



Full Length Article

Efficiency of Vigor Tests and Seed Elemental Concentrations to Estimate Field Emergence in Soybean (*Glycine max*)

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Abstract

The relationship between seed vigor test and seedling emergence is crucial to predict easily the seed performance under field conditions and the seed producers continuously explore the best vigor test to enhance stand establishment. This study was conducted to determine the suitable seed vigor test and mineral element in the seed to predict accurately the field performance of soybean. Fifty seed lots of 22 commercial soybean cultivars were assessed for seedling emergence under field conditions and germination, emergence, cool and cold tests, electrical conductivity (EC) and pH for 4, 8 and 24 h, accelerated ageing (AA) at 41, 43 and 45°C for 48, 72, 96, 120 and 144 h in the laboratory. Initial seed weight, oil, protein, P, K, Ca, Fe, Mn and Zn concentration in seed lots were also investigated. Their results were correlated with field emergence percentage of seed lots to detect the most suitable indicator for vigorous seed lot. The germination percentage varied from 56% to 100%, while the field emergence percentage ranged between 0% and 84%. No significant correlation was noticed between field emergence and seed weight, oil and protein contents. AA test at 41°C for 72 h gave the highest significantly positive correlation with $r=0.831^{**}$, but the highest significant and negative correlation coefficient ($r=-0.801^{**}$) with field emergence percentage were observed in EC test for 24 h. Only Mn concentration of six elements investigated was significantly correlated with field emergence ($r=-0.376^{*}$). It was concluded that the AA condition of 72 h at 41°C are recommended as an ideal seed vigor test to estimate closely field emergence performance of soybean seeds. © 2016 Friends Science Publishers

Keywords: *Glycine max* L.; Germination; Seedling emergence; Accelerated ageing; Seed quality

Introduction

Soybean (*Glycine max* L.) is an economically important protein rich oilseed crop with variety of uses, including wide use in poultry industry of Turkey. Its production is supported by the Turkish government to avoid import of soybean for local uses (Kolsarıcı *et al.*, 2015; TUIK, 2016). It has been extensively cultivated as a main and second crop after winter wheat or barley in the Mediterranean region of Turkey (Çopur *et al.*, 2009). The difference between planting dates lead to exposing the seeds to adverse environmental conditions, namely, water lodging, low temperature and crust formation in the main crop, or high soil temperature, drought and poor seedbed preparation in double-crop planting (Khaliliaqdam *et al.*, 2013). For these reasons, the vigorous seed production is obligatory for regular germination, seedling emergence and a good stand establishment.

Seed suppliers consider the standard germination test as an evidence of seed quality; that is conducted under optimal conditions in terms of temperature and moisture for germination. It doesn't completely estimate the seedling emergence performance because field conditions are rarely

optimum for germination, which adversely affect the emergence and subsequent seedling growth (Hampton and TeKrony, 1995; Kolasinska *et al.*, 2000; Khan *et al.*, 2010). Tekrony and Egli (1977) reported the standard germination exhibited a significant correlation with field emergence of soybean only if favorable field conditions have been available while Makkawi *et al.* (1999) demonstrated the standard germination was the only test to be useful for predicting field emergence of lentil. Usha and Dadlani (2015) found that germination test was the worst test predicting field performance of soybean among the investigated vigor tests.

Several seed vigor tests have been developed for better prediction of field performance of seed lots (Hampton and Coolbear, 1990; ISTA, 2002; Milošević *et al.*, 2010; Arif *et al.*, 2014). Colete *et al.* (2004) and Vieira *et al.* (2004) reported that electrical conductivity (EC) may be efficient to determine seed vigor in soybean while Salinas *et al.* (2010) the minimum 19 hours must require for the stable EC results in soybean. Marcos-Filho *et al.* (2001) found the accelerated ageing and controlled deterioration tests could be used to select vigorous seed lots in soybean. Vieira *et al.* (2010) suggests that the cold test conducted between papers at

10°C should be used for an alternative procedure to rank soybean seed lots for seed vigor. On the other hand, Gamiely *et al.* (1990) have evaluated seed element concentrations for the viability and vigor of the seed in onion and Kaymak *et al.* (2012) found a significant correlation between field emergence and some element concentrations in bean seed. Although there are several kinds of researches on the vigor tests, different tests have been advised for estimating seedling emergence in soybean by Kulik and Yaklich (1982), Colete *et al.* (2004), Torres *et al.* (2004), Vieira *et al.* (2010) and Santorum *et al.* (2013) in consequence of using limited seed lots or several lots of a variety. Consequently, the present study was carried out to assess the efficiency of various seed vigor tests and seed mineral element concentrations to rank seed lots, and to identify the most suitable test which could forecast the field performance using fifty seed lots of soybean.

