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Correlation of Laboratory Seed quality with Seedling Performance of Wheat (*Triticum aestivum* L.) in the Field

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Farmers and seed producers have long recognized that the labeled germination percentage often overestimates the actual seedling emergence in the field of seed lots. The aim of this study was to determine correlation between laboratory tests and seedling performance of in the field. Laboratory tests included standard germination, Tetrazolium tests for viability, electrical conductivity, speed of germination and seedling evaluation. The experimental design was Randomized Complete Block Design with four replications and five crops as the treatments. Parameters measured in the field were emergence, seedlings growth rate and speed of germination. Accession1285 had lower laboratory germination of 87.5 % than seedling emergence in the field of 89.8 %. Accession 50 had 81.8 % and 83% laboratory germination and seedling emergence respectively. In this study biochemical seed vigor tests of electrical conductivity and tetrazolium, seedling evaluation and seedling growth tests were correlated with seedling performance in the field. Correlation coefficient (r) and coefficient of determination (R^2) between laboratory and field tests were obtained and tested at 0.05 and 0.01 probability. Laboratory germination was significantly lower ($P \leq 0.05$) than seedling germination in the field.

*Key words***:** Wheat, laboratory germination, field emergence, seedling growth rate, performance, cereal and vegetable crops.

Introduction

The seed market has experienced important changes such as commercial globalization and an increase in economic value during the last decades (Contreras, 2002). Seed is the basic unit of reproduction and it carries the germ of new individual. Seed is also a principal means to secure crop yields in less favorable production areas and a medium for rapid rehabilitation of agriculture in cases of natural disaster (Feistrizer, 1975). Seeds as reproductive units are expected to produce healthy plants in the field. High quality seeds play a pivotal role in increasing productivity of all other agricultural inputs (Economic Review of Agriculture, 2006).

Good quality seed is important because seed is the first link in the food production chain. Seed quality refers to the genetic, physical, physiological and sanitary status of the seed. Plant characteristics that result from genetic constitution of the embryo determine the genetic quality. Physical seed quality refers to the percentage of other seed species, impurities and undamaged seed of the desired variety of a specific crop in a seed lot. Physiological seed quality refers to the germination capacity, viability, characteristics related to dormancy and

vigor of the seed. Sanitary seed quality refers primarily to the presence or absence of disease causing organisms such as fungi, bacteria, viruses, and animal pests (Hampton, 2000).

Kenya produces 0.27 million tones of wheat per year which is lower than the annual consumption of 0.54 million tones (Impact on New Farming Systems, 2003). The shortfall in wheat production is met by imports. The Ministry of Agriculture and KARI release new varieties, and it is a common practice for farmers to buy recommended seed varieties suited for their agro- ecological zone (Impact on New Farming Systems, 2003).

International Seed Testing Association (ISTA) has the mandate of standardizing and publishing rules and methods of seed testing that should be used with uniformity in different countries. Membership is composed of participating countries throughout the world. Kenya is a member country of ISTA, thus Kenya Plant Health Inspectorate Services (KEPHIS) does seed testing as per ISTA rules and methods. In this study physiological seed quality aspect of standard germination, vigor and tetrazolium tests were carried out in the laboratory. Performance in the laboratory of wheat seed lots were correlated with seedling performance in the field in emergence and seedling growth rate.

Materials and Methods

Experimental site and materials

The research was carried out at Chepkoilel Campus of Moi University, 10 Km North of Eldoret town. The institution is situated in Uasin Gishu district within Rift Valley Province of Kenya at latitude 0^0 30N, and longitude 35^0 15 E at 2180 M above sea level. The area is within Uasin Gishu plateau, which is in agro ecological zone Lower Highland 3 (AEZ-LH 3) where wheat is grown among other crops (Jaetzold and Schmidt, 1982). Seed samples of wheat were obtained from Kenya Plant Health Inspectorate Services (KEPHIS), Lanet, Nakuru.

Laboratory tests

The laboratory tests included standard germination, seedling evaluation, Tetrazolium tests for viability, speed of germination, and electrical conductivity test for vigour according to procedures laid down by ISTA (2004) rules as follows. Field seedling emergence, seedling growth rate and speed of germination were obtained. Laboratory germination was correlated with field emergence, and laboratory seedling growth rate was correlated with field seedling growth rate.

