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Rosette Disease of Groundnut—Transmission Studies

by

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Introduction: The occurrence of a virus disease of groundnut in the Madras State was first reported by Sundararaman (1926) at Palur Agricultural Experimental Station and he named it as "clump disease". As very little is known about the various aspects like transmission, mode of spread, influence of various agronomic practices like manuring, spacing, roguing, weeding *etc.* on the incidence of rosette disease occurring in Madras State, a scheme was initiated at the Agricultural College and Research Institute, Coimbatore with the financial assistance of Indian Central Oilseeds Committee. The pattern of spread of rosette disease and the assessment of crop loss due to rosette disease were dealt in the previous two papers (1965 & 1967). This paper deals with the different modes of transmission of the disease.

Materials and Methods: Rosette disease culture obtained from Coimbatore location was used in these studies. The culture was maintained by using *Aphis craccivora*, as vector under insect-proof glass house conditions. All transmission studies were carried out under insect-proof glass house conditions. TMV 2, a bunch variety of groundnut, was selected for these studies.

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I. *Graft transmission:* Approach and side grafting methods were followed. Fortyfive days old healthy groundnut plants were used in the studies. Scions were obtained from rosette diseased groundnut plants and grafted on to healthy plants. One hundred plants were used for each method of grafting.

II. *Sap transmission:* Fifteen days old healthy groundnut plants were selected for the study. Sap was extracted from freshly infected young leaves at the rate of one gram of the material in 3 c.c. of the following solutions, namely, 0.1 M dipotassium phosphate solution at pH 7.0, 0 5 per cent sodium sulphite solution and 0.1 M potassium dihydrogen phosphate solution. The infective sap was filtered through a fine muslin cloth and the filtrate was used for transmission studies. Three methods, namely, rubbing, pin prick and brush inoculation were tried separately in sap transmission studies. Fifty seedlings were taken for each type of sap extraction in each method. Suitable controls were maintained for each method. Carborundum (600 mesh) was used as an abrasive and dusted on to the young unfolded leaves (2 to 3 in number) prior to inoculation.

In another method, eight hundred one week old plants were selected for the study. A set of 400 plants were etiolated by keeping them in darkness for 48 hours and another set of 400 by keeping them in darkness for 96 hours. The infective sap was rubbed on to the carborundum dusted young leaves of etiolated plants in each set. The inoculated leaves of half the number of plants in each set were washed with distilled water immediately after inoculation and those of the other half in each set were left unwashed. Suitable controls were maintained for each set.

III. *Insect transmission:* a. *Aphids: Aphis craccivora* normally found on healthy groundnut plants were collected and a colony of virus free aphids was raised from a single aphid. Adult apterious form of the aphids were used in transmission studies. Fifteen days old healthy groundnut seedlings were selected as test plants. The aphids were allowed to have a preacquisition fasting period of 24 hours and test feeding period of 24 hours on healthy groundnut plants. The acquisition feeding period on rosettee diseased plants varied from 24 hours to 96 hours in different methods. Ten viruliferous aphids were allowed to feed on each test plant for 24 hours and they were then killed by spraying with Folidol 0.025%. Fine camel hair brush was used for the transfer of aphids from plant to plant.

a) In one method, the aphids were allowed to have an acquisition feeding period of 24 hours on rosette diseased groundnut plants.

b. *Half seed method of transmission:* In another method, the aphids were allowed to have acquisition feeding periods of 24, 48, 72 and 96 hours.

Groundnut seeds were allowed to germinate for 3 days in petri-dishes. On the 4th day, one of the cotyledons was removed from each seedling. Later, these halved seedlings were used for insect transmission studies.

A.craccivora collected from healthy *Gliricidia maculata* plants were allowed to have a pre-acquisition fasting period of 24 hours, acquisition feeding period of 48 hours on rosette diseased plants and test feeding period of 24 hours on healthy ground seedlings. *A.craccivora* normally found on healthy *G.maculata* plants were collected and a colony of virus free aphids was raised from a single aphid on healthy groundnut plants. These aphids were then allowed to have a pre-acquisition fasting period of 24 hours, acquisition feeding period of 48 hours on rosette diseased plants and test feeding period of 24 hours on healthy groundnut seedlings.

