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Effect of seed infection on seed quality and longevity under storage of three rice varieties produced at different environments

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Abstract
The study was conducted to elucidate the Effect of seed infection on seed quality and longevity under storage of three rice varieties produced at different environments at College of Agriculture, Acharya N. G. Ranga Agricultural University and Indian Institute of Rice Research, Hyderabad (India). In this present investigation, seed infection percentage in rice cultivars produced at two different environmental conditions have been studied and effect of seed moisture content on germination percentage and seed infection in rice varieties. The seed produced in arid environment i.e., at IIRR, Hyderabad showed minimum percent seed infection (15.26, 10.01, 18.51%), seed moisture content (11.33, 11.03, 11.14%) and maximum germination percentage (72.55, 76.66, 74.12%), whereas the seed produced in humid environment *i.e.,* at Central Rice Research Institute, Cuttack, showed maximum percent seed infection (19.49, 14.01, 22.14%), seed moisture content (11.98, 11.70, 12.31%) and low germination percentage (61.97, 64.84, 44.92%) was recorded in IR 36, IR 64 and Annada varieties respectively at nine monthly storage in gunny bags at ambient conditions. With respect to the age of seed, the freshly harvested seeds recorded low percent seed infection, seed moisture content and high percent of seed germination than one year old seed of three varieties at the end of the storage (9 months after storage). Among treatments, seeds treated with carbendazim @ 2.0g/kg showed high germination percentage and minimum percent seed infection and low moisture content percentage in all the three cultivars used for study at the end of storage experiment. Interaction among production environment, seed age and chemical treatments also showed significant difference for quality parameters. This investigation helps in understanding the potential role of production environment, seed age and means to control the seed from deterioration during storage.

Keywords: Germination percentage, Moisture content, Rice, Seed age, Seed infection, Seed treatment

Introduction
Grain production of a country depends on good quality seeds. Quality seeds play a very important role for the production of healthy crop. Healthy and pathogen free seeds are the basic requirements for disease free crop. Seeds are stored for a considerable period of time in order to catch the correct season. It is reported that 25% of the world's crops are affected by mould or fungal growth. Fungi growing on stored seeds, can reduce the germination rate along with loss in the quantum of carbohydrate, protein and total oil content, induces increased moisture content, free fatty acid content and enhancing other biochemical changes. The tropical climate with high temperature and high relative humidity along with unscientific storage conditions adversely affect the preservation of cereal grains, oilseeds, etc., which lead to the total loss of seed quality (Begum *et al*. 2013) [2].

Rice (*Oryza sativa* L.) is one of the most important staple food of more than 60 percent of the world's population and over 90% of rice is produced and consumed in Asia. Among the rice growing countries, India ranks second in production after China. India alone produces one fourth (22%) of the world rice and is grown in an area of 43.39 millon ha with the production and productivity levels of 104.32 million tonnes and 2404 kg/ha, respectively during 2015-16 (DAC&FW, 2016; http://eands.dacnet.nic.in). Rice being a major food grain throughout the world requires storage for one or more planting seasons before cultivation. In some countries like India have a preference for stored rice whilst others (e.g. Japan and China) favours fresh rice for consumption (Zhou *et al*. 2002) [46]. During storage, a number of physiochemical and physiological changes occur, this is usually termed ageing. These changes which include pasting properties, color, flavor and composition affect rice quality.

Seed health is a major consideration in any seed production programme next to vigour and viability of seeds. Seed borne fungi not only reduce the germination and vigour of the seedling but also act as a source of inoculums for the development of diseases in the field (Srinivas *et al.* 2001) [43]. For successful crop production seed must be sound and free from mycoflora which are likely to interfere with germination, emergence and subsequent performance of the crop in the main field. Under field conditions seeds are known to harbor several fungi, which affect their health seriously causing germination failure and partial to complete death of seedlings. Seed borne pathogens declines seed viability and vigour both in storage and field conditions and causes yield losses subsequently in the field (Anuja and Aneja, 2000) [4]. The increase in seed infestation might be due to loss of membrane integrity, lipid peroxidation and invasion of fungi. Bewley and Black (1983) [5] reported that extensive leakage of metabolites was one of the reasons for increased infestation during ageing process. Increase in leakage of free sugars, amino acids etc. from deteriorated seeds indirectly hasten the destruction by encouraging the growth of micro organisms. It is possible that fungi, which were already present in the seed sample even in low quantities, multiplied and were distributed among the seeds during storage.

During storage, number of biotic and abiotic factors also influenced the storage potential of seeds and results in gradual seed deterioration and ultimately death of the seeds (Kumar *et al.* 2014) [23]. Among several biotic factors threatening rice production, fungal diseases account for major losses (Grover *et al.* 2003) [10]. In general most of the fungicides act by inhibiting the energy metabolism, blocking biosynthesis or altering cell membranes of fungus. Carbendazim (Benzimazimidazoles) a systemic fungicide with curative and protection action, extensively used in agriculture, inhibit the development of germinal tube, formation of the aspersoria and growth of nucleus of fungus (Paweri *et al.* 2016) [32].

Being a staple food grain, it is grown in different parts of the country. It is assumed that as the production environment and storage conditions changes the spectrum and type of infection by the pathogens tend to change which ultimately causes severe losses during storage and also reduces the seed quality. Therefore, it is important to examine the effect of environmental variation on rice quality over a wide geographical domain with respect to the occurrence of seed borne fungi and seed infection under certain storage duration of those seed produced at different environmental regimes. With this context, investigations were undertaken to know the effect of seed infection on seed quality and longevity under storage of three rice varieties produced at different environments.

Materials and Methods

The storage experiment to understand the effect of seed infection on seed quality and longevity under storage of three rice varieties produced at different environments was conducted at ICAR - Indian Institute of Rice Research (IIRR) and College of Agriculture, Acharya N. G. Ranga Agricultural University, Hyderabad (India).

Seed source

In the experiment three rice varieties were used *viz.*, IR- 36, (V1), IR-64 (V2) and Annada (V3). The seed lots of these three varieties were procured from two production environment conditions *viz.*,Q_1 – Central Rice Research Institute (CRRI), Cuttack (humid region) and Q_2 –IIRR, Hyderabad (arid region). The weather data during the production period was also collected (Table 1 and Fig. 4). The one year old (P_1) and freshly harvested (P_2) Seeds of above three varieties were subjected to treatment with fungicides (carbendazim @2.0 g/kg of seed (T_1), mancozeb@ 2.5 g/kg (T_2) and seed without treatment was considered as control (T_3). Immediately after treatment the seeds were stored in gunny bags at ambient conditions (33°C and 57% RH). The samples were drawn at 0, 3, 6 and 9 months intervals to study physiological and seed health parameters. The experiment was conducted in Completely Randomized Block Design with Factorial concept (FCRD) and three replications have been taken in each parameter.