Standard germination test

Four hundred seeds in replicates of 100 seeds of wheat were taken at random from the well mixed pure seed. Replicates were divided into sub-replicates of 50 or 25 seeds depending on the size. Seeds of wheat were planted on moistened paper substrates. The seeds were germinated between two layers of papers placed in transparent petri dishes. Evaporation was minimized by placement of a tight fitting lid. The relative humidity of 70 %, temperature of 25 ^oC and light of 700 lux were maintained in the cabinet (LMS cooled incubators, Jencons-PLS). The number of seeds that germinated was recorded daily after first day of germination.

Seedling evaluation

Seedlings were evaluated when all essential structures could be accurately assessed. Abnormal seedlings were left on the substrate until the final count (ISTA, 2004). Seedling evaluation was carried out on each crop as specified by ISTA (2004) in Table1. Seedlings were classified as either normal, abnormal or dead.

Tetrazolium test

A test was carried out on four replicates of 100 pure seeds drawn at random of pure seed fraction. Seeds were premoistened at 20 ^{0}C for 24 hours and stained with an aqueous solution of 2,3,5-triphenyl tetrazolium chloride (Tetrezolium salt) of pH 6.5-7.5. Concentration of 1.0% was used and then incubated for 3 hours between 35^{o}C to 40^{o}C. Seeds were evaluated and classified as viable or non viable on examination.

Germination speed coefficient

The germination speed coefficient was calculated according to Kotowski (1926) was also calculated. This is a measure of the number of seeds germinating daily, over the period it takes the seed in a seed lot to germinate. If in day one 2 seeds germinated and day two 40 seeds germinate, the germination speed coefficient is calculated by adding one out of two and forty out of two.

Seed vigour testing by electrical conductivity method

Four replicates of 50 seeds each were drawn at random from pure seed fraction directly with moisture content of between 10 % and 14% and weighed to two decimal places. Four flasks were added 250 ± 5 ml water for each sample to be tested. The flasks were covered to prevent contamination and equilibrated to 20 ± 2^{0}C for 18 to 24 hours prior to placing the seeds in water. Two control flasks/beakers with each test run were included containing only deionised water for background reading and rinsing of conductivity cell between each measurement. Each weighed replicate was placed into a flask with 250 ± 5 ml water. Each flask was gently swirled to ensure that all the seeds were completely immersed. Each flask was covered with aluminium foil before placing at 20 ± 2^{0}C for 24 hours. The conductivity of soak solution was measured after 24 hours (± 15 minutes) soak period using a Fieldlab- LF conductivity meter and LF 513 T- Electrode dip-type cell (Schott Gerate Glass Company, Mainz, Germany). The conductivity measured was expressed per gram of seed weight (µ S cm^{-1} g^{-1}) for each replicate and calculated after accounting for the background conductivity of the original water. The average of the four replicates provided the seed lot test results. Thus for each replicate, conductivity was calculated as per ISTA 2004 as follows:

$$\frac{\text{Conductivity reading } (\mu S\ cm^{-1}) - \text{background reading}}{\text{Weight (g) of replicate}} = \text{Conductivity } (\mu S\ cm^{-1} g^{-1})$$

Field Experiment

Field layout

The experimental design was Randomized Complete Block Design (RCBD) with four replications and five crops as the treatments. All the crops were planted with 200 kg of Single Super phosphate (SSP 12.0) per hectare. French bean seed lots each replicated four times were planted on 17th November 2004 on plot sizes of 3.5 m x 2.4 m size with fertilizer

rate of 0.168 kg per plot. Wheat was planted on 18th November 2004 on plot sizes of 2.4m x 1.4m with fertilizer rate of 0.067 kg P_2O_5 per plot and maize on 6 m x 5.25 m plot sizes at fertilizer rate of 0.630 kg P_2O_5 per plot. Each plot had 7 rows comprising of 5 rows and 2 guard rows. Each row had 20 seeds planted and thus contained of 100 seeds per plot.

Data collection

The number of emerged seedlings at the first leaf stage was recorded daily until no further seedling appeared. Seedling growth rate was obtained by taking plant heights weekly for seven weeks and dividing plant height by the number of days. Germination speed of coefficient was calculated by measuring the number of seeds emerging daily over the period seed takes to emerge in the field and stand establishment was also measured by taking average percentage emergence of seedlings of the replicates.

Statistical analysis

The results were subjected to Analysis of Variance for samples in the laboratory and field. Correlation and regression coefficients were used to determine the relationship between laboratory quality test results of standard germination and seedling growth rate, field emergence and seedling growth rate using computer packages of Scientific Package for Social Scientists (SPSS). Standard error bars were used to compare points at the same conditions. Laboratory germination was correlated with field seedling emergence while as laboratory was correlated with field seedling growth rate.

