Indian Forester, **143** (11) : 1105-1111, 2017
http://www.indianforester.co.in
ISSN No. 0019-4816 (Print)
ISSN No. 2321-094X (Online)

EFFECT OF STRATIFICATION AND GIBBERELLIC ACID ON GERMINATION AND SEEDLING GROWTH OF *ACER ACUMINATUM* IN NORTH WESTERN HIMALAYAS

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ABSTRACT

The presence of physiological dormancy in *Acer acuminatum* (Wall. ex D. Don) seeds poses a challenge for its germination. In the present study we investigated the effect of stratification and gibberellic acid (GA_3) on seed germination and seedling growth of *A. acuminatum* for two years. The seeds were subjected to five stratification periods of twenty day interval i.e. 0, 20, 40, 60 and 80 days; three stratification temperatures viz., room temperature, 3±1°C and -3±1°C and finally treated with distilled water and 200 ppm GA_3 prior to sowing. Half of the seeds were sown in the laboratory conditions while the other half was sown in polybags in the shade net house. The results showed that the seeds subjected to 60 day stratification provided significantly maximum germination (43.61% and 38.06%) in both conditions. Similarly, 3±1°C stratification provided significantly best germinability and seedling growth. GA_3 (200 ppm) application also proved to enhance germination of the seeds in both conditions.

Key words: *Acer acuminatum*, Dormancy, Stratification, Germination, Seedling growth.

Introduction

Maples are a diverse and versatile group of trees and shrubs, well known for their autumnal colour. Although it is primarily a temperate group, a few maples grow into the tropics with only one species, *Acer laurinum* extends into the southern hemisphere. With a natural distribution across the globe, having centre of diversity in China, maples range across the northern hemisphere from North America to Japan including much of Europe, the very north of Africa, the Middle East, Central Asia, Himalayas and East Asia (Gibbs and Chen, 2009). Of maples, *Acer acuminatum* (Wall. ex D. Don) is endemic to the Himalayas, distributed in northern India, Nepal, northern Pakistan and China. *A. acuminatum* is a small to moderate sized (up to 15 m tall), deciduous, dioecious tree (Renner *et al*., 2007), growing in open ravines on shady aspects between 2400-3300m asl. But, the species is least explored, yet very useful species in the locality, especially as fodder for domestic animals during winters. Fruit ripening also takes place in this season but due to heavy lopping, very few fruit bearing trees are left for natural regeneration being successful. The continuous exploitation of this species is leading to decrease in its population to a greater extent. Additionally, the presence of dormancy further reduces the chances of its seed germination. Different degree of seed dormancy and germinability is found in the genus *Acer* (Pinfield and Dungey, 1985). The range extends from non dormant seed of *A. saccharinum* (Tomaszewska, 1979a) through a number of intermediate classes, to the deeply dormant seeds of *A. tataricum* (Nikolaeva, 1969).

Mass scale propagation through conventional and non-conventional methods and their *in-situ* rehabilitation may help in its conservation and perpetuation. But, germination of fresh as well as non-stratified seeds of *A. acuminatum* is very poor. Stratification is a method employed to break dormancy of such seeds and to ensure uniform and quick germination of seeds in many forest species (Tiwari *et al*., 2016). To break seed dormancy, seeds are usually incubated on a moist substrate at low temperature (0-10^{0}C). For many species, 5^{0}C is optimal (Baskin and Baskin, 2014), but in some species temperature lower than 5^{0}C is more effective. Therefore, to break dormancy, the stratification treatments and GA_3 might be helpful in this species to enhance its germinability and growth performance under laboratory as well as nursery conditions. But, till date, no work has been done in this regard in the species, so an exploratory study was undertaken to enhance the germination and to find suitable stratification methods.

The cold moist stratification for a period of 60 days at 3^{0}C provided better germination and seedling growth of *Acer acuminatum*.

