




Augmenting thermo-hydro-chemical seed priming and germination dynamics on genotype/cultivar-Arka Shubra: a spineless green manure legume cover crop *Mucuna pruriens* L. DC. var. utilis

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Abstract

The exploration of seed priming techniques represents a critical avenue for enhancing agricultural productivity, particularly in leguminous cover crops like *Mucuna pruriens* L. DC. var. utilis is commonly known as the velvet bean. This spineless green manure legume has significant potential for sustainable agricultural systems, offering multiple ecosystem services including soil fertility improvement, erosion control, and nitrogen fixation. Over 2022–2023, an experiment was conducted to evaluate the impact of diverse seed treatments on the germination dynamics and seedling growth of trichomeless (spineless) *Mucuna pruriens*. The experiment was designed in an RCBD with three replications, occurred under shaded conditions. Various treatments were applied, including scarification, soaking in hot and normal water, H₂SO₄, KNO₃ + HNO₃, GA₃, and thiourea. The results indicated that H₂SO₄ treatment significantly enhanced the germination rate (57.14% ± 2.49%) and reduced the mortality rate (42.86% ± 2.49%) compared with the other treatments. Additionally, the H₂SO₄ treatment exhibited higher values for VI I (2.91 ± 0.21) and VI II (51.18 ± 4.41) than the other treatments. Furthermore, the application of H₂SO₄ resulted in a greater seedling drying weight (0.90 ± 0.04 g) in comparison to other treatments, with GA₃ showing similar trend at 0.82 ± 0.02 g. The observed PCA plot indicates that PC-1 and PC-2 collectively explained 96.17% of the total variation. These findings highlight the effectiveness of H₂SO₄ in promoting favorable germination and seedling growth characteristics in spineless *Mucuna pruriens*.

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Introduction

The velvet bean is a member of the *Fabaceae* family and commonly referred to as '*M. pruriens* L. DC. var. utilis', is an annual climbing leguminous, green manure, legume, and land cover crop plant^[1–4]. *M. pruriens* (L.) DC. grows wild in India^[5–7] and thrives in tropical and subtropical regions of America, Africa, Asia, the Pacific Islands, and India^[8–11]. It has 150 species worldwide, including 15 documented in India. *M. pruriens* is a medicinally valuable crop^[12,13]. In India, two main species of *M. pruriens* (L.) DC. are commonly found: *M. pruriens* var. *pruriens* and *M. pruriens* var. *utilis*. *M. pruriens* var. *pruriens*, a wild variation with a black seed coat, has reddish-brown irritating trichomes on the pod that produce severe itching when in contact with the skin^[14].

Human contact with *Mucuna pruriens* (L.) DC. leads to itchy dermatitis attributed to *mucuna* production^[15]. Due to this, farmers were hesitating mainly in terms of cultivation and toughness in harvesting this crop. The other varieties, like CIM-Ajar, CIM-Nirom (released by CSIR-CIMAP, Lucknow), and Arka Shubra^[13,14,16,17], possess non-irritating trichomes, and their distinctive velvety appearance is due to the dense silky trichomes, earning them the common name 'velvet beans'^[12,18–20].

This study focused on *M. pruriens* (L.) DC., var. *utilis*, a commercially significant plant renowned for treating central nervous system disorders such as dementia, Parkinson's, and Alzheimer's. Almost, all plant parts contain L-DOPA, with seeds having the highest quantity, followed by roots, stems, and leaves. *Mucuna pruriens* (L.) DC., var.

utilis (variety: Arka Shubra, developed by ICAR-IIHR, Hesaraghatta Lake Post, Bengaluru) seeds exhibited notably high L-DOPA content (51.9 mg/g) and proline (1.74 mg/g), along with strong antioxidant activity (86.5%)^[21]. This medicinally valuable crop is used for culinary purposes, with pods consumed as vegetables and leaves utilized as animal feed^[22–24]. The characteristic pods bear fruits, and this plant has been studied in various studies^[23,25,26].

