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Seed Quality Effects on Plant Growth and Dinitrogen Fixation of Red Field Bean

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ABSTRACT

Field bean (Phaseolus vulgaris L.) seeds represent an important nutritional amendment to many diets of people in developing countries. Yet yields of this high protein crop are typically much lower in developing compared to developed regions. This study evaluates the effect of seed quality of an important field bean cultivar, 'Pompadour Checa,' on emergence, plant growth, yield, and dinitrogen fixation. Both naturally and artificially (1, 2, and 3 days at 41°C and 100% RH) aged seeds were planted in 1984 and 1985 in a Crosby silty clay loam (fine, mixed mesic aeric ochraqualf) with an organic matter content of 3 to 5 percent. Percent emergence and normal seedlings decreased while percent abnormal seedlings increased with increasing seed deterioration. Plant growth was evaluated at the first trifoliate (V3), first flower (R6), and pod initiation (R8) growth stages. Decreasing seed quality generally culminated in less top and root growth as assessed by dry weight. Yield was similarly reduced consonant with reductions in seed quality and this decrease was attributed to fewer seeds produced per plant since no significant decrease in individual seed weight was observed. Seeds of lower seed quality also produced fewer nodules, nodule weight, and less dinitrogen fixation as determined by the acetylene reduction assay. The observed differences in plant vegetative and reproductive growth from seeds differing in quality may be associated with the plant's ability to successfully nodulate and fix nitrogen.

Because of the scarcity and cost of quality meat, fowl, and fish, other high protein food sources must be utilized in many developing countries. The field bean, a member of the legume family, is considered one of the principal high protein crops for human consumption, especially in Latin America. As an example, of the total worldwide production of field beans, 15.4 million metric tons or 36% of total production was harvested in Latin America alone (FAO, 1984). The red field bean, 'Pompadour Checa,' eaten daily with rice (Oryza sativa L.) and corn (Zea mays L.), is the primary protein source in many developing countries in Latin America and Africa. A major problem encountered in field bean production in these regions is the low yield compared to potential yield (FAO, 1979). Reported average yields in Latin America are less than 600 kg ha^{-1} compared to monocultural yields of nearly 1,400 kg ha^{-1} in the United States and 2,700 to 4,500 kg ha^{-1} under experimental conditions in Latin America (CIAT, 1979). Bazan (1975) stated that diminished yields for many crops in Latin America were a consequence of the lack of soil nitrogen which is often not supplemented by farmers due to the high cost of nitrogen fertilizer. It is for this reason that legume production and its associated biological dinitrogen fixation becomes even more crucial to maximize crop productivity.

While inoculation with Rhizobium of field beans has enhanced yields in some cases, this has not always been the case. Other forms of biological stress also may be limiting maximum field bean yield through their effect on dinitrogen fixation. One possibility is reduced seed quality. It is well documented that seed deterioration occurs from harvest to planting and poor seed quality results in low seedling survival at planting. These problems are often more pronounced in developing countries where successful harvest and storage of high quality seeds are more difficult. Poor seed quality could lead to slower seedling and plant growth which might retard nodulation and subsequent fixation of atmospheric nitrogen. This may lead to less plant growth culminating in decreased crop yield. The objectives of this study were to determine the influence of field bean seed quality on emergence, plant growth, and yield and to evaluate the effect of seed quality on root nodulation and dinitrogen fixation.

MATERIALS AND METHODS

Seed Quality Determinations. Three seed lots of the red field bean Pompadour Checa which varied in germination and seed quality were obtained for each year of the study from foundation seed stocks in the Dominican Republic. The germination and seed quality levels of these naturally aged seed lots were determined in the following manner: Seed Germination - The standard germination test was conducted according to the AOSA Rules for Testing Seeds (1981), and the seedling vigor classification test was conducted according to procedures described in the AOSA Seed Vigor Testing Handbook (AOSA, 1983). In order to obtain controlled differences in seed quality, the naturally aged high vigor seeds were subjected to artificial aging (41°C, 100% relative humidity) (AOSA, 1983) for 0, 1, 2, and 3 days to produce seed lots of increasing deterioration.

Field Studies.

