

THE CREDIBILITY OF SOME LABORATORY TESTS FOR PREDICTING THE PERFORMANCE OF SOYBEAN (*Glycin max* L. Merrill) SEED UNDER FIELD CONDITIONS

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ABSTRACT

Evaluating of seed viability by Lab. germination and Tetrazolium tests were a valuable approach to predict the number of emerging and surviving plants in the field. The present investigation aimed to determine the value of lab. Germination as prerequisite requirement to sale or purchase the seed in the market from one hand, and determine the value of electrical conductivity and tetrazolium as a quick and suitable lab. tests to predict the performance of soybean seeds under field conditions from the other hand. Lab. tests were conducted at Seed Technology Research Department, Agricultural Research Center, Giza, according to the international rules, while field trails were undertaken at Sakha Research Station (widely adapted for Egyptian Regions) during 2008 and 2009 growing seasons. The results showed highly significant correlation between the numbers of seeds capable of producing normal seedlings under lab. Conditions (germination percentage) and those emerged and surviving plants in the field (field emergence and survival percentages) indicating the important of sowing seeds of high germination capacity to achieve a good field establishment. The correlation coefficient for the relationship between the number of stained seed due to TZ treatment and those resulted in the field was also significant, indicating that the TZ test can provide more quick evaluation of soybean seed quality and be considered at calculating seeding rate. The value of electrical conductivity to predict seed performance in the field cannot be emphasized.

Keywords: TZ, Electrical conductivity tests- field performance- soybean seed.

INTRODUCTION

Evaluation of seed physiological quality is an important tool for decision making at the stages of production process, storage and commercialization. Evaluation of seed physiological quality includes seed germination and seedling vigor. Electrical conductivity (EC) and tetrazolium (TZ) tests have been realized by the International Seed Testing Association (ISTA,1993) as quick and suitable tests and be included in the routine work for evaluating seed viability, particularly if the number of tested samples at the testing station is large (Loeffler *et al.*,1988 and Wilson Jr, 1992). The standard germination test performed under optimal growth conditions and requires a relatively long period (eight days or more according to ISTA rules (1993) to be accomplished, and any deviation from ideal test conditions can affect the results by influencing the development of the essential structures of the seedling or by favoring the spread of microorganisms (Roberts, 1972). Electrical conductivity is based on the cellular membrane integrity of the seed and it provides possible detection of initial phases of the seed deterioration process (Delouche & Baskin, 1973). The values of Electrical conductivity can be taken to reduce or minimize the effect of deterioration and the consequent decrease in the seed physiological potential of a given seed lot (Dias &

Marcos Filho, 1996). The measurement of electrolytes in the leachate after the seeds are soaked in deionized water for a period and temperature depending on the species. Seeds of low conductivity value is more likely to produce normal and vigorous or seedlings (Siddique and Goodwin, 1985). Furthermore, seed germination can be predicted from the conductivity value with better reliability in low than in high and intermediate seed quality (Herter and Burris,1989). On the other hand, the TZ is a biochemical test, which differentiates live from dead seeds based on the activity of the respiration enzymes in seeds. Upon seed hydration, the activity of dehydrogenase enzymes increases resulting in the release of hydrogenions, which reduces the colorless tetrazolium salt solution (2, 3, 5-triphenyl tetrazolium chloride) into a chemical compound called formazan. Formazan stains living cells (respiring) with a red color while dead cells (not respiring) remain colorless. The viability of seeds is interpreted according to the staining pattern of seed tissues (ISTA,1993). Actually, the TZ test is considered the most significant discovery in seed testing in the 20th century, which provides an answer. It determines the percentage of viable seeds within a sample, even if seeds are dormant. The results of the TZ test indicate the amount of viable seeds in a sample that are capable of producing normal plants under suitable germination conditions. Moore (1971), reported that the tetrazolium test provides an accurate and rapid estimate of germination potential and it is relatively free from numerous and variable environmental influences that cause trouble in growth tests. The results of the TZ test for broadcasting field emergence was dependent on the incubated temperature and immersing period regardless of the seed size where the lower temperature and the longer period permitted staining process of the embryos to occur slowly and steady (Elmery and Elrabie ,1996). They added that the results of TZ test were correlated with those of field emergence.

