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Influence of Zeolite Application on Germination and Seed Quality of Soybean Grown on Allophanic Soil

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Abstract: The objective of the present study was to evaluate the effect of zeolite application levels and allophanic soil on seed germination and vigor of soybean cultivars grown under natural green house environment. Seeds of two soybean cultivars (Enrei, [MG] 1V) and Harosoy [MG] 11) were produced in the greenhouse from three zeolite application levels (0, 1 and 2%) and three soil (two allophanic and one paddy) at the Faculty of Agriculture, Ehime University, Japan during summer, 2007. Brown (mature) pods were harvested, threshed and all shriveled and abnormal seeds were removed before determining germination and other vigor tests. Zeolite applied at the rate of 1 and 2% improved germination and vigor of the seed increased linearly ($R^2 = 0.72$) as compared to control treated plots (0%). Seeds obtained from two allophanic soils (KyP and KnP) gave similar trend of increase in germination and vigor as compared to a Paddy Soil. Among the vigor tests, accelerated aging, electrical conductivity and seedling dry weight provided the best estimate of seed vigor for the two soybean cultivars, both for ranking seed lots quality and predicting field emergence. Seeds of Harosoy were more sensitive to high temperature stress than seeds of Enrei and accelerated aging test was more responsive in categorizing seed lots as compared to standard germination. Among cultivars, Enrei gave maximum germination and showed more vigor than Harosoy. Present findings support the results of experiment by demonstrating that zeolite application to soybean crop improves germination and vigor of soybean seeds. KyP and KnP allophanic soils showed its potential for fertility and availability of nutrients for plant growth and development as compared to paddy soil.

Key words: Vigor, germination, zeolite application, allophanic soil, soybean cultivars

INTRODUCTION

Plant nutrients are essential for producing sufficient and healthy food for the world's expanding population. Proper nutrition is essential for satisfactory crop growth and production and is therefore a vital component of any system of sustainable agriculture. Moreover, agricultural intensification requires increased flows of plant nutrients to crops and higher uptake of those nutrients by crops. Balanced use of plant nutrients corrects nutrient deficiency, improves soil fertility, increases nutrient and water use efficiency, enhances crop yield and farmers income. The use of healthy seed is essential for good yields and quality. High quality seeds improve potential response to fertilizer, water and other inputs and thereby increases crop production significantly (McMaster and Smika, 1988; McMaster, 1997). At present, a considerable gap exists between the yields obtained from the use of good quality seeds and poor-quality seeds in farmers' fields. Lack of use of good quality seeds by farmers is one of the biggest production constraints. To reap the benefits of balanced use of plant

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nutrients, it is important to have good quality seed, adequate moisture and better agronomic practices with greater emphasis on timeliness and precision in farm operations. High quality planting seed is a key component of all grain cropping systems and is needed to ensure adequate plant populations, with reasonable seeding rates, in a range of field conditions. Seed quality at planting represents the integrated effects of the environment during seed production and the conditions the seeds were exposed to during harvest, conditioning and storage.

Allophane is a clay-size alumino-silicate mineral with short range order, which occur widely in Spodosols and Andisols (Wada, 1989; Parfitt, 1990). Allophane has a large propensity of sorbing organic substances. Similarly, allophanic clay stabilizes the microbial biomass and its metabolites (Saggar *et al.*, 1996). Andisols are often very fertile and they commonly have physical properties that make them very suitable for agricultural use.

