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A CASE FOR INDUCED MUTATION IN CHICKPEA

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### POR ASCOCHYTA BLIGHT RESISTANCE

M.V. Reddy and Y.L. Nene

International Crops Research Institute for the Semi-Arid Tropics, (ICRISAT), ICRISAT Patancheru P.O., Andhra Pradesh 502 324, India.

Chickpea (Cicer arietinum L.) is an important grain legume grown in several countries. Blight caused by Ascochyta rabiei (Pass) Labrousse is widespread and results in considerable losses in several countries (2, 3, 5, 8, 11, 12, 14, 15, 17, 19, 22, 26, 29, 30, 31, 33, 35, 36, 38, and 40). Extensive efforts have been made by earlier workers to identify resistance sources and minimize losses through the development of resistant/tolerant varieties (1, 4, 6, 7, 9, 13, 16, 20 21, 23, 25, 28, 34, 36, and 38). Most of the screening work has been carried out in fields, either in areas where natural epiphytotics occur or through artificial field inoculations in areas where the disease is not always severe (13, 16, 24, 25, 32, and 37).

Chickpea is one of the crops on which ICRISAT works. We believe that the role of pathologists in the crop improvement program is to help the breeders in identifying stable sources of disease resistance and evolving genotypes in which such a resistance is incorporated. To start with, we found a great need for a rapid and effective laboratory screening procedure. We developed such a procedure, and used it to screen the lines reported to be resistant. When none of these was found resistant, the systematic screening of the germplasm available at ICRISAT (over 11000) was undertaken to identify good resistance sources. So far over 3500 germplasm accessions of Cicer species representing highly varied collections have been screened, but none of the cultivated species of Cicer has been found resistant. It

induced to obtain more variability and possibly better sources of resistance.

## MATERIALS AND METHODS

## Screening apparatus

Three Isolation Plant Propagators obtained from Burkard Manufacturing Company Ltd., Rickmansworth, Herts, England, were used (18). The propagators were located in a screenhouse. Mhen necessary, three evaporative coolers per propagator were operated to keep the temperatures below 30°C. Additional artificial light (four 40 W, 1.2-m- long cool daylight tubes per chamber) was provided whenever necessary to enable normal growth of the seedlings in the lower chambers of the propagator.

Fungus cultures, multiplication, and inoculation

Two isolates of the fungus differing in morphological and pathogenic characters were used. One was obtained from Dr. J.S. Grewal of the Indian Agricultural Research Institute (IARI), New Delhi, India, and the other was isolated from the diseased plants obtained from the Lahaul Valley in Himachal Pradesh state of India. The former is slower-growing, darker in color (due to heavy pycnidial formation), and more virulent than the latter. Cultures were maintained on potato-dextrose-agar slants. Multiplication was done in autoclaved (15 lb; 20 min) chickpea flour broth (40 g/L) incubated at 25°C with 12-hour light. Ten to 15-day-old cultures were used for inoculation. A spore suspension containing 20000 conidia/ml of water, required to kill a susceptible line (ICC-460) was used for inoculation. Inocu-

lation was done by spraying with Gamesh pneumatic hand sprayer (capacity 1L). On an average, each seedling received 1.5 ml of spore suspension.

# Gormplasm screening

For each germplasm accession, 10 seeds were planted in a single pot of the propagator. The pots were filled with a mixture of autoclaved (20 lb; 2 hours) sand and Vermiculite (5:1). Ten to 15day-old seedlings were inoculated. For each line, the time taken for symptom expression and number of plants infected and killed was recorded. The overall disease severity was rated on a 9-point scale two times; first when the susceptible check showed a 9 rating, and second 15 days after inoculation. The scale was adopted from Morrall and McKenzie (26) and is as follows: 1 - resistant (no lesions visible); 2 - resistant to moderately resistant (lesions on few plants, usually not visible); 3 - moderately resistant ( a few scattered lesions), usually seen after careful searching); 4 moderately resistant to tolerant (lesions and defoliation on some plants, not damaging); 5 - tolerant (lesions common and easily observed on all plants, but defoliation and/or damage not great); 6 - tolerant to moderately susceptible (lesions and defoliation common, killing few plants); 7 - moderately susceptible (lesions very common, damaging and killing 25% of plants); 8 - moderately susceptible to susceptible (extensive lesions on all plants, causis; defoliation and drying of branches and killing 50% of plants); 9 susceptible (lesions extensive on all plants, causing defoliation

and drying of branches and killing 75% of plants.).

