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Research Article

Genetic and molecular studies on components of rust resistance in recombinant inbred lines and back-cross populations of peanut (*Arachis hypogaea* L.)

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Abstract

Recombinant inbred lines and backcross populations of GPBD 5 × ICGV 86699 were inoculated with uredospores of rust fungus to study the genetic variation of components of rust resistance, relationships among components of resistance to rust and identification of microsatellite markers linked to rust resistance in peanut. There were highly significant differences among recombinants for incubation period and number of pustules per leaf area in all the population and high genetic gain along with high heritability for all components of rust resistance. Association study revealed all the components of rust resistance were significantly correlated with each other except Incubation period. Bulk segregant analysis in the segregating populations of cross (GPBD 5 × ICGV 86699) indicated SSR marker TC4g10 to be putatively linked to rust resistance. Further, validation of this marker outside the original mapping population could strengthen the reliable association of this marker with rust.

Key words

Components of rust resistance, *Puccinia arachidis*, Backcross breeding, Recombinant inbred lines, SSR marker

Introduction

Peanut is one of the important oilseed crops in the world known for its ability to survive in less favorable agro-climatic conditions. India along with China accounts for half of the world's peanut production. It is the major oilseed crop in India accounting for 45 % of oilseed area and 55 % of oilseed production in the country. Among the biotic stresses, rust causes up to 70% yield loss (Subrahmanyam and McDonald, 1987), particularly if the crop is also attacked by the two major leaf spot fungi, *Cercospora arachidicola* Hori and *Cercosporirium personatum* (Verk. and Curt.) Deighton. Apart from reducing yield, it also reduces oil and protein content of seeds and quantity as well as quality of fodder.

Genetic potentiality towards resistance can be estimated based on the yield component traits but this has complex attributes, generally having negative association with stress tolerance (REF), which may or may not end up with identification of resistance sources. Diseases components are the good indirect measure to pinpoint the plant with desirable disease reaction which can ultimately help in isolating resistant cultivar. Marker assisted selection (MAS) can improve the efficiency of conventional breeding especially in the case of low heritable and recessive traits, where phenotypic selection is difficult, expensive, lack accuracy or precision and even resistant lines can be identified at seedling stage through tightly linked trait specific marker. Recently, a few SSR marker and RAPD marker were found to be putatively linked with rust resistance loci. In the present study linked molecular markers were used to tag the rust resistance in cultivated peanut.

Materials and method

Rust susceptible peanut variety GPBD 5 (TG 49 X GPBD 4) is a Spanish bunch type (*A. hypogaea* subsp. *fastigiata* var. *vulgaris*), was selected for introgression of rust resistance and it is high yielding and bold seeded cultivar but highly susceptible to rust disease (Gowda *et al.* 2002) whereas, ICGV 86699 is is a highly resistant variety to rust and LLS and was selected as a donor parent for development of recombinant inbred lines and backcross populations. It is Virginia Bunch, high-yielding inter-specific derivatives with multiple resistance/tolerance to diseases and was derived from the cross of *Arachisbatizocoi* × *A. duranensis* X *A. hypogaea (cv. NC 2).* The F_1s were selfed to produce F_2 and advanced through single seed descent (SSD) method till F_6 generation. Selected resistant F_2 plants were used to backcross to the recurrent parent GPBD 5 to

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produce backcross (BC_1F_4, BC_2F_3 and BC_3F_2) population.

The identified rust resistant and high yielding recombinants from all (F_6, BC_1F_4, BC_2F_3 and BC_3F_2) the above populations were sown in replicated trial except BC_3F_2 population. Totally 43 lines of F_6, 33 lines of BC_1F_4, 27 lines of BC_2F_3 population and 121 BC_3F_2 individuals were planted in 1 m rows with 30 cm and 10 cm inter and intra-row spacing, respectively, in Randomized Block Design (RBD) with two replications. The two parents of above cross were sown as control after every 50th rows and 1st rows of each population. Components of rust resistance were studied in the identified resistant recombinant lines isolated from RILs (F_6) and BILs (BC_1F_4, BC_2F_3 and BC_3F_2 population) including parents along with susceptible check (TMV 2) and resistant checks (GPBD 4 and ICGV 99005). All the necessary agronomic practices were followed to raise a healthy crop except disease management

The "Infector row technique" was used to create artificial discase epiphytotic conditions. The susceptible check TMV 2 was planted as infector row on every 10th row as well as in border around the field to entice the fungal spores and to aggravate the disease development as suggested by Subrahmanyam *et al*. (1995). In order to encourage disease pressure, artificial inoculation with spraying of spore suspension was done at 30 days after sowing. Rust urediniospores were isolated by soaking and rubbing of infected leaves in water for 30 minutes. The filtered inoculum contained 20,000 urediniospore per ml suspension mixed with tween 8 (0.2 ml per 1.2 litres of water) as mild surfactant was sprayed on the plants using Knapsack sprayer in the evening and high humid condition was created by frequent spraying of water for three days following inoculation. Incidence of rust was recorded on 1-9 scale as suggested by Subbarao *et al*., 1990

