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Effect of Different Concentrations of Gibberellic Acid on Seed Germination Behaviour and Seedling Vigour Index of *Betula utilis* D. Don

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Abstract: A germination trial was conducted to study the effect of different concentrations of Gibberellic acid (GA_3) on seed germination behaviour and seedling vigour of *Betula utilis* under Laboratory condition. The seeds collected from Barda Forest in Baspa Valley of Kinnaur district, Himachal Pradesh were treated with six different concentrations of Gibberellic Acid (GA_3) viz., 50, 100, 200, 300, 400 and 500 µg mL^{-1} and control to study their effect on germination behaviour and seedling vigour index. Significant differences were observed in various germination parameters, seedling length and seedling vigour index of seeds treated with different pre-sowing treatments. The maximum germination (79%), mean daily germination (3.76%), germination value (5.79), germination speed (9.07), seedling length (3.77cm) and seedling vigour index (297.83) were recorded in seeds treated with GA_3 100 ppm whereas maximum peak value (1.58) was recorded in seeds treated with GA_3 400 ppm and maximum germination energy (38.00) was recorded in seeds treated with GA_3 500 ppm. The minimum germination (61.33%), mean daily germination (2.92%) and germination speed (7.07) were recorded in seeds treated with GA_3 500ppm whereas minimum peak value (1.26), germination value (3.78), seedling length (2.25cm) and seedling vigour index (143.35) recorded in seeds which were not given any pre-sowing treatment (control) and minimum germination speed (7.07) was recorded in seeds treated with GA_3 500ppm. The present investigation revealed that seeds of *B. utilis* which possess embryo dormancy should be treated with GA_3 100 ppm before sowing in the nursery for breaking seed dormancy and better germination.

Key words: *Betula utilis, Catkins, Germination Parameters, Seedling Vigour Index.*

Betula utilis D. Don syn. *B. bhojpattra* Wall is a moderate-sized multipurpose tree up to 20 m in height with irregular bole and sometimes also exhibiting shrubby growth in the upper limit of vegetation in the temperate Himalayan region. It is commonly known as "Bhojpattra", "Himalayan Silver Birch", or "Indian Paper Birch". The bark of this tree is smooth, shining, reddish-white or white with white horizontal lenticels. The leaves are ovate acuminate, irregularly serrated with 5.0-

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7.5 cm long. It is found in higher altitudinal areas of the temperate Himalayas chiefly at 3,000 to 4,200 m above msl but sometimes even at to 1800 m altitude above msl (Anonymous, 1988). At higher elevations, it is gregarious, occurring either in pure patches, often with undergrowth of *Rhododendron campanulatum*, *R. anthopogon, Juniperus squamata, J, communis* and dwarf willow. In its gregarious form, it marks the upper limit of tree vegetation on the uplands of the Himalayas before the treeless snowy wastes begin (Troup, 1921). It is mainly found in higher temperate and sub-alpine areas of Shimla, Kangra, Kullu, Chamba, Lahaul & Spiti and Kinnaur Districts of Himachal Pradesh (Chauhan,1999).

It is one of the most valuable trees which has been used by people in their day-to-day life since ancient times. The papery bark of a tree was used as a writing material by saints in ancient times. The papery bark is also used as a covering for umbrellas, hooka pipes, and for packing and roofing material of wooden houses in the Himalayan region. The bark is also used by the local communities in performing various religious rites. The leaves are used in the treatment of urinary tract infections, kidney and bladder stones. The wood is used as fuelwood by the local communities and its tender foliage is also used as fodder for domestic animals during harsh winter months. The fungal growth commonly called Bhurja-granthi is used in local traditional medicine (Singh *et al.,* 2012). The species is recommended for afforestation in avalanche-prone areas (Tiwari *et al*.1984). The germination behaviour study of *Betula utilis* seeds was not reported to date in Himachal Pradesh, however, a maximum of 67% germination was reported from seeds of *Betula utilis* collected from Niti provenance of Uttarakhand (Phondani *et al*., 2010).

Keeping in view the importance of this valuable tree and problem in its germination behaviour, a germination study was conducted to evaluate the effect of different concentrations of Gibberellic acid on seed germination behaviour and seedling vigour of *Betula utilis* seeds in the laboratory so that the best pre-sowing treatment could be identified for getting better seed germination and seedling vigour for production of quality nursery stock.

