



Effect of Seed Treatments on Seed Germination Studies in Ramphal (*Annona reticulata* L.)

Jeevanantham V ^{a++*}, Elakkuvan S ^{b#}, Kumar S ^{c#}
and Karthikeyan A ^{d#}

^a Department of Horticulture, Faculty of Agriculture, Annamalai University, Annamalai Nagar, Chidambaram, Tamil Nadu-608002, India.

^b Department of Horticulture, Agricultural College and Research Institute, Eachangkottai, India.

^c Horticulture and Forestry Research Station, Kodaikanal, India.

^d Regional Research Station, Virudhachalam, Tamil Nadu, India.

Authors' contributions

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

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ABSTRACT

Annona reticulata L., commonly known as bullock's heart, is a tropical fruit-bearing tree belonging to the Annonaceae family. It is prized for its flavourful and nutritious fruit. The plants are used for anti-inflammatory, stress reliever, antimutagenic, curing wounds, anti-anxiety, and spasmolytic properties and have all been used. In this fruit, the seeds are poor due to seed dormancy, and viability is easily lost. To overcome this problem in bullock's heart, an experiment was laid out in the

⁺⁺ Post Graduate Student;

[#] Assistant Professor;

^{*}Corresponding author: E-mail: jjivanchandru@gmail.com;

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Floriculture unit, Department of Horticulture, Faculty of Agriculture, Annamalai University, Tamil Nadu on seed germination studies in Ramphal (*Annona reticulata* L.). Amongst the different pre-sowing seed treatments, treatment with Gibberellic Acid @ 1000 ppm significantly improved the germination percentage (90.63%) and reduced the number of days taken for initiation of seed germination (17.02 days), days taken for 50 percent of seed germination (22.44 days), span of seed germination (25.09 days) as compared to control. This experiment concluded that Gibberellic Acid @ 1000 ppm treated seeds have better seed germination characteristics.

Keywords: Bullock's heart; *Annona reticulata*; gibberellic acid; seed treatment; seed germination.

1. INTRODUCTION

Annona reticulata, commonly known as bullock's heart, is a tropical fruit-bearing tree belonging to the Annonaceae family. This species is native to the tropical regions of the Americas, with its origins traced back to the West Indies, Central America, and Northern South America. Over time, *Annona reticulata* has spread and naturalized across various parts of the world including Southeast Asia, Africa, and India, where it thrives in warm, tropical climates. *Annona reticulata* is prized for its flavourful and nutritious fruit. It is commonly consumed fresh, either scooped out of the skin with a spoon or blended into smoothies and desserts. *Annona reticulata* is a small, glabrous tree that grows in tropical conditions. It has many lateral branches and grows 6.0-7.5 m in height. The stems are cylindrical with shorter and brown-colored hairs and lenticels. Leaves are acute, oblong, lanceolate, rounded, cusped, or obtuse leaf-based. The upper surface of the leaves is glabrous and the lower surfaces have a few spreading hairs. *Annona reticulata* is heart-shaped with the surface rough and the ripening time color changed from yellow to yellowish red. Fruits are delicious and used for diabetes as it has blood glucose-lowering properties. The seeds are dark-colored with a smooth surface (Basappa et al., 2024). The nutritional composition of 100 grams of bullock's heart fruit is as follows: 101 kcal, 25.2 g of carbohydrates, 2.4 g of fiber, 0.6 g of fat, 0.2 g of saturated fat, 1.7 g of protein, 33 IU of vitamin A, 19.2 mg of vitamin C, 0.1 mg of thiamine, 0.1 mg of riboflavin, 0.5 mg of niacin, 0.2 mg of vitamin B₆, 0.1 mg of pantothenic acid, 30 mg of calcium, 0.7 mg of iron, 18 mg of magnesium, 21 mg of phosphorus, and 382 mg of potassium (Karande et al., 2023). This study used some organic and chemical substances like jeevamirtham, gibberellic acid, potassium nitrate, sulfuric acid and thiourea. They are used to play a major role in seed germination. The role is (i) Jeevamirtham - It is believed to stimulate the production of

growth-promoting hormones, which can further accelerate germination and enhance seedling vigor, (ii) Gibberellic acid (GA₃) - It also induces the synthesis of hydrolytic and gluconeogenic enzymes, particularly in monocots and dicots, promoting vigorous seedling growth during early germination stages, (iii) Potassium nitrate (KNO₃) - Using potassium nitrate (KNO₃) in seed treatment has been linked to breaking dormancy, increasing germination rates and improving seedling growth and uniformity across various plant species, (iv) Sulfuric acid (H₂SO₄) - It is commonly used in seed treatment to improve germination by softening or scarifying hard seed coats, allowing water and gases to penetrate more easily. Increased water absorption from sulfuric acid treatment, (v) Thiourea - It is commonly used in seed treatments to improve germination, especially under stress conditions. *Annona spp* are commonly propagated through seeds and the seeds are poor due to seed dormancy, and viability is easily lost. To overcome this problem in ramphal, the study must improve germination characteristics.