The present investigation aimed to study the relationship between laboratory germination as a recommended test by international rules from one hand, and electrical conductivity and tetrazolium as quick tests and seedling emergence under field conditions.

MATERIALS AND METHODS

Seed tested samples of six soybean genotypes namely Giza 111, Giza 22, Giza 82, Giza 35, Giza 83 and Crwoford were obtained from the food legumes research section, field Crops Research Institute, Agricultural Research Center. The soybean crop was grown in the summer season of 2007 and 2008 seasons. Seeds of each soybean genotypes and harvesting season were stored in a cold storage of 10°C until the planting times comes up in 2008 and 2009 seasons. The seeds of each sample were undergo to the following tests at the laboratory of Seed Technology Research Section, Field Crops Research Institute, Agricultural Research Center, Giza, Egypt:

Standard Germination Test: Eight replications of 50 seeds per lot were planted in plastic boxes of 40X20X20 cm dimensions and contained sterilized sand. The boxes were watered and kept at 25 °C in an incubated chamber for 8 days. Normal seedlings were counted at 4, 8 days (first and final

accounts) according to the international rules of ISTA (1999). Germination percentage was calculated using the following formula outlined by Krishnasamy and Seshu (1990), where (Germination (%) = Number of normal seedlings/ Number of seed tested X100). Seedling vigour as measured by the length of normal seedling and its dry matter which was made on 10 seedlings per replicate at the end of germination test and after removal the seed attached. The seedlings were oven-dried at 70°C for 24 hours and weighed. Seedling vigor index was calculated using the following formula outlined by Ruan *et al.* (2002):

$$\text{Seedling vigor index} = \text{SDW} \times \text{G.P}$$

Where S.D.W = Seed Dry Weight.

G.P = Germination percentage.

Electrical conductivity test: The electrical conductivity of the leachate was determined according to procedures described by AOSA (1983). Four sub-samples of 50 seeds of each genotype were weighed and placed in Erlenmeyer flasks (250 ml) containing 200 ml of distilled water and covered by aluminum foil. The flasks were then placed in an incubator chamber at 25°C for 24 hours. The conductivity of the steep water was measured immediately after removal of samples from the incubator with a pipette-type conductivity cell attached to a bulk conductivity meter the conductivity values were expressed as $\mu\text{mhos/gm}$ seed weight.

Tetrazolium test. Four replicates 100 seeds of each sample were soaked in distilled water for 20 hours at room temperature. Then, the seeds were cut with a sharp blade in such a way that the whole embryo of each seed was exposed without any mechanical damage. Fragments of the cotyledon attached to the embryo were removed. The embryos were immersed in 0.5% solution of 2, 3, 5 triphenyl tetrazolium chloride at 25 °C for 6 hours after which they were rinsed with distilled water and transferred to petri dish containing water to avoid drying. Complete-staining embryos of each replicate were calculated through magnifying lens (5 X) under fluorescent light.

Field emergence: Two experiments were conducted at Sakha Agricultural Research Station, Kafr El-Sheikh, (widely adapted for Egyptian Regions and about 130 km from Cairo). A completely randomized design was utilized with four replications of 100 seeds per lot were sown on three different dates 16 May and 6 and 11 June, respectively, during 2008 and 2009 seasons. All cultural practices concerning land preparation and irrigation were done as recommended for growing soybean crop. Seedling emergence and plant survival were recorded at time intervals until constant (30 days from seed sowing) and the highest figures were used. Other calculations were made including relative field emergence or Field emergence index (denote to the percentage of viable seeds produced plants in the field /seed germination as determined in the laboratory), relative field survival or Field survival index (denote to the percentage of viable seeds produced plants survived / seed germination as determined in the laboratory). Data were subjected to standard analysis of variance procedures outlined by Steel and Torrie (1980). Simple

correlation coefficients were calculated to compare the association between values of TZ, EC and seedling vigor under controlled laboratory conditions and field emergence.

