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Short communications

Effect of mid-storage invigoration treatments on biochemical activity of enzyme associated with seed quality of paddy under ambient storage conditions

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Abstract

Mid-storage invigoration treatments improve the physiological condition of the seed and increase storability of the treated seeds. This investigation was performed to assess the biochemical alterations in seed quality of treated seeds stored under ambient condition. The seed quality parameters such as dehydrogenase, alpha-amylase and protease were assessed in these stored seeds at monthly intervals. There were significant differences among the seed treatments with respect to the biochemical enzyme activities as the age and storage period increased.The enzymes which help to maintain the quality and storability of seeds *viz.* dehydrogenase, alpha-amylase and protease were maximum in the seeds treated with neem oil, hydration dehydration and red chilli powder.These treatments outperformed all other treatments in enhancing seed quality, under ambient condition. Therefore the cost effective and ecofriendly mid-storage invigoration treatments provide advantage to the farmers for prolonging paddy seed storage without compromising seed quality.

Key words: Alpha amylase, Dehydrogenase, Mid-storage invigoration, Protease.

Mid-storage seed invigoration treatments play a key role in maintaining seed quality over time. These treatments aim to improve the physiological status of seeds, resulting in enhanced germinability, greater storability, and better overall performance compared to untreated seeds. Essentially, they help maintain seed vigor during the storage period. (Renugadevi et al., 2006). The cost-effective invigoration treatments are practically more feasible in seed storage. Botanicals like neem oil was found superior in maintaining higher seed quality and in prolonging seed storage (Amruta et al., 2015). Seed treatment with red chilli powder slowed down seed deterioration under natural ageing conditions, showed higher dehydrogenase enzyme activity and improved germinability of stored seeds over the untreated seeds (Saha and Mandal, 2016). Midstorage correction seed treatment such as hydration dehydration improved the seed vigour and subsequently storability. Extended soak times for treatment could also help to clear the seed of toxic compounds that have accumulated. (Sundralingam et al., 2023).

The observed modulation in biochemical parameters in midstorage invigorated seeds, stored under ambient conditions, serves as an indicator for the preservation of seed quality. The biochemical parameters, such as dehydrogenase, alpha-amylase, and protease enzyme activity provide insights into enhancing seed quality by maintaining the physiological condition of the seeds. Dehydrogenase enzyme activity reflects the overall metabolic state of seeds which is essential for seed quality. Protease and alpha amylase activity plays a role in mobilization of seed storage reserves such as proteins and starch there by providing energy for germination there by maintain seed quality and longevity (Damaris et al., 2019).Monitoring these enzymes provide information on seed quality, germination potential, and storability (Joshi, 2018).

Therefore, the study was undertaken to identify the effect of seed invigoration treatment administered midway through the storage period on variations in biochemical constituents and ultimately on the fluctuating seed quality.

The research trial was started from October 2022 to March 2023 at the Regional Agricultural Research station, Pattambi in a completely randomized design. Six month old seeds of the paddy variety Jyothi were subjected to ten mid-storage invigoration treatments, as per dosage shown in Table 1. Three replications were studied for each treatment,with a sample size of one fourth kilogram of seeds for each replication. Seeds were soaked in water in hydration dehydration treatment for four hours, ascorbic acid and calcium chloride solutions in other treatments for two hours, and then dried to a moisture content of less than 13 per cent. Seeds were treated with powders of albizzia leaf, neem leaf, red chilli and vayambu leaf in other treatments. Seeds were treated with neem cake, neem oil in the treatments as per dosage. The seeds were then packed in gunny bags and stored in ambient

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conditions. At the monthly intervals, the treated seeds were tested for biochemical enzyme activity such as dehydrogenase activity, alpha amylase activity, and protease activity.

Dehydrogenase activity

The test involved using a 0.5% solution of 2,3,5-triphenyl tetrazolium chloride dissolved in Sorenson's buffer solution as a solvent. This solution was used to assess dehydrogenase activity and seed vigour. After decanting the coloured solution, a blue filter (470 nm) was employed as a blank for measuring colour intensity using a spectrophotometer.The procedure and measurement of OD value was done referred to Marthandan and Jerlin, 2017.

