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Screening Methods and Further Sources of Resistance to Peanut Rust[1]

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ABSTRACT

A germplasm collection of 6000 peanut entries was screened for resistance to rust at ICRISAT, India. Preliminary field screening was done during the 1977 rainy season when a natural epidemic of rust was in progress. The cultivars or lines which were rated between 2 and 5 on a 9-point scale during this screening were further tested during the 1977/78 dry season employing an infector row system of susceptible cultivars and spreader plants systematically interplanted with the test material. High relative humidity was maintained in the field by operating an overhead sprinkler irrigation system. Percentage leaf area damaged on the test material was estimated at 10 day intervals from approximately 90 days after their emergence until harvest. Each entry was also assessed on a scale proposed by Mazzani and Hinojosa. Two land races, NC.Ac. 17090 and EC. 76446 (292) were more resistant than either PI. 259747 or PI. 298115 which were reported resistant by other workers. In addition, NC.Ac. 17030, NC.Ac. 17132, NC.Ac. 17129, NC. Ac. 17135 and NC.Ac. 17124 were moderately resistant. Four cultivars or lines with different levels of resistance in the field were tested in the greenhouse at three different stages in development. The results indicated that resistance increased as the plants aged.

Key Words: *Arachis hypogaea* L., *Puccinia arachidis* Speg., disease resistance, peanut rust, peanut germplasm.

Peanut rust, caused by *Puccinia arachidis* Speg., has become of increasing economic importance over the last few years. Prior to 1969 rust was largely confined to South America and the Caribbean, with occasional outbreaks occurring in the southern most peanut-producing areas of the United States. Since 1969 rust has been reported in all major peanut-producing areas of the world according to Hammons (3) and Subrahmanyam *et al.* (8).

Various methods have been used to assess the reaction of cultivars and species to rust. Kenknight (6) screened cultivars under natural field conditions and by artificial inoculation with uredospores. Bromfield and Cevario (1) used uredospores, previously stored in a liquid nitrogen refrigerator, to inoculate 4 to 5 week-old greenhouse grown peanut plants. After inoculation the plants were either transferred to dew chambers for 16 to 20 hours or were covered with polyethylene sheeting and misted overnight. Bromfield and Cevario (1) also used detached leaflets to assess rust damage and they observed similar reactions to those obtained from leaves of intact plants. Cook (2) screened material in the greenhouse and in the field.

Previously reported sources of resistance to rust are summarized in Table 1. These sources include cultivars, natural hybrids, and wild *Arachis* species. Two species, *Arachis glabrata* Benth., and *A. villosulicarpa* Hoehne, were rated as immune by Bromfield and Cevario (1) and Hammons (3) respectively.

In this paper the results of field and greenhouse screening are reported.

Table 1. Reported sources of peanut rust resistance (Bromfield and Cevario, 1970: Hammons, 1977)

Identification	PI Number	Botanical type	Origin
Tarapoto	259747, 341879, 350680, 381622, 405132	Valencia	Tarapoto, Peru
Israel Line 136	298115, 315608	Virginia	Introduction to Israel from USA
DHT 200	314817	Valencia	Juanji, Peru
FESR 1-14	Hybrids between 298115 and unknown pollen donor	Segregating	Puerto Rico (USDA rust nursery)
A. glabrata	118457, 231318, 262287	Section *Rhizomatosae*	Brazil
	262141*	"	Bolivia
	262801	"	Argentina
A. villosulicarpa	336985	Section *Extranervosae*	Brazil
*A. monticola***	263393	Section *Arachis*	Brazil

*Bromfield and Cevario (1970) give this PI number for *A. glabrata*. However, according to Gregory *et al.* (1973) PI. 262141 is *A. cardenasii* in section *Arachis*. pp. 98 in *Peanuts - culture and uses*, American Peanut Research and Education Association, Inc.

**Small, weakly sporulating pustules only--but another accession of *A. monticola* (PI 405933) was killed by rust at Tifton, Georgia.

Materials and Methods

Seeds were obtained from the ICRISAT germplasm collection.

Preliminary field screening in 1977-rainfed crop:

During the 1977 rainy season a peanut germplasm collection of 6000 entries was screened at ICRISAT center which is situated some 25 km northwest of Hyderabad in the Indian state of Andhra Pradesh. The entries were unreplicated but systematic checks of a rust susceptible cultivar TMV2 (Spanish) were used. Each germplasm entry, consisting of at least two 5 m rows, was scored prior to harvest on a 9-point field scale (1 = free from rust and 9= 50 to 100% defoliation caused by rust).

