Inheritance of Resistance in Peanut to Mixed Infections of Groundnut Rosette Virus (GRV) and Groundnut Rosette Assistor Virus and a Single Infection of GRV

P. E. OLORUNJU, Department of Plant Science, Institute for Agricultural Research Samaru, Ahmadu Bello University, PMB 1044, Zaria, Nigeria; C. W. KUHN and J. W. DEMSKI, Division of Plant Pathology, University of Georgia, Athens 30602; and S. M. MISARI and O. A. ANSA, Institute for Agricultural Research Samaru, Ahmadu Bello University, PMB 1044, Zaria, Nigeria

ABSTRACT

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Inheritance of resistance to the green form of groundnut rosette was studied in two ways—aphid inoculation in the field with a mixed culture of groundnut rosette virus (GRV) (plus its satellite RNA) and groundnut rosette assistor virus (GRAV) and mechanical inoculation in the greenhouse with GRV (plus its satellite RNA). Crosses were made with two resistant and six susceptible genotypes in a diallel test. In most crosses, the resistance was conditioned by two recessive genes. Furthermore, results were similar both with mixed infections and the single GRV infection, thus providing direct evidence that genetic control of resistance is to GRV and not to GRAV. In the RMP 12 × M1204.78I cross (and its reciprocal), F_2 progeny segregated 1:3 susceptible/resistant. Mechanical inoculation with GRV provided an acceptable and rapid procedure to screen segregating populations for resistance to the virus.

The discovery of sources of resistance to groundnut rosette by Sauger and Catharinet (18,19) and de Berchoux (5) was a major breakthrough for the improvement of peanut (*Arachis hypogaea* L.). Efficient use of the sources of resistance requires an understanding of the genetic control of resistance and a knowledge of the amount of genetic variability available for selection. Two studies on inheritance of resistance have been reported. Both de Berchoux (6) and Nigam and Bock (11) worked with the chlorotic form of groundnut rosette and found that resistance was controlled by two recessive genes. In a report to the African Groundnut Council in 1977, C. Harkness, working with three crosses, found a similar genetic control for the green form of groundnut rosette.

General manipulation of the viruses associated with groundnut rosette has been difficult because of the following complicating factors: 1) two viruses, groundnut rosette virus (GRV) and groundnut rosette assistor virus (GRAV), exist together in diseased plants in nature (7,13), 2) a GRV satellite RNA is responsible for both rosette symptom production (9,10) and aphid transmission (8), 3) at least two major forms of the disease (chlorotic and green) occur in nature, 4) GRV is dependent on GRAV for vector transmission (persistent) (7), and 5) there has been erratic success with mechanical inoculation of GRV (17). Improvement in the mechanical inoculation procedure for GRV now allows 100% infection of plants of susceptible peanut genotypes (14,15). This procedure was extremely useful in studying the inheritance pattern of resistance to a single virus, GRV.

The objectives of this study were to determine the inheritance of resistance in peanut to mixed infections of GRV (green rosette) and GRAV in the field and to a single viral infection of GRV through mechanical inoculation in the greenhouse, to determine the effectiveness of a field screening procedure, and to study the feasibility of using mechanical inoculation of GRV as a quick screening procedure in a breeding program.

On the basis of electrophoresis diagnostic tests, we believe all references in this paper to infections of GRV should also include its satellite RNA (8–10).

MATERIALS AND METHODS

Genotypes. The field reaction of eight peanut genotypes to GRV and GRAV has been described (16). Two of the genotypes, RMP 12 and RG 1, have a high level of resistance to groundnut rosette. The other six genotypes (55-437, ICGS-56(E), JL 24, M1204.78I, MK 374, and RRB) are susceptible to rosette.

Hybridization. Hybridizations were made in Athens, GA, in 1987. F_1 full-

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and half-sib families were produced by crossing all eight parents in the greenhouse. Crosses were made following the method of Norden (12). Pollinated flowers and pegs that developed were marked with a waterproof paint. F_1 plants were also backcrossed to both resistant and susceptible parents. Seeds for F_2 plants were produced, harvested, and taken to Samaru, Nigeria, for field evaluation under local seasonal conditions.