Table (1): The pedigree of the soybean cultivars:

Cultivar	Pedigree
Giza 21	Crowford X Columbus
Giza 22	Selected from MBB-133-9 Union x L76-0038 Williams x PI 161451
Giza 35	Crowford x M.Presto
Giza 82	Crowford x Celest
Giza 83	Crowford x Celest
Giza 111	Crowford x Forrst
Clark	Lincoln (2) x Richland
Crowford	Williams x Columbus

RESULTS AND DISCUSSION

Table (2) shows that there was seasonal variation of varietal performance as to seed germination at first and final accounts, which included higher figures in 2008 compared with 2009 season. There was also varietal variation in seed germination within each season where seeds of Giza 111, Giza 22 had higher germination values (87%) and Clark, Crowford, Giza 82 had lower germination values (79,80 and 81%,respectevly). The averages of seed germination of seeds of all varieties were 82.4 and 80.6 % in 2008 and 2009 seasons, respectively. The correlation coefficient for the relationship between the germination values at the first and final accounts was significant ($R= 0.958^{**}$ and 0.952^{**} in 2008 and 2009 seasons, respectively) indicating the importance of the number of faster germinated seeds when evaluating seed quality. The figures of seedling length and its dry matter showed similar tendency to those of seed germination at both first and accounts were the higher seed germination the taller and the heavier seedling. Furthermore, the correlation coefficient for the relationship between the germination values and those of seedling length and its dry matter was significant ($R= 0.808^{**}$ and 0.607^{*} in 2008 and 2009 seasons, respectively, indicating the importance of various seed germination aspects. As a result vigor index was reflected the capability of seeds to produce vigorous seedling under lab conditions.

Table (3) shows that seasonal variation concerning the values of seed conductivity and the number of seeds stained by red color in TZ test. The values of seed conductivity in 2008 season were comparable low to those of 2009 season. In contrast, the percentage of stained seeds of the same variety in 2008 season was higher than that of 2009 season. Similar results were obtained by Eraky *et al.*, (2010) and Elemery and Elrabie (1996). The seeds of high electrical conductivity value might indicates their poor quality as in the cases of those of the varieties Giza 83, Giza 21 and Giza 82, while Seeds of low values revealed good quality seeds as in the cases of the varieties Giza 111and Giza 35. Other varieties show intermediate values of electrical conductivity and percentage of stained seeds either. The correlation

coefficient for the relationship between the germination values and those of electrical conductivity was negatively significant ($R= 0.827^{**}$ and 0.886^{**} in 2008 and 2009 seasons, respectively). Whereas, The correlation coefficient for the relationship between the germination values and number of stained seed due to TZ treatment was significant ($R= 0.827^{**}$ and 0.670^* in 2008 and 2009 seasons, respectively), indicating that both EC and TZ tests can provide more quick evaluation of soybean seed quality. The results obtained by Ali (1979) came in large extent to the same conclusion.

Table (2): Seed germination (%), seedling length and its dry matter and vigor index of some soybean varieties during 2008 and 2009 seasons

Genotype	Seed germination %		Seedling Length (Cm)	Seedling dry Matter (g)	Vigor index
	4 Days	8 Days			
Season 2008					
Giza 21	31	82	18.4	0.137	11.2
Giza 22	34	87	20.2	0.147	12.8
Giza 35	32	83	19.6	0.149	12.4
Giza 82	28	80	18.8	0.139	11.1
Giza 83	28	81	19.1	0.142	11.5
Giza 111	34	87	20.8	0.154	13.4
Clark	28	79	18.2	0.142	11.2
Crowford	28	80	18.2	0.141	11.3
L.S.D. at 5%	2.2	2.2	2.1	0.012	
Season 2009					
Giza 21	27	81	16.5	0.127	10.3
Giza 22	29	82	17.5	0.136	11.2
Giza 35	29	83	18.7	0.141	11.7
Giza 82	26	79	17.1	0.131	10.3
Giza 83	25	79	17.2	0.131	12.0
Giza 111	29	84	19.1	0.143	10.6
Clark	25	79	17.4	0.134	10.5
Crowford	25	78	17.8	0.135	0.135
L.S.D. at 5%	2.6	2.5	2.3	0.014	

Table (3): Seed conductivity ($\mu\text{bmhos/g}$) and embryo staining (%) of some soybean genotypes.

