Oxidative inactivation of auxin by DAO1 regulates growth in *Arabidopsis thaliana*

Jun Zhang[1], Jinshan Lin[2], Chinchu Harris[1], Fernanda Campos Mastrotti Pereira[3], Fan Wu[1], Joshua Blakeslee[2] and Wendy Peer[1]

[1]University of Maryland, [2]The Ohio State University, [3]São Paulo State University

Tight homeostatic regulation of the phytohormone auxin (indole-3-acetic acid, IAA) is essential to plant growth and survival. The *Arabidopsis thaliana* enzymes that function in auxin biosynthesis and conjugation to sugars and amino acids for temporary or permanent inactivation have been identified, but the enzyme that catalyzes oxidation of IAA to its primary catabolite 2- oxindole-3-acetic acid (oxIAA) remains uncharacterized. Here we show that DIOXYGENASE for AUXIN OXIDATION1 (DAO1) catalyzes formation of oxIAA *in vitro* and *in vivo* and that this mechanism regulates auxin homeostasis and plant growth. Null *dao1-1* mutants contain 95% less oxIAA compared to wild type, and complementation of *dao1* restores wild-type oxIAA levels, indicating that DAO1 is the primary IAA oxidase in seedlings. Further, *dao1-1* plants have phenotypes associated with increased auxin levels including elongated organs (hypocotyls, primary roots, rosette leaves, inflorescence stems), increased lateral root density and delayed sepal opening compared to wild type. These phenotypes are complemented by transformation with *DAO1pro*:YFP-DAO1. The dominant *dao1-2D* overexpression line has increased oxIAA levels, thus supporting DAO1 IAA oxidase function *in vivo*. *DAO1pro*:YFP-DAO1 expressed in *dao1-1* produces signals in the root tip as well as in all juvenile and mature vascular and epidermal tissues, especially in the sepal. A second isoform, DAO2, is very weakly expressed in seedling root apices. Together, these data confirm that IAA oxidation by DAO1 is the principal auxin catabolic process in Arabidopsis and that DAO1 is an important regulator of auxin homeostasis during plant morphogenesis.