Zeolites are microporous, crystalline, hydrated aluminosilicates of alkali and alkaline materials that have a high internal surface area (Rollman and Valyocik, 1981). The unique cation exchange, adsorption, hydration-dehydration and catalytic properties of zeolite have prompted slow-release of fertilizers and other materials (Mumpton and Fishman, 1977; Pond and Mumpton, 1984). The poor utilization of fertilizers by crops is largely caused by losses of N through denitrification, ammonia volatilization, runoff and leaching (Craswell and Vlex, 1979). Natural compound such as zeolite minerals have been reported as ameliorants for coarse soils to modify soil Cation Exchange Capacity (CEC) to decrease N leaching and to increase fertilizer recovery (Huang and Petrovic, 1994). This property allows the grower to use this less costly form of Nitrogen in larger and less frequent applications during the growing season. The retention and timely release of needed nutrients by zeolite improves overall crop yield. It provides a lasting reservoir of nutrients allowing the user to reduce added fertilization while achieving better plant and vegetable performance. Zeolites hold nitrogen in the soil, moderate nitrification of ammonia and make it available to plants as needed. Seed quality at harvest is primarily a function of disease, temperature and moisture conditions. We are not aware of reports describing effects of the plant's environment (soil conditions, nutrient availability) on seed quality, so variation in seed quality in the absence of disease can be related to soil condition and nutrients availability during seed development and maturation. Consequently, our objective was to evaluate the relationship between zeolite application levels, KyP and KnP allophanic soil and cultivar on soybean seed germination and seed quality from plants grown under natural greenhouse environment.

MATERIALS AND METHODS

To assess the effect of zeolite application on germination and vigor of soybean seed grown on Japanese two allophanic soils (KyP and KnP) and a paddy soil (Table 1), a pot experiment was carried out at the Faculty of Agriculture, Ehime University, Matsuyama Japan, during summer, 2007. The experiment was carried out in completely randomized design in greenhouse under natural environment. Ceramic cylinders pots (h = 20 cm, Ø = 10 cm) were filled with 2 kg of air-dried, sieved soil samples. A 3 factor (3×3×2) factorial experiment of three soils and zeolite amendment levels on two soybean varieties were used. A Paddy soil was collected from Ehime University Agriculture Research Farm Hojo and was used as control. The pumice grains for KyP having low Si/Al ratio were collected from

Table 1: Some physical and chemical properties of the different soil used in the experiment

| Soil type | pH (H ₂ O) | pH (KCl) | CEC | Na | K | Ca | Mg |
|-----------------------|--------------------------------------|----------|------|-----|-----|-----|-----|
| | ----- (cmol kg ⁻¹) ----- | | | | | | |
| Paddy soil | 6.0 | 5.1 | 13.8 | 0.6 | 0.5 | 7.0 | 1.4 |
| Allophanic soil (KyP) | 5.7 | 5.0 | 17.2 | 1.8 | 0.3 | 0.3 | 0.2 |
| Allophanic soil (KnP) | 6.2 | 4.8 | 30.1 | 1.3 | 1.9 | 7.2 | 2.6 |

