

Feature Review

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Gene Expression Analysis and Regulatory Mechanisms of Pine Seed Germination

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Abstract Pine species are crucial to forest ecosystems and the timber industry, yet understanding their seed germination processes presents both challenges and opportunities. This study explores gene expression dynamics and regulatory mechanisms during pine seed germination, providing insights into key stages, major genes involved, and the techniques used for analysis. We investigate hormonal, environmental, genetic, and epigenetic factors influencing gene expression, supported by detailed case studies and comparative analyses across different pine species. Technological advances in transcriptomics, genomics, and bioinformatics have been leveraged to elucidate gene expression profiles. The findings have significant implications for enhancing germination rates, genetic engineering, and sustainable forestry practices. By integrating gene expression data into conservation strategies, this study aims to improve pine seed conservation and develop informed policies for species preservation. Future research directions highlight emerging trends, potential collaborations, and ongoing research needs in pine seed germination and conservation. This study contributes to a deeper understanding of pine biology, offering practical applications for forestry and conservation.

Keywords Pine seed germination; Gene expression; Regulatory mechanisms; Genetic analysis; Conservation strategies

1 Introduction

Pine species are integral components of forest ecosystems and hold significant value in the timber industry. Understanding the gene expression and regulatory mechanisms during pine seed germination is crucial for improving forest management practices and enhancing the adaptability of these species to changing environmental conditions (Salazar et al., 2018; Chen et al., 2023). Pine species, such as the Loblolly pine (*Pinus taeda* L.) and Scots pine (*Pinus sylvestris* L.), play a pivotal role in forest ecosystems by contributing to biodiversity, carbon sequestration, and soil stabilization (Galibina et al., 2023). They are also economically important due to their use in the timber industry for producing wood, paper, and other forest products (Nawrot-Chorabik et al., 2021). The Loblolly pine, for instance, is widely used in afforestation projects due to its rapid growth and adaptability to various environmental conditions (Mao et al., 2021). Additionally, pine species are sources of valuable metabolites with pharmacological activities, further underscoring their ecological and economic significance.

Despite their importance, the molecular mechanisms underlying pine seed germination remain poorly understood (Zaborowska et al., 2022). Factors such as genetic introgression, environmental stressors, and dormancy-breaking treatments significantly influence seed germination and seedling development (Nawrot-Chorabik et al., 2021). For example, genetic introgression between closely related pine species can impact seed morphology and germination fitness, highlighting the need for comprehensive studies on genetic and environmental interactions (Liu et al., 2021; Zhang et al., 2022). Moreover, dormancy-breaking treatments, including stratification, scarification, and the application of phytohormones, have been shown to enhance germination rates, but the underlying gene expression changes require further investigation.

This study aims to explore the gene expression and regulatory mechanisms involved in pine seed germination. By integrating transcriptome and metabolome analyses, this study identifies key genes and metabolic pathways that regulate germination processes in pine species; specifically, examines the role of differentially expressed genes (DEGs) and differentially accumulated metabolites (DAMs) in germination, as well as the impact of

environmental factors and genetic variations on these processes. Understanding these mechanisms will provide valuable insights for improving seed germination and seedling establishment, ultimately contributing to the sustainable management and conservation of pine forests.

2 Gene Expression Dynamics in Pine Seed Germination

2.1 Overview of key stages in pine seed germination

Pine seed germination is a complex process that involves multiple stages, each characterized by distinct physiological and molecular changes. The initial stage involves imbibition, where the seed absorbs water, leading to the reactivation of metabolic processes. This is followed by the activation of various signaling pathways and the mobilization of stored reserves to support the growth of the embryo (Stasolla et al., 2004; Ratnaparkhe et al., 2009). The final stage is marked by the emergence of the radicle, signifying the completion of germination and the beginning of seedling growth (Han et al., 2019; Xu et al., 2020).

