

Investigation on the *Arabidopsis thaliana* Oligopeptide Transporter 5

Mia Huebner, Keelee Storrud, Sagar Judhav, Monica Borghi

Introduction

The *Arabidopsis thaliana* (*Arabidopsis*) *Oligopeptide Transporters (OPT)* family encode for integral membrane proteins. It has been proposed that *OPT5*, a member of this family, transports amino acids and heavy metals [1]. However, the substrate of *OPT5* transport is currently unknown. A study of the *OPT5* promoter using the GUS reporter revealed that the gene is primarily expressed in flowers, while minimal expression was detected in leaves and other tissues [2]. In this study, we performed *in vivo* and *in vitro* analyses to shed light on the function of *OPT5* in flower tissues.

Results

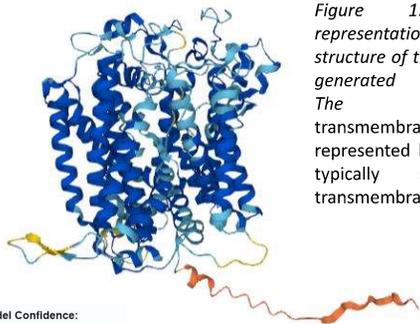


Figure 1: Schematic representation of the 3D structure of the *OPT5* protein generated in UniProt. The numerous transmembrane domains represented by α -helices are typically observed in transmembrane proteins.

Model Confidence:
 Very high (pLDDT > 90)
 Confident (90 > pLDDT > 70)
 Low (70 > pLDDT > 50)
 Very low (pLDDT < 50)



Figure 2: Schematic representation of the OLIGOPEPTIDE TRANSPORTER 5 gene on chromosome 4. The position of T-DNA insertion lines are indicated with a triangle.

Results

Metabolite	Pre-Anthesis	Post-Anthesis	Metabolite	Pre-Anthesis	Post-Anthesis
Gly	1.35478	N/A	Pyroglutamic acid	1.39296	N/A
Pro	1.50035	N/A	Guanidine	3.24152	3.32656
4-Aminobutanoic acid	1.39076	0.76581	Uracil	0.33901	0.15414
Ornithine	1.30527	0.69381	Erythrose	0.73387	0.71869
Phe	1.39576	N/A	Sucrose	1.35653	N/A
Asp	2.18467	0.60688	Ala	N/A	0.61964
Tyr	1.28372	N/A	Putrescine	N/A	0.69053
His	1.76686	N/A	Met	N/A	0.73482
Glycolic acid	1.61613	N/A	Trp	N/A	0.46101
Glyceric acid	1.48436	N/A	Glyoxylic acid	N/A	0.68461
Maleic acid	1.78861	1.95011	Glycerol-3-phosphate	N/A	0.80466
Nicotinic acid	0.69766	N/A	Galacturonic acid	N/A	0.68673

Figure 4: Heat map of Log₂ fold changes of metabolites measured in pre- and post-anthesis florets harvested from *opt5* (*sail_184_B04*) and *Col-0* wild-type plants. The red color indicates higher metabolite abundance in *opt5* than *Col-0*, while the blue color indicates higher metabolite abundance in *Col-0* than *opt5*.

AT4G26590 Expression Profiling

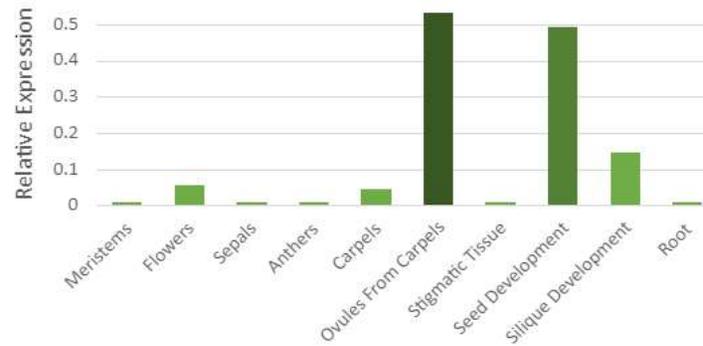


Figure 3: Relative expression of *OPT5* transcripts in different plant organs. *OPT5* is mostly expressed in the female reproductive organs.

Conclusions

- *OPT5* is a transmembrane protein (figure 1) potentially expressed in the chloroplast and believed to export metabolites from the chloroplast to the cytosol.
- The putative localization of *OPT5* is in the female reproductive organs (figure 2). The metabolites shown in figure 5 are significant sources of the essential organic Nitrogen plants need to survive.
- Difference in metabolite abundance in pre- and post-anthesis is shown when *OPT5* is down-regulated (figure 4). This indicates *OPT5* exports the listed metabolites that are more blue in color in post-anthesis. The substrate for *OPT5* transport is currently unknown.

Future Directions

The future of this project will explore the metabolites being transported by the *OPT5* protein and the role they play in seed viability and reproduction. In support to this profile evidence, microscopy will be utilized to visualize the seeds of wild-type *Col-0* and mutant down-regulated *OPT5* gene lines *sail* and *salk*.

References

- [1] Lubkowitz M (2011). The Oligopeptide Transporters: A Small Gene Family with a Diverse Group of Substrates and Functions? *Science Direct*, 4: 407-415.
- [2] Stacey, M.G., Osawa, H., Patel, A. *et al.* Expression analyses of *Arabidopsis* oligopeptide transporters during seed germination, vegetative growth and reproduction. *Planta* **223**, 291–305 (2006). <https://doi.org/10.1007/s00425-005-0087-x>
- [3] UniProt: Q95UA4 · OPT5_ARATH. UniProt. (n.d.). https://www.uniprot.org/uniprotkb/Q95UA4/entry#names_and_taxonomy
- [4] Transcriptome Variation Analysis . Travadb.org. (n.d.). <http://travadb.org/browse/DeSeq/AT4G26590/0-100/AvNorm/Color=RCCount/>