L-DOPA, a valuable compound found in *M. pruriens* is in high demand globally, with the world market reaching 250 tons/year, costing USD\$101 billion/year^[27–29]. *M. pruriens* is a versatile plant with historical, cultural, and medicinal significance, offering a range of pharmacological properties and facing challenges in meeting the high demand for its valuable compound, L-DOPA^[30–32]. The velvet beans exhibit intense itching and dermatitis upon contact^[30,32]. Despite L-DOPA being present in all plant parts, its extraction from wild populations faces challenges due to limited availability, posing difficulties in meeting the growing demand^[33,34].

Spineless *M. pruriens* is a tropical leguminous plant that is celebrated for its versatility, serving as a cover crop, forage, traditional medicine source, and high-protein food^[13,35–37]. Similar to other legumes like soybeans, common beans, and mung beans, velvet beans thrive in environments with ample moisture and warmth, both in cultivated and wild varieties^[38–42]. To maximize its potential, understanding and optimizing the germination process are crucial. Seed treatments, including scarification and soaking, offer potential methods to enhance seed germination and, subsequently, crop establishment.

Thermo-hydro-priming, a method of soaking seeds in water to initiate germination without sprouting, has effectively enhanced germination rates and seedling vigor in various crops. This technique activates metabolic processes, improving germination rates and seedling growth. Chemical priming has also been shown to have positive effects on root development and seedling vigor^[2]. It can significantly improve germination rates, with some studies indicating up to 88% germination when combined with mechanical treatments^[40]. Seed priming enhances physiological traits, such as root and shoot length, and biochemical responses, contributing to improved crop establishment and yield. This study aimed to evaluate the effects of different seed priming treatments on germination rate and seedling establishment and determine the optimal seed priming treatment for improving germination dynamics and seedling establishment in *Mucuna pruriens* L. DC. var. utilis (Arka Shubra).

Materials and methods

Experimental site

During 2022–2023, the experiment was conducted under Shade net Nursery conditions, at the CSIR-CIMAP RC, Experimental Farm in Hyderabad (17.25° N latitude and 78.33° E longitude). Spineless *M. pruriens* (L.) DC. Var. utilis, genotype/variety (Arka Shubra) seeds were used in this experiment, and it was designed in a randomized complete block design (RCBD) with seven treatment combinations with three repetitions.

Seed collection

Seeds of spineless *M. pruriens* (L.) DC. Var., genotype/variety: Arka Shubra, were obtained from ICAR-IIHR, Hesaraghatta Lake Post, Bengaluru farm, ensuring genetic purity and viability. The study examined the influence of different seed treatments on seed germination, using the following methods:

Scarification

The Seeds were scarified by chemicals to break the seed coat.

Soaking

The Seeds were soaked in normal water (24 h) and hot water (80 °C) for 5 min.

Treatments

The following seed treatments were used in the experiment: T₁ (control), T₂ (hot water at 80 °C for 5 min), T₃ (normal water for 24 h), T₄ (H₂SO₄ 1% for 5 min), T₅ (KNO₃ 1% + HNO₃ 1% for 24 h), T₆ (GA₃ 500 ppm for 24 h), and T₇ (thiourea for 24 h). A total of 30 seeds for each treatment were subjected to their respective treatments and were sown in separate trays under controlled environmental conditions on June 10th, 2022, and June 22nd, 2023. In this experiment, we utilized sand, vermiculite, and paper towels as substrate. The seeds were exposed to normal daylight conditions, and the temperature was maintained within the range of 20–30 °C to facilitate optimal germination. Germination was monitored daily, and the percentage of germinated seeds was recorded.

Sampling and measurement

To obtain a complete picture of germination behavior, the following parameters were calculated using formulas and methodologies:

Germination (%) was calculated using the following formula^[43]:

$$\text{Germination (\%)} = \left[\frac{\text{No. of seeds germinated}}{\text{Total no. of seeds}} \right] \times 100$$

The mortality (%) was calculated using the following formula:

$$\text{Mortality (\%)} = \left[\frac{\text{No. of seeds ungerminated + Dead seedlings}}{\text{Total no. of seeds}} \right] \times 100$$

The survival (%) was calculated using the following formula:

$$\text{Survival (\%)} = \left[\frac{\text{No. of healthy seedlings}}{\text{Total no. of seeds}} \right] \times 100$$

For survival (%) we use visual observations for healthy seedlings that typically exhibit vibrant green leaves, strong stems, and a well-developed root system. Signs of distress include yellowing leaves, wilting, stunted growth, or discoloration.

