

Resources Available to Arabidopsis Researchers through the Center for Eukaryotic Structural Genomics (CESG)

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CESG (www.uwstructuralgenomics.org) has developed high throughput cloning, expression, purification, and protein structure determination pipelines for both E. coli cell-based and wheat germ cell-free expression systems (see abstract 652 for more details). We have initiated work on over 2700 targets from the annotated Arabidopsis genome, focusing primarily on targets for which 3D structure information is not yet available and on targets suggested by the research community. This effort has yielded a large number of plasmids and other products which are freely available to the scientific community including:

- Over 1500 Gateway Entry Clones and Expression Vectors (primarily as cleavable His Tag-MBP fusions in a pQE backbone)
• Small amounts of purified protein (not all targets are available)
• 43 three dimensional protein structures for Arabidopsis proteins (available through the Protein Data Bank (PDB))
• Optimized protocols for producing recombinant Arabidopsis proteins in both cell-based and cell-free systems (see our web site for details)

Based on data gathered from our project database, the performance of each of the Arabidopsis targets selected for entry into the cell-based (Ec) and/or cell-free (WG) expression systems is summarized below. The PCR step used primarily cDNA isolated from the T87 callus cell line. Entry clones and expression vectors are available for targets with a "+" in the Cloned column. Expression and solubility are rated as high (H), moderate (M), weak (W), positive but level not quantified (+) or negative (-). Structures are determined by either X-ray crystallography or NMR spectroscopy. The initial step in these methods is the determination of protein crystals and analysis of an HSQC spectra, respectively. Contact us at cesginfo@biochem.wisc.edu to nominate a target for study, for additional details on target performance, or for information on obtaining constructs and other resources.

Table with columns: Gene Name, Cloned, Expression, Purification, Structure, and other details. Lists numerous Arabidopsis targets and their experimental outcomes.