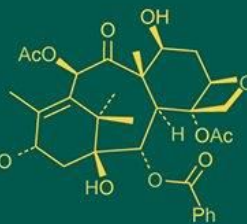
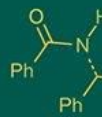
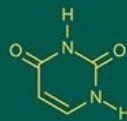
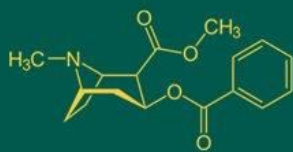


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Sweta Binjhar
Research Scholar, Pt. Kishori
Lal Shukla College of
Horticulture and Research
Station, MGUVV, Durg,
Chhattisgarh, India

Abhay Bisen
Assistant Professor (Fruit
Science), Pt. Shiv Kumar
Shashtri College of Agriculture
and Research Station, IGKV,
Chhattisgarh, India

Corresponding Author:
Sweta Binjhar
Research Scholar, Pt. Kishori
Lal Shukla College of
Horticulture and Research
Station, MGUVV, Durg,
Chhattisgarh, India

Effect of pre-sowing seed treatments on germination, seedling growth and development of Ber rootstock (*Ziziphus rotundifolia* Lamk.)

Sweta Binjhar and Abhay Bisen

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Abstract

The present investigation entitled “Effect of pre-sowing seed treatments on germination, seedling growth and development of Ber rootstock (*Ziziphus rotundifolia* Lamk.)” was carried out at the research cum instructional farm of Department of Fruit Science, Pt. K.L.S. College of Horticulture and Research Station, Rajnandgaon (C.G.). during the kharif season of 2023-24 in completely randomized design and replicated thrice with a total of 14 treatments viz., T₀: Direct seed sowing (control), T₁: Hot water treatment 50 °C for 30 minute, T₂: Hot water treatment 100 °C for 15 minute, T₃: GA₃ @ 250 ppm for 24 hours, T₄: GA₃ @ 500 ppm for 24 hours, T₅: GA₃ @ 250 ppm for 12 hours, T₆: GA₃ @ 500 ppm for 12 hours, T₇: CONC. H₂SO₄ for 5 minute, T₈: CONC. H₂SO₄ for 10 minute, T₉: KNO₃ 1% for 12 hour, T₁₀: KNO₃ 1% for 24 hour, T₁₁: KNO₃ 2% for 12 hour, T₁₂: KNO₃ 2% for 24 hour, T₁₃: Mechanical scarification with sand paper. Among the different treatments, GA₃ @ 250 ppm for 24 hours performed best in term of The days taken for seed germination were significantly the earliest (11.2 days), Seed germination percentage were the maximum at 15 DAS and 30 DAS (58.88 and 91.11%), height of seedling (53.86 cm), diameter of seedling (4.03 mm), intermodal length (32.6 mm), number of leaves per plant (57.06), leaf area (6.24 cm²).

Keywords: Ber pre-sowing, seed treatments, GA₃, CONC. H₂SO₄, KNO₃

1. Introduction

Ber (*Ziziphus rotundifolia* Lamk.) is a wild species of Ber and a hardy fruit tree used as one of the important rootstocks for commercially grown Ber plants in India. It belongs to the family Rhamnaceae; it is tetraploid in nature with chromosome number 2n (4x) = 48. Ber is an arid and semi-arid region fruit crop; it can grow well up to a height of 100 m above mean sea level. The areas receiving annual rainfall of 400–650 mm are ideal for its cultivation. It is known for its ability to withstand adverse conditions such as drought, waterlogging, and saline conditions. Ber grows in cultivated and wild forms in India, Haryana, Punjab, Uttar Pradesh, Rajasthan, Gujarat, Madhya Pradesh, Bihar, Maharashtra, Andhra Pradesh, and Tamil Nadu. The area under Ber cultivation in India is about 52,000 hectares with a production of 539 MT (NHB, 2020). In Chhattisgarh Korba, Raigarh, Dhamtari and Janjgir Champa are major contributor in a total area of 4511000 ha, with a production of 73.586 MT of Ber fruit in Chhattisgarh. (Anon., 2022) [24]

Ber plants raised by sowing of seeds is less uniform, poor and have delayed germination due to hard seed coat (endocarp). Seeds can be extracted from stones either mechanically or chemically but it might damage the embryo, which results in irregular germination. Seeds are most often used to produce rootstock for the purpose of vegetative propagation. Each fruit contains one stone embedded in the pulp at the center of the fruit. The dormancy of seeds might be due to the hard seed coat which can be overcome to a greater extent by physical removal of seed coat or using pre-sowing treatments. Dormancy of the hard seed can be broken by gibberellic acid treatment, chilling treatment, hot water treatment, scarification, stratification, soaking in water and acid treatment depending on the kind of dormancy (Kher and Nataraj, 2015) [10]. Mechanical scarification of seed to improve germination must be done up to seed coat, which is immediately above the tip of cotyledons. (Sheoran *et al.*, 2018) [19]. GA₃ also helps in increasing the availability of reserved mineral elements, which induces the germination process.

