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Prospects of Genetic Modification in Enhancing *Eucommia ulmoides* Production

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Abstract This study aims to explore the potential of genetic modification techniques in improving the production efficiency of *Eucommia ulmoides*. *Eucommia ulmoides* has garnered attention for its medicinal and industrial value, particularly in its role in rubber biosynthesis. Utilizing high-quality genome assembly and transcriptomic analysis, the research identified a series of key genes and metabolic pathways involved in the biosynthesis of rubber and chlorogenic acid, sex differentiation, and stress response. Notably, the study found that the methylerythritol phosphate (MEP) pathway is the primary route for isopentenyl diphosphate synthesis in *Eucommia ulmoides*, while the mevalonate (MVA) pathway serves this role in the Brazilian rubber tree (Hevea brasiliensis). Additionally, the *EuAP3* and *EuAG* genes are associated with sex differentiation, and the high expression of the ω-3 fatty acid desaturase-encoding gene *EU0103017* is related to the biosynthesis of α-linolenic acid. The study also revealed that the promoter activity of the small rubber particle protein (SRPP) gene is regulated by methyl jasmonate, gibberellins, and drought pathways, indicating these factors as potential targets for gene enhancement. Moreover, the *EuTIL1* gene was identified as a key gene for enhancing cold tolerance, providing a molecular basis for expanding the cultivation range of *Eucommia ulmoides*. The findings suggest that genetic modification techniques hold great potential in improving the yield and quality of *Eucommia ulmoides*. By modifying specific genes and metabolic pathways, it is expected to increase rubber yield, enhance stress resistance, and improve other economically important traits to meet the growing demand for this valuable resource.

Keywords *Eucommia ulmoides*; Genetic modification; Rubber biosynthesis; Chlorogenic acid; Sex differentiation; Stress tolerance; *SRPP* gene; *EuTIL1* gene

1 Introduction

Eucommia ulmoides, commonly known as the hardy rubber tree, is a species of significant botanical and economic importance. As the only species within the family Eucommiaceae, it has garnered attention for its ability to produce trans-polyisoprene, known as Eu-rubber (Wang et al., 2016; Wuyun et al., 2017). In traditional Chinese medicine, *Eucommia* is widely used for its health benefits, such as anti-inflammatory and antihypertensive effects (Li et al., 2020). Agriculturally, *Eucommia* holds substantial value in pharmacology, horticultural landscaping, and as a natural rubber resource, and it is extensively cultivated in China (Jin et al., 2020; Zhang et al., 2023b).

Despite its development potential, the cultivation of *Eucommia ulmoides* still faces numerous challenges. One of the primary issues affecting its commercial viability is the limited yield of *Eucommia* rubber (Zhao et al., 2023b). Environmental stressors such as drought and climate change complicate the cultivation process, further impacting the growth and rubber production of *Eucommia* (Jin et al., 2020; Zhao et al., 2023b). Additionally, with the increasing market demand for natural rubber and medicinal products derived from *Eucommia*, there is an urgent need to improve cultivation techniques to meet these demands (Wang et al., 2016; Wuyun et al., 2017).

Genetic modification offers potential solutions to the challenges faced in *Eucommia ulmoides* cultivation. Advances in genomic technologies, such as high-quality genome assembly and the identification of key genes related to rubber biosynthesis, have laid the foundation for genetic engineering research (Wuyun et al., 2017; Li et al., 2020). By regulating genes associated with stress resistance, growth traits, and rubber biosynthesis, the yield and adaptability of *Eucommia ulmoides* can be improved (Jin et al., 2020; Zhang et al., 2023a; 2023b). For instance, the identification of sex-related molecular markers contributes to more efficient breeding practices, while the regulation of transcription factors like NAC and MYB helps optimize rubber production (Wang et al., 2020; Hu et al., 2023; Zhang et al., 2023b).

This study aims to explore the prospects of genetic modification in enhancing the production of *Eucommia ulmoides*. The research objectives include analyzing existing genomic data, identifying key genes related to rubber biosynthesis and stress resistance, investigating the potential of genetic engineering in improving yield and environmental adaptability, and assessing the impact of specific genetic modifications on overall growth and rubber production in *Eucommia ulmoides*. Additionally, the study seeks to provide a comprehensive overview of the molecular mechanisms underlying genetic modification, offering insights for future research and practical applications. By achieving these goals, this study aims to contribute to the sustainable and efficient cultivation of *Eucommia ulmoides* to meet the demands of both agricultural and industrial sectors.

