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STRESS TOLERANCE IN HORTICULTURAL CROPS

CHALLENGES AND MITIGATION STRATEGIES



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Speed breeding: a potential tool for mitigating abiotic stresses

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4.1 Introduction

The rising human population, climate changes, decreasing arable land, as well as the increasing demand for valuable natural compounds have significantly increased concern for global food security. Abiotic stresses such as salinity, drought, flood, heat, nutrient deficiency, ozone, heavy metals, and ultraviolet radiation affect plant growth, productivity, and decrease the quality of horticultural crops worldwide (Toscano et al., 2019). The frequent changes in weather expected to be more common in the future, presenting a huge challenge for researchers globally. Abiotic stresses can strongly increase crop yield losses, ranging from 50% to 70% (Francini and Sebastiani, 2019). Plants modulate various physiological, biochemical, and molecular mechanisms to cope with the abiotic-stress conditions. However, the degree of adaptability and tolerance varies among species and varieties (Mariani and Ferrante, 2017). Investigations of these mechanisms lead to a better understanding of stress tolerance and identify the new sources of stress-tolerant traits. Further, breeders translate this information into stress-tolerant varieties using available tools like conventional breeding, marker-assisted selection, and plant transformation (Sarkar et al., 2019). However, horticulture crops typically have long breeding cycles, and breeders require longstanding breeding programs and years for the development and introduction of improved cultivars (van Nocker and Gardiner, 2014).

The development of the new climate-resilient elite varieties takes several years in most of the crops. After hybridization of selected parental lines and even in the case of the advancement of transgenic lines, it requires four to six successive cycles of inbreeding to achieve desirable homozygosity (Watson et al., 2018). Earlier, the shuttle breeding, developed by Norman Borlaug in the 1950s, was the most known approach in wheat to harvest about two generations per year (Ghosh et al., 2018). Other approaches were also being used like physiological stress for early flowering, embryo rescue to shorten generation cycle, and embryo rescue coupled with the application of plant-growth regulators and double haploid (DH) to obtain homozygous lines only in two generations, which otherwise takes six or more generations (Ghosh et al., 2018). Among these approaches, DH was most extensively used in breeding programs for a number of plant species (Hooghvorst et al., 2020). Shortening breeding cycles allow plant breeders to fast-track genetic improvements such as yield gain, biofortification, disease resistance, and climate resilience (Ghosh et al., 2018).

The rapid development of functional genomics and gene technologies over the past decade, particularly sequencing technologies, availability of the complete genome of various crops along with high-throughput analysis tools the genomics have entered its pick stage (Varshney et al., 2009). However, available genetic information has not been effectively exploited due to the archaic phenotyping techniques and the time-consuming traditional cultivation methods, which have not kept pace with the flourishing high-throughput genotyping tools (Yang et al., 2020). Traditional plant breeding is a slow process, attributed partly to the time required to complete the plant life cycle (Fiyaz et al., 2020). Further developing stable lines, plants are grown for many generations, and each new generation is targeted to breed out undesirable traits while keeping the desirable ones (Nawade et al., 2018). The quicker they can take a generation from seed-to-seed, the quicker they can remove undesired traits while promoting wanted ones (Ghosh et al., 2018; Watson et al., 2018). Indeed, it is apparent that generation time in most plant species has become a new bottleneck in crop breeding. This has driven intense efforts by the scientific community of agriculture researchers and engineers to

adapt newer technologies for generation advancement (Ghosh et al., 2018; Watson et al., 2018). Adapting horticulture for faster development of new cultivars is essential to meet evolving consumer preferences, varying climatic conditions, increasing demand for horticultural products, which requires tremendous research efforts from multiple disciplines.

The present chapter discusses the perspectives of speed breeding in horticultural research, emphasizing current development in speed-breeding techniques.

4.2 Speed breeding: a concept to rationality

About 100 years back, Pfeiffer (1926), a botanist, first reported that plants can be grown under an artificial source of light. The effects of continuous supplemented light by using incandescent and electric lamps were studied by Siemens (1880). Subsequently, it was reported that continuous light induces early flowering in several plant species, including cereals, pulses, weed species, vegetables, herbs, and ornamentals (Davis and Burns, 2016).