2. *Jassids: Amrasca devastans* collected from healthy groundnut plants was used in transmission studies. These Jassids were allowed to have a pre-acquisition fasting period of 2½ hours, acquisition feeding period of 24 hours on rosette diseased plants and test feeding period of 24 hours on 15 days old healthy groundnut seedlings after which the jassids were killed by spraying with Folidol 0.025%. Twenty five healthy groundnut seedlings were selected for the study.

IV. *Seed transmission:* Five hundred plants were raised from seeds collected from each of healthy and rosette diseased plants respectively. The plants were kept under observation till they were 120 days old.

V. *Soil transmission:* Soil was collected round about rosette diseased and healthy groundnut plants. Two hundred plants were raised in each type of soil. The plants were observed for the incidence of the disease till they were 120 days.

VI. *Dodder (Cuscuta reflexa) transmission: Cuscuta reflexa* plants were allowed to trail on rosette diseased groundnut plants till they have established well on them. Then the tips of these parasites were allowed to trail on 30 days old healthy groundnut plants. The test plants were under observation for 90 days.

Results: 1. *Graft transmission:* Ninety and eighty per cent complete graft union was obtained in approach and side grafting methods, respectively. One hundred plants were used for transmission of the virus in each grafting method. The results are given in the table below :

Grafting method	Number in which there was successful graft union	Number infected	Percentage infected
Approach grafting	90	90	90
Side grafting	80	80	80

Hundred per cent success was obtained in the transmission of disease by grafting wherever graft union was successful.

II. *Sap transmission:* Rosette disease was not found to be transmitted to healthy groundnut plants by any of the sap transmission methods tried, *viz.*, rubbing, pin prick and brush inoculation methods though there was a very negligib'e percentage of success (2%) when 0 5% Sodium sulphite buffer was used.

In another experiment, a set of 400 plants were etiolated for 48 hours and another set for 96 hours. The young unfolded leaves (2–3 numbers) of these plants were inoculated with the infective sap extracted in 0.1 M potassium dihydrogen phosphate solution or in 0.5% sodium sulphite solution as per the method already described. Two per cent infection was obtained in the case of plants etiolated for 48 hours when inoculation was made with infective sap extracted in 0.5% sodium sulphite solution and not washed with distilled water subsequent to inoculation.

III. *Insect transmission:* 1. *Aphid (A. craccivora):* a) In insect transmission studies, the aphids were allowed to have 24 hours each of pre-acquisition feeding period and test feeding period. Fifteen out of fifty plants inoculated were found to be infected. Thirty per cent infection was thus obtained by this method.

b) *Half seed method of transmission:* The experiment was conducted as per the method already described. The acquisition feeding periods alone were varied, namely, 24, 48, 72 and 96 hours. The results are given in the following table :

Acquisition feeding period	Number of plants inoculated	Number infected	Percentage infected
24 hours	20	9	30
48 hours	20	8	40
72 hours	20	8	40
96 hours	20	8	40

The results in the table above show the maximum infection of 40 % was obtained in 48, 72 and 96 hours of acquisition feeding periods. Thirty per cent infection was obtained when the acquisition feeding period was 24 hours.

None of *A. craccivora* collected directly from *Gliricidia maculata* plants, transmitted the rosette disease to healthy groundnut plants. In another experiment, *A. craccivora* collected from *G. maculata* plants, were allowed to

breed on healthy groundnut plants. Then these aphids were allowed to have a pre-acquisition fasting period of 24 hours, acquisition feeding period of 48 hours and test feeding period of 24 hours. Twenty out of fifty plants inoculated by this method gave positive infection. The percentage of infection was 40.

2. *Jassids: (Amrasca devastans):* Negative results were obtained in the transmission of rosette disease of groundnut by using *Amrasca devastans*, as vector.

IV. *Seed transmission:* None of the plants raised from seeds collected from rosette diseased plants showed symptoms of the disease.

V. *Soil transmission:* The symptoms of the rosette disease were not seen in any one of the plants raised in soil collected round about rosette diseased plants.

VI. *Dodder (Cuscuta reflexa) transmission:* No transmission was evident when *Cuscuta reflexa* was used in the transmission of rosette disease.