Scarification of Ber seeds using 250 ppm GA₃ for 24 hrs performed best in terms of germination and seedling growth under lab conditions (Sheoran *et al.*, 2018) [19].

Sulphuric acid increases the germination of seed by softening the seed coat and allowing easy permeability to air and water (Brijwal *et al.*, 2013) [7]. Potassium nitrate (KNO₃) is the most widely used chemical for promoting seed germination. Nitrate ions activate the germination of dormant seeds (Alboresi *et al.*, 2005) [1]. A Solution of 1% and 2% KNO₃ gives good results during seed germination of Ber in lab condition as compared of direct sowing (Sheoran *et al.*, 2018) [19]. It is therefore highly necessary to facilitate the seed germination and seedling growth of promising rootstock species with pre-sowing treatments.

2. Materials and Methods

The present investigation entitled, “Effect of pre-sowing seed treatments on germination, seedling growth and development of Ber rootstock (*Ziziphus rotundifolia* Lamk.)” was carried out in the Horticulture Research Farm, under Pt. K. L. S. College of Horticulture and Research Station Rajnandgaon (C.G.) during the year 2023-24. The experiment was laid out in Completely Randomized Design (CRD) with 14 treatments and three replications. Fruits were kept in water for 2-3 days to remove peel and adhered pulp portion from the stone. The extracted seed and stone were cleaned thoroughly, dried in shade and stored at room temperature. Seeds treated with different pre-sowing treatments viz., T₀: Direct seed sowing (control), T₁: Hot water treatment 50 °C for 30 minute, T₂: Hot water treatment 100 °C for 15 minute, T₃: GA₃ @ 250 ppm for 24 hours, T₄: GA₃ @ 500 ppm for 24 hours, T₅: GA₃ @ 250 ppm for 12 hours, T₆: GA₃ @ 500 ppm for 12 hours, T₇: CONC. H₂SO₄ for 5 minute, T₈: CONC. H₂SO₄ for 10 minute, T₉: KNO₃ 1% for 12 hour, T₁₀: KNO₃ 1% for 24 hour, T₁₁: KNO₃ 2% for 12 hour, T₁₂: KNO₃ 2% for 24 hour, T₁₃: Mechanical scarification with sand paper. These treatments were applied to the ber seeds in mentioned concentration during July. These treated seeds were sown in the polybags, which were filled with sand, soil and vermicompost in 1:1:1 ratio. The cultural operations during experiment were similar for all the treatments. Observation on days taken for seed germination were counted regular and seed germination (%) at 15 DAS and 1 month after sowing. It was calculated based on the below mentioned formula:

$$\text{Germination (\%)} = \frac{\text{Total no. of seed germinated}}{\text{Total no. of seeds sown}} \times 100$$

Seedling were measured for seedling height, seedling diameter, internodal length, number of leaves per plant, leaf area at 60, 90 and 120 DAS. The height of 5 randomly selected seedlings was measured from the collar area to the tip of the shoot, and the average height was given in cm. Seedling diameter was measured with the help of electronic vernier caliper. Internodal length was measured with the help of meter scale and expressed in millimeter. Fully formed leaves were physically counted from randomly selected seedling. To compare the effects of the treatments, an average number of leaves was gathered. Leaf area per leaf of 5 randomly selected seedling was measured. It was calculated using a leaf area meter and reported in square centimeters. The data was statistically analyzed using the completely randomized block design by applying the technique of analysis of variance by Fisher (1958) [25]. The

mean values of several parameters make up the data in the thesis and the crucial difference is used to compare the significance level. The OPSTAT portal, which is accessible on the CCS HAU website, was used to do statistical analysis.

