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Doubled Haploids: A Revolutionary Technology in Maize Breeding

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Abstract

As the global population continues to rise and the demand for staple crops like maize (Zea mays L.) increases, innovative breeding strategies are essential for ensuring food security. Doubled Haploid (DH) technology represents a breakthrough in maize breeding, allowing the rapid development of pure lines for hybrid breeding programs. Unlike traditional methods that require multiple generations of self-pollination to achieve homozygosity, DH technology significantly reduces this timeframe, producing completely homozygous lines within two generations.

Introduction

Maize (Zea mays L.) is a staple crop, serving as a vital source of food, feed and industrial raw material. With a growing global population and challenges posed by climate change, innovative breeding strategies are essential to enhance productivity and resilience of maize. Doubled Haploid (DH) technology has emerged as a key tool in maize breeding.

DH technology facilitates the rapid development of fully homozygous inbred lines, which are essential for hybrid breeding programs. Traditional methods often require several generations of self-pollination to achieve homozygosity, a time-consuming process (Odiyo et al., 2014). In contrast, DH technology allows for the direct production of homozygous lines in just two generations, enhancing efficiency of selection for desirable traits and accelerating genetic improvement (Prigge et al., 2012; Prasanna, 2012). The history of DH technology in maize began with the discovery of the first spontaneous haploid in 1929. Over the decades, advancements have improved the haploid induction rates and led to the development of key inducer lines, such as Stock 6. Today, DH technology is integrated into various stages of maize breeding, enhancing genomic selection and facilitating the release of high-performing hybrids.

Importance of Doubled Haploid Technology

DH technology plays a critical role in maize breeding, a crop that is a global staple and a key component of food security. DH technology facilitates faster genetic gains, enabling the development of superior hybrids that are better adapted to changing environmental conditions, resistant to diseases, and capable of achieving higher yields. Furthermore, DH technology aligns with modern breeding approaches, including genomic selection and gene editing, making it a powerful tool for improving complex traits such as drought tolerance and nutrient use efficiency.

Key Steps in DH Production

The process has four significant steps (Chaikam et al., 2019)

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1. Haploid Induction: Haploid induction in maize can occur through various methods, including spontaneous induction, in vivo induction, and genetic engineering approaches.

1. **Spontaneous Haploid Induction:** This process is linked to meiosis induction and genetic factors like ggi1 (chromosome 1) and ig (chromosome 3). Delayed pollination enhances parthenogenesis, but chromosome doubling remains inefficient, necessitating both in vitro and in vivo methods.

2. **In Vivo Haploid Induction:** This involves crossing adapted lines with haploid inducers, significantly improving haploid induction rates (HIR).
   - Paternal Haploid Induction: Relies on the ig1 mutant gene (chromosome 3), promoting anucleate egg cells that develop into paternal haploids.
   - Maternal Haploid Induction: The most widely used method, where haploid-inducing male lines pollinate donor females, producing maternal haploids.

3. **Genetic Engineering and Gene Editing:**
   - Centromere Engineering: Modifying the CENH3 gene induces genome elimination, but its efficiency varies across genetic backgrounds.
   - CRISPR/Cas9 Gene Editing: Targeting genes like MATRILINEAL (MTL) (ZmPLA1/NLD) and ZmDMP significantly increase haploid induction, accelerating DH line development.

2. Identification of Haploids

Haploid seeds are identified using phenotypic traits such as kernel morphology, biomarkers (R1-navajo), genetic markers (SSR & SNP), or advanced techniques like flow cytometry for more precise identification (Maqbool et al., 2020 & Dermail et al., 2024).

3. Chromosome Doubling

Haploid plants are sterile due to their incomplete chromosome set. Chromosome doubling is necessary to restore fertility and can be achieved through chemical treatments such as colchicine or newer alternatives like nitrous oxide, as well as other antimicrotubular agents like trifluralin and oryzalin.

4. Self-Pollination

Once chromosome doubling is successfully achieved, the resulting plants are self-pollinated to produce completely homozygous seeds for use in hybrid production or further breeding programs

(Indu et al.2023)

Advantages of DH Technology

1. Accelerated Breeding Cycles: Rapidly generating homozygous lines significantly reduces the time required to develop new maize varieties.