2 Current State of *Eucommia ulmoides* Production

2.1 Traditional cultivation methods

Eucommia ulmoides, also known as the hardy rubber tree, is widely cultivated in China for its medicinal and industrial applications. Traditional cultivation methods of *Eucommia* primarily rely on seed and cutting propagation. However, these methods have certain limitations. The seeds of *Eucommia* exhibit dormancy, which significantly limits their germination rate, complicating the propagation process. Research has shown that the seed coat of *Eucommia* restricts water absorption, making it necessary to break dormancy using treatments such as scarification, stratification, and the application of plant hormones like gibberellin (GA3) (Deng et al., 2021).

Additionally, traditional cultivation methods are significantly affected by environmental factors. Studies have shown that factors such as the average annual temperature and duration of sunlight in the planting area play a key role in the growth and economic traits of *Eucommia ulmoides*. Therefore, selecting suitable genotypes and planting regions is crucial for improving the performance and stability of *Eucommia* traits (Deng et al., 2022). Despite taking corresponding measures, the low yield of *Eucommia* rubber remains a major challenge, limiting its application as a source of natural rubber (Zhao et al., 2023b).

2.2 Market demand and applications

Eucommia has broad application prospects in industrial, pharmaceutical, and ecological fields, attracting global attention. Its medicinal value has been fully utilized in traditional Chinese medicine, especially in the treatment of various diseases. The bark and leaves of *Eucommia* are rich in bioactive compounds, such as chlorogenic acid, which have significant pharmacological effects (Deng et al., 2022).

2.2.1 Industrial and ecological fields

In the industrial field, *Eucommia* shows significant advantages as a potential source of natural rubber. The small rubber particle protein (SRPP) gene plays a key role in the biosynthesis of *Eucommia* rubber, and its expression is regulated by endogenous hormones and environmental stresses, such as drought (Zhao et al., 2023b). This makes *Eucommia* a strong candidate for sustainable rubber production in regions where traditional rubber trees cannot grow.

Rui-fang Yan and her team at the Institute of Chemistry, Chinese Academy of Sciences, were the first to successfully vulcanize natural *Eucommia* rubber into high-elasticity rubber internationally. Currently, *Eucommia*-derived materials include orthopedic materials, new shape memory materials, corrosion-resistant materials, rubber materials, chemical functional materials, daily functional materials, aerospace-specific materials, and high-impact performance materials, among others.

Ecologically, *Eucommia* plays an active role in environmental greening, wind protection, and soil stabilization, making it an important species in environmental protection and land management (Jin et al., 2020). Its high genetic diversity and outcrossing reproduction system further enhance its adaptability and stress resistance, ensuring its long-term sustainability and practicality (Zhang et al., 2013).

2.2.2 Medicinal field

The dried bark of *Eucommia* has been used as a top-grade herbal medicine for thousands of years. It is described in traditional Chinese medicine as “spicy in flavor and neutral in nature” and is often combined with other medicines in clinical practice. It has the effects of “alleviating pain in the waist and knees, replenishing vital energy, strengthening muscles and bones, stabilizing pregnancy, and promoting longevity with long-term use.” *Eucommia* has special efficacy in treating hypertension, particularly its unique ability to “regulate blood pressure in both directions,” a feat no chemical drug can match. Recent studies also suggest that *Eucommia* can lower cholesterol levels, prevent arteriosclerosis, and has marked anti-aging, anti-cancer, and body-function-enhancing effects.

Research results have shown that chlorogenic acid exhibits strong scavenging effects on DPPH, ABTS, and OH free radicals (Yu et al., 2022). In vitro experiments have demonstrated that *Eucommia* leaf polysaccharides and polyphenols also have antioxidant activity (Wu, 2022). Research has also shown that the flavonoid components in 70% ethanol extract of *Eucommia* leaves have anti-neuroinflammatory activity in lipopolysaccharide-induced BV-2 mouse microglial cells and neuroprotective effects against H2O2-induced PC12 cells (Hu et al., 2023). Additionally, another study suggests that a triterpene compound from *Eucommia* leaves can reduce neuroinflammatory responses in microglial cells and improve neuroinflammation. Chlorogenic acid and geniposidic acid in *Eucommia* leaf extract also have excellent lipid-lowering effects, reducing white fat content in mouse tissues and leading to weight reduction (Hirata et al., 2011).