Discussion and Conclusion: There was hundred per cent success in the transmission of rosette disease of groundnut whenever graft union was complete irrespective of the method of grafting employed. The symptoms of rosette disease appeared in 15–40 days. Storey and Bottomley (1928) reported that the disease was readily transmitted by grafting and the symptoms were produced within 26–60 days from the date of grafting. Brunt and Bonney (1964) transmitted the disease by cleft grafting and stated that six of the seven in which graft union was successful showed typical leaf symptoms within six weeks.

Rosette disease was not transmitted to healthy groundnut plants in sap inoculation studies by any of the methods tried. This result is in confirmity with the findings of other workers. Sundararaman (1931) reported that sap transmission of the disease by pin-prick method was not successful. Storey and Bottomley (1928) could not transmit the disease by mechanical inoculation of the infective juice. Storey and Ryland (1955) failed to transmit the disease by inoculating the juice from diseased leaves into normal green plants with or without carborundum or celite as an inoculation aid.

Two percent success was obtained when one week old seedlings etiolated for 48 hours (by keeping in darkness) were inoculated with the infective sap extracted in 0.5% sodium sulphite solution and without washing the inoculated leaves in distilled water. Storey and Ryland (1955) obtained 10.6 % infection by using celite and etiolated plants. Bawden (1951) infected 10% of his plants by this method. Brunt and Bonney (1964) reported that they could get 32.2%

infection in mechanical inoculation studies by inoculating infective sap with the addition of Hyflosupercel celite to young healthy etiolated groundnut plants. According to them the susceptibility of groundnut seedlings was increased by keeping in darkness.

A. craccivora is the vector of groundnut rosette virus and 30% infection was obtained by using this aphid as a vector. The symptoms of the disease appeared in 26–48 days. Storey and Bottomley (1928) first proved that *A. laburni* was the vector of groundnut rosette virus in South Africa. Brunt and Bonney (1964) stated that the groundnut rosette virus was redily transmitted from naturally infected plants to healthy groundnut seedlings by using 15–20 aphids (*A. craccivora*). They also reported that quite high transmission rates (around 50%) can be obtained even when using single aphid. The symptoms of the disease appeared in 16-40 days later. Sundararaman (1931) could not transmit the disease by using *A. medicagenis* (*A. craccivora*) collected from infected plants. *A. craccivora* Koch. (*A. leguminosae*. Theo.) is the only insect so far shown to transmit the disease (Storey and Bottomley, 1928).

In insect transmission studies conducted by using *A. craccivora* as a vector and by using half seed method, acquisition feeding periods of 48, 72 and 96 hours were found to yield maximum infection of 40%. Vanderveken (1961) found that the minimum feeding period required for the aphid was 48 hours and he could occasionally get transmission with one hour feeding period. In the present study even a feeding period of 24 hours has given 30% infection.

Rosette disease could not be transmitted to groundnut by using *A. craccivora* collected directly from healthy *Gliricidia maculata* plants. But 40% success was obtained by using *A. craccivora* collected from *G. maculata* plants and then bred on healthy groundnut plants. This is in confirmity with the findings of Davies and Kasule (1964).

Rosette disease was not transmitted by *Amrasca devastans*. Failure of transmission of this disease by leaf hoppers has also been reported by Storey and Bottomley (1928).

Rosette disease was found to be neither seed borne nor soil borne. This is in confirmity with the findings of Sundararaman (1931), Storey and Bottomley (1928) and Hayes (1932).

No transmission was evident when *Cuscuta reflexa* was used in the transmission of rosette virus.

Summary: Rosette disease of groundnut collected from Coimbatore is readily graft transmissible. It is transmitted by the aphid, *Aphis craccivora* and not by the jassid, *Amrasca devastans*. 2% success was obtained on etiolated one

week old seedlings in sap transmission studies by using 0.5% sodium sulphite solution and carborundum powder. It is neither seed borne nor soil borne. The disease is also not transmitted by dodder, *Cuscuta reflexa*.

Acknowledgements: Grateful acknowledgements are due to Dr. K. Ramakrishnan, Dean, Agricultural College, Coimbatore, Dr. C. V. Govindaswamy, Dean, Agricultural College, Madurai and Thirumathi Soumini Rajagopalan, Plant Pathologist AICRIP, Coimbatore for their valuable guidance, suggestions and encouragement during this investigation and in the preparation of this paper. Thanks are also due to the Indian Central Oilseeds Committee as the studies were conducted during a tenure of a grant from this authority.

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