

Review Article

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## Seed Germination Enhancement through Breaking Seed Dormancy: A Review in Tropical and Temperate Tree Species

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### ABSTRACT

The most important factor for the success of plantation is readily availability of quality seeds. In favorable climatic conditions with optimum moisture and temperature most of the tree species germinate quickly and in unfavorable conditions some degree of seed dormancy occurs. There are various kinds of dormancy occur in trees species such as; exogenous dormancy due to hard seed coat, seed coat impermeable to water, presences of chemical; endogenous dormancy due to immaturity of embryo, oxidation and presence of inhibitors; and double dormancy which include both seed coat and embryo dormancy. In most of tropical tree seeds, physical dormancy occurred because of hard seed coat whereas temperate trees seeds mainly have physiological dormancy. There are various methods to deal with different kinds of dormancy i.e. through Scarification, Stratification, Chemical, Biological, and Irradiation methods.(I) Scarification which is done in hard seed coated species such as *Acacia spp.*, *Albizia lebbeck*, *Cassia spp.*, *Santalum album*, *Terminalia arjuna* etc. Mechanical scarification and concentrated H<sub>2</sub> SO<sub>4</sub> have been widely used to improve germination of several hard seed species, but the level of seed coat thickness among various species is the reason for different responses to all treatments. (II) Stratification is done to remove physiological and morphological dormancy. Most of the Conifers generally have physiological dormancy, stratification for 0-5°C for 60-70 days is better to overcome this kind of dormancy. (III) Chemical methods include use of growth regulator and chemicals for dormancy breaking. Growth hormones such as gibberellins, IAA and Kinetin etc. are mostly associated with seed germination and seedling physiology. Among all growth regulators GA<sub>3</sub> is most commonly used for germination enhancement @ 100-200 ppm for 1-2 h in most of the species. KNO<sub>3</sub> is important chemical for germination enhancement in many species such as *Acacia nilotica*, *Melia azedarach* etc. (IV) Biological method by passing through digestive tracts of animals like seeds of *Acacia senegal* and (V) Irradiation method irradiate seeds with like x-rays, light rays, gamma rays etc. So, breaking seed dormancy is necessary for seed germination enhancement in nursery.

### Keywords

Dormancy,  
Growth regulators,  
Scarification,  
Stratification,  
Irradiation, etc

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## Introduction

A Seed has been defined as a “mature ovule” or a reproductive unit formed from fertilized ovule consisting of an embryo, reserve food and a protective cover. Based on storage seeds are mainly two kinds viz. Orthodox seed having low moisture content (<5 to 10%) and successfully stored at low or sub-freezing temperature for long time e.g. *Acacia*, *Prosopsis*, *Pinus*, *Eucalyptus*, *Picea*, *Betula* etc. Recalcitrant seed having high moisture content (20-25%), which cannot be stored successfully for long period. Seed are sensitive to low temperature, chilling damage and death may occur if stored in low temperature e.g. *Azadirachta*, *Quercus*, *Willow*, and *Poplar* etc. Other than this there is also an intermediate seed which exhibit the drying tolerance characteristic of orthodox seed but are sensitive to low temperature storage like recalcitrant seed.

Dormancy is an inherent state of growth that is found among all life forms (Footitt *et al.*, 2011; Finch- Savage and Leubner-Metzger, 2006). However, dormancy of seeds in plants elucidated as inborn limitation on germination under circumstances that would otherwise encourage germination in non-dormant seeds. In favorable conditions, dormancy is state which enable seeds to avoid germination. Plants also disperse their progeny across time to avoid risk against uncertain environmental conditions by producing seeds with varying levels of dormancy (Venable, 2007; Poisot *et al.*, 2011). Rather, non-dormant seed germinate as quickly as commendatory conditions occurs and thereby reduce the risk of seed predation and escalate the growing season. Factor affecting seed dormancy are internal chemical inhibitors which may need to degrade or need to leach out, immature embryo needing an after-ripening period for internal chemical changes, oxygen content reaching the embryo, hard impermeable seed

coats disallowing water imbibition, light and darkness requirement involving light intensity level or wavelength. Fundamentally, dormant or non-dormant seeds govern the habitat experiences by plants and it is key constituents of plant life histories (Donohue *et al.*, 2005).