Kurayoshi, Tottori prefecture, near Mt. Daisen. While for KnP having high Si/Al ratio were collected from Kakino, Kumamoto Prefecture, near Mt. Aso. Three zeolite levels of 0, 20 and 40 g were applied one day before planting of the crop. The zeolite was obtained from Maeda Kensetsu, Co. Ltd. CEC is 260 cm kg^{-1} and main zeolite species is Na.P1. Determinate cultivar (Enrei, [MG] 1V) and indeterminate cultivar Harosoy [MG] 11) were planted at 30 mm depth in the above soils. Maximum seed were planted to obtain the required plant population density that should be quite enough to study the required parameters. A basal dose of 5 g N and 10 g each of P_2O_5 and KCl were applied one day before starting the experiment. The pumice grains for KyP and KnP and paddy soils were ground and then sieved through a 2 mm sieve before application. The 18 combinations were replicated 3 times so there were $18 \times 3 = 54$ experimental units. The 54 pots were arranged within 18×3 arrays of rows with 20 cm distance between pots. Normal cultural practices for raising a successful crop were applied uniformly to all the experimental units. The plots were hand weeded at different vegetative and reproductive stages. Irrigation was applied at weekly intervals or as when needed. Seeds were evaluated for seed quality, immediately after harvest by various vigor tests i.e., by Standard Germination Test (SGT), Accelerated Aging (AA), Electrical Conductivity (EC), Seedling Axis Dry Weight (SADW) and expected field emergence test under stress conditions were carried out. For SGT, two 50 seeds samples from each field plot were planted in rolled paper towels and placed in a germinator at 25°C for 7 days (Association of Official Seed Analysts, 2002). The (AA) test was conducted as described by Hampton and TeKrony (1995). Two lots of 50 seeds from each treatment replicated four times were kept for 72 h at 42°C . Germination was determined after aging by planting 50 seed sample in rolled moist towels at 25°C as described by ISTA (1995) and final count was made at 7 days. For the conductivity test two replicates of 50 weighed seeds incubated for 24 h in 250 mL flask containing 200 mL of deionized water at $20 \pm 2^\circ\text{C}$. The electrical conductivity was measured with conductivity meter and expressed as $\mu\text{S/cm/g}$ (International Seed Testing Association, 1995). Seedling Axis Dry Weight (SADW) was determined by obtaining normal seedling on day 5th after germination in the dark. Cotyledons were detached before drying and were placed in paper bag, dried at 70°C for 48 h and weighed (Association of Official Seed Analysts, 2002). Field emergence test was carried out under stress environmental conditions with four replications of 100 seeds of each variety were planted by hand in 4-meter rows at 3.5 cm depth. The pots were irrigated through spring irrigation system to maintain relatively uniform soil moisture conditions. Daily counts were made as soon as the seedlings begin to emerge and continued until emergence was completed. Seedlings were considered as emerged, when the cotyledons were free of the soil surface. Soil temperature and average maximum and minimum air temperature at soil surface and one meter above the soil were recorded daily. An Emergence Index (EI) was calculated using the following formula:

$$EI = \{T_i N_i / S\}$$

where, T_i is the number of days after sowing, N_i is the number of seeds germinated on day i and S is the total number of seeds planted (Scott *et al.*, 1984). Final emergence was calculated as a percentage of the number of seeds planted. All tests were performed by using a completely randomized design. Analysis of variance was performed and significant differences among treatments were determined by the LSD test at 0.05 level of probability.

RESULTS AND DISCUSSION

Germination (%)

Zeolite application and allophanic soil significantly affected the germination of soybean varieties (Table 2). Seed harvested from 20 and 40 g zeolite gave maximum germination (77.79%) as compared to control treated plots. Maximum germination of seeds from zeolite treated plots may be due to more

Table 2: Germination test of soybean varieties as affected by zeolite application and soil type

| Soil type | Cultivars | Zeolite doses (g) | | | Mean* |
|--------------|-----------|-------------------|--------|--------|--------|
| | | 0 | 20 | 40 | |
| S×V×Z | | | | | |
| Paddy soil | Harosoy | 65.30 | 71.00 | 73.40 | 69.09a |
| | Enrei | 69.50 | 72.10 | 74.60 | 72.06b |
| KyP | Harosoy | 71.40 | 74.90 | 76.80 | 74.36c |
| | Enrei | 73.90 | 76.50 | 78.30 | 76.23d |
| KnP | Harosoy | 72.90 | 76.90 | 81.30 | 77.03e |
| | Enrei | 75.50 | 80.50 | 82.40 | 79.46f |
| S×Z | | | | | |
| Paddy soil | | 67.04 | 71.55 | 74.00 | 70.86a |
| KyP | | 72.65 | 75.05 | 77.55 | 75.09b |
| KnP | | 74.02 | 78.07 | 81.85 | 76.27b |
| Z×V | | | | | |
| | Harosoy | 69.86 | 74.27 | 77.16 | 73.76a |
| | Enrei | 72.96 | 76.36 | 78.43 | 75.91b |
| Mean | | 71.41a | 75.31b | 77.79c | |