Field experiments in 1988. Virus inoculum from green rosetted plants growing near Samaru, Nigeria, consisting of a mixture of GRV and GRAV, was estabished by aphid inoculation as described previously (14,16). The parental lines, F_1, F_2, and backcross populations were planted on research land of the Institute for Agricultural Research Samaru, Ahmadu Bello University at Zaria, Nigeria, on 17 July 1988 in a randomized complete block design with four replications. Seeds were treated with thiram for protection against fungi. Each plot consisted of 17 rows 3.6 m long. Four rows were planted with F_2 plants, two had parental plants, and another two had a mixed population of F_1, backcrosses, reciprocals, and parents. The remaining nine rows were used as infector rows and planted with susceptible genotype F 452.4, which was planted in alternate rows with the test material. Spacing between plants was 20 cm with 16 plants per row and rows were spaced 75 cm apart. Nine days after the experimental material was planted, peanut seedlings infected with GRV and GRAV, produced in the greenhouse and heavily infested with viruliferous aphids, were transplanted at a 1.5 m spacing at three plants per infector row.

Beginning 13 days after the rosetted plants were transplanted to the infector rows, disease evaluations were made at weekly intervals during the first 4 wk and every 2 wk thereafter. Individual plants with symptoms were identified at each evaluation date. At 45 days after inoculum was introduced, a disease rating system, described previously (16), was used to assign a specific number of 1–5 (1 = symptomless and 5 = severely diseased) to each test plant.

Field experiments in 1989. The general field design with alternate rows of test and infector plants in 1989 was similar to that of 1988. The experiment consisted of plots that had three rows of F_2 plants, two rows of parental plants, and six rows of F 452.4 infector plants (one row between each test row) planted in a complete block design with three replications. Planting was on 6 June and seedlings infected with groundnut rosette viruses were transplanted (1.5-m spacing) into infector rows 15 days later.

Disease observations began 8 days after exposure to inoculum. The 1988 disease rating system (16) was modified slightly to take into account the large number of resistant plants that developed symptoms. Rating 3 was separated into 3a and 3b whereby 3a = rosette leaf symptoms appearing between 27 and 42 days after exposure to inoculum, and 3b = rosette leaf symptoms appearing at least 27 days after exposure to inoculum. Plants rated 3a were slightly stunted and those rated 3b were markedly stunted. Index values were determined as described previously (16) but with a slightly different weighting system, e.g., $(A + 2B + 2.7C + 3.3D + 4E + 5F)$/the number of plants per plot where A, B, C, D, E, and F equal the number of plants with ratings of 1, 2, 3a, 3b, 4, and 5, respectively.

Mechanical inoculation of GRV. Ten of the F_2 crosses from the rosette hybridization program were used in this study. Genotype F 452.4 was used as a susceptible check, whereas F_2 plants of RMP 12 × RG 1 (resistant × resistant [R × R]) were the resistant check. Resistant × susceptible (R × S) crosses were RMP 12 × JL 24 and RMP 12 × RRB; S × R crosses were 55-437 × RMP 12, M1204.78I × RMP 12, and ICGS-56(E) × RMP 12. Susceptible × susceptible (S × S) crosses were M1204.78I × JL 24, RRB × JL 24, MK 374 × JL 24, and RRB × ICGS-56(E). Because of seed shortage, unequal numbers of plants per cross were tested, and none of the RG 1 crosses could be tested.

GRV was isolated from peanut plants infected with both GRV (green rosette) and GRAV. Serial transfers of GRV, including its satellite RNA, by mechanical inoculation were made to and from susceptible F 452.4 plants until 100% infection of the plants routinely occurred (about four transfers). Thereafter, F 452.4 plants served as the source of inoculum that was maintained by inoculating new seedlings each week. Peanut seeds were planted into 10-cm-diameter pots and then maintained in a greenhouse treated weekly to control aphids. Five- to seven-day-old peanut seedlings, in the two-leaf stage, were kept in the dark for 24 hr before inoculation. Seedlings were then removed from the dark and kept for at least 1 hr before inoculation to enable closed leaflets to open. Inoculum preparation and inoculation procedure were reported previously (14,15).

Inoculated plants were observed daily for first symptom appearance, which was recorded for each plant. Plants that did not show symptoms 12 days after inoculation were reinoculated. Plants were rated on a scale of 1–5 where 1 = no symptoms, 2 = leaf symptoms with no stunt, 3 = leaf symptoms with stunt ranging from slightly discernible to about 15%, 4 = leaf symptoms with stunt of about 15–50%, and 5 = leaf symptoms with stunt greater than 50%. Disease index values were determined according to the procedure described earlier.