Planting - Field experiments were conducted during 1984 and 1985 at the Kenny Road OSU Agronomy Farm, Columbus, OH. The experimental design was a randomized complete block with six seed quality levels replicated three times. The six seed quality levels were high, medium, and low seed quality lots of naturally aged seeds and three artificially aged seed lots from 1, 2, and 3 days artificial aging. Captan ([(trichloromethyl)thio]-4-cyclohexene-1,2-dicarboximide) was applied to the seed surface at a rate of 2.53 g kg^{-1} seeds as a seed protectant prior to planting. The soil type was a Crosby silty clay loam (fine, mixed, mesic aeric ochraqualf) with an organic matter content of 3 to 5 percent.

Seeds were hand planted on 3 July 1984 and 9 June 1985 in 5 m rows, 10 cm between plants and 60 cm between rows with four rows per treatment. Soil implant Rhizobium phaseoli (DG-2 strain) granular inoculant (Nitragin Co., Milwaukee, WI) was applied in the seed furrows at the recommended rate of 5.61 kg ha^{-1}. Irrigation was provided as necessary to maintain adequate soil moisture. After emergence (cotyledons above the soil surface), the seedlings were thinned to 10 cm apart to provide uniform plant populations for all treatments. Normal and

abnormal seedling determinations were made according to the criteria described by ACSA (1986). Plots were hand weeded as necessary.

Sampling - Developmental growth stages of field beans proposed by CIAT (1983) were used at three sampling times: V3, R6, and R8 growth stages. V3 was the plant displaying the first trifoliate leaf which was completely unfolded (about 30 days after planting (DAP) for 1984 and 22 DAP for 1985), R6 was the plant showing the first opened flower (about 45 DAP for 1984 and 40 DAP for 1985), and R8 was the plant initiating pod fill with the oldest pods having developed seeds (about 70 DAP for 1984 and 65 DAP for 1985). Yield determinations consisted of pod number, number of seeds, and weight of seeds which were assessed at the R8 growth stage. Seed yields were determined by harvesting two square meters of plots and the seed moisture content adjusted to 15% moisture. The top and root portions of the plants were harvested at each of the three growth stages and dried immediately after harvest in a forced-air oven for 72 h at 65°C. Nodule dry weights were similarly obtained after the nodules were removed, weighed, and counted.

Acetylene Reduction Assays. Samples were taken on sunny days in the late morning or early afternoon to ensure that photosynthesis was occurring. Five plants from each of the inner rows for each treatment were selected. A 10 x 15 cm soil core containing the plant roots was collected for each of the five plants. The roots were shaken and the loose soil crumbled over sieves to retrieve any detached nodules. The shoot was separated from the root at the soil-air interface and the roots placed in 470 mL wide mouth, gas-tight mason jars sealed with a screw-cap lid fitted with a rubber sampling plug. Acetylene (60 mL) was added to create a slight positive pressure and the samples incubated at ambient temperature. After 5, 10, and 15 min, samples were taken in sterile, 4 mL vacutainer tubes (Becton, Dickinson & Co., Rutherford, NJ) as described by Streeter (1974). The ethylene content of the sample was determined with a Hewlett Packard 5720A gas chromatograph equipped with a flame ionization detector (FID) set at 175°C and the results quantified with a Columbia Scientific Industries 38 integrator for the 1984 analyses. For the 1985 analyses, a Varian Model 3700 gas chromatograph equipped with a FID and set at 175°C

was used and peaks quantified with a Hewlett Packard 3392A integrator. In both years, a stainless steel column, 95 cm length and 0.5 cm i.d., packed with 80/100 mesh alumina was used. Helium, at a rate of 30 mL min^{-1}, was the carrier gas. Column temperature was 150°C. Samples (1.0 mL) were injected into the chromatograph and the ethylene produced was determined from the integrated peaks using an ethylene standard.

Statistical Analyses. Analysis of variance and least significant difference (LSD) tests were performed for mean separations of all data according to Steele and Torrie (1980).