Dormancy includes; Exogenous dormancy (Seed coat D) which is further divided into physical, chemical and mechanical dormancy a) Physical dormancy occurs where seed coat is impermeable to water and gaseous due to hardened endocarp. Such seed coats develop during the last stage of seed development e.g. *Acacia*, *Cassia*, *Prosopsis*, *Albizia* etc., b) Chemical dormancy occurs due to presence of chemical inhibitor in outer covering of many seeds. It may be present in other tissue surrounding the embryo like endosperm e.g. *Betula* etc. c) Mechanical dormancy due to hard seed coat does not allow embryo to expand during germination e.g. *Terminalia* spp., *Melia* spp. etc. Another is Endogenous dormancy which includes physiological and morphological dormancy a) Physiological dormancy in which seed coat resist transfer of oxygen to embryo and presence of inhibitors like phenols which fix oxygen by oxidation and limit the availability of oxygen to embryo and leads to problem in germination which is further of three kinds; shallow, deep and intermediate dormancy. Shallow dormancy is for short term and lasts for 1-6 months. It may be due to inhibitors and control of gas exchange (oxygen). It occurs due to high temperature *i.e.* Thermo dormancy (*Quercus*, *Pinus*, *Eucalyptus*, *Fagus* etc.) and due to light *i.e.* Photo dormancy (*Betula*, *Eucalyptus*, *Ficus*, etc.). Deep dormancy controls by the factors within the embryo itself. It occurs in most temperate trees. It can be controlled by natural stratification or cold Stratification or moist chilling e.g. *Apple*, *Juniper*, and *Taxus* etc. Intermediate dormancy the control mechanism thought to be in seed coats and in

tissues surrounding embryo. It can be broken down by chilling, stratification and Gibberlllic acid. (E.g. Conifers). b) Morphological dormancy in which embryo is not fully developed at the time of ripening. It needs additional period for embryo growth after the seed is separated from the plant. This dormancy is broken by alternate warm and cold temperature, gibberlllic acid treatment. E.g. *Fraxinus*, *Ginkgo* etc. Ex-Endo/combined dormancy is caused by factor related to seed coat as well as embryo. It may be caused either by physiological deficiency in the embryonic axis or due to presence of metabolic blocks within the cotyledons. E.g. *Emblica officinalis*, *Gmelina Arborea* etc.

Seed germination enhancement is the application of physical, physiological, biological and chemical agents to the seed in order to enhance the physical, physiological, genetical, biochemical and health qualities of seed. It can be achieved through Physical methods/Scarification, Physiological method/Stratification, Chemical method/ soaking in growth regulator or chemicals, biological method and irradiation method. Moreover, there is also germination augmenting treatments like Seed fortification, Seed Priming Hardening, Bio fertilizer treatment etc.,

Seed coating treatments like Seed pelleting, Coloring etc., Protective seed treatments like Insecticide, Fungicide etc., but here we focused on dormancy part only. Dormancy breaking treatments, literature findings, and discussion are described in this review paper as in following sub-headings:

Scarification/ Physical Method

Stratification / Physiological Method

Chemical Method

Biological method

Irradiation method

### Scarification/ physical method

It is a method of disrupting an impermeable seed coat of seeds. This method includes followings types mention in Table 1.

Kullar (1991) reported that seeds of many leguminous families having impermeable seed coat such as species of *Cassia*, *Acacia*, and *Albizia* etc. In majority of the forestry species seed coat dormancy occurs which is due to structure surrounding the embryo. Gehlot and Sen (1998) studied that embryo endure dormant stage while covered in the seed coat but germinate if seed coat is punctured or isolated and cultured on the appropriate medium. Study was conducted on *Acacia mangnium* due to rapid growth and liberality to very acid and infertile soil and is most suitable species for commercialization, reforestation and environment protection. Experiment was performed with different scarification treatment of conc.  $H_2SO_4$  for (0.5, 1, 2, 3, 5, 10, 15 minutes) and control. Results were observed after 10 days of treatment. They found that maximum germination percent 90.67 % was obtained in 10 minutes scarification followed by 5 minutes (89.33%) and 3 minutes (82.67%). But in control (without scarified seed) minimum germination percent (6.67%) was obtained.

Azad et al (2010) conducted study in five different treatments i.e. control, cold water (4°C) immersion in for 12 h, hot water (80°C) immersion in for 10 min, scarification of seeds with sand paper, concentrated  $H_2SO_4$  (80%) immersion in for 20 min, to determine methods that maximized germination percentages. Poly-bags media taken was in the ratio of 3:4:1:1 (contains topsoil, coconut husk compost, coarse sand and fine sand). Germination percentage observed in scarification (80%), concentrated  $H_2SO_4$  80 % immersion for 20 min (74%), hot water

treatment (69%), it shows no significant difference with each other rather all the treatments differed significantly with control i.e. 48%. In  $H_2SO_4$  treatment, germination started first (on 8<sup>th</sup> day), in hot water (on 9<sup>th</sup> day), in control and sand paper scarification (on 10<sup>th</sup> day) and, in cold water treatment (11<sup>th</sup> day). Similar germination pattern is due to thinned outer coat of the seeds.