Diagnostic tests. Leaf samples for both nucleic acid and serological tests were taken in 1989 from field plants rated 1–2 from all crosses. The leaf samples were dried over calcium chloride in a desiccator and transported to Georgia for diagnostic tests: electrophoresis for GRV satellite RNA (3) and enzyme-linked immunosorbent assay (ELISA) (potato leafroll virus antiserum provided by R. Casper, Institute for Plant Virus Dis-

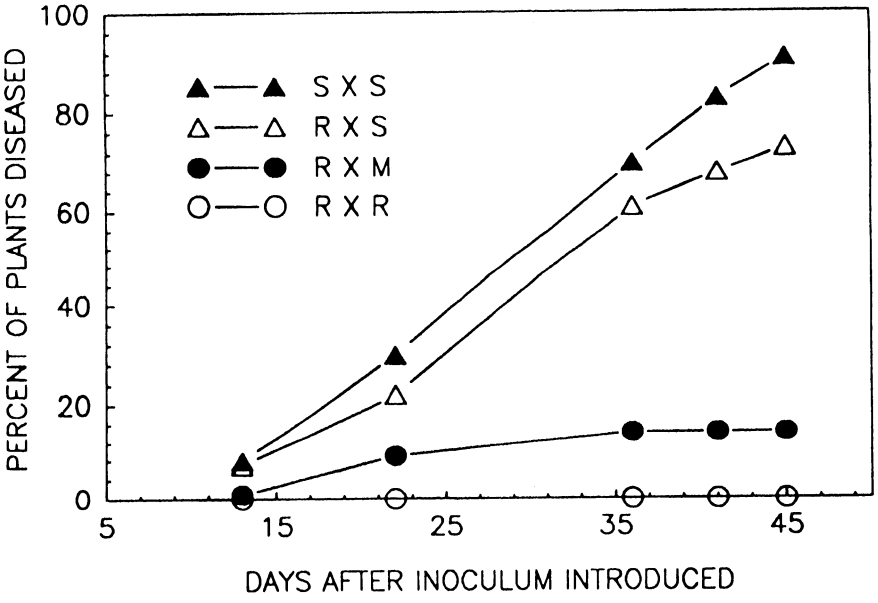

Fig. 1. Disease progress curves of rosette resistant and susceptible peanut crosses grown in the field at Samaru, Nigeria, in 1988. Inoculum originated from viruliferous aphids and infected plants transplanted into infector rows (every other row of plot) of susceptible F 452.4. S × S = susceptible × susceptible, R × S = resistant × susceptible (also includes susceptible × resistant), R × M = RMP 12 × M1204.78I, and R × R = resistant × resistant. Data points represent the average percentage of diseased plants (disease ratings of 2–5) of four replications of F_2 populations.

eases, Braunschweig, Germany) for GRAV (4). Specific procedures for use with peanut tissue have been described previously (14,16). Samples were handled in compliance with a shipping permit issued by the Georgia Department of Agriculture and the Animal and Plant Health Inspection Service, U. S. Department of Agriculture.

RESULTS

Field experiments in 1988. Rosette disease conditions were moderate to severe in 1988. In S × S crosses, the final disease incidence was 88% (Fig. 1), similar to susceptible parents (3,455 of 3,924 plants diseased), and the disease severity index was a moderate 3.2 (scale of 1–5). About 10% of the plants of resistant parents had very mild leaf symptoms. Disease incidence was lower for R × S and S × R crosses than for S × S crosses (Fig. 1). Also, the disease severity index was less for R × S and S × R crosses (2.9 ± 0.1) than for S × S crosses (3.2 ± 0.1). One R × S cross (RMP 12 × M1204.78I) (labeled R × M in Fig. 1) was distinctly different from all other R × S crosses (Fig. 1). Disease incidence was only 13%, compared with an average of 72% (range of 53.5–90.3%) for the other R × S and S × R crosses, and the disease severity index was only 2.0 (± 0.2). Segregation occurred in all R × S and S × R crosses and no diseased plants were observed in the R × R cross.

Field experiments in 1989. Disease incidence in plants (3,846) of susceptible parents and infector genotype F452.4 in the field in 1989 reached 100% and disease severity was very high (Fig. 2, Tables 1 and 2). Symptoms appeared as early as 8 days (no observations were made before this date) after infector plants were transplanted into the field, and 100% infection of some S × S crosses was observed 22 days later. Disease incidence increased rapidly for the R × S (and S × R) and S × S crosses with 50–60% infection by the first observation date. Even though incidence and disease severity were high, the R × S and S × R crosses had a lower disease incidence than S × S crosses from day 8 to days 40–65 when resistant plants in R × S and S × R crosses began to show mottle but not stunting symptoms (Fig. 2).