Seed moisture content (%)

Moisture content of seed was determined as per ISTA rules (ISTA, 1985). Five grams of seed was weighed, grounded and put in aluminium cups. The aluminium cups along with ground seed material was dried in hot air oven maintained at 130 ± 1ºC temperature for two hours. The moisture content was determined on dry weight basis by using the following formula.

$$\text{Moisture content (\%)} = \frac{M_2 - M_3}{M_2 - M_1} \times 100$$

Where, M_1 - Weight of empty container with its cover (g)
M_2 - Weight of container with its cover and ground seeds before drying (g)
M_3 - Weight of container with its cover and ground seeds after drying (g)

Seed germination (%)

Germination test was conducted on pure seed fraction using 100 seeds in three replicates following between paper (BP) method at 25ºC temperature and 93±2% relative humidity as per ISTA,1985 (Anonymous, 1999) [3]. The numbers of normal seedlings were counted on 5th day (first count) and 14th day (final count) of germination from all the replications. The average of three replications was expressed as germination percentage. The germination per cent was calculated based on the number of normal seedlings produced.

$$\text{Germination (\%)} = \frac{\text{Number of normal seedlings}}{\text{Total number of seeds planted}} \times 100$$

Seed infection (%)

During storage period of nine months, periodical observations on seed infection were recorded in terms of infected seeds. Observations were recorded at initial and at tri-monthly intervals from the seed of all treatments stored in gunny bags for 9 months under ambient conditions. Fungal infestation in the seed sample was determined by using standard blotter method (ISTA, 1993) [15]. Three discs of blotter paper (9 cm diameter) were dipped in beaker containing sterile distilled water and placed in petri plates on which seeds were placed with the help of forceps. Four hundred seeds of each variety were tested in 16 petriplates having twenty five seeds per plate. After labelling, these petri-plates were incubated at 25±1ºC under alternate cycles of 12 hours light and 12 hours darkness for seven days in BOD incubator. On seventh day, these plates were examined under stereo binocular microscope

and the percentage of total number of fungal colonies was calculated and the infected seeds were counted and identified the causal organism under binocular microscope.

$$\text{Seed infection (\%)} = \frac{\text{Number of infected seeds}}{\text{Total number of seeds planted}} \times 100$$

Statistical analysis

In each experiment, 100 seeds in each replication for germination test and seed infection (%) and 5 gm of seed for moisture content estimation have been used and each experiment was repeated three times. Standard errors (SEs) of the arithmetic means were calculated for each treatment. Three factorial analysis was made by using Indostat software package 8.0 version and Analysis of variance (ANOVA) constructed as per Panse and Sukhatme (1954) [33].

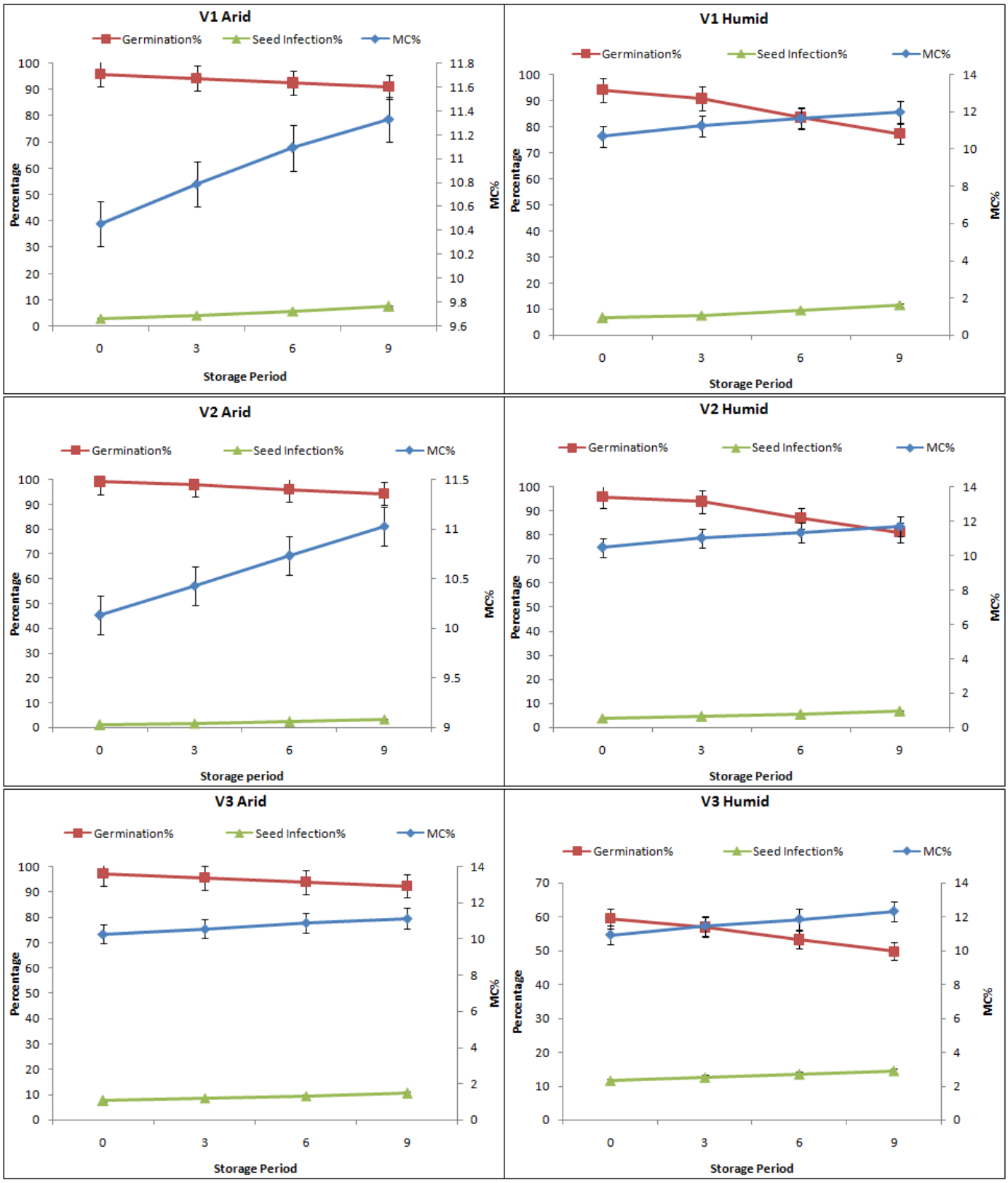

Fig 1: Effect of Moisture content on germination and seed infection% in three rice varieties after different months of storage period in humid and arid regions.