2.2 Major genes involved in germination and their functions

Several key genes play crucial roles in the regulation of pine seed germination. For instance, MADS-box transcription factors such as MADS11 and DAL1 have been identified as important regulators of the vegetative-to-reproductive transition in pine, which is closely linked to the germination process. These genes are involved in the aging pathway and have distinct roles in flowering regulatory networks (Ma et al., 2021). Additionally, genes associated with cell wall degradation, biosynthesis, and remodeling are highly expressed during pollen germination, indicating their importance in the germination process (Salazar and Fernando, 2019). DNA methylation and gene expression variations in genes such as DNMTs and circadian clock genes also contribute to local adaptation and germination in Scots pine populations (Alakärppä et al., 2018).

2.3 Techniques used for gene expression analysis in pine seeds

Various techniques have been employed to analyze gene expression in pine seeds. Temporal dynamic transcriptome analysis is one such method, which involves sampling at different developmental stages to identify differentially expressed genes and regulatory modules (Ma et al., 2021). Yeast secretion trap (YST) coupled with computational prediction has been used to isolate cDNAs encoding secretory proteins associated with pollen germination, providing insights into the diversity and functions of these proteins (Salazar and Fernando, 2019). Comparative transcriptomic analysis has also been utilized to study transcriptional changes at different stages of seed development and germination, helping to identify key regulatory genes and pathways (Han et al., 2019; Xu et al., 2020). These techniques collectively enhance our understanding of the molecular mechanisms underlying pine seed germination.

3 Regulatory Mechanisms Influencing Gene Expression

3.1 Hormonal regulation during seed germination

Hormonal regulation plays a pivotal role in seed germination, with abscisic acid (ABA) and gibberellins (GA) being the primary hormones involved. ABA generally acts as an inhibitor of germination, maintaining seed dormancy, while GA promotes germination by breaking dormancy and stimulating growth processes (Bogamuwa and Jang, 2013). The balance between these hormones is crucial for the regulation of seed germination. For instance, the PIF1-miR408-PLANTACYANIN repression cascade in *Arabidopsis thaliana* demonstrates how light signals are translated into hormonal profiles that control germination by modulating ABA and GA levels (Jiang et al., 2021). Additionally, the expression of ABA receptors such as PYR1/PYL/RCAR is regulated by transcription factors like ABI5, which modulates ABA sensitivity during germination (Figure 1) (Zhao et al., 2020). In rice, similar hormonal pathways involving ABA and GA have been identified, highlighting the conserved nature of these regulatory mechanisms across different plant species (Gong et al., 2022).

The research of Zhao et al. (2020) illustrates the role of PYLs (abscisic acid receptors) and ABI5 in seed germination and their interaction under the influence of abscisic acid (ABA). During germination without ABA, SnRK2, a kinase, is inhibited by PP2C, preventing the phosphorylation of ABI5. As a result, ABI5 is degraded, and ABA-responsive genes are not activated, allowing germination to proceed. In the presence of ABA, ABA binds to PYLs, which inhibit PP2C, releasing SnRK2 from inhibition. Activated SnRK2 phosphorylates ABI5,

stabilizing it and preventing its degradation. The phosphorylated ABI5 then activates ABA-responsive genes, leading to the suppression of germination. This feedback loop highlights the critical role of ABA in regulating seed germination. PYLs maintain the stability of ABI5, which in turn regulates the expression of ABA-responsive genes and PYLs, ensuring a tightly controlled germination process in response to environmental cues. This mechanism ensures seeds only germinate under favorable conditions.

