Original Research Article

PHYSIOLOGICAL SEED QUALITY RESPONSES OF THREE RICE VARIETIES TO DIFFERENT STORAGE MATERIALS UNDER AMBIENT CONDITIONS

ABSTRACT

Rice is an important staple crop which provides essential dietary nutrients for the well being of humans but the seeds lose viability rapidly in storage before the next planting season. This study was conducted at the seed and pathology laboratories in CSIR-Crops Research Institute (Fumesua) to determine the physiological seed quality responses of three rice varieties to different storage materials under ambient conditions. The experiment was set up in 3x4 factorial arranged in a completely randomized design with three replications under ambient conditions. Factor one was variety of rice at three levels (Agra, Amankwatia and Jasmine) and factor two was packaging materials at four levels (plastic bucket, single-layered nylon sack, un-layered nylon sack and double-layered nylon sack). Moisture content, vigour, vigour index, germination percentage and health test were the major parameters determined before and after the experiment. Results indicated that all the varieties stored in the un–layered packaging materials recorded the highest moisture content of 13.2%. Rice seeds in single-layer nylon sack had the best germination percentage (98%), seed vigour (48.04) and vigour index (20689) whereas seeds stored in un-layered nylon sack had the least germination percentage (84%), vigour (37), and vigour index (13193). Un-layered packaging materials recorded the highest population of *Sitophilus oryzae* while plastic Bucket storage material recorded the least insect infestation. Twelve fungal pathogens were identified in total, of which nine were pathogenic and three saprophytic. The study concluded that to ensure good quality of rice seeds, rice seeds should be stored in single layered packaging materials for three month under ambient condition.

Keywords: Staple, malnutrition, hunger, hygroscopic, infestation and deterioration.

1. INTRODUCTION

Rice (*Orya sativa* L.) is grown in >100 countries with 90% of the total global production from Asia[1]. Rice, as a staple food, is splendidly incomparable because it feeds more people (>3.0 billion) than any other crop and grows in wet environments that do not support the sustenance of other crops. Wet environments located abundantly across Asia may be one reason that makes rice the most popular staple food in countries such as Japan, China, Philippines, Thailand, Indonesia, Vietnam, Myanmar, India, and Bangladesh. Japanese people and people from other Asian countries obtain >75% of their daily calorie intake through consumption of polished WR, which is visually more appealing, and discard rice bran [2]. FAO[3] posited that, rice contains 275 of energy and 20% alimentary protein. The germ and the husk which is eliminated during milling are rich in vitamins–especially vitamin B1, minerals, fiber and enzymes. Besides the one third production input by local rice farmers in Ghana, government spends about 500 million dollars to import the remaining two thirds to meet the country's requirement annually [4]. This situation which is highly unacceptable, has arisen due to the low yields recorded by the local farmers. In explaining this adverse situation, Asante *et al.,* [5] contended that, in Ghana the purity and quality of seeds are compromised due to the storage practices employed by the farmers. Substantial amount of seeds stored become unviable when planted in the next season. Good quality seed has however been reported to increase yield by 5-20% [6] and that good crop establishment is directly linked to the quality of seed used [7]. Yet in Ghana, farmers store their seeds, irrespective of the source, under ambient conditions and in different storage materials. Some of the commonly used storage materials are jute bags, polyethylene bags, clay pots, and nylon fertilizer bags. Such poorly stored seeds result in poor seed quality leading to poor stand establishment, low seedling vigor and low grain yield [8]. For good and quality storage of seeds, the storage material used is also considered as one of the most vital factors influencing viability and longevity of seeds [9]. There aresome types of storage material which may accelerate the exchange of energy and mass between the stored seeds and the storage environment [10] advancing the rate of deterioration, reduction in vigor and loss of viability of the stored seeds [11;12]. In most developing countries, research information on the successful storage of rice seeds in these materials is scanty and hence the need for this study. The overarching objectives of the study was to determine the effect of different storage materials on seed physiological and health qualitycharacteristics of stored rice seeds.

2. MATERIALS AND METHODS

2.1 STUDY AREA

The laboratory experiment was conducted at the CSIR-CRI Seed Science and Pathology Laboratories.