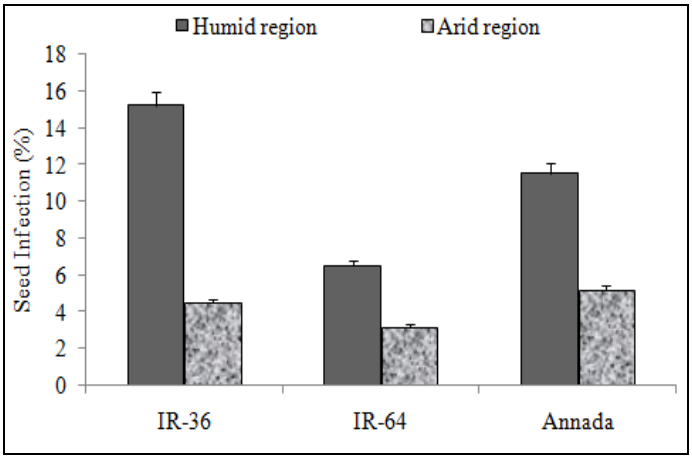

Fig 2: Percentage seed infection recorded in humid and arid region in three rice varieties

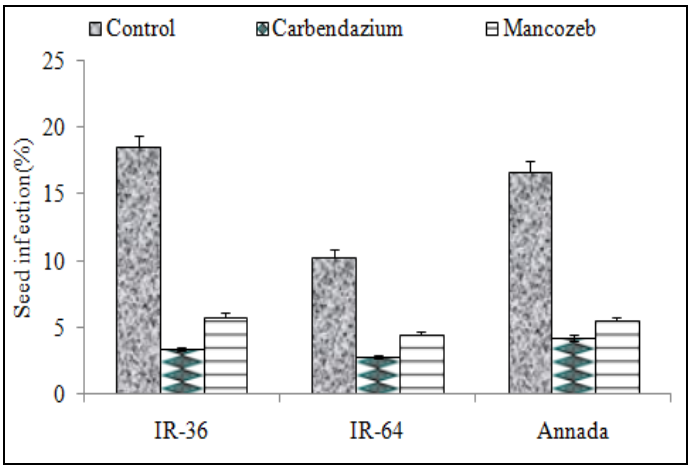

Fig 3: Effect of fungicides for the control of seed infection in three rice varieties

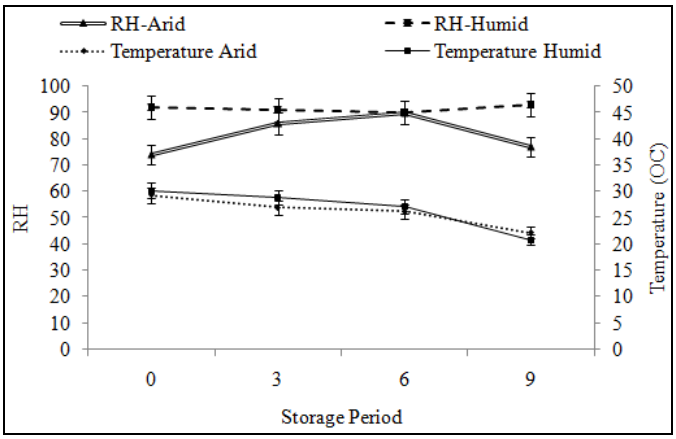

Fig 4: Average temperature and Relative Humidity (%) of the Humid and Arid region during nine months of storage period

Results and Discussion

The results on the seed infection (%), seed moisture content and Germination test in three rice varieties *viz.* IR-36, IR-64 and Annada as influenced by production environment, seed age and seed treatment and their interaction effect at different months of storage period are presented in Figures 1,2 and 3.

Production environment (Q)

Percent seed infection differed significantly among the two production environments in all the months of storage period. Among the production environments, seed produced at IIRR (Q_2) has recorded the lowest pathogen infection in all three varieties *i.e.* IR-36 (7.56%), IR-64 (3.39%) and Annada (10.51%) over seed produced at humid environment, CRRI (Q_1) *i.e.* IR-36 (11.62%), IR-64 (6.72%) and Annada (14.55%) at the end of storage period. With the advance of the storage period, the percent seed infection showed gradual increase in both the production environments (Fig 1).

Percent moisture content differed significantly among the two production environments in all the months of storage period. Seed produced at arid region (Q_2) has recorded the lowest moisture content in all three varieties *i.e.* IR-36 (11.33%), IR-64 (11.03%) and Annada (11.14%) over seed produced at humid environment (Q_1) *i.e.* IR 36 (11.98%), IR-64 (11.70%) and Annada (12.31%) at the end of storage period. With the advance of the storage period, the percent moisture content showed gradual increase in both the production environments (Fig 1).

Percent germination differed significantly among the two production environments in all the months of storage period. Seed produced at arid region (Q_2) has recorded the highest germination in all three varieties *i.e.* IR 36 (72.55%), IR-64 (76.66%) and Annada (74.12%) over seed produced at humid environment (Q_1) *i.e.* IR-36 (61.97%), IR-64 (64.84%) and Annada (44.92%) at the end of storage period. With the advance of the storage period, the percent germination showed gradual decrease in both the production environments (Fig 1).

In Humid region, the seed infection was highest in all the three varieties as compared to arid region. In humid region, germination% and seed infection rate was also depending on seed moisture content. According to storage period, the germination% was drastically decreased as moisture content was increase in all the verities on both the regions as we used in this study (Fig 2). After 9 months of storage period, seed infection was also increased and seed viability was decreased due to seed moisture (Fig 1). In Humid region, temperature and RH values was also highest as compared to the Arid region environment during the study throughout the storage periods (Fig 4) which help the increase seed deterioration and infection rate in all the varieties.

Seed age (P)

The seeds of one year old (P_1) and freshly harvested (P_2) varied significantly for per cent seed infection throughout the storage period. At the end of the storage period freshly harvested (P_2) seed has recorded lower seed infection per cent (7.16, 3.00 and 10.12%) over one year old (P_1) seed (12.02, 7.11 and 14.94%) Similarly, for seed moisture content (%) freshly harvested seed recorded lower values *i.e.*, 11.62, 11.29 and 11.64% over one year old seed (11.68, 11.43 and 11.81%) for IR 36, IR-64 and Annada respectively.

Whereas the percent germination recorded higher in freshly harvested rice seed (68.43, 71.54 and 64.08%) than one year old seed (66.09, 69.96 and 54.95%) in IR-36, IR-64 and Annada respectively at nine months after storage.

Seed treatment (T)

Per cent seed infection differed significantly among the seed treatments in all the months of storage period. Seed infection per cent was found to be decline with the advancement of storage period in all seed treatments. Among the seed treatments, seeds treated with carbendazim (T_1) recorded very low (5.75, 1.92 and 8.67%) pathogen infection. whereas, it was 9.26, 4.58 and 12.25 per cent in T_2. Higher seed infection of 13.76, 8.67 and 16.68 per cent was noticed in the treatment T_3 (untreated seed) at the end of storage period for IR 36, IR

64 and Annada respectively. Among the three rice varieties, IR 64 recorded lower seed infection percent values over IR 36 and Annada (Fig 2).