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Material and Methods

This experiment was conducted in the Department of Silviculture and Agroforestry, Dr. Y.S. Parmar UHF, Nauni, Solan–Himachal Pradesh (India), located at $30^0 51'$ North latitude and $76^0 11'$ East longitude at an elevation of 1250m asl. The mature seeds were collected from Shimla Forest Circle during Oct-Nov of 2010 and 2011; packed in gunny bags and transported to the University. As the frequency of parthenocarpic fruits was observed to be very high (about 60-65%); a flotation test was performed to separate viable seeds from non-viable seeds. Wings of the samaras were first removed and the wingless fruits (seeds hereafter) were soaked in water for 24h, after which seeds that did not sink were discarded.

The seeds were stratified by keeping them in moist sand in polythene bags for entire period. Three batches of seeds were placed for stratification for 0, 20, 40, 60 and 80 days (P_1, P_2, P_3, P_4 and P_5, respectively) at room temperature, 3 ± 1^0C and -3 ± 1^0C ($T\text{-}_1$, $T\text{-}_2$ and $T\text{-}_3$, respectively). Thereafter treated with distilled water (G_1) and 200ppm GA_3 (G_2). The germination study was carried out by putting the seeds in germinator at 20±1°C. While to know the seedling growth, half of the seeds (from each treatment) were sown in the polybags in the shade net house. The observations were recorded during the end of first growing season. Same experiment was again repeated in 2011, and the data were analyzed using SPSS 16.0 package after pooling.

All the germinability parameters were recorded as per standard practice. The germination value was calculated as given by Czabator (1962), while, the germination index was calculated by dividing the total number of seeds germinated at the end of the experiment with the time taken for 50% germination. The Dickson quality index of the seedling was calculated as given by Dickson *et al*. (1960):

$$\text{Quality index} = \frac{\text{Total dry weight of seedling (g)}}{\dfrac{\text{Seedling height (cm)}}{\text{collar diameter (cm)}} + \dfrac{\text{Shoot weight (g)}}{\text{Root weight (g)}}}$$

Results and Discussion

Effect of stratification period

Acer acuminatum seeds were stratified for 0 (P_1), 20 (P_2), 40 (P_3), 60 (P_4) and 80 days (P_5) to observe its effect on various germination parameters viz., germination per cent (GP), germination capacity (GC), germination value (GV) and germination index (GI). It was found that germination increased significantly as the stratification period increased from 0 to 60 days. The results (Table 1) revealed significantly highest GP (43.61%), GC (86.39%), GV (4.10) and GI (2.44), when seeds were stratified for 60 days (P_4). The results are in line with that for *Acer trautvetteri* (Mustafa, 2007), *A. opalus* (Gleiser *et al*., 2004) for 3 months, *A. pseudoplatanus* for 12 weeks and *A. platanoides* for 18-19 weeks (Pinfield *et al*., 1990) stratification, all of which resulted in higher germination parameters than that of non-stratified seeds.

The similar trend was observed for seedling growth parameters under nursery conditions (Table 2), for which the significantly maximum germination (38.06%), plant height (15.05 cm), collar diameter (3.13 mm), total dry weight (0.463 g), root:shoot ratio (0.54) and quality index (0.071) was recorded when seeds were stratified for 60 days (P_4). This represent 80.67% more plant height, 299.14% more total dry weight and 294.44% more quality index over the respective control treatments for these parameters. Similarly, 75 days stratification of *Cedrus deodara* seeds provided maximum growth and biomass of the seedlings in nursery for Sofi and Bhardwaj (2008). Lakhanpal and Kumar (1995) have also reported higher shoot height, root length and stem diameter growth ranging from 11.6 cm to 20.6 cm, 8.3 cm to 14 cm and 4.2 mm to 5.0 mm, respectively for *Pinus gerardiana* seedlings.