In sharp contrast to 1988, most plants of the resistant parents (85%) and R × R crosses (70%) developed rosette leaf symptoms (Fig. 2 and Table 1). Symptoms on these plants were mild and appeared only on young leaves of a few branches. Stunting was observed on only 6–7% of resistant plants. Although these few plants had a susceptible disease rating (3b–5) (Table 1), we believe they are genetically like the other resistant plants and develop severe symptoms only because of early infection and a long incubation period. Disease incidence gradually increased to 67% by 65 days (Fig. 2). Again in 1989, the RMP 12 × M1204.78I cross was intermediate between the R × R and the R × S and S × R crosses but closer to the R × R cross except on the last evaluation date (Fig. 2). Despite the high incidence of rosette in the field, the F_2 population of the R × S and S × R group of crosses (excluding RMP 12 × M1204.78I) had a range of 6–26% of their plants showing resistance (Table 1). The cross RMP 12 × ICGS-56(E) had more resistant plants than the other R × S crosses. Also, resistant plants were observed in the S × S cross ICGS-56(E) × JL 24, where 8% of the plants developed symptoms late and were rated resistant (Table 1).

Disease severity was higher in 1989 (Table 1) than in 1988. Populations of the R × R and RMP 12 × M1204.78I crosses had index values similar to those of resistant parents and different from the rest of the crosses.

Because of the 100% disease incidence in the susceptible genotypes in 1989, field results of the inheritance pattern of F_2 populations were more meaningful than those of 1988 (Table 2). Chi-square analysis of the breeding data showed that F_2 plants in four of the eight R × S crosses segregated at a 15:1 S/R ratio (Table 2). Two crosses (RRB × RMP 12 and RG 1 × M1204.78I) segregated 13:3 S/R, and one (RMP 12 × ICGS-

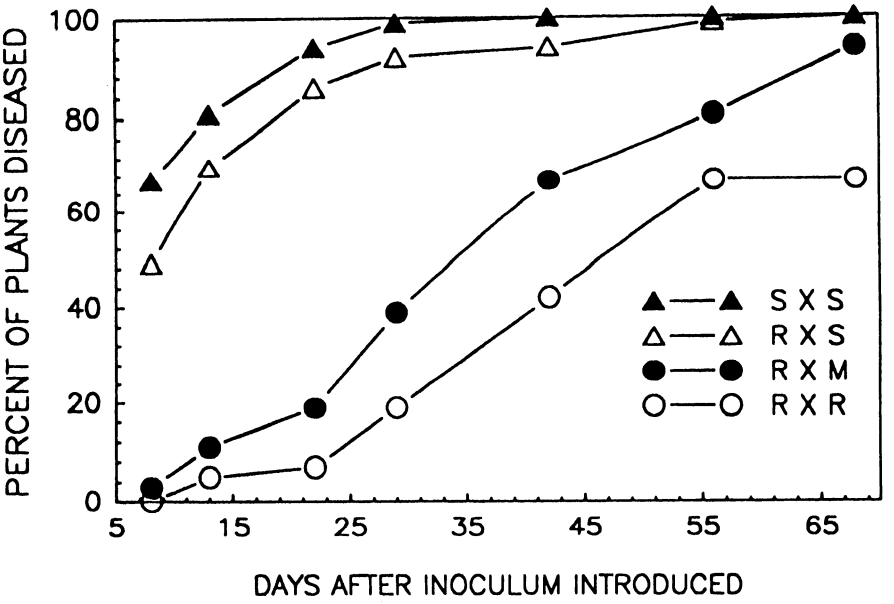

Fig. 2. Disease progress curves of rosette resistant and susceptible peanut crosses grown in the field at Samaru, Nigeria, in 1989. Inoculum originated from viruliferous aphids and infected plants transplanted into infector rows (every other row of plot) of susceptible F 452.4. S × S = susceptible × susceptible, R × S = resistant × susceptible (also includes susceptible × resistant), R × M = RMP 12 × M1204.78I, and R × R = resistant × resistant. Data points represent the average percentage of diseased plants of three replications of F_2 populations.

