



Orchestrating Growth Cycles: The Plant Physiology that Fuels Speed Breeding

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Abstract

To achieve food security, quick investments in crop improvement are required to increase their resistance for both biotic and abiotic threats while sustaining high-quality and optimal yields and this is the gap to be addressed by the concept of speed breeding. From carbon arc to LED lamp, the journey of speed breeding is empowered by the tools of plant physiological factors such as photosynthesis, respiration, flowering and crop phenology. The modifications in environmental growth factors not only allow reducing the seed to seed time interval but also combining with high throughput phenotyping, genome editing and transgenic pipelines. This literature is mainly focusing on the driving energy of speed breeding exploiting from the plant physiological phenomena.

Keywords: Phenotyping, Photosynthesis, Plant Physiology, Speed Breeding

Introduction

Studies have shown that the current momentum of global agricultural advancement will not be sufficient to replenish the hungry plate for the mushrooming global population by 2050, which is likely to meet a 10 billion growth at 25% rate (Andrus, 1963). The cumbersome and time-taking conventional plant breeding strategies can't keep the pace with this exponentially rising demand for food production. Speed breeding is a breakthrough in plant developmental technique that accelerates the plant life cycle from the vegetative to reproductive stage in high-density planting (HDP) by utilizing controlled environments. In this technique, plants are grown in manipulated environmental conditions such as higher and diffused light intensity, longer light duration, optimum light quality, higher temperature, elevated CO₂ levels and addition of hormones and nutrients to hasten development of new varieties (Potts *et al.*, 2023). About 150 years ago, the use of carbon arc lamps for growing plants under artificial light held the probability of speed breeding through manipulating the growing environment. Gradually with the implementation of light-emitting diodes (LED) in indoor plant propagation

systems, fast-growing varieties from NASA made the field for University of Queensland to introduce speed breeding in 2003 for the first time (Potts *et al.*, 2023). In recent days, IRRI has developed the pioneer protocol for rice speed breeding named 'Speed Flower'. This technique speeds up the growth and development from "seed to seed" by advancing the plant physiological process like cell division and elongation, germination and photosynthesis rate. The primary goal of this technique is to shorten the breeding cycle significantly by improving photosynthetic efficiency & biomass accumulation, stimulating flowering and enhancing seed production.

Factors Influencing Physiological Responses in Speed Breeding Technology

1. Lighting Conditions

Photoperiods

Plants are grown in enclosed growth chambers with modified light exposure (light quality, intensity, direction and duration) by using artificial lighting sources (blue and red LED lights) to extend the hours of day length, stimulating photosynthesis, growth rate and flowering

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time. Lettuce shows higher growth rate under red LEDs and blue fluorescent lights and 2 weeks early flowering occurs in petunia and pansies grown with far-red light (Davis and Burns, 2016).

2. Temperature and Humidity

Optimized Conditions

Temperature and relative humidity (60-70%) influence the germination rate, growth rate, flowering and seed set duration. Temperature range of 17-30 °C and 25-35 °C are the most suitable for germination and developmental processes respectively. For speed breeding, an optimal temperature regime for each crop is recommended. The combined effect of optimum temperature and photoperiod accelerates the flowering. In wheat, numbers of generations are enhanced by low temperature in dark and high temperature in light. Alterations in soil moisture status influence the generation time of crop species in speed breeding protocol. When exposed to the moisture stress, early flowering and seed setting are occurred in most of the crops (Table 1).

Table 1: List of crops where speed breeding has cut down the generation time

Plants	Factors affecting the plant response	Generation year ⁻¹
Wheat	H ₂ O ₂ + low temperature	4-6
Spring wheat	Long day	4-6
Rice	Short day	4-5
Clover	Immature seed	3-6
Chickpea	Long day	6
Lentil	Plant growth regulators	8
Soybean	CO ₂	5
Groundnut	Short day	4

3. CO₂ Level

Optimum Level of CO₂

Maintaining optimum level of CO₂ in the controlled environment is essential to reduce the duration of vegetative phase. Critical range of CO₂ varies with the genotypes. CO₂ supply (≥400 ppm) accelerates the breeding cycle of crop plants due to increase in biomass accumulation and earliness of flowering. In the plant growth chamber, the growth of soybean is augmented through CO₂ supplementation which affects the parameters such as plant height, total length of branches, total leaf area, dry weight of leaf, stem and root.

