The Use of Covariance Technique in Screening Mungbean Accessions for Waterlogging Resistance

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ABSTRACT

Flooding is often a problem in rainfed lowland under pre-rice or post-rice conditions. A field study was established to identify mungbean genotypes for waterlogging resistance. The experiment involved 220 mungbean accessions obtained from Chai Nat Field Crops Research Center, Thailand and the Institute of plant Breeding, UPLB, Philippines. The 220 accessions included the variety Pag-asa 5 and Pag-asa 1 as resistant (R) and susceptible (S) check, respectively. The S-check was also planted alternately after every five plots of the test genotypes to monitor variation in the level of flooding stress over an experimental area. The experiment was designed in randomized complete block with 2 replications. Plants were subjected to flooding twice, at 15 and 30 days after emergence. Flooding at each time lasted for six days. Data was analyzed by the analysis of covariance using an average of the 2 nearest S-checks as covariate.

Substantial variability among the 220 mungbean accessions was observed in most plant characteristics. Results of the analysis showed that covariance technique can be used as an efficient method to adjust for variability between the test plots. Based on yielding ability after flooded condition, six out of the 220 accessions were classified as resistant varieties, i.e., IPB M 85 22-2, IPB M 85 22-8, IPB M 79 22-108, IPB M 82 22-2, IPB M 85 18-13 and IPB M 82 21-28. Seed yields of these varieties ranged from 354.3 to 466.0 kg/ha while the yield of resistant check (Pag-asa 5) and susceptible check (Pag-asa 1) were 268.7 and 158.8 kg/ha, respectively.

Keywords : analysis of covariance, mungbean, waterlogging resistance

INTRODUCTION

breeding crops resistant to waterlogging requires an efficient method for rapid screening of a large number of varieties. Generally, field performance of plants is regarded as the most reliable standard by which crop response under excessive moisture is assessed since yielding ability and stability from season to season are the main interest of farmers. However, it is usually difficult to maintain a uniform level of flooding stress over an experimental field. In most cases, blocking also cannot adequately reduce the experimental error. Alternatively, analysis of covariance is useful as supplementary procedure to take care of sources

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of variation that cannot be accounted for by blocking (Gomez and Gomez, 1984). This paper proposes an effective use of covariance technique in adjusting variability between mungbean genotypes after flooded field condition.

MATERIALS AND METHODS

Field study was conducted during 1991 at IRRI experimental farm. The Soil was silty clay loam with pH of 6.2. Soil nitrogen was low (0.12% total N) while available P (12 ppm) and exchangeable K (0.9 me/ 100 g) were adequate. During the experiment period, minimum and maximum temperature were 21.9 and 32.1 °C, respectively: there were less rain while the relative humidity ranged from 77 to 82% (Fig. 1).

The experiment involved 220 mungbean accessions including resistant and susceptible checks obtained from Chai Nat Field Crops Research Center, Thailand and the Institute of Plant Breeding, UPLB, Philippines. The resistant (R) and susceptible (S) checks were Pag-asa 5 and Pag-asa 1, respectively, The 220 accessions were laid out in a randomized complete block design with two replications. Each replication was divided into 11 blocks wherein each block was bunded for effective control of flooding treatment. Each block had 25 plots, 20 for the materials to be evaluated and five for the S-check which was used for covariance analysis. The S-check was planted at both ends of the blocks and after every five plots. Seeds were sown in tow-row plots (40 cm apart, 3 m long) and seedlings were thinned to 21 plants per linear meter. Fertilizer consisting of 30-30-30 kg/ha (N-P_2O_5-K_2O) was incorporated during sowing. Pests and diseases were controlled with the application of decis (Deltamethrin) and Benlate (Benomyl). Furadan 3 G (2,3-dihydro-2,2-dimethyl 1-7-benzofuranyl methylcarbamate) at 0.5 kg a.i.ha^{-1}) was also applied before seeding for early insect control. Hand weeding was done twice, at 2 weeks after emergence and at the onset of flowering. Sprinkler irrigation was supplied during germination.

Plants were subjected to flooding stress twice : first, during vegetative stage (15 DAE) and second, at reproductive stage (30 DAE). During the flooding period, furrow irrigation system was provided to maintain the water depth of 3-5 cm above the soil surface. Flooding at each time lasted for six days.

