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HOW ARE DGK1 AND DGK2 INVOLVED IN MEMBRANE CONTACT SITES?

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Abstract: Eukaryotic cells have regions of interaction between two organelles where some proteins, which act as tether, bring both membranes closer (10-30 nm) without fusion, named membrane contact sites (MCS). Two organelles that can form MCS are endoplasmic reticulum (ER) and plasma membrane (PM). ER-PM CS play important metabolic functions such as communication between both membranes, lipid homeostasis and Ca^{2+} influx. Our group has identified that AtDGK1 and AtDGK2 (Diacylglycerol kinase 1, AT5G07920 and Diacylglycerol kinase 2, At5g63770) form a complex with a well-known protein located at ER-PM CS, Synaptotagmin1 (SYT1, At2g20990).
Upon perception of stress, phospholipase C (PLC) is activated at the plasma membrane to hydrolyse PIP(4,5)P2 or PI4P in order to generate DAG and inositol phosphates. Diacylglycerol (DAG) is phosphorylated by diacylglycerol kinases (DGKs) to produce phosphatidic acid (PA). DAG and PA are important cell signalling molecules. There are seven DGKs encoded in Arabidopsis thaliana genome, but only DGK1 and DGK2 have a transmembrane domain that anchors them to the endoplasmic reticulum, the rest are cytoplasmic. DGK1 and DGK2 appear to play a role in stress response as both are induced by exposure to low temperatures and wounding. Also, we found that *dgk2* knockout mutant produces lower resistance to freezing. Using confocal microscopy, we have analysed the subcellular localization of these two proteins and investigated their interaction with SYT1 and between them using FRET and co-immunoprecipitation studies. Additionally, we report that the mutation of DGK1 is lethal in homozygosity. Our studies suggest that DGK1 and DGK2 act in concert with SYT1 to regulate the production of PA at ER-PM CS and highlight the importance of these proteins for the correct response to stress tolerance.

Key words: Membrane Contact Sites, Synaptotagmins, Stress.

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References:

Arisz, S. A., Testerink, C., & Munnik, T. (2009). Plant PA signaling via diacylglycerol kinase. Biochimica et Biophysica Acta - Molecular and Cell Biology of Lipids, 1791(9), 869–875. https://doi.org/10.1016/j.bbalip.2009.04.006

Schapire, A. L. et al. 2008. "Arabidopsis Synaptotagmin 1 Is Required for the Maintenance of Plasma Membrane Integrity and Cell Viability." the Plant Cell Online 20(12): 3374–88.

Winter, D., Vinegar, B., Nahal, H., Ammar, R., Wilson, G. V., & Provart, N. J. (2007). An "electronic fluorescent pictograph" Browser for exploring and analyzing large-scale biological data sets. PLoS ONE, 2(8), 1–12. https://doi.org/10.1371/journal.pone.0000718