

Unravelling the true identity of XIP1;1 α in *Nicotiana benthamiana*

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Aquaporins (AQPs) are membrane proteins which can be found across all kingdoms of life and play a crucial role for our survival. They control a highly complex network across the plasma membrane, regulating the influx and efflux of polar molecules and ions. AQPs form hetero or homotetramers whereas each monomer is characterized by six transmembrane α -helices, with a substrate conducting pore in each monomer (1). In higher plant systems, phylogenetic studies demonstrate that AQPs can be divided into five independent subfamilies, whereof one of the subfamilies, the X intrinsic proteins family (XIPs), remains largely unexplored and structures are yet unknown (2).

XIPs are predominantly found in plants and fungus, however they are not found in commonly used plants, e.g., *Arabidopsis thaliana*, suggesting that these types of plants are well functioned without XIPs present. They are known to exhibit transport of smaller solutes, such as H₂O₂, with a variable activity for water. Phylogenetic studies suggest that XIPs are closely related to orthodox AQPs, thus their biological significance and function remain largely unknown (1). By integrating single particle cryo-EM, we aim to determine the first structure of a XIP member, the splice variant NbXIP1;1 α recombinantly expressed in *P.Pastoris*, and characterize its functional importance further. These studies will be supplemented with mass photometry to support our future findings.

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2. Ampah-Korsah H, Anderberg HI, Engfors A, Kirscht A, Norden K, Kjellstrom S, et al. The Aquaporin Splice Variant NbXIP1;1 α Is Permeable to Boric Acid and Is Phosphorylated in the N-terminal Domain. *Frontiers in Plant Science*. 2016;7.