3. Result and Discussion

3.1 Days taken for seed germination

As per the data displayed in Table 1, it is clear that the minimum days taken to start germination (11.2 days) was confirmed under the treatment T₃ (GA₃ @ 250 ppm for 24 hours) The maximum (25.2 days) days taken to start germination was noted under control (T₀). This was followed by T₁₁ (19.4 days). Data in Table 1 demonstrate that early seed germination was achieved by Ber seeds treated with GA₃ before sowing. Possible explanations for this include increased cell wall flexibility, improved water absorption, and GA₃'s role in the activation of cytological enzymes. Through encouraging protein synthesis, elongating coleoptiles and leaves, and aiding in the generation of ethylene, GA₃ directly affects embryos, releasing them from dormancy. Because of the ethylene, hydrolases are synthesized, particularly amylase, which promotes seed germination. Similar results were also found by Vasantha *et al.*, (2014) [23] in tamarind, Lalitha *et al.*, (2020) [11] in aonla and Athani *et al.*, (2013) [4] in guava.

3.2 Germination (%)

As per the data displayed in Table 1, it is clear that the Among different treatment of GA₃, T₃ (GA₃ @ 250 ppm for 24 hrs) significantly had the highest percentage of germination at 15 and 30 DAS, 58.88% and 91.11% and it is followed by T₄ (GA₃ @ 500 ppm for 24 hrs) 52.21% and 85.55%. While, the minimum germination (%) was observed in control (T₀) at 15 and 30 DAS (24.44 and 53.33%). GA₃ promotes seed germination by causing the enzyme amylase to be formed, which turns insoluble starch into soluble carbohydrates. According to Gillard and Walton (1973), GA₃ also starts the proliferation of radicals by eliminating some metabolic barriers. The current study's findings indicated that seeds responded favorably to GA₃ treatment up to 250 ppm, however the greater dose of GA₃ may have partially inhibited or slowed the germination process. These results are consistent with those obtained by Hore and Sen (1994) [8]. Similar results were reported by Rajamanickam *et al.*, (2002) [14] in aonla and Singh *et al.*, (2023) [21] in custard apple.

3.3 Height of seedling (cm)

The results revealed that among all treatments, the seedling height increased significantly in comparison to control. The maximum seedling height after 60, 90 and 120 DAS was recorded as 38.8, 47.06 and 53.86 cm, respectively, with T₃ (GA₃ @ 250 ppm for 24 hours); which was followed by T₄ (GA₃ @ 500 ppm for 24 hours) was recorded as 38.13, 43.66 and 49.4 cm. Minimum seedling height at 60 DAS, 90 DAS and 120 DAS was observed as 29.53, 32.26 and 35.13 cm, in untreated seed control (T₀).

The hormone GA₃ may have increased osmotic uptake of nutrients, causing cell elongation and subsequent height growth in the seedlings. It may also have increased stem diameter by increasing cell division and elongation at the stem portion (Sen *et al.*, 1990) [15]. Similar results were reported by Singh *et al.*, (2023) in custard apple and Hota *et al.*, (2018) [9] in jamun.

3.4 Diameter of seedling (mm)

Diameter of seedling (mm) as influenced by various pre-sowing seed treatments on Ber are presented in Table No. 1. Among different treatments, maximum seedling diameter at 60, 90 and 120 DAS (3.52, 3.92, 4.03 mm, respectively) was recorded when the seeds of Ber were dipped in GA₃ @ 250 ppm (T₃), which was followed by T₄ (3.26, 3.78, and 3.88 mm). The minimum seedling diameter was observed in control at 60, 90 and 120 DAS (1.96, 2.70, and 2.88 mm, respectively). The result is showed that the pre-sowing seed treatments noticeably improved Ber seedling diameter over time. Dipping seeds in GA₃ at 250 ppm for 24 hrs consistently resulted in the thickest stem diameter at all stages, promoting cell expansion and cell division. The plant growth hormone likely stimulated morphological development more effectively than other treatments by augmenting cambial activity. Similar results were reported by Hota *et al.*, (2018)^[9] in jamun and Manekar *et al.*, (2011)^[12] in aonla.