Seed yield (kg/ha) was converted from total seed yield (g/plot) harvested in bulk at 12% moisture content. Yield components, i.e., number of pods per plant, number of seeds per pod and 100 seed weight were taken at random. Survival percentage was calculated from number of plants that survived after first flooding, second flooding and at harvest. Seedling and plant vigor were recorded at 10 days after emergence and at flowering, respectively using the standard rating scale of 1-9 where 1=extra vigorous seedling/ plant, 3=vigorous, 5=normal/intermediate, 7=less vigorous than normal and 9=very weak and small seedling/plant. Scoring for leaf senescence was done at flowering stage using the standard rating scale of 1-9 where 1=no or very less senescence, 3=less senescence, 5=intermediate, 7=more senescence than normal and 9=very high senescence. Plant height was measured from ten randomly selected plants during flowering stage. Days to 50% flowering and days to maturity were

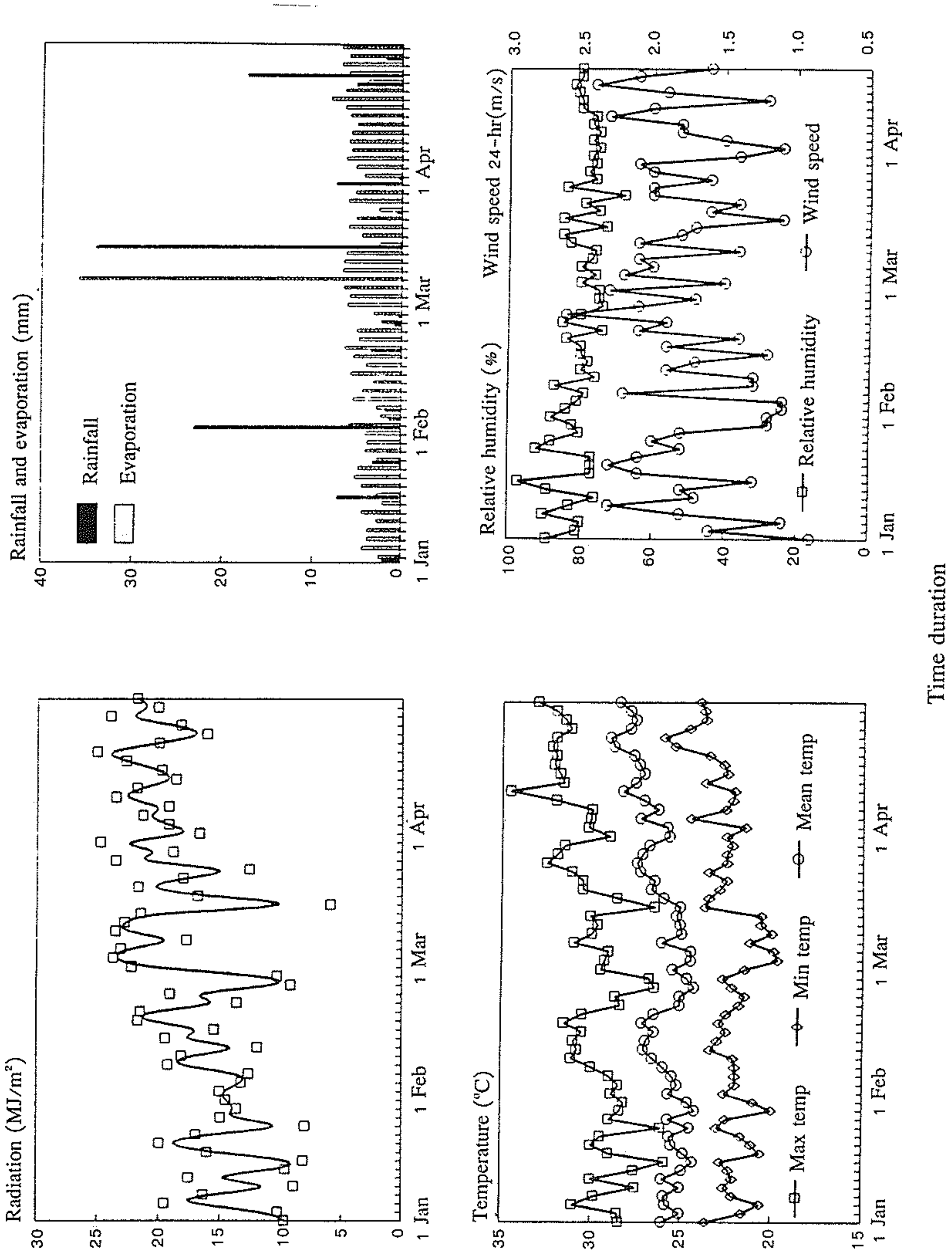

Figure 1. Meteorological data from sowing to maturity of mungbean grown at IRRI (1991).

