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## Arabidopsis flower dip transformation

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1

Works for me

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### ABSTRACT

A standard protocol how to transform Arabidopsis plants using a flower-dip method.

For higher rates of transformation, plants may be dipped two times at 5-7 day intervals. We suggest one dip two days after clipping, and a second dip one week later.

THIS PROTOCOL ACCOMPANIES THE FOLLOWING PUBLICATION

<ftp://ftp.arabidopsis.org/Protocols/Simplified-Arabidopsis-transformation-Protocol.pdf>

Bechtold, N., Ellis, J., and Pelletier, G. (1993). *In planta Agrobacterium*-mediated gene transfer by infiltration of adult *Arabidopsis thaliana* plants. C. R. Acad. Sci. Paris, Life Sciences 316:1194-1199.

Clough SJ and Bent AF, 1998. Floral dip: a simplified method for *Agrobacterium*-mediated transformation of *Arabidopsis thaliana*. Plant J 16:735-43.

Additional commentary can be found by searching the Arabidopsis newsgroup archives: [http://genome-www.stanford.edu/cgi-bin/biosci\\_arabidopsis](http://genome-www.stanford.edu/cgi-bin/biosci_arabidopsis)

- 1 Grow healthy Arabidopsis plants until they are flowering.
- 2 Prepare *Agrobacterium tumefaciens* strain carrying gene of interest on a binary vector. Grow a large liquid culture @ 28C in LB with antibiotics to select for the binary plasmid, or grow in other media.
- 3 Spin down *Agrobacterium*, resuspend to OD600 = 0.8-1.0 (can be higher or lower) in LB with 5% Sucrose. You will need 250 ml for each four small pots to be dipped.
- 4 Before dipping, add Silwet L-77 to a concentration of 0.05% (125 ul/250mL) and mix well.
- 5 Dip above-ground parts of plant in *Agrobacterium* solution for 10-30 seconds, with gentle agitation. You should then see a film of liquid coating plant.
- 6 Place dipped plants under a cover in a dark O/N to maintain high humidity (plants should be laid on their side).
- 7 Water and grow plants normally. Stop watering as seeds become mature.
- 8 Harvest dry seed. Transformants are usually all independent, but are guaranteed to be independent if they come off of separate plants.
- 9 Select for transformants using antibiotic or herbicide selectable marker.
- 10 Transplant transformants to soil.



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