Inspired by the National Aeronautics and Space Administration (NASA) techniques for growing plants on space station, scientists from the University of Queensland Australia, the John Innes Centre, United Kingdom and collaborators optimized the technique for rapid plant growth and development naming it “speed breeding.” This technique has been successfully utilized for a range of crops like wheat, barley, chickpea, and canola, which uses controlled modified glass-houses fitted with light-emitting diode (LED) to grow plants under extended photoperiods to accelerate plant growth, early flowering, and seed maturation consequently reducing plant generation time by 5 times compared to field conditions and by 2.5 times compared to regular greenhouse (Ghosh et al., 2018; Watson et al., 2018). This advanced technology that shortens the plant breeding cycle and accelerates breeding research is receiving much attention worldwide.

4.3 Speed-breeding components

Speed-breeding techniques consist of setting up a controlled environment that meets all the plant needs and influences its growth at every stage of its development (Ghosh et al., 2018). The basic components of speed breeding include the use of growth chambers with supplemental LED for prolonged photoperiods, controlled temperature, and humidity (Fig. 4.1).

Environmental factors such as light, temperature, and humidity are significant in relation to determine plant’s growth and health levels. Optimization of plant growth involves controlling these environmental factors to encourage and boost photosynthesis and vegetative and reproductive growth (Fig. 4.1). In the case of horticulture crops, various technology-driven approaches are employed to grow plants in highly controlled closed environments where various abiotic parameters essential for plant growth can be optimized and maintained throughout development (Goto, 2003).

4.3.1 Light

Light is the source of energy for plant photosynthesis and growth. Light characteristics such as intensity, duration, spectral wavelengths, and direction can influence plant growth and development (Bayat et al., 2018). Light intensity influences photosynthesis, stem length, leaf color, and flowering (Yoshida et al., 2012). Artificial light is provided as a photoperiodic light to control flowering and as a supplementary light to reduce the plant development time and obtain higher produce quality and yield (Bergstrand and Schussler, 2013). Runkle and Heins (2001) demonstrated that far-red light promotes flowering in several long-day ornamental plants. The petunias and pansies, grown under red:blue:far-red light mixtures, showed earlier flowering up to 2 weeks in plants treated with far-red light compared to plants grown without far-red light (Davis and Burns, 2016). Begonia and poinsettia flowering times were advanced in response to

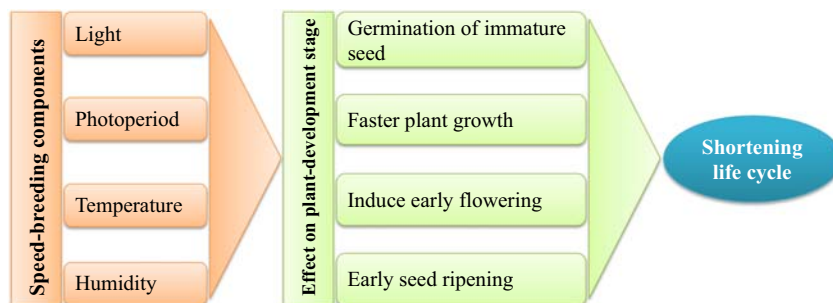


FIGURE 4.1 Speed breeding components and their effect during plant development.

red + white + far-red light treatments (Davis and Burns, 2016). In recent years the use of LEDs has become an increasingly attractive option for lighting in horticultural systems because of its high radiant efficiency, long life, low heat emission, narrow spectrum, capacity to meet the light intensity, and wavelength requirements of different plant species (Bugbee, 2016). The recent achievements in horticulture with the use of LEDs have been reviewed in detail (Viršilė et al., 2019; Bantis et al., 2018; Davis and Burns, 2016). These reviews discuss the application and advances of LED light in horticulture to control flowering and increase transplant success, quality of preharvest, and postharvest produce. The use of LEDs in amending phytochemical content for improvement of nutritional values in horticultural crops also been discussed in the above reviews.