2. MATERIALS AND METHODS

The experiment was conducted in the Floriculture unit, Department of Horticulture, Faculty of Agriculture, Annamalai University, Cuddalore, Tamil Nadu during the year 2024 (January - June). Selecting the seeds from uniform-sized quality and disease-free Ramphal fruits. Seeds are collected from local cultivar varieties in Vaiyampatti village, Tiruchirappalli district of Tamil Nadu. The seeds are extracted after being washed in water and selected as good-quality seeds. The experiment is laid out in Completely Randomized Design (CRD) with 16 treatments and 3 replications. The different treatments like T₁ - Jeevamirtham @ 5%, T₂ - Jeevamirtham @ 10%, T₃ - Jeevamirtham @ 15%, T₄ - GA₃ @ 500 ppm, T₅ - GA₃ @ 1000 ppm, T₆ - GA₃ @ 1500 ppm, T₇ - KNO₃ @ 1000 ppm, T₈ - KNO₃ @ 2000 ppm, T₉ - KNO₃ @ 3000 ppm, T₁₀ - H₂SO₄ @ 1%, T₁₁ - H₂SO₄ @ 2%, T₁₂ - H₂SO₄

@ 3%, T₁₃ - Thiourea @ 1%, T₁₄ - Thiourea @ 2%, T₁₅ - Thiourea @ 3% and T₁₆ - Control. After the treated seeds were sowed in a 2-3 cm depth in polybags filled with media comprising red soil, sand, and Farm yard manure (FYM) (2:1:1). The observations about seed germination characteristics were recorded daily up to 150 days after sowing *i.e.* germination percentage, days taken for initiation of seed germination, days taken for 50 percent of seed germination and span of seed germination. The experimental data recorded on various characteristics during the investigation were analyzed statistically using the method of analysis of variance (ANOVA) for Complete Randomized Design (CRD) by Fisher and Yates (1963). Analysis of variance (one-way classified data) for each characteristic was performed using 'WASP 2.0' software. The significance of various treatments was judged with the help of the "F" value (test) at a 5% level of significance. To elucidate the nature and the magnitude of effects, Standard Error of Difference (S.Ed) and Critical Difference (C.D) at a 5 per cent level of significance were included in the summary table.

3. RESULTS AND DISCUSSION

The data relating to germination percentage, days taken for initiation of seed germination, days taken for 50 percent of seed germination, and span of seed germination in Ramphal as influenced by the different pre-sowing treatments are given in Table 1. The highest germination percentage (90.63) is registered in T₅ (GA₃ @ 1000 ppm) followed by T₈ (KNO₃ @ 2000 ppm) which showed 87.68 and the lowest germination

percentage (50.18) was recorded in T₁₆ (control). Gibberellic acid's antagonistic impact on germination may be the cause of the increase in germination (Brain and Hemming, 1958; Wareing et al., 1968) and GA₃ has a role in the cytological enzyme activation process that triggers the α -amylase enzyme which transforms insoluble starch into soluble sugars. Energy from these sugars is needed for several metabolic and physiological processes connected to germination. The results conform with the earlier reports by Hazarika et al. (2023) in papaya, Hossain et al. (2023) in papaya, Sabarad et al. (2023) in wood apple, Anagha et al. (2024) in dragon fruit, Bahadure et al. (2024) in lime and Deshi et al. (2024) in apple.

The minimum days taken for initiation of seed germination is registered in T₅ (GA₃ @ 1000 ppm) followed by T₈ (KNO₃ @ 2000 ppm) which showed 17.02 days and 17.85 days respectively to T₁₆ control (33.98 days). GA₃ synthesized by the enlarged embryo acts on the living cells and causes *de novo* synthesis of hydrolyzing enzymes particularly α -amylase and protease which converts the starch into simple sugars during germination (Paleg, 1965). The enzymes that weaken the seed coat and permit the axis to break through are the other enzymes that GA₃ activates. For the radicle to break through the endosperm and seed coat that limits its growth, GA₃ also increases cell elongation (Hartmann and Kester, 1979). The results conform with the earlier reports by Garg and Bahadur (2023) in guava, Hossain et al. (2023) in papaya, Anagha et al. (2024) in dragon fruit, Bahadure et al. (2024) in lime and Deshi et al. (2024) in apple.