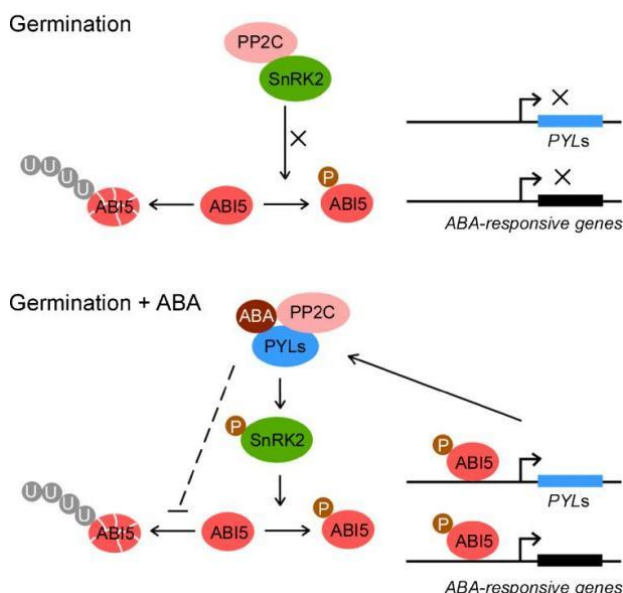


Figure 1 A working model for the cooperation of PYLs and ABI5 in seed germination (Adopted from Zhao et al., 2020)

Image caption: The abscisic acid (ABA) receptor PYLs maintain the stability of ABI5 to activate the expression of ABA-responsive genes and ABI5 plays a role in the feedback regulation of ABA signaling by maintaining the expression of PYLs (Adopted from Zhao et al., 2020)

3.2 Role of environmental factors in gene regulation

Environmental factors such as light and temperature significantly influence gene expression during seed germination. Light, for example, is a critical environmental cue that affects the degradation of repressors like PHYTOCHROME INTERACTING FACTOR 1 (PIF1), thereby promoting germination through hormonal changes (Jiang et al., 2021). High temperatures can delay germination by reactivating the expression of transcription factors like FUSCA3 (FUS3), which in turn modulates ABA levels to inhibit germination under suboptimal conditions (Chiu et al., 2012). Furthermore, the histone H3K27 demethylase REF6 has been identified as a positive regulator of light-initiated seed germination, indicating that epigenetic modifications also play a role in how environmental signals are integrated into the germination process (Wang et al., 2023).

3.3 Interaction between genetic and epigenetic factors

The transition from seed to seedling involves complex interactions between genetic and epigenetic factors. Key transcription factors such as LEAFY COTYLEDON1 and 2 (LEC1 and LEC2), ABSCISIC ACID INSENSITIVE3 (ABI3), and FUS3 are crucial for maintaining seed dormancy and need to be suppressed for germination to proceed. This suppression is mediated by chromatin remodeling complexes like POLYCOMB REPRESSIVE COMPLEX 1 and 2 (PRC1 and PRC2) (Smolikova et al., 2021). Additionally, histone modifications, such as the demethylation of H3K27 by REF6, play a significant role in activating gene expression necessary for germination (Wang et al., 2023). These epigenetic changes ensure that the genetic programs required for seed maturation are turned off, while those needed for seedling development are activated, thereby facilitating a smooth transition from dormancy to active growth.

In summary, the regulation of gene expression during pine seed germination is a multifaceted process involving hormonal signals, environmental cues, and intricate genetic and epigenetic interactions (Miransari and Smith, 2014). Understanding these mechanisms provides valuable insights into the fundamental processes governing plant development and adaptation.

4 Case Studies: Gene Expression Profiles in Pine Seeds

4.1 Detailed analysis of specific gene expression studies

Several studies have provided insights into the gene expression profiles in pine seeds, focusing on different aspects of seed development and germination. For instance, a comprehensive transcriptomic analysis of 14 different tissues from 1-month-old maritime pine (*Pinus pinaster*) seedlings (Figure 2) identified 39 841 new transcripts, with 2 376 ubiquitously expressed across all tissues, suggesting these as core 'housekeeping genes' in pine (Cañas et al., 2017). Another study on loblolly pine (*Pinus taeda*) identified genes associated with pollen germination, highlighting the roles of cell wall degradation, biosynthesis, and remodeling, as well as stress and defense responses (Salazar and Fernando, 2019). These findings underscore the complexity and specificity of gene expression during different stages of pine seed development and germination.