Table 1. Groundnut rosette disease rating number frequency and disease index of F_2 plants from 11 peanut crosses in field tests at Samaru, Nigeria, in 1989

		Plants (%) in each rating no.[b]					
		Resistant			Susceptible		Disease index value
Genotype	Phenotype[a]	1	2	3a	3b	4-5	
RMP 12	R	17	67	10	4	2	2.0 ± 0.13
RG 1	R	12	67	14	2	5	2.1 ± 0.11
RMP 12 × RG 1	R × R	30	52	11	5	2	1.9 ± 0.18
RMP 12 × M1204.78I	R × S	5	58	6	11	11	2.5 ± 0.07
RMP 12 × RRB	R × S	0	2	4	25	70	3.7 ± 0.10
RMP 12 × ICGS-56(E)	R × S	0	17	9	3	71	3.7 ± 0.12
RMP 12 × 55-437	R × S	0	5	3	43	49	3.5 ± 0.10
RMP 12 × MK 374	R × S	0	7	0	8	85	4.0 ± 0.05
RRB 12 × RMP 12	S × R	0	13	6	20	61	3.7 ± 0.18
RG 1 × RRB	R × S	0	8	3	18	71	3.9 ± 0.21
RG 1 × M1204.78I	R × S	0	10	3	6	81	3.8 ± 0.09
RRB × 55-437	S × S	0	0	0	27	73	3.8 ± 0.07
ICGS-56(E) × JL 24	S × S	0	0	8	18	74	3.8 ± 0.09

[a] R = resistant; S = susceptible.

[b] Rating scale: 1 = no symptoms; 2 = leaf symptoms, no stunt; 3 = symptoms plus stunt (general plant size) ranging from slightly discernible to about 30%; 4 = symptoms plus stunt of about 30–70%; 5 = symptoms plus stunt greater than 70%. Ratings 3a and 3b were distinguished by time of first symptom appearance and degree of stunting.

Table 2. Reaction to groundnut rosette in parents and F_2 populations and segregation ratios of F_2 populations in field tests (mixed infections of groundnut rosette virus and groundnut rosette assistor virus) at Samaru, Nigeria, in 1989

Genotype	Phenotype[a]	Total no. of plants	Susceptible reaction (%)		Best fitting model (S/R)	χ^2	Probability
			Observed	Expected			
Infector plants	S	3,366	100.0	100.00			
Susceptible parents	S	480	100.0	100.00			
Resistant parents	R	432	5.0	0.00			
RMP 12 × RG 1	R × R	130	6.9	0.00	0:1	0.6231	0.500–0.250
RRB × 55-437	S × S	77	100.0	100.00	1:0	0.0000	
ICGS-56(E) × JL 24	S × S	125	93.6	100.00	15:1	0.0048	0.980–0.950
RMP 12 × RRB	R × S	114	94.7	93.75	15:1	0.1894	0.750–0.500
RMP 12 × 55-437	R × S	128	91.4	93.75	15:1	1.2000	0.500–0.250
RMP 12 × MK 374	R × S	119	93.3	93.75	15:1	0.0454	0.900–0.750
RG 1 × RRB	R × S	109	89.9	93.75	15:1	2.7456	0.100–0.050
RRB × RMP 12	S × R	122	81.1	93.75	13:3	0.0008	0.990–0.970
RG 1 × M1204.78I	R × S	109	87.2	93.75	13:3	2.4956	0.250–0.100
RMP 12 × ICGS-56(E)	R × S	92	75.0	93.75	3:1	0.0000	
RMP 12 × M1204.78I	R × S	109	22.9	93.75	1:3	0.2477	0.750–0.500

[a] R = resistant; S = susceptible.