# Multilocation testing

Field testing of the lines was facilitated through the International Chickpea Ascochyta Blight Nursery (ICABN) operated by us and cooperators in several countries.

Cultivars and germplasm accessions that repeatedly showed an average rating of 5 or less, and where enough seed was available, were sent for testing in endemic areas in India, Pakistam, Ethiopia, Syria, Iram, Tunisia, Turkey, and Algeria through ICABN. Some entries reported resistant by earlier workers were also included. During the 1977-78 season, 24 such entries were tested at 10 locations in the aforesaid countries. Each entry was grown in two single-row replications. Fifty seeds were planted per row. After every two test rows one of the two susceptible checks i.e., ICC-460 or ICC-4991, was grown. Artificial inoculations were carried out in some cases. A detailed report has been published earlier (27).

### RESULTS

Screening

## Cicer arietinum L.

A total of 3461 lines involving germplasm accessions and cultivars were screened in a 3-year period. None of the accessions and cultivars screened was resistant. Eighteen lines namely, ICC-120, 377 666, 693, 727, 780, 788, 867, 931, 964, 1203, 1273, 2173, 4552, 4716, 4934a, 4989, and 6067, were moderately resistant. One hundred and thirty-two lines were tolerant. The remaining 3311 lines showed

anderstely susceptible to susceptible reaction.

The lines that showed 3 to 5 ratings in the preliminary screening, and for which enough seed was available, were retested in the propagator in a replicated trial (three replications). Thirty-four ICC lines showed an average rating of 5 or less. These were ICC-120, 130, 204, 229, 377, 462, 468, 539, 559, 567, 595, 599, 600, 693, 703, 704, 724, 781, 816, 838, 931, 1009, 1465, 1911, 1915, 2153, 2156, 2160, 2237, 2600, 4716, 4939, 4989, and 6250. All the other lines except ICC-1908 showed a 7 or less rating. None of the lines showed a 9 rating.

# Wild Cicer species

Eight wild species of <u>Cicer</u> namely, <u>C. chorassanicum</u>, <u>C. pinnatifidum</u>, and <u>C. anatolicum</u>, were screened. <u>C. anatolicum</u> was resistant. <u>C. bijugum</u> showed a 3 rating. <u>C. cumeatum</u> and some collections of <u>C. judaicum</u> (Nos. 182 and 185) showed a 5 rating. Collections of <u>C. reticulatum</u> showed different reactions. Collection JM-2106 showed a 3 rating; JM-2106-A-1 showed a 4 rating, and No. 205 showed a 5 rating.

# International Chickpea Ascochyta Blight Nursery (ICABN)

Seventeen of the 24 ICC entries tested, ICC-280, 788, 1903, 4716
4934, 4935, 4939, 4989, 5006, 5127, 5784, 7513, 7514, 7520, and 7523,
showed a 5 rating or less at one or more locations. Two entries,
ICC-1903 and ICC-4935, showed a 3 or less rating at all five locations.
Two entries, ICC-5127 and ICC-7520, showed a 3 or less rating at four
locations. Three entries, ICC-4939, 7515, and 7514, showed a 3 or

less rating at three locations.

Reactions to the Lahaul isolate of lines that were found underately resistant to tolerant to IARI

Seventeen of the chickpea lines and two wild species (<u>C. reticulatum</u> and <u>C. anatolicum</u>) were tested against the Lahaul isolate.
Two chickpea lines, ICC-150 and ICC-229, showed 3 rating. Five ICC lines, 204, 272, 780, 788, and 4716 showed a 5 rating. Other lines showed 7 and above rating. Both the wild species were resistant.

#### DISCUSSION

The Isolation Plant Propagator was found to be an excellent tool for studies on blight all year around at Hyderabad, where the disease does not occur naturally. Even though the entire germplasm has not yet been screened, genotypes representing the genetic diversity have been tested. None of the 3461 accessions of <u>C. arietinum</u> screened was found resistant, indicating the difficulty in obtaining a high level of resistance in the germplasm.

