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## Indirect Selection for Genetic Improvement of Seed Yield and Biological Nitrogen Fixation in Iranian Common Bean Genotypes (*Phaseolus vulgaris* L.)

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**Abstract:** Common bean is one of the most important crops in majority regions of Iran. Determination of effective traits on biological nitrogen fixation and seed yield as indirect selection criteria is very important in breeding this valuable crop. For this reason 160 common bean genotypes were cultivated in two separate augmented design. Genotypes were inoculated with bacteria *Rhizobium leguminosarum* biovar *Phaseoli* isolate L-109 only in one of the experiments. Second experiment was considered as check for first. Analysis of variance indicated non-significant difference between blocks for all the traits. Correlation analysis showed that majority of the traits had positive and highly significant correlation with percent of nitrogen fixation. Step-wise regression designated that traits percent of total nitrogen of shoot, number of nodule per plant and biological yield accounted for 91.6% of variation 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. Results of correlation analysis also revealed that all the traits studied correlated positively and significantly with seed yield. Amongst, traits harvest index, biological yield, number of nodule per plant and percent of nitrogen fixation entered to regression model and verified 95% of seed yield variation. Path analysis indicated that harvest index and biological yield have considerable and positive direct effect on seed yield. Therefore, these traits recommend as the best indirect selection criteria for improvement of seed yield especially in early generations.

**Key words:** Common bean, *Rhizobium leguminosarum* biovar *Phaseoli*, biological nitrogen fixation, seed yield

### 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).

Adams (1967), Duarte and Adams (1972), Ulukan *et al.* (2003), Gebeyehou *et al.* (1982), Williams *et al.* (1990), Dofing and Knight (1992) and Guler *et al.* (2001) determined the direct and indirect effects of various plant characters on yield and yield components using path analysis in common bean, faba bean, durum wheat, navy pea, chickpea, winter barley cultivars and small grains. Katiyar and Singh (1990) determined that pod number plant<sup>-1</sup>, harvest index, grain number pod<sup>-1</sup> and grain weight were the major yield components for selection in faba bean. Dimova and Svetleva (1992) reported significant correlation between independent variables seed number plant<sup>-1</sup>,

seed number pod<sup>-1</sup>, 100-seed weight, pod number plant<sup>-1</sup> and pod weight with seed yield as dependent variable and possibility of indirect selection via these traits for genetic improvement seed yield specifically in early generations.

Altinbas (1993) emphasized on traits pod number plant<sup>-1</sup> and seed number pod<sup>-1</sup> as effective indirect selection criteria for breeding seed yield in spotted bean. Miller *et al.* (1986) found that nodule weight and nodule number plant<sup>-1</sup> are the best effective criteria for genetic improvement of biological nitrogen fixation in spotted bean, while pod number plant<sup>-1</sup> has been proposed for this purpose in common bean genotypes (Ali *et al.*, 2000). Attewell and Bliss (1985) reported higher seed yield in common bean genotypes having higher amount of biological nitrogen fixation. Ghasemi-Pirbalouti *et al.* (2005) emphasized on traits nodule number plant<sup>-1</sup>, nodule weight, pod number plant<sup>-1</sup> 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<sup>-1</sup> and grain number pod<sup>-1</sup> 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<sup>-1</sup> and grain number pod<sup>-1</sup> upon biological yield were positive. These traits determined as selection criteria for genetic improvement of biological yield.

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

## 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, Branch of Khorasgan, Iran during 2005. 160 Iranian common bean genotypes were selected from the Tehran university gene bank. Seeds from each genotype were sowed in 3 m long rows. The field experiment was established as an observed design (modified augmented design) along with Talash cultivar as control for adjusting effect of blocks on traits.

