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Original scientific paper

MULTIVARIATE ANALYSIS AND DETERMINATION OF THE BEST INDIRECT SELECTION CRITERIA TO GENETIC IMPROVEMENT THE BIOLOGICAL NITROGEN FIXATION ABILITY IN COMMON BEAN GENOTYPES (*PHASEOLUS VULGARIS* L.)

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In order to determine the best indirect selection criteria for genetic improvement of biological nitrogen fixation, sixty four common bean genotypes were cultivated in two randomized complete block design. Genotypes were inoculated with bacteria *Rhizobium legominosarum* biovar *Phaseoli* isolate L-109 only in one of the experiments. The second experiment was considered as check for the first. Correlation analysis showed positive and highly significant correlation of majority of the traits with percent of nitrogen fixation. Step-wise regression designated that traits

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percent of total nitrogen of shoot, number of nodule per plant and biological yield accounted for 92.3 percent of variation exist in percent of nitrogen fixation. Path analysis indicated that these traits have direct and positive effect on percent of nitrogen fixation. Hence, these traits are promising indirect selection criteria for genetic improvement of nitrogen fixation capability in common bean genotypes especially in early generations.

Key words: common bean, hierarchical cluster analysis, multivariate linear regression, nitrogen fixation, nodulation

INTRODUCTION

Biological nitrogen fixation is one of the most important sources for production approximately 65% of nitrogen used in agriculture (THOMAS *et al*., 1997). Indirect selection in early generation through traits correlated with seed yield and biological nitrogen fixation is important strategy in common bean breeding.

Correlation coefficient analyses help researchers to distinguish significant relationship between traits. Step-wise regression can reduce effect of non-important traits in regression model, in this way traits accounted for considerable variations of dependent variable are determined (AGRAMA, 1996). Path analyses that present by Li (1956) have been extensively used for segregating correlation between yield and its components in field crops. Path analysis is used to determine the amount of direct and indirect effects of the variables on the dependent variable (ULUKAN *et al*., 2003).

MILLER *et al*. (1986) found that nodule weight and nodule number plant^{-1} are the best effective criteria for genetic improvement of biological nitrogen fixation in spotted bean, while pod number plant^{-1} has been proposed for this purpose in common bean genotypes (ALI *et al*., 2000). ATTEWAL and BLISS (1985) reported higher seed yield in common bean genotypes having higher amount of biological nitrogen fixation.

GHASEMI *et al*. (2005) emphasized on traits nodule number plant^{-1}, nodule weight, pod number plant^{-1} and pod weight as the most important criteria for breeding biological nitrogen fixation in common bean cultivars.

ULUKAN *et al*. (2003) reported positive and significant relationships between biological yield with plant height, pod number plant^{-1} and grain number pod^{-1} in faba bean genotypes. The total coefficient of determination was found as 63.6% in the regression model for biological yield as dependent variable. Direct effects of plant height, pod number plant^{-1} and grain number pod^{-1} upon biological yield were positive. These traits determined as selection criteria for genetic improvement of biological yield.

In this research, relationships between biological nitrogen fixation and some metric traits were investigated to determine which characters directly affected biological nitrogen fixation in Iranian common bean genotypes and determination of the effective selection criteria for genetic improvement of this trait.

MATERIALS AND METHODS

The study was carried out at the experimental fields of the department of agronomy and plant breeding, faculty of agriculture, Islamic Azad University, Khorasgan branch, Isfahan, Iran during 2009-2010. Sixty four common bean genotypes were selected from the Karadj gene bank. Seeds from each genotype were sown in 3m long rows.

The field experiment was established as the randomized complete block design. Experiment envolve two separated augmented design. In one of them seeds were inoculated with *Rhizobium legominosarum* biovar *phaseoli* isolate L-109 as check for another (GULER *et al*., 2001; KATIYAR and SINGH, 1990). In this way, trait biological nitrogen fixation is estimated by using nitrogen-difference method proposed by TAMIMI (2002). Isolate L-109 had been recognized as the compatible isolate with Iranian common bean genotypes in this province at the basis of results given from previous study (GHASEMI *et al*., 2005).

Measurements of investigated traits were done on ten normal plants, which have been randomly chosen from the middle-row of each plot. The following measurements were achieved on; seed yield plant^{-1}, seed number pod^{-1}, pod number plant^{-1}, pod yield plant^{-1}, 100-seed weight, biological yield, harvest index, nodule number plant^{-1} in 50% flowering, total nitrogen of shoot and biological nitrogen fixation. Biological nitrogen fixation was calculated using the formula suggested by TAMIMI (2002).

Relationships between traits investigated using simple correlation coefficients. Step-wise regression analysis was achieved for determination of the best model which accounted for variation exist in biological nitrogen fixation as dependent variable. Path analysis based on the method given by DEWEY and LU (1959) was done on the traits entered to regression model. The cluster analysis based on ward's method was also used to classify the traits and determine the effective criteria for genetic improvement of biological nitrigen fixation. Data analysis was done by using softwares SAS and SPSS.

RESULTS AND DISCUSSION

Correlation analysis revealed that all the traits except seed number pod^{-1} have positive and significant relationship with biological nitrogen fixation. Step-wise regression analysis for biological nitrogen fixation as dependent variable and other traits as independent variables (Table 1) indicated that traits total nitrogen of shoot, nodule number plant^{-1} and biological yield accounted for 92.3% of variation exist in biological nitrogen fixation. Amongst, trait total nitrogen of shoot accounted for 60% of variation of biological nitrogen fixation.

