

Screening of pigeonpea genotypes for *Fusarium*-wilt resistance*

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A set of 226 pigeonpea (*Cajanus cajan* (L.) Millsp.) genotypes were screened for their resistance against wilt disease. 105 genotypes were found to show resistant reaction (0 - 10% wilt), 33 moderately resistant (10 - 30% wilt) and 88 susceptible reaction (> 30% wilt). Estimation of total phenols and sugars in root exudates of resistant and susceptible genotypes showed marked difference in the quantity. Resistant genotypes had higher phenol and sugar content than susceptible ones. In resistant genotypes total phenol content was appreciably higher in leaves and roots of the resistant genotypes as compared to the susceptible ones. But in seeds the difference was not so marked.

Key words : Pigeonpea, wilt, *Fusarium udum*, root exudates resistance, total phenol, total sugar

INTRODUCTION

Pigeonpea or redgram [*Cajanus cajan* (L.) Millsp.] is known to be affected by more than hundred pathogens (Nene *et al.*, 1989) but only a few of them cause economic losses (Kannaiyan *et al.*, 1984) of which *Fusarium* wilt caused by *F. usarium udum* is a serious problem. The disease first reported in India by Butler (1910). Study was taken to screen and identify the resistant source of *Fusarium*-wilt of pigeonpea, which could benefit breeding programmes and varietal introduction.

MATERIALS AND METHODS

Evaluation of wilt resistance was carried out in a well developed and maintained sick plot at the University Farm, Pulses Scheme, UAS, GKVK, Bangalore. A total of 226 lines obtained from IIPR, Kanpur, ICRISAT, Hyderabad and BARC, Mumbai were screened. Seeds were shown in the first fortnight of July during 1996-97 and 1997-98 in a randomized block design replicated twice. The lines

were sown in reverse order in the second year (1997-98). For each line, 50 seeds were sown in 5 m row, spaced at 30-45 cm apart, depending upon maturity group. After every two rows of test entries, a row of susceptible cultivar (Bahar or ICP-2376) was sown for monitoring the wilt incidence as check and maintaining the sickness of soil. Final wilt incidence was recorded at maturity by calculating the percentage of wilted plants. Genotypes with wilt incidence of 10 per cent and below was considered as resistant, an incidence of 11 to 30 per cent as moderately resistant and above 30 per cent regarded as susceptible (Anon, 1997).

The method described by Rovira (1956) was used for collection of root exudates. Sterilised 10 cms diameter Giffy pots were filled with washed clean sand. In each pot four seeds were sown at equidistance in 2 cms depth with ICP 8863 (resistant) and TTB 7 (susceptible) seeds. The pots were periodically watered with sterile water. The water leachate of 15 day old seedlings was collected. The seedlings were gently removed

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without any injury to the roots. The removed seedlings were kept for a day in the removed leachate. Thus, collected concentrated root exudates were used for the study.

One gm of dried seed, root or leaf powder was taken in a test tube, 100 ml ethyl alcohol added to and boiled over a hot water bath for 5-10 minutes. The extracts were filtered through Whatman No. 41 filter papers. Extractions was repeated for three times and the extracts were pooled for biochemical analysis. Alcohol was evaporated on a hot water bath till alcoholic smell disappeared and the final volume made upto known volume.

Folin-ciocalteau reagent (FCR) method was followed to estimate total phenols in the plant samples (Anon., 1970). One ml of the plant extract was taken in a test tube and 1 ml of FCR was added to it followed by 2 ml of 20 per cent of Na_2CO_3 solution and heated in a boiling water bath for one minute. The tube was cooled and the content made to 25 ml with distilled water and the absorbance was measured at 650 nm. A standard curve was made from different concentrations of catechol. Total phenol content was calculated by comparing absorbance value with standard curve.

The total sugars in the root exudate extraction sample were estimated by the phenol sulphuric acid method. The samples were centrifuged and the supernatant was used for the analysis of total sugars.

Aliquot of extract was made upto 1 ml with distilled water and to this 0.5 ml of phenol reagent was added and mixed well. Further 5 ml of sulphuric acid was added and placed in water bath at 30°C for 10 minutes and the absorbance was read at 490 nm. The amount of total sugars in the sample was estimated by comparing the results with a standard glucose curve (0 to 100 µg/ml) and prepared in a similar way.