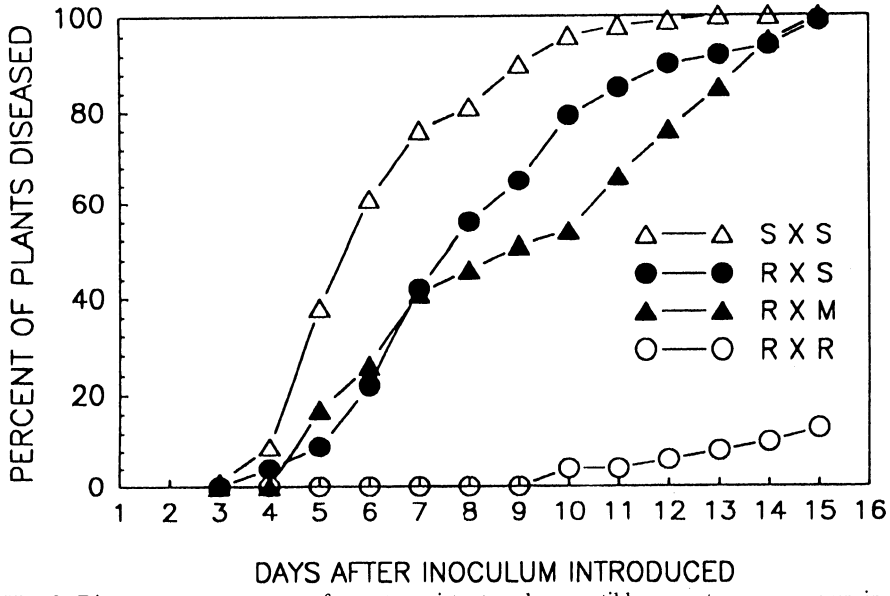

Fig. 3. Disease progress curves of rosette resistant and susceptible peanut crosses grown in the greenhouse and mechanically inoculated with groundnut rosette virus. S × S = susceptible × susceptible, R × S = resistant × susceptible (also includes susceptible × resistant), R × M = RMP 12 × M1204.78I, and R × R = resistant × resistant cross. Data points represent the average percentage of diseased plants of F_2 populations.

Table 3. Groundnut rosette disease rating number frequency and disease index of plants in parental and F_2 populations mechanically inoculated with groundnut rosette virus (green strain) in the greenhouse

Genotype	Phenotype[a]	Plants (%) in each rating no.[b]					Disease index value
		Resistant			Susceptible		
		1	2	3	4	5	
RMP 12	R	33	67	0	0	0	1.7
RG 1	R	17	75	0	8	0	1.9
RMP 12 × RG 1	R × R	88	8	4	0	0	1.2
M1204.78I × RMP 12	S × R	0	59	15	26	0	2.7
RMP 12 × RRB	R × S	7	0	0	31	62	3.7
RMP 12 × JL 24	R × S	0	4	0	26	70	3.8
ICGS-56(E) × RMP 12	S × R	0	7	0	49	44	3.4
55-437 × RMP 12	S × R	0	6	0	12	82	3.8
M1204.78I × JL 24	S × S	0	0	0	15	85	4.1
JL 24 × RRB	S × S	0	0	0	6	94	4.9
RRB × ICGS-56(E)	S × S	0	0	0	8	92	4.4
JL 24 × MK 374	S × S	0	0	0	6	94	4.5

[a] R = resistant; S = susceptible.
[b] Rating scale: 1 = no symptoms; 2 = leaf symptoms, no stunt; 3 = symptoms plus stunt (general plant size) ranging from slightly discernible to about 15%; 4 = symptoms plus stunt of about 15–50%; 5 = symptoms plus stunt greater than 50%.

56(E)) segregated at a 3:1 S/R ratio. RMP 12 × M1204.78I had a ratio of 1:3 S/R plants. All chi-square values were within the acceptable probability limit for the models tested (Table 2).

No seeds were available in 1989 to evaluate F_1 and backcross plants. Segregation evaluation of F_3 families was incomplete because very few seeds from individual F_2 plants, particularly those with ratings of 3–5 (16), were available because of high disease incidence and severity. However, families from plants rated 1 and 2 in crosses (RMP 12 × RG 1, RMP 12 × M1204.78I, RMP 12 × 55-437, RMP 12 × ICGS-56(E), RMP 12 × MK 374, RG 1 × M1204.78I, RG 1 × RRB, RG 1 × ICGS-56(E), and RG 1 × MK 374) of the F_2 generation were tested by mechanical inoculation with GRV. Even after four repeated inoculations, all 115 and 253 progeny from plants rated 1 and 2, respectively, were symptomless (resistant). F 452.4, the susceptible control, had 48 of 48 plants with a susceptible reaction (one inoculation only).

Mechanical inoculation. When peanut plants were mechanically inoculated with GRV in the greenhouse, patterns of disease incidence developed that were similar to those observed in the field in 1989 where plants were infected with both GRV and GRAV (Fig. 3). First symptoms appeared as early as 3 days after inoculation on the susceptible crosses and as late as 10 days for the R × R cross (Fig. 3). When 100% infection was observed on plants of all S × S crosses 12 days after inoculation, only 6, 76, and 90% of the R × R, RMP 12 × M1204.78I, and R × S and S × R crosses, respectively, were infected. As in the 1988 and 1989 field experiments, four rather distinct disease progress curves were observed in GRV-infected plants (Fig. 3). However, the RMP 12 × M1204.78I curve was much closer to the R × S curve than the R × R curve, which was different from the field studies.