3.5 Internodal length (mm)

Internodal length of seedlings (mm) at 60, 90 and 120 DAS as influenced by different pre-sowing seed treatments on Ber is presented in Table 2. After 60 DAS, the Ber seedling with the longest inter-nodal length (24.2 mm) was measured using GA₃ @ 250 ppm for 24 hours (T₃). This result was at par with GA₃ @ 500 ppm for 24 hours (T₄) (22.43). The control (T₀) had the lowest intermodal length (17.03 mm). A Ber seedling at 90 DAS had the longest inter-nodal length (30.1 mm) when GA₃ @ 250ppm was used for 24 hours (T₃). This result was statistically at par with 28.63 mm when GA₃ @ 500 ppm for 24 hours (T₄) was used. While the control (T₀) had the shortest intermodal length (23.13). The GA₃ at 250 ppm for 24 hours (T₃) produced the longest inter-nodal length of Ber seedlings (32.6 mm) at 120 DAS, which was at par with (T₄) GA₃ @ 500 ppm for 24 hours (31.13 mm). The smallest inter-nodal length of seedling (26.43 mm) at 120 DAS was observed in control. Early germination may have ultimately led to a rise in seedling height, which could account for the increase in intermodal length observed with pre-sowing seed treatments. Thus, the longest intermodal length was associated with more heighted plants. Increased apical dominance, increased leaf growth, and increased inter-node extension are the three

most distinctive impacts of GA₃ on shoot growth. (Brian *et al.*, 1959)^[6]. The results are in agreement with Sheoran *et al.*, (2018)^[19].

3.6 Number of leaves per plant

The maximum number of leaves at 60, 90 and 120 DAS (35.26, 52.8 and 57.06, respectively) were recorded when the seeds of Ber were treated with GA₃ @ 250 ppm (T₃), which was statistically at par with T₄ (33.66, 48.33, 52.73) at 60, 90 and 120 days. The minimum number of leaves was noted in control at 60, 90 and 120 DAS (20.13, 24.8 and 26.13, respectively). Maximum number of leaves per plant seen in seed soaked in 250 ppm soaked 24 hours of GA₃.

This observation may be explained by the increased nucleoprotein system that results from GA₃ in the apical meristem, which increases leaf initiation and expansion (Simao *et al.*, 1960)^[17]. More leaf output may arise from improved seedling growth as well as increased GA₃ activity at the apical meristem, which increases nucleoprotein synthesis and increases leaf initiation (Sen and Ghunti, 1976)^[16]. Similar results were found by Singh *et al.*, (2011)^[20] in cashew rootstocks.

3.7 Leaf area (cm²)

The perusal of data presented in Table 2 indicated leaf area per leaf at 60, 90 and 120 DAS. The highest leaf area per leaf of Ber seedling at 60, 90 and 120 DAS (4.65, 5.73 and 6.24 cm², respectively) were recorded when the seeds of Ber were treated with GA₃ @ 250 ppm for 24 hrs (T₃), which was at par with T₄ (4.44, 5.54 and 6.13 cm²) at 60, 90 and 120 DAS and T₆ (6.07 cm²) at 120 DAS. The lowest leaf area per leaf of Ber seedling was noted in control at 60, 90 and 120 DAS (3.38, 4.22 and 4.61 cm², respectively). The increase in leaf area with pre-sowing seed treatments may be due to the reason that early germination eventually increased the vigour of the seedling. Therefore, plants that were more vigorous were linked to larger leaf areas. The most characteristic effects of GA₃ on shoot growth are increased inter-node extension, increased leaf-growth and enhanced apical dominance. (Brian *et al.*, 1959)^[6]. Similar results were found by Barche *et al.*, (2010)^[5] in papaya, Muralidhara *et al.*, (2016)^[13] in mango and Singh *et al.*, (2011)^[20] in cashew rootstocks.