α-amylase activity

To extract enzymes, three replicates of 500 mg from each pre-germinated seed sample underwent homogenization in 1.8mL of cold 0.02M sodium phosphate buffer (pH6.0). After centrifugation at 20,000 rpm for 20 minutes, a spectrophotometer measured the colour change at 620 nm.The output of the calculation in mg maltosemin^{-1} and the procedure was done referred to Naikand Chetti, 2017; Marthandan and Jerlin, 2017.

Protease activity

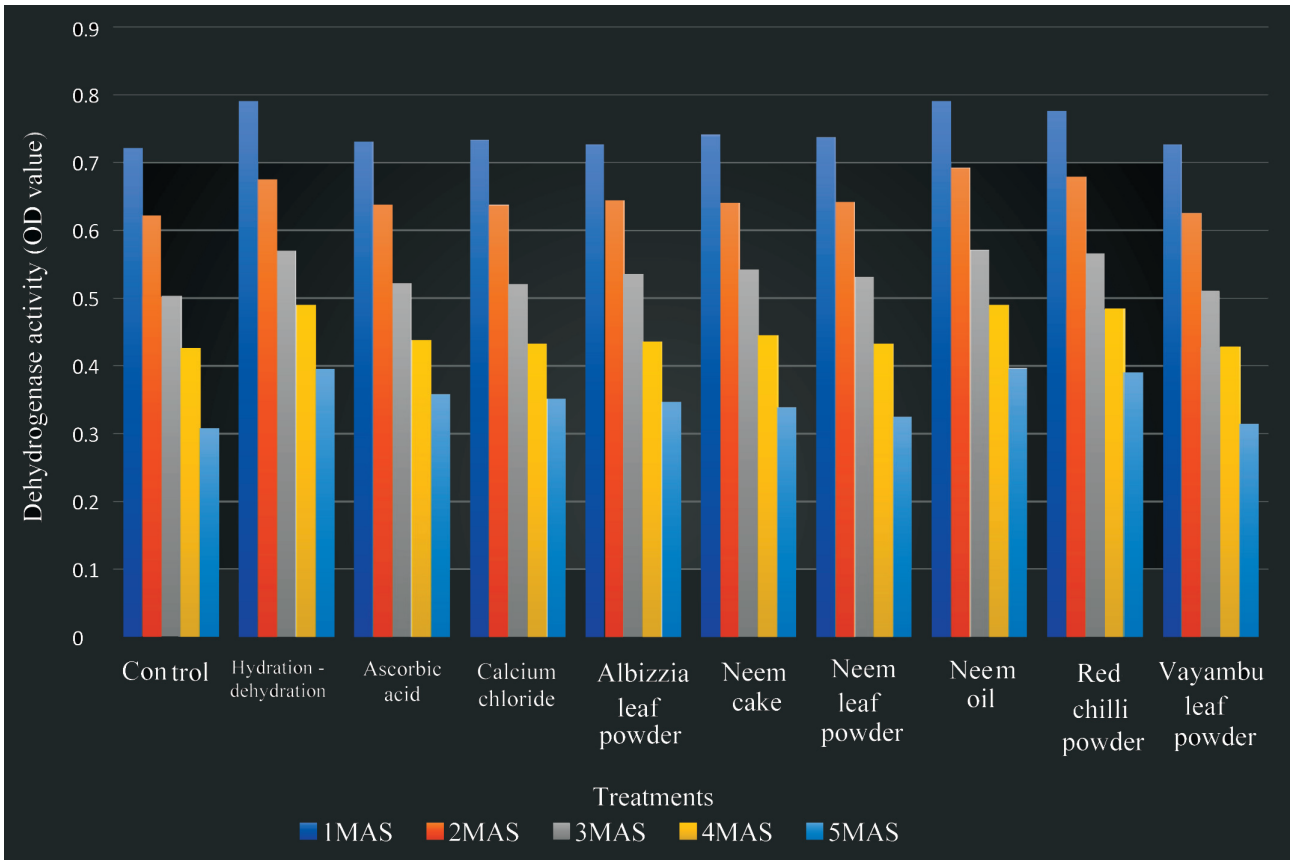
One millilitre of embryo extract was combined with 0.5 millilitres of 1% casein solution in 0.1M Tris-HC1 buffer (pH8.5). Following a 30 minute incubation period at 37°C,1.25ml of five per cent tricarboxylic acetic acid (TCA) was added to halt the process. One protease activity unit was defined as an increase of 0.01 absorbance units at 280 nm under the assay's parameters. The OD value was used to calculate the protease activity (Naik and Chetti, 2017; Marthandan and Jerlin, 2017).