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Five components of rust resistance were measured *viz*., 1. *Incubation period:* Number of days taken from inoculation to appear 50 % of the pustules on leaf surface area. 2. *Sporulation index:*It was measured on a 1-5 scale as given by Mehan *et al.,* (1994) where, 1 = No sporulation evident, 2 = 1-25 per cent pustules area covered with spores, 3 = 26-50 per cent pustules area covered with spores, 4 = 51-75 per cent pustules area covered with spores and 5 = 76-100 per cent pustules area covered with spores. A rating of 5 indicates that the uredinium was fully open and entire uredinium was covered with urediniospores. 3. *Number of pustules per leaf area:* Total numbers of pustules on entire leaf area were counted and recorded. 4. *Leaf area damage* (%)*:* It was estimated by comparing leaves with diagrams depicting leaves with known percentages (0.5, 5, 10, 20, 35, 50, 75 and 100 %) of their areas affected (%). 5. *Ruptured pustules* (%): Mean number of uredosori ruptured at 30 days after inoculation was recorded and it was expressed in percentage.

Genomic DNA isolation was carried out with 2 g of tender leaf tissue from recombinant inbred lines and backcross (BC_1F_4, BC_2F_3 and BC_3F_2) inbred lines and their respective parents using "cetyltrimethyl ammonium bromide (CTAB)" method (Saghai-Maroof *et al*., 1984) (buffer containing 1M Tris-HCL buffer pH 8, 4M NaCl, 0.5M ethylene diaminetetraacetic acid (EDTA) with few modification). DNA quality was checked and quantified on 0.8% agarose gel with known concentration of uncut lambda DNA as standard.

Polymerase Chain Reactions (PCR) were performed by using a Touch-Down PCR profile and DNA amplification was performed in 20 µl reaction mixture containing 20ng/µl template DNA (1 µl), 10 pM / µl SSR primer pair (0.5 µl each Forward and Reverse), 2 mMdNTP's(1 µl), 25 mM $MgCl_2$ (Qiagen)+10X PCR buffer (2 µl) (Qiagen), 5U/µl Taq DNA polymerase (0.33 µl) (Qiagen) and water(14.67 µl) . Touch - Down PCR amplification using a program, in which the annealing temperature is lowered from 65 to 60 by 1°C every cycle, followed by 40 additional cycles at 59°C. After initial denaturation for 5 min at 95°C each cycle comprised 30 sec. denaturation at 94°C, 45 sec. anneaniling at 65°C and 1 min. extension at 72°C with final extension for 10 min. at 72° C at the end of 40 cycles.

The PCR products were mixed with 2µl of loading dye (0.25% bromophenol blue with 40% sucrose) and were loaded into each well and separated on 1.4 per cent agarose gel using 1X TAE buffer of pH 8.0 containing ethidium bromide. The gel was documented using white/2UV Trans-illuminator of Ultra Violet products, London. The agarose did not give high resolution for low size PCR products but the markers that showed less base pair size difference on agarose were arrayed on 4% metaphore agarose gel.

Totally one hundred and fifty gene specific SSR primers were used for screening between two parental genotypes ***viz.***, GPBD-5 and ICGV 86699. Single marker analysis (SMA) (Haley and Knott, 1992) was performed to tag and confirm potential SSR markers linked to the trait based on phenotypic and genotypic data pertaining to the RILs and backcross populations.

An analysis of variance (ANOVA) was carried out separately for each component of rust resistance. Genotypic coefficient of variation and phenotypic

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coefficient of variation were computed by the method suggested by Burton and Devane (1953), heritability by Hanson *et al.,* (1956), genetic advance (Robinson 1949), genetic as a per cent over mean (GAM %) were worked out as suggested by Johnson *et al.,* (1955).

Results and Discussion

There was decrease in the mean rust score disease that can be considered as desirable trait and it was pronounced in recombinants derived from cross of RILs (F_6) with mean rust score of 3.85 on 1-9 scale in RILs (F_6) compared to mean rust score of 4.3 in recombinants derive from BC_1F_4, 4.33 in BC_2F_3 and 4.32 in BC_3F_2 derived recombinants. The range of rust score (3-8.5) and incubation period (7.5-27 days), number of pustules per leaf area (1-16-7.5), sporulation index (1-5), leaf area damage (0.75-7.56 %), mean number of ruptured pustules (0.53-7.56) were quite broad in recombinants derived from both RILs (F_6) and backcross population. This indicated that presence of recombinants with reduced rust resistance in RILs (F_6) and backcross derived recombinants to make effective selection for this trait.