recorded by observing the whole plot. In case of harvesting 2 times, days to maturity was computed using the formula:

$$H = \frac{H1Y1 + H2Y2}{Y1 + Y2}$$

where H = days to maturity

H1 = days from emergence to first harvest

H2= days from emergence to second harvest

Y1 = yield at first harvest

Y2 = yield at second harvest

Covariance analysis was employed using the mean value of the 2 nearest S-checks as covariate. The relative efficiency of covariance analysis (ANACOVA) over that of analysis of variance (ANOVA) was computed by the formula given in Gomez and Gomez (1984). Mean differences were determined by Least Significant Difference Test. General linear models procedure (PROC GLM) and analysis of variance procedure (PROC ANOVA) of the statistical analysis system (SAS, 1982) were used to analyze the data.

Based on mean yield difference between the test entry and the check, the accessions were classified for their reaction to waterlogging as follows : Highly resistant (HR) = significantly higher than R-check, Resistant (R) = not significantly different from R-check but significantly higher than S-check, Susceptible (S) = not significantly different from S-check, and Highly susceptible (HS) = significantly lower than S-check.

Table 1. Variation on plant traits among 220 mungbean accessions (No. of observations = 440).

Character	Minimum	Maximum	Mean	Standard deviation
1. Seedling vigor	1.0	7.0	3.7	1.0
2. Plant vigor	1.0	9.0	5.0	2.3
3. Senescence	1.0	9.0	3.4	2.2
4. Plant height at flowering (cm)	7.5	41.4	18.5	7.1
5. Days to 50% flowering	32.0	51.0	42.0	5.6
6. Days to maturity	63.0	94.0	73.6	4.7
7. % survival after first flooding	41.1	100.0	87.8	9.4
8. % survival after second flooding	32.3	100.0	78.1	14.0
9. % survival at harvest	11.2	100.0	61.7	18.1
10. No. of pods/plant	0.5	10.5	2.6	1.5
11. No. of seeds/pod	2.9	13.3	6.9	1.8
12. 100 seed wt. (g)	2.5	7.8	5.3	0.8
13. Yield (kg/ha)	2.9	964.2	165.5	185.1

RESULTS AND DISCUSSION

Variation on Plant traits

Simple statistics for all traits measured in terms of minimum, maximum and mean values including standard deviation (SD) are shown in Table 1. Seedling vigor ranged from extra vigorous to less vigorous than normal with mean of 3.7±1.0. Plant vigor ranged from extra vigorous to very weak and small plant with mean of 5.0±2.3. Leaf senescence ranged from very less to very high senescence with mean of 3.4±2.2. The greater mean of plant vigor over seedling vigor indicated flooding effect over all the test entries.

Plant height at flowering ranged from 7.5 to 41.1 cm with mean of 18.5±7.1 cm. Plant height reduction was widely observed in water logged plants of mungbean, soybean, tomatoes and corn (Del Rosario and Fajardo 1989, Del Rosario and Santos 1982) In other mungbean experiment, Aguilar and Villareal (1989) found that plant height was significantly reduced from 55.7 cm in open-upland environment to 43.6 cm under condition of exceeded water.

Days to 50% flowering ranged from 32 to 51 with mean of 42.0±5.6. the delay of flowering from the usual period (30-35 DAE) was common to all accessions. Days to maturity, ranging from 63 to 94 DAE with mean of 73.6±4.7, was also delayed from the normal period of 65-70 DAE. The delay of maturity would have significant impact on cropping pattern, especially under pre-rice condition. The pre-rice environment is characterized by puddled soil without cultivation. Planting is only done when moisture content of the soil becomes minimal but may become excessive as crop reaches near maturity (Pun 1984). To fit into this condition, suitable mungbean varieties should have early maturing character or the days to maturity is less affected by waterlogging.

Survival percentage was recorded three times : after first flooding (22 DAE), after second flooding (37 DAE) and at harvest. The mean values of percent survival decreased from 87.8±9.4 to 78.1±14.0 and 61.7±18.1, respectively while the range widened.