4. Plant Growth Regulators

Hormonal Application

The application of growth hormones regulates the developmental process such as growth and development, flowering and seed set. Plant hormones such as auxin and cytokinin stimulate cell division and elongation, ethylene promotes aging related metabolisms, gibberellins hasten flowering and seed maturity under optimum environmental conditions. With some modifications in temperature (22 °C light/ 18 °C dark) and photoperiod (18 h light/ 6 h

dark), the application of plant growth regulators such as auxin, cytokinin and flurprimidol facilitates to obtain eight generations per year in lentils and Faba beans (Mobini et al., 2015).

5. Cultural Practices

High Density Planting

High density planting helps to maintain a large plant population in a cost minimal way under a speed breeding system. The high competition among the plants leads to the early induction of flowering and seed maturity as reported in rice, sorghum and cotton. The genotypic analysis is highly required for validating the effect of HDP on early flowering as a modulator of speed breeding.

Immature Seeds

In wheat and barley, the immature seeds from speed breeding methods exhibit to be viable in extended duration of photoperiod (22 h of light). The early harvest of seeds subsequently lowers down total production time. The immature seeds (37 days after anthesis) from plants have grown under CO₂ supplementation show higher germination rate in soybean.

Physiological Responses of Crop Plants in Controlled Environment

To optimize the speed breeding protocols, the ideal phenotypes under suitable environments with increased photosynthesis and early flowering are to be selected. These physiological phenotypes are acquired by environmental quantification. The understanding of interaction between plant physiology and environmental cues, helps to optimize suitable conditions to grow plants in controlled growth chambers (Figure 1).

There are various physiological responses which are considering to enhance desirable traits in speed breeding.

Germination

Moisture of growth media acts as a limiting factor to the germination and crop stand by affecting the seed imbibition. Specific temperature and light exposure also work as treatments in initiating germination in seeds. The premeditated application of phytohormones amplifies cell division, elongation, vascular differentiation, chlorophyll biosynthesis and hastens overall crop growth. Deviation from optimized environmental conditions leads to alteration in metabolic, transcriptomic, enzymatic and hormonal level which further slows down the growth. The germination response of wheat seeds is influenced by H₂O₂ treatment at a low temperature (11 °C) and, the 12-23 days reduction in generation time was varied with cultivars.

Root System Architecture (RSA)

Plants in manipulated environments exhibited changes in root length density and branching to quickly access nutrients and water. The plasticity and adaptability of plants which modifies the growth cycle is highly dependent on the RSA adjustment. The impact of controlled environment on RSA determines the genetic and metabolic mechanisms in plants from the speed breeding view. It is also correlated with the yield and yield stability.