The difference between PCV and GCV values were high for sporulation index recorded in recombinants derived from direct as well as all backcross derived recombinant lines, mean rust score in F_6 and BC_1F_4 derived recombinants indicating higher environmental influence in expression of these traits in the population. High genetic variation along with high heritability and Comparison of genetic advance as per cent mean value in recombinants derived from RILs and backcross population revealed very higher expected genetic mean for all component traits (Table 1).

Number of pustules per leaf area, sporulation index, per cent leaf area damage and ruptured pustules were significant positive correlation with each other and with mean rust scores in recombinants derived from RILs (F_6) as well as backcross (BC_1, BC_2 and BC_3) population. Incubation period was negatively and significantly correlated with all other components and with mean rust scores in recombinants derived from direct backcross generations at both genotypic and phenotypic level. Association analysis for components of rust resistance indicated that longer incubation period, low number of infection sites coupled with low sporulation resulting in low leaf area damage (%) which imparts better resistance in genotypes and thus these can be considered as important components of rust resistance that can helps in identification of resistant lines (Table 2).

In F_6 population recombinant line number 43 had infected after 15 days (incubation period) with 2.50 numbers of pustules per leaf area, rating 1 sporulation index and it is immune (3.5 rust score) reaction to rust. Recombinant line number 8 was infected very late 19.5 days after incubation and it had only 8 numbers of pustules per leaf area with 1 rating sporulation index and leaf area damage was only 0.9 per cent and 0.5 per cent ruptured pustules in BC_1F_4 population. In BC_3F_2 population recombinant line number 12 even though it infected 19 days after incubation with more (21.5) numbers of pustules per leaf area but sporulation index is 1. All recombinants (resistant lines) mentioned here, the rust resistant components (5 components of rust resistance) tend to reinforce one another. It is believed that long incubation period and low sporulation index slow down rust development and production of urediospores in the field (Table 3a and 3b).

In the present investigation also ICGV 86699 is highly resistant to rust showed much longer (> 25 days) incubation period as compared to earlier report (12.33 days) by Dwivedi *et al.*,(2001). The possible causes of this variation are the pathogen population, variation in temperature and humidity. All these can substantially influence components of resistance, particularly incubation period, sporulation index and leaf area damage. Mehan *et al.,* (1994), who reported >60 % leaf area damage and Liao *et al.,* (1990) reported 82-83 per cent leaf area damage. It is noteworthy that the susceptible check TMV 2 showed much greater leaf area damage (>98 %) in present study. The fact that components of rust resistance are not fully complementary is highlighted by several recombinants, recombinant line number 4 had more incubation period 20 days and more numbers pustules (21.50) and leaf area damage 2.5 per cent and 1.5 per cent ruptured pustules in F_6 population.

From above results it is emphasized that some recombinants may have partial resistance due to all components, whereas others have partial resistance due to some of the components. Thus while there may be a correlation among components (reinforcement), certain lines contribute genes for different traits in cross used to develop lines with better resistance.

The variation existed in the all population for components of rust resistance and mean rust scores was represented graphically using bar chart from each direct (F_6) and backcross BC_1F_4, BC_2F_3 and BC_3F_2 populations. The recombinants including parents (GPBD 5 and ICGV 86699) resistant variety ICGV 99005 and GPBD 4 and also a susceptible check (TMV 2) were plotted on *X*-axis against incubation period (days) number of pustules per leaf area, leaf area damage and ruptured pustules (%) and sporulation index and

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mean rust score (score shown by line diagram) on *Y*-axis with equal class intervals (Fig.1 to 4).

In the present investigation, 150 specific SSR primers were used for screening parental genotypes *viz.*, GPBD-5 and ICGV 86699. Out of 150 primers only 23 primers were polymorphic indicating low polymorphism in the parental genotypes used for the study using SSR markers. SSR primers polymorphic between the parents of the tagging population were subjected to bulk segregant analysis to identify putatively linked markers for rust disease resistance. Out of the 23 SSR markers that were polymorphic between the parents. Only two SSR markers were found to be polymorphic between resistant and susceptible bulks. For reconfirmation of these two markers were analyzed on individual eight extreme resistant and susceptible plants. Of these two markers one SSR primer (TC4g10) (Plate 1) was found to be polymorphic indicating that this marker is putatively linked to rust disease resistant gene. Rust resistant lines were identified based on this marker in recombinant inbred lines (F_6) population and backcross (BC_1F_4, BC_2F_3 and BC_3F_2) populations (Plate 2).

Single marker analysis (SMA) was used simple linear regression method to find out the significant marker trait. Single marker analysis revealed that TC4g10 marker accounted for 72.40 per cent variation in F_6 population, 67.10 per cent in BC_1F_4 population, 38.40 per cent in BC_2F_3 population and in BC_3F_2 population, and 61.30 per cent of the total variation for the rust resistance (Table 5). Resistance to rust reported to be governed by recessive genes. MAS can save one generation of selfing to select recessive genes using linked markers.