Materials and Methods

Experimental Details and Treatments

Experimental material: This study was performed at the experimental fields and Seed Science Laboratory at the Field Crops Department, Faculty of Agriculture, Eskişehir Osmangazi University, Turkey. Fifty seed lots from 22 soybean varieties collected from seed suppliers were used in the study. The seeds lots were stored at 4°C until the start of the experiment.

Treatments: Standard germination was performed with four replications of 50 seeds from each seed lot. The seeds were placed in three filter papers moistened with 8 mL of distilled water. To avoid moisture loss, each rolled paper was put into a sealed plastic bag. The seeds were germinated at 25±1°C for 10 days under the dark conditions. The seeds were considered germinated when the emerging radicle was at least 2 mm long. The germination counts were recorded every 24 h for 10 days and the mean germination time (MGT) was calculated to determine the germination speed as described by the rules of ISTA (2003).

The cold test was applied to four replications of 50 seeds placed in three layer filter papers and moistened with distilled water as mentioned in germination test. The rolls were then wrapped and placed in an upright position into plastic packages and kept in a cold chamber at 10°C for seven days. Then the packages were transferred to incubators at 25°C (Loeffler *et al.*, 1985). Germination counts were done daily to determine the speed of germination under cold stress.

The cool test was performed continuously at 18°C using filter papers (Hampton and TeKrony, 1995). The seeds were incubated for 10 days under dark conditions and the germinated seeds were counted daily.

For laboratory emergence percentage, 4×50 seeds from each seed lot were sown at 3 cm depths in a seedling tray (30 cm × 20 cm × 7 cm) using peat. The trays were

placed in a growth chamber at 25±1°C and 70% relative humidity for 10 days. The seedlings emerged at the surface were counted 10 days after sowing. The field emergence test was also conducted with four replications of 50 seeds for each seed lot and the seeds were sown in rows spaced at 45 cm at a depth of 2.5–3.5 cm in a 3 m long plot at the experimental field of the Field Crops Department. The emerged seedlings with cotyledon leaves were counted 30 days after sowing and represented the field emergence percentage.

The electrical conductivity (EC) and pH tests were performed by using four replications of 50 weighed seeds from each lot. The weighed seeds were immersed in 200 mL of deionized water at 20±1°C for 24 h (ISTA, 2003) to detect the electrolyte leakage from seeds. After 4, 8 and 24 h incubation, the EC and pH of the soaked water were measured using a conductivity meter (WTW Cond 7310i, Germany) and pH meter (WTW pH 7110). The EC results were expressed in $\mu\text{S cm}^{-1} \text{ g}^{-1}$ to account for the variability in the seed weight among the seed lots.

Two hundred seeds were sampled for the accelerated ageing (AA) test from the each seed lot. The AA test was performed with different temperature and time combination of 41, 43 and 45°C and approximately 100% air relative humidity for 24, 48, 72, 96, 120 and 144 h in plastic boxes (11 × 11 × 4 cm) with 40 mL of distilled water as described by Hampton and Tekrony (1995). The seeds were placed on a 10 × 10 × 3 cm wire mesh tray and placed in a box after they were uniformly distributed. Each box was used for only one temperature and time combination in each seed lot. After ageing incubation, standard germination test using 50 seeds per replicate were applied at 25±1°C in a dark growth chamber for 10 days.

For seed element analysis, dry seeds were ground with a ball mill, then all samples were dry ashed at 550°C for 5 h in a muffle furnace. The ash was dissolved in 3.3% (v/v) HCl (Risser and Baker, 1990). The Fe, Cu, Zn and Mn concentrations in digesting samples were measured using an atomic absorption spectrometer (AAS) (Analytic Jena Type NovAA-350, Germany) (Daghan *et al.*, 2008). Potassium (K) concentrations by a flame photometer (BWB XP, England) and phosphorus (P) concentrations by UV/VIS spectrophotometer (Thermo Aquamate, England) were measured. Nitrogen was analyzed by Kjeldahl method (Kacar, 1995).