With a well-established platform, artificial growth further could be optimized for shorting the generation time of horticulture crops. According to the recommendation of Ghosh et al. (2018), any light that produces a spectrum that covers photosynthetic active radiation (PAR:400–700 nm), consisting blue, red, and far-red ranges, is suitable to use for speed breeding. The LEDs, or a combination of LEDs, could be used to get an appropriate spectral range.

4.3.2 Temperature

Temperature is a primary factor affecting the rate of plant development. The responses to temperature differ according to plant species and their phenological stage (Makhmale et al., 2015). The vegetative growth of plants increases with a rise in temperature but within the required optimum level temperature. In general, in most of the plant species, vegetative phase generally requires a higher optimum temperature than its reproductive phase (Hatfield and Prueger, 2015). The dormancy in temperate-zone fruits and nuts is naturally overcome by using extended periods of low winter temperature under high-moisture conditions (van Nocker and Gardiner, 2014). There is a strong interaction between temperature and photoperiod for influencing flowering in many species. The combination of optimum temperature and favorable photoperiod was used to accelerate flowering within critical limits (Adams et al., 1999). Environmental control of flowering by light and temperature has been practiced for many years (Hatfield and Prueger, 2015).

For speed breeding, it is recommended that an optimal temperature regime (maximum and minimum temperatures) should be applied for each crop (Ghosh et al., 2018). A higher temperature during the photoperiod and lower temperature during the dark period successfully accelerated the generation times of wheat (Ghosh et al., 2018).

4.3.3 Humidity

Relative humidity directly impacts the water relations of plants and indirectly affects leaf growth, photosynthesis, and pollination. Plant stomatal movements are based on the vapor pressure deficit and air humidity (Taiz and Zeiger, 1991). At high air humidity, plant's water-usage efficiency slows down even when the stomates are open. While at low humidity, transpiration is too high; therefore plants close the stomatal openings to reduce water loss, and wilting consequently slows the photosynthesis resulting in stunted plant growth (Georgii et al., 2017).

Humidity is the most difficult environmental factor to control in controlled-environment chambers, but a reasonable range of 60%–70% is ideal (Ghosh et al., 2018). For crops that are more adapted to drier conditions, a lower humidity level may be advisable (Ghosh et al., 2018).

4.4 Advances in the optimization of conditions for speed breeding

Speed-breeding components are optimized according to crop for reducing generation time, and crop-specific protocols are developed. Mainly three types of approaches were used for speed breeding: controlled environment chamber, use of glasshouse conditions, and benchtop growth cabinet (Watson et al., 2018). The benchtop growth cabinet can be used to conduct experiments before it scaled-up to larger glasshouse conditions (Ghosh et al., 2018). Watson et al. (2018) used a prolonged photoperiod with supplementary lighting and temperature control in the glasshouse or fully enclosed growth chambers and reported that they can grow up to six generations of long-day or day-neutral crops in a year, while traditional techniques only enable one to two generations. The phenotypes and key growth stages in wheat and barley were not affected under speed-breeding conditions (Watson et al., 2018). Till now, speed breeding is successfully implemented for long-day and day-neutral plants that do not require vernalization (Table 4.1). The optimized conditions accelerate the germination of the immature seed, the developmental rate of plants, and induce early flowering and maturity, thereby reducing generation time (Chiuurugwi et al., 2019; Ghosh et al., 2018; Watson et al., 2018) (Fig. 4.1).

TABLE 4.1 Speed breeding protocols optimized for shortening the breeding cycle.

