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Determination of Variability in PPSMV (Pigeonpea Sterility Mosaic Virus) Isolates through Host Differentials

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Abstract

Ten pigeonpea host differentials *viz*., Bahar, BRG-1, BRG-2, BRG-3, BSMR-736, ICP2376, ICP7035, ICP8863, IPA 8F and Purple-1 were used to assess the variability of PPSMV prevailing in different pigeonpea growing locations *viz.*, Gulbarga, Bengaluru, Badnapur, Coimbatore, Rahuri and Varanasi in *kharif* 2015. Bahar, BRG-1, BSMR-736, ICP7035 and IPA8F showed resistant reaction at Badnapur and Varanasi, whereas same entries showed susceptible reaction at Bengaluru and Coimbatore. BRG-3 exhibited resistant reaction at Badnapur, Bengaluru and Rahuri and same line exhibited susceptible reaction at Coimbatore. Whereas, in *kharif* 2016, BSMR-736, ICP7035 and ICP8863 showed resistant reaction at Varanasi and same lines expressed susceptible reaction at Coimbatore, Bengaluru and Badnapur. ICP2376, BSMR736 and ICP8863 showed 100 per cent disease incidence and highly susceptible reaction at Bengaluru and they were expressed resistant reaction at Rahuri and Varanasi. Based on the pigeonpea host differential reaction to PPSMV, distinctly six strains/ isolates *viz*., Bengaluru, Badnapur, Coimbatore, Varanasi, Rahuri and ICRISAT isolates were identified.

Keywords: Pigeonpea, Sterility mosaic disease, Pigeonpea sterility mosaic virus

Pigeonpea [*Cajanus cajan* (L.) Millspaugh] is a hard lived legume belonging to *Cajaninae* sub tribe of the economically most important leguminous tribe *Phaseoleae*. It is a multipurpose grain legume crop grown extensively for food in the Asian and African countries. India is considered as the primary centre of origin for pigeonpea due to the presence of ample variability in local germplasm and wild relatives (Saxena *et al*., 2008). In recent years, the crop is gaining importance due to its inherent ability to perform well under marginal input conditions and also its adaptability to drought and other abiotic stresses.

Although, India leads the world both in area and production of pigeonpea, its productivity is lower than the world average which may be attributed to various abiotic (e.g. drought, salinity and water-logging) and biotic (e.g. diseases like Fusarium wilt, sterility mosaic and insects like pod borers) factors. Among diseases, Fusarium wilt and sterility mosaic diseases are the major constraints to pigeonpea production worldwide. This is a matter of concern since the domestic demand of pigeonpea is rapidly increasing.

Sterility mosaic disease (SMD), considered as the "most destructive disease" caused by *pigeonpea sterility mosaic virus* (PPSMV) and the virus is transmitted by the vector eriophyid mite, *Aceria cajani* is one of the major biotic factors in the Indian subcontinent which leads to heavy yield loss at crop growth stage (Pallavi, 2014).

In almost all the pigeonpea growing areas of the country the disease results in significant yield reduction up to 100 per cent (Manjunath *et al*., 2012). In spite of various control measures, SMD has continued to be major constraint in pigeonpea production. A lot of variation exists among the isolates of the virus exhibited in different regions. These variations render it difficult to evolve a common management strategy to control SMD epidemics. Therefore, a number of molecular or biological approaches are available to differentiate the variability of the virus isolates. These biological techniques were used to study the variability in plant pathogenic virus.

Material and Methods

Testing of host differentials at different locations

Ten pigeonpea host differentials namely Bahar, BRG-1, BRG-2, BRG-3, BSMR-736, ICP 2376, ICP7035, ICP8863, IPA 8F and Purple-1 were tested to know the variability of PPSMV in different pigeonpea growing locations *viz.*, Gulbarga, Bengaluru, Badnapur, Coimbatore, Rahuri and Varanasi in *Kharif* season 2015 and 2016. Each entry was sown in two replications of five meters length. Susceptible check was planted after every two test rows and inoculated at two leaf stage by leaf stapling technique.

Observations on disease incidence and symptom type were recorded. Mite counts were taken on younger leaves (trifoliate) of five randomly selected plants for each differential variety.

The per cent disease incidence was calculated based on the number of plants infected out of total plant examined in each sector.

$$\text{Disease incidence} = \frac{\text{Number of plants infected}}{\text{Total number of plants examined}} \times 100$$

Leaf stapling technique

In this method leaflets from diseased plants infested with mites are stapled onto the primary leaves of the test seedling. Mite from the diseased seedling migrates onto the test seedling results in virus transmission onto the test plant. This is the most efficient method for PPSMV transmission to Pigeonpea.