Path analysis for biological nitrogen fixation based on traits entered to regression model (Table 2) revealed considerable positive and direct effect of these traits on biological nitrogen fixation, while their indirect effects are smaller than direct effects. Because of that, total nitrogen of shoot, nodule number plant^{-1} and biological yield are suggested as the best indirect selection criteria for genetic improvement of biological nitrogen fixation in common bean genotypes. GHASEMI *et*

al. (2005), YADEGARI (2003) and AMINI *et al*. (2004) reported similar results for these traits.

Table 1. Step-wise regression for biological nitrogen fixation (dependent variable) in Common bean genotypes

Variable	$b_{(1)}$	S.E	R^2	t	Prob
Total nitrogen of shoot (%)	0.516	1.083	0.600	4.415	0.000
Nodule number plant^{-1}	0.325	0.403	0.762	4.002	0.000
Biological yield (gr)	0.218	2.231	0.923	2.392	0.000
Intercept	-42.214	7.503		-4.956	0.013

(1): b values have been tested relative to zero.

Table 2. Path analysis for biological nitrogen fixation in Common bean genotypes

Variable	(1)	(2)	(3)	Sum of effects
(1) Total nitrogen of shoot (%)	0.651	0.157	0.051	0.860
(2) Nodule number plant^{-1}	0.220	0.417	-0.051	0.638
(3) Biological yield (gr)	0.072	-0.073	0.549	0.550
Residual effects	0.214			

The dendrogram obtained from cluster analysis (Figure 1) showed relative consistency between the result of classify traits given by using two multivariate methods. The similar findings were obtained from other researches (ABDOLLAHI *et al*., 2009; BARRON *et al*., 1999; GHASEMI *et al*., 2005; TADESSE and BEKELE, 2001).

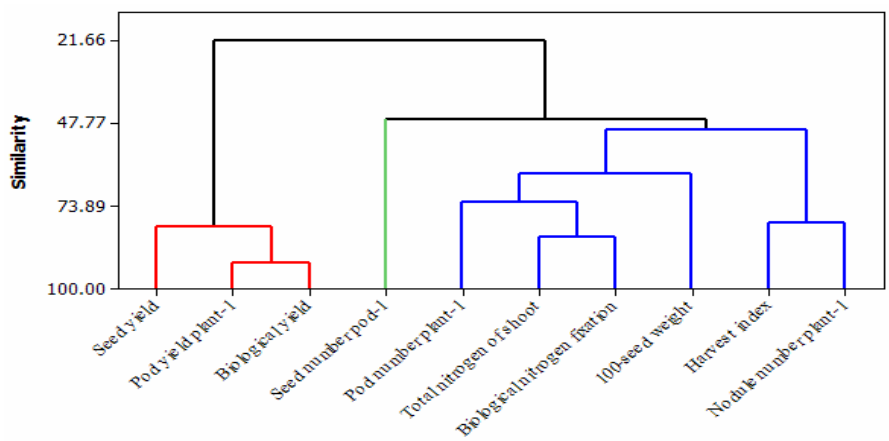

Figure 1. Dendrogram of cluster analysis to classify variables based on ward's method

In conclusion, we can suggest indirect selection in early generations via traits that have the highest direct effect on dependent variables. These traits usually determine by means of statistical procedure like correlation, regression and path analysis. In this research, revealed that traits total nitrogen of shoot, nodule number plant^{-1} and biological yield are the best indirect selection criteria for genetic improvement of biological nitrogen fixation in common bean genotypes.

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ANALIZA MULTIVARIJANSE I UTVRĐ]IVANJE NAJBOLJEG INDIREKTNOG KRITERIJUMA SELEKCIJE NA GENETIČKO POBOLJŠANJE SPOSOBNOSTI BIOLOŠKE FIKSACIJE AZOTA KOD RAZLIČITIH GENOTIPOVA PASULJA (*PHASEOLUS VULGARIS* L.)

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Da bi se utvrdili najbolji indirektni kriterijumi selekccije na genetičko poboljšanj sposobnosti biološke fiksacije azota, šesdeset četiri genotipa pasulja (*Phaseolus vulgaris* L.) su gajena u dva ista slučajna blok sistema. Inokulacija biljaka je vršena bakterijom *Rhizobium legominosarum* izolata L-109 biovarieteta *Phaseoli* samo u jednom blok sistemu dok je drugi blok sistem bio kontrola. Analiza korelacije je okazala pozitivnu i visoko značajnu zavisnost većine ispitivanih osobina i procenta biološke fiksacije azota. Stepenasta regresija je pokazala da se osobina procenta ukupnog azota biljke, broj nodula po biljci i biološki prinos se pripisuju sa 92.3 % variranju koje postoji u procentu fiksacije azota. Pat analiza ukazuje da te osobine te osobine pozitivno I direktno zavisse od fiksacije azota. Zaključeno je da su te osobine obećavajući indirektni kriterijumi sselekcije na genetičko poboljšanje fiksacije azota genotipova pasulja (*Phaseolus vulgaris* L.), posebno u ranim generacijama.

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