2.2.3 Food application field

As a food and medicine homologous herb, *Eucommia* leaves are safe and have been widely applied in food. *Eucommia* leaf tea, made by frying fresh leaves, has certain antioxidant activity (Xu et al., 2021). *Eucommia* leaf brick tea is prepared by fresh leaves following a process consistent with the production of brick tea, including pressing, fermentation, and aging. The types and content of flavonoid and phenolic acid components in the tea are associated with microorganisms during the fermentation process (Zeng et al., 2021). Adding ultra-fine *Eucommia* leaf powder in steamed buns improves hardness, stickiness, and chewiness compared to regular buns and also offers certain nutritional benefits (Li et al., 2022).

2.2.4 Feed additive applications

Research shows that adding 4% to 6% *Eucommia* leaf powder to Yellow River carp feed can increase amino acid content in the fish meat, improving taste and nutritional value (Li et al., 2021). Studies also found that adding 4% *Eucommia* leaf powder to Green Shell chicken feed improves meat quality (Mao et al., 2021). Research by Li et al. (2022) also indicates that adding *Eucommia* leaf fermentation products and extracts to chicken feed promotes chick development, improves meat quality, and enhances immunity. In Nagano Prefecture, Japan, a local company added 2% to 3% *Eucommia* leaf powder to regular feed to produce high-quality eel feed. The fish fed with this feed had tender, delicious meat, distinct from those fed with regular feed. Using *Eucommia* leaves as a feed additive can greatly improve the economic benefits of the aquaculture industry. In summary, as market demand for *Eucommia* increases and its application fields continue to expand, improving cultivation techniques and conducting genetic modification to enhance yields to meet global demand has become an urgent priority.

3 Genetic Modification Techniques Relevant to *Eucommia ulmoides*

3.1 CRISPR-Cas9 and other gene editing tools

The existing genome editing systems primarily include the Zinc Finger Nuclease (ZFNs) system, the Transcription Activator-Like Effector Nuclease (TALENs) system, and the CRISPR/Cas9 (Clustered Regularly Interspaced Short Palindromic Repeat/CRISPR-associated protein 9) system. CRISPR-Cas9 technology, due to its simplicity, efficiency, and specificity, has become a revolutionary tool in the field of genetic improvement. This technology allows precise genome modification by targeting specific DNA sequences and introducing cuts. Through repair mechanisms, desired genetic changes can be introduced. CRISPR-Cas9, with its high precision in promoting targeted gene editing, has been widely applied in various fields, including plant biotechnology (Li et al., 2020; Sharma et al., 2020; Guo et al., 2023).

In addition to CRISPR-Cas9, other gene-editing tools such as zinc finger nucleases (ZFNs) and transcription activator-like effector nucleases (TALENs) have also been used for genetic improvement. Although these tools are effective, they are generally more complex and less efficient compared to CRISPR-Cas9. The development of these technologies has significantly advanced our ability to manipulate plant genomes, including the *Eucommia ulmoides* genome, thereby promoting the improvement of desirable traits such as growth rate, stress resistance, and production of metabolites (Li et al., 2020; Sharma et al., 2020). The establishment of the CRISPR/Cas9 gene-editing system in *Eucommia ulmoides* successfully achieved gene editing in *Eucommia* cells for the first time. This research provides a new approach for gene function identification and genetic improvement in *Eucommia ulmoides*.

3.2 Potential target genes for modification

Identifying and targeting specific genes associated with key traits is crucial for the successful genetic improvement of *Eucommia ulmoides* (Figure 1). Genes involved in growth regulation, stress resistance, and metabolite biosynthesis are priority candidates for modification. For instance, genes regulating hormone pathways, such as those involved in gibberellin and auxin signaling, can be targeted to enhance growth rate. Similarly, stress resistance genes, including those responsible for synthesizing heat shock proteins or antioxidant enzymes, can be modified to improve the plant’s adaptability to environmental stresses (Han et al., 2020; Li et al., 2020a; Sharma et al., 2020).