Genotype	Season 2008		Season 2009	
	EC	TZ	EC	TZ
Giza 21	0.13	86	0.18	84
Giza 22	0.11	89	0.14	86
Giza 35	0.10	87	0.13	85
Giza 82	0.13	85	0.19	82
Giza 83	0.14	83	0.17	80
Giza 111	0.09	88	0.12	86
Clark	0.12	81	0.17	78
Crowford	0.11	86	0.16	80
L.S.D. at 5%	0.02	2.6	0.03	2.8

It is worthy to mention that Oregon Seed Certification Program in USA allows tagging seeds based on the TZ test until the germination test results are available.

Field emergence of seeds of soybean varieties during 2008 and 2009 seasons at three different dates is shown in table (4). There was seasonal and varietal variation where field emergence from sowing the seeds at 6th of June was slightly high compared with sowing dates at 16th of May and 26th of June. The results almost followed the same patterns of germination percentage at first and final accounts. Andric *et al.* (2004), Ghuge *et al.*, (2007) and Eraky *et al.*, (2010), reported similar results. Field emergence was considerably high from sowing seeds of high germination values, i.e. Giza 22, Giza 35 and Giza 111. The correlation coefficient for the relationship between the germination values and those of field emergence from different sowing dates was significant ($R=0.868^{**}$, 841^{**} , 0.944^{**} in 2008 season, and, $R=0.870^{**}$, 0.872^{**} , 0.806^{**} in 2009 season), indicating the importance of these values when purchasing the seed from the market. The averages of field emergence of seeds of all varieties were 72.1 and 70.8 % in 2008 and 2009 seasons, respectively. Therefore, precaution should be taken when calculating seeding rate to achieve the indented plant population. The germination percentage of seeds is available on the tag of seed package. This specially true for the varieties Clack and Crowford were field emergence below 70% (an average of different sowing date and season). Generally, an accurate plant population can be obtained from seeds of high germination percentage under laboratory conditions. Whereas, the correlation coefficient for the relationship between the field emergence and number of stained seed due to TZ treatment was significant ($R=0.874^{**}$, 735^{**} , 0.873^{**} in 2008 season, and, $R=0.881^{**}$, 0.880^{**} , 0.904^{**} in 2009 season). The correlation coefficient for the relationship between the field emergence was negatively significant in a few cases and insignificant in most ones. These results revealed indicating that the TZ test can provide more quick evaluation of soybean seed quality and be considered at calculating seeding rate. The value of electrical conductivity to predict seed performance in the field cannot be emphasized.

Table (4): Field emergence (%) of some soybean genotypes from different seed sowing dates during 2008 and 2009 seasons.

Genotype	Sowing date /Season 2008				Sowing date /Season 2009			
	16 May	6 June	26 June	Average	16 May	6 June	26 June	Average
Giza 21	72	75	74	73.7	71	72	73	72
Giza 22	75	77	76	76	73	75	75	74.3
Giza 35	73	76	75	74.7	71	74	74	73
Giza 82	70	73	71	71.3	67	69	68	68
Giza 83	69	70	68	69	66	68	66	66.6
Giza 111	75	77	76	76	72	76	74	74
Clark	67	72	68	69	67	71	68	68.6
Crowford	68	71	69	69.3	69	71	69	69.6
L.S.D. at 5%	2.5	2.2	2.4		2.3	2.7	2.2	

Almost, the results of relative field emergence, field survival and relative field survival were followed by the trend of field emergence (tables 5, 6 and 7). The higher field emergence the higher field survival and relative field survival. While, the averages of field survival were 69.5 and 67.7 % in 2008 and 2009 seasons, respectively, the averages of field emergence were 72.1 and 70.8 %.

Table (5): Field survival (%) of some soybean genotypes from different seed sowing dates during 2008 and 2009 seasons.

