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Exploring Seed Dormancy Focussing on Medicinal Plants: A Comprehensive Review

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Authors' contributions

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

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Review Article

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ABSTRACT

This review explores the complex field of studies on medicinal plants during dormancy, aiming to provide a comprehensive overview of research findings and methodologies. Dormancy, a crucial physiological phase in the growth stage of plants, plays a master role in regulating growth, development and survival strategies. In medicinal plants, understanding dormancy is of paramount

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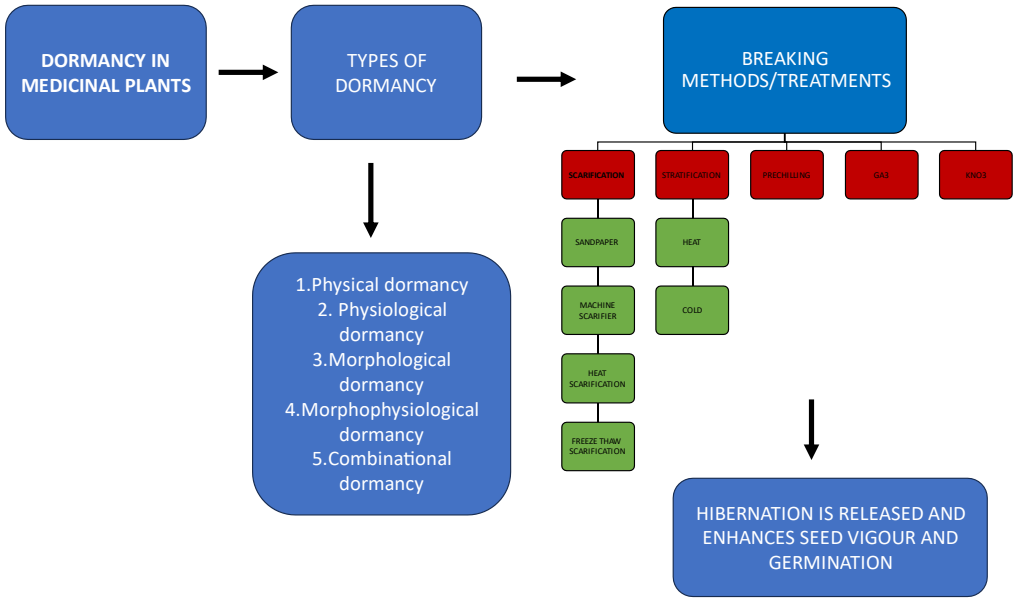
importance due to its implications for cultivation, propagation and the production of some compounds that are in high demand for pharmaceutical industry. This review focuses on methodologies in dormancy research, elucidating the underlying mechanisms and exploring strategies to manipulate dormancy for enhanced medicinal plant cultivation and utilization. Furthermore, it discusses the challenges in dormancy studies, highlighting the potential for novel approaches and interdisciplinary collaborations to unravel the complexities of dormancy and harness its benefits in medicinal plant research and industry.

Graphical Abstract



Keywords: Hormones; physiology; regulation; scarification; secondary metabolites; stratification.

1. INTRODUCTION

Therapeutic plants are essential to both traditional and contemporary medicine because they are rich in bio active substances. Further to being used for medical intensions, medicinal plants can supply pharmaceutical businesses with raw ingredients [1]. Nevertheless, issues with seed quality, germination rates and genetic stability arise in the long-term production of therapeutic plants. To tackle these issues and promote the study culture, this analysis seeks to showcase the latest developments in seed technology. Medicinal plants constituting 4.4% of the total worldwide seed market, in which the United States makes up 27%, China 20%, France 8% and Brazil 6%. Medicinal plants are those that are often used to cure and prevent specific diseases and disorders [2]. Plants are the green factories of medication and a vital part of global health. Indigeneous health care systems are still commonly used in many contexts. The attention on utilizing plant materials as a source of pharmaceuticals for a diverse applications of human needs has increased as a result of infectious diseases. These include an insufficient supply of medicine due to population growth, excessive therapy costs, unfavorable effects of different synthetic drugs, and the establishment of drug resistance to frequently prescribed drugs [3].

These plants are used as a raw material for the pharmaceutical sector, which has led to their recent major relevance in agriculture, medicine and exports. The majority of medicinal plants in the field have inconsistent seed quality, leading to problems with germination and stand establishment. Techniques for improving seed germination and seedling establishment will improve the performance of medicinal plant production because of their significance in the plant life cycle. Recently, the value of several plants for agriculture and medicine has been

retrieved in an attempt to save people's livelihoods worldwide [4].

Offering solutions to enhance seed germination and seedling establishment will contribute to improved concert in the cultivation of medicinal plants, since the life cycle of plant depends on these mechanisms. Globally, curative plants are a tremendous source of novel pharmaceuticals [5]. In the United States, over 118 of the top 150 prescription medications are extracted from natural sources, while over 1300 medicinal plants are explored in Europe, with 90% of them being gathered from wild resources[6]. Moreover, approximately 25% of recommended medications in wealthy nations come from wild plant species, while up to 80% of individuals in underdeveloped nations completely rely on herbal remedies for their primary health needs [7,8]. A conservative estimate suggests that the current rate of plant species loss is between 100 and 1000 times higher than the natural extinction rate and approximately one potential significant drug is lost every two years [9]. The International Union for Conservation of Nature and the World Wildlife Fund report that globally, there are between 50,000 and 80,000 flowering plant species used for medicinal purposes, with about 15,000 species endangered by expansion due to over harvesting and habitat destruction [10]. Despite awareness of this threat for decades, the accelerated loss of species and habitats globally poses an increased risk for the extinction of medicinal plants, especially in countries like Kenya [11], Nepal [12] India [13], Uganda [14], Tanzania [15] and China [5].Over 50,000 plant species more than one-tenth of all plant species are utilized in medications and healthcare items [16]. However there are geographical differences in the global distribution of therapeutic plants [17]. With 11,146 and 7,500 species, respectively, China and India have the highest numbers of medicinal plants used. In plant seeds, dormancy signifies a temporary cessation of visible growth, serving as an adaptive mechanism to endure adverse environmental conditions. This dormancy phase notably impacts the production of secondary metabolites, often reducing their synthesis. To improve germination in medicinal plants, knowledge on dormancy, types of dormancy and dormancy breaking treatments are essential. In this context, the information on above points are reviewed hereunder for the benefits of farmers, stakeholders and interested parties. Understanding dormancy patterns in medicinal plant seeds is essential for cultivators seeking to optimize germination success and crop yield. Tailoring germination protocols to address specific dormancy types can significantly enhance cultivation outcomes. Additionally, knowledge of dormancy can inform seed storage and handling practices to maintain seed viability over time. Understanding the germination behavior of medicinal plant seeds is fundamental for successful cultivation. One significant aspect to consider is seed dormancy, which can greatly influence germination rates and plant establishment.

2. DORMANCY – THE HIDDEN PULSE OF BOTANICAL VITALITY

Seed dormancy, characterized as the failure of a viable seed to complete germination under right circumstances, is influenced by environmental factors such as light, temperature and the duration of seed storage [18]. Seed dormancy is commonly understood as a hindrance to the complete germination of a viable seed even under favorable conditions. However, some literature has reported potential sources of confusion. Ecological studies, for instance, have encountered confusion between seed dormancy and soil persistence [19, 20] stemming from differing views on dormancy, particularly regarding the role of light in either terminating dormancy or inducing germination.

A formulated dormancy classification system acknowledging the interplay of both morphological and physiological features in seed dormancy was given by [21]. Expanding upon this framework, they introduced a comprehensive classification system comprising five main classes of seed dormancy: Physical, Physiological, Morphological and Combinational dormancy. This hierarchical system, outlined below, further categorizes these classes into various levels and types. Dormancy in seeds is not just escaping the germination; rather, it refers to the seed's characteristic that determines the potential required for germination [20, 22]. Any environmental issue that alters the conditions which is necessary for germination is considered to be altering dormancy. Moreover, when the seed no longer requires specific environmental cues, it is considered to be nondormant. Researchers have shown that various environmental factors can influence dormancy. For instance, while some suggest that only temperature can alter physiological dormancy following dispersal [23] demonstrated the induction of secondary dormancy in *Nicotiana attenuata* seeds by naturally occurring chemical

signals, such as abscisic acid (ABA) and other terpenes found in leachate from litter covering the seeds in their habitat. A seed that is entirely non dormant possesses the ability to germinate across the broadest range of normal physical environmental factors like temperature, water availability, nutrients that are feasible for its genotype [24].

