Resistance and yield stability of groundnut cultivars on Ralstonia wilt endemic areas in central of Java, Indonesia

Novita Nugrahaeni[1]*, Joko Purnomo[1], Muji Rahayu[1], Pasupuleti Janila[2]

[1]Center of Excellence of Legumes and Tuber Crops, Indonesian Legumes and Tuber Crops Research Institutes, Jl. Raya Kendalpayak KM 8, Malang 65101, Indonesia
[2]ICRISAT, Patancheru 502 324, Andhra Pradesh, India

Abstract

Bacterial wilt caused by *Ralstonia solanacearum* is an important production constraint of groundnut over large areas in some countries of Asia including Indonesia. Seventeen wilt resistant lines including 11 breeding lines developed from ICRISAT germplasm, five improved cultivars, as well as a susceptible check (MLGG 0627) were tested for their pod yield and stability of resistance to bacterial wilt in five bacterial wilt endemic areas. The plant wilt intensity acros locations were high, indicated severe incidence of the disease. Among the improved cultivars only Gajah that showed resistance to the disease and resistance is stable across the five locations, where as the other improved cultivars were susceptible. Eight out of the 11 breeding lines were highly resistant, comparable or even higher than Gajah's resistant level. The level of resistance to bacterial wilt disease was highly contributes to the ability to deliver higher pod yield in bacterial wilt endemic areas.

Keyword: *Arachis hypogaea*, high yield, bacterial wilt, resistance.

1. Introduction

Bacterial wilt caused by *Ralstonia solanacearum* is an important production constraint of groundnut (*Arachis hypogaea* L.) over large areas in some countries of Asia including China, Indonesia, and Vietnam [1]. In Indonesia, the bacterial wilt disease has long been existed on groundnut planting areas. It was reported that since 1920 all soil in Java has been contaminated by the bacterium [2]. In a survey conducted in 1990, high disease intensity was still found in groundnut plant in West Sumatra, Lampung, West Java, Central Java, East Java, Bali and South Sulawesi [3]. These areas are accounted for almost 70% of the total groundnut production in Indonesia [4]. Yield loss caused by the disease ranges between 15-35% for resistant varieties and 60 %–100 % for susceptible varieties [5, 6] when planted under high disease intensity areas.

Farmers in wilt endemic areas, still plant old improved varieties which were released around 1950, such as Macan, Jepara, Gajah, Kidang cultivars. The lower level of resistance to wilt disease is the main reason of farmers in the area for not planting new varieties. During the period of 1950 to 2013, as many as 39 high yielding varieties of groundnut have been released, 26 of them were declared as bacterial wilt disease resistant. Of the 26-high yielding bacterial wilt resistant varieties, 20 of them derived from Schwarz 21 resistant variety, either directly or indirectly. Schwarz 21 is the first released high yielding bacterial wilt resistant variety in Indonesia by Dutch scientist who worked in Indonesia in 1925 [7]. However, resistance expression of those high yielding varieties was not on the expected level when planted in wilt endemic areas.

Areas of bacterial wilt disease are potentialy increased as the disease can be spread through seed (*seed borne*), although the rate is low, at 4 %–8 % [8], through irrigation water [9, 10], and the discovery of latent infection symptoms on resistant varieties [11]. Although many high yielding resistant varieties are available, the bacterial wilt disease is remaining a serious problem in most of the groundnut production centers in Indonesia. Ralstonia wilt disease is recently reported from areas that are not formerly reported as endemic areas, i.e. Malang, Probolinggo, Pasuruan, Tuban, and Borneo [12]. Allegedly there has been a decrease in the resistance of the old high yielding varieties [13], whereas the level of resistance of new high yielding varieties are lower than the existing local variety [14]. Komodo and Biawak the high yielding varieties grown in Malang wilted up to 80 %. Wilt intensity of 60 % occurred in Domba and Singa varieties when grown in Banjarnegara. Those suggested that ralstonia bacterial wilt resistance is critical in adoption new groundnut variety in Indonesia.