RESULTS AND DISCUSSION

The wilt incidence was recorded as per the recommendations of IIPR, Kanpur. As many as 105 accessions recorded 10 per cent or less incidence of

wilt of which 46 genotypes viz., ICP 8859, ICP 8863, ICP 12731, TCP 12748, ICPL 89048, ICPL 90097, ICPL 92057, ICPL 93005, ICPL 93011, ICPL 93013, ICPL 94063, AWR 74-75, GPS 30, GPS 33, GPS 36, GPS 52, BWR 153, BWR 254, BWR 370, BWR 377, BWR 378, Sujatha 1-2, Sharan 1-21, PR 5149 sel. BDN 699, PKPH 6190, BSMR 214, BSMR 837, BSMR 838, BSMR 846, BSMR 848, BSMR 850, BSMR 851, BSMR 853, BSMR 855, MAL 10, WRP 82-2, K32-2, WRP 12, WRP 248-1, WRP 232, WRP 247-2, WRP 61-3, WRP 264, WRP 243-2 and AKT-9621 were completely free from wilt incidence. These are considered resistant to wilt disease (Table 1).

Thirty three entries recorded wilt incidence of 11 to 30 per cent and therefore, were considered as moderately resistant. As many as eighty-eight genotypes have consistently recorded more than 30 per cent wilt and thus rated as highly susceptible. This group also includes such popular and released varieties like TT B-7, ICPH-8, Bahar, HY-3C, etc (Table 1).

Total phenol of different genotypes were studied by sampling seeds, roots and leaves (Table 2). Total phenol content varied in different parts of the plants. In general phenol content was highest in leaves followed by roots and lowest in seeds.

Phenol content in seeds of resistant genotypes varied between 7.2 mg/100gm (MAL 10) to 12.1 mg/100gm (DAP 92-1) while that in susceptible genotypes varied between 7.4 mg/100gm (Bahar) and 12.6 mg/100gm (ICP 2367). Thus, there was no appreciable difference of phenol content in the seeds of resistant and susceptible genotypes.

The root samples of the resistant and susceptible genotypes on the other hand showed a striking difference in their phenol content. The resistant genotypes showed a range of 43.22 and 67.13 mg/100 gm of total phenol content in IPA 943 and ICP 8863 respectively. The phenol content in ICP 87119 (56.00), MAL 10 (59.81) and DPA 92-1 (52.64) was intermediate. The susceptible genotypes showed a range of 9.37 mg/100 gm (in TT B-7) to 18.43 mg/100 gm (in ICP 2376).