Seed moisture content (%) differed significantly among the seed treatments in all the months of storage period. Seed moisture content (%) was found to be increase slightly with the advancement of storage period in all seed treatments. Among the seed treatments, seeds treated with carbendazim (T_1) recorded lower moisture content (11.06, 10.85 and 11.14%). Whereas it was 11.61, 11.21 and 11.57 per cent in seed treated with mancozeb (T_2). Higher moisture content of 12.29, 12.03 and 12.46 per cent was noticed in the treatment T_3 (untreated seed) at the end of storage period for IR 36, IR 64 and Annada respectively. Among the three rice varieties, IR 64 recorded lower seed moisture content (%) values over IR 36 and Annada (Fig 1).

Significant differences among the seed treatments with respect to seed germination per cent were recorded in all the months of storage period. Seed germination per cent was found to be decline with the advancement of storage period in all seed treatments. At the end of the storage period, the highest seed germination per cent for IR 36, IR 64 and Annada was observed in carbendazim treated seed T_1 (89.58, 93.33 and 74.66%) followed by seed treated with mancozeb T_2 (86.79, 90.92 and 70.25%). The lowest was observed in untreated T_3 (75.63, 78.58 and 68.33%) respectively (Fig 2). Among the three rice varieties, IR 64 recorded higher seed germination per cent values over IR 36 and Annada.

Interaction effect between factors with respect to seed infection percentage

1. Interaction effect of production environment and seed age (Q x P)

Interaction effect on seed infection due to seed production environments and seed age was found to be significant in all the months of storage period. The seed produced at DRR and freshly harvested (Q_2P_2) recorded minimum seed infection (6.22, 2.56 and 9.12%) followed by Q_1P_2 (seed produced at CRRI and freshly harvested) with 8.11, 3.44 and 11.12 per cent and maximum seed infection (15.12, 10.00 and 18.00%) was noticed in Q_1P_1 (seed produced at CRRI and one year old) for IR 36, IR 64 and Annada respectively at the end of storage period (Table 2).

2. Interaction effect of production environment and seed treatment (Q x T)

Interaction effect on seed infection due to seed production environments and chemical treatments was found to be significant in all varieties throughout storage period except at initial and 3rd month of storage period for genotype Annada and IR 36 respectively. At the end of the storage period significantly lowest seed infection per cent for IR 36 and Annada was noticed in seed produced in arid environment and treated with carbendazim Q_2T_1 (3.68 and 6.50%) followed by seed produced in arid environment and treated with mancozeb Q_2T_2 (7.84 and 10.83%) respectively. At the end of the storage period significantly lowest seed infection per cent for IR 64 was noticed in Q_2T_1 (1.00%) followed by Q_1T_1 (2.83%). The maximum seed infection per cent (16.33, 11.33 and 19.17%) was observed in Q_1T_3 (seed produced at CRRI and untreated) for IR 36, IR 64 and Annada respectively (Table 2).

3. Interaction effect of seed age and seed treatment (P x T)

The interactions between P x T were found to be significant throughout the storage period. There was an increase in seed infection per cent values were recorded from initial to 9th month of storage significantly lowest seed infection per cent was noticed in freshly harvested seed with carbendazim treated P_2T_1 (3.50, 0.83 and 6.32%) followed by freshly harvest seed with mancozeb treated P_2T_2 (7.49, 2.83 and 10.50%) for IR 36, IR 64 and Annada respectively. The highest seed infection per cent was observed in one year old seed with untreated P_1T_3 (17.02, 12.01 and 19.82%) for IR 36, IR 64 and Annada respectively (Table 2).

4. Interaction effect of production environment, seed age and seed treatment (QxPxT)

The interactions of Q x P x T varied significantly for seed infection per cent throughout the storage period. At the end of the storage period significantly the lowest seed infection per cent was recorded in seed produced in arid environment and freshly harvested with treated with carbendazim $Q_2P_2T_1$ in IR 36 (0.00 to 0.66%), IR 64 (0.00 to 0.00%) and Annada (0.66 to 3.35%). Which was followed by seed produced in humid environment and freshly harvested and treated with carbendazim ($Q_1P_2T_1$) in all varieties *i.e.*. IR 36 (1.33 to 6.34%), IR 64 (0.00 to 1.67%) and Annada (6.32 to 9.32%) and $Q_2P_1T_1$ in all varieties IR 36 (1.33 to 6.34%), IR 64 (0.00 to 1.67%) and Annada (6.32 to 9.32%). The next best treatment, were $Q_2P_2T_2$ *i.e.* IR 36 (2.33 to 7.35%), IR 64 (0.00 to 2.67%) and Annada (7.34 to 10.36%) and $Q_1P_2T_2$ *i.e.* IR 36 (2.67 to 7.69%), IR 64 (0.00 to 3.00%) and Annada (7.67 to 10.67%). The highest seed infection per cent was observed in $Q_1P_1T_3$ *i.e.* IR 36 (17.33 to 22.35%), IR 64 (14.33 to 16.33%) and Annada (17.01 to 25.03%) (Table 3).

Seed quality changes due to storage fungi

Observation on seed health status or seed infection percentage showed that with the advance of the storage period, the seed borne pathogens associated during storage showed gradual increase in both the production environments. In this study, seed produced at arid environment showed low level of seed infection than that of humid environment (Fig 2), freshly harvested seed showed low level of seed infection than one year old seed and seeds treated with chemicals showed very low level of seed infection than that of untreated ones and the results are in conformity with Jayaweera *et al.* (1988) [17], Mazen *et al.* (1993) [25] and Habib *et al.* (2012) [12] in rice. Increasing the storage period of groundnut seeds upto nine months decreases the viability, while pathogen activity, moisture and sugar content in seeds increase gradually (Begum *et al.* 2013) [2]. De Frietas *et al.* (2000) [9] reported that with increase in storage period of cotton seeds, there was a linear decrease in viability of seeds and a linear increase in incidence of storage fungi. Raj *et al.* (2002) [35] identified that the species of *Aspergillus*, *Alternaria*, *Rhizoctonia*, *Fusarium*, *Phoma* and *Chaetomium* are affecting germination and emergence in soybean seeds.

Krishnappa *et al.* (2003) [22] reported that groundnut pods stored in gunny bag had recorded maximum infection ranged between 16 and 18% of *Aspergillus flavus*, *Aspergillus niger*, *Fusarium* spp. and *Pencillium* spp. and caused reduction in germination and vigour index. While Nargund *et al*. (2003) [28]

found the presence of *Aspergillus flavus, Aspergillus niger, Rhizoctonia* spp., *Fusarium* sp. and *Sclerotium rolfsii* in all the varieties of groundnut and they caused reduction in germination% and vigour index. In the present study, we observed and listed the different seed borne pathogens affected the rice seed throughout the storage period in different varieties of rice *viz.* in IR-36 (Table. 4), IR-64 (Table. 5) and Annada (Table. 6).

