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Influence of Seed Invigoration with Organic *Kunapajala* on Seed Quality and Biochemical Activity in Late Sown Wheat

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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Original Research Article

ABSTRACT

A laboratory experiment was conducted at GBPUA&T, Pantnagar during *rabi* season of 2020-2021 to study the influence of seed invigoration with organic *kunapajala* on germination and biochemical activity in wheat variety UP-2526. The experiment was laid in completely randomized design which consisted of five treatments viz; no seed priming (control), Hydropriming for 16 hours, *kunapajala* seed priming @10%, 25% and 50% for 16 hours. Results revealed that germination percentage in hydro primed seeds and 25% *kunapajala* primed seeds jointly recorded maximum germination

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percentage (99.7%) which was statistically at par with 10% *kunapajala* priming (99.3%). Seed reserve mobilization efficiency after 4th day and 8th day of incubation was significantly affected by seed invigoration treatments. 25% *kunapajala* primed seeds recorded significantly higher dehydrogenase activity (0.68 OD) which was at par with hydroprimed seeds and 10% *kunapajala* primed seeds (0.63 and 0.62 OD, respectively). So, seed priming with either 10% or 25% organic kunapajala is an effective method to bring about improvement in germination of wheat and predict its field performance.

Keywords: Seed germination; seedling vigour; seed priming; wheat.

1. INTRODUCTION

Wheat is second most important food crop in India and occupies an area of about 31.6 million hectare with total production of 103.88 million tonnes and shares 13% of total production of world [1]. Regardless of its high production, the productivity of wheat is declining due to many abiotic stresses such as low temperature during early growth. In India, around 13.5 million hectare of wheat is grown under heat stressed. Globally, India is the second largest producer of wheat contributing 14.0% in terms of area and 13.6% in production [2]. With increase in population, there is shrinkage on cultivable land. Therefore, the main focus would be on increasing the productivity by adopting improved cultivation practices. Crop establishment is an important factor that depends upon optimum plant population and uniform emergence. Proper crop establishment depends upon quality of the seed in terms of its germination and seedling vigour [3]. Planting time, seed quality material and supply of optimum nutrients during crop growth are some of the important parameters that limit productivity. These elements decide crop establishment, growth and development, which thus decide yielding capability of the harvest. Wheat is predominantly grown during rabi season and has more wider planting window. In the north western plain zone of India, it is planted in second fortnight of October as early planting, first to third week of November as optimum sowing time and up to fourth week of November to end of December. It is always a challenge to achieve desirable yield potential and grain quality under both normal as well as late sowing condition. In normal sowing condition, poor quality seed material and inadequate soil moisture and nutrients may cause reduction in yield potential and seed quality. On the other hand, heat stress both low and high temperature are major reason of declining yield in late sown wheat. Prediction of seed germination, seedling vigour and biochemical activities in seed before sowing is of prime importance for assured yield in late sown condition.

Seed priming is a process of regulating the process germination by managing the temperature and seed moisture content and is the most widely used seed invigoration technique improve field emergence and to crop establishment under adverse environmental condition. Hydropriming is one of the oldest practices used for seed invigoration. This practice has been used widely to stimulate the speed, uniformity of emergence and improve the final plant stand. Nowadays, researchers are focusing more on eco-friendly seed invigoration techniques to improve seed germination and uniform crop establishment. Vrikshayurveda, a part of Ayurvedic history is an ancient science of plant life which deals with healthy growth of plants and its productivity. In vrikshayurveda, herbal based kunapajala is used to enhance the biological efficiency of crop as well as soil. The manure kunapajala (jala=water) or kunapambu, is derived from the Sanskrit word 'kunapa' meaning "smelling like a dead body, stinking" and it is a fermented liquid manures prepared from flesh, animal urine, marrow, etc. [4]. Nowadays, herbal based kunapajala is used as nourishment of seed, crop and soil. It is very effective in nourishing plants at various stages and can be used as a seed priming technique. Seed priming with either 10% or 25% herbal based kunapajala was found to be an ecofriendly technique that improve emergence, seedling development and biochemical activity of wheat over hydropriming and no priming [3]. Therefore, the experiment was conducted to evaluate the influence of herbal kunapajala concentration on seed quality and biochemical parameters of wheat seed.