Advanced field screening - 1977/78 irrigated crop:

During the 1977/78 dry season a field-inoculation technique was developed on the irrigated crop using previously collected uredospores which had been stored at -15°C for about 3 months. Two susceptible cultivars, TMV2 and Robut 33-1, with different maturity dates were sown systematically throughout a 2 ha field as infector rows some 14 days in advance of the test

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material. The planting pattern was an infector row, two test rows and then another infector row. The rows were 75 cm apart and within-row spacing was 20 cm. Susceptible cultivar (TMV2 and Tifspan) check plots were planted to assess the spread of rust from the infector rows. At peak flowering the infector rows were inoculated with uredospore suspensions (approx. 50,000 spores/ml) in tap water containing a wetting agent (Tween 80). The inoculations were made at approximately 1700 hr. after the field had been furrow irrigated. Subsequently the field was irrigated with overhead sprinklers, on alternate days initially, and then at irregular intervals until harvest. Potted "spreader plants", already heavily infected with rust, were also placed systematically throughout the field to act as additional sources of inoculum.

Approximately 90 days after emergence, 20 to 25 plants of each entry were sampled by taking one leaf each from the lower, middle, and upper parts of the plant. These samples were combined and scored for percentage of leaf area damaged by rust. Subsequent samples were collected at approximately 10 day intervals until harvest. Percentage leaf area damaged by rust was estimated with an "intensity grade scale" similar to the method described by Hassan and Beute (4) for assessing leafspot (*Cercospora arachidicola* Hori.) damage to peanuts. Each entry was also assessed on a scale proposed by Mazzani and Hinojosa (1961) as follows:

R_0 = no leaves heavily infected
R_1 = less than 25% leaves heavily infected
R_2 = 25 to 50% leaves heavily infected
R_3 = 50 to 75% leaves heavily infected
R_4 = more than 75% leaves heavily infected

A further rating was done by using a modified Cobb's scale from 0 to 5 (where 0 = no infection and 5 = more than 51% leaf area affected).

Greenhouse screening:

(a) Whole Plants

Plants were raised in 15 cm diameter pots and inoculated with a uredospore suspension (as previously described) at different physiological stages of growth i. e. at the four-leaf stage, at peak flowering, and nearing maturity. After inoculation the plants were kept in dew chambers for 24 hours at approximately 24-34°C inside the greenhouse. Each set of test plants included a susceptible check cultivar (TMV2). The percentage leaf area damaged by rust was assessed on a total plant basis 30 days after inoculation (except for new leaves which had subsequently emerged). Ratings were also made, at the same time, using the scale proposed by Mazzani and Hinojosa (7).

(b) Detached Leaves

Fully expanded leaves from the lower, middle, and upper portions of the plant at the 4 leaf, at peak flowering, and nearing maturity stages were detached and placed with their petioles immersed in glass vials of a nutrient solution (5) supplemented with 20 ppm. Kinetin. The leaves were inoculated and incubated as described for the whole-plant method.

Results and Discussion

Preliminary field screening 1977 rainfed crop:

During the 1977 rainy season a heavy natural infestation of rust occurred. The entries selected as promising sources of resistance are shown in Table 2. All entries were rated between 2 and 5 on the 9 point scale. Two entries Tarapoto (PI. 259747) and Israel Line No. 136 (PI. 298115), previously reported as resistant were not included in this screening because of a shortage of seed.

Advanced field screening 1977/78 irrigated crop:

All the promising entries previously selected, plus Pl 259747 and Pl 298115, were screened during this season. Considerable rust occurred on the artificially inoculated infector rows. Rust spread from the infector rows to the indicator rows of the susceptible TMV 2 and Tifspan cultivars was successful and typical susceptible reactions were obtained.

Table 2. Rust reactions of 11 peanut cultivars in field screening trial at ICRISAT, Hyderabad

Cultivar	Source	Seed color	Preliminary field Screening 1977 rainy season	Advanced field screening 1977/78 dry season		
			9-point field scale	9-Point field scale	Modified Cobb's scale	Mazzani & Hinojosa scale
NC Ac 17090*	Peru	Tan	2	2	1	R_0
EC 76446 (292)	Uganda	Purple	3	3	1	R_0
Pl 259747	Peru	"	-	3	2	R_1
Pl 298115	Israel	Pale Tan	-	4	3	R_2
NC Ac 17129	Peru	Tan	4	4	3	R_2
NC Ac 17130	Peru	Tan	4	4	3	R_2
NC Ac 17132	Peru	Purple	4	4	3	R_2
NC Ac 17135	Peru	Purple	4	4	3	R_2
NC Ac 17124	Peru	Tan/ Purple var.	4	4	3	R_2
TMV 2	India		9	9	5	R_4
Tifspan	USA		-	9	5	R_4