Paderes *et al.* (1997) [31] studied the relationship of moisture content and storage period to fungal population, seed germination, grain whiteness and translucency of rice was determined. The fungi observed were *Aspergillus flavus-oryzae, A. glaucus, A. nidulans, A. candidus, A. versicolor, A. terreus* and *A. niger* and unidentified species of *Penicillium, Trichoconiella, Curvularia, Fusarium, Syncephalastrum and Verticillium*. The predominant storage fungi were *A. flavus-oryzae* and *A. Candidus* and he reported a significant negative correlation between percentage germination and moisture content was observed. At 14.5-18.33% moisture content, the germination of stored paddy decreased with a marked increase of storage moulds. During storage, viability and vigour are lost due to many biotic factors like microflora. The storage fungi cause considerable damage and are responsible for deterioration and reduction in storage potential of seed. The probable reason for differences in storability of seeds treated with various chemicals might be due to variations in their effectiveness in combating the seed borne pathogen and also might be due to the persistence of these chemicals on seed surface during storage for longer time. The results are in conformity with the work of Kaur *et al.* (1990) [21].

Among the age of seeds, better quality was observed from freshly harvested seeds as compared to one year old seeds. During seed ageing, a number of physiochemical and physiological changes occur (Noomhorm *et al*. 1997) [30]. These changes in seed lead to reduce viability, growth efficiency and seedling characteristics after germination and quality. Gao *et al.* (2016) [16] and Bewley *et al*. (2013) [6] reported that rice seeds deteriorate during storage, leading to significant losses due to decreased viability and germination rates. Similar findings were also reported in rice (Zhou *et al.* 2002) [46] and soybean (Wang *et al.* 2012) [24].

Jiang Ling *et al.* (2007) [19] conducted a comparative experiment with one year stored and newly harvested seeds between W017 with LOX-3 null allele and Wuyujing 3 with LOX-3 function allele as a check. After 5 days of preageing, the germination rate of one year stored W017 was 87.3%, while that of one year stored Wuyujing 3 was only 14%, which indicated that Wuyujing 3 seeds were very sensitive to preageing after one year's storage, while newly harvested Wuyujing 3 seeds were insensitive to preageing.

The present study revealed that, with the advance in the storage period and seed treatments with chemicals, all the seed quality parameters were gradually decreased. Similar findings were reported in marigold (Kumar *et al*. 2014) [23] and chickpea (Chormule *et al.* 2015) [8]. Among chemicals, carbendazim constituent may inhibit the pathogen activity and favours for germination and quality parameters as compared to mancozeb (Fig. 3). The results of this study were confirmed with Prasad *et al*. (2010) [27] in rice. This is in accordance with the findings of Singh *et al.* (1996) [41] in onion who have reported carbendazim as an effective fungicide against *Alternaria alternata, Rhizopus* spp, *Fusarium* spp and exhibited higher germination and vigour index in seeds of onion and other crop seeds. Similar results were made by Pameri *et al*. (2016) [32] in rice. Fungicide seed treatments containing fludioxonil and azoxystrobin can improve germination, plant population, growth, and yield in maize (Solorzano *et al*. 2011) [42], and with thiram and delson in eggplant (Reddy and Reddy, 1994) [39] and scented rice Cv. Mugad Sugandha (Raikar *et al.* 2008) [34]. Similarly, Butt *et al.* (2011) [7] investigated the effect of four chemical fungicides namely antracal, topsin, mancozeb and derosal on seed-borne mycoflora of rice.

The seed treatment had significant effect on vigour during all the months of storage period. The seeds treated with carbendazim gave higher vigour followed by mancozeb. This might be due to maintenance of cell membrane integrity leading to less susceptibility to peroxidation and free radical reactions, as these chemicals possess antimicrobial agents hence the deterioration level in these treated seeds was less which is evident with lower EC values and seed infection with higher activity of dehydrogenase enzyme. Similar findings were also reported by Ravi *et al.* (2007) [38] in chilli seed treated with potassium iodide and $CaOCl_2$.

Seed treated with carbendazim was found to preserve the quality of seed by its antifungal effect. Carbendazim (Fig 3) also protects the seed from fungal and insect attack finally contributing to seed quality parameters (Taylor and Eckenrode, 1993). Similar findings were also reported by Jayaraj *et al.* (1988) [16] in capsicum, Gupta and Dharamsingh (1990) [11] in vegetable mung and cowpea seeds for bavistin and thiram, Raju and Sivaprakasam (1994) [36] in cabbage, and Sathishkumar, (2005) [40] in brinjal.

Seed treatment become more economical and effective when it is carried out with respect to nature of pathogen and level of infection percentage (Neergaard, 1979) [29]. Carbendazim is effective against storage rot of groundnut caused by *A. flavus* (Rathod *et al*, 2010) [37]. Since carbendazim is systemic in nature it inhibits the colony growth and sporulation of fungi and eradicates both the external and internally seed borne pathogens (Mohanna and Sharma, 1991; Habib *et al*, 2007) [26, 13]. Vasundhara and Gowda (1999) [45] found that groundnut seeds treated with thiram had higher germination over control after 12 months of storage.

Conclusion

Storage fungi influence the seed quality parameters and decrease the germination potential of the seeds during storage. From the investigation it is concluded that, in the context where rice has to be stored at least for a year or more, before it's planting in the field, so the factors studied helps in understanding and control the seed deterioration and infection during storage. Overall, interaction effect of production environment, seed age and chemical treatments (QxPxT) showed significance difference. The minimum percent seed infection and lower seed moisture content and maximum percent germination was observed from ($Q_2P_2T_1$) arid environment, freshly harvest seeds and carbendazim treatment @2.0g/kg seeds. Overall, in Humid region, the seed infection was highest in all the three varieties as compared to arid region due to seed moisture content. Temperature and RH values observed maximum in Humid region, as compared to the Arid region environment during the study throughout the storage which caused seed deterioration and infection rate in all the varieties. The successful and fruitful crop production depends on the availability of good quality seed and this sort of study would definitely helps in recommending the possible means to improve the storability and quality of rice seed, without which the expensive and well planned crop production approach may fail.