Statistical Analysis

The experimental data consisted of a completely randomized design with four replicates and 50 seeds per replicate. Data for the germination percentages were subjected to an arcsine transformation before analysis of variance was performed using the MSTAT-C program (Michigan State University). The single vigor test predicting field emergence was identified using the correlation coefficient values (r) at 5% and 1% level of significance.

Results

The results showed that there were significant differences among soybean seed lots in all laboratory tests and field emergence percentage (Table 1). Among seed properties of the investigated soybean seed lots, thousand seed weight, oil and protein content showed a considerable variation. The lowest oil content was 13.9%, while the maximum value was 25.5%. Initial seed viabilities of the lots ranged between 56% and 100%, while field emergence percentage changed between 0% and 84%. The least electrical conductivities were measured after 4 h and extended time that resulted in increase in EC values of the seed lots.

Simple correlation coefficients calculated among the vigor tests showed that most of the tests were correlated with each other except for thousand seed weight, oil and protein content (Table 2). No significant effects of weight, oil and protein content of seeds were observed on germination and emergence performance of soybean. The laboratory and field emergence possessed negative correlation coefficients with the vigor tests apart from the cool and cold test. The highest significant and positive correlation ($r=0.896^{**}$) for standard germination was detected with the cold test. Also, there was a significant positive correlation between the cold test and laboratory emergence. Field emergence was negatively correlated with EC, pH and MGTs and positively with the standard germination, laboratory emergence, cool and cold tests. Increased exposure duration of EC test caused an increase in correlation coefficient, and the highest correlation was measured for 24 h. Similarly, germination and emergence of soybean seeds were related to pH measurements, but lower correlation coefficient than the other tests was determined. Among the vigor tests, field emergence performance was better predicted with EC for 24 h and cold test than the standard germination and laboratory emergence because the correlation coefficient peaked. MGT calculated in the standard germination, cool and cold tests showed a significant negative correlation with field emergence percentage, but higher coefficients were calculated with standard germination. Among them, MGT in the cold test was better in predicting field emergence percentage with the coefficient $r=0.658^{**}$.

A series number of time and temperature combinations in AA were significantly correlated with germination and emergence percentage of soybean while lower temperature and time gave higher correlations compared to the extended time and increased heat (Table 3). Because any germination was not recorded for 144 h at 41, 43 and 45°C, the results were not mentioned. All the correlation coefficients calculated were positive. The highest coefficients were observed at 41°C for 48 h in germination ($r=0.807^{**}$) and laboratory emergence ($r=0.742^{**}$), and at 41°C for 72 h in field emergence ($r=0.831^{**}$). Increased time and temperature in AA conditions caused decreasing correlation coefficient. Also, the correlations with standard germination were much lower

than that of field emergence.

Mineral element concentrations of seed lots of soybean correlated with germination, laboratory and field emergence were shown in Table 4. A significant and negative correlation was determined between Mn concentrations in the seed and germination and field emergence, while Fe concentration showed a positive significant correlation with laboratory emergence. Only one element, Mn, was significantly correlated with field emergence. Compared to seed vigor tests, seed element concentration showed lower and insignificant correlations with germination and emergence percentages.

The relationship among the vigor tests with the highest correlation coefficients was determined with field emergence (Fig. 1). The R^2 values (0.760^{**} , 0.691^{**} and 0.711^{**}) of the cold test, AA and EC results were significant while Mn concentration in the seed was insignificant. An increase in germination percentage in the cold and AA related with increasing field emergence. Also, increased EC resulted in decreasing the field emergence.