2.2SOURCE OF RICE SEEDS AND STORAGE MATERIALS

Twelve kilograms each of three rice varieties (AGRA Rice, Amankwatia and Jasmine 85) were collected from farmers one month after harvest in September, 2017. A total of K. 36kg of rice seeds were obtained and kept in brown envelope labeled sealed and transported to Fumesua CSIR-CRI. All varieties were dried to a moisture content of 12%. The quality of the seed was analyzed at the crops Research Institute seed science laboratory. 500g of seed sample of each variety was weighed and stored in four different packaging materials each measuring 30cmx 30cm. These storage containers were Double layer in nylon sack with thickness of 0.13mm, Single layer in nylon sack with thickness 0.25mm, No layer nylon sack with thickness of 0.1 mm and Plastic buckets 2.03 mm.

Jasmine 85: It is an indica rice which has yield potential of 4-6-5 tones/hectare and matures in 120-130days. It has short round seeds.

Amankwatia: It is an indica rice whichhas yield potential of 2.75 tons/hectare and mature 110-120 days it has medium seed length.

AGRA rice: It is an indica rice whichhas yield potential of 8 tons/hectare and mature between 120-130 days. It has medium seed length.

2.3 EXPERIMENTAL PROCEDURE

The seed storage experiment was set up in 3x4 factorial arrangement in a completely randomized design. The first factor was varieties at three levels (AGRA Rice, Amankwatia and Jasmine 85). The second factor was packaging materials at four levels (Un-layered in nylon sack, Single layer in nylon sack, Double layer in nylon sack, and Plastic buckets. The experiment was replicated three times under ambient conditions.

2.4 DATA COLLECTION

Storage humidity and temperature readings were taking daily for the storage room using Lascar (Microdaq humidity and temperature Data Logger).

2.4.1 Ambient and relative humidity of storage environment

The storage temperature ranged between 29.3° C and 28.7 °C while the relative humidity ranged between 68% and 56% Minimum and maximum temperature and relative humidity of storage environment (Table 1).

Table1: Storage Temperature and Relative Humidity

Month	Min Temp (^{o}C)	Min RH (%)	Max Temp (^{o}C)	Max RH
January	29.3	56	32.8	58
February	28.8	68	34.1	70

March	29.1	57	33.2	59
April	28.7	67	33.8	67

2.4.1 Moisture Content

The seeds moisture content was taken before, monthly and after storage using the moisture digital grain moisture meter. Seeds were put in to the moisture meter and pressed and reading were taken three times consistently and the average taken as the moisture content of the rice samples.

2.4.2 Vigour Test

Vigour test was conducted using the first count method. The test was conducted alongside with the normal germination test. The number of normal seedlings, germinated on the first count day, as indicated in the germination test for each species are counted. The number of normal seedlings gives an indication of the vigour level in the sample. Higher number of normal seedling the better the seed vigour. Vigour was measured as the percentage of the seeds that had germinated by the 5th day of incubation [13]. Data gathered were averaged for each sample.

The vigour index was calculated using the formula of Abdul-Baki and Anderson (1973) as follows:

Vigour Index = (Shoot length + Root length) x Germination Percentage

2.4.3 Seed Health

The seed health test was done using the Blotter method procedures of Mathur and Kongsdal[14].

2.4.4 Germination Test

The germination test was performed using the procedures of ISTA [15].The test was conducted using one hundred seeds. Twentyfive seeds of each sample were placed in four 9cm petri dish lined with three pieces of filter paper soaked in distilled water. Germinated seeds were counted and recorded at 4 day and 7days after sowing. After the 7 days, seedlings were classified in normal and abnormal seedlings percentage germination was calculated as

$$\% \text{ Germination} = \frac{\text{Number of Normal seedlings} \times 100}{\text{Total number of seed.}}$$

2.5 DATA ANALYSIS

Data collected were however subjected to Analysis of Variance (ANOVA) using Statistix software version 10. Significant differences between treatments means were set at 0.01 probability level using the Turkey’s Honestly Significant Difference (HSD) test.

3. RESULTS

3.1 PHYSIOLOGICAL SEED QUALITY CHARACTERISTICS OF RICE SEEDS AS INFLUENCED BY PACKAGING MATERIALS

3.1.1 Initial seed quality of the three rice varieties

There were no significant difference in germination, vigour and vigour index (Table 2). However, germination percentage ranged between 99.3% and 100% vigour ranged between 80 and 84.33 and vigour index ranged between 17469.00 and 19420.00.