Table 1 : Reaction of pigeonpea genotypes against *Fusarium udum*

| Resistant (R) < 10 % | |
|---|--|
| ICP 6997, ICP 8858, ICP 8859, ICP 8860, ICP 8861, ICP 8863, ICP 9174, ICP 12731, ICP 12745, ICP 12748, ICPL 87119, ICPL 89048, ICPL 89049, ICP 90097, ICPL 91057, ICPL 91058, ICPL 92057, ICPL 93004, ICPL 93005, ICPL 93011, ICPL 93012, ICPL 93013, ICPL 94063, AWAR 74-15, B-Palera, GPS-3, GPS-7, GPS-30, GPS-33, GPS-36, GPS-42, GPS-52, Godu 1-1, BWR-153, BWR-254, BWR-370, BSMR 214, Jows-5, Jows-6, Sujata 1-2, PI 397480 sel, PR 5149 sel, KPBR 80-2-1, Pusa-941, BDN 699, PKPH 6190, KA 26-4, MA-3, VSMR 380, KA 91-25, PWR 171, MA-6, TAT 9402, WRP 82-3, K 30-2, WRP 12, WRP 248-1, WRP 232, WRP 247-2, WRP 61-3, WRP 264, WRP 237-2, WRP 243-2, AKWR 3, JJA 33, TAT 97-16M, KM 128, KA 91-25, BWR 375, BWR 378, BSMR 837, BSMR 838, BSMR 839, BSMR 841, BSMR 843, BSMR 844, BSMR 839, BSMR 846, BSMR 847, BSMR 848, BSMR 849, BSMR 850, BSMR 851, BSMR 852, BSMR 853, BSMR 854, BSMR 855, MA-3, TAT 94-2, IAR 98-03, WRP-1, AKT 9221, APA 94-3, K 26-4, MA-10, VRG-19, TT 9802, PT 8707-29-1, AKT 9621, TAT 9803, DPA 92-1, ALP 9221. | |
| Moderate Resistant (MR) 10-30% | |
| Pusa 944, AL 600, AL 1340, AL 1361, AKPH 9180, KM 34, MPH 9343, C-11, DA 92-4, VRG 13, KM 118, DA 46, DA 93-1, DA 93-2, NDA 94-4, METH 103, ARG 102, TAT 97-69, TT 97-48, TAT 93-47, WRP 235, AKWR-2, AKWR 6, KM 125, BWR 376, BSMR 942, AKT 9221, AKPH 1150, AKPH 2080, TAT 43-47, TAT 97-48, PT 8202-16, AKT 9713. | |
| Susceptible (S) > 30% | |
| ICPL 332, Pusa 945, Pusa 961, Pusa 962, Pusa 9, Pusa 21, Pusa 1322, Pusa 323, Pusa 325, Pusa 326, Pusa 330, Pusa 331, Pusa 333, Af 239, Af 284, Af 293, ICPH 8, Bahar, KA-3, NDA 93-1, KA 32-2, DA 41, DA 42, DA 93-5, NDA 94-6, METH 104, DA 45, NDA 94-1, ICP 2376, AF 345, WRG 5, KA 3, KE 108, KM 108, Pusa 971, METH 121, AL 1313, GAUT-971E, GAUT 97-20, H 91-7, H 82-1, H 91-19, KDF 2, LRG 38, WRG 13, WRG 27, VRG 4, VRG 7, VRG 13, ARG 1001, ARG 1002, MA 7, MTH 9508, MTH 9607, MTH 9608, P 945, MTH 9611, NDA 93-2, Pusa 27, H 90-13, H 90-14, KDF 2, FS 90, TTB 7, IPA 951, KA 3, KM 137, GAUT 9801, KM 138, AKT 9011, AL 1340, AL 1381, KP 2376, MA 4, MTH 6111, KA 32-2, Pusa 327, Pusa 334, Pusa 335, AF 239, AF 284, P 782, P 981, TAT 9801, WRG 1, KE 108, METH 121. | |

The leaf samples showed much higher amount of total phenol as compared to seed and root samples of the test genotypes. The resistant genotype MAL 10 (587.86 mg/ 100 gm) showed maximum amount of phenol, whereas, ICP 87119 (198.01 mg/100 gm) showed least amount among the five resistant genotypes analysed. Total phenol in susceptible genotypes was, in general, was comparably lower.

Table 2 : Estimation of phenols in resistant and susceptible genotypes

| Genotypes | Host reaction | Phenols (mg/100 gm) | | |
|-----------|---------------|---------------------|-------|--------|
| | | Seeds | Roots | Leaf |
| ICP 8863 | R | 8.08 | 67.13 | 228.95 |
| ICP 87119 | R | 9.43 | 56.00 | 198.01 |
| MAL 10 | R | 7.27 | 59.81 | 587.86 |
| DAP 92-1 | R | 12.12 | 52.64 | 396.03 |
| IPA 943 | R | 11.72 | 43.22 | 541.45 |
| TTB 7 | S | 8.35 | 9.37 | 187.18 |
| ES 90 | S | 8.52 | 14.16 | 197.14 |
| ICP 2376 | S | 12.63 | 18.43 | 218.69 |
| Bahar | S | 7.44 | 12.92 | 182.42 |
| IPA 951 | S | 10.31 | 12.68 | 206.93 |

Table 3 : Estimation of phenols and total sugars in root exudates of susceptible and resistant pigeon pea genotypes

| Genotypes | Host reaction | Phenols (mg/100 gm) | Total Sugars (mg/ml) |
|--------------------|---------------|---------------------|----------------------|
| TTB-7 | S | 1.73 | 2.18 |
| ICP 8863 (Maruthi) | R | 4.34 | 6.20 |

The analysis of the root exudates collected from susceptible genotype TTB-7 and resistant genotype ICP 8863 (Maruthi) showed a marked difference in their content of total phenol and sugar. Total phenol and sugar contents were higher in resistant genotype than the susceptible one (Table 3).

The results are in conformity with the reports of Bidari *et al.*, (1996) and Reguchander and Arjunan (1996) on host reaction.

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