RESULTS AND DISCUSSION

The standard germination and seedling vigor classification tests demonstrated that significant differences existed in the seed quality of the field bean seed lots for the two years of the study (Table 1). When the hiqh quality naturally aged seed lot was subjected to artificial aging for up to three days, expected differences as assessed by the seed quality tests were observed with the one day aging seed lot performing better than the seed lot subjected to three days aging. The number of seeds planted in the field was adjusted according to these laboratory results in order to approach uniform stands for all seed quality levels. These values and the number of seedlings emerged and percent emergence as well as the percent normal and abnormal seedlings for each seed quality level as a result of natural and artificial aging are presented in Tables 2 and 3. While the seedlings emerged were essentially equivalent for each seed quality level as a consequence of the changes in seeding rate; on a percent emergence basis, decreased seed quality led to an overall reduction in field emergence, decreased percent normal seedlings, and a concomitant increase in percent abnormal seedlings (Tables 2 and 3).

These results demonstrate that seed deterioration in field bean is a significant seed quality problem with respect to field emergence. Field bean seed quality can be reduced by a number of factors. For example, Siddique and Goodwin (1980) studied the influence of maturation temperature on seed quality for 10 snap bean genotypes and their susceptibility to mechanical damage. They found that better quality seeds and resistance to mechanical injury can be obtained when seeds matured at low temperatures. Temperatures in the range of 18/13°C and 21/16°C during seed maturation were ideal for the production of high quality and mechanically damage resistant bean seeds for a number of genotypes. Harvesting field bean seeds prior to maturity also influences seed quality. Wijandi and Copeland (1974) showed significant reductions in germination and seedling vigor when field beans were harvested early. Thus, differences in field bean seed quality can be observed naturally and have a subsequent effect on field performance.

While no reports to our knowledge are available demonstrating the effect of artificial aging on field bean performance, Edge and Burris (1971) investigated this relationship by

reducing the vigor of high quality soybean [Glycine max (L.) Merr.] seed lots through artificial aging for periods up to 16 days. They found a decrease in field emergence and stand establishment with increased artificial aging as was observed here for field beans.

Differences in the number of days to seedling emergence varied among both naturally and artificially aged seeds. For example, high quality naturally aged seeds emerged faster than did low quality naturally aged seeds (5 and 9 days relative to 6 and 11 days for 1984 and 1985, respectively). One day artificially aged seeds required 6 and 10 days for emergence whereas three days artificially aged seeds required 7 and 14 days to emerge during 1984 and 1985, respectively. Similar results have been reported for soybeans (Smith and Ellis, 1980) which show that decreased seed quality retards emergence and slows seedling growth. While the exact mechanisms governing this reduced growth are unknown, these changes in metabolism may be associated with physiological events governing seed deterioration (McDonald and Nelson, 1986). It is known that proteins play an important role in seed germination and seedling development. Grange (1980) reported changes in solubility properties of proteins with age in field bean seeds indicating that alterations in protein structure indeed occur. These may hinder optimum functioning of germination specific enzymes. Reductions in ATP levels also have been observed in other seeds which have been related to either naturally or artificially induced seed aging (VanOnckelen et al., 1974; Anderson, 1977). Similar reductions in ATP levels may occur as well in field bean and imply reductions in the synthesis of many cellular constituents such as proteins, lipids, and carbohydrates that use ATP as their major energy source.

It also was of interest to determine how persistent this seed quality effect was on later plant performance. To assess this, determinations were made of top and root growth (Table 4). Top and root dry weight were significantly affected by seed quality for the three growth stages examined in 1984 and 1985 with the exception of root dry weight at the V3 stage in 1984. In all other instances, decreased seed quality led to overall reductions in vegetative growth under both aging regimes. Maximum top and root dry weights were generally noted at the R8 growth stage.

Pod number, pod dry weight, and yield decreased with decreasing seed quality for both naturally and artificially aged seeds over the two-year study (Table 5). In contrast, no significant

difference in weight per 100 seeds for the differing seed quality levels was observed. Thus, the decrease in yield was attributed to decreased seed number as a consequence of fewer pods being produced rather than a significant decrease in seed weight.