Best pre sowing seed treatment for *Acacia catechu* seeds and *Elaeocarpus floribundus* seeds studied by Das N (2014). It is based on six pre sowing treatments at the nursery stage that includes: (i) control, (ii) immersion of seeds in hot water for 10 min in 80°C, after that dipping in cold water 24 hrs. (iii) Immersion of seeds in cold water 4°C for 24 hrs. (iv) Immersion of in hot water 100°C for 12min after that dipping in cold water for 24 hrs. (v) Immersion of seeds for 20min in concentrated  $H_2SO_4$  (80%) (vi) Scarification of seeds with sand paper. Highest germination percentage (91.26%) in *Acacia catechu* was observed in hot water immersion (80°C) 10 min after that dipping in cold water for 24 hrs. which was only (69% in control).

It might be due to it require hot water firstly to break dormancy and after that dipping in cold water for 24 hrs. after hot water treatment makes softer seed coat. Whereas, in case of *Elaeocarpus floribundus* highest germination 89.8 % with  $H_2SO_4$  (80%) 20 min and 86.35% with immersion of seeds in hot water 100°C for 12 min, after that dipping in cold water for 24 hrs.

Effect of pre-sowing treatment varies from species to species. Levels of thickness in seed coat among various species are the reason of different degree of response to different pre sowing treatment.

Lots of work has been done by many different scientist for breaking the hard seed coat of

different forest species and they found that in *Albizia labbeck*, *Cassia fistula*, and *Pinus roxburghii* etc. the main reason for the increase in imbibition and germination is seed coat and microphyler plug disintegration. In leguminous tree species, acid treatment shows maximum germination percentage, quick and early emergence of seeds which results from the seed coat softening that allows movement water into the seed and enlargement of the embryo that is induced water absorption and gaseous exchange.

### Stratification/physiological method

Physiological dormancy occurs mostly in conifer seed. So, in coniferous species stratification/physiological method is one of the widely used methods to enhance germination percentage by breaking seed dormancy. It can be done with various temperatures and in humid and dry conditions that last for few weeks and months. It is done by two methods i.e. cold stratification and warm stratification.

### Cold stratification

In this method, Seeds are imbibed at low temp (3-5°C) for a period of time (48 h) and then draining of water and seeds are mixed with moist (sand, sand peat mixture etc.) 2-4 times of their volume and stored at cold temperature 3-5°C.

Biochemical activity due to high moisture and low temp, transforms complicated food material to simple forms that is taken by embryo, and micro-organisms activity is also reduced. It is done to break physiological and morphological Dormancy. Study shows that cold ST induces a response to gibberellic acid and initiated cell division resulting in release of physiological dormancy and subsequent germination in *Taxus chinensis* seeds Zhang et al (2013).

### Warm stratification

In this method, Seeds are imbibed in cold water at 3-5°C for a period of time (48 h) and then draining of water and seeds are mixed with moist or water retained medium (sand, sand peat mixture etc.) 2-4 times of their volume and stored at warm temperature 20-25°C. Partial digestion of the outer layer and final weakening of inner layer results in increased oxygen uptake and energy to embryonic growth, cell expansion, increase GA<sub>3</sub> and decreased ABA level. It is done to break physical, mechanical and morphological Dormancy. Studies shows that warm ST increased the growth of embryo by cell expansion and enlargement in *Taxus chinensis* seeds (Zhang *et al.*, 2013).

Conifers seed in generally has extreme dormancy even in the favorable environment conditions for its germination as studied by Jull and Blaich (2000). Dormancy in seeds is due to internal and external factors. For seed germination improvement cold stratification before sowing has been widely used for disrupting dormancy (Baskin and Baskin, 2004). Muglo (2013) conducted study with different stratification temperature and period to observe germination percentage. Different treatments take were Stratification period *i.e.* 15, 30, 45, 60, 75, 90, 105, 120 days and Stratification temperature 0±1°C, 4±1°C, control. Highest germination percentage (62%) was observed in 0±1°C temperature between 60 to 90 days, thereafter germination period decrease with increase in stratification period.