Table 2: Initial seed quality of the three rice varieties

Variety	Germination	Vigour	Vigour Index
Jasmine	99.33[a]	84.33[a]	19420.00[a]
Amankwatia	100.00[a]	82.67[a]	19126.00[a]
AGRA	100.00[a]	80.00[a]	17469.00[a]
HSD (0.01)	2.4407	20.126	3257.2

3.1.2 Germination Percentage

There were no significant packaging*variety for germination percentage (Table 2). The germination percentage ranged from 81% to 99.67%. However, across the packaging materials, there were significant differences in the germination percentage of stored rice seeds. (Table 3). Germination percentage was significantly highest in rice seeds stored in single–lined packaging material and the least was recorded by rice seeds stored in unlined-nylon sack and which was similar to double-layered nylon sack.

Table 3: Effect of Packaging materials on the Germination Percentage of Stored Rice Seeds

Packaging materials	Germination Percentage (%)
Single-layered	98.15[a]
Double-layered	94.07[b]
Bucket	93.03[b]
No layer	84.11[c]

HSD (0.01)	3.954

3.1.3 Vigour

There were no significant packaging material*variety interaction for vigour. The vigour ranged from 38.333 to 52.333. However, across the packaging materials there were significant differences in the vigour of stored rice seeds (Table 4) Significantly highest vigour was recorded by rice seed stored in single-layered packaging material and the least was recorded in un- lined nylon sack which was similar to double–layered nylon sack.

Table 4: Effect of Packaging Materials on the Vigour of Stored Rice Seeds

Packaging materials	Vigour
Single-layered	48.04^{a}
Double layered	44.70^{a}
Bucket	44.37^{a}
No layer	37.26^{b}
HSD (0.01)	5.681

3.1.4 Vigour Index of Stored Rice Seeds

There were no significant packaging material*variety interaction for vigour index. The vigour index ranged from 13186 to 21755. However, across the packaging materials, there were significant differences in vigour index of stored rice seeds (Table 5). Significantly highest vigour index was recorded by rice seeds stored in single layered nylon sack and the least was those stored in unlined nylon sack which was similar to double-layered packaging materials.

Table 5: Effect of Packaging Materials on the Vigour Index of Stored Rice Seeds

Packaging materials	Vigour Index
Single-layered	20689^{a}
Double-layered	16791^{b}
Bucket	16698^{b}

No layer	13193[c]
HSD (0.01)	**2313.0**

3.1.5 Moisture Content of Stored Seeds

There were no significant packaging material*variety interaction for moisture content. The moisture content ranged from 10.8% to 13.2%. However, across the packaging materials there were significant differences in the moisture content of stored rice seeds (Table 6). Significantly highest moisture content was recorded by rice seeds stored in un-line nylon sack and the least was recorded by single layered nylon sack which was similar to seeds stored in plastic bucket.

Table 6: Effect of Packaging Material on Moisture Content of Stored Rice Seeds

Packaging materials	Moisture Content
Single-layered	11.39[b]
Double layered	11.61[b]
Bucket	11.63[b]
No layer	12.72[a]
HSD (0.01)	0.66

3.2 EFFECT OF PACKAGING ON SEED HEALTH CHARACTERISTICS OF STORED RICE SEEDS

3.2.1 Insect Infestation of Stored Rice Seeds

There were no significant package x varietyinteractions for insect infestation. The insect populations ranged from 0.00 to 13.00. However, across the packaging materials there were significant differences in the insect infestation of the stored rice seeds (Table 7). Insect infestation significantly was highest in rice seeds stored in un-lined nylon sack whiles the least was stored in double-layered nylon sack. The infestation in the double lined material was similar to that of the plastic bucket.

Table 7: Effect of Packaging Materials on the Insect Infestation of Stored Rice Seeds

Packaging materials	**Insect infestation**
Single-layered	1.9[b]
Double-layered	1.7[b]

Bucket	2.9^{b}
No layered	9.0^{a}
HSD (0.01)	4.76

3.2.2 Microbial Infection of Stored Rice Seeds

A total of twelve fungi were found out of which nine were pathogenic. These were *Alternaria* spp., *Bipolaris* spp., *Curvularia lunata, Phoma* spp., *Nigrospora* spp., *Fusarium moniliforme, fusarium pallidoroseum,Tricoderma* spp. *and Myrothecium* spp. *C*urvularia *lunata* recorded the highest percent incidence (12%) whiles *Nigrospora* recorded the least (5.0%). The saprophyte fungi were *Rhizopus* spp., *Aspergillus flavus and Penicillium* spp. (Table 8).

