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ENHANCING SEED GERMINATION AND VIGOR IN RICE, BLACKGRAM, AND BHENDI THROUGH ULTRASONIC TREATMENT: A STUDY OF OPTIMAL TREATMENT DURATIONS AND THEIR IMPACT ON SEED QUALITY PARAMETERS

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ABSTRACT
Seed dormancy, a natural adaptation that prevents premature germination under adverse conditions, poses significant challenges for agricultural productivity. This study evaluates the effects of ultrasonic treatment—a non-chemical, high-frequency sound wave technique—on seed germination and vigor in rice, blackgram, and bhendi. Seeds were exposed to ultrasonic waves at 40 kHz and 43°C for durations of 10, 20, 30, and 40 minutes. Germination percentage, root and shoot lengths, dry matter accumulation, vigor indices, and speed of germination were assessed. Results revealed significant improvements in all seed quality parameters, particularly at 40 minutes of exposure, with germination rates reaching 98% (rice), 91% (blackgram), and 83% (bhendi). Enhanced root/shoot development and seedling biomass also correlated positively with exposure time. These findings demonstrate the effectiveness of ultrasonication in breaking seed dormancy and enhancing early seedling growth. As a simple, eco-friendly alternative to chemical treatments, ultrasonic seed priming holds promise for improving crop establishment and yield in sustainable agriculture. Future research should focus on optimizing treatment parameters and assessing field-level performance.

KEYWORDS seed dormancy, ultrasonic waves, seed priming, germination, vigor index, sustainable agriculture, rice, blackgram, bhendi

INTRODUCTION
Ensuring the germination percentage for proper crop establishment to increase the yield and production, should be the primary goal. Most of the crops, with their dormancy, reduce the plant population and crop establishment and ultimately the yield. Seed dormancy is a natural mechanism that prevents seeds from germinating under unfavourable conditions. While this adaptation is crucial for plant survival in the wild, it can pose challenges in agriculture where rapid and uniform germination is desired. Various methods have been developed to break seed dormancy, and among these, the use of ultrasonics has gained attention in recent years.

One of the major reasons for physical dormancy include the presence of hard cell wall which prevents the penetration of water and oxygen into the cells and ultimately reduced seed germination percentage. Ultrasonic seed treatment is one of the physical approaches to break seed dormancy and improve germination rate. These high-frequency sound waves when applied to seeds, can induce various physical and biochemical changes that help overcome dormancy and promote germination. By harnessing the power of high-frequency sound waves, this method offers a non-chemical means to enhance germination and seedling establishment. As research in this field continues to advance, ultrasonic treatment may become an increasingly important tool in modern agriculture, helping to ensure rapid, uniform crop establishment and potentially contributing to improved yields and food security.

The advantages of the ultrasound technique are that it is simple, inexpensive, and environmentally friendly (Goussous et al., 2010). Ultrasound technology can also be used in the solid-phase microextraction and the detection of bioactive compounds in plants (Asfaram et al., 2019). Researchers have discussed the use of ultrasound technology to stimulate germination of various seeds such as maize, barley, rice, and sunflower seeds and emphasized its importance (Ding et al., 2018; Rifna et al., 2019). Liu *et al.,* (2016) (Liu et al., 2016) demonstrated that the use of ultrasound waves increased and improved the growth of tall fescue (*Festuca arundinacea*) and Russian wild rye (*Psathyrostaehys juncea* Nevski) seedlings. Likewise, germination of sesame (*Sesamum indicum* L.) seeds treated with ultrasound waves showed increased germination and growth (SHEKARI et al., 2015). Similarly, a research was performed to determine the effect of ultrasonic treatment on seed germination and seedling emergence in seeds of watermelon, melon, leek, pepper, carrot, tomato, and aubergine (Memiş et al., 2022).