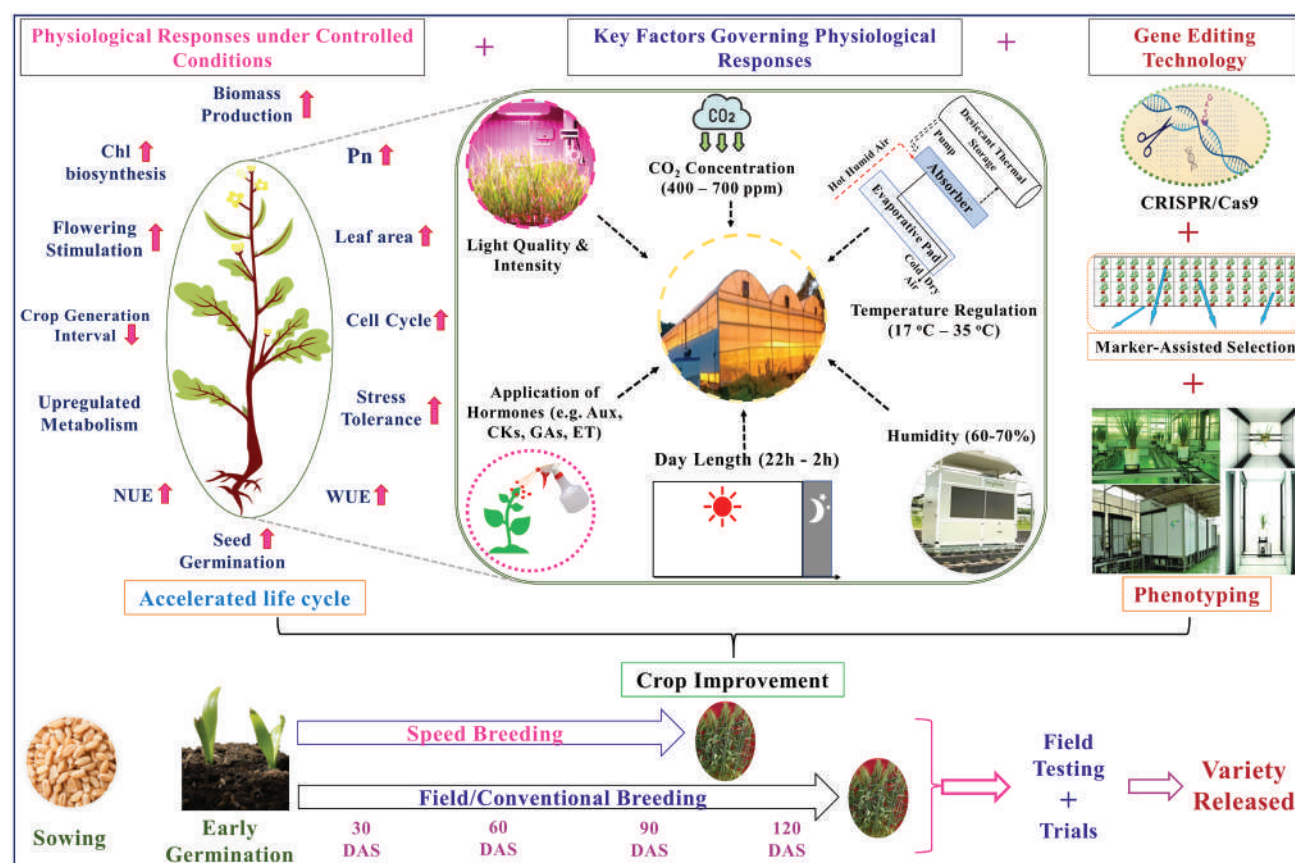


Figure 1: Key physiological responses, environmental factors, gene editing, marker assisted selection and phenotyping tools to accelerate crop improvement in speed breeding [Pn: Photosynthesis rate; Chl: chlorophyll; WUE: water use efficiency; NUE: nutrient use efficiency; CKs: cytokinins; GAs: gibberellic acids; ET: ethylene; Aux: auxins]

Phenology

Photoperiod regulates the transition between different growth stages of plants such as vegetative, flowering, maturity and senescence through photosynthetic performance for determining the speed breeding outcome. Flowering time and morphogenesis are specifically controlled by the light quality and exposure. The hormonal treatments such as auxin and gibberellins shorten the vegetative growth period and induce early flowering, crucial for rapid seed production and simultaneously improves intercellular CO₂ concentration and growth parameters.

Morphology

Light intensity, light quality and light duration influences leaf morphology (size & shape) which results in optimized photosynthetic rate. Red light can induce bud growth and branching whereas, blue light hinders the stem elongation and leaf expansion. The far-red light induces plant internode length, petiole length and plant height. These parameters facilitate speed breeding through photoreceptor perception and triggering rapid reproductive developments.

Photosynthesis

In speed breeding technology, elevated CO₂ levels and standardized illumination is maintained to reduce transpiration alongside maximizing photosystem efficiency in crops. Oversaturated with high light intensity, leaves can't fix excess CO₂ in the dark phase of photosynthesis as it causes

loss of quantum and lower radiation utilization efficiency (RUE), but diffused light with high intensity improves RUE by even distribution of quantum. Temperature and CO₂ level also affects the photosynthesis and transpiration along with nitrogen and phytohormones application.

Biomass Production

Enhanced day light and temperature accelerates germination, stem elongation and thickness in crop plants, providing better support to the plant as it matures quickly. Rapid growth rate in plants under speed breeding setups, promotes the partitioning of more photosynthates to reproductive parts than to vegetative parts. Some studies reported that elevated levels of CO₂ decrease photorespiration and increase the electron flow to RuBisCO carboxylation that results in enhanced biomass production.