The results presented here indicate that the influence of seed quality in field beans is expressed far beyond emergence. Every plant growth stage monitored depicted an effect of seed quality on plant performance. These results are consistent with another report which indicated that the early stages of plant de 'elopment were slower when naturally aged seeds of broad bean (Vicia faba L.), peas (Pisum sativum L.), and barley (Hordeum vulgare L.) were sown (Abdalla and Roberts, 1969). However, this study also noted that the differences between plants from young and old seeds were less evident at later stages of plant development in contrast to the results reported here. Other studies have reported that yield per plant decreased with declining seedling emergence for snap beans naturally differing in seed quality (Toole et al., 1957) or aged in open storage in Brazil (Viera, 1966). But these effects could be attributed to decreased stands as a consequence of poor emergence. In our study, we corrected for stand differentials by increasing the number of seeds planted with decreased seed quality (Table 2) and still noted differences in vegetative and reproductive development attributed to seed quality. Thus, the beneficial effects of using high quality seeds in field bean production may be not only to achieve uniform stands but also to maximize yield potential of individual plants in the field.

While these findings clearly indicate the importance of seed quality for enhancing yield in field beans, they fail to ascribe a physiological mechanism associated with this effect. Rennie and Kemp (1984) emphasized that early nodule development is crucial for providing nitrogen for vigorous vegetative growth and support of higher seed yield in bean. In addition, Smith and Ellis (1980) investigated the influence of seed quality on soybean root nodulation. They found that high quality seeds of three soybean cultivars which were artificially aged (40°C and 100% relative humidity) for 1, 2, 3, and 4 days resulted in delayed seedling emergence and decreased top dry weight, parameters noted here (Tables 2, 3, and 4). They further found that these artifically induced changes in seed quality culminated in reduced number of tap root nodules per plant and nodule dry weight per plant. The authors emphasized that the initial, rapid growth of high

quality seeds resulted in early nodulation of the tap root. The number of nodules or nodule dry weight per plant during later growth of the root system were less influenced by changes in seed quality. To determine whether this association also existed in field beans, measurements were made of the nodule numbers, nodule dry weight, dinitrogen fixing capacity of roots from seeds differing in seed quality.

Nodulation was low at the V3 growth stage, increased during the R6 growth stage, and decreased during the R8 growth stage (Table 6). It also was noted that nodule formation was four times lower in 1984 compared to 1985. This effect was primarily attributed to the lack of indigenous rhizobia capable of infecting Pompadour Checa which was planted in Ohio for the first time. By 1985, the rhizobia had been successfully established which culminated in more effective nodulation. Maximum nodule dry weight occurred between the R6 and R8 growth stages. These findings confirm other reports which indicate maximum nodulation at the R6 growth stage (Graham, 1979; Graham and Rosas, 1977; 1979; Rennie et al., 1985). However, Graham and Rosas (1977) reported average values over 300 mg dry weight nodule tissue per plant in field bean bush-type cultivars occurring between flowering and the start of pod fill. So, these values are considered low for field bean. A possible explanation for the low nodule dry weight found in this study is that the cultivar, Pompadour Checa, is considered among the poorer nodulating field bean cultivars under tropical conditions whereas those cultivars examined by Graham and Rosas (1977) are among the higher nodulating cultivars. The decrease in nodulation observed during the R8 growth stage probably is a consequence of nodule senescence concurrent with an overall decrease in root growth (Table 4). The process of pod filling results in a reallocation of photosynthate and usually signals nodule senescence (Waters et al., 1980).

Nodulation was substantially enhanced in plants produced from high quality seeds relative to plants from low quality seeds regardless of the aging regime (Table 6). Low quality seeds generally possessed the least number of nodules and lowest nodule dry weight, medium quality seeds were intermediate, and high quality seeds the highest values for these parameters. However, these differences were less evident in 1984 when no significant effect of seed quality on nodule number and weight was observed at the R8 growth stage.