Considering the above mentioned things, the present study was therefore conducted to observe the effects of ultrasonic seed treatment in reducing seed dormancy and to ascertain the appropriate duration of ultrasonic treatment on seed quality parameters in rice (*Oryza sativa*), blackgram (*Vigna mungo* L. Hepper), and bhendi (*Abelmoschus esculentus*).

Achieving rapid and uniform germination is a critical prerequisite for successful crop establishment, which directly influences yield and productivity. However, seed dormancy, a naturally evolved survival mechanism, frequently impedes agricultural efficiency by reducing plant population and uniform emergence. One of the key contributors to physical dormancy is the impermeable seed coat, which restricts water and oxygen uptake.

Recent advancements have highlighted the potential of ultrasonic seed treatment as a physical, non-chemical method to overcome dormancy. Ultrasonication involves the application of high-frequency sound waves, which induce both mechanical and biochemical changes in seed structure, thereby enhancing germination and vigor. Notably, this approach is simple, cost-effective, and environmentally benign (Goussous et al., 2010).

Several studies have reported positive effects of ultrasonication on various crops, including maize, barley, rice, and sunflower (Ding et al., 2018; SHEKARI et al., 2015; Liu et al., 2016). Given the increasing need for sustainable seed enhancement techniques, this study investigates the efficacy of ultrasonic treatment on rice, blackgram, and bhendi seeds, aiming to determine optimal treatment durations and their effects on key seed quality parameters.

MATERIALS AND METHODS

Collection of seeds

Seeds of rice (*Oryza sativa*), blackgram (*Vigna mungo* L. Hepper), and bhendi (*Abelmoschus esculentus*) were received from the Department of Seed Science and Technology, Department of Pulses and Department of Vegetable Science, TNAU, Coimbatore, respectively. These seeds contributed the base material of research work.

Seed treatment

Seeds were subjected to Ultrasonic treatment was conducted using a GT Sonic ultrasonic machine operating at 40 kHz and 43°C for 10, 20, 30, and 40 minutes. present at the Department of Plant Biotechnology, CPMB, TNAU, Coimbatore.

Germination test

After imposing seed treatment, seeds were analyzed for their physiological seed quality parameter at the Department of seed Science and Technology, Tamil Nadu Agricultural University, Coimbatore.

Germination % (ISTA, 2013)

The germination test was performed by inclined plate method using four replicates in each fraction of treatment in a germination room maintained at 25 ± 2°C temperature and 95 ± 5% Relative Humidity (RH) illuminated with fluorescence light. After final count days (for paddy-14 days, for black gram- 7 days and for bhendi-21 days) the normal seedling were counted in each replication and a mean number of normal seedlings were arrived and based on the mean number, the mean germination was calculated and expressed in percentage (%) (ISTA, 2013).

Root length

Per replication, ten normal seedlings were taken at the final count day of germination test randomly and the root length was calculated by measuring the distance between the collar and the tip of the root and mean value was expressed in centimeter (cm) (ISTA, 2013).

Shoot length

In order to determine the length of the shoots, seedlings were measured between collar and tip of the primary leaves and mean values were expressed in centimeter (cm) (ISTA, 2013).

Dry matter Production

Ten normal seedlings after seedling measurement were ensured for removal of cotyledon and then placed in a brown paper cover and kept for shade drying. After 24 hours, they were dried in hot air oven maintained at 80°C for 1 hour. After the end of time period, cover containing seedlings were cooled in a desiccator for 30 minutes and then the dried seedlings were weighed in an electronic balance and expressed in g 10 seedlings^{-1} (Gupta, 1993).

Vigour Index

Vigour Index values were estimated using the following formulae suggested by Abdul-Baki and Anderson (1973) and expressed as whole number.

Vigour Index I = Germination % x Total Seedling length (cm)

Vigour Index II = Germination % x Dry matter production (g 10 seedlings^{-1})

Speed of germination (Maguire, 1962)

After the ultrasonic treatment, seeds were put forth to check speed of emergence on sand tray.