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Table 1: Genetic variability for component of rust and Mean rust score (1-9 scale) in F_6, BC_1F_4, BC_2F_3 and BC_3F_2 population of GPBD-5 x ICGV 86699

Components of rust	Mean				Range				PCV (%)			
Populations	**F_6**	**BC_1F_4**	**BC_2F_3**	**BC_3F_2**	**F_6**	**BC_1F_4**	**BC_2F_3**	**BC_3F_2**	**F_6**	**BC_1F_4**	**BC_2F_3**	**BC_3F_2**
Incubation period (days)	17.73	17.41	18.22	18.35	8.5-27	7-29	8-26	6.5-27	20.52	29.04	26.61	26.89
Number of pustules per leaf area	22.92	30.72	28.30	28.38	1-170	1-173.5	1-167.5	1-160.5	160.11	167.56	179.21	173.10
Sporulation index (1-5 scale)	1.55	1.69	1.750	1.64	1-5	1-5	1-5	1-5	60.01	73.08	72.49	77.96
Leaf area damage (%)	1.62	1.85	2.00	1.86	0.4-3.38	0.75-7.75	0.5-7	0.75-8	42.98	115.58	102.57	116.65
Ruptured pustules (30 DAI)	1.38	1.57	1.60	1.61	0.53-8.5	0.54-7.56	0.55-7.8	0.4.3-7	115.45	138.90	136.26	139.11
Mean rust score (1-9 scale)	3.85	4.3	4.33	4.32	3-9	3-8.5	3-8	3-7.5	27.76	30.63	30.11	31.39

Components of rust	GCV (%)				h^2 bs (%)				GA				GAM (%)			
populations	**F_6**	**BC_1F_4**	**BC_2F_3**	**BC_3F_2**	**F_6**	**BC_1F_4**	**BC_2F_3**	**BC_3F_2**	**F_6**	**BC_1F_4**	**BC_2F_3**	**BC_3F_2**	**F_6**	**BC_1F_4**	**BC_2F_3**	**BC_3F_2**
Incubatio period (days)	20.30	28.87	26.40	26.76	97.91	98.80	98.44	99.00	7.34	10.29	9.83	10.06	41.39	59.11	53.96	54.85
Number of pustules per leaf area	159.96	167.43	178.96	172.97	99.81	99.85	99.72	99.84	75.47	105.88	104.21	101.05	329.20	344.66	368.16	356.03
Sporulation index (1-5 scale)	56.29	70.76	67.73	75.71	87.98	93	87.31	94.31	1.69	2.39	2.28	2.49	108.77	141.16	130.38	151.47
Leaf area damage (%)	41.95	115.54	102.28	116.49	95.29	99.94	99.43	99.73	1.36	4.40	4.21	4.46	84.37	237.95	210.09	239.65
Ruptured pustules (30 DAI)	115.13	138.77	136.12	139.02	99.46	99.82	99.80	99.87	3.26	4.49	4.48	4.62	236.53	285.63	280.13	286.19
Mean rust score (1-9 scale)	26.06	30.50	29.64	31.26	88.16	99.20	96.93	99.20	1.94	2.69	2.60	2.77	50.42	62.59	60.12	64.14

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Table 2: Phenotypic (above the diagonal) and genotypic (below the diagonal) correlation coefficient for component of rust and Mean rust score (1-9 scale) in F_6, BC_1F_4, BC_2F_3 and BC_3F_2 populations

Components of rust	Populations	GPBD-5 x ICGV 86699					
		PI	NP	SI	LAD	RP	MRS
PI	F_6	1.000	-0.66**	-0.65 **	-0.61**	-0.69 **	-0.66**
	BC_1F_4		-0.71**	-0.60**	-0.73**	-0.71**	-0.75**
	BC_2F_3		-0.86**	-0.81**	-0.84**	-0.84*	-0.87**
	BC_3F_2		-0.85**	-0.80**	-0.85**	-0.85**	-0.87**
NP	F_6	-0.66**	1.000	0.93 **	0.42 **	0.98 **	0.90**
	BC_1F_4	-0.71**		0.97**	0.98**	0.99**	0.95**
	BC_2F_3	-0.84**		0.97**	0.98**	0.99**	0.96**
	BC_3F_2	-0.85**		0.97**	0.98**	0.99**	0.96**
SI	F_6	-0.69**	0.99**	1.000	0.31	0.92**	0.84**
	BC_1F_4	-0.69**	0.99**		0.94**	0.97**	0.94**
	BC_2F_3	-0.85**	0.99**		0.98**	0.98**	0.94**
	BC_3F_2	-0.84**	0.91**		0.98**	0.98**	0.94**
LAD	F_6	-0.18	0.42**	0.36	1.000	0.36	0.22
	BC_1F_4	-0.73**	0.98**	0.98**		0.97**	0.93**
	BC_2F_3	-0.85**	0.99**	0.98**		0.99**	0.94**
	BC_3F_2	-0.85**	0.98**	0.99**		0.99**	0.95**
RP	F_6	-0.69**	0.98**	0.98**	0.37	1.000	0.91**
	BC_1F_4	-071**	0.99**	0.99**	0.97**		0.95**
	BC_2F_3	-0.83**	0.99**	0.99**	0.99**		0.94**
	BC_3F_2	-0.86**	0.99**	0.99**	0.99**		0.95**
MRS	F_6	-0.71**	0.96**	0.93**	0.26**	0.97**	1.000
	BC_1F_4	-0.76**	0.95**	0.97**	0.94**	0.96**	
	BC_2F_3	-0.84**	0.97**	0.98**	0.97**	0.97**	
	BC_3F_2	-0.88**	0.97**	0.98**	0.95**	0.96**	