While these values indicate the physical evidence for dinitrogen fixation and its association with seed quality, they do not provide direct support for the rate of dinitrogen fixation occurring in the root nodule. In fact, Ruschel et al. (1982) concluded that nodule weight in Phaseolus spp. was a poor indicator of dinitrogen fixation. To determine whether this was the case, a study of dinitrogen fixation using the acetylene reduction assay was conducted. Greatest dinitrogen fixation activity was generally detected at the R6 growth stage (Table 7). Plants from high quality naturally aged seeds fixed greater quantities of nitrogen than plants from low quality seeds at the R6 and R8 growth stages in 1984. This increase in dinitrogen fixation in 1984 for high quality seeds relative to low quality seeds was observed through all growth stages in 1985. Plants from medium quality naturally aged seeds were similar in dinitrogen fixation activity to plants from low quality seeds at the V3 and R8 growth stages in 1984. However, they fixed more nitrogen at the R6 growth stage in 1984. No significant differences were observed among these two seed quality levels at all sampling times in 1985.

For seeds exposed to artificial aging, dinitrogen fixation was greater for plants from one day artificially aged seeds than plants from three days artificially aged seeds only at the R6 growth stage in 1984 (Table 7). However, data for 1985 further confirmed lower dinitrogen fixation in plants produced from three days artificially aged seeds compared to plants produced from one day artificially aged seeds at the V3 and R6 growth stages, respectively. The greater dinitrogen fixation levels for plants produced from one day artificially aged seeds was most pronounced at the R6 growth stage in 1984 and at the V3 growth stage in 1985.

Plants from high quality naturally aged seeds generally fixed greater amounts of nitrogen than plants from low quality seeds at the R6 and R8 growth stages in 1984 and at all growth stages in 1985, paralleling those results observed for nodule numbers and nodule dry weight (Table 6). Similar trends were noted for the artificially aged seeds. These data also clearly indicate an association between dinitrogen fixation activity and nodule number and nodule dry weight which is not in accord with the report by Ruschel et al. (1982).

These results document that seed quality influences dinitrogen fixation in field beans. However, the mechanisms by which nodulation and/or dinitrogen fixation are reduced in low

quality naturally or artificially aged seeds is not known. We postulate that at least three potential mechanisms may be involved.

First, decreased membrane integrity as a result of reduction in seed quality may enhance solute leakage from the seeds (Larson, 1968; Simon, 1978). Typically, these exudates contain storage polymers such as proteins, lipids, and starches (McDonald, 1976) which, although having little influence on total seedling emergence, strongly encourage microbial proliferation. Rhizobia, for example, are known to be more active in the vicinity of the rhizosphere (Broughton, 1978), presumably a consequence of increased availability of organic molecules exuded from the root. Similarly, the proliferation of other microflora is encouraged due to increased seed leakage. Species of field fungi such as Alternaria, Fusarium, Cephalosporium, Helminthosporium, and Pythium increase under these conditions (Chonkar and Subba-Rao, 1966; Gupta, 1973). These fungi not only compete for organic sources, but also may colonize or rot seedlings and reduce rhizobial populations surrounding the root (Chowdhury, 1976; Schlub and Lockwood, 1981; Schlub et al., 1981). Thus, the reduced nodulation of low quality field bean seeds observed in this study may be a consequence of increased seed leakage which encourages the development of antagonistic microorganisms thereby reducing the number and/or ability of rhizobia to infect the host root.

Second, farmers have long recognized that some seed lots produce stands which are delayed in emergence and exhibit poor uniformity even though germination test results indicate the seed lot to be of acceptable quality. This delayed emergence leads to shorter plants which are at a competitive disadvantage because of less exposure to direct sunlight and less consequent photosynthesis compared to taller, more rapidly growing plants, parameters noted in this study. Thus, low quality seeds produce plants which synthesize less photosynthate available for nodule growth and subsequent nitrogen fixation. According to Rawsthorne et al. (1980), high levels of photosynthate are translocated to the nodules and catabolized to supply ATP and the reducing power for dinitrogen fixation which then generates carbon skeletons for amino acid and ureide synthesis (Streeter, 1985) and supports nodule subsistence. Lack of root growth may also have an additional effect on nodule formation. Since rhizobial infection depends on root hair availability

and total root mass, seedlings produced from low quality seeds simply may not provide sufficient sites for rhizobial invasion; thus decreasing the infection rate and subsequent dinitrogen fixation.