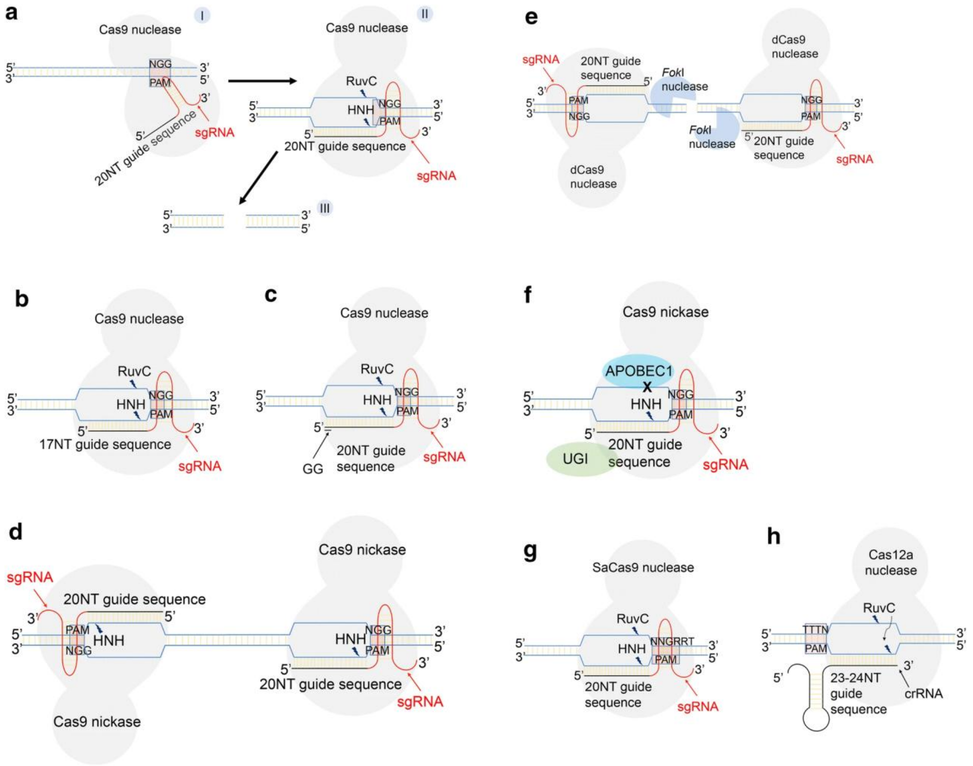

Figure 1 Stagewise schematic representations of target site recognition in CRISPR/Cas9-mediated genome editing with modifications to the sgRNA and Cas9 endonuclease to reduce OTEs (Adopted from Han et al., 2020)

Image caption: a The Cas9 endonuclease first scans the genomic DNA and binds to canonical PAM sequences (I). This induces a structural change in the sgRNA that allows the guide sequence to search and hybridise to complementary target sites upstream of the PAM (II). sgRNA-DNA hybridisation activates the Cas9 nuclease domains which then cleaves both strands of DNA (III). b sgRNA can be truncated at the 5′-end by 2–3 nucleotides or c modified at the 5′-end to contain 2 guanine nucleotides to improve the specificity of the guide sequence. d Cas9 nickase with only 1 active catalytic domain can be paired and e catalytically deactivated Cas9 fused to FokI nuclease that requires dimerisation for nuclease activity can be used to minimise off-target indels (insertion/deletion). f Base editors that convert a single cytosine base to thymine without requiring DSBs are less promiscuous at off-target sites. g Cas9 orthologs from other bacteria such as SaCas9 and h other Cas nucleases such as Cas12a that recognises alternative PAMs can be used to target novel DNA sequences and improve specificity (Adopted from Han et al., 2020)

Moreover, genes involved in the biosynthesis of valuable metabolites, such as those encoding enzymes in the terpenoid and flavonoid pathways, can be targeted to increase the production of these compounds. By leveraging the precision of CRISPR-Cas9 and other gene editing tools, it is possible to introduce specific mutations or insertions that enhance the expression or activity of these key genes, thereby improving the overall productivity and quality of *Eucommia ulmoides* (Park et al., 2022; Guo et al., 2023)

3.3 De novo genome assembly contributions

The application of de novo genome assembly techniques has greatly advanced the development of genetic improvement technologies for *Eucommia ulmoides*. High-quality genome assemblies provide a comprehensive reference for identifying target genes and their regulatory networks. In recent years, research has shifted towards generating detailed genomic maps of *Eucommia ulmoides*, including annotations of genes, regulatory elements, and structural variations.