Effect of stratification temperature

Regarding stratification temperature significantly maximum GP (33.92%), GV (3.24) and GI (1.88) were observed by stratifying seeds at 3 ± 1^0C (Table 1). The increase in seed germinability might be due to increase of gibberellins in the seeds due to cold-moist treatment (Koornneef *et al*., 2002). Low temperature treatment also influences the balance of germination promoting (GA) and germination inhibiting phytohormones (ABA). For the oriental beech, eight weeks of cold-stratification reduced the ABA level in the embryonic axis by 15-fold compared to the control treatment, thus indicated an inverse relationship between ABA level and germination process (Soltani, 2003). The combination of low temperature and high moisture level appear to trigger off biochemical changes in seeds to transform complex food substances into simpler forms, which are utilized by the growing embryo during germination (Zhang *et al.,* 2012). Similarly, Drăghici and Abrudan (2011) obtained highest percentage of germination in *A. platanoides* when seeds were stratified at 3^0C in the moist medium. It was also apparent that there was less germination and highest GC at -3±1°C in comparison to 3 ± 1^0C. Freezing the seeds (-5 to -10^0C) of *Acer campestre, A. ginnala, A. negundo, A. palmatum, A. platanoides* and *A. pseudoplatanus* during stratification also prevented germination completely with both scarified and intact samaras (Toth and Garrett, 1989).

Table 1: Effect of stratification and GA_3 treatments on germinability of *Acer acuminatum* seeds.

Treatment	Germination (%)	Germination capacity (%)	Germination value	Germination index
Stratification period (P)				
P_1 : Control	7.08 (15.32)	53.61 (47.07)	0.21	0.30
P_2 : 20 days	22.50 (27.99)	64.72 (53.85)	1.49	1.23
P_3 : 40 days	32.64 (34.65)	82.92 (65.79)	2.64	1.78
P_4 : 60 days	43.61 (41.21)	86.39 (68.56)	4.10	2.44
P_5 : 80 days	36.81 (37.20)	80.69 (64.07)	3.21	2.05
$CD_{0.05}$	**1.60**	**1.58**	**0.27**	**0.16**
Stratification temperature (T)				
T_1 : Room temp.	21.58 (24.90)	68.67 (56.48)	1.37	1.21
T_2 : 3±1°C	33.92 (32.38)	73.00 (59.59)	3.24	1.88
T_3 : -3±1°C	25.83 (27.35)	79.33 (63.53)	2.25	1.41
$CD^{0.05}$	**1.24**	**1.23**	**0.21**	**0.13**
Gibberellic acid (G)				
G_1 : Control	24.06 (26.28)	71.56 (58.35)	1.85	1.32
G_2 : GA_3 (200ppm)	30.17 (30.14)	75.78 (61.39)	2.73	1.67
$CD_{0.05}$	**1.01**	**1.00**	**0.17**	**0.10**

Figures in parentheses are arc sine transformed values.

Under nursery conditions, the seedling growth followed a more or less similar trend (Table 2). Stratifying seeds at 3±1°C (T_2) resulted in significantly maximum germination (29.17%), plant height (14.19 cm), collar diameter (3.03 mm), root length (14.78 cm), total dry weight (0.369 g), root:shoot ratio (0.46) and quality index (0.055) compared to other treatments. For Polat (2003), scarification and stratification (7°C) of *Juglans regia* resulted in the highest emergence rate (96.66%) and the lowest number of days to emergence (90 days). The results thus find support from the work of Sofi (2005) who reported that moist stratification at 2-3°C exhibited better germination and seedling characteristics in *Cedrus deodara*. Similar findings have also been reported by Malik (2007) for *Pinus gerardiana* and Pawak (1993) for *Callitropsis nootkatensis* seedlings.

Effect of gibberellic acid (GA_3)

Application of gibberellic acid at 200ppm concentration (G_2) demonstrated a marked influence on germination (Table 1) and seedling growth parameters (Table 2) of *Acer acuminatum* seeds. Significantly highest GP (30.17%), GC (75.78%), GV (2.73) and GI (1.67) resulted when seeds were treated with 200ppm GA_3 (G_2). The high germinability of GA_3 treated seeds might be attributed to increase in gibberellins content of seeds during treatment (Taylor and Wareing, 1979). Similarly, the application of exogenous GA_3 (100ppm) promoted dormancy breaking and increased the final germination of *Pinus gerardiana* seeds (Kumar *et al*., 2013). Meghwal (2007) also reported significant increase in germination after application of 250 and 500ppm GA_3 in *Cordia myxa*. Whereas, Nasiri (2008) for *Acer monospessulanum* and Ertekin *et al*. (2011) for *A. platanoides* did not find any significant increase in seed germination by the application of GA_3 after chilling. Goldstein and Loescher (1981) also observed that exogenous gibberellin, cytokinin and ethylene were ineffective in breaking dormancy of *A. macrophyllum* seeds.