Table 1: Monthly meteorological data recorded at ARI, Rajendra Nagar and CRRI, Cuttack from April, 2011 to March, 2012

Month	Temperature (^{0}C)				Mean temp (^{0}C)		Relative humidity (%)				Rainfall (mm)		Sunshine (hrs)		Evaporation	
	Max		Min				I		II							
	A	B	A	B	A	B	A	B	A	B	A	B	A	B	A	B
Apr-11	36.7	35.6	21.9	24.8	29.3	30.2	74.0	92.0	35.0	52.0	0.0	10.6	8.2	7.2	5.0	4.8
May-11	39.2	36.9	25.4	29.0	32.3	33.0	56.0	90.0	28.0	58.0	1.0	175.6	8.5	8.1	6.4	5.2
Jun-11	35.0	33.2	24.4	28.5	29.7	30.9	74.0	89.0	45.0	63.0	4.0	310.2	6.1	5.3	6.1	5.5
Jul-11	31.2	30.5	22.7	27.2	26.9	28.9	86.0	91.0	62.0	73.0	11.0	315.9	4.8	3.2	4.8	4.0
Aug-11	30.2	30.1	22.7	27.0	26.4	28.6	90.0	93.0	71.0	80.0	9.0	375.6	3.9	2.6	4.4	4.1
Sep-11	30.7	29.5	22.0	26.0	26.3	27.8	88.0	92.0	74.0	76.0	5.0	368.6	5.5	2.9	2.8	4.3
Oct-11	32.1	31.6	20.3	22.8	26.2	27.2	90.0	90.0	69.0	61.0	4.0	30.5	6.6	8.8	2.7	4.0
Nov-11	30.1	30.0	15.4	19.8	22.8	24.9	82.0	92.0	45.0	49.0	1.0	0.0	8.2	9.0	2.4	3.8
Dec-11	29.9	28.7	12.6	15.9	21.3	22.3	83.0	89.0	40.0	42.0	0.0	0.0	8.5	5.8	2.3	3.7
Jan-12	30.1	26.5	14.2	15.0	22.2	20.8	77.0	93.0	37.0	56.0	0.0	100.2	8.5	6.2	2.3	3.9
Feb-12	33.0	31.2	15.4	19.3	24.2	25.3	73.0	91.0	25.0	40.0	0.0	0.0	9.5	8.5	3.2	4.1
Mar-12	36.9	34.8	17.3	24.5	27.1	29.7	62.0	95.0	21.0	43.0	0.0	0.0	8.9	7.3	4.4	4.8

* A – Agricultural Research Institute, Rajendra Nagar, Hyderabad and B - Central Rice Research Institute, ICAR, Cuttack

Source: ARI, Agro Climatic Research Centre, Rajendra Nagar, Hyderabad- 500 030 and CRRI, Annual Report 2011-12, Central rice research institute, ICAR, Cuttack, (Odisha), 753006, India.

Table 2: Influence of production environment, seed age and seed treatment on seed infection (%) in three different varieties of rice during storage.

Varieties												
Treatments	IR 36				IR 64				Annada			
	Months of storage											
	0	3	6	9	0	3	6	9	0	3	6	9
Q x P Interaction												
Q_1P_1	10.11 (17.89)	11.12 (18.92)	13.13 (20.81)	15.12 (22.55)	7.11 (14.16)	8.12 (15.56)	9.12 (16.78)	10.00 (17.75)	15.12 (22.55)	16.00 (23.30)	17.01 (24.10)	18.00 (24.88)
Q_1P_2	3.10 (9.66)	4.11 (11.38)	6.12 (14.14)	8.11 (16.43)	0.57 (5.04)	1.00 (5.74)	1.78 (7.23)	3.44 (10.28)	8.11 (16.43)	9.11 (17.47)	10.13 (18.46)	11.12 (19.40)
Q_2P_1	3.89 (10.84)	4.90 (12.42)	6.89 (15.01)	8.90 (17.21)	1.44 (6.60)	2.00 (7.66)	2.89 (9.10)	4.22 (11.32)	8.89 (17.21)	9.89 (18.21)	10.90 (19.17)	11.88 (20.08)
Q_2P_2	2.33 (8.41)	3.34 (9.82)	4.78 (11.68)	6.22 (13.31)	0.56 (5.09)	1.00 (5.98)	1.67 (7.18)	2.56 (8.71)	6.22 (13.31)	7.12 (14.50)	8.12 (15.72)	9.12 (16.94)
Mean	4.86	5.87	7.73	9.59	2.42	3.03	3.86	5.06	9.58	10.53	11.54	12.53
SEd+	0.648	0.609	0.667	0.666	0.643	0.667	0.676	0.737	0.667	0.648	0.646	0.647
C.D (0.05)	1.337	1.256	1.376	1.375	1.327	1.376	1.395	1.521	1.376	1.337	1.333	1.337
Q x T Interaction												
Q_1T_1	2.83 (9.27)	3.85 (11.04)	5.84 (13.85)	7.85 (16.18)	0.67 (5.30)	1.16 (6.40)	1.83 (7.56)	2.83 (9.41)	7.84 (16.18)	8.85 (17.23)	9.85 (18.22)	10.84 (19.17)
Q_1T_2	5.67 (13.18)	6.67 (14.53)	8.68 (16.84)	10.67 (18.87)	2.83 (8.89)	3.50 (9.77)	4.50 (11.29)	6.00 (13.66)	10.67 (18.87)	11.66 (19.80)	12.68 (20.70)	13.66 (21.57)
Q_1T_3	11.33 (18.87)	12.34 (19.8)	14.34 (21.3)	16.33 (23.43)	8.00 (14.1)	9.01 (15.78)	10.01 (17.17)	11.33 (18.97)	16.32 (23.43)	17.16 (24.13)	18.18 (24.92)	19.17 (25.68)
Q_2T_1	0.83 (5.69)	1.34 (6.71)	2.51 (8.53)	3.68 (10.07)	0.00 (4.06)	0.00 (4.06)	0.33 (4.62)	1.00 (6.00)	3.67 (10.07)	4.51 (11.42)	5.52 (12.84)	6.50 (14.26)
Q_2T_2	2.83 (9.57)	3.84 (11.22)	5.84 (13.94)	7.84 (16.23)	0.33 (4.62)	0.83 (5.69)	1.83 (7.55)	3.17 (10.06)	7.83 (16.23)	8.83 (17.27)	9.84 (18.26)	10.83 (19.20)
Q_2T_3	5.67 (13.61)	7.18 (15.42)	9.18 (17.5)	11.18 (19.47)	2.67 (8.84)	3.67 (10.71)	4.67 (12.26)	6.00 (13.97)	11.17 (19.47)	12.15 (20.37)	13.18 (21.24)	14.17 (22.08)
SEd+	1.122	1.147	1.155	1.155	1.114	1.155	1.171	1.276	1.155	1.122	1.120	1.121
C.D (0.05)	2.315	2.367	2.384	2.384	2.299	2.384	2.417	2.634	2.384	2.315	1.231	2.313
P x T Interaction												
P_1T_1	3.00 (9.66)	4.02 (11.35)	6.01 (14.08)	8.02 (16.37)	0.67 (5.30)	1.17 (6.40)	2.00 (7.84)	3.00 (9.71)	7.99 (16.37)	9.01 (17.41)	10.02 (18.39)	11.02 (19.33)
P_1T_2	6.00 (13.75)	7.01 (15.02)	9.02 (17.25)	11.01 (19.22)	3.17 (9.45)	4.00 (10.85)	5.01 (12.31)	6.3 (14.15)	11.01 (19.22)	12.02 (20.14)	13.03 (21.02)	14.01 (21.87)
P_1T_3	12.00 (19.68)	13.01 (20.64)	15.00 (22.40)	17.02 (24.00)	9.00 (16.37)	10.01 (17.59)	11.00 (18.68)	12.01 (19.73)	17.02 (24.07)	17.81 (24.72)	18.82 (25.49)	19.82 (26.23)
P_2T_1	0.67 (5.30)	1.17 (6.40)	2.34 (8.31)	3.50 (9.87)	0.00 (4.06)	0.00 (4.06)	0.17 (4.34)	0.83 (5.69)	3.50 (9.87)	4.35 (11.24)	5.32 (12.67)	6.32 (14.09)
P_2T_2	2.50 (9.01)	3.52 (10.73)	5.51 (13.53)	7.49 (15.88)	0.00 (4.06)	0.33 (4.62)	1.33 (6.54)	2.83 (9.57)	7.49 (15.88)	8.48 (16.94)	9.52 (17.94)	10.50 (18.90)
P_2T_3	5.00 (12.79)	6.50 (14.67)	8.51 (16.88)	10.50 (18.86)	1.67 (7.08)	2.67 (8.91)	3.67 (10.75)	5.33 (13.22)	10.50 (18.86)	11.51 (19.78)	12.48 (20.67)	13.51 (21.52)
Mean	4.86	5.87	7.73	9.59	2.42	3.03	3.86	5.06	9.58	10.53	11.54	12.53
SEd+	0.793	0.811	0.817	0.816	0.788	0.817	0.828	0.903	0.817	0.793	0.795	0.793
C.D (0.05)	1.638	1.673	1.685	1.684	1.626	1.685	1.708	1.863	1.685	1.638	1.640	NS*

