Genetic Control of Leaf Development

Summary: This kit can be used to demonstrate the concepts presented in the Teaching Tools in Plant Biology module "Leaf Development 2 (Genetic Control of Leaf Development)". This module explores the genetic controls that influence how a leaf is formed. The ten stocks included in this kit include seven mutants with their corresponding reference strains. By growing and observing this set of seeds students will see how mutations can affect various areas of leaf development including identity, polarity, size and shape, and differentiation.

Recommended Grade Level: College

Supporting Resources

All teaching materials related to this kit are available via *Teaching Tools in Plant Biology*, a publication of the American Society of Plant Biologists.

- Teaching Tools in Plant Biology - https://academic.oup.com/plcell/pages/teaching-tools-plant-biology
- Module - https://academic.oup.com/plcell/article/doi/10.1105/tpc.109.tt1209/6096142

Seed Strain Details

Columbia (Col-1, CS28169) – This reference strain of Arabidopsis is closely related to Col-0, which has been completely sequenced and is used as a basis of comparison with other natural variants. Col-1 is a laboratory strain used to generate many mutants, including *ago1-46, as1-1, cuc2-1d, gl1-1,* and *kan1-11* used in this module.

***ago1-46* (CS67862)** – This strain carries an ethylmethane sulfonate (EMS)-induced mutation in *ARGONAUTE 1*, a protein required for a micro RNA (miRNA) function. The resulting mutant, which lacks miRNA function, shows disrupted leaf polarity. The mutant phenotype is reflected in large, serrated leaves, premature production of abaxial trichomes, and abnormal arrangement of leaves in the inflorescence[1].

***as1-1* (CS69042)** – This strain carries an X-ray induced mutation resulting in a single base pair deletion in *ASYMMETRIC LEAVES 1*, coding for a transcription factor. The mutant phenotype is reflected in asymmetrical and lobed rosette leaves[2].

***cuc2-1d* (CS16485)** – This strain carries an EMS-induced mutation in the transcription factor *CUP-SHAPED COTYLEDON 2* miRNA target site. This mutant shows a pleiotropic phenotype with enlarged vegetative and reproductive lateral organs, reflected in enlarged and lobed leaves, elongated inflorescence stems, pedicles and petals, short and curled siliques with thin projections along the valve margins, and lower fertility resulting from reduced seed set[3].

***gl1-1* (CS28175)** – This strain carries a point mutation in the *GLABRA 1* gene which encodes for a transcription factor involved in trichome (leaf hair) formation. The phenotype of the mutant is reflected in the absence of trichomes on the leaf and stems.

***kan1-11* (CS67887)** - This strain carries an EMS-induced mutation in KANADI 1, which is a transcription factor with a function to promote abaxial cell fate. The phenotype of this mutant is reflected in the premature production of abaxial trichomes, and leaves that are either curled upwards or flat in comparison to the reference strain[4].

Columbia (Col-5 (gl1), CS1644) – The whole genome of this reference strain has been sequenced by the Arabidopsis Biological Resource Center. The phenotype of this plant is reflected in slightly serrated leaf margins, and the absence of trichomes on the leaves and stems. This strain was used to generate the *tmm-1* mutant used in this module.

***tmm-1* (CS6140)** – This strain carries an EMS-induced mutation in a receptor protein *TOO MANY MOUTHS* affecting stomatal patterning. The mutant has clustered stomata. The effect of this mutation is stronger in the cotyledons than in true leaves. Inflorescence stems contain very few stomata. The mutant plant has slightly smaller siliques and lacks trichomes on leaves and stems[5].

Nossen (No-0, CS77128) – This reference strain is a natural accession collected in Halle, Germany. No-0 was used to generate the *rev-1* mutant used in this module.

***rev-1* (CS3826)** – This strain carries an EMS-induced mutation in *REVOLUTA* transcription factor, which influences both vegetative and reproductive development. The effect of this mutation on the development of vegetative parts includes abnormally large and misshaped rosette leaves developing after bolting, narrow, downward-curved cauline leaves with rolled-under margins, darker green leaves and primary shoots, lacking secondary and axillary shoots in the rosette and cauline axils. The effect of this mutation on reproductive development includes a reduction in fertile flowers, with most floral meristems forming sterile flowers lacking pistils and stamens, or filamentous structures resembling abortive pedicels[6].

Sources

1. Smith, M.R., Willmann, M.R., Wu, G., Berardini, T.Z., Moller, B., Weijers, D. and Poethig, S. (2009). Cyclophilin 40 is required for microRNA activity in *Arabidopsis*. PNAS, 106(13), 5424-5429.

2. Byrne, M.E., Barley, R., Curtis, M., Arroyo, J.M., Dunham, M., Hudson, A. and Martienssen, R.A. (2000). *ASYMMETRIC LEAVES1* mediates leaf patterning and stem cell function in *Arabidopsis*. Nature, 408, 967-971.

3. Larue, C.T., Wen, J. and Walker, J.C. (2009). A microRNA-transcription factor module regulates lateral organ size and patterning in Arabidopsis. The Plant Journal, 58, 450-463.

4. Wu, G., Lin, W., Huang, T., Poethig, R.S., Springer, P.S. and Kerstetter, R.A. (2008). *KANADI1* regulates adaxial-abaxial polarity in *Arabidopsis* by directly repressing the transcription of *ASYMMETRIC LEAVES2*. PNAS, 105(42), 16392-16397.

5. Nadeau, J.A. and Sack, F.D. (2002). Control of stomatal distribution on the *Arabidopsis* leaf surface. Science, 296(5573), 1697-1700.

6. Talbert, P.B., Adler, H.T., Parkers, D.W. and Comai, L. (1995). The *REVOLUTA* gene is necessary for apical meristem development and for limiting cell divisions in the leaves and stems of *Arabidopsis thaliana*. Development, 121, 2723-2735.