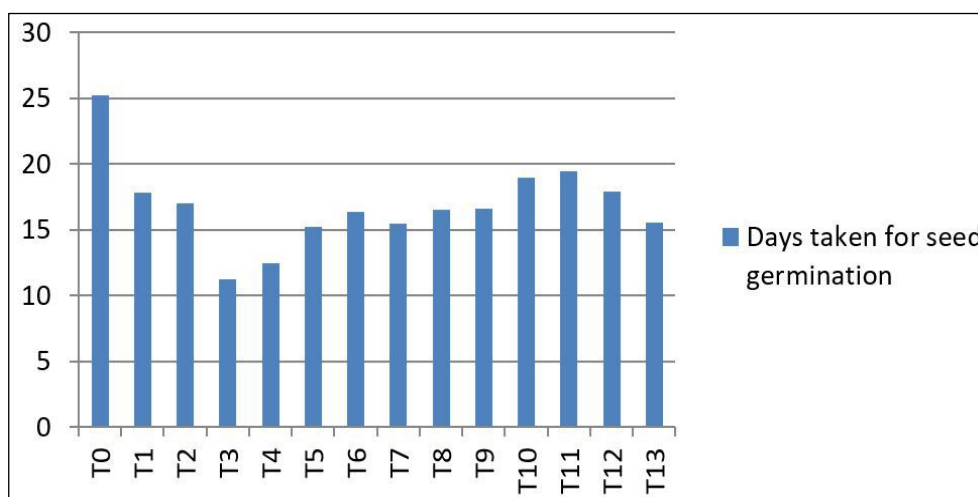
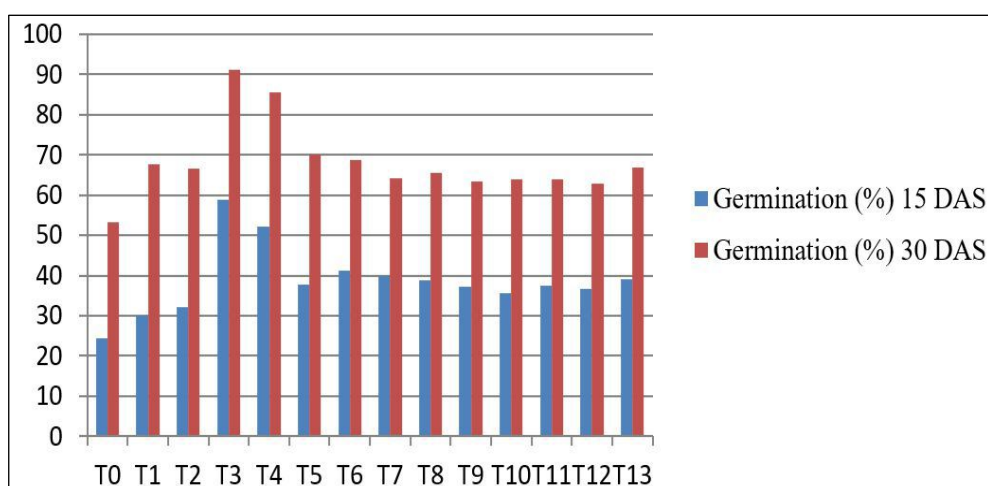
Table 1: Effect of pre-sowing seed treatments on germination, seedling growth and development of Ber rootstock (*Ziziphus rotundifolia* Lamk.)

Notations	Treatment details	Days taken for seed germination	Germination (%)		Seedling height (cm)			Diameter of seedling (mm)		
			15 Das	30 Das	60 Das	90 Das	120 Das	60 Das	90 Das	120 Das
T ₀	Direct seed sowing (control)	25.2	24.44 (4.99)	53.33 (7.34)	29.53	32.26	35.13	1.96	2.70	2.88
T ₁	Hotwatertreatment 50 °C for 30 minute	17.8	29.99 (5.52)	67.77 (8.26)	35.60	40.6	45.26	2.75	3.37	3.54
T ₂	Hotwatertreatment 100 °C for 15 minute	17	32.22 (5.72)	66.66 (8.19)	34.20	39	42.33	2.89	3.48	3.60
T ₃	GA ₃ 250 ppm for 24 hour	11.2	58.88 (7.71)	91.11 (9.57)	38.80	47.06	53.86	3.52	3.92	4.03
T ₄	GA ₃ 500 ppm for 24 hour	12.43	52.21 (7.26)	85.55 (9.28)	38.13	43.69	49.4	3.26	3.78	3.88
T ₅	GA ₃ 250 ppm for 12 hour	15.23	37.77 (6.19)	69.99 (8.40)	36.49	41.75	46.2	2.87	3.41	3.503
T ₆	GA ₃ 500 ppm for 12 hour	16.33	41.11 (6.45)	68.72 (8.32)	35.53	39.46	42.53	3	3.25	3.45
T ₇	CONC. H ₂ SO ₄ for5minute	15.43	39.99 (6.36)	64.13 (8.05)	33.66	38.53	41.73	2.73	3.05	3.35
T ₈	CONC. H ₂ SO ₄ for10minute	16.5	38.88 (6.28)	65.48 (8.12)	36.22	40.4	43.21	2.45	3.17	3.29
T ₉	KNO ₃ 1% for 12 hour	16.56	37.23 (6.14)	63.33 (7.99)	35.60	39.46	43.13	2.72	3.27	3.32
T ₁₀	KNO ₃ 1% for 24 hour	18.96	35.55 (6.00)	63.99 (8.03)	34.73	38.8	42.8	2.82	3.47	3.61
T ₁₁	KNO ₃ 2% for 12 hour	19.4	37.43 (6.16)	64.02 (8.04)	34.86	39.12	42.73	2.79	3.3	3.36
T ₁₂	KNO ₃ 2% for 24 hour	17.86	36.54 (6.09)	62.79 (7.96)	34.28	39.23	43.06	2.76	3.12	3.38
T ₁₃	Mechanical scarification with sand paper	15.53	39.12 (6.29)	66.88 (8.21)	35.36	39.2	43.28	2.82	3.19	3.3
	SE(m)	0.59	0.67 (0.05)	1.17 (0.07)	0.97	0.76	0.87	0.05	0.06	0.06
	C.V.	6.09	3.01 (1.47)	2.98 (1.48)	4.78	3.31	3.46	3.18	3.14	3.29
	C.D. at 5%	1.72	1.95 (0.15)	3.40 (0.20)	2.831	2.22	2.55	0.15	0.17	0.19