Crop	Type of plant	Growth parameters under RGA or SB				Generation time (days)		References
		Photoperiod	Temperature	Light and other parameters	Humidity (%)	Field condition	Speed breeding	
Wheat (bread)	LD	22 h	22°C/17°C light/dark	SB-I: 360–500 $\mu\text{mol m}^{-2} \text{s}^{-1}$ SB-II: 440–650 $\mu\text{mol m}^{-2} \text{s}^{-1}$	70	105 87	SB-I: 62 SB-II: 65	Watson et al. (2018)
Wheat (durum)	LD	22 h	22°C/17°C light/dark	360–500 $\mu\text{mol m}^{-2} \text{s}^{-1}$	70	102	62	Watson et al. (2018)
Chickpea	Quantitative LD	22 h	22°C/17°C (day/night)	440–650 $\mu\text{mol m}^{-2} \text{s}^{-1}$ (adult plant stage)	70	115	82	Watson et al. (2018)
Barley	LD	22 h	22°C/17°C light/dark	SB-I: 360–500 $\mu\text{mol m}^{-2} \text{s}^{-1}$ SB-II: 440–650 $\mu\text{mol m}^{-2} \text{s}^{-1}$	70	102–115	55–60	Watson et al. (2018)
Sorghum	SD	Continuous light	30°C	–	–	119	77	Rizal et al. (2014)
Peanut	SD	Continuous light (SVLs)	28°C \pm 3°C/17°C \pm 3°C (day/night)	Continuous	65	145	89	Ochatt et al. (2002)
Oat	LD	12 h initial stage 18 h later stage	21°C \pm 1°C/ 18°C \pm 1°C light/dark	340–590 $\mu\text{mol m}^{-2} \text{s}^{-1}$ (50 cm above the pot)	–	114	100	Ghosh et al. (2018)
Soybean	SD	14 h	30°C/25°C (light/dark)	220 $\mu\text{mol m}^{-2} \text{s}^{-1}$ at the canopy level (CO ₂ supplementation at >400 p.p.m)	50–80	102–132	70	Nagatoshi and Fujita (2019)
Canola (1) <i>Brassica rapa</i> (2) <i>Brassica oleracea</i> (3) <i>Brassica napus</i>	LD	22 h	22°C/17°C (day/night)	440–650 $\mu\text{mol m}^{-2} \text{s}^{-1}$ (adult plant stage)	70	171	98	Watson et al. (2018)
			20°C/15°C day/night	356.8 \pm 16.5 to 956.5 \pm 185.0 $\mu\text{mol m}^{-2} \text{s}^{-1}$ at bench height	–	(1) 112 (2) 169 (3) 109	i- 91ii- 128iii- 91	Ghosh et al. (2018)
Pea	LD	22 h	22°C/17°C light/dark	360–500 $\mu\text{mol m}^{-2} \text{s}^{-1}$	70	84	51	Watson et al. (2018)
	LD	16 h	In vivo 24°C/20°C In vitro 24°C/22°C	In vitro plus in vivo system and embryo axis explants	70	143	67	Ochatt et al. (2002)

Lentil	Quantitative LD	20 h	–	–	–	100–110	56	Lulsdorf Banniza (2018)
	Quantitative LD	18 h	22°C/18°C light/dark	178 $\mu\text{mol m}^{-2} \text{s}^{-1}$, use of plant growth regulators and immature seed	–	104	45	Mobini et al. (2015)
Faba bean	DN or LD	18 h	22°C/18°C light/dark	178 $\mu\text{mol m}^{-2} \text{s}^{-1}$	–	365 field, 120 greenhouse	54	Mobini et al. (2015)
<i>Medicago truncatula</i>	LD	22 h	22°C/17°C light/dark	360–500 $\mu\text{mol m}^{-2} \text{s}^{-1}$	70	90–180	78–80	Watson et al. (2018)
<i>Brachypodium distachyon</i>	SD	22 h	22°C/17°C light/dark	360–500 $\mu\text{mol m}^{-2} \text{s}^{-1}$	70	73	48	Watson et al. (2018)
Sugarcane	SD	11 h 42 min to 12 h 45 min	>24°C	Continuous fertilizer to induce synchronous flowering	–	> 365	–	Hale et al. (2017)
<i>Amaranthus</i>	Quantitative SD	LD—16 h in initial stage SD—8 h later stage	LD—35°C/30°C day/night SD—30°C/25°C day/night	LD—150 mmol SD—150 mmol	–	180	60	Stetter et al. (2016)
Tomato	DN	–	–	Introgressed a continuous light-tolerance gene CAB13, which increased productivity under continuous light	–	80	–	Velez-Ramirez et al. (2014)
Potato	LD or DN	–	–	Speed-breeding protocol is under development at James Hutton Institute	–	140	–	–
Rice	SD	11 h	30°C/25°C day/night	350 $\mu\text{mol m}^{-2} \text{s}^{-1}$ and CO ₂ at 475 ppm (biotron speed breeding)	70	122	80	Ohnishi et al. (2011)
		LD-14 h for 30 days SD 10 h after that	30°C/25°C light/dark	350 $\mu\text{mol m}^{-2} \text{s}^{-1}$	70	135	100	Rana et al. (2019)