3. EXPLORING DORMANCY PATTERNS IN MEDICINAL SEEDS

The exploration of seed germination in medicinal plant species has drawn particular attention from the scientific community, driven by the escalating demand for these plants in the pharmacological industry and the necessity to establish rational crops for herb production [4, 25]. In the domain of medicinal plant cultivation, recognizing and addressing seed dormancy is paramount for achieving successful germination and robust crop establishment. By elucidating the dormancy characteristics of various medicinal plant species, cultivators can refine their techniques and harness the full potential of these valuable botanical resources [26].

4. PHYSICAL DORMANCY

This type of hibernation arises from water-impermeable layers of palisade cells in the seed or fruit coat, which regulate water movement. Physical dormancy can be broken through mechanical or chemical scarification methods. Examples of species exhibiting this dormancy include *Melilotus* and *Trigonella* (Fabaceae) [27].Some of the plants with this type of dormancy can be treated by scarification method in which it alters a seed coat to make it more permeable to water and gases, which helps improve germination rates and speed up water absorption. Physical dormancy and its breaking methods for few crops are listed in Table 1.

Table 1. Physical dormancy of medicinal plants and their treatments

SN	Name of The Medicinal Plant	Breaking Methods	Reference
1	Common wormwood (*Artemisia absinthium*)	Pre soaking in distilled water for 8 h	[28]
2	Amaranthus (*Amaranthus annus*)	Pre soaking of seeds in distilled water for 6 h	[29].
3	Cornflower (*Centaurea cyanus*)	Mechanical scarification until the seed coat is damaged.	[30].
4	Saw Palmetto (*Serenoa repens*)	Scarification by sand paper	[31].
5	St.John's Wort (*Hypericum perforatum*)	Soaking in KNO_3 at 2g/lit for 3h	[32]
6	Andrographis(or King of Bitters) (*Andrographis paniculata*)	Acid scarification using H_2SO_4 for 4min.	[33]
7	Greater Burdock (*Arctium lappa*)	Leaching in fresh water	[34]
8	Chinese Lantern (*Physalis alkekengi*)	Soaking in GA_3 250ppm for 6h	[35]
9	Summer Savory (*Satureja hortensis*)	Soaking in KNO_3 at 2g/lit for 5h	[36]
10	Securigera (*Securigera securidaca*)	Leaching in fresh water	[36]
11	liquorice (*Glycyrrhiza glabra*)	Acid scarification for 5 min using HCl	[36]
12	Tanacetum (*Tanacetum sp*)	Soaking in KNO_3 at 2g/lit for 3h	[36]
13	Valerian (*Valeriana officinalis*)	Cold stratification at 5 ℃ for 8 days	[37]

5. PHYSIOLOGICAL DORMANCY

It is the most common type of hibernation found in seeds of gymnosperms and all major groups of angiosperms. It is especially prevalent in seeds from temperate regions [38]. It can be divided into three groups based on the depth or intensity of dormancy deep, intermediate and nondeep. Deep dormancy requires more prolonged exposure to cold stratification or other treatments to break dormancy, while nondeep physiological dormancy can be broken with shorter periods of stratification. The varying depths of this dormancy allow plants to stagger germination and protect themselves against unforeseen conditions. Its nuanced nature, with differing depths of dormancy, provides an adaptive advantage for plants in temperate climates with seasonal variations in climate [39]. Physiological dormancy has many reasons and background to occur and differs from one to on. Some of them are narrated below in Table 2.

Table 2. Physiological dormancy and their breaking treatments

SN	Name of the medicinal plant	Breaking methods	Reference
1	Dill (*Anethum graveolens*)	Pre chilling at 5 ℃for 6 days	[40].
2	Colocynth (*Citrulus colocynthis*)	Fluctuating temperatures	[41].
3	Wild sweet William (*Saponaria officinalis*)	Warm stratification	[42]
4	Ammoniacum (*Dorema ammoniacum*)	Cold stratification at 5 ℃ for 6 days	[43]
5	Purple cone flower (*Echinacea purpurea*)	Prechilling at 5 ℃ for 6 days	[44]
6	Galbanum (*Ferula gummosa*)	Prechilling at 5 ℃ for 6 days	[45]
7	Hyssop (*Hyssopus officinalis*)	Prechilling at 4 ℃ for 4 weeks	[46].
8	Tree Mallow (*Malva dendromorpha*)	Stratifcation (alternating temperatures)	[47]
9	High Mallow (*Malva silvestrys*)	Prechilling at 5 ℃ for 6 days	[48]
10	White Horehound (*Marrubium vulgare*)	Stratifcation (alternating temperatures)	[49].
11	Lemon Balm (*Melissa officinalis*)	Prechilling at 5 ℃ for 6 days	[50].
12	Oregano (*Origanum vulgare*)	Warm stratification	[51].
13	Flea Plant (*Plantago psyllium*)	Prechilling at 5 ℃ for 6 days	[36]
14	Black-eyes Susan (*Rudbeckia hirta*)	Prechilling at 5 ℃ for 6 days	[36]
15	Tobacco Sage (*Salvia dorrii*)	Cold stratification at 3-5 ℃	[36]
16	Common Sage (*Salvia officinalis*)	Pre chilling at 5 ℃ for 6 days	[36].
17	Ginseng (*Panaxginseng*)	cold stratification	[36]
18	Echinacea (*Echinacea purpurea*)	Stratification and Scarification	[36]
19	St. John's Wort (*Hypericumperforatum*)	Moist stratification at 4 ℃	[36].
20	Chamomile (*Matricari chamomilla*)	Stratification followed by pre soaking for 4h	[36]

SN	Name of the medicinal plant	Breaking methods	Reference
21	Feverfew (*Tanacetum parthenium*)	Warm stratification	[52]
22	Peppermint (*Mentha piperita*)	Stratification and prechilling treatment	[53]
23	AloeVera (*Aloebarbadensis*)	Chemical soaking in KNO_3 for 6h	[54]
24	Arnica (*Arnica montana*)	Warm stratification	[55]
25	Catnip (*Nepeta cataria*)	Cold stratification at 5 ℃ for 5 days.	[56]
26	Perwinkle (*Vinca rosea*)	Chemical soaking followed by warm stratification	[57]

6. MORPHOLOGICAL DORMANCY

It is characterized by seeds possessing underdeveloped embryos in terms of size, yet these embryos are differentiated into structures such as cotyledons and hypocotyl-radical. Seeds exhibiting this kind of dormancy are not physiologically dormant but rather require time for growth and germination. For instance, celery (*Apium graveolens*) exemplifies this dormancy [58]. The assertion that underdeveloped embryos represent the ancestral state of dormancy in seed plants suggests that this form of dormancy predates other forms such as physiological dormancy, morphophysiological dormancy and physical dormancy [59] . Seeds exhibiting morphological dormancy have evolved strategies to persist in the soil seed bank until all the requirements are fit for germination and seedling establishment. This dormancy mechanism allows plants to optimize their reproductive success by timing germination with favorable environmental conditions, thereby increasing the likelihood of seedling survival and establishment. Stratification methods also helps to overcome this type of dormancy.

7. MORPHO PHYSIOLOGICAL DORMANCY

It has a lot in common with morphological dormancy because it involves seeds that have immature embryos. Nonetheless, seeds exhibiting this also have a physiological aspect to their dormancy, which calls for a dormancy-breaking intervention such as the application of gibberellic acid or warm and/or cold stratification. There are eight recognized levels. *Trollius* (Ranunculaceae) and *Fraxinus excelsior* (Oleaceae) species are two examples [55] .

