

A rapid method for pigeonpea wilt resistance screening

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Abstract : A standardized rapid method for inoculating pigeonpea seedlings with *Fusarium udum* was developed to study host plant resistance under controlled conditions and to demonstrate the pathogenic variability in two *Fusarium udum* isolates.

Keywords : *Fusarium udum*, pathogenic variability

Fusarium wilt caused by *Fusarium udum* Butler is an economically important disease of pigeonpea [*Cajanus cajan* (L.) Millsp.] in the Indian subcontinent and Africa (Kannaiyan *et al.*, 1984). Nene *et al.* (1981) described field- and pot-screening techniques to identify sources of resistance to wilt. However, these methods were not satisfactory for studies on pathogenic variability in *F. udum* isolates, and inheritance of wilt resistance because of variation in the number of days required for wilting among plants. Also, nearly 60-90 days were normally required for wilting of all plants in the wilt-susceptible cultivar, ICP 2376. Therefore, a root-dip technique was developed for rapid screening of pigeonpea genotypes for wilt resistance and to determine the pathogenic variability amongst *F. udum* isolates.

MATERIALS AND METHODS

Single-spore culture of *F. udum* from wilted pigeonpea plants were maintained on acidified

(pH 5) potato-dextrose-agar in culture tubes. Of them, isolates 1 and 2 were used for inoculations. Seeds of pigeonpea test genotypes (ICP 2376, ICP 8518 and ICP 6997) obtained from selfed plants were surface sterilized with 2.5 per cent sodium hypochlorite for 2 min. The seeds were sown in polythene bags containing autoclaved river-bed sand. Seven-day old seedlings were used for testing (Nene *et al.*, 1981).

Root-dip technique

The fungus was multiplied in 100 ml potato-dextrose-broth in 250 ml conical flasks that were placed on a rotary shaker for 7 days at 25°C. The entire fungal contents of each flask was macerated in a Waring blender for one minute and then used as inoculum. Roots of the test seedlings were dipped in the inoculum for 1 min. The seedlings were then transplanted in 15-cm plastic pots containing autoclaved sand, Vertisol, or Alfisol soil. Uninoculated seedlings transplanted in uninoculated sand or soil served as control.

Pot-culture techniques

The fungus was multiplied on 100 g of sand-pigeonpea meal medium in 250 ml conical flasks

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at 25°C for 20 days. A fungus-soil mixture was prepared as described by Nene *et al.*, (1981) by mixing 200 g of the inoculum with 2 kg of autoclaved sand, Vertisol or Alfisol. The soil mixture was placed in 15-cm plastic pots and watered. Surface sterilized seeds were sown in the fungus-infested soil mixture. Also, 7-day old seedlings were transplanted in the soil mixture. Seeds sown or transplanted in uninoculated sand or soil served as control.

Three pots or replications were used for each treatment in each inoculation technique. Each pot had 10 seeds or seedlings. The pots were placed in a growth chamber at 30°C under fluorescent light (cool, 120-cm long tubes, 40 W) with 12 h day length. Test plants were watered daily with sterile deionized water to keep the

sand or soil moist. Observations on wilt incidence were recorded at 5-day intervals. The experiment was repeated twice with reproducible results.

The root-dip technique was then used to demonstrate the pathogenic variability in two isolates of *F. udum*. Five pigeonpea genotypes (ICP 8863, -12737, -8518, -6997 and -2376) were used.

RESULTS AND DISCUSSION

In the present studies, genotypic differences in time taken for wilting of pigeonpea were observed (Table 1). There was no effect of soil type on wilting of the three cultivars in the root-dip technique (Table 1). However, it was easier to transplant seedlings in sand than in Alfisol or Vertisol. There was a considerable delay in wilting

Table 1 : Time required for wilting in three pigeonpea genotypes using the root-dip and pot-culture techniques at ICRISAT Center, 1985

Inoculation treatment	Days to wilt*		
	ICP 2376	ICP 8518	ICP 6997
Root-dip technique			
Sand	26 (91.7)	20 (100.0)	26 (100.0)
Vertisol	26 (100.0)	26 (100.0)	26 (100.0)
Alfisol	26 (96.7)	20 (100.0)	26 (100.0)
Pot culture technique			
Direct sowing			
Sand	56 (53.3)	51 (33.3)	60 (33.3)
Vertisol	0 (0.0)	60 (20.0)	0 (0.0)
Alfisol	51 (53.3)	60 (60.0)	56 (33.3)
Transplanting			
Sand	41 (85.0)	26 (86.7)	41 (73.3)
Vertisol	51 (80.0)	46 (86.7)	60 (53.3)
Alfisol	41 (93.3)	46 (86.7)	46 (93.3)
	Inoc. × Soil × Genotype		
SE ±	7.3 (9.19)**		
CV (%)	40.0 (25.3)**		

Figures in parentheses denote maximum wilt incidence (%).

*Wilt-resistant genotypes, ICP 8863 and ICPL 8356, showed no mortality.

**Based on angular transformation.

of pigeonpea plants in the pot-culture techniques—direct sowing and transplanting (Table 1). Also, wilting was not uniform within each genotype. In the root-dip technique, over 90 per cent wilt incidence was observed in 20-26 days in the three genotypes (Table 1) and wilting was uniform within each genotype. ICP 8863 and ICPL 8356 did not show any mortality. These lines were also resistant in field screening at ICRISAT center (Nene and Kannaiyan, 1982). The root-dip technique was successfully used to study the inheritance of resistance to *F. udum* in pigeonpea. The technique is used routinely in greenhouse experiments.

Isolate 1 of *F. udum* obtained from the wilt-susceptible cultivar ICP 2376 is common at ICRISAT Center, Patancheru. The cultivar ICP 8863 was resistant to wilt in field, greenhouse, and laboratory-screening tests to isolate 1 (Nene and Kannaiyan, 1982). During 1985-86 sporadic wilting was observed in ICP 8863 at ICRISAT Center. Isolate 2 was obtained from wilted plants of ICP 8863. ICP 8863 was resistant to isolate 1 but highly susceptible to isolate 2 (Table 2) indicating the presence of races in *F. udum*.

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Table 2 : Per cent wilt incidence in five pigeonpea genotypes inoculated with two *Fusarium udum* isolates by the root-dip technique

Genotype	Wilt incidence (%)*	
	Isolate 1	Isolate 2
ICP 8863	0	100
ICP 12737	50	0
ICP 8518	100	100
ICP 6997	100	100
ICP 2376	100	100

*Wilt incidence at 30 days after inoculation.

REFERENCES

- Kannaiyan, J., Nene, Y.L., Reddy, M.V., Ryan, J.G. and Raju, T.N. (1984). Prevalence of pigeonpea diseases and associated crop losses in Asia, Africa and the Americas. *Trop. Pest Manage.* **30** : 62-71.
- Nene, Y.L. and Kannaiyan, J. (1982). Screening pigeonpea for resistance to *Fusarium* wilt. *Plant Dis.* **66** : 306-307.
- Nene, Y.L., Kannaiyan, J. and Reddy, M.V. (1981). Pigeonpea Diseases—Resistance-Screening Techniques. Information Bulletin No. 9. Patancheru, Andhra Pradesh, India : ICRISAT, 14 pp.