Table1. Treatment details

SI.No	Treatment	Dosage/kg of Seeds
T1	Control	Untreated seeds
T2	Hydration - dehydration treatment	1l
T3	Ascorbic acid	10g/l
T4	Calcium chloride	10g/l
T5	Albizzia leaf powder	2g/kg
T6	Neem cake	5g/kg
T7	Neem leaf powder	2g/kg
T8	Neem oil	2ml/kg
T9	Red chilli powder	1g/kg
T10	Vayambu leaf powder	2g/kg

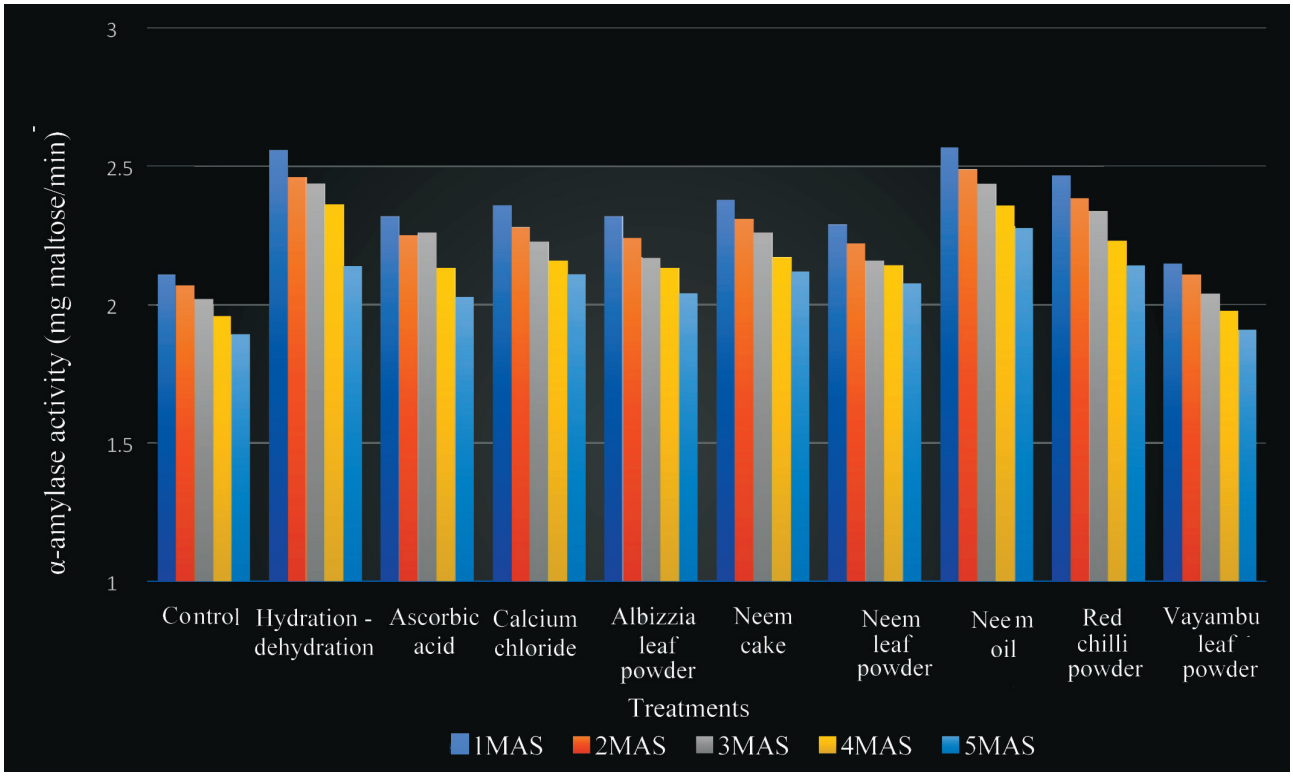
Results and Discussion

On application of midstorage invigoration treatments, six month old paddy seeds were subjected to biochemical analysis at monthly intervals. The results indicated that the storability of the treated seeds extended up to five months after the midstorage invigoration treatments. Under ambient storage conditions, significant variations