The standard germination test is considered as the common test for evaluating seed quality particular seed performance under ideal environmental conditions. However, seed lots may also differ in field emergence potential due to various biochemical activities and hormones. Thus, seed germination test along with enzyme activities is considered powerful when ranking the seed lots based on vigour level groups. Therefore, germination test, dehydrogenase activity in seeds can bring more precise solution to predict field emergence. According to Indian Minimum Seed Certification Standards (IMSCS, 2013), final count of wheat seedling evaluation is 8th day for its germination per cent. With this key points, the present study was carried out to correlate germination per cent with biochemical activity to assess the seed vigour and planting value of late sown wheat variety UP2526.

2. MATERIALS AND METHODS

The laboratory study was conducted at seed physiology laboratory, Department of Agronomy, College of Agriculture, G. B. Pant University of Agriculture and Technology, Pantnagar (29°1' N latitude. 79°29'E longitude and elevation of 231 m above MSL) during rabi season of 2020-21. The experiment was laid out in completely randomized design with five treatments viz; T1: No seed priming, T₂: Seed hydropriming, T₃: 10% seed priming with herbal kunapajala, T₄: 25% seed priming with herbal kunapajala and T_5 : 50% seed priming with herbal kunapajala with six replications. Seed priming was for 16 hours duration with tap water (T2) and herbal kunapajala in T_3 , T_4 and T_5 at their respective concentrations (10, 25 and 50%, respectively). Wheat seed variety UP-2526 was soaked in respective solutions in 1: 2: seed: priming media ratio and seeds were shade dried up to original moisture content (11.8%). Germination per cent was counted [5] at 8th days, root length, shoot length [6], seed reserve mobilization efficiency [7] 8th days after incubation and biochemical parameter like dehydrogenase activity of wheat seeds [8], were evaluated.

In order to assess and compare the quality attributes, preparation of herbal kunapajala and its components used in this study has been adopted based on the recommendation of Y.L. Nene, a modified version of the ancient preparing formulation [9-10]. For herbal kunapajala, the ingredients (10 kg cow dung, 10 L cow urine, 2 L sour butter milk, 2 kg jaggery, 2 kg sprouted urd, 2 kg mustard cakes, 3 L rice bran water, 1 L fresh milk, 3-4 pieces of dry cow dung, 10 kg nettle grass and 10 kg leaves of other grasses) were mixed into a plastic drum of 200 L capacity and the volume of solution was made up to 200 L with water. The contents were mixed thoroughly with a stick and the lid was closed. Twice a day (morning and evening) the contents were mixed until the disappearance of bubbles in the drum. Lack of bubble formation in

the contents indicates that the formulation preparation was complete and can be used for the experiment. Herbal *kunapajala* was ready and was sieved properly with cloth for further use.

2.1 Seed Physiological Test

Germination test was conducted as per standard procedure for germination as described in International Rules for Seed Testing [11]. It was calculated on 8th day on the basis of number of normal seedlings emerged divided by total number of seeds taken into hundred.

For seed reserve mobilization efficiency, fifty seeds of wheat were placed in petri dishes having two moist germination papers. The seeds were then covered with lid of petri dishes. The petri dishes were placed in an incubator for germination maintained at 20±2°C temperature. Five seedlings were randomly selected from each petri dishes at 4th and 8th day after incubation. Then, their root and shoot axes and (endosperm) seed were separated. The separated parts were dried in an oven maintained at 70±2°C for 48 hours. The dry weight of the samples were determined on the attainment of constant weight by drying in oven.

Amount of seed material lost as respired (SMLR) was calculated as follows:

SMLR= SDW-(SHW+RTW+RSW)

Where, SDW= Seed dry weight before germination (g), SHW= Shoot dry weight (g), RTW= Root dry weight (g), RSW= Remaining seed dry weight (g)

Seed Reserve Mobilization Efficiency (SRME) was calculated using the following formula [12].