Most plants (average of 75%) of the resistant parents developed leaf symptoms when inoculated with GRV, but they could be distinguished from susceptible plants by their degree of stunting (disease rating) (Table 3). Only 12% of the F_2 plants of the R × R cross developed symptoms.

In the R × S and S × R crosses, 74% of the M1204.78I × RMP 12 cross were resistant (based on disease ratings), whereas the other four crosses had an average of 6% plants showing resistance (Table 3). All plants from the four S × S crosses were susceptible with stunting greater than 30%. The resistant parents, the R × R cross, and M1204.78I × RMP 12 had low disease indices, whereas indices of the other R × S and S × R crosses were almost twice those of resistant parents. The S × S crosses had disease index values greater than 4.0.

Populations of four of five R × S and S × R crosses mechanically inoculated with GRV segregated at a ratio of 15:1 S/R (Table 4). The fifth one, M1204.78I × RMP 12 (reciprocal of the cross used in field studies), was again an exception with a ratio of 1:3 S/R. All plants of four S × S were susceptible, and 96% of the plants of the R × R cross were resistant.

Diagnostic tests. No 900-bp dsRNA was found in six of six plants from various crosses with a disease rating of 1. However, dsRNA was found in two of six plants rated 2 and 10 of 10 plants rated 5. Therefore, the dsRNA associated with GRV could be detected in some plants with mild leaf symptoms.

GRAV antigen was detected by ELISA in 11 of 15 symptomless plants (disease rating of 1) of the R × R and RMP 12 × M1204.78I crosses. It was also found in seven of eight plants, from R × S crosses, with a disease rating of 2.

DISCUSSION

Genetic variability for resistance to the green form of groundnut rosette in peanut was found among all F_2 populations of R × S and S × R crosses in this study. It is at this generation that maximum expression of genetic makeup is observed in progeny produced from homozygous parents. Three independent experiments, each providing different types of information, led us to conclude that one specific type of resistance in RMP 12 and RG 1 is controlled qualitatively by two recessive genes. In the first experiment conducted in the field in 1988, crosses were tested against a mixed natural infection of GRV and GRAV. Under moderate to severe disease conditions, all F_2 plants of the R × R cross remained disease-free, indicating a high level of resistance. Segregation patterns of R × S and S × R F_2 populations were largely ignored because not all plants of susceptible parents became infected. In the second field experiment in 1989, disease conditions were extremely severe and four R × S crosses segregated at a 15:1 S/R ratio. In the third experiment conducted with plants mechanically inoculated with GRV, results were consistent for a 15:1 S/R ratio for four of five crosses of resistant and susceptible parents.

In all three experiments, the R × R cross did not show segregation in the F_2 generation. In the 1988 field experiments, none of the F_2 plants from this cross developed symptoms, whereas in 1989, when extreme disease conditions occurred, many plants developed mild leaf symptoms. However, only a very few were stunted, indicating an excellent type of resistance in the parents. In the mechanical inoculation experiment, mild leaf symptoms were observed on about 12% of the F_2 progeny of the R × R cross.

In the 1989 field test, one S × S cross (ICGS-56(E) × JL 24) and three crosses of resistant and susceptible parents (RRB × RMP 12, RG 1 × M1204.78I, and RMP 12 × ICGS-56(E)) had more resistant plants in their F_2 populations than was expected for control of resistance by double recessive genes, even though 100% of 3,800 susceptible plants (parents and infector rows) developed a susceptible reaction. An obvious explanation is that a few susceptible plants escaped infection or developed symptoms more than 27 days after exposure to inoculum. In fact, the reciprocal of the RMP 12 × ICGS-56(E) cross had the expected 15:1 S/R ratio in the mechanical inoculation test. It should be noted, however, that all four crosses have either one or both parents of RMP 12 and ICGS-56(E).

An unexpected F_2 segregation pattern was observed for the RMP 12 × M1204.78I cross (and its reciprocal) in which ratios were 1:5, 1:3, and 1:3 S/R in the 1988 field test, the 1989 field test, and the mechanical inoculation test, respectively. As a parent, M1204.78I reacted similarly to other susceptible parents (16), and the predominance of resistant plants was not observed when M1204.78I was crossed with resistant RG 1. Impure breeding lines could explain the phenomenon. However, selection of the parental lines was done carefully, there was no evidence of heterogeneity with regard to disease reaction in the parents (14,16), and the same phenotypic responses occurred with seeds from a second cross, M1204.78I × RMP 12. Additional studies are required to confirm and understand these results.