Results

Seed quality

There was a relationship between electrical conductivity (EC) and seed quality in terms of speed of germination, percentage of normal and abnormal seedlings, and stand establishment as shown in table 1. High EC leads to low speed of germination in both the laboratory and field. It also leads to high abnormalities, low normal seedlings and low stand establishment. Low EC leads to high speed of germination in the laboratory and field, low abnormalities, high normal seedlings and high stand establishment. In this study, results of electrical conductivity predicted seedling performance in the field better than standard germination.

Seedling Emergence

There was a significant ($p \leq 0.05$) increase in percentage of seedling emergence ($p \leq 0.05$) in the laboratory and in the field for both wheat accessions 50 and 1285 (Figure 2). Percentage seedling emergence was faster in the field than in the laboratory in both wheat accessions.

In accession 50, there was a rapid increase in percentage of seedling emergence in the laboratory between the first and third day whereas it was less rapid between the third and seventh day (Figure 2). There were no significant differences ($p \leq 0.05$) in laboratory seed germination between the third and seventh day. There was a rapid increase in percentage seedling emergence in the field between the fifth and sixth day and a slower increase between the sixth and eleventh day. There were no significant differences ($p \leq 0.05$) in field emergence between the sixth and eleventh day.

In wheat accession 1285, there was a rapid increase in percentage of seed germination in the laboratory from the first to third day and less rapid between the third and seventh day. There were no significant differences ($p \leq 0.05$) in percentage of seed germination from the third to the seventh day (Figure 2). Also there was a rapid increase in the percentage seedling emergence in the field from the fifth to the sixth day and a slower increase between the sixth and the eleventh day was noticed. There were no significant differences ($p \leq 0.05$) in field seedling emergence from the sixth to the eleventh day.

Percentage seedling emergence for wheat accession 50 increased from 47.5% on the second day to 81.8 % on the seventh day after sowing whereas field seedling emergence increased 68.5% on the sixth day to 83% on the eleventh after sowing. Percentage seedling emergence for wheat accession 1285 increased from 51% on the second day to 87.5 % on the seventh day after sowing whereas field seedling emergence increased from 80.8% on the sixth day to 90% on the eleventh after sowing (Figure 2).

		Speed of germination index		Seedling Evaluation (%)		Seedling stand (%)	
Seed Samples	Electro conductivity (μ S cm^{-1} g^{-1})	Lab	field	Normal seedlings	Abnormal seedlings	Dead seedlings	Stand establishment
Wheat 50	1.070	112.16	177.43	60	29	11	83
Wheat 1285	1.068	115.03	208.11	72	20	8	90

Table 1. Seed quality scores of wheat seed samples

Figure 1. *Wheat laboratory seedlings. Seedling is glassy (glassy wheat seedling that is typical of phytotoxicity). Seminal roots are missing, coleoptile is stunted and seeds are dead.*