PI-Incubation period (days) **SI- Sporulation index (1-5 scale)** **LAD- Leaf area damage (%)** **NP-Number of pustules per leaf area**

RP-Ruptured pustules (30 DAI) **MRS- Mean rust score (1-9 scale)**

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Table 3a: Mean components of rust resistance and rust score of selected recombinant lines and backcross population in peanut

Recombinant lines		Incubation period (days)		Number of pustules per leaf area		Sporulation index (1-5 scale)		Leaf area damage (%)		Ruptured pustules (30 DAI)		Mean rust score (1-9 scale)	
F_6	BC_1F_4	F_6	BC_1F_4	F_6	BC_1F_4	F_6	BC_1F_4	F_6	BC_1F_4	F_6	BC_1F_4	F_6	BC_1F_4
42	10	15.0	17	2.00	13	1.0	1	2.00	1.0	2.00	2.0	4.00	4.0
43	8	15.0	19.5*	2.50	8	1.0	1	2.5	2.5	2.5	2.0	3.50	4.0
40	12	16.0	23.5**	6.00	9.5	1.0	1.5	1.5	2	1.5	2.5	4.00	4.0
4	13	20.00*	15.5	21.50	3.5	1.00	1.5	2.5	2.0	1.5	2.5	4.00	4.0
32	35	17.00	15.5	13.00	13.5	1.00	1.5	2.0	2.5	3.0	2.0	3.50	4.0
Mean		17.55	17.41	22.58	32.38	1.44	1.91	2.86	2.86	2.39	2.88	3.82	4.30
GPBD-5		9.00	8.5	161.50	173.5	5.00	5	90.5	100	95.5	90.5	7.00	7.0
GPBD-4		19.00	20.5	17.00	10.5	1.00	1	1.5	1.0	1.0	1.0	4.00	4.0
TMV2		7.50	7.5	167.50	167.5	5.00	5	100.0	100	100.0	100	8.50	8.5
ICGV86699		22.00	22	1.00	1	1.00	1	1.0	1.0	1.0	1.5	3.00	3
ICGV99005		27.00	27	1.50	1.5	1.00	1	1.0	1.0	1.0	1.5	3.00	3
CD at 5%		1.49	1.65	4.49	5.90	2.57	0.62	1.35	0.80	1.32	1.60	1.03	0.35
CD at 1%		2.00	2.27	6.03	8.11	3.44	0.85	1.81	1.10	1.94	2.01	1.38	0.48

* - Significant at 5% probability level **- Significant at 1% at probability level

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Table 3b: Mean components of rust resistance and rust score of selected recombinant lines and backcross in population of peanut

Recombinant lines		Incubation period (days)		Number of pustules per leaf area		Sporulation index (1-5 scale)		Leaf area damage (%)		Ruptured pustules (30 DAI)		Mean rust score (1-9 scale)	
BC_2F_3	BC_3F_2	BC_2F_3	BC_3F_2	BC_2F_3	BC_3F_2	BC_2F_3	BC_3F_2	BC_2F_3	BC_3F_2	BC_2F_3	BC_3F_2	BC_2F_3	BC_3F_2
15	11	21*	21.5*	9.5	5.5	1	1	2.0	2.5	2.0	2	4	4
14	15	20	20	17	3.5	1	1	1.5	1	2.0	1.5	4	4
1	28	17	16	13	8.5	1	1	1.0	2.5	2.0	2	4	4
25	12	15.5	19	8	21.5	1.5	1	2.0	1.5	2.5	1.5	4	4
42	38	17	22	10	11.5	1.5	1.5	1.0	2.5	2.0	1	4	4
Mean		18.22	18.35	28.80	30.05	1.86	1.94	3.19	3.11	3.00	2.88	4.33	4.32
GPBD-5		7.5	7.5	165.5	146.5	5	5	100	95.5	95.5	95.5	7.0	7.0
GPBD-4		22	19.5	2.5	3.5	1.5	1	1	1.5	1	1	4.0	4.0
TMV2		7.5	7.5	167.5	167.5	5	5	100	100	100	100	8.5	8.5
ICGV86699		22	22	1	1	1	1	1	1	1.5	1.5	3	3
ICGV99005		27	27	1.5	1.5	1	1	1	1	2	2	3.5	3
CD at 5%		1.27	1.47	5.69	6.37	0.80	0.53	0.92	1.57	1.44	1.17	0.48	0.36
CD at 1%		1.75	2.03	7.82	8.78	1.10	0.73	1.26	2.17	1.98	1.61	0.66	0.50