In a attempt to develop new groundnut varieties with high bacterial wilt disease resistant with high pod yield genetic background, new sources of the wilt resistant were employed in the breeding program. The new wilt resistant obtained from an extensive screening germplasm were crossed to high yielding genotypes. Development of the segregating populations (F2 to F5) were conducted in endemic areas in Banjarnegara, an endemic area. During the lines development, the susceptible genotypes were completely wilted. Selected lines were further tested for their yield and a number of promising resistant lines have been obtained. The aim of the present study was to evaluate the resistance stability and pod yield of those promising lines under Ralstonia wilt disease endemic areas.

2. Material and Methods

2.1. Planting materia

The experiments were conducted in five locations where known as endemic areas in central Java province,i.e. Tayu, Ngetuk, Blingoh, Tulakan, and Wonogiri during dry season of 2013. Planting material used in this experiment were seventeen groundnut genotypes consisted 11 breeding lines developed from ICRISAT germplasm, five improved cultivars (Gajah, Bison, Kancil, Hypoma 1, and Tuban), and a susceptible check (MLGG 0627). Those varieties are high yielding varieties with high yield potential, ranges between 2.4 and 3.7 t/ha, and are farmers preference because of the high productivity and good pod and seed characteristics. Pre-planting isolation indicated that soil bacterial populations in the experimental locations in Tayu and Ngetuk were quite high (2.1 and 2.6 x106 cfu/g), whereas that in Blingoh and Tulakan were lower (0.75 and 1.36 x106 cfu/g) [15].

2.2. Experimental design

The experiment in each location was arrange in a randomized block design, repeated three times. Each genotype was planted in a plot of 2.4m x 5m, plant spacing was 40cm x 10cm, one plant per hill. Fertilizers of Phonska 300kg/ha + 100kg SP36/ha, applied entirely at planting time

2.3. Disease rating and pod harvesting

Wilt disease observed weekly from a week after planting to harvesting time. Disease incidence was calculated based on ratio between number of wilted plants and number of plant established and multiplied by 100%. The plants resistance were classified on the basis of their percentage of disease incidence into the following categories according disease rating [16], i.e. disease incidence (DI) of 0-15% wilted plants, DI>15-25%, DI>25-35%, DI>35%, respectively for resistant, moderately resistant, moderately susceptible and susceptible categories. Plants were harvested at 90 days. Dry pod yield were converted to tones per hectare (t/ha).

2.4. *Data analysis*

Stability parameters were estimated following the Eberhart and Russell model [(1966) based on a linear model as follows:

$$Yij = Ui + Bi + Ij + dij \ , \ i = 1,2 \ , \ldots \ g \qquad (1)$$

Where Y_{ij} = yield average of line i at test site j, U_i = overall mean, B_i = slope of response of lines on locations, I_j = location index, d_{ij} = deviation of the regression ith lines on j_{th} location. A genotype with high mean seed yield, regression coefficient (b_i) close to unity and deviation from regression (δ^2_i) near to zero was defined as a stable cultivar [17].

3. Result and Discussion

First symptoms of bacterial wilt disease was observed a week after planting and the symptoms develop through harvest on susceptible genotypes. Disease symptoms initially occured on a few leaves, then the plants die suddenly in a state of the leaves are still green. The presence of dark brown color on xylem and the flow of bacteria mass of stems cross-wise in the water is a diagnostic characteristic of this disease [18]. In the resistant genotype wilt symptoms stopped at 3-4 week old plants. The highest wilt disease incidence obtained in Blingoh-Jepara, followed Tulakan, Tayu, Wonogiri, and Ngetuk (Table 1). Average rank wilt intensity was not in line with the level of bacterial populations observed in soil samples taken prior to planting [15].