*NC Ac. numbers are germplasm lines received from North Carolina State University

The percentage leaf area affected by rust increased with plant age, but the reaction varied from entry to entry (Fig. 1). Two land races, NC Ac 17090 (originally collected in Peru by Dr. W. C. Gregory) and EC 76446 (292) (received from Uganda by the National Bureau of Plant introduction of India in 1969), were more resistant than either Tarapota (PI 259747) or Israel Line 136 (Pl 298115) when scored on a modified Cobb's scale. On the Mazzani and Himojosa (1961) scale they were rated R_0. Tarapoto was rated as resistant (R_1) and Pl 298115 as moderately resistant (R_2). In addition, five other land races (NC Ac. 17130, NC Ac. 17132, NC Ac. 17129, NC Ac. 17135 and NC Ac. 17124) were rated as moderately resistant (R_2).

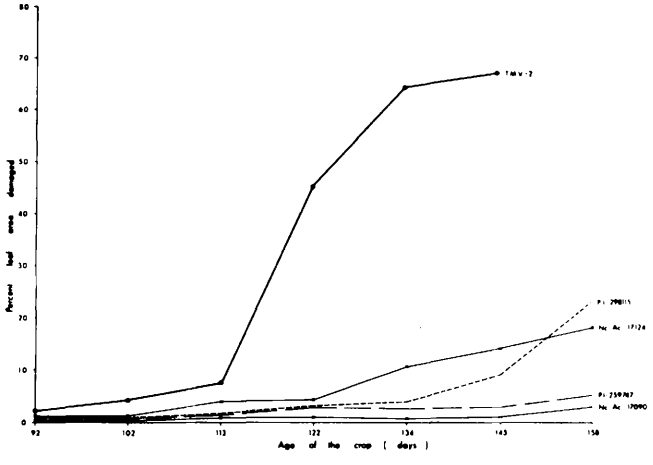

Fig. 1. Leaf area damage at different growth stages.

In the field, susceptible cultivars produced numerous large sori on the lower surface of the leaflet which were elevated and sporulated profusely. A colony of sori developed around the original sorus and the leaflet turned yellow and abscissed. On NC Ac. 17090 and EC 76446 (292) the sori were small, few in number, and without necrotic areas; sporulation was sparse and sori on older leaves often remained somewhat depressed and unruptured with practically no defoliation. On the other resistant and moderately resistant entries sori were again weakly sporulating but a zone of necrotic tissue varying in color from reddish-brown to purplish-black developed around the sori. Colonies of sori did not develop around the original infection site as described in susceptible cultivars.

Greenhouse screening:

Four land races or lines (NC Ac. 17090, PI 259747, NC Ac. 17129 and TMV2) showing disease reactions varying from resistant to susceptible in the field were selected for whole plant testing in the greenhouse. Plants inoculated at the seedling stage or at the peak flowering stage develped more rust by 30 days after inoculation than plants inoculated at maturity (Table 3). However, NC Ac. 17090 showed very little damage even when inoculated at the seedling and flowering stages as compared with other entries. The entry Pl 259747, which was identified as resistant in the field, showed 50.83% leaf area damaged 30 days after inoculation at the seedling stage, 30.83% damage when inoculated at the flowering stage, but only 2.86% when the mature plants were inoculated. Cook (2) suggested that the decline in susceptibility to infection was associated with a corresponding decrease in leaf wettability. It appears that resistance is physiological, as already reported by Bromfield and Cevario (1) and Cook (2).

Hassan and Beute (4) have reported that greenhouse grown plants produced more leafspot lesions in the greenhouse than plants raised in the field and subsequently transferred into the greenhouse. It seems important, therefore, to test material both in the greenhouse and under field conditions, as greenhouse results alone may not reveal field resistance. Results obtained from detached leaves in the greenhouse were similar to those obtained from whole plants, which confirms the observations made by Bromfield and Cevario (1).

Table 3. Rust reactions of four peanut cultivars 30 days after inoculation at three physiological stages of development in the greenhouse.

	Per cent leaf area damaged by rust (mean of five plants)		
	Plant stage at inoculation		
	Seedling	Peak Flowering	Nearing Maturity
NC Ac 17090	4.00	6.46	2.83
NC Ac 17129	26.66	38.05	5.91
Pl 259747	50.83	30.83	2.86
TMV 2	100.00	85.50	41.11

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