* - Significant at 5% probability level **- Significant at 1% at probabil

Table 4: Single marker analysis of TC4g10 SSR marker with rust resistance in recombinant inbred lines (F_6) and backcross inbred lines (BC_1F_4 , BC_2F_3 and BC_3F_2) of GPBD 5 X ICGV 86699

Trait	Marker	Populations	R^2 adjusted (%)
Rust	TC4g10	F_6	72.40
		BC_1F_4	67.10
		BC_2F_3	38.40
		BC_3F_2	61.30

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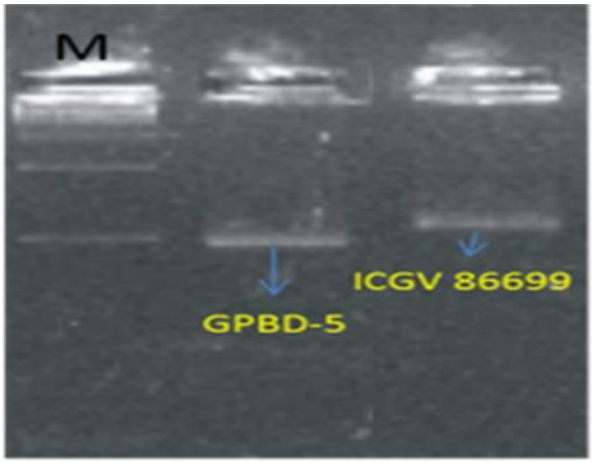

Plate 1. TC4g10 primer showing polymorphism between parent GPBD-5 and ICGV 86699

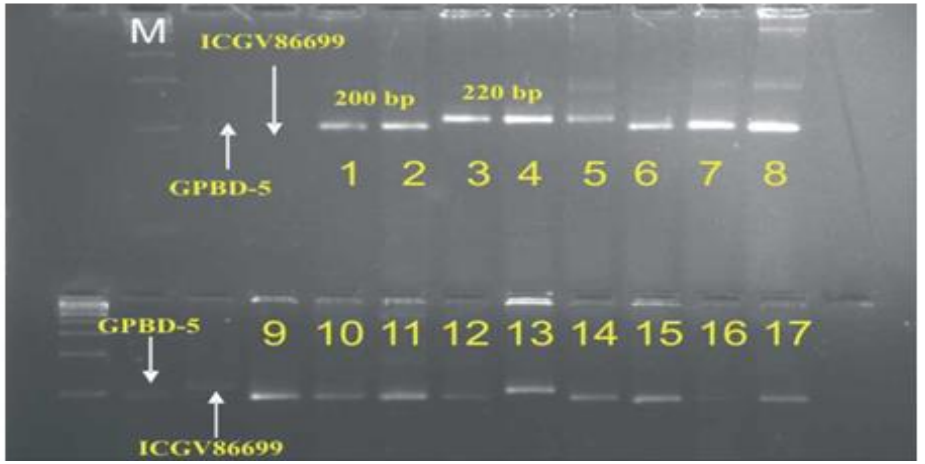

Plate 2. Tc4g10 showing polymorphism between resistant(RR) and susceptible (SS) in BC_2F_3 population of GPBD 5 X ICGV 86699