Procedure

- Leaflets were collected from SMD infected pigeonpea plant. After assessment of the infected leaves by mites were observed under Steriobinocular microscope.
- The diseased leaf was folded in such a way that under side of the diseased leaf comes in contact with both the surface of the test plant and stapled to susceptible check

Results and Discussion

Effect of host differentials and mite population on the incidence of disease at Bengaluru during *kharif* 2015 and 2016

Data in Table 1 clearly indicated there is difference in reaction of host differentials with respect to per cent disease incidence and vector population during the year 2015 and 2016 *Kharif*. At 30 DAS no disease incidences and no mite activity was noticed in all host differentials during both season of *kharif* 2015 and 2016. At 45 DAS almost, all host differentials harbored by the activity of vector *A. cajani., w*here highest number of mites was recorded in ICP2376 (8.3) followed by ICP8863 (6.8) and least was recorded in Bahar (0.80) and IPA8F (0.93). BRG-2, BSMR736, ICP 2376 and ICP8863 showed susceptible reaction (100%) and Bahar and BRG3 were resistant to PPSMV at Bengaluru during *kharif* 2015.

In *kharif* season 2016, vector population was more on BRG2 followed by ICP 2376 and least population was recorded in ICP 7035 and per cent disease incidence was highest in ICP- 2376 (100%) and ICP 8863 (100%) and least incidence was observed in ICP7035 and BRG-3 exhibited resistant reaction to PPSMV. Similar observations on variability of PPSMV have been reported in pigeonpea. Similar observations on variability of PPSMV have been reported in pigeonpea (Gnanesh *et al*., 2011).

Effect of host differentials on mite population and incidence of SMD at Gulbarga during *kharif* 2015 and 2016

At Gulbarga center ten host differentials exhibited different reaction to vector population along with per cent disease incidence. BSMR 736 showed highest per cent disease incidence (100) with highest number of mites per trifoliate leaf (11.7) followed by ICP 8863 (85.1% and 6.9) during *kharif* 2015 @ 115 DAS and same ten host differentials given different reaction during 2016 *kharif*. BRG2 and Purple 1 recorded highest per cent disease incidence of 40.3 and 56.7 per cent with mite population of 1.03 per trifoliate leaf and least incidence was recorded in BRG 3 (16.3%).

TABLE 1

Reaction of pigeonpea host differentials to Pigeonpea Sterility Mosaic Virus (PPSMV) during *kharif* 2015 and 2016 at Bengaluru

Variety	2015									2016								
	45 DAS		65 DAS		95 DAS		115 DAS			45 DAS		65 DAS		95 DAS		115 DAS		
	PDI	MP	PDI	MP	PDI	MP	PDI	MP	Mean of MP	PDI	MP	PDI	MP	PDI	MP	PDI	MP	Mean of MP
Bahar	0.00	1.00	0.00	1.00	00.0	0.70	00.0	0.50	0.80	1.05	10.70	10.0	6.30	8.30	1.50	9.80	1.60	5.20
BRG 1	3.00	3.35	4.45	7.90	14.8	3.00	23.8	0.40	3.60	0.75	3.40	6.50	7.10	2.45	1.70	29.90	1.10	3.32
BRG 2	9.20	0.70	16.6	1.20	39.3	1.10	100	0.90	0.97	6.15	10.9	8.45	12.3	9.75	5.20	87.50	2.30	7.60
BRG 3	6.05	0.70	18.2	1.30	24.5	1.50	0.00	0.60	1.02	00.0	2.60	3.10	0.80	1.80	1.40	00.00	1.30	1.50
BSMR 736	3.55	0.30	6.85	11.6	24.7	5.80	100	0.40	4.50	20.0	4.40	13.2	11.3	16.5	8.40	97.50	1.20	6.30
ICP 2376	9.95	11.0	11.5	15.2	21.3	3.20	100	4.00	8.30	1.75	11.0	16.6	1.20	15.0	11.3	100	4.40	6.90
ICP 7035	0.00	0.30	15.0	1.00	19.5	0.65	21.0	0.25	0.55	1.70	0.25	7.95	0.50	15.2	0.10	9.50	1.30	0.53
ICP 8863	1 1.6	10.3	13.3	11.1	13.5	0.70	100	5.30	6.80	3.20	12.3	5.65	7.30	7.65	3.20	100	1.90	6.10
IPA 8F	2.75	0.40	4.90	1.30	11.1	1.25	15.1	0.80	0.93	0.50	3.80	12.6	5.20	9.75	0.90	45.50	5.90	3.90
Purple 1	3.15	0.95	13.0	1.40	14.0	1.50	25.8	0.95	1.20	1.05	7.95	9.00	3.40	0.00	1.30	47.80	1.40	3.50
LSI	4.2		11.4		20.4		44.9			5.9		17.0		11.4		52.7		