Third, lectin is a plant protein produced in both roots and seeds (Stacey et al., 1980) which is capable of binding Rhizobium to the host plant root. Studies have indicated that lectins may be involved in legume-rhizobium associations by playing important physiological roles such as 1) acting as plant antibodies to neutralize soil bacteria and fungi, 2) protecting legumes against fungal invasion by inhibiting the activity of fungal enzymes, 3) playing a key role in the differentiation and development of cells, and 4) acting as legume-rhizobial specifins (Liener, 1976). While the last suggested role is important in determining legume-rhizobial specificity, the three preceding roles have important implications in the success of infection by nodulating bacteria for seedlings produced from seeds of different quality levels. Lectins produced from low quality field bean seeds may be unique from the isomeric tetramers composed of varying L- and R-subunits reported for Phaseolus vulgaris (Miller et al., 1975). These conformational alterations could modify endemic microorganism levels through direct soil effects or by modifying cell differentiation and development so that the root is no longer recognized by the bacteria.

This study has shown that differences in seed quality whether induced by natural or artificial aging have major effects on field bean emergence, top growth, and root growth; these effects persisted to yield. Low seed quality also reduced nodule number, weight, and nitrogen fixation suggesting one possible mechanism for the resultant loss in vegetative and reproductive matter. Further studies are needed to determine the mechanism of this association between seed quality and dinitrogen fixation.

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Table 1. Naturally and artificially aged field bean seed quality evaluations determined by the standard germination (SG) and seedling vigor classification (SVC) tests for 1984 and 1985.

	1984		1985	
Seed Quality	SG	SVC	SG	SVC
	-------%-------		-------%-------	
Naturally Aged				
High	89	78	93	86
Medium	81	69	81	73
Low	72	61	73	67
LSD (P=0.05)	4	7	4	4
Artificially Aged				
1 Day	81	72	84	77
2 Days	58	46	65	54
3 Days	44	33	52	38
LSD (P=0.05)	7	7	13	12

Table 2. The effect of seed quality on field emergence as determined by percent emergence, number of normal seedlings, number of abnormal seedlings for naturally and artificially aged field bean seeds in 1984.

Seed Quality	Seeds Planted	Seedling Emergence	Emergence	Normal Seedlings	Abnormal Seedlings
	--#--	---#---	----%----	----%----	----%----
Naturally Aged					
High	240	222	93	91	2
Medium	260	224	86	80	6
Low	360	244	81	75	6
LSD (P=0.05)		3	4	4	1
Artificially Aged					
1 Day	260	218	84	77	7
2 Days	280	215	77	66	11
3 Days	600	354	59	48	11
LSD (P=0.05)		6	4	6	4

Table 3. The effect of seed quality on field emergence as determined by percent emergence, number of normal seedlings, number of normal seedlings for naturally and artificially aged field bean seeds in 1985.

Seed Quality	Seeds Planted	Seedling Emergence	Emergence	Normal Seedlings	Abnormal Seedlings
	--#--	---#---	----%----	----%----	----%----
Naturally Aged					
High	240	210	88	85	3
Medium	260	212	82	78	5
Low	360	241	69	61	8
LSD (P=0.05)		4	5	5	2
Artificially Aged					
1 Day	260	211	81	76	5
2 Days	280	182	65	56	9
3 Days	600	312	52	36	16
LSD (P=0.05)		3	12	12	3

Table 4. Seed quality effects on plant top and root dry weight of field bean seeds at the V3, R6, and R8 growth stages in 1984 and 1985.

	1984			1985		
Seed Quality	V3	R6	R8	V3	R6	R8
	----------Top dry weight (g plant^{-1})----------					
Naturally Aged						
High	10.3	19.2	36.4	14.2	31.4	47.0
Medium	8.5	12.8	24.2	23.0	29.8	41.8
Low	8.2	12.9	20.3	12.2	25.2	33.1
LSD (P=0.05)	0.5	3.5	3.9	1.6	4.9	11.4
Artificially Aged						
1 Day	10.2	17.3	36.6	13.3	31.4	34.3
2 Days	9.4	15.4	34.0	11.9	28.9	37.4
3 Days	7.4	13.6	25.4	8.4	20.6	25.8
LSD (P=0.05)	0.9	2.5	7.2	4.4	8.6	8.4
	----------Root dry weight (g plant^{-1})----------					
Naturally Aged						
High	1.7	2.7	2.6	2.6	3.3	3.8
Medium	1.6	2.1	2.4	2.3	2.5	3.7
Low	1.5	2.2	2.2	2.0	2.6	3.2
LSD (P=0.05)	NS	0.2	0.3	0.3	0.4	0.4