The speed of germination was assessed from the first count day of each crop and continued till last count day.

Crop	**First count (Days)**	**Final count (Days)**
Rice	5	14
Black gram	4	7

Bhendi	4	21

The seeds showing radicle protrusion were counted daily after first count day pertaining to each crop until their respective final count. From the number of seeds germinated on each day, the speed of germination was calculated using the following formula

$$\text{Speed of germination} = \frac{X_1}{Y_1} + \frac{X_2 - X_1}{Y_2} + \ldots + \frac{X_n - X_{n-1}}{Y_n}$$

Where,
X_1 - Number of seeds germinated at first count
X_2 - Number of seeds germinated at second count
X_n - Number of seeds germinated on n^{th} count
Y_1 - Number of days from sowing to first count
Y_2 - Number of days from sowing to second count
Y_n - Number of days from sowing to n^{th} count

3.3. Statistical Analysis

In order to determine significance of the aforementioned parameters, the data were analyzed using analysis of variance as per Panse and Sukhatme (1954). The percentage values were transformed into arcsine values before statistical computation. Using a 5% probability level (P=0.05), the critical difference (CD) was calculated, and non-significant values are denoted as 'NS'.

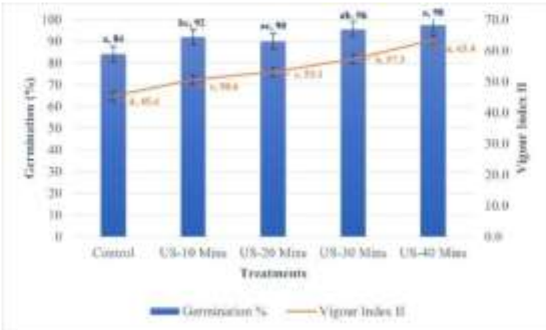

Fig 1. Effect of Ultrasonic treatment on seed germination (%) and seedling vigour index II in rice under laboratory condition (US-Ultrasound wave treatment). Error bars above means were the standard error of four replicates. Bars and lines sharing a common letter did not differ significantly at p < 0.05.

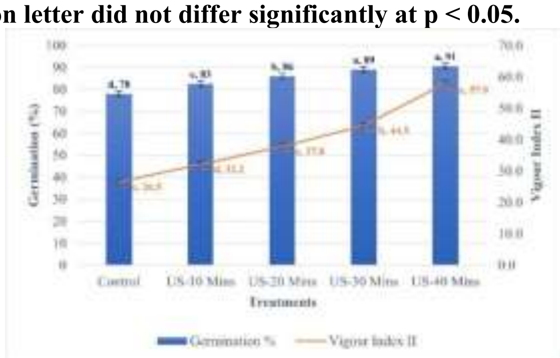

Fig 2. Effect of Ultrasonic treatment on seed germination (%) and seedling vigour index II in black gram under laboratory condition (US-Ultrasound wave treatment). Error bars above and below means were the standard error of four replicates. Bars and lines sharing a common letter did not differ significantly at p < 0.05.

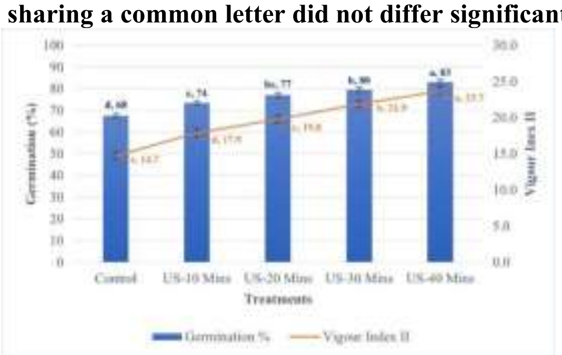

Fig 3. Effect of Ultrasonic treatment on seed germination (%) and seedling vigour index II in bhendi under laboratory condition (US-Ultrasound wave treatment). Error bars above means were the standard error of four replicates. Bars and lines sharing a common letter did not differ significantly at p < 0.05.