Table 8: Percentage of occurrence of pathogenic fungi identified during storage rice seed

Pathogen	**Percentage of occurrence (%)**
Curvularia lunata	12
Phoma spp.	7. 2
Bipolari spp.	5.2
Phoma spp.	4.8
Myrothecium spp.	3.7
Fusarium moniliforme	3.2
Fusarium pallidoroseum	3.2
Tricoderma spp.	3.0
Nigrospora spp.	0.5

4. DISCUSSION

Moisture content increased in rice seeds stored in un-lined nylon sacks. This may be attributed to the porous nature of packaging material which permit seeds to absorb moisture from the atmosphere. This is in line with Rai *et al* [16] who pointed out that, to minimize moisture absorption in seeds from the environment due to hygroscopic nature of seeds it is better to store seeds in moisture proof storage material such as aluminum foil, polyethylene bags, or any sealed container to preserve the seeds for a longer time.Rice seeds stored in unlined nylon sack also recorded the lowest germination percentage and vigour after the storage period. This could be as a result of the storage material that, permit moisture absorption by the seeds that made conducive condition for the decline in seed quality which lead to poor germination percentage. Single layered packaging material had the highest germination percentage, since it was lined it did not makeit porous as compared to the un-lined packaging materials and hence did not create conducive environment for seed deterioration leading to germination loss. Rai *et al* 16] reported similar work, which indicated that seeds stored in impervious sealed containers stored well compared to porous storage materials.Seed vigour is directly proportional to seed viability and that a decline in seed vigour leads to a loss in seed germination [17] The results of the present confirmstheir findings.

Sitophilus oryzae (rice weevil), were the insects found   after the storage duration. Among the four packaging materials, those un–lined were highly susceptible to insect infestation probably due to their mode of feeding or action which is characterized mouth parts snout or rostrum used to for boring holes and laying eggs. According to de Sousa *et al*., [18] rice weevil are one of the insects that can chew holes in to packaging materials are referred to as penetrators. The rice weevil is one of the most widespread and destructive insect pests found in stored cereals worldwide, and the interaction with rice involves all life stages of the insect, with the larvae being the most destructive stage. These insects cause rice losses and affect their quantity and quality [19]. The results of the present study confirm their findings about the rapid manner in which seeds were destroyed. The results of the present study indicated that out of the pathogenic fungi *Curvularia lunata* was the highest and among the saprophytic fungi Rhizopus ssp was the highest. According to Agarwal and Sinclair 1996[20] these fungi are termed "storage fungi" that may be present in the deterioration of seeds at storage. Agarwal [21] pointed out that, seed–borne microflora living in association with seeds did not cause disease automatically. Agrios [22] reported that, for a disease to occur, the three major components of disease cycle (host, pathogen and environment) must interact. When any of the three components is absent, disease may not occur. Agrios, [22] further indicated that, when any of the component changed it affected the level of disease severity within a particular host Though the pathogens found after storage, the number of seeds infected did not exceed economic injury level to cause disease condition to reduce in seed quality especially three months of storage.

5. CONCLUSION

Seeds in nylon sack with single layer had the highest germination percentage (98%), vigour (48.037) and vigour index and seeds stored in un-lined nylon sack had the least germination percentage of 84%, vigour 37, vigour index 13193. Un-layered nylon sack recorded the highest number of *Sitophilus oryzae* while Bucket recorded the least insect infestation. Twelve pathogens were identified, of which nine were pathogenic (*Alternaria spp, Bipolaris spp, Curvularia lunata, Phoma spp, Nigrospora spp Fusarium moniliforme, fusarium pallidoroseum,Tricoderma spp and Myrothecium)* and three saprophytic (*Rhizopus sp, Aspergillus flavus and Penicillium* sp). After three months of storage, seed viability for all the three varieties was above 80% proving the orthodox seed storage behavior of rice seeds.