As far as seedling growth parameters were concerned (Table 2), the application of 200ppm GA_3 provided significantly maximum germination (24.67%) and all other seedling growth parameters, studied. Similarly, Sofi and Bhardwaj (2008) observed maximum germination and seedling growth of *Cedrus deodara* after the application of 300 ppm GA_3 for 24h. Similar findings have also been reported by Beyhan *et al*. (1999) in *Corylus* sp., Kabar (1989) in *Pinus bracta* and *Thuja orientalis* and Verma and Tandon (1988) in *Pinus khasiana* seedlings.

Interaction effects

The interaction effect of stratification period and temperature (PxT) treatment on germinability parameters of *A. acuminatum* seeds has been depicted in Table 3. The results revealed maximum GP (55.00%), GC (88.33%), GV (5.79) and GI (3.06), when seeds were stratified for 60 days at 3±1°C (P_4T_2). However, the inferior values were recorded for non stratified seeds kept at room temperature (P_1T_1) were sown. Similarly, moist cold stratification at 1–5°C for 3 months was required to break dormancy of *Acer platanoides* and *A. pseudoplatanus* samaras (Pawłowski, 2010). On the other hand, samaras of *A. platanoides* required 17 week cold stratification at 1°C for full germination (Tomaszewska, 1979b). The results were also found to be in agreement with the findings of Tanaka *et al*. (1991) for *Alnus rubra* and Chien *et al*. (1998) for *Taxus*

Table 2: Effect of stratification and GA_3 treatments on germination and seedling growth of *A. acuminatum*.

Treatment	Germination (%)	Plant height (cm)	Collar diameter (mm)	Root length (cm)	Dry shoot weight (g)	Dry root weight (g)	Total dry weight (g)	Root: shoot ratio	Dickson quality index
Stratification period (P)									
P_1 : Control	6.67 (14.83)	8.33	2.12	9.08	0.085	0.032	0.116	0.37	0.018
P_2 : 20 days	16.11(23.31)	11.66	2.47	12.18	0.152	0.062	0.213	0.40	0.030
P_3 : 40 days	22.50 (27.89)	14.14	2.81	13.41	0.195	0.084	0.279	0.43	0.038
P_4 : 60 days	38.06 (38.00)	15.05	3.13	15.81	0.297	0.167	0.463	0.54	0.071
P_5 : 80 days	28.61 (32.12)	13.74	2.98	14.66	0.233	0.109	0.342	0.46	0.051
$CD_{0.05}$	**1.46**	**0.31**	**0.07**	**0.32**	**0.007**	**0.005**	**0.011**	**0.01**	**0.003**
Stratification temperature (T)									
T_1 : Room temp.	17.17 (23.79)	10.50	2.38	11.18	0.145	0.062	0.207	0.41	0.030
T_2 : 3±1°C	29.17 (31.69)	14.19	3.03	14.78	0.245	0.124	0.369	0.46	0.055
T_3 : -3±1°C	20.83 (26.21)	13.06	2.70	13.12	0.186	0.086	0.272	0.44	0.039
$CD_{0.05}$	**1.13**	**0.24**	**0.05**	**0.25**	**0.006**	**0.004**	**0.009**	**0.01**	**0.002**
Gibberellic acid (GA3)									
G_1 : Control	20.11 (25.60)	12.01	2.54	12.29	0.177	0.082	0.258	0.43	0.038
G_2 : GA_3 (200ppm)	24.67 (28.85)	13.16	2.86	13.76	0.208	0.100	0.307	0.45	0.045
$CD_{0.05}$	**0.92**	**0.20**	**0.04**	**0.20**	**0.005**	**0.003**	**0.007**	**0.01**	**0.002**

Figures in parentheses are arc sine transformed values.

