

Doubled Haploid Technology

Accelerating Genetic Gain in Plant Breeding

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Received: July, 2025; Accepted: August, 2025; Published: October, 2025

Introduction

Doubled haploid (DH) technology represents a significant advancement in modern plant breeding, enabling the rapid development of homozygous lines in a single generation. By escaping multiple generations of inbreeding, DH methods significantly reduce breeding cycles, thereby accelerating genetic gain. This review explores the principles, methodologies, applications, and recent advancements in DH technology, with a focus on its practical implementation in major crop species.

Principle of Doubled Haploid Technology

DH technology involves the induction of haploid cells (n) from gametophytic tissues followed by chromosome doubling to restore diploidy ($2n$).

Conventional breeding requires several generations of selfing to achieve homozygosity. Doubled haploid (DH) technology provides a powerful alternative by producing completely homozygous lines in one step, thus enhancing selection efficiency and reducing time and cost. First reported in **Datura** by Blakeslee and Avery (1937), DH technology has evolved into a mainstream approach in many breeding programs.

The resulting lines are genetically fixed, making them ideal for direct use in breeding and genetic studies.

Methods of DH Production

- **In Vivo Techniques: Wide hybridization and chromosome elimination:** For example, in wheat, maize pollen is used to fertilize wheat ovules, followed by embryo rescue and colchicine treatment (Laurie&Bennett,1988). **Haploid inducer lines:** Used effectively in maize, where inducer lines can trigger haploid formation during fertilization (Rober, Gordillo, & Geiger, 2005).
- **In Vitro Techniques: Anther culture:** Microspores are cultured to form embryoids, commonly used in barley and rice. Isolated microspores are cultured directly, often more efficient and genotype independent. **Ovary/ovule culture:** Used in crops like sugar beet where male gametophyte culture is less effective (Segui-Simarro, 2010).

Applications in Plant Breeding

- Rapid development of homozygous lines for hybrid programs.
- Genetic mapping and QTL analysis using uniform DH populations.
- Genomic selection and marker-assisted breeding, where DH lines improve selection accuracy.
- Pre-breeding and introgression of exotic traits, particularly in recalcitrant germplasm.
- Reverse genetics and mutagenesis studies, utilizing DH lines to fix mutations quickly.

Advantages of DH Technology:

- Complete homozygosity in one generation.
- Time and cost efficiency.
- Enhanced accuracy in phenotypic and genotypic selection.
- Effective in producing uniform, stable lines for multilocation trials.

Limitations and Challenges

Genotype dependency: Not all genotypes respond equally well to DH protocols. Required specialized facilities and skills.

Case Studies:

Barley: Anther and microspore cultures successfully used for breeding and functional genomics (Forster et al., 2007).

- Rice: Challenges with albino plantlets, but progress with genotype screening and culture media optimization (Jain et al., 2022).

- *Brassica napus*: Efficient microspore culture protocols for rapid DH line development used in hybrid canola breeding (Ferrie & Möllers, 2011).
- Sugar beet: Ovule culture combined with flow cytometry for haploid identification (Paesold et al., 2012).

Conclusion

Doubled haploid technology is a transformative tool in plant breeding. It offers unparalleled speed, accuracy, and efficiency in the

development of pure lines. While challenges remain, ongoing innovations continue to expand its utility across crop species.

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