

In Vitro Flowering in Poplar for Rapid Reproductive Trait Improvement

Background

- Woody perennials, including *Populus* spp., have a juvenile phase that ranges from several years to decades. This and the slow floral development process are major impediments to rapid breeding and fundamental research.
- Early flowering based on heat inducible *FT* (*Flowering Locus T*) expression is time- and labor-intensive, with inconsistent results and sometimes detrimental effects to microsporogenesis.

Approach

- We developed an induction-free early flowering system based on CRISPR knockout of a floral repressor CEN (CENTRORADIALIS). Multiplex editing was then used for speedy investigation of reproductive traits.

Results

- *In vitro* flowering of *cen* mutants was observed within ~4 months of *Agrobacterium* transformation. Single flowers developed directly from axillary buds of node culture within days (Fig. 1).
- We successfully switched sex of *P. tremula* × *alba* (Fig. 1c,d) with a hybrid sex configuration (♀, XZ) to show sex is governed by the same ARR17 master regulator as previously reported in some XY and ZW poplars.
- Multiplex editing resulted in glabrous mutants which were also devoid of seed trichomes (Fig. 2a-d). This shows for the first time all aerial organ trichomes are regulated by the same MYBs and provides a molecular basis for engineering hairless seeds for genetic confinement or for reducing allergen spread in urban/plantation forestry.
- We present evidence for the occurrence of trimonoecy in poplar, with male, perfect (male + female) and female flower development dictated by the node position on the mother (♀) plant (Fig. 2e-h).

Significance

- Our system effectively compressed flowering time to mere months and the year-long floral organ development period to a timescale of days. It promises to fast-track reproductive trait research and accelerate rapid-cycle breeding and rapid-cycle genomic selection of long-lived woody perennials.

Ortega M.A. et al. *New Phytologist* (2022) doi.org/10.1111/nph.18624

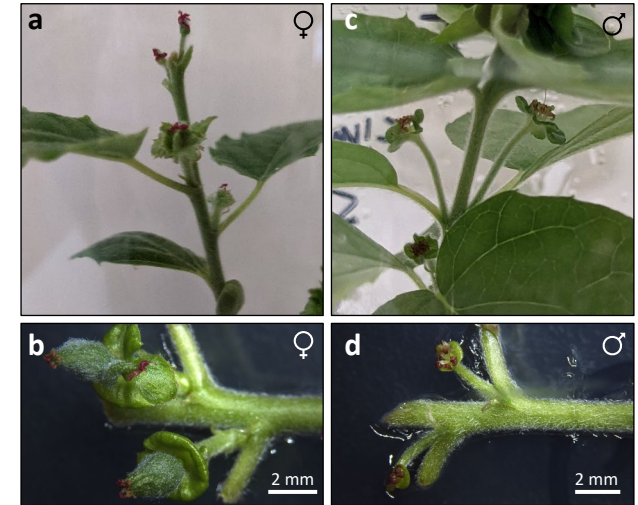


Figure 1. *In vitro* flowering of ♀ (a, b) and ♂ (c, d) poplar mutants.

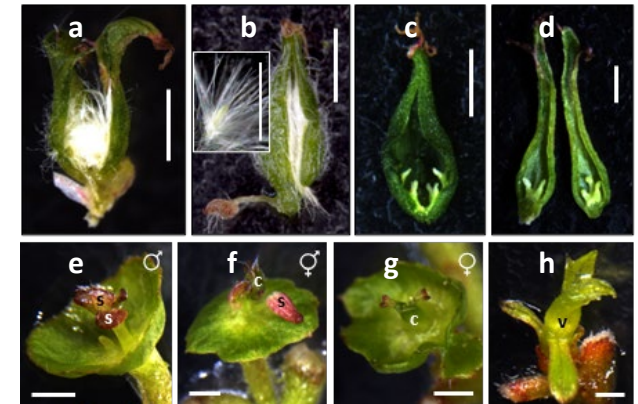


Figure 2. *In vitro* carpels with (a, b) or without (c, d) intra-ovarian (seed) trichomes. A representative plant showing ♂ (e), perfect (f), and ♀ (g) flowers or vegetative (h) growth (bars = 1 mm).