Table 3: Effect of stratification period and temperature (PxT) on germinability of *A. acuminatum* seeds.

Treatment (PxT)	Germination (%)	Germination capacity (%)	Germination value	Germination index
P_1T_1	7.08 (15.32)	51.67 (45.94)	0.29	0.36
P_2T_1	22.50 (28.15)	54.58 (47.64)	1.21	0.98
P_2T_2	27.50 (31.53)	60.83 (51.30)	2.25	1.48
P_2T_3	17.50 (24.30)	78.75 (62.60)	1.01	1.24
P_3T_1	25.83 (30.42)	77.92 (62.05)	1.67	1.46
P_3T_2	40.83 (39.66)	83.75 (66.33)	3.84	2.23
P_3T_3	31.25 (33.87)	87.08 (68.98)	2.41	1.64
P_4T_1	31.26 (33.89)	83.75 (66.38)	2.29	1.77
P_4T_2	55.00 (47.87)	88.33 (70.30)	5.79	3.06
P_4T_3	44.58 (41.86)	87.09 (69.01)	4.21	2.48
P_5T_1	28.33 (32.06)	75.42 (60.37)	1.90	1.59
P_5T_2	46.25 (42.81)	82.50 (65.28)	4.32	2.61
P_5T_3	35.83 (36.72)	84.17 (66.56)	3.42	1.93
$CD_{0.05}$	**2.77**	**2.74**	**0.47**	**0.28**

Figures in parentheses are arc sine transformed values.

mairie seeds. Similarly, Pinfield *et al*. (1990) found better germination of *A. pseudoplatanus* and *A. platanoides* seeds after stratifying them at 5°C for 12 and 18-19 weeks, respectively.

Similarly, under nursery conditions, the interaction effect of P_4T_2 (Table 4) revealed significantly better germination (45.42%), plant height (17.77 cm), collar diameter (3.51 mm), root length (18.15 cm), total dry weight (0.667 g), root:shoot ratio (0.62) and quality index (0.102) when seedlings were raised from seed stratified for 60 days at 3±1°C. The results thus, get support from the work of Sofi (2005) for *Cedrus deodara* seeds. Similar observations were also made by Borghetti *et al*. (1986) in *Pinus leucodermis* and Tanaka *et al*. (1991) in *Thuja plicata* seedlings.

The present investigation also revealed the effective influence of stratification period and gibberellic acid (PxG) treatments on germination parameters (Table 5). The significantly maximum germination of 47.78% resulted when seeds stratified for 60 days were treated with GA_3 (P_4G_2). The other germination parameters viz., GC, GV and GI also exhibited a similar trend. The high germinability in P_4G_2 combination might be linked to gibberelllin increase as reported by Tomaszewska (1976) for *Acer platanoides* and Taylor and Wareing (1979) for *Pinus lambertiana* seeds during treatment. The effect of GA_3 might have resulted in increased synthesis of amylase or breakdown of starch into monosaccharide resulting in better germinability. The almost similar results have been

Table 4: Effect of stratification period and temperature (PxT) on germination and seedling growth of *A. acuminatum*.