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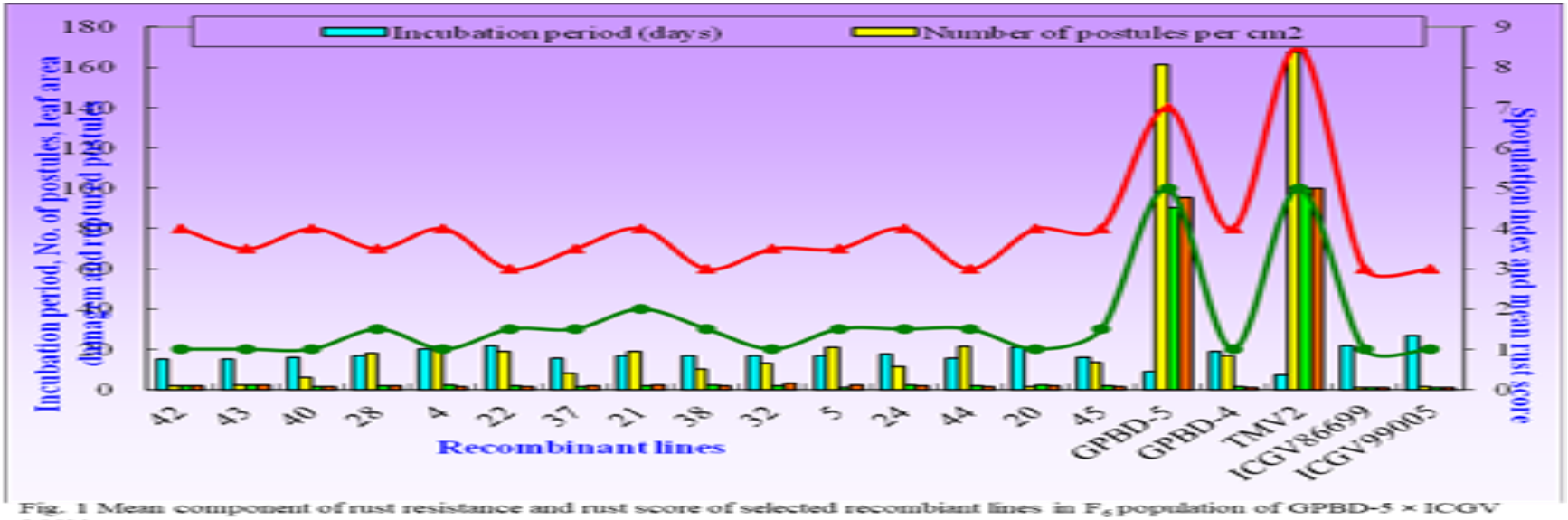

Fig. 1 Mean component of rust resistance and rust score of selected recombiant lines in F_6 population of GPBD-5 × ICGV 86699

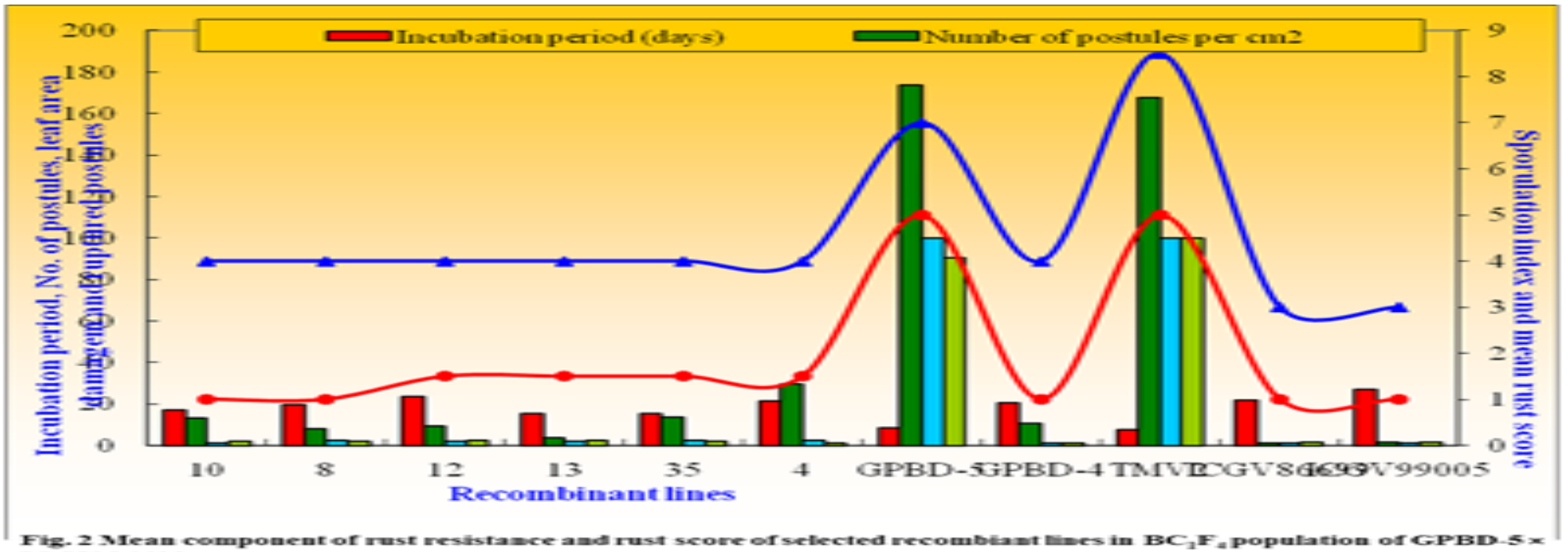

Fig. 2 Mean component of rust resistance and rust score of selected recombiant lines in BC_1F_4 population of GPBD-5 × ICGV 86699