SRME= (SHW + RTW)/SMLR

2.2 Biochemical Parameter

2.2.1 Dehydrogenase activity

Dehydrogenase activity was measured as per the standard procedure given by Kittock and Law [8]. Fifty seeds were soaked in a beaker of 50 mL kept at $25\pm1^{\circ}$ C for overnight in order to excise the embryonic axes easily. Then 10 embryonic axes in three replications were kept in 1 mL of tetrazolium solution (1% solution) and incubated in dark at 30^oC for 4 hours. Drained out the excess solution and washed coloured embryonic axes in distilled water twice. Thereafter these axes were soaked the axes in 10 ml of methyl cellosolve until axes became colourless. The intensity of colour developed was read at 480 nm in a UV-VIS Spectrophotometer. Methyl cello solve was used as blank.

2.3 Statistical Analysis

The study was set up in Completely Randomized design with six replications and the data was analyzed by using OPSTAT, designed and developed by O.P. Sheoran, Professor Statistics, COBS&H CCS HAU, Hisar. Standard error of means (SEm±) was computed. The critical difference at 5% level of probability was calculated for testing the significance of difference between any two means whenever the F test was found significant.

3. RESULTS AND DISCUSSION

All the physiological and biochemical parameters of wheat seeds were significantly affected by different seed invigoration treatments.

3.1 Effect of Seed Invigoration on Physiological Parameters

In the present experiment, hydro primed seeds and 25% *kunapajala* primed seeds jointly recorded maximum germination percentage

(99.7%) which was statistically at par of 10% kunapajala priming (99.3%) (Fig. 1). Hydropriming and seed priming with herbal kunapaiala were beneficial due to advancement of reactions leading to faster germination and stimulation of biochemical potential of cell and repair mechanisms triggered in seeds after 1st day, 2nd day and 4th day after incubation (Fig. 2). Faster production of germination metabolites and better genetic repair during hydration seems the basis of improved germination rate. The results are in line with the findings that seed priming with 10% herbal kunapajala being at par with hydropriming (87.3%). recorded significantly hiaher germination percentage (90.7%) in chickpea [13].

The result indicated in Fig. 3 revealed that seed reserve mobilization efficiency after 4th day and 8th day of incubation significantly affected by seed invigoration treatments. After 4th day, the highest seed reserve mobilization efficiency was recorded with 10% herbal kunapajala priming (2.99 g/g) which was shown statistically at par with 25% herbal kunapajala priming (2.98 g/g) as well as hydropriming (2.80 g/g) whereas the lowest values were recorded with control (1.92 g/g) but at par with 50% kunapajala priming (2.00 g/g). On 8th day after incubation, significant seed mobilization efficiency was recorded in 10% kunapajala priming (0.96 g/g) but at par with hydropriming (0.92g/g) and lowest recorded from no priming treatment (0.45 g/g). Higher seed reserve mobilization efficiency indicates that higher efficiency in seeds as loss in respiratory

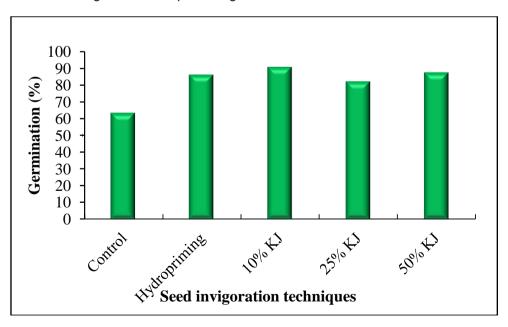
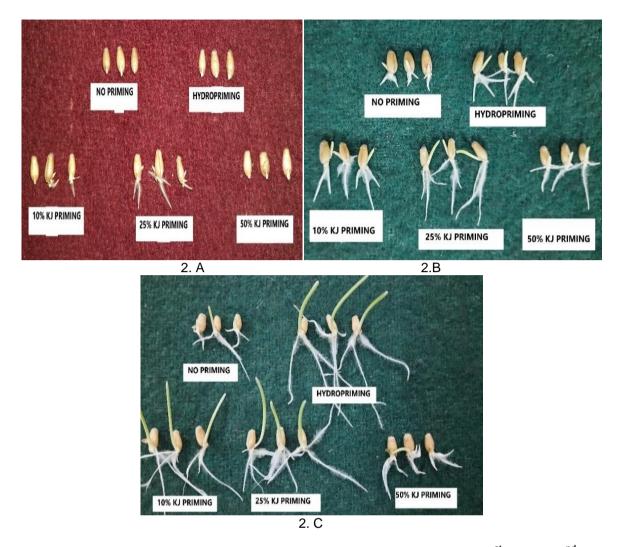