Results

Ultrasonication increased germination percentages significantly compared to control in all three crops ($P<0.05$). The highest germination percentage of 98 %, 91% and 83% were observed in the seeds treated with ultrasound wave for 40 minutes (T_4) in rice, blackgram and bhendi respectively followed by the seeds treated with ultrasound wave for 30 minutes (T_3) with the germination percentage of 96% in rice and 89% in blackgram and 80% in bhendi (Fig. 1, Fig. 2 and Fig. 3). Results of the germination test demonstrated that germination capacity improved progressively with longer exposure to ultrasonic waves across all crops, indicating a positive correlation between treatment duration and germination percentage. Root and shoot lengths also showed improvement with longer ultrasonic treatments, indicating enhanced early seedling growth. With the highest root and shoot length was observed in with ultrasound wave treatment for 40 minutes (T_4) in all crops. Ultrasonication accelerated the germination process, with the fastest germination observed in T_4 treatment across the studied crops where highest speed of germination of 3.05, 3.20 and 3.45 were observed in rice, blackgram and bhendi respectively (Table 1, Table 2 and Table 3).

Dry matter production was recorded highest in the T_4 treatment in rice, blackgram and bhendi with the values of 0.653, 0.640 and 0.285 g/10 seedlings respectively followed by T_3, T_2 and T_1 across all crops (Table 1, Table 2 and Table 3).

Fig. 1, Fig. 2 and Fig. 3 shows that the vigour index II increases progressively with increase in duration of treatment and is highest in ultrasound wave treatment for 40 minutes (T_4) across all crops studied.

From tables 1, 2 and 3, it is evident that the highest vigour index I was recorded in the seeds treated with ultrasound wave treatment for 40 minutes (T_4) followed by the treatment duration of 30 minutes (T_3).

RESULTS AND DISCUSSION

The study investigated the effects of ultrasonic treatment on seed germination and seedling vigor in rice, blackgram, and bhendi. The findings revealed a significant enhancement in germination percentages across all three crops when compared to control groups, with statistical significance at $P<0.05$. Notably, the highest germination percentages were observed with ultrasound waves treatment for 40 minutes (T_4), yielding germination percentages of 98%, 91%, and 83% for rice, blackgram, and bhendi respectively. A shorter treatment duration of 30 minutes (T3) also showed better results compared to the control (T_0) with germination percentages of 96% for rice, 89% for blackgram, and 80% for bhendi (Fig. 1, 2 and 3). The data illustrated a clear trend: longer exposure to ultrasonic waves positively correlated with increased germination capacity. This enhancement suggests that ultrasonication effectively breaks seed dormancy by inducing both physical changes, such as microfractures in the seed coat, and biochemical modifications, such as enhanced enzyme activity within the seeds (Kavak, n.d.). Research has demonstrated that ultrasonically treated seeds show a higher germination percentage compared to untreated seeds. For example, treatments at specific frequencies and durations have resulted in significant increases in germination rates across various species, including barley and rice (Foschi et al., 2023; Nazari & Eteghadipour, 2017). The effectiveness of ultrasonication is often comparable to or exceeds traditional methods such as hydropriming, making it a valuable technique in seed technology (Memiş et al., 2022). Previous research supports this notion, indicating that ultrasonic waves facilitate water penetration and improve oxygen availability within seeds, both of which are critical for successful germination (Huang et al., 2024).