*Means of the same category followed by different letters are significantly different at 5% level of probability using LSD test

protein content in the seed and larger size of the embryo, which lead toward more viability and vigor of the seed (Gibson and Mullen, 1996a). TeKrony *et al.* (1984) stated that late maturing varieties produced seed of high quality and seed vigor increased as harvest maturity was delayed. KyP and KnP allophanic soil significantly affected the germination percentage of soybean varieties. Seed harvested from KyP and KnP gave higher germination (76.27%) than paddy soil (Table 2). Higher germination rate from KyP and KnP allophanic soils may be attributed to less competition among plants for plant food material, more leaf area development leading to greater total photosynthesis and more assimilates, which indirectly resulted in large and vigorous seed. The lower germination of seeds from control plots of zeolite and paddy soil could be due to lower availability of nutrients during seed filling period for proper growth and development of seed. Significant difference was observed between germination of the two varieties. Enrei gave maximum germination than Harosoy. The interaction between different soils × varieties reveal that seeds of both varieties gave minimum germination on paddy soil as compared to KyP and KnP allophanic soils. A linear increase in germination percentage of both cultivars was observed, when seeds were obtained from KyP and KnP allophanic soils.

Accelerated Aging Test

This test incorporates many of the important traits desired in a vigor test. Initially proposed as a method to evaluate seed storability, the accelerated aging test subject unimbibed seeds to conditions of high temperature (41°C) and relative humidity (100%) for short periods (3 to 4 days). The seeds are then removed from the stress conditions and placed under optimum germination conditions. Zeolites application and allophanic soil significantly affected the AA test (Table 3). Zeolites treated plots gave higher germination (40.41%) than control treated plots (33.99%). The higher germination percentage of seeds from zeolites treated plots may be due the greater variation in resistance to field weathering and tolerance to high temperature during germination which exists among varieties with different seed coat structures, seed sizes, seed weights and seed coat color. TeKrony *et al.* (1984) and Miles *et al.* (1988) stated that late maturing varieties produce seed of high quality and vigor. KyP and KnP allophanic soils gave higher germination percentage than paddy soil. Higher germination percentage from seeds of KyP and KnP may be attributed to vigor and viability, which prove its resistance to stress conditions as compared to paddy soil. The lower germination of seeds from paddy soil could be due to more favorable conditions for seed infection during seed filling duration. Zeolites treated plots improved seed germination and vigor and reduced the levels of seed infection by *Phomopsis* sp. Enrei gave higher germination percentage than Harosoy.

Table 3: Accelerated aging test (percent germination) of soybean varieties as affected by zeolite application and soil type

| Soil type | Cultivars | Zeolite doses (g) | | | Mean* |
|--------------|-----------|-------------------|--------|--------|----------|
| | | 0 | 20 | 40 | |
| S×V×Z | | | | | |
| Paddy soil | Harosoy | 31.25 | 35.88 | 37.88 | 35.01a |
| | Enrei | 33.23 | 37.34 | 40.34 | 36.97ab |
| KyP | Harosoy | 33.88 | 36.25 | 38.00 | 36.04ab |
| | Enrei | 34.38 | 39.75 | 42.25 | 38.79bc |
| KnP | Harosoy | 34.13 | 37.13 | 40.38 | 37.38abc |
| | Enrei | 34.13 | 37.13 | 40.88 | 37.38abc |
| S×Z | | | | | |
| Paddy soil | | 32.24 | 36.61 | 39.11 | 35.98a |
| KyP | | 34.13 | 38.23 | 40.12 | 37.49b |
| KnP | | 35.63 | 38.67 | 42.05 | 38.78b |
| Z×V | | | | | |
| | Harosoy | 33.08 | 36.42 | 38.92 | 36.14a |
| | Enrei | 34.91 | 39.01 | 41.90 | 38.61b |
| Mean | | 33.99a | 37.76b | 40.41c | |

*Means of the same category followed by different letters are significantly different at 5% level of probability using LSD test

