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Expression patterns of auxin response factors in *Arabidopsis thaliana*

The plant hormone auxin is important for plant growth and development. Exogenous application of auxin can cause a change in the expression of specific genes in as little as 5 minutes. A family of 23 genes, auxin response factors (ARFs), has been identified in the plant *Arabidopsis thaliana* as transcription factors involved in the auxin signal transduction pathway. Five of these ARFs are known to activate auxin-responsive genes in the presence of auxin and the rest are likely to repress auxin-responsive genes. Since these transcription factors play such an important role in controlling gene expression in response to auxin, Dr. Guilfoyle's lab has become interested in finding out when and where in the *Arabidopsis* life cycle these ARF genes are active. Two approaches are being used to determine the activity of the ARF genes in the *Arabidopsis* life cycle. Continuing another undergraduate's work, the promoters of two ARF genes, 16 and 17, were individually fused to the reporter gene *fl*-glucuronidase (GUS), and the constructs containing the fused gene were infiltrated into *Arabidopsis*. The transgenic plants will be stained for GUS activity and examined microscopically to determine when and where the promoters of ARF16 and ARF17 genes are active in *Arabidopsis*. The other approach used an *in situ* hybridization analysis of *Arabidopsis* seedlings. For these experiments, a digoxigenin-labeled antisense RNA probe was generated from the unique middle region of ARF19 and reacted with whole seedlings. Following hybridization, the probe and endogenous ARF19 mRNA interaction was detected using antidigoxigenin antibodies that contain an alkaline phosphatase domain. The alkaline phosphatase catalyzed a subsequent chromogenic reaction, which stained the plant organs and tissues. The staining patterns were viewed under the microscope, and allowed us to see where the ARF19 mRNA had been localized. The results from these experiments will indicate where the ARF19 genes are turned on in *Arabidopsis*.