Gosling (2015) conducted study on *Betula utilis* (Himalayan birch) seeds having intermediate dormancy. Mir *et al* (2018) studies on germination ability and performance of *Betula utilis* under field. Stratification and GA<sub>3</sub> treatment is method to overcome embryo dormancy. Stratification of

seeds at 4 ± 1°C for different time period *i.e.* 0, 30, 60, 90 days and then these seeds were treated with GA<sub>3</sub> *i.e.* 0ppm, 50ppm and 100 ppm for 2 h. Results show that with increasing stratification duration and GA<sub>3</sub> concentration, there is significant increase in germination percentage and energy. The maximum and minimum germination (89.11%, 53.33%) and germination energy (38.22, 28.22) were recorded in stratified seeds for (90 days and with 100 ppm GA<sub>3</sub>) and control.

Gosling, 2015 suggests different species requires different stratification period as shown in figure 1: *Abies alba*, *A. balsamea*, *A. concolor*, *A. fraseri*, *A. grandis*, *A. procera* require 8 weeks of stratification. *Cedrus atlantica*, *Cedrus deodara* also requires 8 weeks stratification. *Juniperus communis* needs 24 weeks of stratification. *Picea abies*, *P. omorika*, *P. pungens*, *P. sitchensis* requires 6 weeks of stratification, *Pinus contorta*, *Pinus muricata*, *Pinus nigra*, *Pinus pinaster*, *Pinus radiata*, *Pinus sylvestris* also requires 6 weeks of stratification whereas, *Taxus baccata* needs 40 weeks of stratification.

Various physiological changes total sugars and starch content occur in seeds during stratification method. Stratification activating enzymes such as amylase, protease and lipase which is liable for solubilizing spare food material in the form of starch, protein in seed and deliver energy and other food material to germinating embryo thus allowing dormancy release. Higher germination as compared to control *i.e.* without stratification might be due to conversion of starch (used for embryo growth and development). Growth promoting substances such as increase in GA or cytokines level inhibit the effect of retardants like ABA (Paul *et al.*, 1973). Low temperature and stratification period results in biochemical changes in seeds that is useful during germination by transforming

complicate food material into simpler forms (Malik *et al.*, 2008). Tanaka *et al* (1991) also reported that during stratification lower temperature enhance germination ability in conifers. Physiological method reduces the rate of enzymatic reactions in the seed and cause distinctive changes i.e. in enzyme concentrations or production and phase changes i.e. in membrane fluidity. It also triggers enzymes and hormones production thereby release dormancy reported by Bewley and Black (1994). Increase in seed germination may be due to increase in soluble protein and sugar with increase in stratification duration. This biochemical changes may be responsible for high germination parameter of seed or it is due to increase in GA level in the seed which result in hydrolase synthesis in the seed which causes breakdown of starch into sugar. Allen (1962) reported that in coniferous seed longer ST period is better.

### Chemical Method

This method is also widely used in different species it consists of soaking of seeds in growth regulator, chemicals, new generation plant growth regulators which are represented in table 3, 4, and 5.

Azad *et al.*, (2010) conducted study on *Melia azedarach* a highly valuable multipurpose tree species, so for quality planting stock their vigor's seed with uniform germination is necessary but, main difficulty in this species is its poor seed germination. So, this study was conducted to obtain highest germination percentage with different treatments Viz., Cold water (24hrs), Hot water (10 min), Concentrated  $H_2SO_4$  (10 min dipping),  $KNO_3$  (200 M for 24 h), Gibberllic acid (100 ppm for 8 h), Cow dung slurry (1:4 for 6 days), Biogas slurry (6 days), Mini-sachet method (5 days) and Control. After 60 day of sowing, among the all treatments, highest germination

percentage was observed from 84% in  $KNO_3$  followed by 70.7% in biogas slurry and 70% in cow dung slurry these were significantly better than other treatments and at par with each other. Ali *et al* (2010) reported that increased germination with  $KNO_3$  solution may be due to; reducing the growth retardant (ABA) and enhanced imbibition of water into cotyledons that create hormonal balance in the seed. Whereas, in case of biogas slurry and cow dung slurry, germination percentage increase is due to increase in microbial population, due to anaerobic condition and moderate temperature seed germination is triggered (Sujatha *et al.*, 2007).