PDI: Per cent disease incidence, MP: Mite population, LSI: Local severity Index, DAS: Days after sowing

TABLE 2

Reaction of pigeonpea host differentials to pigeonpea sterility mosaic virus during *kharif* 2016 at Gulbarga

Variety	2015									2016								
	45 DAS		65 DAS		95 DAS		115 DAS			45 DAS		65 DAS		95 DAS		115 DAS		
	PDI	MP	PDI	MP	PDI	MP	PDI	MP	Mean of MP	PDI	MP	PDI	MP	PDI	MP	PDI	MP	Mean of MP
Bahar	0.00	0.80	0.00	0.80	0.00	0.80	0.0	1.40	0.95	19.2	1.00	23.5	0.70	17.1	1.60	21.9	1.00	1.07
BRG 1	11.40	6.00	20.0	7.80	47.80	3.10	50.3	1.10	4.50	10.5	0.90	17.1	0.40	26.8	1.10	19.1	0.90	0.82
BRG 2	22.00	11.70	39.7	11.9	32.50	1.10	67.3	1.70	6.60	15.8	1.30	35.3	0.60	29.8	1.30	40.3	1.30	1.10
BRG 3	3.75	0.50	4.50	1.10	6.50	0.95	9.8	1.90	1.10	12.1	1.20	21.1	0.90	11.1	1.00	16.3	1.20	1.07
BSMR 736	15.90	13.30	33.7	11.10	54.20	5.98	100	11.7	10.5	0.00	1.60	5.9	0.40	10.1	1.20	25.8	1.60	1.20
ICP 2376	23.70	4.70	31.0	9.90	44.20	11.90	64.0	6.20	8.10	11.1	1.20	19.1	1.00	26.1	1.40	30.1	1.20	1.20
ICP 7035	2.20	0.80	6.00	0.90	18.50	0.90	13.8	1.80	1.10	19.1	1.00	7.90	0.25	13.1	1.30	18.5	1.00	0.88
ICP 8863	31.90	9.00	47.0	16.90	66.6	5.15	85.1	6.90	9.40	23.1	1.10	29.1	0.30	25.3	1.90	27.5	1.10	1.10
IPA 8F	2.65	8.80	15.5	6.10	18.7	11.0	23.1	5.00	7.70	15.1	1.30	21.1	0.80	30.6	0.90	36.5	1.30	1.07
Purple 1	5.15	1.90	5.00	1.40	9.10	2.30	9.50	1.60	1.80	30.2	1.40	35.8	0.95	47.8	1.40	56.7	1.40	1.03
LSI	13.0		10.3		22.2		23.4			14.6		18.5		21.7	0.60	26.6		

PDI: Per cent disease incidence, MP: Mite population, LSI: Local severity Index, DAS: Days after sowing

TABLE 3

Reaction of pigeonpea host plant differentials to PPSMV at different locations during *kharif* 2015

Entries	SMD Per cent disease incidence (%) and disease reaction at different locations during *kharif* 2015											
	Badnapur	DR	Bengaluru	DR	Coimbatore	DR	ICRISAT	DR	Rahuri	DR	Varanasi	DR
Bahar	0.00	R	0.00	R	34.30	S	1.60	R	2.50	R	0.00	R
BRG 1	0.00	R	23.8	MR	37.50	S	0.00	R	1.50	R	0.00	R
BRG 2	45.70	S	100	S	45.90	S	0.00	R	42.30	S	0.00	R
BRG 3	5.60	R	0.00	R	48.60	S	-	-	1.60	R	-	-
BSMR 736	0.00	R	100	S	44.20	S	6.80	R	12.90	MR	10.50	R
ICP 2376	0.00	R	100	S	52.30	S	5.80	R	70.90	S	100	S
ICP 7035	-	-	0.00	R	-	-	0.00	R	4.70	R	0.00	R
ICP 8863	100	S	100	S	55.8	S	92.00	S	100	S	100	S
IPA 8F	0.00	R	0.00	R	7.70	R	0.00	R	1.30	R	0.00	R
Purple 1	-	-	25.8	MR	-	-	14.30	MR	55.00	S	66.70	S
LSI	18.90		44.90		40.70		13.90		29.30		30.80	

LSI: Local severity Index, DR: Disease Reaction, R-Resistant, MR: Moderately Resistant, S: Susceptible

Three distinct isolates have been characterized, *viz*., Bengaluru, Patancheru and Coimbatore. The Patancheru and Coimbatore isolates are mild strains while the Bengaluru isolate is the most virulent. Pallavi and Ramappa (2014) reported that the variety ICP 7035 is free of mite vector and ICP 8863 recorded the highest number of mite population (13.58 mites/ trifoliate leaf). Out of twenty varieties, five varieties *viz.,* BRG 3, ICP 7035, Bahar, IPA 8F and IPA 15F grouped as resistant (< 1 mite/trifoliate leaf) and rest of the fifteen genotypes were grouped as susceptible (≥1 mite/trifoliate leaf) Effect of host differentials on mite and disease incidence revealed in Table 2.