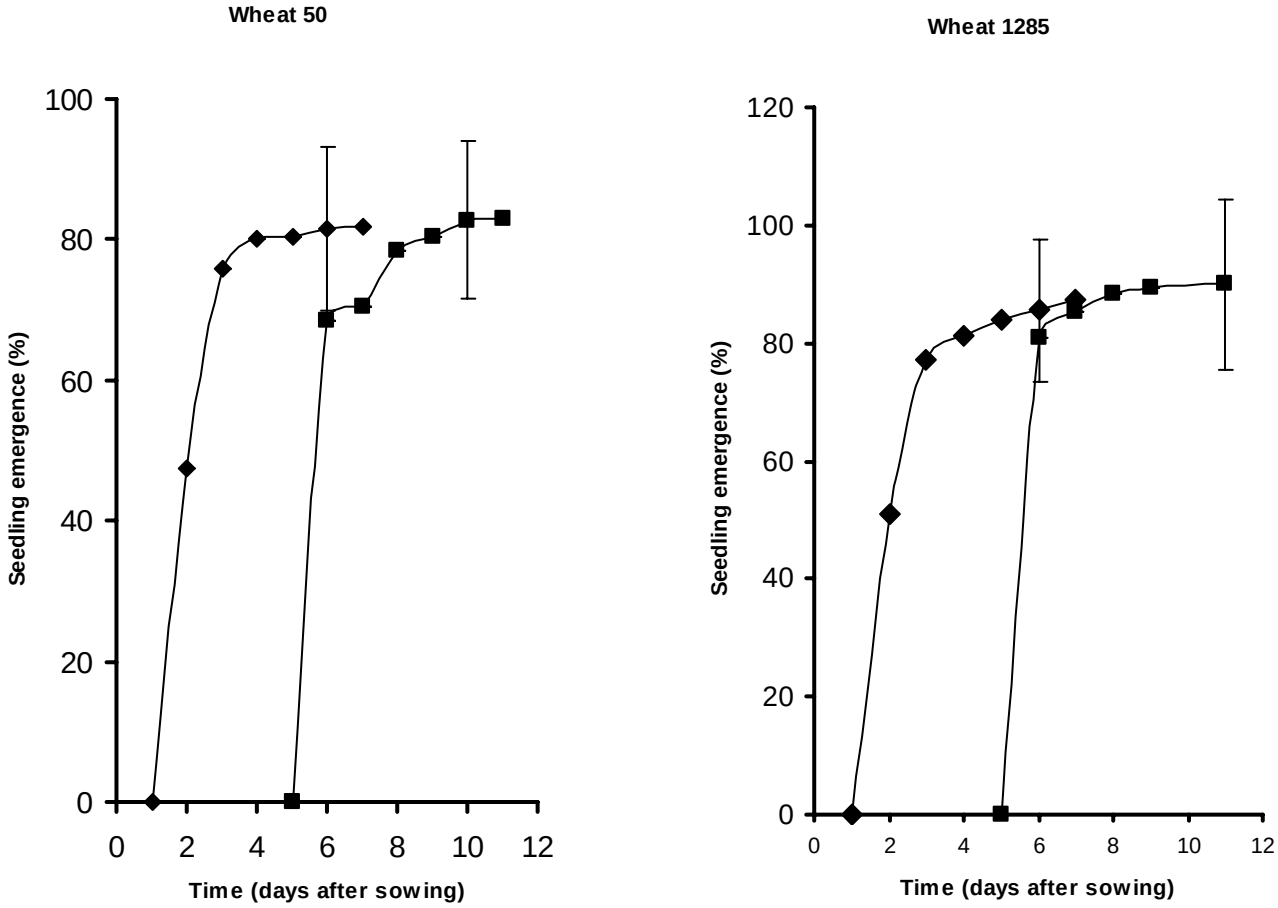


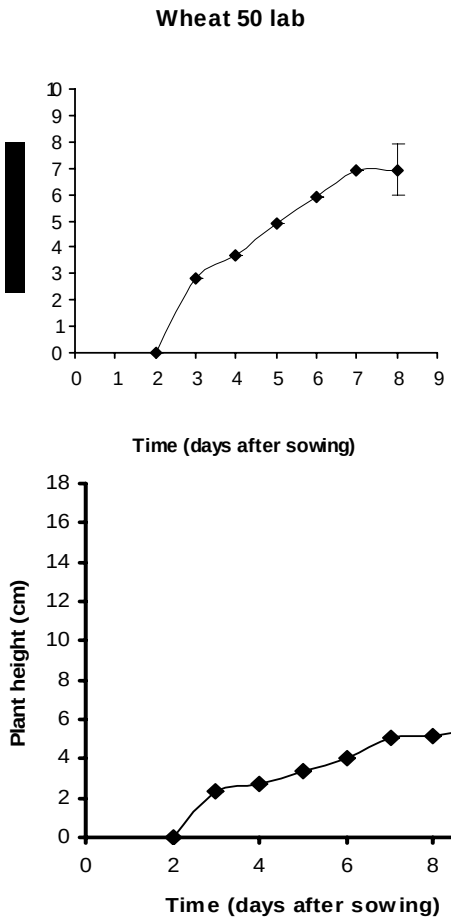
Figure 2. *Laboratory germination (% (♦) and field seedling emergence (% (■) for two wheat (50 and 1285) accessions over time (days after sowing).*

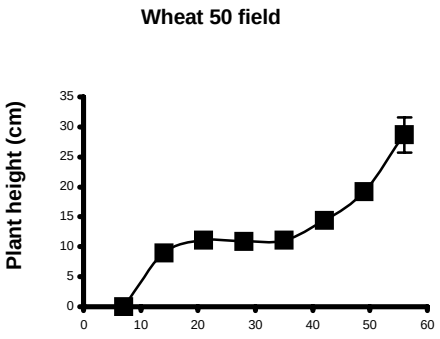
Plant height

There was a significant increase ($p \leq 0.05$) in plant height in the laboratory and in the field for both wheat accessions 50 and 1285 (Figure 3). Growth in plant height was faster in the field than in the laboratory in both wheat accessions. Growth in plant for both wheat accessions showed the same trend for laboratory and field seedling growth rate (Figure 3). Increase in height in the field was faster than in the laboratory (Figure 3).

