparation was started in Sep (one field partially tilled); by the end of Nov, 9 out of 21 fields were partially or completely tilled. No rosette symptoms were noticed on volunteers or ground keepers. Small aphid colonies (less than 50 aphids) were found on volunteer plants only in 3 fields (3 out of 83 plants) on 18 Sep and in one field (one out of 66 plants) on 8 Oct.

This survey showed that groundnut volunteer plants can survive during the dry season, and do not support the data from previous studies suggested (Bock and Nigam 1988, Hildebrand et al. 1991). However, the role of groundnut volunteers as carriers of aphids and rosette inoculum in Central Malawi is yet to be determined.

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Detection of Tomato Spotted Wilt Tospovirus Infection of Groundnut by Immunocapture RT-PCR

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Tomato spotted wilt tospovirus (TSWV) is a major constraint to groundnut production in southern USA. Rapid and sensitive detection of TSWV infection of groundnut by such molecular techniques as reverse transcription and polymerase chain reaction (RT-PCR) has been hampered by technical difficulties in obtaining suitable RNA preparations from groundnut tissue. Immunocapture RT-PCR (IC-RT-PCR) is another highly rapid and sensitive technique widely being used for the detection of plant virus infections of many crop plants. After introducing some modifications, we assessed the potential application of this technique for the detection of TSWV in groundnut.

IC-RT-PCR was performed in pre-treated microfuge tubes (200 µL capacity, thin-walled tubes, Research Products International, Mount Prospect, IL, USA). To ensure the binding of the primary antibody, the tubes were treated for 15 min each in 0.1 N hydrochloric acid and 4 N sodium hydroxide. After each treatment, tubes were rinsed with PBST buffer (0.137 M sodium chloride, 8 mM sodium phosphate-dibasic, 1.7 mM potassium phosphate-monobasic, 2.7 mM potassium chloride, and 0.05% (v/v) Tween-20) and finally washed with 95% ethanol for 15 min, before they were air-dried at room temperature (B.G. Cassidy, personal communication).

Pre-treated tubes were first coated with 100 µL of the antibody (1:200 dilution) specific to the nucleocapsid protein (Agdia Inc., Elkhart, IN, USA) by incubating overnight at 4°C. The antibody solution was discarded and the tubes were washed three times with sterile distilled water before adding the tissue extract. Groundnut plants were collected from the fields in south Georgia. Leaves and roots were processed separately by grinding in an extraction buffer (0.01 M sodium sulfite, 2% polyvinylpyrrolidone, MW 24–40 000, 0.2% sodium azide, 0.2% powdered egg albumin, and 2% Tween-20 dissolved in 1 L PBS, pH adjusted to 7.4). The extract was clarified and the supernatant (100 µL each) was added to the antibody-coated microfuge tubes. The tubes were incubated at room temperature for 3 h, rinsed three times with sterile distilled water, followed by the addition of 64 µL sterile distilled water to each tube. The tubes were subjected to three cycles of freezing (-80°C for 10 min) and thawing (70°C for 5 min). The tubes were then transferred to ice before adding RT-PCR components. RT and PCR were done in the same tube without any buffer changes as described by Pappu et al. (1993), except that Q Taq (Qiagen Inc., Chatsworth, CA, USA) was used as the DNA polymerase. The primer pair specific to the TSWV-nucleocapsid gene (N gene), and RT-PCR conditions used were those described previously (Pappu et al. 1996).

RT-PCR showed the presence of a major DNA fragment of ca. 800 bp expected as a result of specific amplification of TSWV-N gene (Fig. 1). Occasionally, a smaller, ca. 600 bp fragment was seen, possibly due to non-specific amplification. However, this did not interfere with the interpretation of the results. Both roots as well as leaves from infected groundnut plants provided a template suitable for TSWV detection by IC-RT-PCR

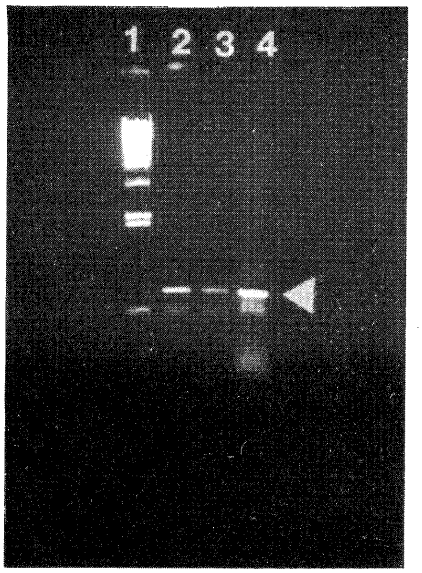

Figure 1. Agarose gel (0.8%) electrophoresis of immunocapture RT-PCR reaction products. Lane 1, Lambda DNA digested with HindIII; Lane 2, tissue extract from infected peanut leaf; Lane 3, tissue extract from infected groundnut root; Lane 4, TSWV N gene amplified from a DNA clone used as a size standard. Presence of the ca. 800 bp DNA fragment (with the arrowhead) in Lanes 1 and 2 indicated TSWV infection.