Table 1. The incidence of Ralstonia wilt in groundnut genotypes at five test sites. Dry season (DS) 2013

No	Genotype	Wilt incidence (%) at					Average	Lowest value	Highest value
		Blingoh	Ngetuk	Tayu	Tulakan	Wonogiri			
1	ChiIc -1	8.97	2.68	1.24	3.05	3.37	3.86	1.24	8.97
2	ChiIc -3	3.82	0.60	2.32	3.51	0.84	2.22	0.60	3.82
3	ChiIc -8	5.18	2.75	1.57	1.80	3.22	2.91	1.57	5.18
4	LPTR -10	4.90	1.20	1.04	3.24	2.53	2.58	1.04	4.90
5	LPTR-12	2.83	1.82	1.63	0.67	1.08	1.61	0.67	2.83
6	ChiLP 14	2.61	4.92	1.71	3.20	2.10	2.91	1.71	4.92
7	LPTr -21	2.15	1.12	1.44	1.06	0.68	1.29	0.68	2.15
8	IcLP-24	8.21	1.28	1.35	0.53	0.33	2.34	0.33	8.21
9	IcLP -25	8.68	2.62	1.56	4.91	1.91	3.94	1.56	8.68
10	IcLP-27	6.10	0.89	3.37	3.57	0.47	2.88	0.47	6.10
11	Chico-s	7.80	2.36	1.57	2.53	0.91	3.03	0.91	7.80
12	Bison	54.67	26.67	28.89	58.32	35.11	40.73	26.67	58.32
13	Hypoma 1	64.71	37.99	49.10	59.00	15.64	45.29	15.64	64.71
14	Kancil	43.79	36.90	45.33	59.20	33.08	43.66	33.08	59.20
15	Tuban	63.18	33.68	40.11	67.71	46.40	50.21	33.68	67.71
16	MLGA0627	82.33	53.23	64.29	77.32	63.55	68.14	53.23	82.33
17	Gajah	11.19	0.75	1.46	0.80	0.37	2.92	0.37	11.19
	Average	22.42	12.44	14.59	20.61	12.45			

Disease incidence in the field is the result of pathogen x environment interactions. Such as occurs in aflatoxin contamination, crop x pathogen interaction is very complex because the pathogen lives in the soil and pathogen infestations occur in soil [18]. Environment (temperature and humidity), pathogen virulence and aggressiveness are all the factors that influence the interaction [19].

The wilt incidence on susceptible check genotype (MLGA0627) was high ranging from 53.23% to 82.33% with an average of 68.14%. Referring to the resistance classification [15] Gajah and the other promising lines were classified as resistant with average wilt disease intensity ranged from 1.29 to 3.94%. The average wilt intensity on Gajah was 2.92% (Table 1). All high yielding varieties tested, except Gajah, were classified susceptible with average wilt disease intensity ranging from 40.73%-50.21%. The disease intensity represented by number of plant die from bacterial wilt Ralstonia, accordingly the direct effect of the high intensity bacterial wilt is the lower plant population. That wilt effect gave rise to negative correlation between dry pod yield and intensity of wilting (Table 2). In this population, high wilt intensity obtained in genotypes with relatively larger grain size, lower plant height, and higher number of pods (Table 2). The high-intensity wilt cause lower population, plants grown at wider spacing so the opportunity to get more nutrition that enables the development of pods and seeds better. Higher dry pod yield genotypes present in relatively higher plants, higher pod number with relatively smaller seed size.

Table 2. Correlations between agronomic and Ralstonia wilt incidence in groundnut genotype tested at five endemic bacterial wilt locations. DS 2013

	Dry pod yield (t/ha)	Wilt intensity (%)	100seed weight (g)	Plant height (cm)	No. branches/ plant
Wilt intensity (%)	-0,71**				
100 seed weight (g)	-0,16**	0,25**			
Plant height (cm)	0,17**	-0,25**	-0,02ns		
No. of branches/plnt	0,02ns	0,12ns	-0,14*	0,17**	
No. filled-seed/plnt	-0,13*	0,23**	-0,08ns	-0,31**	0,36**