Materials and Methods

***Seed Collection and Processing*:** The catkins of *Betula utilis* were collected from the mature, healthy trees from the natural forest of *Betula utilis* at Barda in Rakchham village of Baspa Valley, Kinnaur, Himachal Pradesh during the first week of October 2022. The seed collection site was situated at 31°21'32.9" N latitude and 78°22' 32.3" E longitude at an altitude of 3358m above msl. The catkins were subsequently brought to the laboratory of ICFRE-Himalayan Forest Research Institute, Shimla. The catkins were dried in the laboratory under ambient room temperature for a week and thereafter seeds were processed and extracted from the catkins. The seeds were then spread on the filter paper and dried in the laboratory. The seeds were then separated from the impurities and subsequently stored in an airtight plastic container under refrigerated conditions. The moisture content of the seeds was taken by oven dry method by placing the seeds in the oven at a temperature of 103 ± 2°C for 17 hours as per International Seed Testing Association (ISTA, 1985, 2010, 2020) and was recorded at 3.80 % for seeds.

***Germination Test*:** Seed germination test was conducted in the Seed Laboratory of HFRI, Shimla. The seeds were pre-soaked in six different concentrations of Gibberellic acid (GA_3) and control for 24 hours before sowing in Petri dishes to study their effect on seed germination. The petri dishes were placed in seed germinator. The details of treatments were Control; GA_3 50 µg mL^{-1}; GA_3 100 µg mL^{-1}; GA_3 200 µg mL^{-1}; GA_3 300 µg mL^{-1}; GA_3 400 µg mL^{-1}; GA_3 500 µg mL^{-1}.

A total of 300 seeds per treatment were sown in Petri dishes containing moist filter paper in the seed germinator in a lot of 100 seeds per replication using a completely randomized design (CRD) as per ISTA (1985, 2010, 2020). 100 seeds were sown in each petri dish and total 21 petri dishes were placed in the seed germinator at a time. The petri dishes containing seeds were irrigated as and when required to keep the seeds moist during the germination period. The seeds were then allowed to germinate at a constant temperature of 20°C in the seed germinator under laboratory conditions. The seed germination was noticed when a radicle emerged. The germination data were taken

daily after the commencement of germination until it was over and constant germination was obtained. The germination started after 5 days of seed sowing and culminated within 28 days. To study the effect of different pre-sowing treatments on various germination parameters viz., germination percentage (GP), mean daily germination (MDG), peak value (PV), germination value (GV), germination speed (GS) and germination energy (GE), the final seedling count was considered. The total germination percentage was calculated at the end of the experiments.

$$GP = \frac{\text{No. of seeds germinated}}{\text{Total no. of seeds sown}} \times 100$$

The Mean daily germination (MDG) was calculated as the cumulative percentage of all the seeds germinated at the end of the test divided by the number of days from sowing to the end of the test.

$$MDG = \frac{\text{Cumulative per cent of seed germinated at the end of the test}}{\text{Days since sowing to the end of the test}}$$

The Peak value (PV) was calculated as the maximum mean daily germination reached at any time during the period of the test (Czabator, 1962).

$$PV = \frac{\text{Cumulative germination per cent}}{\text{Days since sowing}}$$

Germination Value = Peak Value x Final MDG (Czabator, 1962).

Germination energy (GE) was calculated as the percentage of the number of seeds in a given sample which germinate up to the time of peak germination (William, 1985). Germination speed (GS) was calculated as per the method given by Panwar and Bhardwaj (2007).

Germination speed= $\sum (n/t)\ GI{=}i{=}1 \sum n(tiGi)\ i{=}1$

where, n = no. of newly germinated seed at time t; t = no. of days since sowing

15 days after the completion of germination, ten seedlings from each replication of all the treatments were randomly selected and measured for total seedling length.

Seedling Vigour Index = Germination percent x Total seedling length (cm) (Abdul-Baki and Anderson, 1973).

The data of germination per cent, mean daily germination (MDG), peak value (PV), germination value (GV), germination energy (GE), germination speed (GS), total seedling length and seedling vigour index were subjected to one-way analysis of variance (ANOVA) to establish the significance of differences between the treatments ($p \leq 0.05$) by Duncan's test using SPSS software - a statistical package for social sciences.