*Figures within the parentheses indicate arcsine transformed values. ** Non significant, Q_1: Seed produced in humid environment, Q_2: Seed produced in arid environment, P_1: One year old seed after harvest (*Kharif, 2011*), P_2: Freshly harvested seed (*Rabi, 2012*), T_1: Seed treated with carbendazim@ 2.0g/kg, T_2: Seed treated with mancozeb@ 2.5g/kg and T_3: Untreated (control).

Table 3: Interaction between production environments, seed age and seed treatments on percent seed infection in three rice varieties.

Varieties												
Treatments	**IR 36**				**IR 64**				**Annada**			
	Months of storage											
	0	**3**	**6**	**9**	**0**	**3**	**6**	**9**	**0**	**3**	**6**	**9**
Q x P x T Interaction												
$Q_1P_1T_1$	4.33 (12.00)	5.34 (13.34)	7.34 (15.70)	9.34 (17.78)	1.33 (6.54)	2.33 (8.75)	3.33 (10.50)	4.00 (11.48)	9.34 (17.78)	10.34 (18.75)	11.36 (19.67)	12.34 (20.56)
$Q_1P_1T_2$	8.67 (17.10)	9.68 (18.10)	11.68 (19.96)	13.67 (21.69)	5.67 (13.73)	6.67 (14.93)	7.67 (16.05)	9.00 (17.44)	13.66 (21.69)	14.68 (22.51)	15.68 (23.31)	16.66 (24.10)
$Q_1P_1T_3$	17.33 (24.57)	18.34 (25.33)	20.34 (26.78)	22.35 (28.18)	14.33 (22.21)	15.33 (23.02)	16.33 (23.81)	(17.0124.31)	22.35 (28.18)	23.00 (28.65)	24.02 (29.33)	25.03 (29.99)
$Q_1P_2T_1$	1.33 (6.54)	2.34 (8.75)	4.34 (11.10)	6.34 (14.57)	0.00 (4.06)	0.00 (4.06)	0.33 (4.62)	1.67 (7.33)	6.32 (14.57)	7.35 (15.70)	8.35 (16.77)	9.32 (17.78)
$Q_1P_2T_2$	2.67 (9.27)	3.68 (10.96)	5.68 (13.73)	7.69 (16.05)	0.00 (4.06)	0.33 (4.62)	1.33 (6.54)	3.00 (9.88)	7.67 (16.05)	8.66 (17.10)	9.69 (18.10)	10.67 (19.05)
$Q_1P_2T_3$	5.33 (13.18)	6.34 (14.43)	8.34 (16.68)	10.33 (18.68)	1.67 (7.01)	2.67 (8.55)	3.67 (10.53)	5.67 (13.63)	10.33 (18.68)	11.32 (19.61)	12.34 (20.51)	13.35 (21.37)
$Q_2P_1T_1$	1.67 (7.33)	2.68 (9.36)	4.68 (12.46)	6.67 (14.95)	0.00 (4.06)	0.00 (4.06)	0.67 (5.18)	2.00 (7.95)	6.67 (14.95)	7.66 (16.07)	8.69 (17.12)	9.65 (18.11)
$Q_2P_1T_2$	3.33 (10.40)	4.34 (11.94)	6.34 (14.54)	8.35 (16.75)	0.67 (5.18)	1.33 (6.77)	2.33 (8.56)	3.67 (10.86)	8.33 (16.75)	9.34 (17.77)	10.35 (18.73)	11.34 (19.66)
$Q_2P_1T_3$	6.67 (14.80)	7.68 (15.95)	9.68 (18.03)	11.68 (19.91)	3.67 (10.53)	4.67 (12.16)	5.67 (13.55)	7.00 (15.14)	11.67 (19.91)	12.66 (20.79)	13.65 (21.65)	14.66 (22.48)
$Q_2P_2T_1$	0.00 (4.06)	0.00 (4.06)	0.34 (4.62)	0.66 (5.18)	0.00 (4.06)	0.00 (4.06)	0.00 (4.06)	0.00 (4.06)	0.66 (5.18)	1.33 (6.77)	2.35 (8.56)	3.35 (10.40)
$Q_2P_2T_2$	2.33 (8.75)	3.34 (10.50)	5.34 (13.34)	7.35 (15.70)	0.00 (4.06)	0.33 (4.62)	1.33 (6.54)	2.67 (9.27)	7.34 (15.70)	8.34 (16.77)	9.34 (17.78)	10.36 (18.75)
$Q_2P_2T_3$	4.67 (12.42)	6.68 (14.90)	8.68 (17.08)	10.67 (19.03)	1.67 (7.15)	2.66 (9.27)	3.67 (10.96)	5.00 (12.81)	10.67 (19.03)	11.68 (19.95)	12.66 (20.83)	13.67 (21.68)
Mean	4.86	5.87	7.73	9.59	2.42	3.03	3.86	5.06	9.58	10.53	11.54	12.53
SEd+	1.122	1.147	1.155	1.155	1.114	1.155	1.171	1.276	1.155	1.122	1.120	1.121
C.D (0.05)	2.315	2.367	2.384	2.384	2.299	2.384	2.417	2.634	2.384	2.315	1.231	2.313

Figures within the parentheses indicate arcsine transformed values. *Non-significant, Q_1: Seed produced in humid environment, Q_2: Seed produced in arid environment, P_1: One year old seed after harvest (*Kharif, 2011*), P_2: Freshly harvested seed (*Rabi, 2012*), T_1: Seed treated with carbendazim@ 2.0g/kg, T_2: Seed treated with mancozeb@ 2.5g/kg and T_3: Untreated (control).