Mugloo J A (2013) reported the effects of various treatment on germination of spruce using different treatments viz., control, cold water for 24 and 48 hr,  $GA_3$  (50ppm, 100ppm, 150ppm, 200ppm, 250 ppm) for 24 and 48 hour. Significantly highest germination percent was obtained from  $GA_3$  200 ppm for 48 h treatment which was 30% in case of control and 35 % in cold water treatment, these both shows lesser percentage compared to other treatments. Increase in germination percentage in  $GA_3$  is might be due to increase the growth potential of embryo and by weakening of layers surrounding the seed that weaken the tissue around the radicle which results in improved growth.

Study was conducted by Rawat *et al* (2006) at three districts viz., Pauri, Tehri and Chamoli of Garhwal Himalaya. Aim is to understand the role of  $GA_3$  treatment for seed germination in different provenances of *Abies pindrow*, *Cupressus torulosa* and *Picea smithiana*. Five provenances each of three important coniferous species (15 provenances) were taken. They were subjected to different treatments *i.e.* soaking of seeds in distilled water at 25 °C for 24 hr, soaking of seed in  $GA_3$  100 ppm for 24 hr. In *Abies pindrow* 45 % germination percentage

was achieved with GA<sub>3</sub> which was only 26% in control. In *Cupressus torulosa* 57% germination percentage was gained after treatment with GA<sub>3</sub> which was 25% in control. Whereas, in case of *Picea smithiana* 56% germination was obtained with GA<sub>3</sub> and 27% in control. Seed germination improvement with gibberllic acid might be due to hydrolase synthesis (especially amylase synthesis) which solubilize spare food material in seed in the form of starch or protein and deliver energy and food material to germinating embryo. Growth of embryo further results in more gibberllic acid synthesis, which induced hydrolase synthesis.

Many plant regulators are linked with seed germination enhancement and seedling growth in favorable conditions, but the key hormones are gibberellic acid, Indole-3-Acetic-Acid and Kinetin etc. (Faridi *et al.*, 2000). Growth hormones increase in growth potential of embryo, weaken the structure covering the embryo through cell wall hydrolases, counteracts the effect of ABA, reduces photo-D, promotes germination in thermo-D through ABA metabolism, initiation of enzymes, mobilize food substances which leads to cell division, promotes germination in viable seeds through cell division, cell elongation and embryo growth (Pandiya, 1989).

In the course of germination and growth, the total carbohydrates are employed to encounter the pre-condition of respiration, protoplasm material and synthesis of cell wall for dividing cells (Black, 1992). Cytokinins promote germination through ethylene biosynthesis (Matilla, 2000). Ethephon (ethylene releasing compound) take part in radicle cell expansion, Increase seed respiration and water potential, decreasing the sensitivity of ABA etc.

### Biological methods

It involves animals, insects, and microorganism. Pods are fed to sheep or goats. Hard seed coat is softening by the combination of moisture, warmth and chemical action of the digestive juices. It is hard to utilize these agents as a controlled pretreatment of seed, but in some cases positive results were also reported.

Goor and Barney (1976) found that pretreatment of seeds of *Acacia Senegal* and *Ceratonia siliqua* collected from droppings of penned goat, overcame seed dormancy and seed germinate quickly when placed in favorable environment condition, which is due to the action of the strong digestive chemicals in digestive tracts of goats. Troup (1921) reported improved germination due to digestive actions of sheep and goat in *Acacia nilotica* seeds that were discharged after rumination.

In many part of the tropics termites are also a key component for cracking the seed dormancy. In Thailand, after teak fruits collection it is spread in ground in a 5 cm thick layer and then covered it with cardboard and after about 5 weeks the termites had removed the exocarps. Germination of removed exocarp seed is significantly improved as compared to with intact exocarps [Bryndum (1966), Sompherm (1975)]. Groome *et al.*, (1957) reported that termites also used the same way to shatter sturdy winged and stubble fruits of *Pterocarpus angolensis*.

Partial fermentation is also beneficial in some species by overcoming seed coat dormancy. Wunder (1966), studied on *Tectona grandis* fruits that improved germination is obtained through laying the fruits of *Tectona* on the floor throughout the monsoon season which results in mild fermentation. Then gathered these seeds and stratified in ditch with

different layers of seed, organic matter and soil and waters it for 10 days. Ahmed, 1986 also found that seeds of *Acacia* species obtained from goat dropping are non-

dormant than seeds which were not feed to goat. It is useful in *Acacia senegal*, *Acacia nilotica*, *Gmelina arborea*, *Tectona grandis* etc.