Treatment (PxT)	Germination (%)	Plant height (cm)	Collar diameter (mm)	Root length (cm)	Dry shoot weight (g)	Dry root weight (g)	Total dry weight (g)	Root: shoot ratio	Dickson quality index
P_1T_1	6.67 (14.83)	8.33	2.12	9.08	0.085	0.032	0.116	0.37	0.018
P_2T_1	12.50 (20.58)	9.40	2.21	10.12	0.114	0.045	0.159	0.39	0.023
P_2T_2	22.08 (27.90)	13.04	2.83	14.43	0.197	0.081	0.277	0.41	0.040
P_2T_3	13.75 (21.46)	12.53	2.36	11.99	0.144	0.059	0.203	0.41	0.026
P_3T_1	15.00 (22.73)	10.62	2.34	11.36	0.150	0.063	0.212	0.42	0.030
P_3T_2	33.33 (35.16)	16.37	3.34	15.78	0.239	0.103	0.342	0.43	0.048
P_3T_3	19.17 (25.79)	15.44	2.74	13.09	0.198	0.085	0.283	0.43	0.036
P_4T_1	30.42 (33.42)	12.18	2.65	13.03	0.202	0.096	0.297	0.48	0.045
P_4T_2	45.42 (42.35)	17.77	3.51	18.15	0.409	0.258	0.667	0.62	0.102
P_4T_3	38.33 (38.23)	15.21	3.22	16.26	0.280	0.147	0.426	0.53	0.066
P_5T_1	21.25 (27.40)	11.98	2.56	12.33	0.177	0.074	0.251	0.42	0.036
P_5T_2	38.32 (38.21)	15.43	3.33	16.47	0.297	0.147	0.445	0.50	0.067
P_5T_3	26.25 (30.73)	13.81	3.06	15.18	0.226	0.106	0.332	0.47	0.050
$CD_{0.05}$	**2.53**	**0.53**	**0.11**	**0.55**	**0.013**	**0.008**	**0.020**	**0.03**	**0.005**

Figures in parentheses are arc sine transformed values.

Table 5: Effect of stratification period and gibberellic acid (PxG) on germinability of *A. acuminatum* seeds.

Treatment (PxG)	Germination (%)	Germination capacity (%)	Germination value	Germination index
P_1G_1	5.83 (13.91)	51.39 (45.78)	0.21	0.30
P_1G_2	8.33 (16.73)	55.83 (48.35)	0.42	0.42
P_2G_1	19.44 (25.73)	64.44 (53.76)	1.15	1.05
P_2G_2	25.56 (30.26)	65.00 (53.93)	1.83	1.42
P_3G_1	28.61 (32.16)	80.28 (63.83)	2.07	1.52
P_3G_2	36.67 (37.13)	85.56 (67.74)	3.20	2.03
P_4G_1	39.44 (38.72)	83.33 (65.97)	3.31	2.20
P_4G_2	47.78 (43.69)	89.44 (71.16)	4.88	2.67
P_5G_1	32.78 (34.77)	78.33 (62.39)	2.71	1.85
P_5G_2	40.83 (39.63)	83.06 (65.75)	3.72	2.24
$CD_{0.05}$	**2.26**	**2.24**	**0.38**	**0.23**

Figures in parentheses are arc sine transformed values.

Table 6: Effect of stratification period and gibberellic acid (PxG) on germination and seedling growth of *A. acuminatum*.

Treatment (PxG)	Germination (%)	Plant height (cm)	Collar diameter (mm)	Root length (cm)	Dry shoot weight (g)	Dry root weight (g)	Total dry weight (g)	Root: shoot ratio	Dickson quality index
P_1G_1	5.83 (13.91)	8.05	1.90	8.00	0.077	0.028	0.104	0.35	0.015
P_1G_2	7.50 (15.74)	8.62	2.35	10.15	0.092	0.036	0.127	0.38	0.020
P_2G_1	13.89 (21.45)	11.04	2.29	11.58	0.144	0.057	0.201	0.40	0.027
P_2G_2	18.33 (25.17)	12.27	2.64	12.77	0.159	0.066	0.224	0.41	0.032
P_3G_1	19.17 (25.68)	13.47	2.59	12.79	0.183	0.077	0.260	0.42	0.034
P_3G_2	25.83 (30.11)	14.82	3.02	14.03	0.208	0.090	0.298	0.43	0.041
P_4G_1	35.83 (36.67)	14.30	3.03	14.99	0.261	0.143	0.404	0.53	0.062
P_4G_2	40.28 (39.33)	15.80	3.22	16.63	0.332	0.190	0.522	0.54	0.079
P_5G_1	25.83 (30.32)	13.19	2.90	14.09	0.219	0.102	0.321	0.46	0.048
P_5G_2	31.39 (33.91)	14.29	3.06	15.22	0.248	0.116	0.364	0.46	0.054
$CD_{0.05}$	**2.07**	**0.44**	**0.09**	**0.45**	**NS**	**0.007**	**NS**	**NS**	**NS**