LD, linkage disequilibrium; RGA, rapid generation advancement; SB, speed breeding; SB-I, speed-breeding method I; SB-II, speed-breeding method II; SD, Short Day; LD, Long Day; DN, Day Neutral; SVL, Sodium Vapour Light.

4.4.1 Acceleration of plant growth

In general, physiological stresses, namely, nutrient deficiency, water access, and plant exposure to intense light induce growth and early flowering (Taiz and Zeiger, 1991). Speed breeding combines some of these techniques avoiding the deficiencies and stress with a good quantity supply of water and nutrients during the growth. The excess of light and heat during daytime gets offset by cooler temperatures at night, and this extended photoperiod is the basis to hasten plant growth (Ghosh et al., 2018). Under 22-h photoperiod and a controlled temperature regime, generation times were substantially reduced for wheat, barley, chickpea, pea, and canola as compared to those of plants grown in a field or a glasshouse with no supplementary light (Table 4.1) (Ghosh et al., 2018; Watson et al., 2018).

4.4.2 Induction of early flowering

A higher temperature is maintained during the photoperiod, while a fall in temperature during the dark period can hasten the flowering (Pazos-Navarro et al., 2017). In the case of amaranth (*Amaranthus* spp. L.) extended light was applied before and following a shortened photoperiod to induce flowering, and hasten initial vegetative growth (Stetter et al., 2016).

4.4.3 Induction of early seed ripening

Seed ripening gets accelerated by higher temperature or water shortage, enabling harvest 1 week after the seed has set in the plant (Ghosh et al., 2018). Optimal values may vary by species, cultivar, and planting conditions. The best results were observed with sowing at 1000 plants m^{-2} , 22 h of LED light at 22°C, 2 h of night at 17°C, and the light spectrum for spring barley (Ghosh et al., 2018).

4.5 Abiotic stresses: where speed breeding can be implemented

Abiotic stresses are always being serious constraints of crop production and alone account for nearly 50% yield loss in agricultural crops worldwide (Theilert 2006; Wang et al., 2007; Shinwari et al., 2020). Plants are frequently exposed to different abiotic stresses such as drought, salinity, high and low temperatures, flood, heavy metal toxicity (e.g., Al^{3+} , Cl^{-} , Cd^{2+} , Fe^{2+} , and Na^{+}), and mineral deficiency (e.g., Fe^{3+} , N, P, S, and Zn^{2+}) (Mickelbart et al., 2015). Among these, drought, salinity, cold, and heat lead to nearly 70% yield reduction of food crops across the globe (Kaur et al., 2008; Thakur et al., 2010). These abiotic stresses have a major impact on crop productivity and production (Roy et al., 2011). Approximately 40% yield loss is caused due to high-temperature stress, 20% yield loss occurs due to salinity, and 17% and 15% yield loss is caused by drought and low temperature, respectively (Ashraf et al., 2008). Other abiotic stresses like low temperature, excess water, heavy metal, mineral deficiency, and radiation also lead to considerable yield penalty in crop production. Further global warming and climate change has increased the impact of abiotic stresses on crop yield.