These genomic resources are invaluable for designing precise gene editing strategies. For example, the identification of conserved sequences and regulatory motifs can guide the selection of target sites for CRISPR-Cas9 editing. Additionally, genome assemblies facilitate the study of gene function and interaction, enabling researchers to predict the outcomes of genetic modifications more accurately. The integration of genomic data with gene editing technologies holds great promise for optimizing the genetic improvement of *Eucommia ulmoides* (Han et al., 2020; Li et al., 2020a; Sharma et al., 2020).

In conclusion, the application of advanced gene-editing tools such as CRISPR-Cas9, combined with the identification of key target genes and the integration of genomic data, offers immense potential for improving the yield and quality of *Eucommia ulmoides*. Continued research and development in these areas will pave the way for more efficient and precise genetic improvements, ultimately benefiting the cultivation and utilization of this valuable plant species.

4 Enhancing Productivity through Genetic Modification

4.1 Improving growth rates: strategies for modifying genes related to faster growth

The improvement of *Eucommia ulmoides* growth rate through genetic modification techniques involves the targeted regulation of specific genes responsible for plant development and growth. One promising approach is the overexpression of aquaporin genes such as *EuPIP1;1*. Studies have shown that constitutive overexpression of *EuPIP1;1* in *Arabidopsis thaliana* promotes leaf growth, accelerates bolting and flowering, and up-regulates genes related to growth and flowering, such as *AtPIF4* and *AtTCP14* (Chen et al., 2022a). Additionally, gene pyramiding, which involves the simultaneous introduction of multiple beneficial genes, has been demonstrated to significantly enhance plant growth. For instance, the combined overexpression of *AVP1*, *OsSIZ1*, and *Fld* in creeping bentgrass resulted in superior growth performance under both normal and stress conditions (Zhao et al., 2023a).

4.2 Enhancing stress resistance: genetic pathways to enhance drought, pest, and disease resistance

Enhancing the stress resistance of *Eucommia ulmoides* can be achieved by modifying genetic pathways that confer tolerance to drought, pests, and diseases. The overexpression of aquaporin genes such as EuPIP1;1 and EuPIP1;2 has been shown to improve drought and salt stress tolerance in transgenic plants. These genes help maintain ion homeostasis, reduce membrane damage, and improve osmotic adjustment under stress conditions (Figure 2) (Chen et al., 2022a; 2022b). Additionally, the NAC transcription factor family plays a crucial role in plant adaptation to environmental challenges. The identification and characterization of 69 *NAC* genes in *Eucommia ulmoides* suggest that these genes can be targeted to enhance drought stress tolerance (Wang et al., 2023b). Furthermore, the use of plant growth-promoting rhizobacteria (PGPR) can activate plant adaptive defense systems, improving drought tolerance and overall growth (Gowtham et al., 2022).

4.3 Increasing metabolite production: genetic modifications to improve production of valuable compounds such as gutta-percha

Increasing the production of valuable compounds like gutta-percha in *Eucommia ulmoides* can be achieved through genetic modifications that enhance the biosynthesis pathways of these metabolites. The small rubber particle protein (SRPP) gene is a key player in the biosynthesis of *Eucommia ulmoides* gum. Studies have shown that the promoter activity of the EuSRPP gene is regulated by methyl jasmonate (MeJA), gibberellin (GA3), and drought pathways, suggesting that manipulating these pathways can increase gum content (Zhao et al., 2023b). Moreover, the overexpression of genes involved in the biosynthetic pathways of osmoprotectants, such as betaine aldehyde dehydrogenase (BADH) and flavodoxin (Fld), can enhance stress resistance and productivity, thereby indirectly promoting the production of valuable metabolites (Niazian et al., 2021). By utilizing the aforementioned gene improvement strategies, the productivity, stress resistance, and metabolite yield of *Eucommia ulmoides* can be significantly enhanced, thus increasing its commercial feasibility and application value.