Genotype	Sowing date /Season 2008				Sowing date /Season 2009			
	16 May	6 June	26 June	Average	16 May	6 June	26 June	Average
Giza 21	70	73	69	70.6	69	70	65	68
Giza 22	71	75	74	73.3	69	71	66	68.7
Giza 35	70	74	71	71.7	70	72	66	69.3
Giza 82	66	69	68	67.7	67	69	65	67
Giza 83	65	66	66	65.7	64	66	66	65.3
Giza 111	72	74	72	72.7	70	72	68	70
Clark	64	69	67	66.7	67	68	66	67
Crowford	66	69	67	67.3	65	68	65	66
L.S.D. at 5%	2.1	2.2	2.2		2.3	2.3	2.2	

Table (6): Relative field emergence (%) of some soybean genotypes from different seed sowing dates during 2008 and 2009 seasons.

Genotype	Sowing date /Season 2008			Sowing date /Season 2009		
	16 May	6 June	26 June	16 May	6 June	26 June
Giza 21	87.8	91.5	90.2	86.6	87.8	89.0
Giza 22	86.2	88.5	87.3	83.9	86.2	86.2
Giza 35	87.9	91.6	90.4	85.5	89.2	89.2
Giza 82	87.5	91.3	88.8	83.8	86.3	85.0
Giza 83	85.1	86.4	83.9	81.5	83.9	81.5
Giza 111	86.2	88.5	87.4	82.8	90.5	88.1
Clark	84.8	91.1	86.1	84.8	89.9	86.1
Crowford	85.0	91.0	86.3	86.3	88.8	88.5

Table (7): Relative field survival (%) of some soybean genotypes from different seed sowing dates during 2008 and 2009 seasons.

Genotype	Sowing date /Season 2008			Sowing date /Season 2009		
	16 May	6 June	26 June	16 May	6 June	26 June
Giza 21	85.4	89.0	84.2	84.2	85.4	79.3
Giza 22	81.6	86.2	85.1	79.3	81.6	75.9
Giza 35	84.3	89.2	85.5	84.3	86.8	79.5
Giza 82	82.5	86.3	85.0	83.8	86.3	81.3
Giza 83	80.2	81.5	81.5	79.0	81.5	81.5
Giza 111	82.8	85.1	82.8	80.5	82.8	78.2
Clark	81.0	87.3	84.8	84.8	86.1	83.5
Crowford	82.5	86.3	83.8	81.3	85.0	81.3

By comparing these results, the reduction of emerged plants capable of surviving was 2.6 and 3.1% % in 2008 and 2009 seasons, respectively. But, such reduction was 12.9% if the comparison is made with the results of seed germination (82.4 and 80.6 %% in 2008 and 2009 seasons, respectively). These comparisons emphasize the important of sowing seeds of high germination capacity, taking into consideration planting date.

REFERENCES

- Ali, A.G., A. (1979). Electrical and chemical evaluation of planting seed quality. Ph.D. Thesis, Fac. North Carolina State Univ. U.S.A.
- Andric,L.; T. Cupic; T. Teklic; B. Simic and H. Plavsic. (2004). Field emergence index (FEI) of soybean depend on cultivar, seed age, seed treatment and planting date. Sjemearstvo, 21 (3\4): 127-133. (C.F. Soybean. Txt, CAB abstracts record 60 of 245.
- AOSA (1993). Association of Official Seed Analysis. Seed Vigor Testing Hand Book, No-32 P.160.
- Leoffler, T.M.; D.M.Tekrny and D.B. Egli (1988). The bulk of conductivity as an indicator of soybean seed quality. J. Seed Technol., 12, 37-53.
- Delouche, G.F. and D. C.C. Baskins (1973). Accelerated aging techniques for predicting the relative storeability of seed lots. Seed Sci.Technol.1, 427-452.
- Dias,D.C.F.S. and J. Marcos Filho. (1996). Electrical conductivity tests to evaluate the vigour of soybean (*Glycine max* L. Merril) seeds. Scientia-Agricola, 53 (1):31-42.
- Elemery, M.I. and H.G. Elrabie (1996). The value of Tetrazolium (TZ) and electrical conductivity (EC) tests for forecasting seed viability and vigor of some faba been (*Vicia faba* L.) genotypes. Annals Agric. Sci.,41 (2): 837-847.
- Eraky, A. M.; Hania A.A.G. Ali; H. A. Rabie and M.I. Elemery. (2010). The relationship between seed quality measurements and field emergence of soybean. Zagazig J. Agric. Res., 37 (4): 829-855.
- Ghuge, T. D.; S. B. Jodhav and A.K. Gore (2007). Effect of location and packaging materials on storability of soybean. J. of Soils and Crops, 17(2):427-428.
- Herter,V. and J. S. Burris (1989). Evaluating drying injury on corn seed with a conductivity test. Seed Sci. Technol., 17, 625-638.
- International Seed Testing Association (ISTA, 1993). International Rules for Seed Testing. Seed Sci. and Technol., 21: 187-209.
- International Seed Testing Association (ISTA, 1999). International Rules for Seed Testing. Handbook of Vigour Test Methods. 3rd Edition, 22-35.
- krishnasamy, V. and D.V. Seshu (1990). Phosphine fumigation influence on rice seed germination and vigor. Crop Sci., 30:28-85.
- Moore, R.P. (1971). Seed peanuts potential vs. actual germination percentages. J. Amer. Peanut Research and Education Assoc.Inc., 3(1):168-171.
- Robert, H.E.(1972). Viability of seeds. Chairman and Hall Ltd. London:197pp.