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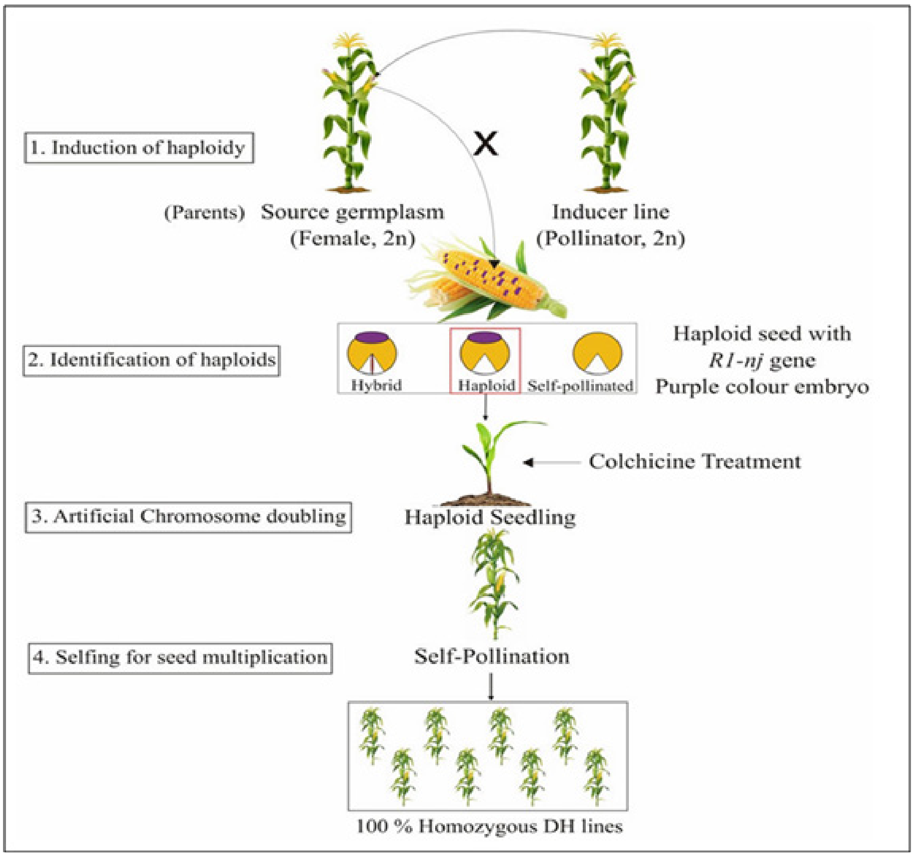

2. Enhanced Genetic Gains: The technology improves the precision of selecting desirable traits, increasing breeding efficiency.

3. Improved Hybrid Development: DH technology strengthens genomic selection, leading to the faster release of high-performing hybrids to meet global food demands.

Challenges in DH Technology

1. Low Haploid Induction Rates: Some genetic backgrounds exhibit lower induction rates, reducing the number of haploid progeny produced.

2. Chromosome Doubling Efficiency: Many chromosome doubling methods have low success rates, affecting the overall efficiency of DH production.

3. High Cost and Resource Requirements: Establishing DH production facilities is expensive, limiting its widespread adoption, particularly in developing countries.

Future Prospects of DH Technology

Ongoing research is focused on overcoming these challenges by improving haploid induction rates, enhancing chromosome doubling methods, and developing more efficient haploid inducer lines. Advances in molecular techniques, such as CRISPR/Cas9, offer potential solutions to increase the efficiency of haploid induction and streamline DH production. As these innovations continue, DH technology is expected to play an increasingly vital role in the development of high-yielding, climate-resilient maize varieties, ultimately contributing to global food security.

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Conclusion

Doubled Haploid technology represents a transformative advancement in maize breeding, providing a pathway to faster and more efficient development of inbred lines which are necessary for the development of superior hybrids. As research and technological advancements continue to refine DH methods, its potential to revolutionize maize breeding and enhance global food security remains immense.

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