8. COMBINATIONAL DORMANCY

It is characterized by seeds possessing water-impermeable coats, as seen in physical dormancy, along with physiological embryo dormancy. Examples of species displaying dormancy include *Geranium* and *Trifolium* [24]. Medicinal plants often have specific requirements for germination, which may include exposure to particular temperature regimes, light conditions, or even chemical cues. Combinational dormancy in medicinal plant seeds may arise as an

Table 3. List of combinational dormancy and their breaking methods

SN	Medicinal Plant	Type of dormancy	Breaking methods	Reference
1	Anise (*Pimpinella anissum*)	Combinational dormancy	Scarification by sand and stratification at 20 ℃	[36]
2	Rue (*Ruta graveolens*)	Combinational dormancy	Stratification and scarification in mechanical scarifier for 12 min	[36]
3	**Milk Thistle (*Silybum marianum*)**	Combinational dormancy	Scarification and stratification at 3℃ for 48 days.	[61]

strategy to ensure that germination occurs under favorable conditions for seedling establishment and survival [60] . For example, seeds may exhibit both physiological dormancy, where internal physiological processes inhibit germination until certain conditions are met, as well as physical dormancy, where impermeable seed coats prevent water uptake and germination until they are sufficiently scarified or broken down (Table 3).

9. CONNECTING THE DOTS - DORMANCY AND QUIESCENCE IN BIOLOGICAL ADAPTATION

A condition of dormancy during which seeds are not actively growing or germination but are still viable is known as the quiescence period in seeds. At this time, seeds are essentially in a "resting" state, with little metabolic activity and little cellular activity. Quiescence is the character of seeds to survive for long periods of time in unfavourable environments until they are ready for germination. Quiescence is distinct from other types of dormancy in seeds, such as embryo dormancy or hibernation imposed by the seed coat, in that it is not always brought on by environmental cues like light or temperature. Rather, it is a natural characteristic of some seeds and is frequently genetically programmable. Seeds can emerge from their quiescent state and resume active growth and germination when the proper circumstances are met [62].

10. PLANT HORMONES AND THEIR MULTIFACETED ROLES IN SEED DORMANCY REGULATION

The opposing forces ABA and Gibberellic acid (GA) control seed dormancy. While mutants lacking GA biosynthesis or signaling have more dormancy, mutants lacking ABA biosynthesis or signaling often have less dormancy in seeds. This suggests that whilst GA promotes germination, ABA serves to induce dormancy. GA is hypothesized to have two effects on dormancy. It does this by first causing the expression of enzymes that weaken the endosperm and make room for the embryo to grow. Secondly, it directly increases the embryo's capacity for growth. In an experiment, [63] found that ABA may control GA responses in the endosperm, where GA operates predominantly to stimulate the break of dormancy . There is strong evidence that the ABA and GA signaling pathways interact in the embryo. The degree of dormancy in an embryo may be influenced by the balance of ABA and GA sensitivity, as mutations that damage one pathway might partially attenuate abnormalities in the other [64] his partial suppression, however, suggests that distinct ABA and GA pathways exist in parallel. Thus, ABA and GA have opposing and complementary effects on endosperm and embryo in the intricate hormonal regulation of seed dormancy [65] . Genetic regulatory networks of great complexity are involved in the crosstalk between ABA and GA_3, While DELLA (aspartic acid–glutamic acid–leucine–leucine–alanine) proteins are important in GA signaling pathways, transcription factors like ABI3 (ABA-INSENSITIVE 3) and ABI5 are crucial in mediating the effects of ABA on dormancy. In the promoter regions of target genes, interactions between ABA-responsive elements (ABREs) and GA-responsive elements (GAREs) coordinate the transcriptional responses to ABA and GA_3 during dormancy regulation [66]. Known for its role in senescence in plants and fruit ripening, ethylene has been found to be essential in a variety of physiological processes, such as the escape of dormancy in seeds, the development of root hairs, and reactions to viruses and wounds, among other stimuli [66]. Many plant species, including peanuts, apples, redroot pigweed, cocklebur, lamb's quarters, *Amaranthus retroflexus*, and sunflowers, have been discovered to break their dormancy when exposed to ethylene. Notably, research with the Arabidopsis ga-1 mutant has demonstrated that, in light settings, ethylene gas can cure germination problems; however, it will cause the seedlings to exhibit the distinctive triple response (Curved apical hook, swollen nodes and horizontal growth). This implies that certain ethylene signaling components are involved in the process of germination. The exact genetic processes by which ethylene affects seed germination, however, are still unknown [67].

It may become clearer whether ethylene promotes germination by modifying ABA or GA levels or sensitivity, or whether it works through different signaling components, by investigating its interactions with important regulators of ABA and GA production and signaling pathways in more detail. There might also be linkage with other signaling channels that are not yet known to exist according to traditional ethylene response mutant screening [68].

11. IMPACT OF DIFFERENT DORMANCY BREAKING TREATMENTS

Seed germination is a multifaceted physiological process shaped by environmental cues like water availability, light exposure, and other variables. The challenge of limited seed germination significantly hampers the cultivation of threatened medicinal plants, especially in harsh cold desert environments. Natural inhibitors derived from compounds like benzoic acid, cinnamic acid, coumarin, naringenin, jasmonic acid, and abscisic acid (ABA) play pivotal roles in regulating germination [65].Certain temperate species experience a warm phase that is followed by a cold one, which breaks their dormancy division into groups. The most common correlation between this reaction and morpho-physiological dormancy is that seeds that exhibit this response have embryos that are not fully formed [69].

To expedite this method further, it can be enhanced through the integration of additional treatments such as chemical applications or mechanical seed coat removal [70]. Various researchers have explored the impact of exogenous growth regulators on seed germination. For instance, Gibberellins have been found to eliminate the chilling requirements of peach and apple seeds, thereby enhancing their germination rates [70].Recent investigations have indicated that cold stratification directly influences the production of gibberellins (GAs) in seeds of Arabidopsis thaliana [71] externally applied GA has been shown to overcome seed dormancy in multiple species [4] and to stimulate germination in species that typically necessitate cold stratification, light exposure, or after-ripening [72] standard procedures for enhancing the germination of dormant seeds involve pre-chilling, scarification, and treatments with gibberellic acid (GA_3) or potassium nitrate (KNO_3).

Various methods, including scarification, using plant growth regulators as a pretreatment (PGRs), and temperature shocks, are employed to break dormancy, depending on the plant species and dormancy type [73,74]. Seed germination is influenced by both intrinsic and extrinsic factors, with certain PGRs playing a crucial role. Abscisic acid (ABA) is involved in germination inhibition, while gibberellins (GAs) participate in terminating seed dormancy [75,76]. PGRs commonly enhance seed germination capacity, increase biomass yield, and confer resistance to diseases and adverse growth conditions (Papadopoulos et al., 2006). GAs, synthesized by seeds, are believed to hydrolyze storage nutrients and directly impact embryo growth . External application of PGRs, including auxins like IAA, IBA and NAA, has been found to break seed dormancy and enhance seedling establishment in various aromatic and medicinal plants [72,77,78].

The study on *Ochradenus arabicus* seeds revealed GA_3 as the most effective in overcoming dormancy, resulting in 80-89% germination when treated with 100µM GA_3 and stored for up to 12 months. Increasing GA_3 concentration showed an inhibitory effect on seed germination, and seeds treated with different chemicals exhibited asynchronous germination. These findings suggest that GA_3 is the most effective in promoting seed germination in *O. arabicus*, emphasizing the necessity for hormone application to break dormancy, as documented by [79]. The dormant nature of *O.* arabicus seeds may be attributed to inhibitors, a hard seed coat, low internal hormones, or underdeveloped embryos, aligning with reports on the application of gibberellic acid to alleviate innate and environment-induced dormancy [80] .The seeds of glory lily soaked in boiled water (100 ℃) for 40 minutes recorded the highest germination of 62 per cent accompanied with faster rate of germination. This is because the seed coat gets weakened by the hot water and eventually the lignins and pectins present in the epidermal layer of seed gets dissolved [81]. Hence there are numerous ways to break down the dormancy present in seeds according to its nature.