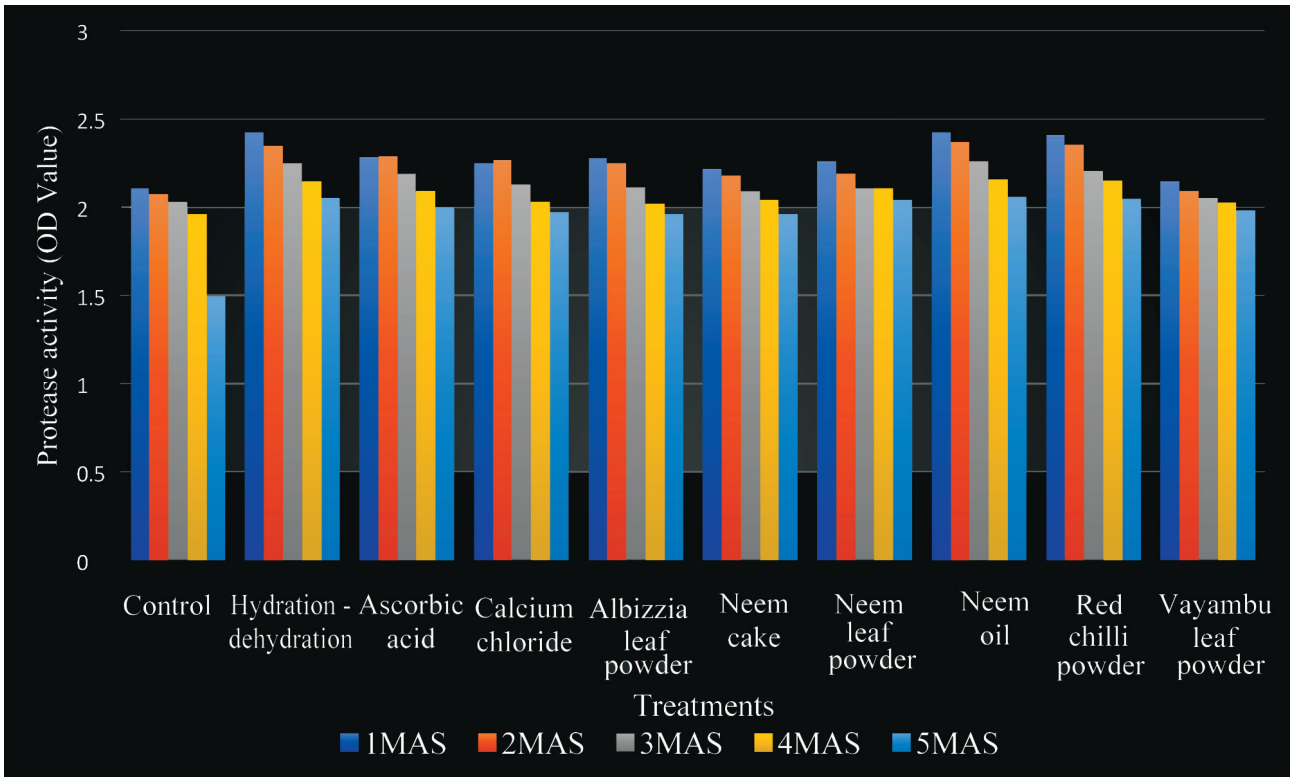
MAS- Months After Storage

Figure1. Impact of treatments on dehydrogenase activity under ambient storage condition in six month old treated seeds



MAS- Months After Storage

Figure 2. Impact of treatmentson α-amylase activity under ambient storage condition in six month old treated seeds



MAS- Months After Storage

Figure3. Impact of treatments on protease activity under ambient storage condition in six month old treated seeds

were noted in the biochemical parameters of treated paddy seeds. The enzymes dehydrogenase, alpha amylase and protease maintain the seed in an active metabolic state without losing the germination potential. Therefore the higher levels of enzymes contribute to improved storage potential for paddy seeds (Marthandan and Jerlin, 2017). The enzymes which help the storability of seeds were maximum in the seeds treated with neem oil, hydration dehydration treatment and red chilli powder.