Yield components were recorded from 10 random plants within each plot : number of pods per plant ranged from 0.5 to 10.5 with mean of 2.6±1.5 , number of seeds per pod ranged from 2.9 to 13.3 with mean of 6.9±1.8 ; 100 seed weight ranged from 2.5 to 7.8 g with mean of 5.3±0.8 g. Much of variations was found on yielding ability which ranged from 2.9 to 964.2 kg/ha with mean of 165.5±185.12 kg/ha. The big difference on seed yields was due to variation on flooding stress level as well as differential responses among the test entries.

Analysis of Covariance

The average values of the 2 nearest S-checks (Pag-asa 1) were used as covariates in the analysis of covariance. F-test and coefficient of variation from covariance analysis were compared to those of the standard analysis of variance, and the relative efficiency was computed (Table 2).

With covariance analysis (COVA), F-test for 13 plant traits showed highly significant differences for 11 traits and significant differences for two traits. With the analysis of variance (ANOVA), F-tests for days to 50% flowering and yield were not significant. The F-values were relatively greater with covariance analysis.

Coefficient of variation (CV) was relatively small with covariance analysis indicating that the experimental error was reduced : therefore, the precision for comparing treatment means was increased. Coefficient of variation for 13 plant traits ranged from 4.8% for days to maturity to 56.9% for yield. It should be noted that CV for yield was too high. This is mainly due to big difference in water depth between replications since 80% of the area in replication I was higher than that of replication II. However, ANACOVA showed tendency of decreasing CV from that of ANOVA.

The relative efficiency for all traits was quite high ranging from 100.2 to 342.6%. This result indicates that using average of the 2 nearest S-checks as covariate would increase the degree of precision compared to the standard analysis of variance.

According to Gomez and Gomez (1984), the reaction of the susceptible check nearest to, or surrounding each test entry can be used as covariate. However, Yates (1970) suggests that the weighted mean of the two controls with weights inversely proportional to their distances would be more suitable. Thus, the suggested covariates for the first five experimental plots would be 5/6 C_1+1/6 C_2,

Table 2. Relative efficiency and the comparison of F-test and coefficient of variation (CV) from the analysis of variance (ANOVA) and covariance (COVA)

Character	F-test		CV (%)		Relative efficiency (%)
	ANOVA	COVA	ANOVA	COVA	
1. Seedling vigor	2.1**	2.1**	22.3	22.3	100.2
2. Plant vigor	1.8**	2.3**	35.2	25.0	196.7
3. Senescence	2.5**	2.5**	46.8	42.0	123.3
4. Plant height at flowering (cm)	1.3**	2.5**	30.3	16.3	342.6
5. Days to 50% flowering	1.0^{ns}	1.2*	9.6	7.4	169.2
6. Days to maturity	1.7**	1.8**	5.0	4.8	105.0
7. % survival after first flooding	1.0^{ns}	1.2**	10.0	9.0	122.7
8. % survival after second flooding	1.4**	1.7**	14.6	12.9	126.7
9. % survival at harvest	1.5**	2.0**	23.3	18.1	163.2
10. No. of pods/plant	1.4**	1.7**	48.3	34.3	196.8
11. No. of seeds/pod	1.2^{ns}	1.3**	21.2	17.9	138.9
12. 100 seed wt. (g)	7.3**	11.5**	8.1	6.4	160.5
13. Yield (kg/ha)	1.2^{ns}	1.4**	91.5	56.9	256.8

ns = not significant
* = significant at 5% level
** = significant at 1% level

Table 3. Coefficient of variation and relative efficiency corresponding to the types of covariate used for adjusting seed yield.

Types of covariate	Coefficient of variation (%)	Relative efficiency (%)
1. Average value of the 2 nearest checks	56.9	257
2. Weighted mean of the 2 nearest checks (Yates 1970)	58.6	242
3. Average plant vigor score of the 2 nearest checks	59.4	236
4. Average senescence score of the 2 nearest checks	64.4	201

4/6 C_1+2/6 C_2, 3/6 C_1+3/6 C_2, 2/6 C_1+4/6 C_2, and 1/6 C_1+5/6 C_2, respectively where C_1 and C_2 represent the 2 nearest S-checks.

Owing to systematic arrangement of S-checks in this study, four types of covariate were tested for their efficiency in adjusting seed yield. These four types are : the average value of the 2 nearest checks, the weighted mean of the 2 nearest checks as suggested by Yates (1970), the average plant vigor score of the 2 nearest checks, and the average senescence score of the 2 nearest checks. The analysis using these covariates resulted in the coefficient of variation of 56.9, 58.6, 59.4 and 64.4% respectively ; the corresponding relative efficiency was 257, 242, 236 and 201%, respectively. (Table 3). Results indicate that any of these covariates can be used in the analysis of covariance.