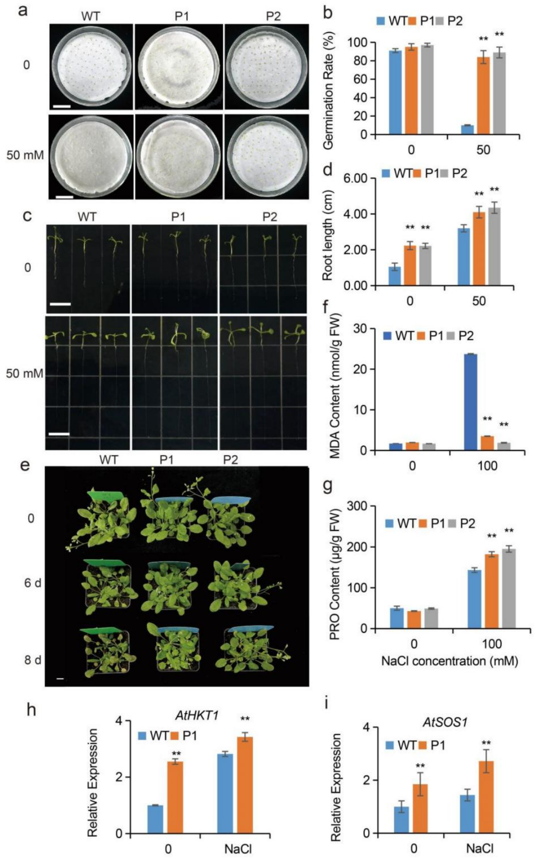

Figure 2 Effect of NaCl treatment on the germination and root growth of WT and transgenic lines (P1 and P2) (Adopted from Chen et al., 2022a)

Image caption: (a) The germination rate of the seeds from WT and transgenic lines (P1 and P2) under 0 or 50 mM NaCl treatment. Bar = 1 cm. (b) The germination rate. (c) Phenotypes of seedlings from WT and transgenic lines (P1 and P2) treated with 0 or 50 mM NaCl treatment. Bar = 1 cm. (d) The length of root. (e) Phenotypes of seedlings from WT and transgenic lines (P1 and P2) treated with 0 or 100 mM NaCl treatment at 6 d and 8 d. Bar = 1 cm. (f) The MDA content. (g) The PRO content. Expression level of AtHKT1 (h) and AtSOS1 (i) in WT and P1 treated with NaCl. (** $p < 0.01$, one way ANOVA) (Adopted from Chen et al., 2022a)

5 Case Studies of Genetic Modification in Related Species

5.1 Rubber tree (*Hevea brasiliensis*)

The rubber tree (*Hevea brasiliensis*) is an important source of natural rubber and has become a central subject of numerous genetic studies aimed at improving its yield and growth characteristics. With the development of genomic tools, such as genome-wide association studies (GWAS) and marker-assisted selection (MAS), deeper insights into the genetic basis of these traits have been provided. For instance, a study integrating GWAS and RNA-sequencing (RNA-Seq) methodologies identified key quantitative trait loci (QTLs) and gene networks associated with rubber tree growth under water stress conditions. This multiomics approach has enhanced our understanding of the molecular mechanisms underlying growth and has potential applications in breeding programs to improve rubber yield and stress resilience (Francisco et al., 2021).

Furthermore, the high-quality chromosome-level genome sequence analysis of wild rubber tree species revealed significant genetic markers associated with latex yield. The study identified 155 trait marker associations and 326 candidate genes, including genes related to sugar transport, metabolism, and ethylene biosynthesis. These findings provide valuable resources for genome-assisted breeding, aimed at improving rubber yield through targeted gene enhancement (Figure 3) (Cheng et al., 2023).

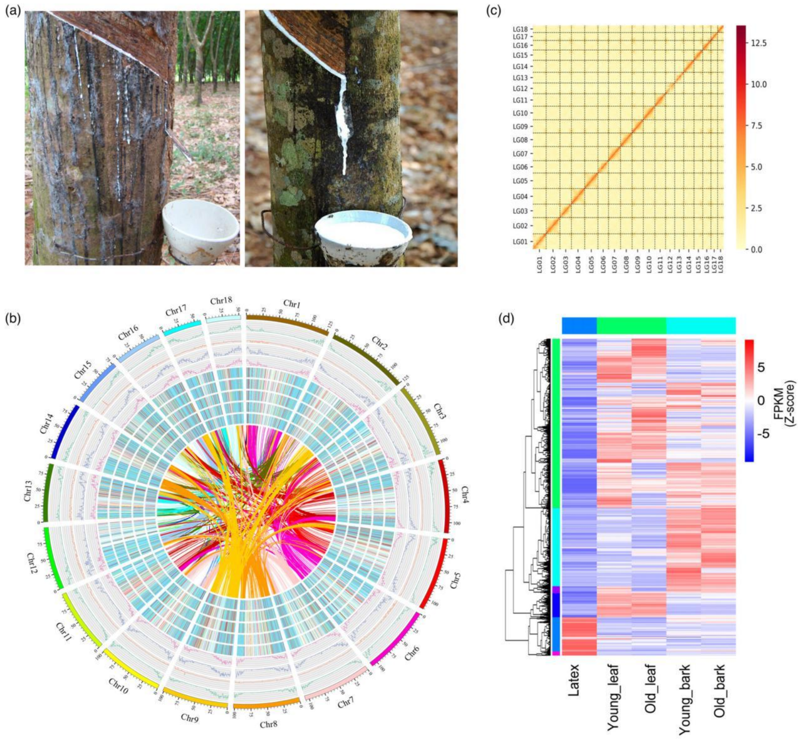

Figure 3 Assembly of rubber tree genome (Adopted from Cheng et al., 2023)

