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Meeting Abstract

P15-4 Sat Jan 2 **Hyperosmolality induces nuclear translocation of osmotic stress transcription factor 1 in Oreochromis mossambicus cells** *MacNiven, L*; Hamar, J; Kültz, D; University of California, Davis, Animal Science; University of California, Davis, Animal Science; University of California, Davis, Animal Science*
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Euryhaline fish tolerate a wide range of environmental salinity by employing molecular mechanisms for coping with the associated osmotic stress. We have previously shown that osmotic stress transcription factor 1 (OSTF1) is part of these mechanisms. OSTF1 is transiently and rapidly upregulated in gill epithelial cells of tilapia (*Oreochromis mossambicus*) exposed to hyperosmolality. Hyperosmotic induction of OSTF1 in tilapia gills was reproduced in the tilapia OmB cell neuroepithelial cell line. OSTF1 shares the signature sequence of the TSC-22 family suggesting that it is a transcriptional repressor. If, in fact, OSTF1 is a transcription factor, we hypothesize that it will localize to the nucleus during hyperosmotic stress. Using standard cloning procedures, OSTF1 was tagged with enhanced green fluorescent protein (EGFP) at either the C- or N-terminus. Using fluorescent microscopy we show that the fusion proteins are retained in the cytosol under iso-osmotic conditions. To evaluate potential nuclear translocation of OSTF1 during hyperosmotic stress, we subjected OmB cells expressing the OSTF1:EGFP fusion protein to hyperosmotic media and imaged at time intervals from 5 minutes to 4 hours using a Leica Dmi8 microscope with automated scanning stage. At four hours and 650 mOsmol/kg, subcellular localization quantified by LASX image analysis (Leica) demonstrated that OSTF1:EGFP was mostly localized to the nucleus. This result supports our hypothesis that OSTF1 is indeed an osmotically inducible transcription factor. Current work evaluates influence of specific OSTF1 domains on nuclear localization. Funded by a NSF grant IOS-1656371.