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Assessment of Germination Parameters, Accelerated Ageing and Conductivity Tests on Seeds of Upland Rice (*Oryza sativa* L.)

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ABSTRACT

The study was to determine the germination percentages of upland rice and to investigate the possible effects of accelerated ageing upon upland rice varieties. Seed of seven upland rice varieties were subjected to accelerated ageing treatment for 72 hours at 43^{0}C and 100% relative humidity at Seed Science laboratory of Obafemi Awolowo University, Ile Ife, Osun State, Nigeria. The experiment was laid out in a randomized complete block design with four replicates. Observations were made on germination %, germination index, germination rate index of both aged and unaged rice seeds, conductivity test and growth analysis test. The data were subjected to Analysis of Variance (ANOVA) and means were separated using LSD 5% level of probability, mean comparison and mean ranking were done among the varieties. Correlation analysis was also done. Accelerated ageing percentage showed significant positive correlation with seedling traits (fresh weight, dry weight,). It was discovered that germination % of a variety reduced drastically at 72 hours of ageing. In addition conductivity test had significant negative correlations with fresh weight, and significantly positive correlated with dry weight. Accelerated ageing and conductivity test should be incorporated into commercial seed testing rather than the usual use of germination test.

Key words: *Rice, Germination percentage, Accelerated ageing, Conductivity, Correlation.*

INTRODUCTION

Rice (*Oryza sativa* L.) belongs to the grass family *Poacea*e, and is of the genus *Oryza* in the tribe *oryzae. Oryza sativa* is one of the most important cereals in the world, with a global production of 590 million tons[6]. Rain-fed upland rice is a major crop of importance in sub-Saharan Africa (SSA) where it accounts for over 40% of the land under rice cultivation[15]. It has increasingly become popular amongst first time rice growers in non-traditional rice producing regions of SSA like Nigeria.

The rice seed consists of the true fruit (brown rice) and the hull, which encloses the brown rice. Brown rice consists mainly of the embryo and endosperm.

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The surface contains several thin layers of differentiated tissues that enclose the embryo and endosperm. Germination and seedling development start when seed dormancy has been broken and the seed absorbs adequate water and is exposed to a temperature ranging from about 10 to 40°C. Physiologically, germination is defined as the time when the radicle or coleoptile (embryonic shoot) emerges from the ruptured seed coat. Under aerobic conditions the seminal root is the first to emerge through the coleorhiza from the embryo, and this is followed by the coleoptile. Under anaerobic conditions, however, the coleoptile is the first to emerge, with the roots developing when the coleoptile has reached the aerated regions of the environment. The production of rice at different ecologies depends on vigour, rate of seedling emergence, uniformity and crop establishment to bring out the potential yield based on variety[9].

During prolonged seed storage, Deterioration of seed and loss of germination sets in. This has been explained by so many theories of seed ageing. Abiotic factors such as fungi attack and biotic factors like loss of vitamins or hormones, degradation of nucleic acids, proteins or membranes[11]. According to McDonald[10], relative humidity of the air which control the seed moisture content and temperature are the two most important environmental factors that is influencing the rate of deteriorative process in seed ageing. According to Abdul-Baki, 1980, high temperature, ambient relative humidity and seed moisture content are the main factors influencing seed storage capability. The process of deterioration under accelerated ageing and normal condition are similar, but the only difference is that the rate of deterioration for accelerated ageing is faster thereby making it easier to predict[4,8]. In order to understand the mechanism of ageing and associated deterioration processes of seed, accelerated ageing techniques have great potential[10].

Research has shown that the use of germination percentage overestimates field emergence, hence, there is need to explore other avenues in determining field emergence. Therefore, the objectives of this study was to determine the germination percentages of upland rice and to investigate the possible effects of accelerated ageing upon upland rice varieties.

MATERIAL AND METHODS

Seeds of seven upland rice varieties FARO55, FARO58, FARO59, FARO63, FARO64, FARO65 and NERICA 2 were used for two studies. The seeds were obtained from the Africa Rice Centre substation at the International Institute of Tropical Agriculture (IITA), Ibadan, Nigeria. The studies were conducted in the Seed Science Laboratory of Obafemi Awolowo University (O.A.U), Ile-Ife, Osun State, Nigeria. Laboratory evaluation included germination test, accelerated ageing test, and bulk conductivity test. In each case, the experiment was laid out in a randomized complete block design. Germination Test***:*** Fifty seeds were planted in a bucket filled with sterilized riverbed sand in four replicates for each variety. Germination counts were taken from 5 to 10 days after planting (DAP) and were used to calculate germination percentage (G %), germination index (GI), and germination rate index (GRI)