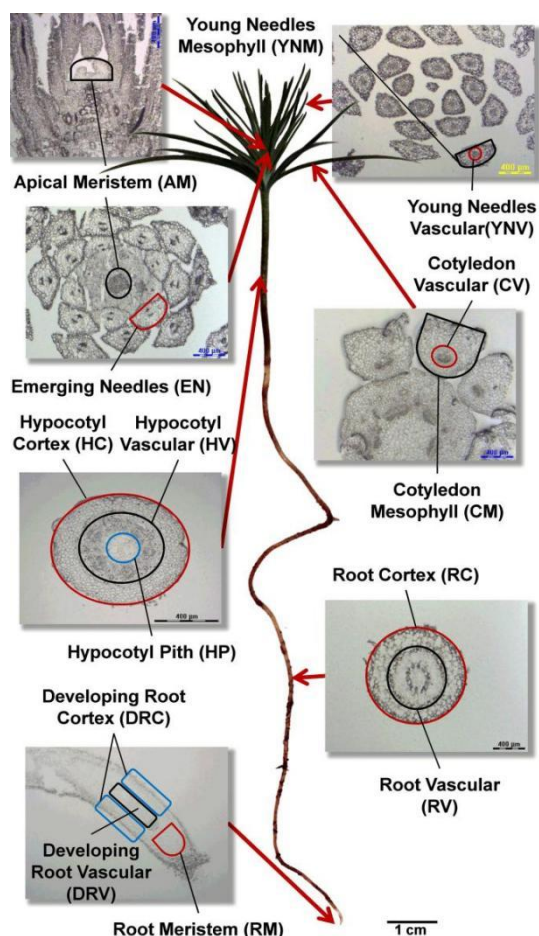


Figure 2 Diagram and histological sections showing the tissues isolated using laser capture microdissection (LCM) (Adopted from Cañas et al., 2017)

The research of Cañas et al. (2017) illustrates various tissues isolated from a plant using laser capture microdissection (LCM). The central image shows the plant's anatomy, with arrows pointing to specific sections where tissue samples were taken for histological analysis. Key areas sampled include the apical meristem (AM), young needles mesophyll (YNM) and vascular (YNV) tissues, cotyledon mesophyll (CM) and vascular (CV) tissues, emerging needles (EN), and various parts of the hypocotyl and root. Each inset image shows a magnified view of the isolated tissues, highlighting their cellular structures. For example, the apical meristem is essential for plant growth, containing undifferentiated cells that give rise to new tissues. The young needles' mesophyll and vascular tissues are crucial for photosynthesis and nutrient transport, respectively. Similarly, the root meristem (RM) is vital for root growth and development, while the root cortex (RC) and vascular (RV) tissues support nutrient absorption and transport. This detailed mapping and histological analysis enable a deeper understanding of plant development and cellular function, providing insights into the complex processes governing plant growth and adaptation.

4.2 Comparative gene expression analysis across different pine species

Comparative studies have also been conducted to understand gene expression differences across various pine species. For example, a study comparing radiata pine (*Pinus radiata*) and maritime pine (*Pinus pinaster*) under gravitropic stress identified differentially expressed genes involved in hormone regulation, the phenylpropanoid pathway, and signal transduction (Ramos et al., 2012). Additionally, research on Scots pine (*Pinus sylvestris*) populations from different climatic regions of Finland revealed variations in DNA methylation and gene expression, particularly in genes related to climate adaptation (Figure 3) (Alakärppä et al., 2018). These comparative analyses provide valuable insights into the genetic and epigenetic mechanisms underlying species-specific responses to environmental stresses.