Artificially Aged						
1 Day	1.5	2.3	2.8	2.2	3.0	3.5
2 Days	1.4	2.1	2.4	2.0	3.0	3.5
3 Days	1.4	2.0	2.4	1.5	2.1	2.6
LSD (P=0.05)	NS	0.3	0.3	0.5	0.8	0.9

Table 5. Effect of seed quality on plant pod number, pod dry weight, weight/100 seeds, and yield of field beans for 1984 and 1985.

	1984				1985			
Seed Quality	Pod No.	Pod Dry Wt.	Wt/100 Seeds	Yield	Pod No.	Pod Dry Wt.	Wt/100 Seeds	Yield
	# plant^{-1}	g plant^{-1}	--g--	kg ha^{-1}	# plant^{-1}	g plant^{-1}	--g--	kg ha^{-1}
Naturally Aged								
High	11	15.8	37.6	1262	17	27.8	39.8	1880
Medium	6	9.4	38.1	841	13	21.5	38.7	1447
Low	4	7.2	36.5	787	12	20.7	38.0	1424
LSD (P=0.05)	1	2.2	NS	376	2	3.9	NS	353
Artificially Aged								
1 Day	10	15.3	38.5	1271	15	24.7	39.1	1633
2 Days	10	13.9	37.2	1131	13	18.1	38.4	1617
3 Days	6	10.2	36.4	858	7	12.0	37.8	1144
LSD (P=0.05)	2	2.7	NS	336	3	4.3	NS	419

Table 6. Effect of seed quality on root nodule number and nodule dry weight of field beans at the V3, R6, and R8 growth stages in 1984 and 1985.

	1984			1985		
Seed Quality	V3	R6	R8	V3	R6	R8
	---------nodule number plant^{-1}----------					
Naturally Aged						
High	9	9	5	42	64	80
Medium	9	26	5	36	89	48
Low	2	6	4	37	72	39
LSD (P=0.05)	3	3	NS	NS	NS	15
Artificially Aged						
1 Day	2	14	4	44	103	54
2 Days	5	4	4	34	80	41
3 Days	2	7	4	26	58	39
LSD (P=0.05)	2	4	NS	16	40	14
	-------nodule weight (mg plant^{-1})--------					
Naturally Aged						
High	2.7	11.4	5.8	19.2	56.9	114.5
Medium	4.4	32.5	4.0	22.6	72.4	63.6
Low	1.3	6.5	5.4	9.3	46.8	42.1
LSD (P=0.05)	2.4	6.5	NS	7.3	NS	23.2

Artificially Aged						
1 Day	0.7	14.6	6.1	16.6	73.4	54.0
2 Days	2.0	2.6	4.5	6.8	53.3	42.7
3 Days	0.6	10.1	4.5	5.2	49.5	38.8
LSD (P=0.05)	0.7	5.5	NS	7.3	21.4	13.7

Table 7. Seed quality effect on nitrogen fixation activity as determined by acetylene reduction of field beans at the V3, R6, and R8 growth stages in 1984 and 1985.

	1984			1985		
Seed Quality	V3	R6	R8	V3	R6	R8
	-----umol C_2H_4 nodule g f.w.$^{-1}$ h^{-1}-----					
Naturally Aged						
High	5.6	13.1	15.3	11.0	12.3	9.7
Medium	5.1	11.0	8.4	9.2	10.6	5.8
Low	3.3	6.7	7.2	8.2	9.2	5.8
LSD (P=0.05)	NS	2.0	4.1	2.7	1.9	3.2
Artificially Aged						
1 Day	4.3	11.7	9.5	13.0	10.4	6.5
2 Days	4.1	6.6	8.7	7.4	9.4	5.4
3 Days	2.9	6.0	8.9	5.5	6.8	4.2
LSD (P=0.05)	NS	2.3	NS	5.9	3.2	NS