Experiment involve two separate augmented design. In one of them seeds were inoculated with *Rhizobium leguminosarum* 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 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-Pirbalouti *et al.*, 2005). Measurements of investigated traits were done on five normal plants, which has been randomly chosen from the mid-row of each plot. The following measurements were achieved on; seed yield plant<sup>-1</sup>, seed number pod<sup>-1</sup>, pod number plant<sup>-1</sup>, pod yield plant<sup>-1</sup>, 100-seed weight, biological yield, harvest index, nodule number plant<sup>-1</sup> in 50% flowering, total nitrogen of shoot and biological nitrogen fixation. Biological nitrogen fixation was calculated as follows (Tamimi, 2002):

$$\text{Bnf} = \frac{N_1 - N_0}{N_1} \times 100$$

Where;  $N_1$  and  $N_0$  consist of total nitrogen of shoot for each genotype in inoculation and non-inoculation conditions, respectively. Biological yield was considered as shoot dry weight and harvest index was estimated using below formula (Chowdhry *et al.*, 1999):

$$\text{HI} = \frac{\text{Seed yield}}{\text{Biological yield}}$$

Unbalanced one-way ANOVA was carried out for assessment of homogeneity of blocks using Talash cultivar as control. 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 seed yield and biological nitrogen fixation as dependent variables in separate analysis. After the test of homogeneity of the variance-covariance matrix of the common bean genotypes, the calculations related to path analysis were done with 160 genotypes and traits entered to regression model. In this study path analysis was carried out based on method given by Dewey and Lu (1959). Data analysis was done using software SPSS and Minitab.

## RESULTS AND DISCUSSION

Analysis of variance indicated non-significant difference between blocks, while didn't need to adjust measurements for effect of blocks. Mean and standard deviation of traits studied have been showed in Table 1. 100-seed weight has the highest standard deviation probably related to more amount of this trait.

Table 1: Mean and standard deviation for all the traits of common bean genotypes

	Seed yield (g)	Seed No. pod <sup>-1</sup>	Pod No. plant <sup>-1</sup>	Pod yield plant <sup>-1</sup> (g)	100-seed weight (g)	Biological yield (g)	Harvest index (%)	Nodule No. plant <sup>-1</sup>	Total nitrogen of shoot (%)	Biological nitrogen fixation (%)
Mean	0.23	3.18	10.05	16.47	37.32	0.64	0.37	5.27	9.13	9.63
Standard deviation	0.06	0.35	2.28	5.40	6.14	0.18	0.06	1.83	0.83	0.88

Table 2: Correlation coefficients for traits studied in common bean genotypes (n = 160)

Variables	(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
(1) Seed yield (g)	1									
(2) Seed number pod <sup>-1</sup>	0.322**	1								
(3) Pod number plant <sup>-1</sup>	0.597**	0.118	1							
(4) Pod yield plant <sup>-1</sup> (g)	0.709**	0.073	0.417**	1						
(5) 100-seed weight (g)	0.356**	0.073	0.403**	0.372**	1					
(6) Biological yield (g)	0.626**	0.125	0.295**	0.837**	0.331**	1				
(7) Harvest index (%)	0.427**	0.265**	0.288**	-0.142	0.044	-0.406**	1			
(8) Nodule number plant <sup>-1</sup>	0.388**	0.007	0.454**	0.064	0.208**	-0.203**	0.584**	1		
(9) Total nitrogen of shoot (%)	0.529**	0.212**	0.547**	0.272**	0.407**	0.216**	0.339**	0.466**	1	
(10) Biological nitrogen fixation (%)	0.524**	0.078	0.464**	0.296**	0.299**	0.272**	0.226**	0.517**	0.671**	1

\*, \*\*: Significant at 0.05 and 0.01 probability levels, respectively

Results of correlation analysis (Table 2) revealed that all traits except seed number pod<sup>-1</sup> 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 3) indicated that traits total nitrogen of shoot, nodule number plant<sup>-1</sup> and biological yield accounted for 91.6% of variation exist in biological nitrogen fixation. Amongst, trait total nitrogen of shoot accounted for 64% of variation of biological nitrogen fixation.

Path analysis for biological nitrogen fixation based on traits entered to regression model (Table 4) 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<sup>-1</sup> and biological yield are suggested as the best indirect selection criteria for genetic improvement of biological nitrogen fixation in common bean genotypes. Yadegari (2003) and Amini *et al.* (2004) reported similar results for these traits.

Correlation analysis (Table 2) showed positive and highly significant relationships of all traits with seed yield. Step-wise regression analysis for seed yield as dependent variable (Table 5) revealed that traits harvest index, biological yield, nodule number plant<sup>-1</sup> and biological nitrogen fixation accounted for 95% of variation exist in seed yield. Amongst, traits harvest index and biological yield accounted for 78% of total variation designated importance of these traits to explain variation of seed yield.