Results and Discussion

The results of the germination parameters, seedling length and seedling vigour as affected by different concentrations of Gibberellic acid (GA_3) are given in Table 1 and Table 2**.** A perusal of data from Table 1 and 2 revealed that significant differences were observed in germination parameters viz., germination percentage, mean daily germination, germination value, germination energy,

Table 1. Germination Parameters of Betula utilis as affected by different Concentration of Gibberellic acid under Laboratory condition

Pre-sowing Treatments GA3 (µg mL^{-1})	Germination Percent (%)	Mean daily germination	Peak value	Germination Value	Germination Energy (%)	Germination Speed (%)
Control	63.00^{de}	3.00^{d}	1.26^{a}	3.78^{b}	37.00^{a}	7.29^{d}
50	66.67^{d}	3.33^{c}	1.33^{a}	4.47^{ab}	36.33^{a}	7.40^{cd}
100	79.00^{a}	3.76^{a}	1.54^{a}	5.79^{a}	33.67^{ab}	9.07^{a}
200	75.00^{b}	3.57^{b}	1.33^{a}	4.76^{ab}	27.67^{b}	7.84^{bc}
300	71.00^{c}	3.38^{c}	1.35^{a}	4.56^{ab}	34.33^{ab}	7.87^{b}
400	62.00^{e}	2.95^{d}	1.58^{a}	4.66^{ab}	28.00^{b}	7.15^{d}
500	61.33^{e}	2.92^{d}	1.33^{a}	3.90^{b}	38.00^{a}	7.07^{d}
($p \leq 0.05$)	<.0001	<.0001	>.0001	>.0001	>.0001	<.0001
S. Em ±	1.73	0.08	0.15	0.55	3.23	0.20
C. D. Value at 5% Level	3.78	0.17	0.34	1.19	7.03	0.44

Note: Mean with the same letters are not significantly different @5% level of significance

germination speed, seedling length and seedling vigour index of *Betula utilis* seeds treated with different concentrations of Gibberellic acid. The maximum germination of 79.00% was recorded in seeds treated with gibberellic acid 100 µg mL^{-1} and is significantly higher than all other treatments and the minimum germination of 61.33% was recorded in seeds treated with gibberellic acid 500 µg mL^{-1}.

The maximum mean daily germination (3.76) was recorded in seeds treated with gibberellic acid 100 µg mL^{-1} and is significantly higher than all other treatments whereas the minimum mean daily germination (2.92) was recorded in seeds treated with gibberellic acid 500 µg mL^{-1}. The maximum peak value (1.58) was recorded with gibberellic acid 400 µg mL^{-1} and is statistically at par with all other treatments whereas the minimum peak value (1.26) was recorded in untreated seeds. The maximum germination value (5.79) was also recorded with gibberellic acid 100 µg mL^{-1} and is significantly higher than all other treatments whereas the minimum germination value (3.78) was recorded in untreated seeds.

The maximum germination energy (38.00%) was recorded with gibberellic acid 500 µg mL^{-1} and is statistically at par with other treatments control, 50, 100 and 300 µg mL^{-1} but is significantly higher than the treatments of 200 and 400 µg mL^{-1} whereas the minimum germination value (27.67%) was recorded in seeds treated with gibberellic acid 200 µg mL^{-1}. The maximum germination speed (9.07%) was also recorded with gibberellic acid 100 µg mL^{-1} and is significantly higher than all other treatments whereas the minimum germination speed (7.07%) was recorded with gibberellic acid 500 µg mL^{-1}.

Similarly, a perusal of data from Table 2 revealed that the maximum average seedling length of 3.77 cm was recorded with gibberellic acid 100 µg mL^{-1} and is significantly higher than all other treatments and is followed by 2.86 cm with gibberellic acid 200 µg mL^{-1}. The minimum seedling length of 2.25 cm was recorded in untreated seeds.

The seedling vigour index (SVI) also varied significantly with different treatments. The maximum seedling vigour index of 297.83 was recorded in seeds treated with gibberellic acid 100 µg mL^{-1} and is significantly higher than all other treatments and followed by seedling vigour index of 214.66 with gibberellic acid 200 µg mL^{-1}, 197.64 with gibberellic acid 300 µg mL^{-1}, 188.68 with gibberellic acid 50 µg mL^{-1}, 169.93 with gibberellic acid 400 µg mL^{-1} and 167.43 with gibberellic acid 500 ppm µg mL^{-1} in a decreasing order whereas the minimum seedling vigour index of 143.35 was recorded in untreated seeds µg mL^{-1}.