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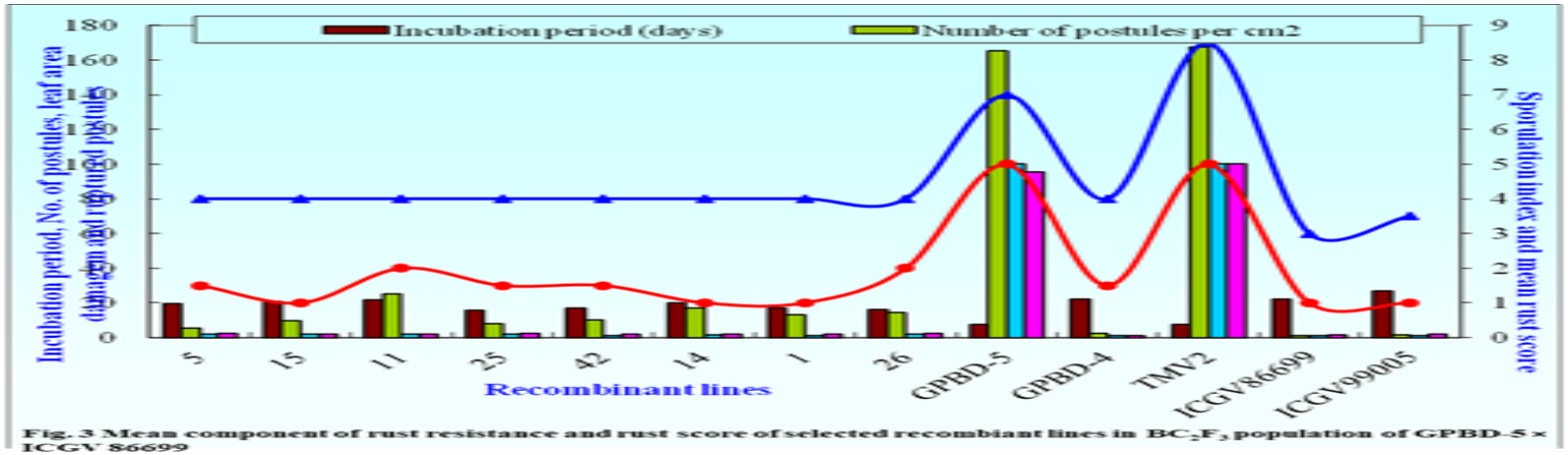

Fig. 3 Mean component of rust resistance and rust score of selected recombiant lines in BC_2F_3 population of GPBD-5 x ICGV 86699

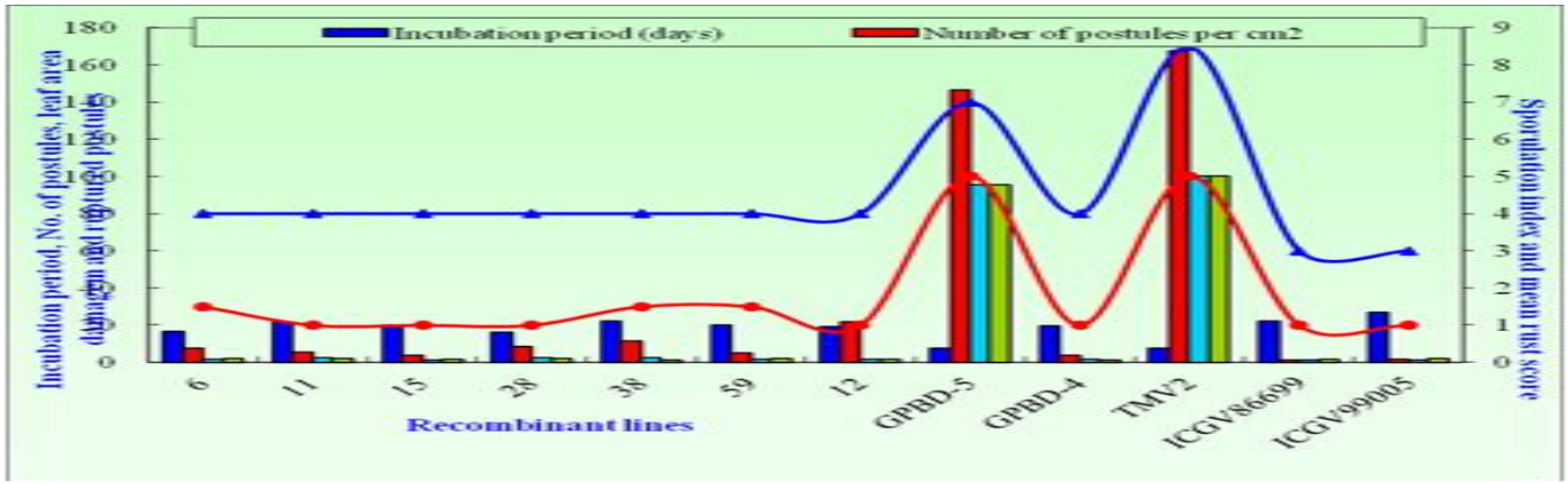

Fig. 4 Mean component of rust resistance and rust score of selected recombiant lines in BC_2F_2 population of GPBD-5 x ICGV 86699