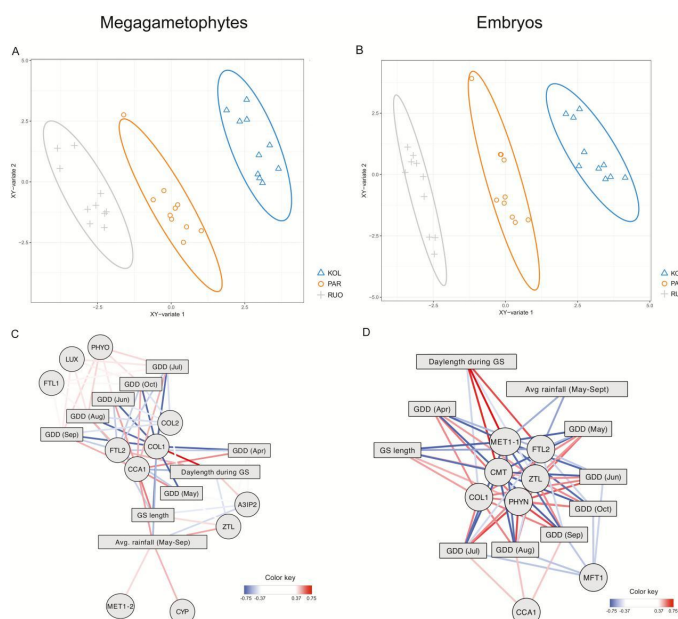


Figure 3 Sparse partial least squares (sPLS)-based classification of the different samples corresponding to the KOL, PAR, and RUO populations (Adopted from Alakärppä et al., 2018)

Image caption: Considering the changes in gene expression and environmental variables (averages from 1977 to 2007) in megagametophytes (A) and embryos (B). Components 1 and 2 corresponding to the XY space are represented. Ellipses represent a confidence interval of 0.9. (C, D) sPLS networks constructed using relative gene expressions and environmental variables in megagametophytes (C) and embryos (D). Color keys represent the value of the correlation between gene expression and environmental variables (Adopted from Alakärppä et al., 2018)

The research of Alakärppä et al. (2018) showcases a study on the correlation between gene expression and environmental variables in megagametophytes and embryos from different plant populations (KOL, PAR, and RUO). The sparse partial least squares (sPLS) plots (A and B) reveal distinct clustering patterns for each population, indicating significant differences in gene expression profiles. These differences are influenced by environmental factors, as shown by the clear separation in the XY-variate space. The sPLS network diagrams (C and D) further illustrate the relationships between gene expressions and environmental variables. Strong correlations are depicted with color-coded lines, where red and blue lines represent positive and negative correlations, respectively. Key genes like *CCA1*, *COL1*, and *FTL2* show significant interactions with environmental factors such as day length, growing season length, and average rainfall. These findings suggest that environmental conditions from 1977 to 2007 have influenced the genetic expression in these plant populations, providing insights into how plants adapt to changing climates and environmental conditions over time. This understanding is crucial for predicting plant responses to future environmental changes.

4.3 Implications of gene regulation studies for seed viability and vigor

Understanding gene regulation in pine seeds has significant implications for seed viability and vigor. For instance, the identification of differentially expressed genes in response to environmental stresses can inform breeding programs aimed at enhancing seed resilience. The study on Scots pine demonstrated that differential DNA

methylation and gene expression contribute to local adaptation, potentially enhancing the fitness of trees under changing climatic conditions (Alakärppä et al., 2018). Moreover, the identification of core 'housekeeping genes' in maritime pine provides a foundation for further research into the genetic basis of seed vigor and viability (Cañas et al., 2017). These insights are crucial for developing strategies to improve seed quality and ensure the sustainability of pine forests.

5 Technological Advances in Genetic Analysis

5.1 Innovations in transcriptomic and genomic technologies

Recent advancements in transcriptomic and genomic technologies have significantly enhanced our understanding of gene expression and regulatory mechanisms in pine seed germination (Cullum et al., 2011; Tarazona et al., 2011). One notable innovation is the use of laser capture microdissection followed by transcriptomic analysis, which allows for the precise isolation and examination of specific tissues and cells. This technique has been successfully applied to maritime pine (*Pinus pinaster*) seedlings, resulting in the identification of 39 841 new transcripts and the characterization of 2 376 ubiquitously expressed genes, which are considered core 'housekeeping genes' in pine (Cañas et al., 2017). Additionally, suppression subtractive hybridization (SSH) has been employed to identify differentially expressed genes in response to environmental stimuli, such as stem inclination in young pine seedlings. This method has facilitated the discovery of genes involved in hormone regulation, the phenylpropanoid pathway, and signal transduction (Ramos et al., 2012).