Table 4: Electrical conductivity ($\mu\text{S}/\text{cm}/\text{g}$) of soybean varieties as affected by zeolite application and soil type

| Soil type | Cultivars | Zeolite doses (g) | | | Mean* |
|--------------|-----------|-------------------|--------|--------|--------|
| | | 0 | 20 | 40 | |
| S×V×Z | | | | | |
| Paddy soil | Harosoy | 36.70 | 34.50 | 32.40 | 34.53e |
| | Enrei | 25.20 | 23.80 | 21.20 | 23.04d |
| KyP | Harosoy | 15.40 | 13.80 | 12.90 | 14.03c |
| | Enrei | 14.10 | 12.80 | 11.50 | 12.08b |
| KnP | Harosoy | 14.30 | 12.60 | 11.50 | 12.8b |
| | Enrei | 12.40 | 11.20 | 8.70 | 10.76a |
| S×Z | | | | | |
| Paddy soil | | 32.95 | 29.15 | 26.80 | 28.66c |
| KyP | | 14.75 | 13.30 | 12.20 | 13.43b |
| KnP | | 13.35 | 11.90 | 10.10 | 11.78a |
| Z×V | | | | | |
| | Harosoy | 22.13 | 20.03 | 18.93 | 20.36b |
| | Enrei | 17.23 | 15.93 | 13.08 | 15.41a |
| Mean | | 19.68c | 17.98b | 16.05a | |

*Means of the same category followed by different letters are significantly different at 5% level of probability using LSD test

Electrical Conductivity

Seed electrical conductivity tests are usually assumed to reflect cellular membrane integrity and are determined by potential measurements of hydrated seeds or on seed steep water. These measurements show that the increases in steep water conductivity correlates well with other indicators of declines in seed vigor. The data regarding electrical conductivity of seed produced from zeolite application and KyP and KnP allophanic soil are reported in Table 4. Minimum electrical conductivity of seeds ($19.68 \mu\text{S}/\text{cm}/\text{g}$) was recorded from 20 and 40 g zeolite applications as compared to control treated plots. Paddy soil and control treated plots of zeolite produced seeds with maximum electrical conductivity ($23.6 \mu\text{S}/\text{cm}/\text{g}$). Maximum electrical conductivity of seed leachate from control treated plots of zeolite and paddy soil may be due to poor seed coat development during seed filling duration and at physiological maturity. Seeds developed in plots treated with 0 g zeolite and paddy soil were exposed to more competition for nutrients and greater diurnal temperature amplitude due to lower canopy density. These findings are in agreement with those of Bhering *et al.* (1991) and Zanakis *et al.* (1994a, b). They reported that seed quality was generally better with later sowing dates

Table 5: Seedlings dry weight (mg/50 seeds) of soybean varieties as affected by zeolite application and soil type

| Soil type | Cultivars | Zeolite doses (g) | | | Mean* |
|--------------|-----------|-------------------|--------------|--------------|---------|
| | | 0 | 20 | 40 | |
| S×V×Z | | | | | |
| Paddy soil | Harosoy | 3.00 | 3.40 | 3.80 | 3.04a |
| | Enrei | 3.20 | 3.50 | 3.90 | 3.53ab |
| KyP | Harosoy | 3.50 | 3.80 | 4.30 | 3.87b |
| | Enrei | 3.60 | 3.90 | 4.60 | 12.08bc |
| KnP | Harosoy | 3.90 | 4.00 | 4.80 | 4.23d |
| | Enrei | 4.10 | 4.60 | 4.90 | 43.53d |
| S×Z | | | | | |
| Paddy soil | | 3.10 | 3.45 | 3.85 | 3.46a |
| KyP | | 3.55 | 3.85 | 4.45 | 3.95b |
| KnP | | 4.00 | 4.03 | 4.85 | 4.29b |
| Z×V | | | | | |
| | Harosoy | 3.46 | 3.73 | 4.30 | 3.83a |
| | Enrei | 3.63 | 4.00 | 4.46 | 4.03b |
| Mean | | 3.55a | 3.86b | 4.38c | |

*Means of the same category followed by different letters are significantly different at 5% level of probability using LSD test

than earlier sowing dates because the maturation of early planted crop coincided with high rainfall and temperature. Significant differences were observed between varieties with respect to electrical conductivity and Harosoy showed more leachate than Enrei.

