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VIGOR TESTS USED TO RANK SEED LOT QUALITY AND PREDICT FIELD EMERGENCE IN WHEAT

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Abstract

The objective of the present study was to examine the relationship between various seed quality tests and field emergence of the new and old wheat cultivars. Laboratory and field tests were conducted during 2003 and 2004 in NWFP Agricultural University Peshawar, Pakistan, to investigate the suitability of various laboratory vigor tests, to rank quality of commercial seed lots, and to predict seedling field emergence (FE) of 32 samples of 4 wheat varieties. Seeds of four wheat cultivars Takbeer 2000, Haider 2000, Bakhtawar-92 and Fakhri Sarhad were produced in 8 different location of NWFP, Pakistan during 2003 and 2004. Mature seeds were harvested, threshed and cleaned before determining standard germination and other vigor tests. Results showed that among all tests, germination index (GI), Accelerated aging (AA) and Electrical conductivity (EC) provided the best estimate of seed vigor for the four wheat cultivars, both for ranking seed lots quality and predicting field emergence. The GI, AA and EC tests better indicated seed lot quality and predicted FE than SG of the four cultivars over the 2-years followed by Radical length (RL). The electrical conductivity (EC) result was not only poorly related to FE, but also poorly related to the standard germination (SG) of a wide range of seed lots of the four varieties that varied in viability. Initial count of standard germination (SGi) generally performed more poorly than the other vigor tests. From this study and previous work on wheat crop, we conclude that GI, AA and EC test for wheat crop have the potential to be developed as improved vigor tests for ranking seed lot quality and predicting seeding performance under temperate regions of the world.

Introduction

The evaluation of vigor tests for predicting seed planting value is important in providing better results for ranking the quality and for indicating planting value of seed lots than the standard germination test. Many vigor test methods have been proposed by different organization viz., Anon., (1993), Anon., (92002) and the results obtained from their studies showed differences among vigor tests used. However, the correlation coefficients between germination, vigor, and seedling emergence varied according to species, vigor test methods and the field sowing conditions. Many research workers have reported significant correlation coefficients between field emergence and standard laboratory germination tests but they have also reported inconsistencies and difficulties with the prediction of field emergence. The aim of the SG test is to estimate the germination potential of a seed lot, which can then in turn be used to compare the quality of different lots and also estimate the field planting value of all crops (Anon., 2002). The methodology of SG has been refined to a high level of reproducibility and reliability; however, SG does not always indicate seed lot potential performance, especially if field conditions are less than optimal (Hampton & TeKrony, 1995). Seed lots that do not differ in germination may differ in deterioration level and may differ substantially in field/ storage performance (Powell & Matthews, 1984; Kolasinska *et al*., 2000). Seed vigor tests therefore have been proposed to detect differences in potential seed lot performance. The critical requirements of a vigor test include (i) it must provide a more sensitive index of seed quality than does SG (McDonald, 1980; Perry, 1984) and (ii) it must better predict planting value of high germinating seed lots than does SG (Hampton & TeKrony, 1995). Although some vigor test methods have been developed and successfully used for several major agronomic crops (AOSA, 1993; Hampton & TeKrony, 1995), these methods have not been studied extensively in forage crops (Wang *et al*., 2001). Wang & Hampton (1994) reported that CD (Controlled Deterioration) and EC tests were more sensitive and accurate for predicting red clover (*Trifolium pratense* L.) storability and field emergence than the SG and germination index tests. Similar results were obtained for alfalfa (Wang *et al*., 1992, 1996). Happ *et al*., (1993) demonstrated that seedling growth, EC, and accelerated aging tests could differentiate seed quality among commercially acceptable perennial ryegrass (*Lolium perenne* L.) seed lots. Marshall & Naylor (1995) found that CD (Controlled Deterioration), respiration and moisture stress tests could indicate field performance of *Lolium multiflorum* Lam. The objectives of this study were to examine various seed vigor tests for their ability to rank seed lot quality and to predict seedling field emergence in four wheat cultivar viz., Takbeer 2000, Haider 2000, Bakhtawar-92 and Fakhri Sarhad under temperate sowing conditions. These cultivars are important grain and forage crops in temperate regions of the Pakistan and play a significant role in grain and livestock production and environmental protection. Furthermore, little information is currently available on seed vigor tests for these cultivars.