**Table.1** Different kinds of scarification and research findings

Scarification	Description	Research findings
Physical/ Manual	Clipping with knife, piercing with needle, rubbing against rough surface, cracking with hammer etc.	Nicking using secateurs gave maximum germination in <i>Acacia polyacantha</i> reported by Edward M (2014). In India this method is used to overcome seed coat dormancy in <i>Albizzia falcataria</i> , <i>A. lebbek</i> , <i>Acacia catechu</i> , <i>A. nilotica</i> subsp. <i>indica</i> , <i>Cassia fistula</i> , <i>C. javanica</i> , <i>C. nodosa</i> , <i>Delonix regia</i> , <i>Santalum album</i> , <i>Terminalia arjuna</i> , <i>T. tomentosa</i> etc.
Water soaking	Soaking of seed in cold and hot water for few hours to several days	Schmidt L (2000) reported that 82-89% germination percentage in <i>Cassia siamea</i> is obtained by soaking seeds for 1-2 min in warm water (85°C) with subsequent cooling in the water for 12-36 hrs. Soaking of seeds in water for 2-48 hrs based on species accelerates germination in many species such as <i>A. amara</i> , <i>A. procera</i> , <i>G. robusta</i> , <i>T. nudiflora</i> , <i>Acacia mearnsii</i> , <i>A. melanoxylon</i> , <i>A. nilotica</i> subsp. <i>kraussiana</i> etc.
Fire	Light inflammable litter are spread over the fruit and then ignite, done to partially damage seed coat	Dry heat ineffective in increasing germination, whereas moist heat greatly increased both germination rate and total germination of <i>Lespedeza</i> species of seed found by Charles et al (1967).
Acid	Pouring undiluted acid until all seeds are wetted, stirring, immediately washing then cooling	Duguma et al (1988) reported 30 min treatment with $H_2SO_4$ in <i>Leucaena leucocephala</i> gives 95% germination. Some examples like soaking of <i>Intsia palembanica</i> for 60 min, <i>Parkia javanica</i> for 15 min, <i>Dialium maingayi</i> for 5 min, <i>Acacia albida</i> for 20 min, <i>Acacia nilotica</i> for 60–80 min, <i>Acacia senegal</i> for 40 min, <i>Acacia planifrons</i> for 2 hours and <i>Prosopis tamarugo</i> for 7 min of tropical species which respond well to Sulfuric acid treatment. <i>Pterocarpus angolensis</i> hulled seeds when treated with acid it gave 60 % germination between 4 -19 days, in comparison to control it was 16 % between 11-37 days.

**Table.2** Stratification effect on some important conifers

Species	Temperature °C	PT days	Germination %	Reporting
<i>Cedurs deodara</i>	5	75	82.10	Sofi and Bhardwaj (2008)
<i>Abies spectabilis</i>	1-5	30-120	70	SFRI information bulletin (1999)
<i>Juniperous recurva</i>	1-5	30-120	50	Mir et al (2014)
<i>Pinus geradiana</i>	4±1	60	83	Malik and Shamet (2009)
<i>Pinus wallichiana</i>	1 - 5	30 -120	82	Mir et al (2014)

**Table.3** Different kind of growth regulators taking part in dormancy breaking, their functions and research findings

Growth regulator	Functions	Research findings
Gibberellic acid (found in mersitem of apical bud and root, embryo)	Promote seed and bud germination, take part in Stem elongation and leaf growth, Stimulate flowering an development of fruits, Affect root growth and differentiation etc.	Mirzavandiani (2016) reported 92% germination in <i>Pinus radiata</i> with GA <sub>3</sub> 300ppm+ KNO <sub>3</sub> (.3) for 48 h whereas Rawat (2006) studied 57% germination in <i>Cupressus torulosa</i> with GA <sub>3</sub> 100ppm@ for 24hrs.
Kinetin (it is present in developing seeds)	Take part in Promotion of cell division in embryo, Release dormancy, Effect root growth and differentiation	In <i>Ziziphus mauritiana</i> BA (100ppm) stimulated both total germination and vigor of seeds reported by Murthy and Reddy (1989).
Auxins (found in embryo of seed, meristem of apical buds)	Stimulate stem elongation, take part in Root growth, Enhance apical dominance etc.	Highest seed germination (91%) observed in <i>Pinus massoniana</i> with the application of IAA 100ppm and GA3 50ppm reported by Zhao Guangwu, (2014).
Ethylene (found in ripening fruit, nodes of stem)	Stimulate germination, Alleviates primary and light induced dormancy etc.	Ethylene plays a key role in dormancy release in numerous species, the effective concentrations allowing the germination of dormant seeds ranging between 0.1 to 200 µL L(-1) reported by Francoise C (2014).
Fusicoccin( <i>Fusicoccin amygdali</i> ) it is a toxin produced by fungus	Breaking seed dormancy, Stimulate germination, Eliminate primary Dormancy	The application of germination regulating chemicals like fusicoccin, proline and nitrate almost completely alleviated the salinity effect on seed germination studied by Ajmal K (2009)