5.2 Application of next-generation sequencing (NGS) in seed biology

Next-generation sequencing (NGS) technologies have revolutionized seed biology by enabling comprehensive analyses of gene expression during critical developmental stages (Jain, 2012). For instance, global microarray analysis using NGS has revealed differential gene expression between quiescent and germinated maize embryo stages, highlighting the translational regulation of ribosomal protein mRNAs during germination (Jiménez-López et al., 2011). In *Arabidopsis*, pathway-based analysis of transcriptomic datasets has identified key regulatory actors and alternative metabolic routes involved in seed germination, demonstrating the genetic plasticity of this process (Ponnaiah et al., 2019). These studies underscore the power of NGS in uncovering the complex regulatory networks that govern seed germination and development.

5.3 Advances in bioinformatics for gene expression data analysis

The integration of bioinformatics tools has been pivotal in analyzing and interpreting the vast amounts of gene expression data generated by modern genomic technologies (Jia et al., 2017; Hwang et al., 2018). Bioinformatic analysis has been instrumental in identifying and categorizing differentially expressed genes, as seen in the study of stem inclination responses in pine seedlings, where 942 unigene elements were identified and classified into functional categories (Ramos et al., 2012). Furthermore, bioinformatics has facilitated the development of pathway-based analysis methods, which allow for the quantification of pathway deregulation and the identification of regulatory mechanisms in seed germination (Ponnaiah et al., 2019). These advances in bioinformatics are essential for translating raw sequencing data into meaningful biological insights, thereby advancing our understanding of gene expression and regulatory mechanisms in pine seed germination.

6 Impact of Research on Forestry Practices

6.1 Enhancing germination rates through genetic insights

Recent research has significantly advanced our understanding of the genetic mechanisms underlying seed germination, which has direct implications for improving germination rates in forestry practices. For instance, the study on scots pine (*Pinus sylvestris* L.) demonstrated that combining stratification/scarification with growth regulator treatments, such as gibberellic acid (GA3), indole-3-acetic acid (IAA), and 1-naphthaleneacetic acid (NAA), can significantly enhance germination rates and seedling development (Nawrot - Chorabik et al., 2021). Additionally, the gene expression landscape of pine seedling tissues has been mapped, revealing core 'housekeeping genes' and tissue-specific expression profiles that are crucial for understanding the physiological processes during germination (Cañas et al., 2017). These insights can be leveraged to develop targeted treatments and breeding programs aimed at enhancing seed germination rates in forestry.

6.2 Genetic engineering approaches to improve seed germination

Genetic engineering offers promising approaches to improve seed germination and overall forest productivity. Advances in tree genetic engineering have enabled the introduction of genes that enhance biotic and abiotic stress tolerance, improve wood properties, and promote root formation (Harfouche et al., 2011). For example, the overexpression of the *OsMYBAS1* transcription factor in rice has been shown to significantly increase germination rates under stress conditions, suggesting that similar genetic modifications could be applied to forest species to improve germination and seedling establishment (Wu et al., 2022). Furthermore, the regulation of seed germination through epigenetic mechanisms, such as DNA methylation and histone modification, has been explored in species like rice, where the *OsJMJ718* gene was found to positively regulate seed germination through the ABA and ethylene signal transduction pathways (Jia et al., 2023). These genetic engineering strategies can be adapted to forest species to enhance germination rates and resilience to environmental stresses (Park et al., 2010).