Table 4: Fungal pathogens associated with IR- 36 variety as influenced by production environment, seed age and seed treatment during storage

Factor	Sub factor	Seed borne fungi (%)	Fungal flora observed in different treatments
Production environment (Q)	Humid environment (CRRI, Cuttack) (Q_1)	15.2	*Aspergillus flavus, A. niger, Cercospora oryzae, Helminthosporium oryzae, Alternaria padwickii, Fusarium oxysporum, Curvularia lunata, Pyricularia oryzae, Penicillium* and *Rhizopus stolinifer etc.*
	Arid environment (IIRR, Hyderabad) (Q_2)	4.5	*Aspergillus flavus, Aspergillus niger, Fusarium oxysporum, Curvularia lunata etc.*
Age of seed (P)	One year old seed (P_1)	10.2	*Helminthosporium oryzae, Bipolaris oryzae, Alternaria padwickii, Aspergillus flavus, A. niger, Fusarium oniliforme, F. oxysporum, Curvularia lunata* and *Penicillium oxalicum* and *Rhizopus* spp. *etc.*
	Freshly harvested seed (P_2)	4.2	*Bipolaris oryzae, Cercospora oryzae, Aspergillus flavus, Curvularia lunata etc.*
Treatments (T)	Carbendazim @ 2.0g/kg (T_1)	3.4	*Curvularia lunata, Aspergillus flavus, Alternaria padwickii etc.*
	Mancozeb @ 2.5g/kg (T_2)	5.8	*Fusarium moniliforme, Bipolaris oryzae, Aspergillus flavus, A. niger* and *Penicillium oxalicum etc.*
	Untreated (control) (T_3)	18.5	*Fusarium moniliforme, Fusarium semitectum, Phoma* sp., *Cercospora oryzae, Alternaria padwickii, Alternaria longissima, Curvularia* sp., *Aspergillus* sp., *Tilletia berclayana Sarocladium oryzae, Helminthosporium oryzae, Penicillium oxalicum, Pyricularia oryzae* and *Rhizopus* spp. *etc.*

Table 5: Fungal pathogens associated with IR- 64 variety as influenced by production environment, seed age and seed treatment during storage

Factor	Sub factor	Seed borne fungi (%)	Fungal flora observed in different treatments
Production environment (Q)	Humid environment (CRRI, Cuttack) (Q_1)	6.5	*Penicillium oxalicum, Aspergillus flavus, A. niger, Helminthosporium oryzae, Fusarium moniliforme, Curvularia lunata etc.*
	Arid environment (IIRR, Cuttack) (Q_2)	3.2	*Aspergillus flavus, A. niger, Curvularia lunata etc.*
Age of seed (P)	One year old seed (P_1)	7.5	*Helminthosporium oryzae, Alternaria padwickii, Aspergillus flavus, A. niger, Fusarium moniliforme, Curvularia lunata* and *Penicillium oxalicum etc.*
	Freshly harvested seed (P_2)	4.6	*Bipolaris oryzae, Cercospora oryzae, Aspergillus flavus, Curvularia lunata etc.*
Treatments (T)	Carbendazim @ 2.0g/kg (T_1)	2.8	*Curvularia lunata, Aspergillus flavus etc.*
	Mancozeb @ 2.5g/kg (T_2)	4.5	*Fusarium moniliforme, Aspergillus flavus, A. niger* and *Penicillium* spp. *etc.*
	Untreated (control) (T_3)	10.3	*Fusarium moniliforme, Fusarium semitectum, Cercospora oryzae, Alternaria padwickii, Curvularia lunata, Aspergillus niger, Helminthosporium oryzae, Penicillium oxalicum, Pyricularia oryzae* and *Rhizopus stolinifer etc.*

Table 6: Fungal pathogens associated with Annada variety as influenced by production environment, seed age and seed treatment during storage

Factor	Sub factor	Seed borne fungi (%)	Fungal flora observed in different treatments
Production environment (Q)	Humid environment (CRRI, Cuttack) (Q_1)	11.5	*Aspergillus flavus, A. niger, Cercospora oryzae, Helminthosporium oryzae, Alternaria padwickii, Fusarium oxysporum, Curvularia lunata, Pyricularia oryzae, Penicillium oxalicum, Bipolaris oryzae* and *Rhizopus* spp. *etc.*
	Arid environment (IIRR, Cuttack) (Q_2)	5.2	*Aspergillus flavus, Aspergillus niger, Fusarium oxysporum, Curvularia lunata, Penicillium oxalicum etc.*
Age of seed (P)	One year old seed (P_1)	10.6	*Helminthosporium oryzae, Bipolaris oryzae, Aspergillus flavus, A. niger, Fusarium moniliforme, F. oxysporum, Curvularia lunata, Penicillium* spp. and *Rhizopus stolinifer, Alternaria padwickii etc.*
	Freshly harvested seed (P_2)	6.4	*Aspergillus flavus, A. niger, Curvularia lunata, Bipolaris oryzae, Cercospora oryzae, Fusarium moniliforme*
Treatments (T)	Carbendazim @ 2.0g/kg (T_1)	4.2	*Curvularia lunata, Aspergillus flavus, A. niger, Alternaria padwickii etc.*
	Mancozeb @ 2.5g/kg (T_2)	5.5	*Fusarium moniliforme, Bipolaris oryzae, Aspergillus flavus, A. niger* and *Penicillium oxalicum etc.*
	Untreated (control) (T_3)	16.7	*Fusarium moniliforme, Fusarium semitectum, Phoma* sp., *Cercospora oryzae, Bipolaris oryzae, Alternaria padwickii, Alternaria longissima, Curvularia* sp., *Aspergillus flavus, A.niger, Tilletia berclayana, Sarocladium oryzae, Helminthosporium oryzae, Penicillium oxalicum, Pyricularia oryzae* and *Rhizopus stolinifer etc.*

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