Image caption: (a) Rubber tree yield collected by tapping. Left, low-yield wild accession MT/VB/25A 57/8; Right, high-yield Wickham clone Reyan7-33-97. (b) Circos plot of repeat sequences and expression patterns in the rubber genome. The curves connect the colinear blocks between different chromosomes. The heatmap illustrates the gene expression levels (Log2FPKM) in young leaves, young bark, old leaves, old bark and latex of rubber (from inside to outside). The red and green colours indicate high and low expression levels, respectively. The density of line charts represents gene density, transposable element (TE), short tandem repeat (STR) and simple sequence repeat (SSR) across 18 chromosomes in the rubber tree (from inside to outside). The colour density indicates a high density of interactions. Distinct chromosomes are separated by dotted lines. (c) Genome-wide all-by-all highest-throughput chromosome conformation capture (Hi-C) interaction heatmap for the 18 linkage groups in *Hevea brasiliensis*. Heat map showing the density of the Hi-C interactions between chromosomes. (d) Heatmap of the DEGs (differentially expressed genes) expression levels in leaf, bark tissues and latex. In total, 4192 DEGs were identified to be differentially expressed (Adopted from Cheng et al., 2023)

5.2 Poplar species

The species of poplar have been extensively studied due to their rapid growth and wide adaptability, making them ideal candidates for genetic improvement to enhance biomass production and stress resistance. Although poplar research is not directly related to rubber production, the genetic findings from these studies can be applied to other tree species, including *Eucommia ulmoides*.

Poplar research primarily focuses on unraveling the genetic basis of traits such as growth rate, wood quality, and stress resistance. Techniques such as genome-wide association studies (GWAS) and transcriptome analysis have been used to identify key genes and regulatory networks involved in these processes. For example, studies have highlighted the roles of specific transcription factors and hormone signaling pathways in regulating poplar growth and development. These findings can provide a theoretical basis for genetic improvement strategies in *Eucommia* *ulmoides* to enhance its productivity and stress resistance (Li et al., 2020b). By leveraging the genetic knowledge and methodologies developed in poplar research, scientists can better understand and manipulate the genetic pathways in *Eucommia ulmoides*, ultimately improving its industrial and medicinal applications.

6 Potential Benefits and Risks

6.1 Economic impact

The genetic modification of *Eucommia ulmoides* holds significant promise for enhancing its productivity, which could have a profound economic impact. By improving growth traits such as tree height, ground diameter, and crown diameter through the identification and manipulation of specific quantitative trait loci (QTLs), the overall yield of *E. ulmoides* can be substantially increased (Jin et al., 2020). This increase in productivity could lead to a more robust supply of *E. ulmoides*, thereby stabilizing market prices and making the products derived from this tree more accessible. Additionally, the enhanced growth rates could reduce the time required for trees to reach maturity, further accelerating economic returns for farmers and stakeholders involved in the cultivation and processing of *E. ulmoides*.

6.2 Environmental benefits

Genetically modified *E. ulmoides* can play a crucial role in environmental conservation efforts, particularly in reforestation and sustainable agriculture. The species is already valued for its ecological benefits, such as wind sheltering and sand fixation (Jin et al., 2020). By enhancing its growth traits through genetic modification, *E. ulmoides* can be more effectively used in reforestation projects, contributing to the restoration of degraded landscapes and the stabilization of soil. Furthermore, the increased resilience and growth rates of genetically modified *E. ulmoides* can support sustainable agricultural practices by providing a reliable source of raw materials without the need for extensive land use, thereby preserving natural habitats and biodiversity.