$$G\% = \frac{\text{(Number of seedlings emerged 10 DAP)}}{\text{Total number of seeds planted}} \times 100$$

$$GI = \frac{\sum (Nx)\,(DAP)}{\text{Number of Seeding that emerged 10 DAP}}$$

Where Nx = number of seedling that emerge on day x after planting

$$GRI = \frac{\text{Germination index}}{\text{Germination percent}}$$

At 10 DAP, ten seedlings were collected from different standard germination test trays and seedling analysis was conducted to obtain the following data; mesocotyl length, coleoptile length, shoot length, root length, primary leaf length, fresh root weight, fresh shoot weight, dry root weight and dry shoot weight. Accelerated Ageing***:*** Forty millilitre of distilled water was poured inside each ageing box and wire meshes were suspended in it. Fifty seeds were placed on a mesh after initial weight had been taken and the boxes were covered. The ageing boxes were properly labelled and kept inside the Accelerated Ageing chamber at 43^0C for 72 hours. At the end of the ageing period, the accelerated ageing boxes were removed and cooled at room temperature. The seeds were subjected to germination test. Germination counts were taken at 5 to 10 DAP and these were used to calculate G%, GI, and GRI as described earlier. Conductivity Test: Fifty Seeds of all the varieties were soaked for 24 hours in 100 ml distilled water in four replicates in beakers, each covered with aluminium foil paper to prevent contamination. The beakers, containing water and seeds, were kept under ambient temperature. After 24 hours of soaking, the quantity of the leachate and electrical conductivity were determined with a conductivity meter. The results were expressed in $\mu Scm^{-1}g^{-1}$ of seeds, in accordance with the methodology described by Vieira and Krzyzanowski[14]. The data collected were subjected to Analysis of Variance (ANOVA) and means were separated using LSD 5% level of probability, mean comparison and mean ranking were done among the varieties. Correlation analysis was also done.

RESULTS

Mean comparison of the seven upland rice varieties were significantly different from each other's considering the LSDs for germination and vigour tests (accelerated ageing and bulk conductivity). For germination percentage, FARO 58 is not significantly different from FARO59, FARO55, FARO65, and NERICA2, but it is significantly different from FARO63 and FARO 64 respectively. In-case of accelerated ageing percentage, FARO58 is not significantly different from FARO 55 but significantly different from other varieties. Reverse is the case for bulk conductivity test for FARO 58 which is significantly different from all other varieties. The coefficients of determination (r^2) values ranged from 19.51% to 75.93%. The CVs associated with the vigour tests (accelerated ageing and bulk conductivity) ranged from 43.19 % to 60.22%. The model used for the analysis accounted for over 60% variation for accelerated ageing percentage (Table 1).

Ranking the means of germination percentage and germination rate index, FARO58 was the best among the seven varieties while Nerica 2 was the poorest, in accelerated ageing percentage ranking of means, FARO65 was the best and FARO55 was the poorest. Considering the ranking summation index of germination percentage, germination rate index and accelerated ageing percentage, FARO 58 emerged as the best followed by FARO63. FARO 64 and FARO65 performed the same way followed by FARO 59 .FARO 55 remain the poorest among the varieties (Table 2).

Accelerated ageing percentage correlated positively significantly with seedling fresh weight and seedling dry weight,. Accelerated aging rate index was negatively correlated with seedling dry weight. Conductivity test was negatively correlated with seedling fresh weight but positively correlated with seedling dry weight (Table 3). Result shows that germination rate index is negatively correlated with shoot length (Table 4).

Table 1: Mean Comparison of germination, accelerated ageing and conductivity tests conducted in the Seed Science Laboratory of Obafemi Awolowo University, 2015

VARIETY	G%	GRI	AAPCT	AARI	COND
Nerica 2	83.5	6.8	90.0	6.08	1.29
FARO 64	98.0	5.6	81.0	8.49	1.11
FARO 65	89.5	6.2	100.0	4.99	1.27
FARO 55	88.0	6.6	49.0	12.72	1.25
FARO 58	99.0	5.3	85.5	9.983	0.98
FARO 59	93.0	5.5	78.5	6.83	1.27
FARO 63	93.5	5.5	86.5	7.54	1.24
$LSD_{0.05}$	5.96	0.71	23.60	7.06	0.38
R^2%	75.93	70.78	19.51	60.42	21.11
CV%	4.36	8.00	60.22	31.72	43.19

G%=Germination percent , GI=Germination index, GRI=Germination rate index, AAPCT=Accelerated ageing Percentage, AAI=Accelerated ageing index, AARI= Accelerated ageing rate index, R^2=Coefficient of determination, CV= Coefficient of variation, LSD=Least significant difference.