Of the eight wild species screened, <u>C. anatolicum</u> was found resistant, but it is not crossable with cultivated chickpea. Also there is evidence pointing to the existence of physiological races of the blight pathogen (10, 39). The lack of a high-level of resistance in the existing germplasm and the presence of physiological races in the prthogen prompted us to suggest induced mutation in chickpea as a method to obtain more variability and possibly better sources of resistance. We would suggest that lines which already appear promising should be treated for obtaining mutations. In addition we will continue to screen the remaining germplasm for better resistance.

#### LITERATURE CITED

- AHMAD, T., S.Z. HASANAIN, and A. SATTAR. 19: Some popular method of plant disease control in Pakistan. Agr. Pakistan. 1: 18-22.
- 2. ANONYMOUS. 1938. Plant Diseases. Rep. Dep. Agr. Punjab, 52-56.
- ANONYMOUS. 1931. Plant Pathology Rapport sur le fonctionnement de 1' Inst. des Recherches Agron. pendant 1' annee 1930, 9: 411-445.
- ANONYMOUS, 1963. Quarterly Report for October December, 1962, of the Plant Protection Committee for the South East Asia and Pacific Region. F.A.O. Publ. 15 pp.
- ASKFROV, I.B. 1968. Ascochytosis of chickpea. Zashch. Rast., Mosk. 13(3): 52-53.
- AUJLA, S.S., and P.S. BEDI. 1967. Relative reaction of different varieties of gram to blight disease incited by Phyllosticta rabiei (Pass.). Trot. in the Punjab. J. Res. Punjab. Agr. Univ. 4: 214-216.
- AZIZ, M.A. 1962. C 727 a new blight-resistant gram variety for Barani areas. W. Pakistan J. Agr. Res. 1: 165-166.
- BEDI, K.S. 1961. Outbreak of gram blight in Punjab in 1958-59.
   Plant. Prot. Bull. 12: 24-25.
- BEDI, K.S., and D.S. ATHWAL. 1962. C 235 is the answer to blight.
   Indian Fmg. 12(9): 20-22.
- BEDI, P.S., and S.S. AUJLA. 1960. Variability in Phyllosticta rabisi (Pass.) Trott: the incitant of blight disease of gram. J. Res. Punjab Agr. Univ. 6: 103-106.

- BENLLOCH, M. 1941. Some phytopathological characteristics of the year 1941. Bol. Pat. Veg. Agr., Madr. X, 29-32: 1-14
- BIGGS, C.E.G. 1944. Annual Report, Department of Agriculture, Tanganyika Territory, 1943. 8 pp.
- BUSHKOVA, Mase L.N. 1960. Susceptibility of chickpea varieties to Ascochytosis. Zasch. Rast., Mosk. 5: 55.
- 14. DEMETRIADES, S.D., D.G. ZACHOS, P.T. CONSTANTINOU, C.G. PANA-OOPULOS, and C.D. HOLEVAS. 1958, 1959. Brief report on the principal, plant disease observed in Greece during the years 1957, 1958. Ann. Inst. Phytopath. Benaki, N.S. I. 6: 323-329, 1958; 2. 1: 3-11, 1959.
- GEORGHIOU, G.P., and C. PAPODOPOULOS. 1957. A second list of Cyprus fungi. Tech. Bull. Dep. Agr. Cyprus 5, 38 pp.
- GREWAL, J.S., and S. VIR. 1974. Varietal resistance of gram to Ascochyta blight. Indian Phytopath. 27: 643-645.
- HALFON-MEIRI, A. 1970. Infection of chickpea seeds by Ascochyta rabiei in Israel. Plant Dis. Reptr. 54: 422-445.
- JENKYN, J.F., J.M. HIRST, and Y. KING. 1972. An apparatus for the isolated propagation of foliar pathogens and their hosts. Ann. Appl. Biol. 73: 9-13.
- KAISER, W.J., D. DANESH, M. OKHOVAT, and H. MOSSAHEBI, 1968.
   Disease of pulse crops (edible legumes) in Iran. Plant
   Dis. Reptr. 52: 687-691.
- KAISER, W.J. 1972. Occurrence of three fungal diseases of chickpea in Iran. FAO Plant Prot. Bull. 20: 74-78.