Materials and Methods

Seeds of 4 wheat cultivars viz., Takbeer 2000, Haider 2000, Bakhtawar-92 and Fakhri Sarhad were produced in the field in 8 different locations of NWFP–Pakistan during 2003 and 2004.All samples were kept at 4°C before use. Laboratory and field tests were conducted in 2003 and 2004 in Department of Agronomy, NWFP. Agricultural University Peshawar (Tables 1 & 2). The laboratory germination test was done on four replicates of 50 seeds per seed lot at 25°C. Each day, normal seedling at least 1.5 cm long were counted and removed until no further seeds germinated. Germination index (GI) was calculated by the formula:

$$GI=\sum(Gt/Tt)$$

where Gt = germination percentage at *t*th day, Tt = day of germination test. In addition, number of normal seedlings at the initial count (Table 2) was used to further assess seed quality (SGi). Finally, percentages of normal seedlings (SG) and hard seeds in all varieties of wheat were calculated at the end of the test following ISTA rules (1999); viability of cereal seeds was expressed as a total sum of SG and hard seed. Radical length was measured for 10 randomly selected seedlings per replicate for each lot. The accelerated aging (AA) test was conducted as described by Hampton & TeKrony (1995). Two lots of 50 seeds from each treatment replicated four times were kept for 72 hours at 42°C. Maximum water was used to maintain nearly 100 % R.H. Germination was determined after aging by planting 50 seed sample in rolled moist towels at 25°C as described by ISTA (Anon., 2002) 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 $^{+}_{-}$ 2 ^{0}C. The electrical conductivity was measured with conductivity meter and expressed as µS cm¯1g-1(ISTA.1995).In both years, FE was tested at New Development Farm of the NWFP Agricultural University Peshawar from 15 May to 15 June. The soil of the experimental site was silty clay loam with a clay type montmorillonite, low in nitrogen (0.03-0.04%), organic matter (0.8-0.9%) and alkaline in reaction with a pH 8.0-8.2 (Shah *et al.*, 1993). Field was irrigated before sowing. After planting, plots were irrigated only once at 7 d after sowing and no rainfall occurred during the test. Four replicates of 100 seeds per lot were sown by hand at a depth of 1 cm, with spacing of about 1 cm between seeds and 40 cm between rows. Emergence counts started when the first seedling was visible and continued at 2-d intervals until no further seedlings emerged. Soil moisture content (SMC) and climatic conditions are listed in Table 3. The SMC in the 0- to 10-cm layer was measured weekly from two replicates of 10-g soil samples collected each week. Samples were taken randomly with a 50-cm-diam soil auger. The SMC was determined gravimetrically after drying at 105 to 110°C for 6 h and expressed as percentage of fresh sample weight. An emergence index (EI) was calculated using the formula of Scott *et al.*, 1984:

$$EI = \{TiNi/S\}$$

where Ti is the number of days after sowing, Ni is the number of seeds germinated on day i, and S is the total number of seeds planted. Final emergence was calculated as a percentage of the number of seeds planted.

Statistical analysis: Statistical analysis was performed by the Statistical program (StatSoft, Inc., Tulsa, OK, 1992). Analysis of variance with LSD was performed to rank the quality of seed lots, and arcsine transformation was applied to percentages before analysis. Simple correlation coefficients were calculated to evaluate the associations among laboratory tests and seed field emergence for cultivar.

Table 1. Germination methods for test cultivars.

Wheat cultivars	Substrate	Temperature (°C)	Initial count	Final count	Reference
			Days		
Takbeer-2000	BP	25	7	14	ISTA,2002
Haider-2000	BP	25	7	14	ISTA,2002
Bakhtawar-92	BP	25	7	14	ISTA,2002
Fakhre-Sarhad	BP	25	7	14	ISTA,2002

BP: Blotting papers.

Table 2. Soil moisture content in the 0- to 10-cm layer and weather conditions during the field emergence test.

Location	Sowing date	Test duration	At sowing	Mean of the test period	Daily mean temperature (°C*)
		Days	g/kg		
NWFP. Agric. Uni. Peshawar	May, 15 2003	30	137	87	18.8
NWFP. Agric. Uni. Peshawar	June,15 2004	30	132	58	22.6

*Daily mean temperatures were obtained from the meteorological station of NWFP. Agric.Uni. Peshawar, Pakistan.