Ultrasonic treatment not only improved germination rates but also accelerated the germination process across all crops. The maximum speed of germination values was recorded at 3.05 for rice, 3.20 for blackgram, and 3.45 for bhendi when the seeds were treated with ultrasound waves for 40 minutes (T_4) (Table 1, 2 and 3). This acceleration indicates that prolonged exposure times not only enhance germination rates but also promote quicker seedling emergence, which is essential for uniform crop establishment. Rapid germination is particularly advantageous in agricultural practices where timely planting is crucial to mitigate adverse environmental conditions (Finch-Savage & Bassel, 2016).Enhanced germination speed reflects ultrasonication's potential to synchronize seedling emergence, reducing variability and promoting robust stand establishment, as corroborated by similar findings in sunflower and sesame seeds (SHEKARI et al., 2015; Souza et al., 2022)

Root and shoot lengths serve as vital indicators of early seedling vigor, and these parameters improved significantly with increased ultrasonic exposure. The longest root and shoot lengths were recorded in the T_4 treatment across all crops (Table 1, 2 and 3), underscoring the stimulatory effects of prolonged ultrasonic exposure on early seedling growth. Such enhancements can be attributed to physical and biochemical alterations induced by ultrasonic waves, including cell wall loosening and improved nutrient absorption (Memiş et al., 2022). Enhanced root development is critical as it establishes a robust foundation for nutrient uptake and overall plant health during early growth stages. The trends observed in root and shoot elongation are consistent with earlier research emphasizing the role of ultrasonication in promoting cellular activity and structural changes within plant tissues, leading to increased growth rates (Karagöz & Dursun, 2021).

Dry matter production is a key measure of seedling robustness, with the highest values observed in the T_4 treatment: 0.653 g/10 seedlings for rice, 0.640 g/10 seedlings for blackgram, and 0.285 g/10 seedlings for bhendi. This increase aligns with improvements in root and shoot growth, reflecting an overall enhancement in seedling vigor under prolonged ultrasonic treatment. The consistent rise in dry matter production across treatments highlights the potential of ultrasonic treatment to not only improve germination but also subsequent growth performance (Huang et al., 2020).

From tables 1, 2 and 3 it is evident that the vigor index I was highest in seeds treated with ultrasound waves for 40 minutes (T_4), followed by those treated for 30 minutes (T_3). Similarly, Fig. 1, 2 and 3 showed a progressive increase in vigor index II with longer treatment durations, peaking in the T_4 group across all studied crops. The vigor indices calculated indicate that seeds treated with longer durations of ultrasound not only germinated more effectively but also produced healthier seedlings capable of better survival rates post-emergence. The results indicate that ultrasonication not only improves germination and growth parameters but also ensures the development of vigorous seedlings capable of better establishment under field conditions (Huang et al., 2024; Memiş et al., 2022).

CONCLUSION

The study demonstrates the efficiency of ultrasonic treatment as a non-chemical strategy for enhancing seed germination and seedling vigor in crops such as rice, blackgram, and bhendi. Prolonged exposure, particularly for 40 minutes, resulted in significant increases in germination percentage, germination speed, root and shoot growth, dry matter production, and vigor indices. The results obtained are consistent with prior studies showing that ultrasonication improves seed performance in crops such as tall fescue, sesame, and sunflower (Huang et al., 2024; Memiş et al., 2022).

Ultrasonication is a sustainable and ecologically sound alternative to traditional chemical treatments, coinciding with modern agricultural practices that aim to reduce costs while limiting the impact on the environment. Its ability to break seed dormancy and promote uniform germination enhances crop establishment and yield potential. The positive outcomes observed underscore the potential of this technique for improving seed quality across various species.

Future study should focus on improving ultrasonic treatment parameters such as exposure duration and frequency for individual crops. Furthermore, investigating the long-term impacts of ultrasonication on field performance, yield, and tolerance to biotic and abiotic stresses may give useful insights into its practical uses in agriculture. Ultrasonication, a simple and eco-friendly approach, has the potential to shift seed treatment practices and contribute significantly to sustainable agricultural development.

In conclusion, ultrasonic treatment has emerged as an effective non-chemical approach for improving seed performance. The significant impacts found with longer treatment durations highlight the necessity of optimizing exposure times for different crops. This method not only promotes faster germination but also supports higher yields, making it a promising tool in modern agricultural practices.