Seedling Dry Weight

Seedling dry weight is an important criterion for evaluating seed vigor in maximum crops. Maximum seedlings dry weight of 4.0 mg per 50 seeds was obtained from 20 and 40 g zeolite applied to KyP and KnP allophanic soil (Table 5). Maximum seedlings dry weight from 20 and 40 g zeolite application indicates that seed produced with zeolite application were larger in size and heavier in weight, which ultimately resulted in more seedlings dry weight than control treated plots (Caulfield and Bunce, 1991). KnP and KyP allophanic soils gave maximum seedlings dry weight as compared to paddy soil. Maximum seedlings dry weight from KnP and KyP allophanic soils may be due to less competition among plants for the availability of plant food material from the soil and maximum leaf area per plant with greater photosynthesis, which ultimately resulted in heavier seedlings as compared to paddy soil. A steady decrease in seedlings dry weight was observed with seed produced from paddy soil treated without zeolite. Seedlings dry weight was significantly different among varieties as seeds of Enrei gave significantly more seedlings dry weight than Harosoy due to larger seed size.

Field Emergence

Field emergence is a hard vigor test, used to measure the potential health of the seed lot. Data regarding field emergence of the two soybean varieties as affected by zeolite application and KnP and KyP allophanic soils are presented in Table 6. Minimum field emergence of 57.7% was obtained from control plots of zeolite and paddy soil. A steady increase in field emergence occurred with the addition of zeolite. Forty gram zeolite application produced seeds with maximum field emergence percentage of 70.3% followed by 20 g zeolite applications. Maximum field emergence from higher dose of zeolite may be due to more protein content and heavier seed weight, which helped during germination and emergence of the seedlings. These results are in agreement with those of Steiner (1990) who stated that high vigor seed lots have significantly higher emergence rate than low vigor seed lots. KyP and KnP allophanic soils gave higher field emergence of the soybean seeds than paddy soil. Maximum emergence from KyP and KnP allophanic soils may be due to the maximum viability and vigor of seeds. Interaction between different soil × varieties indicate that seeds of both Enrei and Harosoy gave better emergence on both KyP and KnP allophanic soils. A steady decrease in field emergence was noticed in seeds obtained from plots grown on paddy soil without zeolite application.

Table 6: Field emergence (%) of soybean varieties as affected by zeolite application and soil type

| Soil type | Cultivars | Zeolite doses (g) | | | Mean* |
|--------------|-----------|-------------------|--------|--------|---------|
| | | 0 | 20 | 40 | |
| S×V×Z | | | | | |
| Paddy soil | Harosoy | 52.40 | 55.30 | 57.40 | 55.03a |
| | Enrei | 54.30 | 57.30 | 59.50 | 57.03ab |
| KyP | Harosoy | 55.70 | 58.80 | 61.70 | 58.73b |
| | Enrei | 57.30 | 62.60 | 63.60 | 61.17c |
| KnP | Harosoy | 58.70 | 61.50 | 63.50 | 61.23c |
| | Enrei | 59.40 | 62.40 | 64.30 | 62.03d |
| S×Z | | | | | |
| Paddy soil | | 53.35 | 56.03 | 58.45 | 55.94a |
| KyP | | 56.06 | 60.07 | 62.65 | 59.59b |
| KnP | | 60.04 | 61.95 | 63.09 | 61.69b |
| Z×V | | | | | |
| | Harosoy | 55.06 | 58.53 | 60.86 | 58.15a |
| | Enrei | 57.00 | 60.76 | 62.47 | 60.07b |
| Mean | | 56.03a | 59.64b | 61.66b | |

*Means of the same category followed by different letters are significantly different at 5% level of probability using LSD test

CONCLUSIONS

It is concluded from the present investigation that zeolite application at the rate of 1 and 2% improve the germination and seed quality of soybean cultivars. It is further stated that KnP and KyP allophanic soils show its fertility and high potential for crop cultivation.

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