Results

Quality of seed lots evaluated by different tests: Qualities of seed lots as evaluated by the different vigor tests are given in Table 3. The SG results showed that all seeds had viability ≥80%, which is commercially acceptable for seed in Pakistan. Laboratory tests used were able to rank the seed lots into various quality groups on the basis of LSD means separation; however, the numbers of groups ranked for each variety differed among tests. We assume that a powerful test identifies more seed lot than a weak test. GI, AA and EC provided the most sensitive index in ranking seed lot quality over both years. For example, in 2003, EC divided seed lots of wheat varieties into four groups and whereas SG divided lots of the four wheat varieties into two groups only. Similarly, in 2004, the same vigor tests divided seed lots of the four cultivars into four or three groups, whereas SG divided each of the four varieties into two groups. The AA test ranked seed lots into more groups than SG in both years for the said varieties. The GI differentiated seed lots of each cultivar into more groups than SG only in 2003. The other laboratory tests did not show better or consistently better lot discrimination than SG (Table 3). The most sensitive parameter in ranking seed lot quality of cultivars was GI dividing the seed lots into three groups in both years compared with one group for SG in 2003 and two groups for SG in 2004. The AA, GI and EC tests were more sensitive than SG in discriminating quality among different seed lots of different wheat cultivars. The other tests did not constantly show any advantage over SG for the four wheat varieties (Table 3).Mean FE was generally lower than that of the SG in both years and the differences varied among the cultivars and sowing time. Values of FE expressed as a percentage of SG (relative field emergence) generally followed the pattern: Takbeer-2000>Haider-2000>Bakhtawar-92>Fakhre Sarhad (Table 3)

Correlations between standard germination, vigor and field emergence: The relationships among SG, vigor and FE are listed in Tables 4. For all wheat varieties, all laboratory test results significantly correlated with FE except SG, SGi, and GI in 2004 and RL in both years. However, the AA and EC tests predicted FE better than did SG and the other tests. Two-year correlation coefficients of FE with other tests were smaller than those with SG (Table 4). For Haider-2000, SG, GI, AA, and EC significantly correlated with FE in both years, but correlation coefficients of AA and EC with FE over 2 yr was higher than those with SG. Some of the other vigor tests correlated significantly with FE and predicated FE better than did SG in one year, but this relation did not hold in the other year (Table 4). In Bakhtawar-92, SGi significantly correlated with FE during 2004 and *r* values of the test were smaller than those of SG during both years. The AA correlated significantly with FE during both years. The other laboratory tests were not correlated to the FE in either year. The AA, EC and GI vigor tests predicted FE better than did SG in Fakhri-Sarhad (Table 4).

Table 3. Quality of seed lots of Wheat cultivars evaluated by different vigor tests in 2003 and 2004.

Wheat cultivars	Test	2003				2004			
		No. of lots tested	Mean	Rang	*No. of groups	No. of lots tested	Mean	Rang	*No of groups
Takbeer-2000	Via	8	87	80-90	–	8	82	80-90	–
	SG		82	72-88	2		78	70-86	2
	SGi		57	18-80	2		63	56-70	2
	GI		25.1	6.7-37.0	3		22.6	20.1-25.2	3
	RL		5.0	2.9-6.8	2		1.0	0.4 -1.5	2
	AA		55	40-65	4		52	53-67	3
	EC		107	77-135	3		137	104 -180	3
	FE		69	36-82	4		54	40-57	3
Haider-2000	Via	8	88	80-90	–	8	87	80-90	–
	SG		72	59-79	2		77	62-89	2
	SGi		69	57-75	1		70	55-80	1
	GI		34.5	24.0-43.4	3		30.8	20.3 -38.4	3
	RL		2.4	2.0-3.4	1		1.1	0.7 -1.8	2
	AA		55	50-65	4		50	45-60	4
	EC		126	102-151	3		157	113-213	3
	FE		79	70-86	3		50	43-60	3

Table 3. Quality of seed lots of Wheat cultivars evaluated by different vigor tests in 2003 and 2004.

Wheat cultivars	Test	2003				2004			
		No. of lots tested	Mean	Rang	*No. of groups	No. of lots tested	Mean	Rang	*No of groups
Bakhtawar-92	Via	8	80	75-85	–	8	81	75-90	–
	SG		88	70-94	2		85	70-80	2
	SGi		85	67-92	2		81	58-90	2
	GI		42.6	23.0-54.0	3		37.2	23.2-47.8	3
	RL		11.2	9.0-13.0	2		3.7	2.0-4.8	2
	AA		67	60-84	3		71	65-82	4
	EC		42	38-45	3		43	39-45	3
	FE		37	26-50	2		77	62-85	3
Fakhri- Sarhad	Via	8	80	70-80	–	8	80	75-90	–
	SG		80	72-87	2		80	72-87	2
	SGi		18	10-30	1		18	10-30	1
	GI		12.7	11.0-14.4	3		12.7	11.0-14.4	3
	RL		5.0	4.1-6.1	2		5.0	4.1-6.1	2
	AA		42	30-60	4		51	30-60	4
	EC		61	54-67	3		61	54-67	3
	FE		41	18-53	1		41	18-53	1

SG: Standard germination (%), SGi: Initial count of SG (%), GI: Germination index, RL: Radical length (cm), AA: Accelerated Aging (%), EC: Electrical conductivity (µS cm^{-1} g^{-1}), FE: Field emergence (%), Via: Viability, including germination and hard seed.