Limited success has been achieved in elucidating genetic and molecular mechanisms for abiotic stresses tolerance like drought, salinity, cold, ion toxicity, mineral deficiency (Roy et al., 2011; Bechtold and Field 2018). Understanding of tolerance mechanism to abiotic stress right from the perception of environmental signal to cellular response to put forth the adaptive response is very important in the development crop-improvement strategies for stress tolerance (Suprasanna et al., 2016). In the case of abiotic-stress experiments, generation advancement, or phenotypic evaluation in abiotic-stress studies, speed breeding can be integrated in a better way as it requires controlled growth conditions of glasshouse. A controlled environment can minimize the experimental error and reduce the complexity of interactions between genetic and environmental effects on phenotype ($G \times E$ interaction) (Berger et al., 2010; Roy et al., 2011). Controlled environment allows monitoring the start of some stresses like limiting water in drought stress, the addition of salts to hydroponics in salinity, and excessive watering. On the other hand, control conditions do not completely mimic the natural environment and cost-reduced reality, as plants are being grown in the pot conditions compared to field (Passioura, 2006). Under glasshouse conditions, preliminary abiotic-stress experiments are usually performed like water-deficit stress (drought), excess water, temperature extremes, and salinity. Speed breeding can be a reliable tool and can be incorporated in regular breeding programs. The advanced breeding technologies, focusing on the improvement of abiotic stresses tolerance, are transgenics, marker-assisted breeding, genomic selection, express genome editing, etc. It can be used in various stages in the development of stress-tolerant cultivars. Speed breeding assists in defining target stress and environments, identification of superior parents, standardization of screens for stress tolerance, elucidate mechanisms of stress tolerance, identification and characterization of genes imparting tolerance, pyramiding different tolerance in elite lines through marker-assisted breeding, evaluation of breeding lines in target stress environments, etc. However, the use of speed breeding in abiotic-stress tolerance is in infancy stages.

4.5.1 Drought

Drought is considered one of the major constraints in sustainable crop production (Pennisi, 2008; Berger et al., 2010). Unavailability of sufficient water in the soil creates water-deficit stress to plant. Drought stress has been found to limit productivity by reducing stem diameter, leaf area, plant height, and plant biomass in different crops (Farooq et al., 2009; Zheng et al., 2016). The unpredictable heterogeneous nature of drought stress (variations between seasons and years and between regions) is of major concern. The constraints faced by breeders for developing drought-tolerant varieties can be easily removed by using speed breeding (Mitra, 2001). Speed-breeding platform helps to identify a representative drought stress condition, thereby providing homogenous condition for screening. Evaluation of genotypes in controlled conditions makes selection of promising individual plants. Reproducible and precise screening techniques can be used for the identification of superior genotypes in speed breeding, thereby eliminating the selection of lines having negative association of stress tolerance traits with yield. It is usually observed that there is an effect of multiple stresses on plants; however, speed breeding helps to study each stress independently and elucidates the mechanism of stress tolerance. Speed breeding will help to expand the research on abiotic stress in pulses and oilseed crops as most of the efforts are confined in cereals.

4.5.2 Salinity

Salinity is another major abiotic stress and affects more than 20% of global agricultural irrigated land (Glick et al., 2007). Soil salinity is becoming an acute problem to the agricultural world, because of low water quality in arid and semiarid regions (Flowers, 2004; Ghassemi et al., 1995; Rhoades and Loveday, 1990). Salinity affects plant growth in many ways and slightly similar manner as drought by creating water-deficit stress. Even though water is available in the soil in sufficient amount, it cannot be taken up by the plant roots due to difference in water potential, which is created under saline conditions. Salinity also causes nutrient deficiency to plant and thereby suppress growth, metabolism, and productivity. To develop salt-tolerant cultivars, effort is being made through approaches like convention breeding, transgenics, and marker-assisted breeding. Although transgenic and marker-assisted breeding approaches have shortened the breeding process to a certain extent, still there is a need to boost the breeding process to meet the increasing food demand. Speed breeding can further hasten these processes. For example, salt-tolerant lines have been developed in rice in shorter time through combined marker-assisted breeding and speed-breeding approach (Rana et al., 2019). Speed breeding offers scope to improve salinity tolerance and develop better cultivars in other crops like, cereals, pulses, oilseeds, and vegetables at a faster rate.