Note-SQRT value depicted under brackets

Table 2: Effect of pre-sowing seed treatments on germination, seedling growth and development of Ber rootstock (*Ziziphus rotundifolia* Lamk.)

Notations	Treatment details	Internodal length (mm)			Number of leaves per plant			Leaf area (cm ²)		
		60 Das	90 Das	120 Das	60 Das	90 Das	120 Das	60 Das	90 Das	120 Das
T ₀	Direct seed sowing (control)	17.03	23.13	26.43	20.13	24.8	26.13	3.38	4.22	4.61
T ₁	Hotwatertreatment 50 °C for 30 minute	18.63	25.60	27.96	27.76	39.53	44.66	3.63	4.55	4.77
T ₂	Hotwatertreatment 100 °C for 15 minute	18.33	27.63	28.12	26.66	34.73	40	3.62	4.59	4.81
T ₃	GA ₃ 250 ppm for 24 hour	24.20	30.10	32.6	35.26	52.8	57.06	4.65	5.73	6.24
T ₄	GA ₃ 500 ppm for 24 hour	22.43	28.63	31.13	33.66	48.33	52.73	4.44	5.54	6.13
T ₅	GA ₃ 250 ppm for 12 hour	20.86	28	29.96	27.86	43.46	47.63	3.76	5.35	5.76
T ₆	GA ₃ 500 ppm for 12 hour	20.26	27.46	29.43	27.46	41.06	46.53	3.76	5.31	6.07
T ₇	CONC. H ₂ SO ₄ for5minute	19.30	26.60	28.56	24.66	37.8	40.93	3.69	5.19	5.61
T ₈	CONC. H ₂ SO ₄ for10minute	18.93	26.50	28.36	26.6	36.33	43.86	3.65	5.30	5.58
T ₉	KNO ₃ 1% for 12 hour	20.73	27.26	28.3	26.73	39.6	43.46	4.18	4.70	5.54
T ₁₀	KNO ₃ 1% for 24 hour	18.80	26.50	28.46	25.2	33.6	41.86	4.09	5.17	5.73
T ₁₁	KNO ₃ 2% for 12 hour	19.66	26.86	28.43	25.73	35	43.8	3.60	4.64	5.63
T ₁₂	KNO ₃ 2% for 24 hour	20.46	26.83	27.93	25.46	40.26	42.26	3.68	4.92	5.32
T ₁₃	Mechanical scarification with sand paper	19.86	26.46	27.7	25.93	36.46	41.46	3.55	4.56	5.12
SE(m)		0.72	0.58	0.55	0.49	0.83	1.05	0.08	0.10	0.13
C.V.		6.30	3.73	3.34	3.19	3.69	4.18	4	3.76	4.24
C.D. at 5%		2.11	1.69	1.62	1.45	2.41	3.07	0.25	0.31	0.39

**Fig 1:** Effect of pre-sowing seed treatments on days taken for seed germination of Ber**Fig 2:** Effect of pre-sowing seed treatments on days taken for germination (%) at 15 and 30 DAS of Ber