12. SCARIFICATION

Scarification techniques have been evolved during time and has become practically feasible for all the crops. Some of the notable methods of scarification include heat, freeze-thaw, mechanical and chemical. Heat scarification uses high temperature to break the hard seed coat whereas freeze thaw breaks the seed coat by alternating temperatures to high and low. Mechanical scarification breaks the seed coat or makes an injury over it in order to accelerate the seed's rate of imbibition [82] .

13. HEAT SCARIFICATION

Heat scarification is way more easier method due its easy application. Two main devices that are being applied to scarify are oven and hot water

bath. Its efficacy depends on various factors like treatment, time, temperature and seed used. It differs from seed to seed like some may break at low temperature and some may be subjected to very high temperatures [83]. The primary purpose of heat scarification is to break seed dormancy. For some species, this dormancy is due to a hard seed coat that inhibits water absorption. Heat scarification helps to overcome this barrier, allowing water to penetrate the seed coat and initiate germination. The temperature and duration of heat exposure vary based on the species of seed being treated. Generally, temperatures range from 100 °C to 120 °C (212°F to 248°F), and exposure times can vary from a few minutes to several hours. It's crucial to research the specific necessities for the seeds you are treating, as excessive heat can damage or kill the seeds [84] .

Seeds can be exposed to dry heat using methods such as placing them in an oven, using hot sand, or applying heat from a heat gun. Dry heat methods are suitable for seeds that are tolerant of low moisture conditions. Seeds are subjected to high temperatures in a moist environment, such as steam or hot water [85]. Moist heat scarification is often preferred for seeds that require higher moisture levels for germination. When using heat scarification methods, it's essential to take proper safety precautions to avoid injury or damage to the seeds. This includes using heat-resistant containers or equipment, wearing protective gear when handling hot materials, and closely monitoring the temperature to prevent overheating (Priyadharshini & Lekha, 2021).

After heat scarification, seeds should be cooled and then planted promptly to take advantage of the weakened seed coat and initiate germination. It's important to provide optimal growing conditions, including proper moisture, light and temperature, to support seedling development. Heat scarification can be highly effective for breaking dormancy in certain species of seeds, particularly with hard seed coats or seeds from arid environments (Priyadharshini & Lekha, 2021). However, not all seeds require scarification, so it's essential to research the specific germination requirements of the seeds we are working with. Overall, heat scarification is a valuable technique for promoting germination in seeds that have dormancy mechanisms related to hard seed coats. When done correctly, it can significantly improve germination rates and support successful seedling establishment. Scarification duration differs from crop to crop.

14. FREEZE-THAW SCARIFICATION

The hidden theory behind this method is to make tiny scars on the seed coat and to make it weaken so that germination takes place. This scar making method depends on the seed size, shape, water content, intensity, duration etc. In this freeze thaw technology freezer, carbon dioxide, dry ice, snow, liquid air, acetone, liquid nitrogen are combined and used [90] . It has to be noted that temperature below-80℃ or lower, may be more effective than higher temperatures (above -80℃). Cryo scarification of seeds is a method that involves using liquid nitrogen and sulfuric acid to overcome the hardness of seeds' endocarps and seed coats. Some medicinal plants exposed to freeze thaw are listed below in Table 5.

Table 4. Scarification treatments

Sn	Crop	Treatment	Effect	References
1	Saskoon berry seeds (*Amelanchier alnifolia*)	Scarification at 110℃ with heat for 30 min	Radicle protrusion was earlier	[84].
2	Burclover (*Medicago polymorpha*)	Sandpaper scarification for 10-40min	Breakage of hard seed coat	[86].
3	Barrel Medic (*Medicago truncatula*)	20 min sandpaper scarification followed by mechanical scarifier	Imbibition of water takes place immediately after soaking	[87]
4	Arrow leaf (*Myriopteris intertexta*)	Mechanical scarifier for 15 min	Breakage of hard seedcoat	[88]
5	Loofah (*Luffa aegyptiaca*)	Mechanical scarifier at 100rpm for I1 min	Increases seed germination from 67 to 75%	[89]

Table 5. Freeze thaw treatments

Sn	Name of medicinal plant	Treatment	References
1	Alfalfa (*Medicago sativa*)	Exposing the seeds in -15 for 36h so that the seed coat becomes brittle	[90]
2	Ashwagandha (*Withania somnifera*)	Exposing the seeds at -22 ℃ for 180 days followed by placing the seeds in dry ice at -20℃.	[91]
3	Pepper (*Piper nigrum*)	Placing the seeds in liquid nitrogen for 10 min and soaking the seeds in hot water bath of 100℃ for 10 min.	[92]
4	Brahmi (*Bacopa monnieri*)	Placing of seeds in liquid nitrogen for 180 days.	[93]

15. PRECHILLING TREATMENT

The pre-chilling treatment proved an efficient in overcoming hibernation for *Calendula officinalis, Saponaria officinalis, Echinacea purpurea, Malva silvestrys, Melissa officinalis*, *Plantago psyllium*, and *Rudbeckia hirta*, resulting in increased germination among the 15 species subjected to pre-chilling. The most best results were noticed in *Plantago psyllium,* with germination increasing from 40 to 99%, and in *Saponaria officinalis,* from 0 to 30% [36]. Stratification improved germination in five out of six commercial *E. purpurea* seed lots, with the most significant improvement occurring at 10 °C for 10 days. Highest germination of *Echinacea angustifolia* in response to a combination of 250 ppm GA_3 and 4 weeks of pre-chilling was reported by [34]. Probert et al. 2009 also found that pre-chilling treatment was effective in enhancing *Saponaria officinalis* germination.

An increase in the germination of *Ferula gummosa* with pre-chilling at 5 °C was noticed by [94]. [77] observed the highest germination of *Dorema ammoniacum* in 30 days at 3 - 4 °C. It has been investigated that pre-chilling is effective in breaking dormancy in *Salvia sp.* The pre-chilling was not effective in breaking dormancy for *Calendula officinalis has been reported by* [34]*.* In the case of *Ferula gummosa* and Dorema ammoniacum, seeds did not germinate, and pre-chilling had no effect on breaking their dormancy. Nevertheless, both the application of GA3 and pre-chilling at 5 °C stimulated both the rate and final germination of *Ferula gummosa* [94]. Physiological dormancy in some species can be broken by relatively short periods of cold stratification. Thus, species for which pre-chilling was effective in breaking dormancy might possess physiological dormancy. A range of the prechilling treatments for medicinal plants are mentioned below in Table 6.

16. GIBBERELLIC ACID (GA_3)

The significant improvement in sprouting of seeds with GA_3 treatment in *Plantago psyllium*, *Rudbeckia hirta* and *Satureja hortensis* compared to untreated seeds were studied , inconclusive results were obtained in *Opuntia tomentosa* [103]. Plant growth regulators like GA_3 and KNO_3 seem to play a role in breaking dormancy by establishing hormonal balance and reducing germination inhibitor substances. The results were compatible with [104] observation of an enhanced germination rate in *Cuminum cyminum* with GA_3 application. [105] also reported increased germination of *Hyssopus officinalis* seeds with GA_3 pre-treatment. Loquat (*Eriobotrya japonica L.)* being a prime fruit crop that has potential of curative properties is mainly cultivated through seeds. The exogenous growth regulators can modify the digestibility of seed coat and if treated with GA_3 250 mg.L-1 soaking duration is 36h [77].

17. POTASSIUM NITRATE (KNO_3)

One of the major issue in cultivating medicinal herbs in a commercial way is they readily germinate in natural or native environment but when they are mandatory to test for germination under laboratory conditions they get failed. Dormancy is not a single phenomenon and it contributes together with various aspects [106] KNO_3 was found to be effective in breaking seed dormancy for *Calendula officinalis, Cynara scolymus, Ocimum basilicum, Physalis alkekengi,* and *Satureja hortensis* compared to their germination in H_2O. Similar instances were noted in studies on *Calendula officinalis* and *Echinacea*

purpurea [34] As mentioned earlier, KNO_3, acting as a plant growth regulator, plays a role in breaking dormancy by establishing hormonal balance and reducing germination inhibitor substances. Galbanum seeds when stratified with perlite medium and then subjected to various concentrations of KNO_3 (0, 0.1, 0.2 and 0.3% (v/v)),after 7 weeks of stratification the seeds soaked in 0.3 percent KNO_3 the germination percentile shoot up to 86 % [98].