6.3 Contributions of genetic research to sustainable forestry

Genetic research contributes to sustainable forestry by providing insights into the adaptive mechanisms of tree species and enabling the development of more resilient forest populations (Manso et al., 2013). Studies on the natural variation of DNA methylation and gene expression in Scots pine populations have highlighted the role of epigenetic regulation in local adaptation to climate variables, suggesting that these mechanisms can enhance the fitness of trees under changing climatic conditions (Alakärppä et al., 2018). Additionally, the study on genetic introgression in pine species in Southeast Asia revealed that hybrids exhibit higher germination fitness under low temperatures, indicating that genetic diversity and gene flow can improve adaptability to environmental stresses (Zhang et al., 2022). These findings underscore the importance of incorporating genetic and epigenetic insights into forest management practices to promote sustainable forestry and ensure the long-term health and productivity of forest ecosystems.

7 Integrating Gene Expression Findings into Conservation Strategies

7.1 Applying gene expression insights to enhance pine seed conservation

Gene expression studies have provided significant insights into the molecular mechanisms underlying seed germination and dormancy, which are crucial for developing effective conservation strategies for pine species. For instance, the identification of differentially expressed genes in response to environmental cues and stressors can help in understanding how pine seeds adapt to various conditions. Studies on *Arabidopsis* and rice have shown that gene expression changes during seed germination involve complex regulatory networks, including transcriptional and post-transcriptional modifications, hormone signaling, and metabolic pathways (He et al., 2011; Wei et al., 2015; Ponnaiah et al., 2019). These findings can be applied to pine seeds to enhance their conservation by identifying key regulatory genes and pathways that can be targeted to improve seed viability and storability.

7.2 Development of conservation strategies based on genetic understanding

The development of conservation strategies for pine species can greatly benefit from a genetic understanding of seed germination and dormancy. For example, research on peanut and maize has highlighted the importance of hormone biosynthesis and signal transduction in seed germination, as well as the role of microRNAs in regulating seed storability (Xu et al., 2020; Song et al., 2022). By identifying similar genetic mechanisms in pine species, conservationists can develop strategies that promote seed germination and reduce dormancy, thereby enhancing the success of reforestation and restoration projects (Kovaleva et al., 2013). Additionally, the identification of genes involved in xylem development and wood formation in loblolly pine can inform breeding programs aimed at improving wood quality and resistance to environmental stressors (Palle et al., 2011).

7.3 Role of genetic research in formulating policies for pine species preservation

Genetic research plays a crucial role in formulating policies for the preservation of pine species. The identification of key regulatory genes and pathways involved in seed germination and stress responses can inform policy decisions related to seed storage, germplasm conservation, and reforestation efforts. For instance, the study of gene expression in response to stem inclination in young pine seedlings has provided insights into the molecular mechanisms of gravitropic responses, which can be used to develop strategies for improving seedling

establishment and growth in reforestation projects (Ramos et al., 2012). Furthermore, the creation of a gene expression atlas for different pine tissues can serve as a valuable resource for functional genomics research, aiding in the identification of genes that are critical for the survival and adaptation of pine species in changing environments (Cañas et al., 2017). By integrating gene expression findings into conservation strategies, we can enhance the effectiveness of efforts to preserve and restore pine species, ensuring their survival for future generations.

8 Future Directions in Pine Seed Research

8.1 Emerging trends and new technologies in seed biology

Recent advancements in genomics and transcriptomics have significantly enhanced our understanding of pine seed biology. Technologies such as laser capture microdissection combined with transcriptomic analysis have allowed for detailed mapping of gene expression in various pine tissues, providing valuable insights into the molecular and functional organization of conifer tissues (Cañas et al., 2017). Additionally, the use of suppression subtractive hybridization (SSH) and quantitative PCR has facilitated the identification of differentially expressed genes in response to environmental stimuli, such as stem inclination, which is crucial for understanding stress responses and wood formation in pines (Ramos et al., 2012). The integration of these technologies with bioinformatics tools has enabled the clustering of genes based on their expression profiles, further elucidating the complex regulatory networks involved in pine seed germination and development.