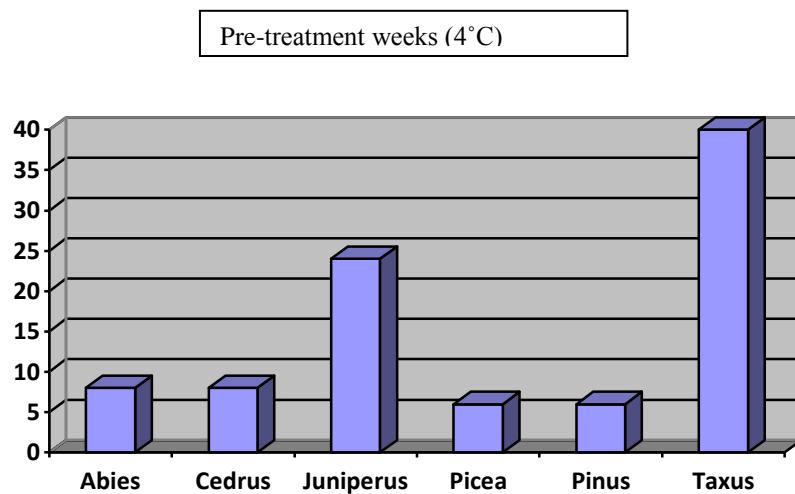
**Table.4** Different chemical used in dormancy breaking and research findings

Chemicals	Scientific findings
Potassium nitrate (It break dormancy, faster seed germination and vigour)	$\text{KNO}_3$ had a strong effect on germination percentage and vigour of <i>Acacia nilotica</i> seeds. At 1% concentration, germination increased 79%, and at 2% conc. It increased to 85% and in control it was 37 % reported by Palani et al (1995).
Thiourea (it also take part in Breaking dormancy, regulator of seed germination in soil)	In <i>Ziziphus mauritiana</i> , thiourea is proved the most effective germination stimulant, soaking of seed in 1% solution for 1day increase total germination percentage 78% at 30°C, 41% in control was found by Lars Schmidt (2000).
Hydrogen peroxide it is an active oxygen source, oxygen can accelerate ABA degradation, enhancing oxygen production thereby promote germination by breaking Dormancy)	Chien and Tsan (1994) found that in <i>Cinnamomum camphorum</i> germination is improved with 15% $\text{H}_2\text{O}_2$ treatment from 0-11% to 51-58%.
Alcohol It also Dormancy breaking, like in the form of ethyl alcohol and ether	Ethanol treatments (3%) enhanced the germination of rice to 100%, which otherwise germinated poorly (30%) after dehusking studied by Miyoshi and Sato (1997).

**Table.5** Different kinds of new generation plant growth regulators used in dormancy breaking and research findings

PGR	Role	Research findings
Brassinosteroids (occurs in every part of plant)	Take part in breaking Seed dormancy, Cell elongation and division, Root and shoot development, Response to stress etc.	BR increase the ability of embryo to grow out of the seed by enhanced rupturing of endosperm reported by Leubner-Metzger (2001). Highest 49.6% germination by BR observed in <i>Fraxinus</i> as compared to control 20% found by Prochazka (2015).
Jasmonic acid	Regulate Photosynthesis, take part in root and shoot growth, Seed germination, Flowering etc.	Jasmonic acid increased the germination and rate of germination by 5-10 days in apple seed as compared to control studied by Yildiz et al., (2007).
Salicylic acid	Promote germination under high salinity, Influences on photosynthesis, enzyme activities, flowering etc.	SA increased germination of wheat by 67% under salt stress as compared to control reported by Bahrani and Pourreza (2012).
Nitric oxide	Induces seed germination in the absence of red light, breaks seed dormancy, and affects growth etc.	82% germination in Empress Trees with $\text{NO}_4^{-}$ 10 M and cold stratification reported by Jia Liu et al (2019).

**Fig.1** Pre-treatment week's requirements for different conifers



### Irradiation method

Seed is packed with the material and energy necessary for inceptive growth, but some external factors are also required to activate that material that is already present in cotyledons. The red region of the spectrum X-rays, gamma rays, light rays and sound waves have been experimented to stimulate germination and to break dormancy.