Classification of Genotypes for Reaction to Water logging

Reaction to waterlogging was based on the difference between adjusted yields of the test entry and the check. Out of the 220 accessions, only six accessions were resistant to waterlogging whereas the rest was classified as susceptible.

Some characteristics of the six resistant genotypes are presented in Table 4. The accessions IPB M 85 22-2 had the highest yield of 466.0 kg/ha followed by IPB M 85 22-8 (422.6 kg/ha), IPB M 79 22-108 (416.8 kg/ha), IPB M 82 22-2 (400.4

Table 4. Characteristics of six mungbean accessions classified as resistant to waterlogging.

Accession	Yield (kg/ha)	Pods/ Plant	Seeds/ Pod	100 seeds WT.(g)	%Survival at harvest
1. IPB M 85 22-2	466.0	4.3	9.1	6.2	64.0
2. IPB M 85 22-8	422.6	4.7	8.2	6.1	68.6
3. IPB M 79 22-108	416.8	4.8	6.3	5.3	72.3
4. IPB M 82 22-2	400.4	3.4	7.4	6.6	66.1
5. IPB M 85 18-13	398.0	5.5	7.3	4.9	61.4
6. IPB M 82 21-28	354.3	3.1	8.1	5.6	74.2
7. Pag-asa 5 (R-check)	268.7	3.7	7.9	5.9	57.5
8. Pag-asa 1 (S-check)	158.8	2.4	6.6	4.8	65.2
LSD.05	186.0	1.8	2.4	0.6	22.1
LSD.01	245.1	2.4	3.2	0.8	29.1

kg/ha), IPB M 85 18-13 (398.0 kg/ha) and IPB M 82 21-28 (354.3 kg/ha), compared to 268.5 kg/ha of Pag-asa 5 (R-check) and 158.8 kg/ha of Pag-asa 1 (S-check).

The six accessions had percent survival at harvest ranging from 61.4 to 74.2 which were not significantly different from R-check (57.5%). Number of pods per plant and number of seeds per pod were not significantly different from R-check, ranging form 3.1 to 5.5 pods per plant and 6.3 to 9.1 seeds per pod, respectively. Seed weight ranged from 4.9 g/100 seeds (IPB M 85-18-13) to 6.6 g/100 seeds (IPB M 82 22-2); only the lowest (4.9 g/100 seeds) and the heighest (6.6 g/100 seeds) were significantly different from R-check (5.9 g/100 seeds)

Results of this experiment indicate variability among the 220 mungbean accessions in most plant characters by which seed yield exhibited most variation regarding the range of 2.9 to 964.2 kg/ha. However, a number of variability were also observed in most plant characters of Pag-asa 1, the susceptible check, indicating that there was a non-uniformity of flooding stress level over the experimental field.

In this study the classification of genotypes for reaction to waterlogging was based solely on seed yield. It was found that the yield of pag-asa 5 (R-check) and Pag-asa 1 (S-check) were not significantly different. Only six out of 220 mungbean accessions were classified as resistant to waterlogging whereas the rest, including Pag-asa 5 and Pag-asa 1, were classified as susceptible. This is in contrast to previous studies in which Pag-asa 5 was classified as resistant variety (Del Rosario and Pandey 1985, Del Rosario and Fajardo 1989)

Covariance analysis (COVA) using average of the 2 nearest S-checks as covariate appears to be an effective method for comparing genotypes in various characters, especially seed yield. COVA results in greater F-values and smaller CV than the standard analysis of variance (ANOVA) The relative efficiency (R.E.) of COVA was quite high in most plant characters (Table 2) indicating that using COVA results in the increased degree of precision over that which would have been obtained with ANOVA

SUMMARY

Field experiment was established to identify mungbean genotypes for waterlogging resistance. Differential responses among varieties were observed in most plant characteristics. Results showed the effective use of covariance technique in adjusting variability between the test plots. Based on adjusted yield, six accessions were classified as resistant varieties, i.e., IPB M 85 22-2, IPB M 85 22-8, IPB M 79 22-108, IPB M 82 22-2, IPB M 85 18-13 and IPB M 85 21-8. The relative coefficient of covariance analysis using the average of the 2 nearest susceptible checks as covariate was 257% indicating that covariance technique would increase degree of precision rather than the analysis of variance. The technique, therefore, would be helpful to plant breeders in screening large number of accessions in the field whereby variation between the test plots would commonly be expected.

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