Reaction of pigeonpea host plant differentials to PPSMV at different locations

During *kharif* 2016, the experiment was carried out at Badnapur, Bengaluru, Coimbatore, ICRISAT, Rahuri and Varanasi to study the variability in PPSMV. The data received from all the centers were considered for interpretation of results. Ten pigeonpea host differentials namely Bahar, BRG-1, BRG-2, BRG-3, BSMR-736, ICP2376, ICP7035, ICP8863 and Purple 1 were evaluated against PPSMV at above said locations. The data presented in Table 3 & 4 revealed that, differential reaction was observed between the locations tested. Bahar, BRG1, BSMR 736, ICP 7035 and IPA 8F showed resistant reaction at Badnapur and Varanasi, whereas same entries showed susceptible reaction at Bengaluru and Coimbatore. BRG-3 exhibited resistant reaction at Badnapur, Bengaluru and Rahuri and same line exhibited susceptible reaction at Coimbatore. Similarly, there was a large variation in disease incidence and symptom expression of the genotypes between the locations and between the seasons, and tests at the respective

TABLE 4

Reaction of pigeonpea host plant differentials to PPSMV at different locations during *kharif* 2016

Entries	SMD Per cent disease incidence (%) and disease reaction at different locations during *kharif* 2016											
	Badnapur	DR	Bengaluru	DR	Coimbatore	DR	Rahuri	DR	Varanasi	DR	ICRISAT	DR
Bahar	-	-	9.80	R	-	-	-	-	-	-	1.60	R
BRG 1	0.00	R	29.90	S	18.70	MR	0.00	R	0.00	R	0.00	R
BRG 2	48.60	S	87.50	S	22.20	MR	9.30	R	0.00	R	0.00	R
BRG 3	13.60	MR	0.00	R	27.40	MR	0.00	R	0.00	R	-	-
BSMR 736	0.00	R	97.50	R	44.20	S	33.00	S	0.00	R	9.80	R
ICP 2376	0.00	R	100	R	52.30	S	16.80	MR	-	-	15.80	MR
ICP 7035	0.00	R	9.50	R	40.90	S	63.40	S	0.00	R	0.00	R
ICP 8863	100	S	100	S	55.80	S	0.00	R	0.00	R	96.10	S
BRG5	0.00	R	45.50	S	29.60	MR	100	S	56.60	S	0.00	R
Purple 1	51.90	S	47.80	S	10.80	R	71.40	S	28.50	MR	17.30	MR
LSI	21.40		52.70		30.10		29.30		42.50		28.10	

LSI: Local severity Index, DR: Disease Reaction, R-Resistant, MR: Moderately Resistant, S: Susceptible

location and at Hyderabad. The variation in symptom expression was comparatively less than disease incidence. PPSMV-1 is prevalent in northern parts of Karnataka, Maharashtra and the three states of north India whereas PPSMV-2 is more prevalent in Coimbatore and Bengaluru in southern India (Basavaprabhu *et al*., 2015)

Whereas in *kharif* 2016, same ten lines expressed different reaction at Gulbarga. BRG-1, 2, 3, BSMR7-36, ICP 7035 and ICP 8863 showed resistant reaction at Varanasi and same lines expressed susceptible reaction at Coimbatore, Bengaluru and Badnapur. ICP2376, BSMR736 and ICP8863 showed 100 per cent disease incidence and highly susceptible reaction at Bengaluru and they were expressed resistant reaction at Rahuri and Varanasi. Therefore, it is evident that, PPSMV strain present at Badnapur, Coimbatore, Rahuri, ICRISAT and Varanasi are different from Bengaluru. Similar observations on variability of PPSMV have been reported in pigeonpea.

A comprehensive study of variability in the sterility mosaic pathogen revealed the occurrence of different isolates of the pigeonpea sterility mosaic virus. However, not much is information available on pathogen variability. In spite of various control measures SMD has continued to be major constraint in pigeonpea production. A lot of variation exists among the genetic isolates of pigeonpea in different regions. These variations render it difficult to evolve a common management strategy to control SMD epidemics. Therefore, it is necessary to know the severity of disease and factors associated with disease development which helps in devising suitable management practices. Therefore, the present study

indicated the existence and variability in PPSMV across pigeonpea growing locations in India. Thus, helpful in developing a variety suited/withstand the pathogen by incorporating the resistance gene.

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