Days to germination were calculated as:

Days to germination = Days to initial emergence – Days to final emergence

Days to Initial Emergence refers to the number of days required for the first seedling to break through the soil surface after planting. In contrast, Days to Final Emergence indicates the total number of days for all seeds within a given treatment to emerge. Additionally, 'Days to Germination' is defined as the time from planting to the first emergence of a seedling, rather than the difference between initial and final emergence.

The speed of germination was calculated using the following formula^[31]:

$$GS = \frac{\sum ni}{\sum di}$$

Here, 'ni' is the number of germinated seeds, and 'di' is the total number of days.

The Vigor index was calculated by the following formula^[32]:

$$VI = \frac{GP \times SL}{100}$$

Here, 'GP' is the germination (%), and 'SL' is the seedling length.

Observations

The following seven attributes were recorded: ASL = average seedling length (cm); DFG = days for germination; Germ (%) = germination (%); Mort (%) = mortality (%); SDW = seedlings dry weight (g); SP = survival (%), and VI = Vigor Index.

Statistical analysis

The mean data were analyzed using IBM SPSS Statistics Ver. 19 software^[44] for DMRT (Duncan's Multiple Range Test). Multivariate PCA was conducted using PAST Ver. 4.3 software to assess the impact of various treatments on the germination of *M. Pruriens*, and correlation analysis was employed to further investigate the relationships between the treatment variables and germination outcomes.

Results and discussion

Germination and mortality (%) (GP and MP)

The germination (%) of spineless *M. Pruriens* exhibited notable variations under different treatments. H₂SO₄ treatment yielded the highest germination rate at 57.14% ± 2.49%, surpassing other treatments. Hot water treatment followed closely, with a germination rate of 44.25% ± 1.35%, comparable to the 42.86% ± 1.87% observed with normal water treatment. In contrast, the control group exhibited a significantly lower germination rate at 22.0% ± 0.96% compared to the rest of the treatments. This outcome suggests that applying H₂SO₄ positively influenced germination, outperforming both hot and normal water treatments. The efficacy of H₂SO₄ in enhancing germination may be attributed to its specific effects on seed coat permeability and the release of dormancy mechanisms^[43,45–47]. The relatively high germination rates observed with hot water and normal water treatments also indicate their potential to promote seed germination.

The markedly lower germination rate in the control group underscores the importance of the applied treatments in optimizing germination conditions for spineless *M. Pruriens* (L.) DC. The obtained results are corroborated with the findings of Wanjekeche et al.^[48] who found that *Mucuna* seeds treated in hot water recorded higher germination compared to the control. Similarly, the mortality percentages in spineless *M. Pruriens* varied significantly across treatments, with the control group recording a markedly higher mortality rate of 78% compared to the other groups.

In contrast, the H₂SO₄ treatment exhibited the lowest mortality rate at 42.86% ± 2.49%, followed by hot water treatment (55.75% ± 1.35%) and normal water treatment (57.14% ± 1.87%) (Table 1). This divergence in mortality rates highlights the potential impact of different treatments on the survival of *Mucuna pruriens*. The significantly higher mortality in the control group suggests that natural conditions or a lack of specific treatments may adversely affect seedling survival. Conversely, the lower mortality rates observed in the H₂SO₄, hot water, and normal water treated plants indicate their potential to enhance seedling survival.

Spineless *Mucuna* seeds commonly display both physical and physiological dormancy, hindering germination in a timely and uniform manner^[49,50]. This dormancy is attributed to the impermeability of the seed coat and the presence of inhibitory substances within the seed. Sulfuric acid is recognized for its capacity to break seed dormancy via scarification, which entails weakening or thinning the seed coat^[51]. The application of H₂SO₄ can effectively target the robust seed coat of Spineless *Mucuna*, promoting water absorption and facilitating the initiation of germination^[34,51].