18. STRATIFICATION OF SEEDS

Exposure to cold temperatures during stratification mimics the natural winter conditions that many seeds would experience in their native habitats. This cold treatment is particularly effective for seeds of plants adapted to temperate climates [107]. The cold temperatures initiate physiological changes within the seed that break dormancy and prepare it for germination when conditions become favorable. In moist stratification, seeds soak up moisture from their environment. This hydration softens the outer covering and triggers biochemical processes within the seed [108]. The cold and sometimes moist conditions of stratification can soften the seed coat, which is often impermeable to water and gases. Softening of the seed coat allows for enhanced water uptake and gas exchange, facilitating metabolic activity within the seed.

Table 6. List of medicinal herbs and their prechilling treatments

Sn	Medicinal plant	Prechilling treatment	Effect	References
1	Corn Poppy (*Papaver rhoeash*)	Wet pre chilling	Improved germination rate	[95]
2	Long-headed Poppy (*Papaver dubium*)	Wet pre chilling	Increased seed germination rate from 46% to 90%	[96]
3	Alfalfa (*Medicago sativa*)	Prechilling followed by soaking of seeds in GA3 at 250ppm for 1 week	Seed germination was easy as the hard seed coat gets softened	[97]
4	California Burclover (*Medicago polymorpha*)	Prechilling at 20 ℃ for a week	Seedcoat was permeable	[98]
5	Yellow Sweet Clover (*Melilotus officinalis*)	Prechilling at 4℃ followed by soaking in GA3 at 750ppm	Physical dormancy was broken	[45].
6	Yellow Sweet Clover (*Melilotus oficinalis*)	Prechilling at 4℃	Maturity of embryo was quick	[96]
7	Douglas Fir (*Psedostuga menziesii*)	Prechilling at 15℃ for 128 weeks	Protrusion of embryo	[99]
8	Asafoetida (*Ferula assafoetida*)	Prechilling at 9℃ for 90 days	Increased germination percentage	[100]
9	Galbanum (*Ferula gummosa*)	Prechilling at 5℃ and soaking of seeds in GA3 at 400ppm	Sprouting of seeds	[101]
10	Colocynth (*Citrulus colocynthis*)	Prechilling at 4℃for 50 days	Dormancy was broken and germination at earlier stage.	[102]

Table 7. Stratification treatments

SN	Medicinal plant	Treatment	References
1	Fennel *Foeniculum vulgare*	Exposing the seeds at 10℃ for about weeks	[109]
2	Echinacea *Echinacea sp*	Exposing the seeds at 4℃ for 10 days	[110]
3	Kalajira *Bunium persium*	Moist stratification at 4℃ for 5 days	[111]
4	Loddon lilly *Leucojum aestivum*	Warm stratification at 30℃ for 6 weeks	[112]

19. SECONDARY METABOLITES (SM) - METABOLIC MAESTROS OF DORMANCY AND LONGEVITY

The duration of dormancy is critical for the secondary metabolites. When seeds remain dormant, they protect as a defensive mechanism against a distinct combination of biotic and abiotic stressors. Certain secondary metabolites, like as phenolic compounds, have antioxidant properties that shield them from oxidative damage. As a result, they provide protection throughout the dormant phase, raising the likelihood of successful germination in the event that favorable conditions continue.

SMs found in medicinal plants are essential components that underpin their clinical curative effects and serve as crucial indicators for evaluating the quality of medicinal materials. The synthesis and accumulation of SMs are intricate processes influenced by both internal developmental genetic circuits (such as regulated genes and enzymes) and external environmental factors (including light, temperature, water, and salinity). While existing literature extensively explores the impact of environmental factors on SM synthesis and accumulation, there is a gap that requires a systematic classification and summary of the effects of developmental growth and genetic factors on SMs. The biosynthesis of SMs initiates from basic pathways such as glycolysis or shikimic acid pathways, with subsequent diversification depending on cell type, developmental stage, and environmental cues [113]. Distinct cells, tissues and organs of medicinal plants may exhibit different medicinal properties at various developmental stages due to the influence of developmental factors on cellular structures involved in SM biosynthesis and storage [52]. Plant growth and development, influenced or impeded by different environmental conditions, significantly impact the accumulation of secondary metabolites [114]. Ashwagandha being an important medicinal plant of Indian origin has numerous secondary metabolites in which Withanaloid increases during saline stress is evident in experimental study and all the secondary metabolites behave in the same way to express their origin during external stress condition [115]. Environmental variations strongly affect SM pathways and their regulation, as the expression of genes involved in SM pathways undergoes alterations in response to different stresses [116]. Secondary metabolites have a heavy interaction with external environment in plants and are initially produced by primary metabolites in higher plants [117]

The plant kingdom boasts approximately 100,000 secondary metabolites, classified into three major groups based on bio synthetic pathways: nitrogen-containing compounds (cyanogenic glycosides, alkaloids, and glucosinolates), phenolic compounds (flavonoids and phenylpropanoids), and terpenes (isoprenoids) [118]. Despite progress in SM biosynthesis and accumulation research, reports on how developmental and environmental factors stimulate the synthesis and accumulation of secondary metabolites in medicinal plants are limited. The stimulation of metabolites biosynthesis in medicinal plants by controlling and optimizing external and internal factors holds promise for advancing bio technologies aimed at high-quality drug production [119].

Secondary metabolite synthesis, accumulation, and distribution patterns in medicinal plants categorize medicinal parts into four main types: roots and stems, leaves, flowers, and fruits and seeds. The complexity and diversity of these metabolites in different parts of medicinal plants end up to the synthesis of different SMs through specific regulatory pathways and transport routes in certain organs, tissues, and cells [42]. Thus, SM biosynthesis and accumulation exhibit organ or tissue specificity.

Fruits and seeds of medicinal materials in many plants, are influenced by their developmental stages, impacting the content and composition of components. For instance, in Citrus fruits, volatile oil content, the main active ingredient, is highest when the fruit is light yellow, serving as a morphological index for harvesting [120]. Essential oil yields in *Citrus medica L. var. sarcodactylis* significantly increase during maturation, and the content of specific compounds varies during maturation stages [121]. Similarly, capsules of *Papaver somniferum L*. reach the maximum morphine content at maturity [122]. Therefore, the developmental stages of plants significantly affect the content and composition of SMs. The synthesis and accumulation of SMs are also closely associated with the developmental stage of medicinal plant seeds. For example, in coffee quinic acid content remains relatively stable, but dicoffee quinic acid content decreases noticeably with seed development. The content of quinic acid, a precursor substance for chlorogenic acid synthesis, is high in the early developmental stage of seeds and decreases significantly later on, related to the expression characteristics of the HQT gene, a key enzyme regulating phenolic acid biosynthesis [123].

