

Single cell molecular toolkit for inducible resistance to complete desiccation

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Ability of larvae of the sleeping chironomid *Polypedilum vanderplanki* represent the most complex organism capable to tolerance to complete desiccation. Upon desiccation, the larvae enter into ametabolic reversible state (*anhydrobiosis*). It was shown that during desiccation, the nonredundant sugar (trehalose) substitutes water in the cells, leading to a "vitrification". This mechanism prevent damage of molecules, cell structures and organelles. It has been demonstrated that anhydrobiosis is the property of individual cells rather than hormonally controlled process (reviewed in Coernette and Kikawada, 2010). One of the recent achievements is the establishment of a protocol of *P. vanderplanki* embryonic cell line, capable to withstand complete desiccation, via inducible anhydrobiosis (Nakahara et.al, 2010). Sleeping chironomid genome sequencing revealed several peculiarities in its structure, associated with the ability to desiccation resistance (Gusev et al., 2014). It is suggested that anhydrobiotic clusters (ARIDs) of genes that were not found in the genomes of other insects, including closely related chironomid *Polypedilum nubifer*, responsible for the formation of a "molecular shield" during dehydration. In the current project we aim to dissect the molecular background of the inducible desiccation resistance in the cells by combining data of whole genome cap analysis gene expression (CAGE) analysis, transcriptomics and comparative proteomics (iTraQ). The first stage of the analysis revealed that in contrast to whole larvae,

characterized by more than 15% of total number of genes altered by desiccation, the inducible anhydrobiosis in the cell line associated with less than 1% of total number of genes is differentially expressed under desiccation. We further found that only selected members of ARIDs “gene islands” are expressed and further up-regulated in response to preconditioning with trehalose and further desiccation in the cell line. Taking together the data suggest that the current approach is effective tool to define the minimum essential gene set needed for induction of anhydrobiosis in stand-alone cell line of chironomid and further would be useful for artificial anhydrobiosis methodology for other eukaryotic cell lines. In addition, tissues or organ specialization might be one of the explanation of anhydrobiosis-related genes paralogization in the sleeping chironomid.

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