Wheat accession 50 had a rapid growth in plant height in the laboratory from 2.8 cm on the third to 6.9 cm on the seventh day and no difference in growth between the seventh and eighth day (Figure 3). There were no significant differences ($p \leq 0.05$) in growth in height for plants germinated in the laboratory between the seventh and eighth day (Figure 3). There was a rapid growth in plants grown in the field in height between the fourteenth and twenty-first day followed by slow growth in plant height between the twenty-first and thirty-fifth day and the rapid growth from the thirty- fifth day to the fifty sixth day. There were significant differences ($p \leq 0.05$)in plants grown in the field in height between the fourteenth to forty-ninth day and fifty-sixth day (Figure 3).

In wheat accession 1285, there was a rapid growth in laboratory plant height from the third to the seventh day, followed by slow growth in height between the seventh and eighth day then rapid growth between the seventh day and eighth day (Figure 3). There were no significant differences ($p \leq 0.05$) in growth of plants germinated in the laboratory in height from the seventh to eighth day in the field, there was a rapid growth in height between the fourteenth and forty second day, followed by a slow plant growth rate between the forty second and forty sixth day and then more rapid growth from the forty sixth to the fifty sixth day (Figure 3). There were significant ($p \leq 0.05$) differences in plants germinated in the field between fourteenth to forty sixth and the fifth sixth day (Figure 3).





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Discussion

Seedling performance in the field in both percent emergence and seedling growth rate was higher than in the laboratory. It was confirmed in that the relationship between laboratory tests and field emergence is not conclusive, and that use of laboratory tests to predict field emergence is variable and strongly dependent on the field environment (Edje and Burris, 1971; Johnson and Wax, 1978; Ludders and Burris, 1979; Kulik and Yaklich, 1982; Duczmal and Minicka, 1989 and Egli and TeKrony, 1995).

Determining the cause of individual abnormal seedlings without a thorough knowledge of the "history" of a seed lot is not possible. However certain types of abnormal seedlings in a test may give valuable hints as to germination conditions or as to handling of the seed. Thickened and shortened roots or coleoptiles of cereals as was seen in this study are usually

the result of over treatment with fungicides and insecticides or treatment at high seed moisture content (ISTA, 2003). This portrays characteristics of over treatment with fungicides (www.ag.ohio.state.edu/~seedsci/svv01.htm). These results were consistent with the reports that application of seed treatment above labelled rates can injure germinating seed and plant stands, possibly reducing yields to less than those produced by untreated seed (Ashley, 2003). The standard germination was done on paper for small seeds while in the field soil was used. Soil is generally not recommended as a primary growing medium. However, it may be used as an alternative to organic growing media when seedlings show phytotoxic symptoms or if the evaluation of seedlings is in doubt on paper or sand (ISTA 2006). Laboratory germination was lower than seedling performance in the field for wheat due to sensitivity of the crop to seed dressing chemicals.

Laboratory seedling growth rate was lower than field seedling growth rate in both wheat accessions. Seedling growth could differentiate seed quality among commercially acceptable perennial rye grass (*Lolium perenne* L.) seed lots (Happ and Danneberger, 1993).

Wheat accessions 150 and 1285 had correlation coefficients (r) for emergence of 0.843* and 0.97**, respectively, between percentage seedling emergence in the laboratory and field. Correlation for accession 50 was significant ($p \leq 0.05$) while accession 1285 was highly significant ($P \leq 0.01$). This shows that percentage seedling emergence in the laboratory could be used to predict percentage seedling emergence in the field. In another study, correlation coefficient (r) between standard germination and field emergence of seed lots of Sudan grass of 0.721** and 0.414 was obtained while in 1999, correlation of 0.780* for Siberian rye grass was also obtained (Wang *et al.*, 2004).

Krishnasamy and Ramaswamy (1987) found that electrical conductivity of sorghum seed leachate correlated negatively with field emergence, standard germination and vigor index. Garcia and Lasa (1991) used electrical conductivity test to predict field emergence of grain sorghum. They found significant difference for seed leaching among genotypes but the correlation with field emergence was not significant.

Wheat accessions 50 and1285 had correlation coefficient for seedling growth rate (r) and 0.70 and 0.83*, respectively, between growth of plants in the laboratory and field. This shows that seedling growth rate in the field can be predicted by seedling growth rate in the laboratory for wheat.

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