4.5.3 Temperature

Extreme temperatures (high/low) exerted a significant negative influence on crop productivity worldwide. High-temperature stress witnessed significant yield loss in different crops. High-temperature stress creates oxidative stress to the crop plants and often occurs along with drought and other abiotic stresses (Mittler, 2006). Like drought and salinity, low temperature or cold stress is also the most harmful abiotic stress (Mboup et al., 2012). Cold stress is divided into two types, namely, chilling stress (0°C–15°C) and freezing stress (<0°C). Agricultural and horticultural crops cultivated in tropical and subtropical regions are more prone to low-temperature stress compared to temperate crops (Ritonga and Chen 2020). Low temperature negatively affects many aspects of plant growth, namely, water transport, cell division, photosynthesis, survival, growth, and at the end crop yield (Hasanuzzaman et al., 2013). Conventional breeding, transgenics, markers assisted breeding, and genome editing approaches are being used by the researchers all over the world for the improvement of temperature-stress tolerance in different crop species. Improvement of crops to these major abiotic stresses (drought salinity and temperature) can be fast-tracked with the integration of speed breeding.

4.6 Integration of speed breeding with advance breeding technologies for abiotic-stress tolerance

Abiotic stresses pose a major challenge for crop production and cause substantial yield reduction worldwide (Wang et al., 2003; Wania et al., 2016). Abiotic stresses adversely affect the vegetative and reproductive stages of plant growth and trigger a series of changes at physiological, biochemical, and molecular levels, often resulting in cellular machinery damage (Rai et al., 2011). Plants respond to abiotic stress by different mechanisms that trigger the plant signaling process and transcriptional regulation and produce several stress-responsive compounds like proteins, antioxidants, and

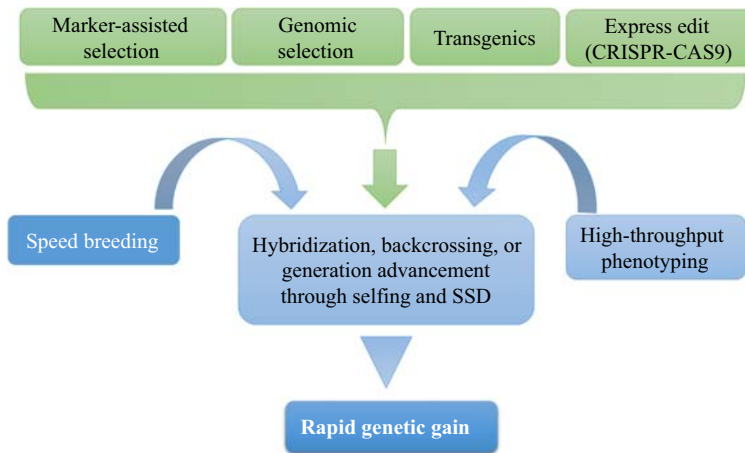


FIGURE 4.2 Integration of speed breeding with advanced breeding technologies for rapid genetic gain.

osmotic solutes (Nakashima et al., 2009). Many genes have been identified from various plants that are responsible for the synthesis of these stress-responsive compounds. These genes are classified into three categories: (1) genes responsible for the synthesis of various osmolytes such as heat-shock proteins, mannitol, proline, glycine betaine; (2) genes that code for ion and water uptake and transport like aquaporins and ion transporter; and (3) genes that regulate transcription and signal transduction mechanism, for example, WRKY1, MAPK, and DREBI (Sarkar et al., 2016; Parmar et al., 2017). Such genes are targeted in breeding programs for the development of stress-resistant cultivars using classical as well as modern breeding techniques like Marker-assisted selection and genetic engineering (Jain, 2015).

However, one of the key limitations in the improved variety development is time. The traditional approaches take more than a decade from making the critical cross to releasing the improved variety. In the current challenges, breeders are facing call for an integration of technologies to enable us to develop crops faster than ever before (Fig. 4.2). Speed breeding is all about growing plants quickly, efficiently, and as cheaply as possible. Speed-breeding technology revolutionizes the concept of growing the plants to keep pace with the recently advanced genomic technology. Speed breeding enables breeders to exploit the collection of germplasms and mutant lines for rapid gene discovery and gene deployment. This approach has been adapted to a range of important crops, particularly field crops, and it is just a matter of optimizing the protocol for inducing early flowering and achieving rapid generation advances.