Dehydrogenase activity of the treated seeds at the start of the storage recorded higher in hydration dehydration, neem oil, and red chilli powder treatments. At the end of storage, hydration-dehydration, neem oil and red chili powder registered higher values (0.396, 0.397 and 0.391 OD Value) (Fig.1). The mean dehydrogenase enzyme activity decreases with an increase in storage of period and age of seeds. Neem oil performed better than other treatment in maintaining seed quality measures, such as dehydrogenase activity, in sorghum seeds, as per Ram et al. (2020).The dehydrogenase enzyme activity is a good stable metabolic marker to estimate the degree of vigour in seedsand have positive association with vigor and viability of seeds. Irrespective of the months of storage after midstorage invigoration treatment, higher values of α-amylase activity (Fig.2) were observed in the seeds treated with neem oil, hydration dehydration and red chilli powder. Neem oil (2.28 mg maltose/min), hydration dehydration treatment (2.14 mg maltose/min) and red chilli powder (2.14 mg maltose/min) showed the higher α-amylase activity value at the end of the storage period. The á-amylase activity in the rice seed endosperm is closely linked to carbohydrate metabolism. The different pattern of seed vigor may due to lower respiratory activity in seeds (Galani et al., 2011). Antioxidant enzyme activity, such as α-amylase and dehydrogenase, declines with an increase in storage time, but provide an active indication of seed quality.

In ambient storage environment, the protease activity value of treated seeds decreased from the first month of storage to the last month of storage and were significantly different among treatments (Fig.3). At the end of the storage period, neemoil treated seeds registered a maximum value (2.06 OD Value) that was comparable to hydration dehydration and red chili powder treated seeds (2.05 OD Value). At the end of the storage period, all treatments showed a progressive loss of activity in the protein degrading enzyme protease. The protease in the treated paddy seed experienced adecline in its capacity to break down and digest proteins over the course of the storage period. This was in accordance with the results of Naik et al. (2017). As the age of the seed increased, the protease activity declined. The variation in protease activity of different midstorage treated seed provided baseline for selecting best midstorage invigoration treatment for better seed quality.

The present study was concluded that paddy seeds under ambient storage condition, subjected to mid storage invigoration with neem oil, hydration dehydration treatment and red chilli powder outperformed all other treatments in enhancing biochemical parameters of seed quality. Maintaining enzyme activity in seed storage ensures seeds in an active metabolic state. The enzyme activity in an efficient manner facilitates the mobilization of the seed reserves such as starch, protein etc and retains the seed in an effective germination phase. The midstorage seed invigoration treatments maintains the enzyme activity in the seed for improved seed quality and increased storage period compared to untreated seeds.

Conclusion

Midstorage invigoration treatments enhances biochemical processes, leading to higher levels of dehydrogenase, α-amylase, and protease enzymes. These enzymes maintain the seed in anactive metabolic state without losing the germination potential. Therefore the higher levels of enzymes contribute to improved storage potential for paddy seeds. In case of midstorage invigoration treatments, selection of cheap organic materials is a necessity. In paddy seed storage, it is worth considering the use of neem oil, hydration dehydration treatment and red chilli powder as midstorage seed treatments. Farmers can enhance seed quality and viability using these eco-friendly treatments. These cost-effective methods potentially increase storage duration and promote organic techniques without seed loss. These treatments are affordable, making them accessible to small-scale farmers. By adopting these methods, farmers can potentially increase seed quality and storage period. Organic techniques avoids synthetic chemicals and benefit seed health. Reduced reliance on agrochemicals helps minimize pollution and environmental impact. In concise, these treatments offer an accessible situation for farmers that provide improved seed quality, prolonged seed storage and a greener approach to agriculture.

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