Enhanced germination observed in spineless *Mucuna* seeds following acid scarification with H₂SO₄ can be attributed to the alleviation of physical and physiological barriers that impede gaseous exchange and water uptake. This treatment likely facilitated improved imbibitions and respiration, which are critical for initiating metabolic processes and fostering early germination. Similar findings have been reported by Bhuse et al.^[52] in *Senna* species, where acid scarification significantly improved germination rates by overcoming seed coat dormancy. The hard seed coat in *Mucuna pruriens* acts as a physical barrier to water and oxygen diffusion. H₂SO₄, a strong acid, chemically erodes the seed coat, creating micro-pores and cracks that enhance permeability. This mechanical breakdown allows for faster and more efficient imbibitions, which is essential for rehydrating seed tissues and activating metabolic processes.

Average seedling length (ASL) cm

The average seedling length of Spineless *Mucuna* exhibited notable variations under different treatments, with the application of H₂SO₄ resulting in a significantly higher seedling length of 19.67 ± 0.83 cm. This length was comparable to that observed after the application of GA₃ (19.39 ± 0.40 cm) and normal water (19.0 ± 0.83 cm) (Table 1). In contrast, the control group exhibited a significantly shorter seedling length of 8.89 ± 0.39 cm.

These findings indicate that H₂SO₄ treatment not only positively influenced the seedling length of Spineless *Mucuna* but also outperformed the effects of both GA₃ and normal water treatments. This suggests that H₂SO₄ may play a crucial role in promoting seedling growth, potentially by enhancing nutrient uptake and improving the physiological processes essential for growth. The effectiveness of H₂SO₄ in stimulating seedling development could be attributed to its ability to break seed dormancy and improve seed coat permeability, thereby facilitating better access to water and nutrients.

Conversely, the significantly reduced seedling length observed in the control group highlights the critical importance of external treatments in fostering the growth and development of *Mucuna pruriens* seedlings. The lack of any treatment in the control group likely resulted in suboptimal conditions for germination and early growth, underscoring the necessity of implementing effective pre-germination strategies to enhance seedling vigor.

These results are consistent with the findings of Fiallos et al.^[53], who also reported the positive effects of various treatments on seedling growth in leguminous species. The implications of this study extend beyond the immediate effects on seedling length; they suggest that the application of treatments such as H₂SO₄ and GA₃ can significantly improve the establishment and overall health of Spineless *Mucuna* seedlings.

Survival (%) (SP %)

The survival rates of spineless *Mucuna* seedlings following treatment with H₂SO₄ were significantly higher, reaching 56.98% ± 1.73%, in contrast to the remaining treatments. Subsequent in efficacy were hot water treatment (43.94% ± 1.16%) and normal water treatments (39.42% ± 1.04%) (Table 1). The control group exhibited a significantly lower survival rate of 20.56% ± 0.53% compared with the other treatments, with thiourea treatment following closely. The results highlight the remarkable impact of H₂SO₄ treatment on the survival of Spineless *Mucuna* seeds, surpassing the effects of both hot and normal water treatments. This underscores the potential utility of H₂SO₄ in enhancing seed viability and germination. The lower survival rate observed in the control group emphasizes the importance of specific treatments in fostering optimal conditions for seedling survival.

These findings underscore the significant positive impact of H₂SO₄ treatment on the survival of Spineless *Mucuna* seedlings, highlighting its effectiveness in enhancing seed viability and promoting successful germination. The superior survival rate associated with H₂SO₄ treatment can be attributed to its role in breaking seed dormancy and improving seed coat permeability, which facilitates better water absorption and nutrient uptake. This treatment likely creates more favorable conditions for seedling establishment, leading to higher survival rates.

The notably lower survival rate observed in the control group emphasizes the critical need for specific treatments to foster

Table 1. Thermo-hydro-chemical seed treatments and germination dynamics of *Mucuna pruriens* L. DC. var. *utilis*.