8.2 Potential collaborative research projects and funding opportunities

Collaborative research projects focusing on the genetic and epigenetic mechanisms underlying pine seed germination and adaptation to climate change are essential. For instance, studies on the natural variation in DNA methylation and gene expression in Scots pine populations have highlighted the role of epigenetic regulation in local adaptation, suggesting potential areas for collaborative research (Alakärppä et al., 2018). Additionally, projects investigating the genetic introgression and hybridization effects on seed morphology and germination fitness in closely related pine species could provide valuable insights into the evolutionary consequences of genetic introgression and its impact on population fitness (Zhang et al., 2022). Funding opportunities for such collaborative projects may be available through national and international research grants focused on forest conservation, climate change adaptation, and sustainable forestry practices.

8.3 Future research needs in pine seed germination and conservation

Future research should prioritize the identification and functional characterization of key regulatory genes involved in pine seed germination and early seedling development. Studies on the expression patterns of genes encoding secretory proteins associated with pollen germination have already provided a foundation for understanding the molecular players involved in this critical reproductive process (Salazar and Fernando, 2019). Additionally, research on the expression of embryogenesis-regulating genes in conifers, such as those in the Araucariaceae family, can offer insights into the conserved molecular mechanisms of embryo development across different taxa (Schlögl et al., 2012). Conservation efforts should also focus on understanding the genetic and epigenetic factors contributing to local adaptation and resilience to climate change, as demonstrated by studies on Scots pine populations (Alakärppä et al., 2018). By addressing these research needs, we can develop more effective strategies for the conservation and sustainable management of pine forests.

9 Concluding Remarks

The studies reviewed provide a comprehensive understanding of the gene expression and regulatory mechanisms involved in pine seed germination. Key findings include the identification of various transcription factors, metabolic pathways, and hormonal interactions that play crucial roles in this process. For instance, the study on *Arabidopsis* seed germination highlighted the importance of transcriptional and post-transcriptional modifications, identifying mechanisms such as RNA transport and vitamin B6 metabolism as critical for seed germination. Similarly, research on rice embryos revealed dynamic transcriptional changes and the significant roles of phytohormones like abscisic acid (ABA), gibberellin (GA), and brassinosteroid (BR) in germination. In pine pollen germination, genes associated with cell wall degradation, biosynthesis, and remodeling were identified,

emphasizing the complexity of the germination process. Additionally, the role of MYB-type transcription factors in regulating seed germination under stress conditions was demonstrated in rice, providing insights into the genetic control of this process.

This study significantly advances our understanding of the molecular mechanisms underlying pine seed germination, which is crucial for both basic biology and practical forestry applications. By identifying key genes and pathways involved in germination, this research provides a foundation for improving seed germination rates and seedling establishment in pine species. The findings on the distinct expression profiles of mature and germinated pollen, as well as the role of transcription factors in stress responses, offer valuable insights for breeding programs aimed at enhancing the resilience and productivity of pine forests. Furthermore, the identification of regulatory networks and metabolic pathways involved in seed germination can inform strategies for managing pine forests under changing environmental conditions.

Future research should focus on further elucidating the complex regulatory networks and interactions between different genes and pathways involved in pine seed germination. Studies employing advanced techniques such as CRISPR/Cas9 for gene editing and single-cell RNA sequencing could provide deeper insights into the specific roles of individual genes and their regulatory mechanisms. Additionally, exploring the effects of environmental factors such as temperature, moisture, and soil composition on gene expression during germination could help develop more effective forestry management practices.

Practical applications of this research include the development of genetically modified pine varieties with enhanced germination rates and stress tolerance. These varieties could be particularly valuable in reforestation efforts and in establishing pine plantations in areas with challenging environmental conditions. Moreover, the insights gained from this study could inform the development of seed treatments and other agronomic practices aimed at improving seedling establishment and growth in pine forests. By integrating these findings into forestry management and breeding programs, we can enhance the sustainability and productivity of pine forests, contributing to both ecological conservation and economic development.

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Conflict of Interest Disclosure

The authors affirm that this research was conducted without any commercial or financial relationships that could be construed as a potential conflict of interest.

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