Path analysis for seed yield (Table 6) based on traits entered to regression model indicated that traits harvest index and biological yield has the highest and positive effects on seed yield. Therefore, these traits are

Table 3: Step-wise regression for biological nitrogen fixation (dependent variable) in common bean genotypes

Variable	b <sub>(1)</sub>	SE	R <sup>2</sup>	t	Prob
Total nitrogen of shoot (%)	0.466	1.091	0.641	4.395	0.000
Nodule number plant <sup>-1</sup>	0.342	0.535	0.787	3.236	0.000
Biological yield (g)	0.209	4.944	0.916	2.180	0.000
Intercept	-50.181	8.463		-5.929	0.033

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

Table 4: Path analysis for biological nitrogen fixation in common bean genotypes

Variable	(1)	(2)	(3)	Sum of effects
(1) Total nitrogen of shoot (%)	0.451	0.166	0.053	0.671
(2) Nodule number plant <sup>-1</sup>	0.210	0.356	-0.051	0.517
(3) Biological yield (g)	0.097	-0.073	0.246	0.272
Residual effects		0.667		

Table 5: Step-wise regression for plant seed yield (dependent variable) in common bean genotypes

Variable	b <sub>(1)</sub>	S.E	R <sup>2</sup>	t	Prob
Harvest index (%)	0.717	0.022	0.495	24.099	0.000
Biological yield (g)	0.921	0.009	0.781	34.244	0.000
Nodule number plant <sup>-1</sup>	0.113	0.001	0.949	3.527	0.001
Biological nitrogen fixation (%)	0.081	0.001	0.952	2.825	0.006
Intercept	-0.180	0.011		-16.904	0.000

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

Table 6: Path analysis for plant seed yield in common bean genotypes

Variable	(1)	(2)	(3)	(4)	Sum of effects
(1) Harvest index (%)	0.717	-0.381	0.081	0.007	0.426
(2) Biological yield (g)	-0.292	0.935	-0.029	0.009	0.625
(3) Nodule number plant <sup>-1</sup>	0.420	-0.191	0.139	0.018	0.388
(4) Biological nitrogen fixation (%)	0.162	0.254	0.072	0.035	0.523
Residual effects		0.187			

introduced as the effective traits for indirect selection of genotypes having higher seed yield specifically in early generations.

Duarte and Adams (1972) and Ghasemi-Pirbalouti *et al.* (2005) in common bean, Guler *et al.* (2001) in chickpea, Singh (1994) and Adak *et al.* (1999) in faba bean and Golparvar (2003) in wheat emphasized on importance of

harvest index and biological yield for genetic improvement of seed yield. Of course, Amini *et al.* (2004) reported inconsistent results for breeding seed yield in common bean genotypes.

Zubair (1985) found strong correlation between 1000 seed weight and pod number plant<sup>-1</sup> with seed yield in mungbean genotypes. The path coefficient analysis indicated that these traits had highest direct effect on seed yield among traits studied. Branch number plant<sup>-1</sup> and pod length were found to have negligible direct effect on seed yield. But the direct effect of cluster number plant<sup>-1</sup> was reduced by branch number plant<sup>-1</sup> and 1000 seed weight in spite of strong correlation with seed yield. Therefore, traits 1000 seed weight and pod number plant<sup>-1</sup> determined as indices for selection of superior genotypes in mungbean breeding programs that is adverse with our results for common bean genotypes.

Nodule number plant<sup>-1</sup> has positive and direct effect on seed yield but its indirect effect on seed yield via harvest index is positive and via biological yield is negative and considerable. Biological nitrogen fixation also exercising positive and indirect effect on seed yield especially via harvest index and biological yield. Therefore, indirect effects of traits nodule number plant<sup>-1</sup> and biological nitrogen fixation on seed yield via harvest index and biological yield must be considered, simultaneously (Farshadfar, 2000; Chaudhary *et al.*, 1999).

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<sup>-1</sup> and biological yield for biological nitrogen fixation while harvest index and biological yield for seed yield are the best indirect selection criteria in common bean genotypes.

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