Treatments	Germination (%)	Mortality (%)	SL (cm)	Survival (%)	DG	SDW (g)
T ₁ : Control	22.00 ± 0.96e	78.00 ± 0.96a	8.89 ± 0.39c	20.56 ± 0.53f	14.26 ± 0.38a	0.30 ± 0.04f
T ₂ : Hot water	44.25 ± 1.35b	55.75 ± 1.35d	17.33 ± 0.76b	43.94 ± 1.16b	11.23 ± 0.54d	0.68 ± 0.05d
T ₃ : Normal water	42.86 ± 1.87bc	57.14 ± 1.87d	19.00 ± 0.83a	39.42 ± 1.04c	10.67 ± 0.81f	0.74 ± 0.03c
T ₄ : H ₂ SO ₄	57.14 ± 2.49a	42.86 ± 2.49e	19.67 ± 0.86a	56.98 ± 1.73a	8.69 ± 0.26g	0.90 ± 0.04a
T ₅ : KNO ₃ + HNO ₃	28.94 ± 1.26d	71.06 ± 1.26bc	16.81 ± 0.73b	27.42 ± 0.87d	13.33 ± 0.34b	0.36 ± 0.08
T ₆ : GA ₃	27.65 ± 0.58d	72.35 ± 0.58b	19.39 ± 0.40a	26.89 ± 0.92d	9.67 ± 0.21e	0.82 ± 0.02b
T ₇ : Thiourea	27.01 ± 0.71d	72.99 ± 0.71b	16.98 ± 0.45b	26.05 ± 0.54de	12.65 ± 0.37c	0.50 ± 0.04de

SL, Seedling length (cm); SP, Survival (%); DG, Days to germination; SDW (g), Seedling dry weight; Average data followed by similar letter in the same column means not significantly different based on 5% of DMRT (Duncan's Multiple Range Test).

optimal conditions for seedling survival. Without these interventions, seedlings may struggle to thrive because of factors such as physical dormancy and inadequate access to moisture and nutrients. This finding reinforces the idea that pre-germination treatments are essential for improving the establishment and growth of *Mucuna pruriens* seedlings.

Days for germination (DFG)

The germination period for spineless *Mucuna* seeds was significantly shorter under the treatment with H_2SO_4 , requiring only 8.69 ± 0.26 days, compared with the other treatments. Following closely in efficiency was the GA_3 treatment. Conversely, the control group exhibited a prolonged germination period of 14.26 ± 0.38 days and followed by $KNO_3 + HNO_3$ treatment (13.33 ± 0.34 d) (Table 1). The notable reduction in germination days observed with H_2SO_4 treatment suggests its effectiveness in expediting the germination process of spineless *Mucuna* seeds. This finding indicates the potential utility of H_2SO_4 in optimizing germination conditions.

The implications of these results are particularly relevant for the cultivation of *Mucuna pruriens*, as faster germination can lead to earlier establishment of the crop and potentially higher yields. Previous studies have shown that treatments such as acid scarification can effectively reduce germination times in various leguminous species by overcoming physical dormancy and enhancing seed coat permeability^[41]. Moreover, the prolonged germination periods observed in the control group and in the $KNO_3 + HNO_3$ treatment underscore the challenges posed by untreated seeds, which may struggle to germinate due to inherent dormancy mechanisms.

Vigour Index (VI)

H_2SO_4 treatment resulted in significantly higher values for VI I and VI II, measuring 2.91 ± 0.21 and 51.18 ± 4.41 , respectively, compared to all other treatments. The hot water treatment was closely followed, with values of 2.56 ± 0.18 and 30.08 ± 1.81 for VI I and VI II, respectively. Conversely, the control group exhibited significantly lower VI I (6.53 ± 0.41) compared to the other treatments, with the $KNO_3 + HNO_3$ treatment recording the second-lowest values of 1.72 ± 0.09 and 10.30 ± 0.94 for VI I and VI II, respectively (Table 1 &