Discussion

There was remarkable variation in minimum and maximum values in terms of seed weight, oil and protein contents among soybean seed lots, however; any significant correlations were not determined with germination and emergence. Contrarily, Gamiely *et al.* (1990) in onion and Pahlavani *et al.* (2008) in cotton observed that there was a significant positive correlation between seed weight and field emergence, while Kulik and Yaklich (1982) reported no significant correlation between seed weight and field emergence because different varieties were used as seed lots in soybean. The cool and cold test showed a positive correlation while the laboratory and field emergence negatively correlated with the other vigor tests. The cold test had the highest significant and positive correlation coefficient with standard germination. Also, a significant positive correlation between the cold test and laboratory emergence was detected. The results of this study are in agreement with the observations of Vieira *et al.* (2010) in soybean and Lovato *et al.* (2005) in maize. They observed that the cold test applied at 10°C gave a significant correlation with field emergence.

Electrical conductivity test has been used for determining seed vigor in pea and soybean because its results are closely related to seedling emergence. In our study, the results of EC test significantly correlated with field emergence of soybean. Similar findings were observed in legumes by Colete *et al.* (2004) and Vieira *et al.* (2004) in soybean, Panobianco *et al.* (2007) and Atak *et al.* (2008) in pea, Makkawi *et al.* (1999) in lentil, Silva *et al.* (2013) in common bean. They noted that the EC was clearly related to germination and emergence percentage, and lower emergence was observed in seeds with higher EC. Germination and emergence of soybean seeds were related

Table 1: Minimum, maximum and mean values of germination and seed vigor tests with field emergence for 50 seed lots of soybean

	Minimum	Maximum	Mean	SE	Probability
Oil content (%)	13.9	25.5	19.2	0.39	<0.01
Protein content (%)	22.4	38.4	32.2	0.60	<0.01
1000 seed weight (g)	125.1	222.0	164.9	3.74	<0.01
Germination percentage (%)	56.0	100.0	89.4	1.73	<0.01
MGT (d)	1.55	3.55	2.22	0.06	<0.01
Laboratory emergence (%)	0.0	99.0	80.6	2.68	<0.01
Field emergence (%)	0.0	84.0	55.7	2.94	<0.01
Cool test (%)	46.5	100	91.3	1.78	<0.01
MGT (d)	2.01	3.57	2.58	0.05	<0.01
Cold test (%)	7.5	99.5	78.3	3.62	<0.01
MGT (d)	3.92	6.00	4.71	0.06	<0.01
EC 4 h	10.3	43.8	23.5	1.02	<0.01
EC 8 h	15.5	59.8	34.7	1.69	<0.01
EC 24 h	24.4	119.9	60.1	3.33	<0.01
pH 4 h	5.95	6.55	6.19	0.02	<0.01
pH 8 h	5.86	6.36	6.07	0.02	<0.01
pH 24 h	5.62	6.22	5.93	0.02	<0.01

Table 2: Correlation coefficients between standard germination, laboratory emergence, field emergence and seed vigor tests in 50 seed lots of soybean

	Standard germination (%)	Laboratory emergence (%)	Field emergence (%)
Laboratory emergence (%)	0.693**	-	-
Field emergence (%)	0.637**	0.722**	-
MGT (d)	-0.886**	-0.698**	-0.634**
Oil content (%)	-0.260 ^{ns}	-0.111 ^{ns}	-0.239 ^{ns}
Protein content (%)	-0.127 ^{ns}	-0.108 ^{ns}	-0.046 ^{ns}
1000 seed weight (g)	-0.057 ^{ns}	-0.085 ^{ns}	-0.170 ^{ns}
Cool test (%)	0.840**	0.681**	0.654**
MGT (d)	-0.731**	-0.648**	-0.690**
Cold test (%)	0.896**	0.768**	0.737**
MGT (d)	-0.755**	-0.594**	-0.658**
EC 4 h	-0.376*	-0.492**	-0.620**
EC 8 h	-0.489**	-0.546**	-0.596**
EC 24 h	-0.620**	-0.751**	-0.801**
pH 4 h	-0.514**	-0.369*	-0.436**
pH 8 h	-0.674**	-0.537**	-0.668**
pH 24 h	-0.476**	-0.341*	-0.470**

*, ** significant at %5 and %1 levels of probability, respectively; ns: not significant

Table 3: Correlation coefficients between germination percentage after AA test conducted at different temperature and time combinations and germination, laboratory and field emergence