20. CONCLUSION

This comprehensive review of medicinal plant seeds and their dormancy mechanisms highlights the critical importance of seed biology in the conservation and cultivation of valuable medicinal species. The diverse dormancy types observed reflect complex evolutionary adaptations, with physiological dormancy being prevalent among many medicinal plants. Environmental factors such as temperature and light play significant roles in regulating dormancy breaking and germination processes. The insights gained from this review can inform more effective strategies for medicinal plant propagation, conservation, and sustainable use, addressing the rising global demand for natural medicines**.** Since prehistoric times therapeutic plants have many distinct purposes. Protection and Defense activity is one of the significant act of medicinal herbs. Seed market is expected to be on top with hybrid varieties developed indigenously for domestic markets and commercial farmer. Studies on these medicinal plant remains to be a growing part for further research. Dormancy pertaining to the seeds must be considered as in advantage or disadvantage way according to the various external factors. Sometimes this bane helps in seeds preservation and in other case it is considered as serious issue for germination. Since the availability of seeds itself is difficult these days, considering these dormancy issues can lead to a big advantage in pharmacological industry for availing drugs at earlier and cost efficient way.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during writing or editing of this manuscript.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Abdallah EM, et al. Back to Nature: Medicinal plants as promising sources for antibacterial drugs in the post-antibiotic era. Plants. 2023;12(17):3077.
2. Zare A, et al. Effect of various treatments on seed germination and dormancy breaking in Ferula assa *foetida L*.(Asafetida), a threatened medicinal herb. Trakia Journal of Sciences. 2011;9(2):57-61.
3. McDonald MB. Seed priming. Seed Technology and its Biological Basis. 2000;287:325.
4. Hassan S, Mathesius U. The role of flavonoids in root–rhizosphere signalling: Opportunities and challenges for improving plant–microbe interactions. Journal of Experimental Botany. 2012;63(9):3429-3444.
5. Chen SL, et al. Conservation and sustainable use of medicinal plants: Problems, progress, and prospects. Chinese Medicine. 2016;11:1-10.
6. Shafi A, Hassan F, Zahoor I, Majeed U, Khanday FA. Biodiversity, management and sustainable use of medicinal and aromatic plant resources. Medicinal and aromatic plants: Healthcare and industrial Applications. 2021;85-111.
7. Balunas MJ, Kinghorn AD. Drug discovery from medicinal plants. Life Sciences. 2005;78(5):431-441.

8. Chacko SM, et al. Beneficial effects of green tea: A literature review. Chinese Medicine. 2010;5(1):1-9.
9. Cole IB, Saxena PK, Murch SJ. Medicinal biotechnology in the genus Scutellaria. *In vitro* Cellular and Developmental Biology-Plant. 2007;43:318-327.
10. Pimm SL, et al. The future of biodiversity. Science. 1995;269(5222):347-350.
11. Heywood VH, Iriondo JM. Plant conservation: Old problems, new perspectives. Biological Conservation. 2003;113(3):321-335.
12. Hamilton AC. Medicinal plants, conservation and livelihoods. Biodiversity and Conservation. 2004;13:1477-1517.
13. Ross IA. Medicinal plants of the world, volume 3: Chemical constituents, traditional and modern medicinal uses. Springer; 2005.
14. Larsen HO, Olsen CS. Unsustainable collection and unfair trade? Uncovering and assessing assumptions regarding Central Himalayan medicinal plant conservation. Biodiversity and Conservation. 2007;16:1679-1697.
15. Zerabruk S, Yirga G. Traditional knowledge of medicinal plants in Gindeberet district, Western Ethiopia. South African Journal of Botany. 2012;78:165-169.
16. Srujana TS, Konduri RB, Rao BSS. Phytochemical investigation and biological activity of leaves extract of plant Boswellia serrata. The Pharma Innovation. 2012;1(5,Part A):22.
17. Schipmann U, et al. Impact of cultivation and collection on the conservation of medicinal plants: Global trends and issues. in III WOCMAP Congress on Medicinal and Aromatic Plants-Volume 2: Conservation, Cultivation and Sustainable Use of Medicinal and 676; 2003.
18. Macchia M, Angelini LG, Ceccarini L. Methods to overcome seed dormancy in Echinacea angustifolia DC. Scientia Horticulturae. 2001;89(4):317-324.
19. Thompson K, et al. Are seed dormancy and persistence in soil related? Seed Science Research. 2003;13(2):97-100.
20. Fenner M, Thompson K. The ecology of seeds. Cambridge University Press; 2005.
21. Baskin JM, Baskin CC. The great diversity in kinds of seed dormancy: A revision of the Nikolaeva–Baskin classification system for primary seed dormancy. Seed Science Research. 2021;31(4):249-277.
22. Vleeshouwers L, Bouwmeester H, Karssen C. Redefining seed dormancy: An attempt to integrate physiology and ecology. Journal of Ecology. 1995;1031-1037.
23. Krock B, et al. Vegetation-derived abscisic acid and four terpenes enforce dormancy in seeds of the post-fire annual, Nicotiana attenuata. Seed Science Research. 2002;12(4):239-252.
24. Baskin CC, Baskin JM. Germinating seeds of wildflowers, an ecological perspective. Hort Technology. 2004;14(4):467-473.
25. Sajjadi SE. Analysis of the essential oils of two cultivated basil (*Ocimum basilicum L.*) from Iran. DARU Journal of Pharmaceutical Sciences. 2006;14(3):128-130.
26. Penfield S, MacGregor DR. Effects of environmental variation during seed production on seed dormancy and germination. Journal of Experimental Botany. 2017;68(4):819-825.
27. Barekat T, et al. A novel approach for breaking seed dormancy and germination in Viola odorata (A medicinal plant). Journal of Novel Applied Sciences. 2013;2(10):513-516.
28. Mannan A, et al. Effect of growth regulators on *In vitro* germination of Artemisia absinthium. Scientific Research and Essays. 2012;7(14):1501-1507.
29. Kępczyński J, Bihun M, Kępczyńska E. The release of secondary dormancy by ethylene in *Amaranthus caudatus* L. seeds. Seed Science Research. 2003;13(1):69-74.
30. Stankiewicz-Kosyl M, Haliniarz M. Diversified germination strategies of Centaurea cyanus populations resistant to ALS inhibitors. Plant Protection Science. 2023;59(4).
31. Schmalzer PA, Foster TE. Dynamics of gaps created by burning in Florida oak–saw palmetto (Quercus, Fagaceae–Serenoa repens, Arecaceae) scrub1, 2. The Journal of the Torrey Botanical Society. 2018;145(3):250-262.
32. Pérez-García F, et al. Hypericum perforatum L. seed germination: Interpopulation variationand effect of light, temperature, presowing treatments and seed desiccation. Genetic Resources and Crop Evolution. 2006;53:1187-1198.
33. Kumar B, Verma SK, Singh H. Effect of temperature on seed germination parameters in Kalmegh (*Andrographis paniculata* Wall. ex Nees.). Industrial