*Number of groups divided by LSD test at $p<0.05$ and arcsine transformation was applied to percentages before analysis.

Table 4. Correlation coefficients (r) between standard germination or vigor and field emergence of seed lots for four wheat cultivars tested in 2003 and 2004.

Quality tests	Takbeer-2000		Haider-2000		Bakhtawar-92		Fakhre Sarhad	
	FE 2003 (n=8)	FE 2004 (n=8)	FE2003 (n=8)	FE2004 (n=8)	FE 2003 (n=8)	FE 2004 (n=8)	FE 2003 (n=8)	FE 2004 (n=8)
SG	0.696**	0.468	0.600**	0.490*	0.494*	0.563**	0.440*	0.463**
SGi	0.693**	0.010	0.295	0.501*	0.460	0.511**	0.394*	0.425**
GI	0.685**	0.086	0.579**	0.615**	0.303	0.267	0.394*	0.370**
RL	0.450	0.100	0.285	0.389	0.242	0.285	0.234*	0.263**
AA	0.767**	0.688*	0.745**	0.583**	0.510*	0.568**	0.435*	0.555**
EC	–0.708**	–0.610*	–0.757**	–0.658**	–0.540*	–0.645**	–0.533*	–0.545**

* Significant at $p<0.05$. ** Significant at $p<0.01$.

SG: Standard germination, SGi: Initial count of SG, GI: Germination index, RL: Radical length, AA: Accelerated Aging, EC: Electrical conductivity, FE: Field emergence.

Discussion

Small seed cereal crop like wheat are generally difficult to germinate under adverse field conditions (Wang *et al*., 2001). Field experiments were conducted under Peshawar climatic conditions during 2003 and 2004. We expected that FE would be variable and substantially lower than SG. This was confirmed particularly in all wheat varieties during both years. This result supports previous findings on small grains legume, such as soybean (Khan *et al*., 2007) and forage species, such as *Lolium multiflorum* (Marshall & Naylor, 1995), *Bromus biebersteinii* Roemer & Schultes (Hall & Wiesner, 1990), *Lotus corniculatus* L., and alfalfa (Wang *et al*., 1996). Poor establishment of the small seeded crop is common worldwide problem; more research is crucial to address this problem. Accelerated aging and electrical conductivity provided a more sensitive test in ranking seed lot quality and showed a higher correlation with FE than did SG in four wheat varieties over 2 years (Tables 1 and 4). Data from previous research on small grain crop similarly indicated that, compared with SG, AA and EC was more sensitive in ranking seed lot quality in alfalfa (Wang *et al*., 1992, 1996) and *Trifolium pratense* (Wang & Hampton, 1991), and both tests could provide a more reliable index for predicating FE of wheat cultivars in drier soils (Wang *et al*., 1996). Our results confirmed that wheat seed lots differ in vigor (Hampton, 1991) and that cell membrane integrity, as estimated by electrolyte leakage by the EC test, is a fundamental cause of vigor degradation (Powell, 1988). In addition, both AA and EC tests have the tremendous advantage of simplicity and rapidity and meets the requirements for a good vigor test (Hampton & Coolbear, 1990). These two tests that have been approved by the 26th ISTA Congress held in France in 2001 to be included in the International Seed Testing Rules as a standard method for testing seeds of *Pisum sativum* L., (TeKrony, 2001). Compared with SG, the EC test was more sensitive in ranking seed lots for quality (Table 1). These findings were supported by the earlier research on *Lolium perenne* (Bennett *et al*., 1998; Cookson *et al*., 2001), *Lolium multiflorum* (Marshall & Naylor, 1995), and *Bromus biebersteinii* (Hall and Wiesner, 1990). However, current results differed from the work on *Festuca arundinacea* Schreber reported by Han *et al*., (1995), who found that EC correlated significantly with FE both in glasshouse (r = –0.930) and field (r = –0.922) studies. Our study, by adding some results from low viability seed lots to the test data set, further demonstrated that the EC was also not correlated with SG results of the grass species. Seeds with higher EC results poor germination. However, the explanation for the poor correlation of EC with FE and SG in most grass seeds is unknown and warrants further investigation. Among the other combinations between the wheat cultivars and laboratory tests used, GI ranked wheat seed lots into more groups and better predicted FE than did SG for both years (Table 3 and 4).

Conclusions and Recommendation: From this study and previous work on small grains cereals, we conclude that AA, EC and GI tests have the potential to be developed as improved vigor tests for ranking seed lot quality and predicting seed performance for small seeded cereals crops. Further work with more seed lots and sowing conditions is needed for confirmation of these findings.

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