Variance analysis on pod yield of 17 groundnut genotypes showed the significant effects of location and genotype. Pooled variance analysis showed significant genotype x environment interaction for dry pod yield, wilt disease intensity and yield components (Table 3). G x L interaction on agronomic traits in crops , including groundnuts, have been widely reported [20, 21], likewise, the genotype x environment interaction on the intensity of wilt disease [22, 23]. Resistance to stress, both biotic and abiotic, could associated to plant performance stability across locations. Crossover interaction is freuently encountered as a manifestation of differences in disease resistance or other characters that have high heritability. The same result is also implied by [24, 25] that resistance or susceptibility to disease is one of the factors contributing to the interaction genotype and environment. The most productive environment on this test, indicated by Ij value, is Tayu, followed by Wonogiri, Ngetuk, Tulakan and Blingoh (Table 4). Productivity of the environment in line with the average intensity of wilt on genotypes tested. The highest average pod yield obtained by LPTR-12, while the highest pod yield potential achieved by ChiIc-8.

Table 3. Pooled analysis of variance over locations for some agronomic characters in groundnut.

Source of variation	Degree of freedom	Mean squares					
		Dry pods (t/ha)	Wilt intensity (%)	100 seed weight (g)	Plant height (cm)	Branch no./plant	No. of pods/plnt
Genotype (G)	16	7.00**	1.397**	569.66**	284.77**	2.08**	154.19**
Location (L)	4	10.44**	0.242**	381.78**	1907.36**	15.85**	704.60**
G x L	64	0.21**	0.016**	89.83ns	50.52**	0.63**	23.94**
Error	168	0.06	0.005	99.35	32.14	0.41	11.99

Parameter used to assess the adaptability and stability of a genotype by [17] is the regression coefficient (βi) and the deviation of the regression (δi2). A genotype is stable if it has a regression coefficient (βi) of unity and the deviation of the regression (δi2) equal to zero. Genotypes that have regression coefficient (βi) > 1 will adapt well to the environment and genotype productive with the regression coefficient (βi) < 1 will adapt well in marginal environments. Value of the regression coefficient (bi) ranged from 0.44 to 1.76 and the amount of deviation from regression (Sdi2) ranged from 0.001 to 0.183 (Table 5). All the tested genotypes have a coefficient equal to unity, except Bison, Kancil, Tuban, and MLGA 0627; while the deviation of the regression is not significant for all the genotypes.

Table 4. Average dry pod yield of groundnut genotypes in adaptation trials at five locations wilt endemic . MK 2013

No	Genotype	Dry pod yield (t/ha)					Average	Minimum	Maximum
		Blingoh	Ngetuk	Tayu	Tulakan	Wonogiri			
1	ChiIc -1	1.31	2.15	2.95	2.30	2.42	2.22	1.31	2.95
2	ChiIc -3	1.41	1.97	2.54	2.32	2.70	2.19	1.41	2.70
3	ChiIc -8	1.51	1.99	3.28	2.40	2.44	2.33	1.51	3.28
4	LPTR -10	1.60	2.15	2.79	2.22	2.46	2.24	1.60	2.79
5	LPTR-12	1.67	2.57	3.11	2.64	2.92	2.58	1.67	3.11
6	ChiLP 14	1.38	1.48	2.48	2.18	2.62	2.03	1.38	2.62
7	LPTr -21	1.53	2.40	2.83	2.11	2.72	2.32	1.53	2.83
8	IcLP-24	1.56	2.39	2.96	2.19	2.78	2.38	1.56	2.96
9	IcLP -25	1.52	1.80	2.66	2.03	2.49	2.10	1.52	2.66
10	IcLP-27	1.85	1.99	2.80	2.44	2.74	2.36	1.85	2.80
11	Chico-s	1.53	1.94	2.60	1.01	2.20	1.86	1.01	2.60
12	Bison	0.57	1.05	2.04	0.65	2.50	1.36	0.57	2.50
13	Gajah	1.55	2.09	2.72	1.92	2.63	2.18	1.55	2.72
14	Hypoma 1	0.34	0.91	1.05	0.67	1.48	0.89	0.34	1.48
15	Kancil	0.85	0.86	1.24	0.41	1.06	0.88	0.41	1.24
16	Tuban	0.79	0.82	1.29	0.48	0.92	0.86	0.48	1.29
17	MLGA0627	0.11	0.28	0.56	0.09	0.67	0.34	0.09	0.67
	Average	1.24	1.70	2.35	1.65	2.22			
	Ij	-0.59	-0.13	0.52	-0.18	0.39			

