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Integration of Rapid Generation Technology in Pulse Crop Breeding

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Abstract : The length of the breeding cycle from seed to seed is a limiting factor in the development of improved homozygous lines for breeding or recombinant inbred lines (RILs) for genetic analysis. The objective of this research was to accelerate the production of field pea RILs through application of rapid generation technology (RGT). RGT is based on the principle of growing miniature plants in an artificial medium under controlled conditions, and allowing them to produce a few flowers which develop seeds that are harvested prior to normal seed maturity. We aimed to maintain population size and genetic diversity in regeneration cycles. The effects of flurprimidol (a gibberellin synthesis inhibitor), plant density, hydroponic system, scheduled fertilizer applications, artificial light spectrum, photoperiod, and light/dark temperature were evaluated in the development of RILs from a cross between cultivars CDC Dakota and CDC Amarillo. The main goal was to accelerate flowering while reducing maintenance and space costs. In addition, embryo rescue of immature seeds was tested for shortening the seed fill period. Data collected over seven generations included plant height, the percentage of plant survival, flowering rate, seed setting rate, the number of seeds per plant, and time from seed to seed. Applying 0.6 µM flurprimidol reduced the internode length. Plant height was decreased to approximately 32 cm allowing for higher plant density without a delay in flowering and seed setting rate. The three light systems (T5 fluorescent bulbs, LEDs, and High Pressure Sodium +Metal-halide lamp) evaluated did not differ significantly in terms of flowering time in field pea. Collectively, the combination of 0.6 µM flurprimidol, 217 plant. m-2, 20 h photoperiod, 21/16 oC light/dark temperature in a hydroponic system with vermiculite substrate, applying scheduled fertilizer application based on growth stage, and 500 µmole.m-2.s-1 light intensity using T5 bulbs resulted in 100% of plants flowering within 34 ± 3 days and 96.5% of plants completed seed setting in 68.2 ± 3.6 days, i.e., 30-45 days/generation faster than conventional single seed descent (SSD) methods. These regeneration cycles were reproducible consistently. Hence, RGT could double (5.3) generations per year, using 3% occupying space, compared to SSD (2-3 generation/year). Embryo rescue of immature seeds at 7-8 mm stage, using commercial fertilizer solutions (Holland's Secret™) showed seed setting rate of 95%, while younger embryos had lower germination rate. Mature embryos had a seed setting rate of 96.5% without either hormones or sugar added. So, considering the higher cost of embryo rescue using a procedure which requires skill, additional materials, and expenses, it could be removed from RGT with a further cost saving, and the process could be stopped between generations if required.

Keywords : field pea, flowering, rapid regeneration, recombinant inbred lines, single seed descent
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