6.3 Ethical and ecological concerns

While the potential benefits of genetically modifying *E. ulmoides* are substantial, there are also significant ethical and ecological concerns that must be addressed. One major concern is the potential for genetically modified trees to crossbreed with wild populations, leading to unintended genetic consequences and the possible disruption of local ecosystems. The introduction of genetically modified traits could also affect the phenotypic variation within natural populations, potentially reducing genetic diversity and resilience to environmental changes (Wang et al., 2023a). Additionally, there are ethical considerations regarding the manipulation of a species that holds cultural and ecological significance. It is crucial to conduct thorough risk assessments and implement stringent regulatory measures to mitigate these risks and ensure that the benefits of genetic modification do not come at the expense of ecological integrity and ethical standards.

7 Future Research Directions

7.1 Innovative genetic techniques

The future of *Eucommia ulmoides* production lies in the innovative application of advanced genetic techniques. Recent advancements in gene editing technologies, such as CRISPR/Cas9, offer promising avenues for enhancing the genetic traits of *E. ulmoides*. These techniques can be employed to target specific genes responsible for desirable traits such as growth rate, disease resistance, and secondary metabolite production. For instance, the high-quality chromosome-level genome assembly of *E. ulmoides* provides a comprehensive genetic blueprint that can be utilized for precise gene editing to improve traits like α-linolenic acid biosynthesis and sex differentiation (Li et al., 2020b; Du et al., 2023). Additionally, the identification of quantitative trait loci (QTLs) associated with growth traits offers a solid foundation for targeted genetic modifications aimed at enhancing growth performance and biomass yield (Jin et al., 2020; Liu et al., 2022). The integration of single-nucleotide polymorphism (SNP) markers in genetic mapping further refines the precision of these modifications, ensuring that the desired traits are effectively expressed (Liu et al., 2022).

7.2 Integration with traditional breeding programs

While genetic modification holds great potential, its integration with traditional breeding programs can lead to the development of more robust and resilient *E. ulmoides* plants. Traditional breeding methods, such as selective breeding and hybridization, have long been used to enhance plant traits. By combining these methods with modern genetic techniques, it is possible to achieve synergistic effects that maximize the benefits of both approaches. For example, the use of triploid breeding has shown significant improvements in vegetative growth and secondary metabolite content in *E. ulmoides*, which can be further enhanced through genetic modification (Li et al., 2016; 2019). Moreover, understanding the genotype × environment interactions and their impact on phenotypic traits can guide the selection of optimal genotypes for specific cultivation areas, thereby improving overall plant performance (Deng et al., 2022). The development of sex-specific markers also facilitates early identification of plant sex, which is crucial for breeding programs aimed at producing high-yielding female plants (Wang et al., 2011). By integrating these advanced genetic techniques with traditional breeding strategies, it is possible to create *E. ulmoides* plants that are not only high-yielding but also resilient to environmental stresses and diseases, thereby ensuring sustainable production and meeting the increasing demand for this valuable species.

8 Concluding Remarks

The potential of genetic modification in enhancing *Eucommia ulmoides* production is vast and promising. Recent advancements in genomic technologies have provided high-quality genome assemblies for both male and female *E. ulmoides*, revealing critical insights into the species' genetic makeup and evolutionary history. These genomic resources have identified key genes involved in rubber biosynthesis, sex differentiation, and other vital metabolic pathways, which can be targeted for genetic engineering to improve yield and quality. Additionally, the development of molecular markers, such as the male-specific AFLP and SCAR markers, facilitates early sex identification, significantly enhancing breeding efficiency. Quantitative trait loci (QTL) analysis has further elucidated the genetic basis of growth traits, providing a foundation for the genetic improvement of *E. ulmoides*. Collectively, these genetic tools and insights could revolutionize the production of *E. ulmoides* by enabling precise and efficient breeding strategies, ultimately leading to higher yields and better quality of rubber and medicinal compounds.

To fully realize the potential of genetic improvement in *Eucommia ulmoides* production, further research and collaboration in this field must be encouraged. Researchers should focus on functional genomics studies to validate the identified genes and their roles in rubber and chlorogenic acid synthesis. Collaboration between geneticists, biotechnologists, and industry stakeholders is crucial for developing and implementing genetic engineering technologies that enhance beneficial traits in *Eucommia ulmoides*. Additionally, expanding genetic linkage maps and QTL analysis will provide deeper insights into the genetic architecture of growth and other economically important traits. By promoting multidisciplinary collaboration and leveraging advanced genomic technologies, we can accelerate the genetic improvement of *Eucommia ulmoides*, ensuring its sustainable production and utilization in various industrial and medical applications.

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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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