4.6.1 Combining speed breeding with marker-assisted breeding and genomic selection

The advancement in genotyping has been huge. The low-cost genotyping and advanced sequencing techniques are offering breeders high-density DNA markers across the genome. Advanced high-throughput technologies have revolutionized plant breeding, for instance enabling scientists to track genes and even develop predictive breeding approaches such as genomic selection (Wang et al., 2018). The speed-breeding technique was successfully integrated with marker-assisted backcross for salinity-tolerance rice (Rana et al., 2019). It took 17 months to achieve six backcross generations with an average of 85 days/generation. The obtained BC₃F₃ lines, further subjected to salinity stress, showed significantly less accumulation of Na⁺, higher survival rate, and biomass compared to recipient parent (Rana et al., 2019). The genomic selection uses sequence information, SNP markers, and algorithms to predict the performance of plants without field testing and reduces the breeding timeframe by the direct identification of parents or lines that can be used for the next breeding cycle. Genomic selection has been combined with speed breeding for the prediction of breeding value of quantitative traits (e.g., yield) in wheat (Watson et al., 2019). Integrated genomic selection and speed-breeding approach were used to develop recombinant inbred lines and enabled indirect phenotypic selection for the improvement of important traits, like height and flowering time, before field trials (Watson et al., 2019). The predicting performance of a plant for any trait, like yield or quality, stress or disease, field-testing operations, and the predictive breeding cycles can go very, very fast. In the future, researchers are planning to use artificial intelligence to predict plant performance based on genotyping, also predicting the best parents to use for crossing. Dr. Lee Hickey from The University of Queensland, Australia and his collaborators developed a new methodology that combines speed breeding and the genomic selection and call it “speed genomic selection.”

4.6.2 Combining speed breeding with genome editing and transgenic pipelines

Biotechnology allows the transfer of the desired genes from any organism, plant, or microorganism into targeted crops, extending the opportunities for stress tolerance by offering new genotypes and phenotypes for breeding purposes, and ultimately the development of new improved cultivars (Wang et al., 2019). The precise genome-editing technique, Clustered Regularly Interspaced Short Palindromic Repeats/CRISPR associated (CRISPR/Cas), have been successfully applied in fruits, vegetables, and ornamental crops like potato, apple, citrus, grape, tomato, pear, banana, kiwifruit, cabbage, carrot, and petunia for gene mutation, repression, activation, and epigenome editing (Corte et al., 2019). There are a lot of bottlenecks in the genome editing and transgenic pipelines that prevent scaling developments up and delivering improved crop varieties. For example, the lengthy tissue culture systems that often require 9 months from particle bombardment to seed production. Moreover, this whole process is limited to just one or two specific genotypes that are very favored for the tissue-culture regeneration process.

The CRISPR/Cas system enables breeders to create multiple targets that can be modified simultaneously in an efficient way and allows immediate pyramiding of beneficial traits into an elite background within one generation (Wolter et al., 2019). This technique with speed breeding could help to utilize the enormous genetic diversity present in wild species, uncultured varieties, and germplasm of crops as a source of allele-mining for various abiotic stresses.

4.7 Challenges ahead

So far, speed breeding has shown the biggest potential in long-day plants that flower in response to longer days, which increase the possibility that it will work with horticultural crops like pepper and radish. According to crop species and breeding objectives, several key components need to be evaluated and customized for the development of speed-breeding protocols. The integrations of multiple disciplines such as, high-throughput genotyping and phenotyping with speed breeding to rapidly improve orphan crops and bring them to the forefront of the quest for a well-nourished world population, in the context of unpredictable environmental and socioeconomic conditions. Speed breeding must, therefore be integrated with other breeding techniques as well as cost-efficient high-throughput genotyping and phenotyping to speed up the generation. Therefore speed breeding could be a pivotal tool for accelerating crop growth and reproduction, through which breeders around the world will be able to breed plants and generate improved cultivars that are better adapted to changing climate to feed the increasing population.

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Stress Tolerance in Horticultural Crops: Challenges and Mitigation Strategies

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