Crops and Products. 2011;34(1):1241-1244.
34. Farhoudi R, Modhej A, Motamedi M. Evalution of Arctium lappa seed dormancy breaking methods; 2021.
35. Asgari A, Moghaddam PR, Koocheki A. Methods for breaking Chinese lantern (*Physalis alkekengi L.*) seed dormancy. Laboratory and greenhouse studies. Revista de la Facultad de Agronomia de la Universidad del Zulia. 2018;35(2):127-151.
36. Aghilian S, Khajeh-Hosseini M, Anvarkhah S. Evaluation of seed dormancy in forty medicinal plant species. International Journal of Agriculture and Crop Sciences. 2014;7(10):760.
37. Heidari Sureshjani Z, Karimzadeh G, Rashidi Monfared S. The first *In vitro* study of seed dormancy breakage in Iranian populations of St. John's Wort (Hypericum perforatum) with different geographical origins. Iranian Journal of Seed Research. 2022;9(1):59-74.
38. Bano H, et al. Effect of pre-sowing treatments using phytohormones and other dormancy breaking chemicals on seed germination of Dioscorea deltoidea Wall. Ex Griseb. An Endangered Medicinal Plant Species of North Western Himalaya. Eco. Env. and Cons. 2021;27:253-260.
39. Zarei-Gavkosh M, Afshari RT, Jahansooz M. Morphophysiological dormancy in smyrnium cordifolium boiss: Germination requirements and embryo growth. Journal of Applied Research on Medicinal and Aromatic Plants. 2022;30:100385.
40. Bukharov AF, et al. Impacts of high temperature on embryonic growth and seed germination of dill (Anethum graveolens). Seed Science and Technology. 2021;49(1):7-17.
41. Saberi M, et al. Comparison the effect of different treatments for breaking seed dormancy of Citrullus colocynthis. Journal of Agricultural Science. 2011;3(4):62.
42. Li Y, et al. The effect of developmental and environmental factors on secondary metabolites in medicinal plants. Plant Physiology and Biochemistry. 2020;148:80-89.
43. Naseri H, Hosseini SA, Lashkari Sanami N. Effect of cooling pre-treatment on germination criteria and drought stress resistance in Dorema ammoniacum. Iranian Journal of Seed Science and Technology. 2019;8(1):241-251.
44. Qu L, et al. Commercial seed lots exhibit reduced seed dormancy in comparison to wild seed lots of Echinacea purpurea. Hort Science. 2005;40(6):1843-1845.
45. Rahnama-Ghahfarokhi A, Tavakkol-Afshari R. Methods for dormancy breaking and germination of galbanum seeds (Ferula gummosa). Asian Journal of Plant Sciences. 2007;6(4):611-616.
46. Arves J, et al. Effect of salinity and temperature interaction on germination of hyssop (*Hyssopus officinalis*). World Applied Sciences Journal. 2013;22(7): 1024-1031.
47. Aghilian S, Khajeh-Hosseini M, Anvarkhah S. Evaluation of seed storage potential in forty medicinal plant species. International Journal of Agriculture and Crop Sciences. 2014;7(10):749-759.
48. Ansari O, et al. Breaking seed dormancy and determining cardinal temperatures for Malva sylvestris using nonlinear regression. Seed Science and Technology. 2016;44 (3):447-460.
49. Javaid MM, et al. Environmental factors affecting the germination and emergence of white horehound (*Marrubium vulgare L.*): A weed of arid-zone areas. The Rangeland Journal. 2018;40(1):47-54.
50. Fahim J, et al. Anti-dormant mycobacterial activity of *Melissa officinalis L.*(Lemon balm). Journal of advanced Biomedical and Pharmaceutical Sciences. 2021;4 (2):95-97.
51. Farashah HD, et al. Germination improvement and alpha-amylase and beta-1, 3-glucanase activity in dormant and non-dormant seeds of Oregano (*'Origanum vulgare'*). Australian Journal of Crop Science. 2011;5(4):421-427.
52. Brown AM, et al. Feverfew (*Tanacetum parthenium L.):* Tissue culture and parthenolide synthesis. Plant Science. 1996;116(2):223-232.
53. Cooper EG, Arana J, Meyers SL. Simulated dormant peppermint (Mentha× piperita) response to mesotrione: A greenhouse study. Weed Technology. 2023;37(2):192-196.
54. Kulkarni MG, et al. Seed germination and phytochemical evaluation in seedlings of Aloe arborescens Mill. Plant Biosystems-An International Journal Dealing with all Aspects of Plant Biology. 2014;148(3):460-466.
55. Balabanova V, Vitkova A, Zheleva-Dimitrova D. Comparative study of

germination and seed antioxidant activity of *Arnica montana L.* and Arnica chamissonis Less.(Asteracea). Comptes Rendus L'académie Bulg. Sci. Sci. Mathématiques Nat. 2013;66:1261-1268.
56. Aćimović M, et al. Nepeta cataria–cultivation, chemical composition and biological activity. Journal of Agronomy, Technology and Engineering Management (JATEM). 2021;4(4):620-634.
57. Mohammadzadeh-Shahir M, et al. The potential use of methyl jasmonate, putrescine and cold atmospheric plasma on genetic variability and seedling growth improvement in medicinal plant *Catharanthus roseus L*. cultivar. Industrial Crops and Products. 2019;140:111601.
58. Jaganathan GK. Defining correct dormancy class matters: Morphological and morphophysiological dormancy in Arecaceae. Annals of Forest Science. 2020;77:1-6.
59. Yaqoob U, Nawchoo IA. Conservation and cultivation of Ferula jaeschkeana Vatke: A species with deep complex morphophysiological dormancy. Proceedings of the National Academy of Sciences, India Section B: Biological Sciences. 2017;87:315-325.
60. Cho JS, Jang BK, Lee CH. Breaking combinational dormancy of Rhus javanica L. seeds in South Korea: Effect of mechanical scarification and cold-moist stratification. South African Journal of Botany. 2020;133:174-177.
61. Pourreza J, Bahrani A. Estimating cardinal temperatures of milk thistle (Silybum marianum) seed germination. American-Eurasian Journal Agricultural Environmental Science. 2012;12(11):1485-89.
62. Velappan Y, Signorelli S, Considine MJ. Cell cycle arrest in plants: What distinguishes quiescence, dormancy and differentiated G1? Annals of Botany. 2017;120(4):495-509.
63. Obroucheva N. Transition from hormonal to nonhormonal regulation as exemplified by seed dormancy release and germination triggering. Russian Journal of Plant Physiology. 2012;59(4):546-555.
64. Brady SM, McCourt P. Hormone cross-talk in seed dormancy. Journal of Plant Growth Regulation. 2003;22:25-31.
65. Shu K, et al. Dormancy and germination: How does the crop seed decide? Plant Biology. 2015;17(6):1104-1112.
66. Finkelstein RR. E4. The role of hormones during seed development and germination. Plant Hormones. 2004;513.
67. Jhanji S, et al. Exploring fine tuning between phytohormones and ROS signaling cascade in regulation of seed dormancy, germination and seedling development. Plant Physiology and Biochemistry. 2024;108352.
68. Seo M, Jikumaru Y, Kamiya Y. Profiling of hormones and related metabolites in seed dormancy and germination studies. Seed dormancy: Methods and Protocols. 2011;99-111.
69. Atia A, et al. Alleviation of salt-induced seed dormancy in the perennial halophyte *Crithmum maritimum L*.(Apiaceae). Pak. J. Bot. 2006;38(5):1367-1372.
70. Eberle CA, et al. Seed germination of calendula in response to temperature. Industrial Crops and Products. 2014;52:199-204.
71. Masferrer A, et al. Overexpression of Arabidopsis thaliana farnesyl diphosphate synthase (FPS1S) in transgenic Arabidopsis induces a cell death/senescence-like response and reduced cytokinin levels. The Plant Journal. 2002;30(2):123-132.
72. Kandari L, et al. Effect of pre-sowing, temperature and light on the seed germination of Arnebia benthamii (Wall. ex G. Don): An endangered medicinal plant of Central Himalaya, India. African Journal of Plant Science. 2008;2(1):005-011.
73. Copeland LO, McDonald MF. Principles of seed science and technology. Springer Science and Business Media; 2012.
74. Hidayati SN, et al. Sympatric species of Hibbertia (Dilleniaceae) vary in dormancy break and germination requirements: Implications for classifying morphophysiological dormancy in Mediterranean biomes. Annals of Botany. 2012;109(6):1111-1123.
75. Dewir YH, El-Mahrouk MES, Naidoo Y. Effects of Some Mechanical and Chemical Treatments on Seed Germination of'Sabal palmetto'and'Thrinax morrisii'Palms. Australian Journal of Crop Science. 2011; 5(3):248-253.
76. Iwasaki M, Penfield S, Lopez-Molina L. Parental and environmental control of seed dormancy in Arabidopsis thaliana. Annual Review of Plant Biology. 2022;73:355-378.
77. Al-Hawezy SMN. The role of the different concentrations of GA3 on seed