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Table 1. Effect of ultrasonic treatment on seed quality parameters in rice

Treatment	Speed of Germination	Root length (cm)	Shoot length (cm)	Dry matter Production (g/10 seedling)	Vigour Index I
T_0	0.24^{e}	12.7^{c}	4.5^{e}	0.544^{c}	1448^{c}
T_1	1.10^{d}	12.9^{bc}	4.8^{d}	0.553^{c}	1696^{b}
T_2	2.62^{c}	13.1^{b}	5.3^{c}	0.609^{d}	1593^{b}
T_3	2.78^{b}	13.8^{a}	5.7^{b}	0.592^{b}	1858^{a}
T_4	3.05^{a}	14.0^{a}	6.1^{a}	0.653^{a}	1962^{a}
Mean	1.96	13.29	5.30	0.590	1709
SEd	0.0201	0.1602	0.1125		49.8662
CD (P=0.05)	0.0428	0.3415	0.2399	0.0131	106.2884

(Means with different letters in the same column were significantly different at 5%)
Treatment DetailsT_0 - Control
T_1 - Ultrasound waves treatment for 10 minutes
T_2 - Ultrasound waves treatment for 20 minutes
T_3 - Ultrasound waves treatment for 30 minutes
T_4 - Ultrasound waves treatment for 40 minutes

Table 2. Effect of ultrasonic treatment on seed quality parameters in black gram

Treatment	Speed of Germination	Root length (cm)	Shoot length (cm)	Dry matter Production (g/10 seedling)	Vigour Index I
T_0	0.64^{e}	17.1^{c}	15.9^{c}	0.340^{e}	2570^{e}
T_1	1.10^{d}	17.3^{bc}	16.5^{bc}	0.390^{d}	2785^{d}
T_2	2.25^{c}	17.3^{bc}	17.1^{b}	0.440^{c}	2962^{c}
T_3	2.60^{b}	17.8^{b}	17.1^{b}	0.500^{b}	3108^{b}
T_4	3.20^{a}	19.8^{a}	19.9^{a}	0.640^{a}	3596^{a}
Mean	1.96	17.9	17.3	0.462	3004
SEd	0.0320	0.2632	0.3371	0.0055	49.4693
CD (P=0.05)	0.07	0.5609	0.7186	0.0117	105.4422

(Means with different letters in the same column were significantly different at 5%)
Treatment Details
T_0 - Control
T_1 - Ultrasound waves treatment for 10 minutes
T_2 - Ultrasound waves treatment for 20 minutes
T_3 - Ultrasound waves treatment for 30 minutes

T_4 - Ultrasound waves treatment for 40 minutes

Table 3. Effect of ultrasonic treatment on seed quality parameters in bhendi

Treatment	Speed of Germination	Root length (cm)	Shoot length (cm)	Dry matter Production (g/10 seedling)	Vigour Index I
T_0	1.4^{e}	14.8^{c}	10.9^{b}	0.218^{e}	1730^{d}
T_1	2.16^{d}	16.7^{b}	11.0^{b}	0.243^{d}	2037^{c}
T_2	3.04^{c}	16.9^{b}	11.1^{b}	0.257^{c}	2154^{bc}
T_3	3.32^{b}	17.0^{b}	11.3^{b}	0.275^{b}	2242^{b}
T_4	3.45^{a}	18.3^{a}	11.9^{a}	0.285^{a}	2501^{a}
Mean	2.68	16.72	11.2	0.256	2133
SEd	0.0462	0.1641	0.1765	0.0037	57.6844
CD (P=0.05)	0.0985	0.3497	0.3763	0.0078	122.9526

(Means with different letters in the same column were significantly different at 5%)

Treatment Details

T_0 - Control
T_1 - Ultrasound waves treatment for 10 minutes
T_2 - Ultrasound waves treatment for 20 minutes
T_3 - Ultrasound waves treatment for 30 minutes
T_4 - Ultrasound waves treatment for 40 minutes