Temperature (°C)	Hours	Germination percentage (%)	Laboratory emergence (%)	Field emergence (%)
41	48	0.807**	0.742**	0.748**
	72	0.719**	0.722**	0.831**
	96	0.485**	0.414**	0.700**
	120	0.261 ^{ns}	0.257 ^{ns}	0.479**
43	48	0.617**	0.538**	0.744**
	72	0.485**	0.390**	0.651**
	96	0.320*	0.346*	0.370*
	120	0.314*	0.202 ^{ns}	0.458**
4	48	0.358*	0.340*	0.458**
	72	0.288 ^{ns}	0.173 ^{ns}	0.453**
	96	0.163 ^{ns}	0.162 ^{ns}	0.375*
	120	0.209 ^{ns}	0.191 ^{ns}	0.377*

*, ** significant at %5 and %1 levels of probability, respectively

to pH measurements, but lower correlation coefficient than the other tests was determined.

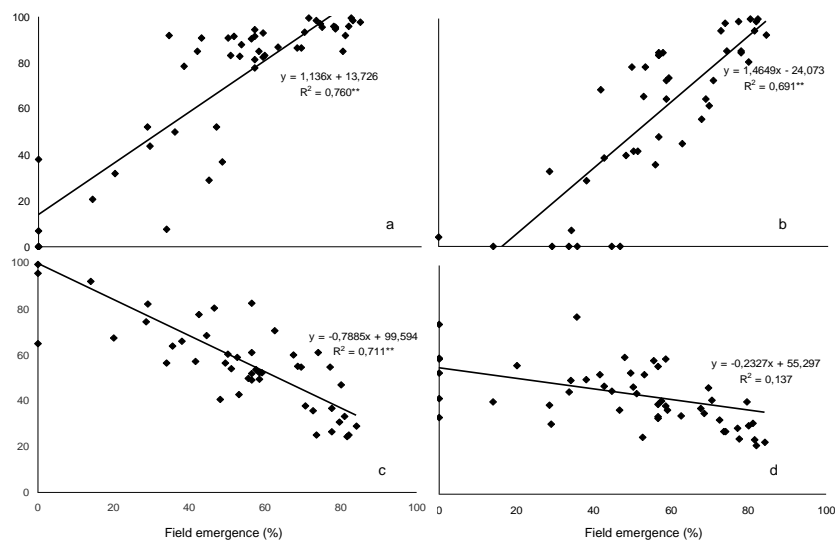
MGT indicates the speed and uniformity of germination and it is evaluated as evidence for seed vigor of several plants. Among the vigor tests, MGT

calculated in the cold test gave higher the coefficient ($r=0.658^{**}$) and better in predicting field emergence percentage; however, this value was lower than that of the cold, AA and EC tests. These results agree with Matthews and Khajeh-Hosseini (2006) and Santorum *et al.*

Table 4: Correlation coefficients between standard germination, laboratory emergence, field emergence and element concentrations of 50 seed lots of soybean

Seed element concentration	Standard germination (%)	Laboratory emergence (%)	Field emergence (%)
K (%)	-0.053 ^{ns}	-0.034 ^{ns}	-0.104 ^{ns}
P (%)	-0.054 ^{ns}	-0.070 ^{ns}	-0.205 ^{ns}
Na (mg L ⁻¹)	-0.297 ^{ns}	-0.152 ^{ns}	-0.164 ^{ns}
Fe (mg L ⁻¹)	0.296 ^{ns}	0.376*	0.223 ^{ns}
Mn (mg L ⁻¹)	-0.369*	-0.317 ^{ns}	-0.376*
Cu (mg L ⁻¹)	0.145 ^{ns}	0.289 ^{ns}	0.076 ^{ns}
Zn (mg L ⁻¹)	-0.070 ^{ns}	0.086 ^{ns}	0.019 ^{ns}

Significant at %5 level of probability; ns: not significant

**Fig. 1:** The relationship between field emergence and cold test (a), AA at 41°C for 72 h (b), EC for 24 h (c) and Mn concentration (d) of 50 seed lots of soybean

(2013), indicating that MGT had potential as an indicator of seedling emergence in maize.

Conclusion

The cold, EC and AA tests can be applied for estimating relative seedling emergence potential of soybean seed lots. Considering the correlation coefficients, the ideal conditions were found at 41°C for 72 h in the accelerated ageing test, which was the best method for predicting field emergence potential in soybean.

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