germination and seedling growth of loquat (*Eriobotrya japonica L*.). IOSR Journal of Agriculture and Veterinary Science. 2013; 4(5):3-6.
78. Sivakumar V, et al. Effects of presowing treatments, desiccation and storage conditions on germination of Strychnos nux-vomica seeds, a valuable medicinal plant. New Forests. 2006;32:121-131.
79. Yücel E, Yilmaz G. Effects of different alkaline metal salts (NaCl, KNO3), acid concentrations (H2SO4) and growth regulator (GA3) on the germination of Salvia cyanescens Boiss. And Bal. seeds. Gazi University Journal of Science. 2009;22(3):123-127.
80. Yarnia M, Tabrizi EFM. Effect of seed priming with different concentration of GA3, IAA and kinetin on Azarshahr onion germination and seedling growth. Journal of Basic and Applied Scientific Research; 2012.
81. Venudevan B, Sundareswaran S, Vijayakumar A. Optimization of dormancy breaking treatments for germination improvement of glory lily (*Gloriosa superba L*.) seeds. Madras Agricultural Journal. 2010;97(jan-mar):1.
82. Khanduri P, Negi K. Effect of scarification to enhance seed germination of certain woody plant species. The Journal of Indian Botanical Society. 2010;89(3and4):308-311.
83. Górnik K, et al. The effect of different stratification and scarification treatments on breaking the dormancy of saskatoon berry seeds. Agronomy. 2023;13(2):520.
84. Debouza NE, et al. Mechanical scarification: The key to optimal germination parameters in nine flowering plants of the United Arab Emirates; 2024.
85. Niu J, et al. The effects of ascorbic acid on breaking the seed dormancy of Malus sieversii. Journal of Plant Growth Regulation. 2019;38:909-918.
86. Alane F, et al. Break dormancy, germination capacity of medics after different techniques of scarification (physical, chemical and mechanical). African Journal of Agricultural Research. 2016;11(5):340-351.
87. Nawrot-Chorabik K, et al. Stratification, scarification and application of phytohormones promote dormancy breaking and germination of pelleted scots pine (*Pinus sylvestris L*.) seeds. Forests. 2021;12(5):621.
88. Choi G, et al. Scarification and stratification protocols for breaking dormancy of Rubus (Rosaceae) species in Korea. Seed Science and Technology. 2016;44(2):239-252.
89. Chokchaichamnankit D, et al. Proteomic alterations during dormant period of Curcuma longa rhizomes. J Proteomics Bioinform. 2009;2:380-387.
90. Farida A, Rabeha C, Aissa A. Effect of different scarification techniques on the germination of some endemic Medicago species in Algeria. PONTE International Journal of Science and Research. 2020;76(8).
91. Kimura E, Islam M. Seed scarification methods and their use in forage legumes. Research Journal of Seed Science. 2012;5(2):38-50.
92. Dosmann MS, Iles JK. Cold stratification improves germination of katsura tree. HortScience. 1997;32(3):447F-448.
93. Aouadj SA, et al. Preliminary study of the pre-germinative treatments of *Juniperus oxycedrus L*. and *Pistacia lentiscus L*. in the Saida region (Western Algeria). Biodiversity Research and Conservation. 2022;67(1):13-20.
94. Nadjafi F, et al. Seed germination and dormancy breaking techniques for Ferula gummosa and Teucrium polium. Journal of Arid Environments. 2006;64(3):542-547.
95. Golmohammadzadeh S, Zaefarian F, Rezvani M. Effects of some chemical factors, prechilling treatments and interactions on the seed dormancy-breaking of two Papaver species. Weed Biology and Management. 2015;15(1):11-19.
96. Balouchi HR, Sanavy S. Effect of gibberellic acid, prechilling, sulfuric acid and potassium nitrate on seed germination and dormancy of annual Medics. Pakistan Journal of Biological Sciences. 2006;9 (15):2875-2880.
97. Gosling PG, Samuel Y, Peace A. The effect of moisture content and prechill duration on dormancy breakage of Douglas fir seeds (Pseudotsuga menziesii var. menziesii [Mirb.] Franco). Seed Science Research. 2003;13(3):239-246.
98. Sharifi H, Nemati A, Gerdakaneh M. Breaking seed dormancy and improve germination of four medicinal species of Apiaceae by gibberellic acid and prechilling treatments; 2017.

99. Ertle JM. Effects of short-term chilling stress on seedling quality and post-transplanting growth of grafted and nongrafted Watermelon. The Ohio State University; 2020.
100. Keshtkar H, Azarnivand H, Atashi H. Effect of prechilling and GA3 on seed germination of Ferula assa-foetida and Prangos ferulacea. Seed Science and Technology. 2009;37(2):464-468.
101. Gastmann, J., et al., Vitis labrusca L. germination: influence of treatments to break dormancy, storage and ripening point of fruits. Vitis, 2023. 62(3).
102. Kahrom N, Farzam M, Mesdaghi M. Autecology of Colocynth (*Citrullus colocynthis L. Schrad*) in Gonabad Desert, Iran. Journal of Rangeland Science. 2022; 12(2):129-140.
103. Olvera-Carrillo Y, et al. Germination of the hard seed coated Opuntia tomentosa SD, a cacti from the México valley. Journal of Arid Environment. 2003;55(1):29-42.
104. Alp S, Ipek A, Arslan N. The effect of gibberellic acid on germination of rosehip seeds (*Rosa canina L*.). in I International Symposium on Woody Ornamentals of the Temperate Zone 885; 2008.
105. Petrić M, et al. The effect of low temperature and GA 3 treatments on dormancy breaking and activity of antioxidant enzymes in Fritillaria meleagris bulblets cultured *In vitro*. Acta Physiologiae Plantarum. 2013;35:3223-3236.
106. Finkelstein R, et al. Molecular aspects of seed dormancy. Annu. Rev. Plant Biol. 2008;59:387-415.
107. Bratcher CB, Dole JM, Cole JC. Stratification improves seed germination of five native wildflower species. HortScience. 1993;28(9):899-901.
108. Milberg P, Andersson L. Does cold stratification level out differences in seed germinability between populations? Plant Ecology. 1998;134:225-234.
109. Hashemirad S, et al. Cold stratification requirement to break morphophysiological dormancy of fennel (*Foeniculum vulgare Mill*.) seeds varies with seed length. Journal of Applied Research on Medicinal and Aromatic Plants. 2023;35:100465.
110. Romero FR, Delate K, Hannapel DJ. The effect of seed source, light during germination, and cold-moist stratification on seed germination in three species of Echinacea for organic production. HortScience: A publication of the American Society for Horticultural Science. 2005; 40(6):1751.
111. Sharma R, Sharma S., Effect of storage and cold-stratification on seed physiological aspects of Bunium persicum: A threatened medicinal herb of Trans-Himalaya. Int. J. Bot. 2010;6(2):151-156.
112. Çiçek E, Aslan M, Tilki F. Effect of stratification on germination of Leucojum aestivum L. seeds, a valuable ornamental and medicinal plant. Research Journal of Agriculture and Biological Sciences. 2007;3(4):242-244.
113. Patra B, et al. Transcriptional regulation of secondary metabolite biosynthesis in plants. Biochimica et Biophysica Acta (BBA)-Gene Regulatory Mechanisms. 2013;1829(11):1236-1247.
114. Zhang X, et al. Endophytic fungus Trichothecium roseum LZ93 antagonizing pathogenic fungi *In vitro* and its secondary metabolites. The Journal of Microbiology. 2010;48:784-790.
115. Sanchita, et al. Physiological performance, secondary metabolite and expression profiling of genes associated with drought tolerance in Withania somnifera. Protoplasma. 2015;252:1439-1450.
116. Borges TH, et al. Comparative analysis of minor bioactive constituents (CoQ10, tocopherols and phenolic compounds) in Arbequina extra virgin olive oils from Brazil and Spain. Journal of Food Composition and Analysis. 2017;63:47-54.
117. Ashraf MA, et al. Environmental stress and secondary metabolites in plants: An overview. Plant Metabolites and Regulation Under Environmental Stress. 2018;153-167.
118. Fang J, et al. Tissue-specific distribution of secondary metabolites in rapeseed (*Brassica napus L*.). Plos One. 2012;7(10):e48006.
119. Perchuk I, et al. Composition of primary and secondary metabolite compounds in seeds and pods of asparagus bean (*Vigna unguiculata (L.) Walp*.) from China. Molecules. 2020;25(17):3778.
120. Li Y, et al. Effects of salt stress on seed germination of Saponaria officinalis. Journal of Northeast Forestry University. 2019;47(9):17-27.
121. Wu X, et al. Drought stress and re-watering increase secondary metabolites and enzyme activity in dendrobium moniliforme. Industrial Crops and Products. 2016;94:385-393.

122. Verma N, Shukla S. Impact of various factors responsible for fluctuation in plant secondary metabolites. Journal of Applied Research on Medicinal and Aromatic Plants. 2015;2(4):105-113.

123. Awada R, et al. Global transcriptome profiling reveals differential regulatory, metabolic and hormonal networks during somatic embryogenesis in Coffea arabica. BMC Genomics. 2023;24(1):41.

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