- Ruan, S.; Q.Xue and K. Tylkowska (2002). Influence of priming on germination of rice (*Oryza sativa* L) seed and seedling emergence and performance in flooded soil. *Seed Sci. and Technol.* 30:61-67.
- Siddique, M. A. and P. B. Goodwin (1985). Conductivity measurements on single seed to predict the firm inability of frenchbean. *Seed Sci. and Technol.*, 13: 643-652.
- Steel, R.G. D. and J. H. Torrie (1980). Principles and procedures of statistics. 2nd Ed., McGraw Hill Book Co., N.Y., USA.
- Wilson, Jr. D. O. (1992). A unified approach to interpretation of single seed conductivity data. *Seed Sci. & Technol.* 20:155-163.

مصادقية بعض الاختبارات المعملية للتنبؤ باداء تقاوى فول الصويا تحت ظروف الحقل

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تقييم حيوية التقاوى ببعض الاختبارات المعملية مثل اختبارا الإنبات والتترازوليم وسيلة فعالة للتنبؤ بعدد النباتات التي تظهر أثناء التكشف الحقلى وتلك التي تستكمل مراحل النمو التالية. أستهدفت الدراسة الحالية تحديد أهمية نسبة الإنبات فى المعمل والتي تعد إحدى المتطلبات الأساسية لتسويق التقاوى من ناحية، وكذلك تحديد جدوى استخدام بعض الاختبارات السريعة مثل التوصيل الكهربى والتترازوليم كبديل لاختبار الإنبات المعملى للتنبؤ باداء تقاوى فول الصويا تحت ظروف الحقل من ناحية أخرى . أجريت الاختبارات المعملية بقسم بحوث تكنولوجيا البذور بمركز البحوث الزراعية بالجيزة ، بينما أجريت التجارب الحقلية بمحطة البحوث الزراعية بكفر الشيخ خلال موسمى النمو ٢٠٠٨ ، ٢٠٠٩ . أوضحت النتائج وجود ارتباط عالى المعنوية بين عدد البذور التي تنتج بادرات طبيعية تحت ظروف المعمل وعدد النباتات التي تظهر ويستمر نموها فى الحقل، وهذا يوضح أهمية استخدام تقاوى ذات نسبة إنبات عالية لإنتاج كثافة نباتية جيدة تحت ظروف الحقل. كذلك كان معامل الارتباط عالى المعنوية للعلاقة بين عدد البذور الملونة نتيجة معاملتها بمحلول ملح التترازوليم كلوريد وعدد النباتات التي تكشفت فى الحقل، مما يجعل من اختبار التترازوليم الذى يستغرق عدد ساعات بديلا جيدا لاختبارات الإنبات المعملى التى يستغرق ثمانية أيام وفقا للقواعد الدولية لفحص البذور. إلا أن درجة مصادقية استخدام اختبار التوصيل الكهربى لمنقوع البذور كبديل لاختبار الإنبات لم ترقى إلى درجة اختبار التترازوليم.

قام بتحكيم البحث

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