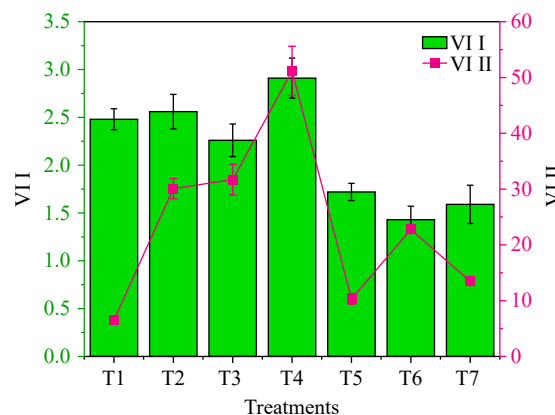


Fig. 1 Vigour index with the different treatments (error bars with standard deviation) in *Mucuna pruriens* L. DC. var. utilis). [T₁ (control), T₂ (hot water 80 °C for 5 min), T₃ (normal water for 24 h), T₄ (H_2SO_4 1% for 5 h), T₅ (KNO_3 1% + HNO_3 1% for 24 h), T₆ (GA_3 500 ppm for 24 h), and T₇ (thiourea for 24 h); ASL = Average seedling length; DFG = Days for germination; GP/Germ% = Germination (%); Mort% = Mortality (%); SDW= Seedling Dry Weight; SP/Sur% = Survival (%); VI I = Vigour index 1; VI II = Vigour Index 2].

Fig. 1). The substantial increase in VI I and VI II observed after H_2SO_4 treatment highlights its effectiveness in promoting these indices, suggesting a positive influence on the physiological and biochemical aspects of treated specimens. The hot water treatment also demonstrated notable effects, albeit to a lesser extent. In contrast, the control group exhibited significantly lower values, highlighting the importance of specific treatments in achieving favorable physiological responses (Figs 2, 3 & 4).

While the hot water treatment was also demonstrated beneficial effects on vigor indices, its impact was less pronounced than that of H_2SO_4 . This suggests that while hot water can be an effective treatment for promoting germination and early growth, it may not be as potent as H_2SO_4 for optimizing physiological responses in Spineless *Mucuna* seedlings.

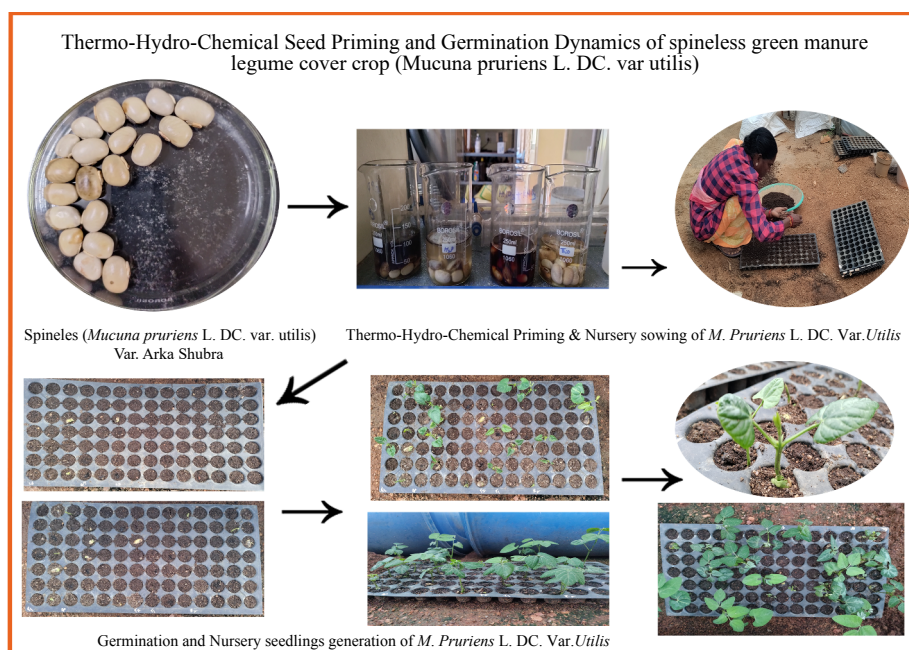


Fig. 2 Thermo-hydro-chemical seed priming and germination dynamics of spineless green manure legume cover crop (*Mucuna pruriens* L. DC. var. utilis).



Fig. 3 Seedling height of *Mucuna pruriens* in (a) (T_4 : H_2SO_4 1% for 5 min), and (b) (T_2 : hot water 80 °C for 5 min) thermo-hydro-chemical seed priming treatments.



Fig. 4 Trichomeless varieties of (a) CIM Ajar, (b) CIM-Nirom, and (c) wild collection with trichomes of *Mucuna pruriens*.