A critical review of results from Table 1 and Table 2 reveals that the highest value of germination per cent (79.00%), mean daily germination (3.76), germination value (5.79), germination speed (9.07), seedling length (3.77 cm) and seedling vigor index (297.83) among different pre-sowing treatments was recorded in the seeds pre-soaked in gibberellic acid 100ppm for 24 hours whereas the highest value

Table 2. Seedling Length and Seedling Vigour Index of Betula utilis as affected by different Concentration of Gibberellic acid under Laboratory condition

Pre-sowing Treatments GA_3 (µg mL^{-1})	Germination Percent (%)	Seedling length (cm)	Seedling vigour index
Control	63.00de	2.25c	143.35f
50	66.67d	2.83b	188.68d
100	79.00a	3.77a	297.83a
200	75.00b	2.86b	214.66b
300	71.00c	2.79b	197.64c
400	62.00e	2.74b	169.93e
500	61.33e	2.73b	167.43e
($p \leq 0.05$)	<.0001	<.0001	<.0001
S. Em ±	1.73	0.07	1.77
C. D. Value at 5% Level	3.78	0.15	3.85

Note: Mean with the same letters are not significantly different @5% level of significance

of germination energy (38.00%) was recorded in the seeds pre-soaked in gibberellic acid 500ppm for 24 hours and peak value (1.58) was recorded in seeds pre-soaked in gibberellic acid 400ppm. The lowest value of germination per cent (61.33%), mean daily germination (2.92) and germination speed (7.07) were recorded with gibberellic acid 500ppm whereas the lowest value of germination value (3.78), seedling length (2.25cm) and seedling vigour index (143.35) were recorded in seeds which were not given any pre-sowing treatments and lowest value of germination energy (27.67) was recorded in the seeds treated with gibberellic acid 200 $\mu g\ mL^{-1}$ for 24 hours.

The highest germination per cent (79%) observed in treatment GA_3 100ppm may be attributed to the optimum concentration of gibberellic acid that helped in overcoming embryo dormancy in *Betula utilis*. The lowest germination (61.33%) observed in seeds treated with gibberellic acid 500ppm for 24 hours may be due to an overdose of gibberellic acid which resulted in a decrease in germination per cent. It is further observed that with the increase in the concentration of gibberellic acid beyond 100 $\mu g\ mL^{-1}$, the germination per cent of *Betula utilis* seeds tends to decrease from a maximum of 79% to a minimum of 61.33%. The declining trend in germination per cent was also observed in seeds of *Hedychium spicatum* treated with GA_3 beyond 400 $\mu g\ mL^{-1}$ (Negi *et al.*, 2021).

The role of gibberellic acid - a potent plant growth regulator and germination promoter has been found to break seed dormancy in many plant species such as *Pinus kesiya* and *Schima khasiana* (Verma and Tandon, 1988), *Picea smithiana* (Singh, 1989), *Quercus leucotrichophora* (Singh *et al*., 1995 and Gautam and Bhardwaj, 2001), *Cassia fistula* (Kamel and Nada, 1986), *Acer oblongum* (Masoodi, 1990), *Quercus glauca* (Sankhyan *et al*., 2008) and *Fraxinus xanthoxyloides* (Negi and Sharma, 2011). The soaking of *Betula utilis* seeds in different concentrations of gibberellic acid for 24 hours resulted in improved germination percentage, seedling length and seedling vigour index. The exogenous application of gibberellic acid in promoting seed germination was also observed in *Picea smithiana* (Chandra and Chauhan, 1976); *Pinus caribaea* and *Pinus patula* (Bhatnagar, 1980); *Picea obovata* and *Pinus sylvestris* (Larionova, 1997); *Pinus wallichiana* (Gera *et al*., 2003), *Aconitum heterophyllum* (Singh *et al*., 2000), *Pinus wallichiana* (Lavania *et al*., 2006) and *Hedychium spicatum* (Negi *et al*., (2021).

Conclusion

The findings of the present investigation reveal that seeds of *Betula utilis* possess embryo dormancy. Therefore, it is recommended that seeds should be treated with gibberellic acid 100 ppm before sowing to break dormancy and to get better germination and seedling vigour in the nursery.

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