NUE and WUE

Maximizing photosynthesis ensures efficient use of nutrients and water, contributing to overall plant health and vigor within the controlled environment of speed breeding. WUE denotes the amount of carbon assimilated as biomass per unit of water used by plants which shows the plants' capacity of survival under different moisture regimes. Developing the NUE and WUE of crops reveals it as a major focal point of breeding and genetic engineering efforts.

Stress Tolerance

Studies reported that hormonal application increases stress

tolerance in crops, enabling them to better withstand the artificial growth conditions in speed breeding techniques. Stress represses photosynthesis through several means of degradation such as leaf shortening, reduced ATP synthase efficiency and upregulated senescence. The application of phytohormones and nitrogenous fertilizers enable plants to increase efficiency of photosynthetic apparatus and other catabolic activity for the growth of roots and leaf area in crops.

Phenotyping

Advanced plant phenotyping is based on the complex traits related to growth, stress tolerance and yield with greater precision, accuracy and scalability. These assessments of growth, development, architecture, physiology, resistance, tolerance, ecology and yield make the field for speed breeding more efficient. The multidimensional interactions of environment and phenotype can be quantified by analyzing plant biomass, root morphology, leaf features, grain traits and protocol can be validated.

Genetic Techniques in Speed Breeding Programme

Following advanced genetic techniques play a vital role in speeding up the selection and advancement of desirable traits.

Gene Editing (CRISPR/Cas9)

Gene editing technologies like CRISPR/Cas9 allow targeted modifications in the genomic DNA, helping the alteration of specific genes associated with desired traits. CRISPR/Cas allows the creation of mutations for different traits at same time. Combination of gene editing technology in speed breeding protocols can accelerate both technological approaches by leveraging the vast genetic diversity found in wild species, uncultivated varieties and crop germplasm as a source of allele-mining for multiple abiotic stresses.

Marker-Assisted Selection (MAS)

MAS involves identifying and selecting plants with desirable traits by analyzing genetic markers associated with those traits, allowing more rapid and accurate trait selection. By directly identifying parents or lines that can be used for the following breeding cycle, genomic selection shortens the breeding period and estimates plant performance using sequence data, SNP markers and algorithms, all without the need for field testing. To anticipate development, speed breeding and genomic selections have been paired and the novel approach is known as “speed genomic selection.” The speed breeding technique is included to enable quick identification and validation of QTLs (quantitative trait loci) also.

Single Seed Descent (SSD)

In field and controlled settings, single-seed descent is a promising selection technique to be used in speed breeding processes. By passing on a single seed from each individual to the following generation, continuous inbreeding of a segregating population paces up the generation of homozygosity. The popular rice cv. Nipponbare was developed by adopting an SSD method with speed breeding

at growth chambers. In legumes, this method is replaced by Single Pod Method.

Difficulties in Adopting Speed Breeding Protocols

Speed breeding techniques for crop improvement face some difficulties as discussed below.

Physiological Challenges

- Extended photoperiod for rapid growth can result in flowering only in long day or day neutral plants.
- Altered light cycle and temperature fluctuation can impact gene expression, hormonal profile and metabolic adjustments along with photoinhibition and oxidative stress.
- Accelerated growth can lead to the deficiency or toxicity of some nutrients in absence of proper nutrient management.
- Sometimes accelerated growth and dense plant populations along with humidity and N-fertilizer make plants more susceptible to pests and diseases.
- Phenotyping can be biased under different environmental exposures as well as immature seed harvesting can also interfere.
- The intensive growth conditions often resulted in limited seed yield.
- Plants grown in controlled condition may not mimic their traits under field condition; therefore, field validation of variety is important.
- While pushing plants to the edge of their ability to alternate physiology, it can lead to the loss of some breeding values at epigenetic level.

Physical Challenges

- Requirement of an advanced growth chamber to control temperature, photoperiod, humidity and CO₂ levels.
- Underdeveloped countries face challenges like lack of modern techniques, financial and policy issues.
- Collaboration with scientists from various organizations is crucial to avoid redundant work, minimize resource investments and facilitate support and sharing of specialized equipment.

Conclusion

The physiological basis of advanced generation improvement in crops explores the metabolic mechanisms of plants that promote extensions in breeding and genetic enhancements. In spite of diverse obstacles, speed breeding has already established itself as game changing technology in plant development to face the changing world scenario which offers the potential of accelerating cultivation, merchandising and commercialization of improved plant varieties for climate resilience, nutritional security and long-term yield.

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