have a coefficient equal to unity, except Bison, Kancil, Tuban, and MLGA 0627; while the deviation of the regression is not significant for all the genotypesBased on the criteria, all the promising lines, except Chico-s, classified to ideal cultivars, i.e stable and have high average dry pod yield (> 2 t/ha) (Table 5). Among the improved varieties, only Gajah belongs to that criteria. Bison belongs to below average stability, which only gives a high yield in a productive environment. In this test, productive environment means low wilt disease intensity. Improved varieties are susceptible to bacterial wilt, were relatively unstable (Table 5). Among the improved varieties, only Gajah belongs to that criteria. Bison belongs to below average stability, which only gives a high yield in a productive environment. In this test, productive environment means low wilt disease intensity. Improved varieties are susceptible to bacterial wilt, were relatively unstable (Table 5).

Table 5. Stability parameters for 17 groundnut genotypes estimated by Eberhart and Russel (1966) model.

	Genotype	Average pod yield	Pod yield range		Regression coefficient (bi) 1)		Regression deviation (Sdi2)1)	
1	ChiIc -1	2.22	1.31-	2.95	1.20	ns	0.050	ns
2	ChiIc -3	2.19	1.41-	2.70	1.03	ns	0.037	ns
3	ChiIc -8	2.33	1.51-	3.28	1.29	ns	0.095	ns
4	LPTR -10	2.24	1.60-	2.79	0.93	ns	-0.004	ns
5	LPTR-12	2.58	1.67-	3.11	1.15	ns	0.030	ns
6	ChiLP 14	2.03	1.38-	2.62	1.09	ns	0.083	ns
7	LPTr -21	2.32	1.53-	2.83	1.12	ns	0.001	ns
8	IcLP-24	2.38	1.56-	2.96	1.19	ns	-0.007	ns
9	IcLP -25	2.10	1.52-	2.66	1.02	ns	-0.009	ns
10	IcLP-27	2.36	1.85-	2.80	0.88	ns	0.018	ns
11	Chico-s	1.86	1.01-	2.60	1.05	ns	0.183	ns
12	Bison	1.36	0.57-	2.50	1.76	**	0.136	ns
13	Gajah	2.18	1.55-	2.72	1.08	ns	-0.020	ns
14	Hypoma 1	0.89	0.34-	1.48	0.82	ns	0.034	ns
15	Kancil	0.88	0.41-	1.24	0.44	**	0.052	ns
16	Tuban	0.86	0.48-	1.29	0.44	**	0.037	ns
17	MLGA0627	0.34	0.09-	0.67	0.53	*	-0.007	ns

1) * and **= siginificant at p = 0,05 and p=0,01, ns = not significant

4. Conclusion

Bacterial wilt disease intensity of susceptible genotypes ranged from 53.2% to 82.3% indicates the trials location is wilt endemic. The breeding lines consistently resistant to bacterial wilt disease across the endemic areas. Eight of the 11 tested lines have comparable or even better resistance than Gajah, the most resistant improved cultivar. The wilt disease intensity of the breeding lines ranged from 1.2 to 2.9%. Stable genotypes is characterized by bacterial wilt disease resistance and high yield. Thus, the level of resistance to bacterial wilt disease contribute greatly to the ability to deliver high pod yield when planting in bacterial wilt endemic areas. Pod yield of resistant genotypes ranged from 1.86 to 2.58 t/ha, while that of susceptible genotypes ranged from 0.34 to 1.56 t/ha. There were two breeding lines (ChiIc-8 and LPTr-12) which have high yield potential, i.e > 3 t/ha), the two lines belongs to ideal cultivar, i.e. stable and high yielder.

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