Table 2: Ranking of means of seven upland rice varieties for vigour test traits obtained from test conducted in the Seed Science Laboratory of Obafemi Awolowo University, 2015.

VARIETIES	G%	GRI	AAPCT	RSI
Nerica 2	7	7	2	16
FARO64	2	4	5	11
FARO65	5	5	1	11
FARO 55	6	6	7	19
FARO 58	1	1	4	6
FARO 59	4	2	6	12
FARO 63	3	3	3	9

Ranking was obtained as the best=1 and the poorest=7, G%=Germination percentage, GRI= Germination rate index, AAPCT=Accelerated ageing Percentage and Ranking summation index

Table 3: Correlation coefficients between laboratory seed vigour traits at Ife 2015

Seed Vigour	FW	DW
GPCT	0.42	0.47
GI	0.25	-0.18
GRI	0.22	-0.27
AAPCT	0.82**	0.79*
AAI	-0.11	-0.10
AARI	-0.72	-0.76*
COND	-0.83*	0.85*

Table 4: Correlation coefficients among seed vigour traits of seven upland rice varieties evaluated at Seed Science laboratory Obafemi Awolowo University, 2015.

	GRI	AAPCT	SL
GRI	1	0.21	-0.75*
AAPCT		1.00	-0.25
SL			1.00

* indicate significance at 0.05 level of probability.

GRI=Germination Rate Index, AAPCT=Accelerated Aging Percentage, SL=Shoot length

DISCUSSION

Result from this study showed variation among the rice cultivars in all parameters studied. This could be due to genetic makeup of the varieties used. There was significant difference observed among the varieties. The variety that has the highest germination percentage does not have the highest percentage when the seed were passed through accelerated ageing; this implies that normal germination percentage cannot predict the performance of the seed when stored for a period before planting this corroborate with the work of Fawad *et al.*[7] who found out that standard germination tests were conducted under an optimal set of temperature and moisture condition which allowed for optimum seed germination with minimal stress. . Variety that performed well under accelerated ageing will definitely give higher germination percentage when planted on the field. Accelerated ageing percentage can predict the field emergence. Similar result was obtained from Suraj Chhetri[13]where ageing condition of 44^0C for 72hrs was suitable and perfect combination which predicted field emergence in rice seed as seed vigour test and recommended for common Thai rice varieties. This study showed differences among the varieties for bulk conductivity. The integrity of the seed coat and the cell membrane, which is determined by deteriorative biochemical changes or physical disruption, can be considered the fundamental cause of differences in seed vigour which are indirectly determined as electrolyte leakage during the conductivity test [3]. Maximum and minimum electrical conductivity of seed leachate was observed in Nerica 2 and FARO 58. This is in line with findings from Abdul-Baki and Anderson[1] who reported that leaching of materials from seeds was passive diffusion and not caused by changes in membrane permeability. Leaching is related to metabolic activity of seeds. Deteriorating seeds would operate at low level of metabolic activity and materials could be leached out. But in seeds with high level of metabolic activity, materials would be utilized rather than getting leached. Thus, high vigour seeds will have high metabolic activity and will contribute to the less leaching of electrolytes. Deteriorating seeds would operate at low level of metabolic activity and materials could be leached out. Accelerated ageing percentage in this studies showed significant positive correlation with seedling trait (fresh weight, dry weight,) which corroborate with Bastians *et al.* [5] that early vigour scores of rice cultivars under field conditions were consistent with the total plant dry weight and leaf area of plant. Result of conductivity test in this study showed that Conductivity test had significant negative correlation with fresh weight and significantly positively correlated with dry weight Ratajezak and Duczmal[12] found out that the electrical conductivity test was the best correlated with field emergence. Considering the result from this study, upland rice varieties seeds that seeds that performed well under accelerated ageing percentage and bulk conductivity test could best predict the field emergence than normal germination test Wilson *et al* [16] discovered that accelerated ageing test and electrical conductivity test could be used in emergence prediction , hence accelerated ageing and conductivity test should be incorporated into commercial seed testing rather than the usual use of germination test.

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