The significantly lower values observed in the control group highlight the critical role of specific treatments in achieving favorable physiological outcomes. Without these interventions, seedlings may experience suboptimal growth conditions, leading to reduced vigor and overall health.

Seedling dry weight (g) (SDW)

Applying H_2SO_4 resulted in a significantly higher seedling dry weight of 0.90 ± 0.04 g compared to all other treatments, with GA_3 followed closely at 0.82 ± 0.02 g. Conversely, the control group exhibited a significantly lower seedling dry weight of 0.30 ± 0.04 g. The substantial increase in seedling dry weight under the H_2SO_4 treatment suggests its efficacy in promoting robust growth and biomass accumulation. The observed higher dry weight in the GA_3 treatment further underscores the positive impact of specific treatments on seedling development. In contrast, the control group's significantly lower seedling dry weight emphasizes the importance of external factors in influencing plant growth. The markedly lower seedling dry weight in the control group underscores the critical importance of external factors, such as pre-germination treatments, in influencing plant growth. Without these interventions, seedlings may struggle to establish themselves, leading to reduced biomass

and overall vigor. This finding aligns with the existing literature that emphasizes the necessity of employing effective treatments to optimize seedling growth and development^[41,54].

Correlation matrix and principal component analysis (PCA)

The correlation matrix provides insights into the degree of association between different variables. From the results, a strong positive correlation ($r = 0.996$) was observed between germination % and survival %, indicating that higher germination rates are strongly associated with better seedling survival. Similarly, a strong positive correlation ($r = 0.956$) was found between germination % and vigor index II, suggesting that seeds with higher germination rates tended to exhibit better overall vigor (Fig. 5). On the other hand, a strong negative correlation was observed between mortality % and germination % ($r = -0.99$), as well as between days to germination and seedling dry weight ($r = -0.991$). This implies that seeds that germinate faster tend to produce seedlings with higher dry weight, indicating that early germination is advantageous for seedling development. Additionally, negative correlations were noted between days to germination and vigor index ($r = -0.896$) and between seedling length and days to germination ($r = -0.812$) (Fig. 5). These findings

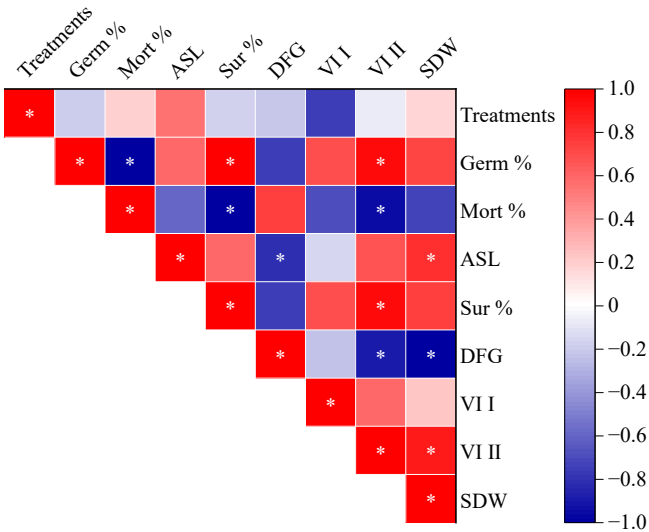


Fig. 5 Correlation matrix of germination traits in *Mucuna pruriens*.

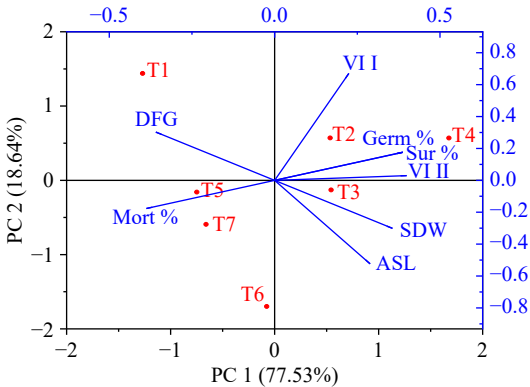


Fig. 6 Principal component analysis (PCA) for treatments with germination-related traits in *Mucuna pruriens* [T₁ (control), T₂ (hot water 80 °C for 5 min), T₃ (normal water for 24 h), T₄ (H₂SO₄ 1% for 5 h), T₅ (KNO₃ 1% + HNO₃ 1% for 24 h), T₆ (GA₃ 500 ppm for 24 h), and T₇ (thiourea for 24 h); ASL = Average seedling length; DFG = Days for germination; GP/Germ% = Germination (%); Mort% = Mortality (%); SDW= Seedling Dry Weight; SP/Sur% = Survival (%); VI I = Vigour index 1; VI II = Vigour Index 2].