Figures in parentheses are arc sine transformed values.

reported by Dogra (2003) in *Picea smithiana* and Sofi (2005) in *Cedrus deodara* seeds. As far as nursery performance of PxG interaction was concerned (Table 6), the significantly higher germination (40.28%), plant height (15.80 cm), collar diameter (3.22 mm) and root length (16.63 cm) was recorded when seedlings were raised from seed stratified for 60 days and treated with 200 ppm GA_3 (P_4G_2). These results also find support from the work of Sofi (2005) and Beyhan *et al*. (1999) for *Cedrus deodara* and *Corylus* seedlings, respectively.

Conclusion

The present study was an exploratory work to see the effect of different stratification temperatures, periods and (gibberellic acid) GA_3 on the germination and seedling growth of *Acer acuminatum*. The results indicated that the cold moist stratification for a period of 60 days at 3°C provided better germination and seedling growth of *A. acuminatum*. The application of GA_3 also helped to break down the dormancy and enhanced the germination of *A. acuminatum* seeds.

Acknowledgements

Authors thank to Dr. N.K. Gupta, Professor and Head, Department of Silviculture and Agroforestry, Dr. Y.S. Parmar UHF Nauni, Solan (HP) for providing critical comments and inputs on paper. Special thanks are to Dr. P.K. Mahajan for providing us important inputs during data analysis. First author is highly thankful to U.G.C. for providing financial assistance in the form of fellowship to carry out Ph.D. research work. Finally, we extend our special thanks to Rohru and Theog Forest Division for providing the valuable inputs and staff for seed collection.

उत्तर-पश्चिमी हिमालयों में *एसर एक्यूमिनेटम* के अंकुरण एवं पौध वृद्धि पर स्तरण एवं जिब्बरेलिक एसिड का प्रभाव

सुरेश कुमार, जी.एस. शमीत, दिनेश गुप्ता, नीना कुमारी एवं राजकुमार

सारांश

एसर एक्यूमिनेटम (वाल एक्स डी डान) बीजों में शारीरिकीय प्रसुप्ति की उपस्थिति इसके अंकुरण में चुनौती खड़ी करती है। वर्तमान अध्ययन में हमने दो सालों के लिए *ए. एक्यूमिनेटम* के बीजांकुरण एवं पौध वृद्धि पर स्तरण एवं जिब्बरेलिक एसिड (जी $ए_3$) के प्रभाव की जांच की। बीजों को बीस दिवसीय अन्तराल, यथा 20, 40, 60 और 80 दिन की पांच स्तरण अवधियों; तीन स्तरण तापमानों, यथा कक्ष तापमान, 3 ± 1 डि. से. और -3 ± 1 डि. से. में रखा गया और अन्त में बोने से पहले आसवित जल और 200 पी पी एम जी $ए_3$ के साथ उपचारित किया गया। आधे बीजों को प्रयोगशाला अवस्थाओं में बोया गया और आधे बीजों को शेड नेट हाउस में पॉली बैगों में बोया गया। परिणामों ने दर्शाया कि 60 दिनों के लिए स्तरण में रखे गये बीजों ने दोनों अवस्थाओं में महत्वपूर्ण रूप से अधिकतम अंकुरण (43.61% और 38.06%) उपलब्ध कराया। इसी तरह 3 ± 1 डि. से. स्तरण ने महत्वपूर्ण रूप से सर्वोत्तम अंकुरणशीलता और पौध वृद्धि उपलब्ध कराई। जी $ए_3$ (200 पी पी एम) अनुप्रयोग ने भी दोनों अवस्थाओं में बीजों के अंकुरण को बढ़ाया।

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