Fig. 1. General view of the experimental plot

Table 1. Effect of seed treatments on germination characteristics of Ramphal seeds at 150 DAS

Treatments	Germination percentage (%)	Days taken for initiation of seed germination	Days taken for 50 percent of seed germination	Span of seed germination (days)
T ₁ -Jeevamirtham @ 5%	57.94 (49.57)	29.32	42.13	45.37
T ₂ - Jeevamirtham @ 10%	55.33 (48.06)	30.00	43.44	46.88
T ₃ - Jeevamirtham @ 15%	52.84 (46.63)	31.48	45.35	48.74
T ₄ - GA ₃ @ 500 ppm	79.75 (63.26)	22.01	28.98	31.83
T ₅ - GA ₃ @ 1000 ppm	90.63 (72.25)	17.02	22.44	25.09
T ₆ - GA ₃ @ 1500 ppm	82.40 (65.20)	20.92	27.36	30.13
T ₇ - KNO ₃ @ 1000 ppm	77.43 (61.64)	23.06	30.58	33.51
T ₈ - KNO ₃ @ 2000 ppm	87.68 (69.46)	17.85	24.14	26.74
T ₉ - KNO ₃ @ 3000 ppm	74.80 (59.87)	24.19	32.29	35.21
T ₁₀ - H ₂ SO ₄ @ 1 %	66.63 (54.72)	26.94	37.18	40.22
T ₁₁ - H ₂ SO ₄ @ 2 %	63.67 (52.94)	27.88	38.78	41.92
T ₁₂ - H ₂ SO ₄ @ 3 %	60.85 (51.27)	28.61	40.68	43.87
T ₁₃ - Thiourea @ 1 %	71.96 (58.03)	25.11	33.92	36.90
T ₁₄ - Thiourea @ 2 %	85.10 (67.29)	19.64	25.75	28.44
T ₁₅ - Thiourea @ 3 %	69.65 (56.58)	26.11	35.60	38.55
T ₁₆ - Control	50.18 (45.10)	33.98	46.90	50.71
S.Ed	1.00 (0.70)	0.26	0.58	0.63
CD at 5 % (p = 0.05)	2.05 (1.43)	0.54	1.18	1.28

Note: Figures in parenthesis are arc sin transformation values



Fig. 2. Selection of quality seeds

T₅ (GA₃ @ 1000 ppm) registered the minimum days taken for 50 percent of seed germination (22.44) whereas control recorded the maximum number of days taken for 50 percent of seed germination (46.90). The reduction in the number of days required for 50% of germination may be due to the synergistic effects of these two inputs combined (GA₃ and soaking period) which enhance protoplasmic physio-chemical

properties, respiration, nucleic acid metabolism, and other aspects of combined growth. GA₃ is thought to have enhanced the de novo synthesis of hydrolyzing enzymes, specifically protease and amylase. The food that had been hydrolyzed was then used to develop the embryo, which improved germination. Findings similar to the present study were reported by Hakimi et al. (2020) in avocado, Singh and Kaur (2021) in

aonla, Harshita et al. (2021) in Indian blackberry, and Bahadure et al. (2024) in lime.

The minimum days taken for a span of seed germination observed in T₅ (GA₃ @ 1000 ppm) followed by T₈ (Potassium Nitrate (KNO₃) @ 2000 ppm) which showed 25.09 days and 26.74 days respectively than T₁₆ control (50.71 days). The shortest number of days required for seed germination in seeds treated with GA₃ may be attributed to the fact that GA₃ initiates starch hydrolysis, whose translocation enables the completion of seed germination. Findings similar to the present study were reported by Harshita et al. (2021) in Indian blackberry, Singh and Kaur (2021) in aonla, and Bahadure et al. (2024) in lime.

4. CONCLUSION

The findings from the present study, titled Seed Germination Studies in Ramphal (*Annona reticulata* L.), indicate that treating seeds with GA₃ at 1000 ppm significantly enhances germination parameters such as germination percentage, time to initiate germination, time to achieve 50% germination, and the overall germination period. Ramphal, also known as Bullock's heart, suffers from physical dormancy due to its hard seed coat, leading to poor germination rates. Treatment with KNO₃ at 2000 ppm also yielded favorable results, but GA₃ at 1000 ppm proved superior. It is recommended that farmers aiming for successful Ramphal cultivation adopt gibberellic acid at 1000 ppm as a pre-sowing treatment. This approach can improve both seed germination and early growth performance. This study confirms that gibberellic acid is the most effective solution for breaking dormancy and enhancing seed germination in Ramphal. These results demonstrate the potential of GA₃ as a reliable strategy to boost germination success.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that generative AI technologies such as Large Language Models, etc. have been used during the writing or editing of manuscripts. This explanation will include the name, version, model, and source of the generative AI technology and as well as all input prompts provided to the generative AI technology

Details of the AI usage are given below:

1. ChatGPT

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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