Table 2. Correlation analysis of germination related traits of *Mucuna pruriens* L. DC. var. utilis.

Characters	G (%)	M (%)	SL (g)	SP (%)	DG	VI I	VI II	SDW (g)
G (%)	1	−0.99	0.596	0.996	−0.747	0.697	0.956	0.739
M (%)		1	−0.600	−0.996	0.746	−0.698	−0.957	−0.738
SL (g)			1	0.591	−0.812	−0.155	0.670	0.802
SP (%)				1	−0.754	0.698	0.958	0.742
DG					1	−0.228	−0.896	−0.991
VI I						1	0.593	0.231
VI II							1	0.887
SDW (g)								1

GP (%), Germination (%); Mort (%), Mortality (%); SP (%), Survival (%); DG, Days to germination; VI, Vigour Index; SL, Seedling length (cm); SDW (g), Seedling dry weight; Correlation is significant at 5%.

highlight that early and high germination rates are critical for overall seedling success. Reducing the time to germination may enhance multiple seedling characteristics, and selecting for high germination percentages is likely to improve survival rates.

To assess the variability among treatment effects and germination characteristics, principal component analysis (PCA) was employed on eight parameters of *M. pruriens*. The resulting PCA plot (Fig. 6) revealed that the first two principal components (PC-1 and PC-2) collectively accounted for 96.17% of the total variation. PC-1 explained 77.53% of this variation, with positive contributions from VI I, VI II, Germination %, and Survival %, and negative contributions from ASL and SDW. PC-2 explained an additional 18.64% of the total variation, with a positive contribution from DFG and a negative contribution from mortality %. Notably, treatments T₂ and T₄ exhibited positive contributions, while the remaining treatments had negative contributions. Furthermore, the correlation matrix depicted significant correlations among various parameters. Germination % exhibited noteworthy correlations with VI 2 and survival (Table 2).

Conclusions

The findings of this study underscore the significant impact of various seed treatments on the germination and growth characteristics of spineless *M. pruriens*. The application of H₂SO₄ emerged as the most effective treatment, leading to the highest germination percentage, reduced mortality rates, enhanced seedling length, and increased seedling dry weight. These results suggested that H₂SO₄ not only facilitates the breaking of seed dormancy but also promotes robust growth and biomass accumulation, thereby optimizing the conditions for seedling establishment. Hot water and normal water treatment also demonstrated positive effects, although to a lesser extent than H₂SO₄. The control group, which lacked any specific treatment, exhibited significantly lower germination rates and higher mortality, highlighting the critical role of targeted interventions in improving seedling survival and growth. Our results align with existing literature, reinforcing the notion that effective seed treatments can substantially enhance the physiological and biochemical responses of *M. pruriens*. Principal component analysis (PCA) further elucidated the relationships among various growth parameters, revealing that specific treatments positively influenced key indices such as vigor and survival rates. The significant correlations identified among germination percentage, seedling length, and survival rates emphasize the interconnectedness of these traits in determining overall seedling performance. The results of this study have implications for developing sustainable agriculture practices, particularly in regions where *M. pruriens* is a key crop.

Author contributions

The authors confirm contribution to the paper as follows: study conception and design: Jnanesha AC; analysis and interpretation of results: Venu Gopal S, Kumar A, Sravya K; draft manuscript preparation: Ranjith Kumar S, Bharath Kumar S; manuscript review and revision: Lal RK. All authors have reviewed the results and approved the final version of the manuscript.

Data availability

All data generated or analyzed during this study are included in this published article.

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Conflict of interest

The authors declare that they have no conflict of interest.

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