There have been reports of improvement in some species including *Tectona* Bhumibhamon (1973), but most of the studies showed hard to obtain reliable outcome and chromosome damage and other malformations may also cause by the treatments (Kemp, 1975). There were reports of many adverse effects from irradiation than favorable effects. None of this procedure is acceptable for actual application at the present time (Lynn, 1967).

Confiding on the levels of irradiation  $\gamma$ -irradiation has intense impact on growth by prompting genetically, biochemical, morphogenetic, physiological and cytological changes in tissues and cells. Small doses of irradiation may elevate the enzymatic

activation and invigorating of the young embryo, by elevating cell division rate and also influences the vegetative growth but it is also a key for the activation of various biochemical, physiological activities.. It can be useful for altering the physiological characters and improving germination and seedling growth. Cobalt-60 was used to irradiate the seeds of *Magnolia champaca* with (0, 5, 10, 15, 20, 40, 60, 80, 100 Gy) does. Three lots, namely germination test, storage test and seedling growth characteristics, were made from treated seeds. LD50 for irradiation was near 30 Gy does and at 80 Gy and higher doses irradiation caused lethal effect to the treatments. At 10 Gy enhancement in all germination parameters were recorded in irradiated seed, and then it was reduced. Up to 80 Gy irradiation dose there is increased in all seedling parameters such as collar diameter, number of leaves, seedling height and dry weight, but the 10 Gy results in more significant and achieve maximum value for all growth parameters studied by (Chandrasekhar *et al.*, (2013).

Akshatha *et al.*, (2013) studied the impact of gamma irradiation on *Terminalia arjuna*. Different doses ranging from 0 to 200 Gy

using the  $^{60}\text{Co}$  source were used and the results were compared with the control plants. In comparison to un-irradiated seeds (control) germination speed at 25 Gy was (0.65) which was double. All the parameters like germination percentage, vigor index etc. was found to be higher at lower dose of radiation treatment as with increase in dose proline content also increased. The chlorophyll content in control was 8.44 mg/g FW whereas, at 100 Gy it was increased to 12.2 mg/g. Bhumibhamon (1973), reported that with use of X-rays, gamma rays, light rays, high frequency sound waves etc. Improvement has been reported in *Tectona* to overcome dormancy and stimulate germination, but it also results in damage of chromosomes and other malformations.

In conclusion the mechanical scarification and conc.  $\text{H}_2\text{SO}_4$  have been widely used to improve germination of several hard seed coat species. It disrupts the barrier for the uptake of water by the seed and permits radicle emergence by weakening coat structure. Acid treatment shows best results in leguminous tree species by breaking hard seed coat that permit unrestricted inflow of water in the seed and expands embryonic parts. Quick and early emergence of seeds by acid treatment is due to softening of the seed coat that induces absorption of water and gaseous exchange. Levels of thickness in seed coat among various species are the reason of different degree of response to different pre sowing treatment. Most of the Conifers generally have physiological dormancy, so stratification for 0-5 °C for 60-70 days is better to overcome morphological and physiological dormancy. Low temperature and stratification period results in biochemical changes in seeds by transforming complicate food substances into simpler forms that are useful at the time of germination. Several growth regulators and chemicals are linked with seed germination and seedling growth, but the most important

being gibberellic acid, Indole-3-Acetic-Acid, Kinetin, Potassium nitrate, Thiourea etc.  $\text{GA}_3$  100-200 ppm for 1-2 days is beneficial for most of the species studied and it overcome embryo dormancy.  $\text{KNO}_3$  is also important chemical for germination enhancement in many species by reducing the effect of growth retardant (ABA) and enhanced imbibition of water into cotyledons that create hormonal balance in the seed. Several other chemicals like thiourea, hydrogen peroxide etc. and new generation plant hormones like jasmonic acid, barssiosteriod etc. are also found to be beneficial in some species. Biological method by feeding pods to animals/microorganisms also shows maximum germination in species like acacia, *Gmelina arborea* etc. Irradiation treatment is used in some species for the activation of biochemical and physiological processes but their negative effects were also reported in studies. Their lower dose up to 10 Gy can be useful to improve seed germination growth of seedling. Therefore, seed dormancy is an important factor while doing any research trail or for large scale production of seedling in nursery. Seed dormancy varies with species, while going for sowing it is must to consider dormancy type and appropriate treatment to obtain good results.

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