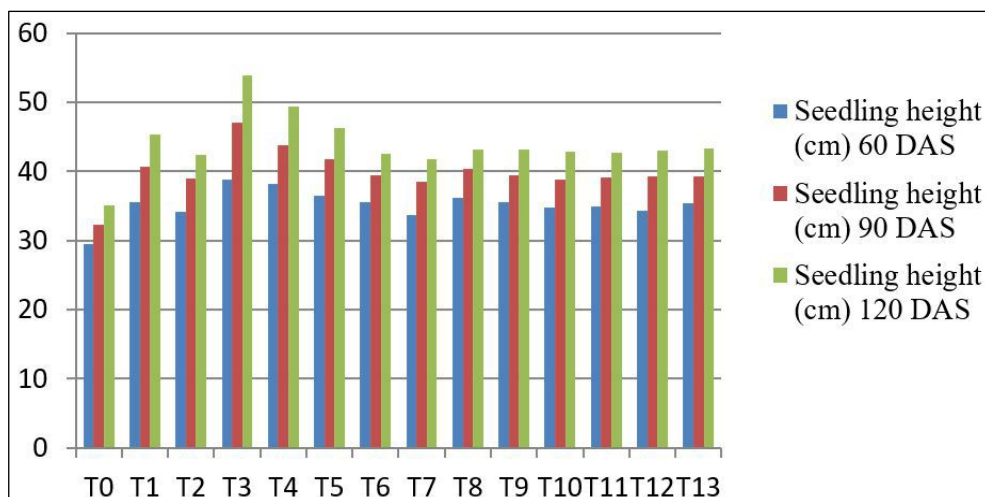


Fig 3: Effect of pre-sowing seed treatments on Height of seedling (cm) at 60, 90 and 120 DAS of Ber

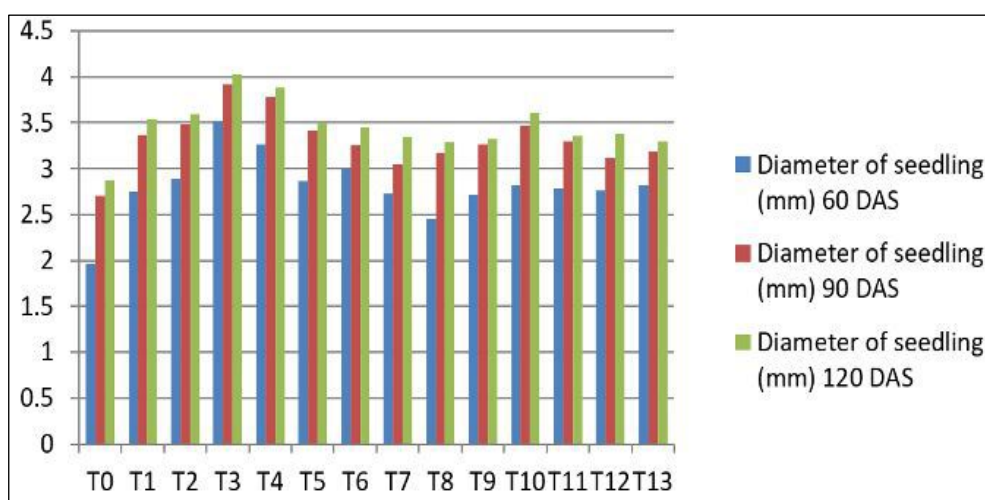


Fig 4: Effect of pre-sowing seed treatments on Diameter of seedling (mm) at 60, 90 and 120 DAS of Ber

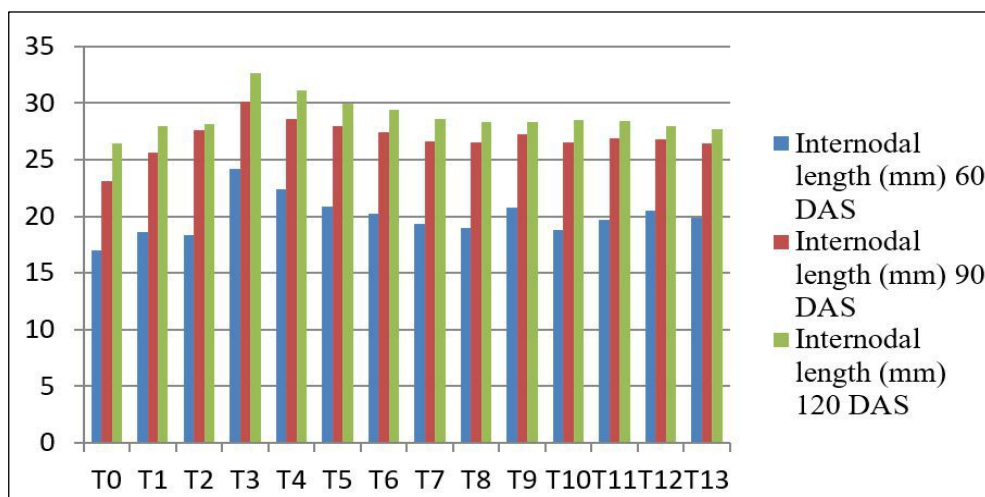


Fig 5: Effect of pre-sowing seed treatments on Internodal length (mm) at 60, 90 and 120 DAS of Ber