REFERENCES

1. Fukagawa NK, Ziska LH. Rice: Importance for global nutrition. Journal of nutritional science and vitaminology. 2019;65(Supplement):S2-3.
2. Park, Hye Lin, Seong Hee Bhoo, Mi Kwon, Sang-Won Lee, and Man-Ho Cho. "Biochemical and expression analyses of the rice cinnamoyl-CoA reductase gene family." *Frontiers in Plant Science* 2017 (8): 2099.
3. FAO/SAA. *NERICA®: the New Rice for Africa–a Compendium. EA Somado, RG Guei and SO Keya (Eds.). Cotonou, Benin: Africa Rice Center (WARDA)*, 2008; 1-9.
4. Akowuah, JO., Addo A and Bart Plunge A. Influence of drying temperature and storage duration on fissuring and milling quality of Jasmine 85 rice variety. Journal of science and technology, 2012; 32;26-33
5. Asante, MD., Asante, BO., Acheampong, GK, Offei, SK, Gracen, V, Adu-Dapaah, H, & Danquah, E. Y. Farmer and consumer preferences for rice in the Ashanti region of Ghana: Implications for rice breeding in West Africa. *Journal of Plant Breeding and Crop Science*, 2013; *5*(12), 229-238.
6. Liliane, Tandzi Ngoune, and Mutengwa Shelton Charles. "Factors affecting yield of crops." *Agronomy-climate change & food security* (2020): 9.
7. Shetty, HS, Seed health testing and its role in plant protection proceedings, 1999–National seminar on seed science and technology, (Shetty, H.S. and H. S Prakash, Eds.)1st Edn Dep. Of Applied Botany and Seed Pathology, University of Mysore, India, 2002; 76-78
8. Bam RK, Craufurd PQ, Dorward PT Asiedu EA., Kumaga FK, and Ofori K. Introducing improved cultivar: understanding Farmers' Seed drying and storage practices in Central Ghana. Exp. 1 Agric. Cambridge University Press Reading, 2002; 43, 301-317.
9. Khalil ES, Ali M, Nour El-Din N, El-Mekawy M. Effect of Periods and Conditions of Preservation in Gene Banks on Seed Viability of *Balanites aegyptiaca* (L.) Del. Catrina: The International Journal of Environmental Sciences. 2018 Jul 1;4(2):21-9.
10. Lima DD, Dutra AS, Pontes FM, Bezerra FT. Storage of sunflower seeds. Revista Ciência Agronômica. 2014;45:361-9.
11. Rocha AJ, Sousa BL, Girão MS, Barroso-Neto IL, Monteiro-Júnior JE, Oliveira JT, Nagano CS, Carneiro RF, Monteiro-Moreira AC, Rocha BA, Freire VN. Cloning of cDNA sequences encoding cowpea (*Vigna unguiculata*) vicilins: Computational simulations suggest a binding mode of cowpea vicilins to chitin oligomers. International journal of biological macromolecules. 2018;117:565-73.

12. Keneni YG, Hvoslef-Eide AT, Marchetti JM. Mathematical modelling of the drying kinetics of *Jatropha curcas* L. seeds. Industrial crops and products. 2019;132:12-20.

13. Gupta, PC. Seed vigour testing. *Handbook of seed testing. New Delhi*, 1993; 242-249.

14. Mathur, SB and Kongsdal, O. Common Laboratory Seed Health Testing Methods for detecting Fungi. First Edition. Danish Government Institute of Seed Pathology for Developing Countries, Copenhagen, Denmark. 2001; 95-104

15. International rules for seed testing: International Seed Testing Association (ISTA). *Seed Sci. Technol.*, *27 2007*

16. Rai, PK., Kumar, G, & Singh, KK. Influence of packaging material and storage time on seed germination and chromosome biology of inbred line of maize (Zea mays L.). *Journal of Agricultural Technology*, 2011; *7*(6), 1765-1774.

17. Rehmani, MS., Xian, B, Wei, S, He, J, Feng, Z, Huang, H, & Shu, K Seedling establishment: The neglected trait in the seed longevity field. *Plant Physiology and Biochemistry*, 2023; 107765.

18. de Sousa, IG, Oliveira, J, Mexia, A, Barros, G., Almeida, C., Brazinha, C., ... & Brites, C. Advances in environmentally friendly techniques and circular economy approaches for insect infestation management in stored rice grains. *Foods*, 2023; *12*(3), 511.

19. Mesterházy,Á; Oláh, J; Popp,J Losses in the Grain Supply Chain: Causes andSolutions. *Sustainability* 2020**;** *12*, 2342.

20. Agarwal, VK. and Sinclair, JB., 1996. *Principles of seed pathology*. Crc Press.

21. Agarwal, RL Seed Technology. Second Edition. Oxford and IBH Publishing Co. Pvt LTD. India. 1995; 829

22. Agrios G.N. 2005. Plant pathology. Elsevier, USA.