- 21. EHIL'50, V.G. 1974. Study on the resistance of chickpes to ascochytosis. Genetike i selektsii. 53(3): 132-136.
- KOVACLEVSKI, I.C. 1936. The blight of chickpea, Mycosphaerelia rabiei n. sp. Issued by Min. Agr. Nat. Domains, Sofia, 80 pp
- LABROUSSE, F. 1931. Anthracnose of chickpea. Rev. Path, Veg. et Ent. Agr. 28: 226-231.
- LABROUSSE, F. 1931. Observations on some diseases of market garden plants. Rev. Path. Veg. et Ent. Agr. 28: 286-289.
- LUTHRA, J.C., A. SATTAR, and K.S. BEDI. 1938. The control of the blight disease of gram by resistant types. Curr. Sci. 7(2): 45-47.
- MORRALL, R.A.A., and D.L. McKENZIE. 1974. A note on the inadvertent introduction to North America of <u>Ascochyta rabiei</u>,
   a destructive pathogen of chickpea. Plant Dis. Reptr.
   58: 342-345.
- 27. PROGRESS REPORT-4, Pulse Pathology 1979. International Chickpea Disease Resistance Testing Program. The second International Chickpea Cooperative Disease Nursery, 1977-78, Report. ICRISAT, ICRISAT Patamcheru P.O., Andhra Pradesh 502 324, India.
- RADKOV, P. 1970. Susceptibility of some chickpea varieties to Mycosphaerella rabiei. Rastit. Zasht. 18(9): 22-27
- RADULESCU, E., E. CAPETTI, E. SCHWIDT, and A. CASSIAN. 1971.
   Contributions to the study of anthracnosis of chickpea
   (Necosphasrella rabiei Kov.) Lucrari stuntifice, 14: 311-321.

- SAREJARNI, J.A. 1939. Amnotated list of the fungi encountered on cultivated plants in Greece. Ann. Inst. Phytopath. Benaki. 3(2): 41-66.
- 31. SATTAR, A. 1933. On the occurrence, perpetuation and control of gram (Close aristinum L.) blight caused by Ascochyta rabisi (pass.) Labrousse, with special reference to Indian conditions. Ann. Appl. Biol. 20: 612-632.
- SATTAR, A., and A. HAFIZ. 1951. A practical method of inoculating gram (Ascochyta rabisi) on a field scale. Proc.
  Third Pakistan Sci. Conf. pp. 1-2.
- SAVULESCU, T. 1932. Phytosanitary conditions in Romania during the year 1930-31. Inst. Cerc. Agron. al Romaniei Publ. 8, 31 pp.
- SCHARIF, G., E. NIEMANN, and M. GHANEA. 1967. Chickpea blight
  in Iran Mycosphasrella rabiel Kovacevski = Ascochyta rabiei
  (Pass) Labrousse. Entomologie Phytopath. Appl. 25: 9-15.
- 35. SOLEL, Z., and J. KOSTRINSKI. 1964. The control of Ascochyta anthracnose of chickpes. Phytopath. Mediterranes. 3: 119-120.
- SORAN, H. 1975. The most important diseases of chickpea in central Anatolia. J. Turkish Phytopath. 4(2): 53-62.
- VEDYSHEVA, R.G. 1966. Evaluation of the resistance of Cicer arietinum to Assochyta rabisi. Vest. Sel. Khoz. Nauki, Mosk. 11(12): 109-111.

- VEDYSHEVA, R.G. 1966. Infection of chickpes with Assochyta
  rabisi in infected ground. Zap. Voronezh, Sel. Khoz. Inst.
  32: 275-281.
- 39. VIR, S., and J.S. GREWAL. 1974. Physiological specialization in Ascochyta rabiei, the causal agent of gram blight. Indian Phytopath. 27: 355-360.
- 40. ZALPOOR, N. 1963. Nyoosphaerella rabisi Kovacevski (Asoochyta rabisi) (Pass.) Lab. Ent. Phytop. Appl., Tehran.
  21: 10-12.

# UNIDENTIFIED : A comment:

As the authors have proposed, the acope for creating new sources of resistence through mutations definitely exists, Even if natural sources of resistence are discovered, there will always be a need for having a larger variability for resistance.