Although qualitative inheritance studies are often complicated by cytoplasmic and/or maternal effects (20), no such factors were observed in these studies. F_2 populations of three R × S crosses segregated like the reciprocal crosses. A fourth reciprocal cross, RRB × RMP 12, segregated 13:3 instead of 15:1 S/R; the 13:3 ratio could have been affected by a low population number of a few susceptible plants not becoming infected under field conditions.

Confirmation of genetic patterns for most traits normally depends on the disease reaction of plants of backcrosses and/or F_3 families. Unfortunately, in our study there were few seeds from the backcrosses, and all were used in the 1988 field study when genetic analysis was not possible because too many susceptible plants did not become infected. In

Table 4. Reaction to groundnut rosette in parental and F_2 populations mechanically inoculated with groundnut rosette virus (green strain) in the greenhouse

Genotype	Phenotype[a]	Total no. of plants	Susceptible reaction (%)		Best fitting model (S/R)	χ^2	Probability
			Observed	Expected			
F 452.4	S	60	100.0	100.00			
Resistant parents	R	33	3.0	0.00			
RMP 12 × RG 1	R × R	48	4.2	0.00	0:1	0.0833	0.80–0.70
M 1204.78I × JL 24	S × S	47	100.0	100.00	1:0	0.0000	
JL 24 × RRB	S × S	111	100.0	100.00	1:0	0.0000	
RRB × ICGS-56(E)	S × S	104	100.0	100.00	1:0	0.0000	
JL 24 × MK 374	S × S	95	100.0	100.00	1:0	0.0000	
RMP 12 × RRB	R × S	39	92.3	93.75	15:1	0.1385	0.80–0.70
RMP 12 × JL 24	R × S	74	94.6	93.75	15:1	0.0901	0.80–0.70
ICGS-56(E) × RMP 12	S × R	113	92.9	93.75	15:1	0.1327	0.80–0.70
55-437 × RMP 12	S × R	52	94.2	93.75	15:1	0.0205	0.95–0.90
M1204.78I × RMP 12	S × R	102	26.5	93.75	1:3	0.1176	0.80–0.70

[a] R = resistant, S = susceptible.

1988, there were 781 symptomless F_2 plants of both R × S (and S × R) and S × S crosses, and the task would have been too burdensome to try to determine which families were segregating and which were not, especially with the uncertainty of achieving 100% infection. In 1989, susceptible plants of all crosses were severely diseased and too few seeds produced per F_2 plant for segregation tests of F_3 families. However, adequate seed numbers were available for F_2 plants that were rated resistant, and no segregation was observed in these families after mechanical inoculation with GRV, thus confirming resistance in the selections. Their behavior under mixed infections (GRV and GRAV) has not been tested, but we believe it should be similar to that of GRV alone. The complete resistance of families from these F_2 plants also supports the validity of the rating system as a means to separate susceptible and resistant plants in segregating populations.

Previous studies of inheritance of resistance to groundnut rosette have been reported by de Berchoux (6) and Nigam and Bock (11). They worked with chlorotic rosette and found that resistance was controlled by two recessive genes, similar to our studies with green rosette. Therefore, it appears that the two recessive genes control both forms of groundnut rosette. Neither previous report indicated any deviation from the F_2 phenotypic ratio of 15:1 S/R, such as the unexpected ratios with the RMP 12 × M1204.78I cross and crosses involving ICGS-56(E). However, our studies used more parents, which could have represented a larger gene pool, including genes that interact differently with the GRV strain causing green rosette than with the GRV strain causing chlorotic rosette.

This is the first attempt to evaluate patterns of inheritance of resistance to a single infection of GRV. It has been known since the studies of Hull and Adams (7) that GRV infection (plus its satellite RNA) is primarily responsible for the rosette symptoms. GRAV can infect both susceptible and resistant genotypes (2,14,16), but it causes no symptoms in either. However, GRAV does appear to play some role in rosette disease development because in a mixed infection, symptoms can be intensified (10) and stunting and yield reduction (1) are greater than with a single infection of GRV (plus satellite RNA). The mechanical inoculation procedure (15) proved to be highly effective in separating resistant and susceptible plants, whereas optimal field conditions for screening are difficult to control. F_2 segregation patterns were similar to and less equivocal than those in field studies with a mixed infection of GRV and GRAV. Furthermore, avoiding the complications of mixed infections with GRV and GRAV seems highly appropriate for breeding programs.

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