## A role for phospholipase D in nitric oxide induced stomatal closure

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Plants regulate the uptake of carbon dioxide and water loss through pores, called stomata, located in the epidermis of the aerial parts of the plants. Stomatal closure is an essential event for plant survival upon drought stress. The closure is regulated by a complex network of signalling events involving numerous intermediates, among them nitric oxide (NO). Previous studies have shown that NO modulates cytosolic calcium concentration and the activation of plasma membrane ion channels. Our results showed that NO also induces stomatal closure via activation of phospholipase D (PLD) with the consequent production of phosphatidic acid (PA). Multiple PLD isozymes exist. Arabidopsis has 12 with distinguishable biochemical and regulatory properties, and also with little functional redundancy. Two isoforms (PLD $\alpha$ 1 and  $\delta$ ) had been related to drought response. Our current goal is to determine whether PLD $\alpha$ 1 and PLD $\delta$  are required for NO induced stomatal closure. As a measure of stomatal closure we evaluate the leaf loss of fresh weight (LFW), since LFW is largely determined by stomatal conductance. NO treated wild type plants shows less LFW than non-treated plants, reflecting the fact that NO induces stomatal closure. In pld $\alpha$ 1 plants, NO treatment partially avoided the reduction of LFW; but it has no effects on LFW of pld $\delta$ and pld $\alpha$ 1-pld $\delta$  mutants. These results were corroborated by direct microscopic stomata observation in epidermal peels of NO-treated wild type and mutants plants. This indicates that during stomatal closure NO could be activating PA accumulation via PLD\delta and partially via PLD $\alpha$ . Futures studies will be done to measure NO induces gene transcription in PLD mutants.

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