Fig. 1. Effect of seed invigoration treatments on germination % of wheat (KJ: kunapajala)



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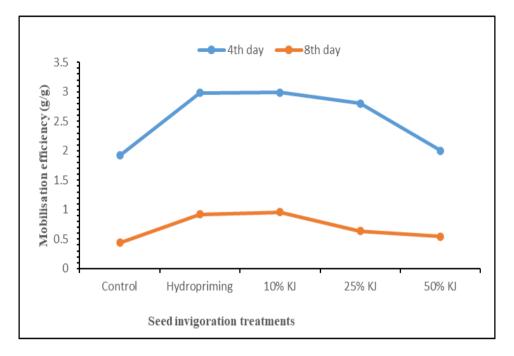
Fig. 2. Effect of seed invigoration treatments on germinatio of wheat after 1st day (A), 2nd day (B) and 4th day after incubation (C)

was minimum and large seed reserves would be used for producing seedlings. The rate of decrease in seed reserve mobilization efficiency was 42.7% in 50% *kunapajala* priming as compared to 10% kunapajala priming on 8th day after incubation. This reduction in 50% *kunapajala*, could be due to lesser uptake of water by seed and decrease the gibberellic acid concentration and other hydrolytic enzymes during germination [14].

Dehydrogenase activity (OD) was significantly influenced by seed invigoration treatments (Fig. 4).

Data from Fig. 4 revealed that 25% *kunapajala* primed seeds recorded significantly higher dehydrogenase activity (0.68 OD) which was at par with hydroprimed seeds and 10% *kunapajala* primed seeds (0.63 and 0.62 OD, respectively).

The lowest value were recorded from 50% kunapajala primed seeds along with control which was statistically found similar (0.44 and 0.4 OD, respectively). Seed germination is an amphibolic process where both catabolic and anabolic processes takes place in the embryonic axes. Catabolic process is controlled by dehydrogenase of group enzymes. Dehydrogenase enzyme activities indicate the level of viability in seed as its component of ETC system which facilitated the electron transport from a substrate to oxygen necessary for the respiratory process. The similar results are also in agreement in wheat that hydropriming priming improved the dehvdrogenase enzymes (20µg/100 seeds) as compared with untreated seeds [15]. Thus, dehydrogenase enzymes improve metabolism of seed reserves by activating hydrolytic enzymes during seed priming [16].



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Fig. 3. Seed reserve mobilization efficiency in wheat seeds (KJ: kunapajala)

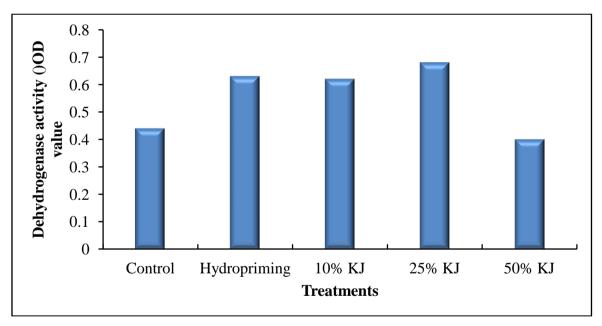


Fig. 4. Dehydrogenase activity as affected by seed invigoration treatments (KJ: kunapajala)

4. CONCLUSION

From the above findings, it may be concluded that seed invigoration with either 25% herbal based *kunapajala* may be an eco-friendly technique that can be used to improve seed quality and biochemical activities of wheat seeds.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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