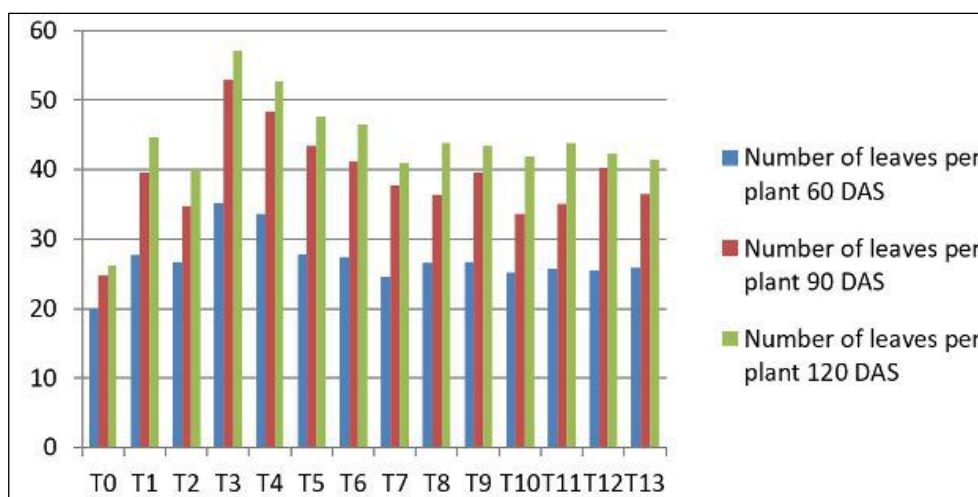


Fig 6: Effect of pre-sowing seed treatments on Number of leaves per plant at 60, 90 and 120 DAS of Ber

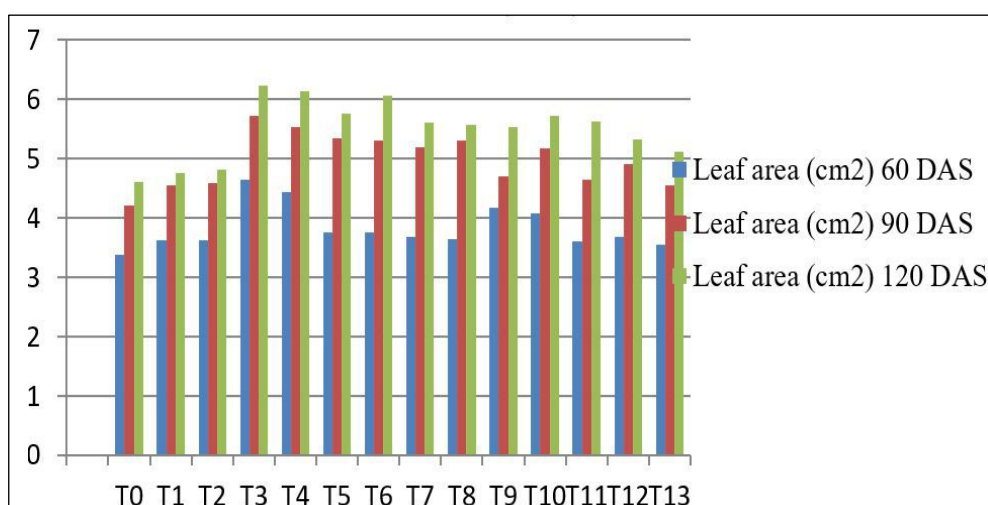


Fig 7: Effect of pre-sowing seed treatments on Leaf area (cm²) at 60, 90 and 120 DAS of Ber

4. Conclusion

On the basis of experimental findings, it can be concluded that among the different of pre-sowing seed treatments soaking in GA₃ @ 250 ppm for 24 hours (T₃) was the most effective treatment for minimum days taken for ber seed germination, highest germination (%), maximum height of seedling, maximum diameter of seedling, longest intermodal length, largest number of leaves per plant and maximum leaf area.

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