## Golgi calcium pump secretory pathway $Ca^{2+}$ ATPase 1 (SPCA1) in breast cancer cells

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Calcium signalling regulates a variety of cellular processes including cell growth, cell migration and cell death. Intracellullar $Ca^{2+}$ levels are controlled by an array of $Ca^{2+}$ proteins, including calcium channels, pumps and exchangers. Aberrant expression of $Ca^{2+}$ transporters is a feature of some cancers, including breast cancer. The secretory pathway $Ca^{2+}$ATPase 1 (SPCA1) is a $Ca^{2+}$ pump, which sequesters $Ca^{2+}$ and $Mn^{2+}$ from the cytosol into the Golgi apparatus. SPCA1 levels are elevated in basal breast cancers, which is a molecular subtype with poor prognosis (Grice *et al.*, 2010). Previous studies have shown that SPCA1 silencing alters the post-translational modification of insulin-like growth factor receptor 1 (IGF-1R) in MDA-MB-231 breast cancer cells (Grice *et al.*, 2010). In these studies, 2D-DIGE was employed to identify other proteins sensitive to SPCA1 silencing in MDA-MB-231 breast cancer cells. Validation of the 2D-DIGE results by western blotting showed that SPCA1 silencing downregulated the expression of transketolase and heat shock protein 60 (HSP60). SPCA1 silencing also reduced mRNA levels of HSP60. SPCA1 silencing had no effect on protein expression of the heat shock proteins HSP90 and HSP70, indicating a HSP60 specific effect. Silencing of HSP60 did not alter the post-translation modification or proteins levels of IGF-1R, suggesting that the alternated post-translational modifications of IGF-1R with SPCA1 silencing are independent of changes in HSP60. SPCA1 silencing increased intracellular manganese concentrations in MDA-MB-231 cells. To determine if elevated manganese concentrations could emulate the effect of SPCA1 siRNA, cells were treated with increasing concentrations of manganese or a manganese chelator (N,N,N',N'-tetrakis (2-pyridylmethyl) ethylenediamine, TPEN) and the expression of HSP60 and transketolase was assessed. Manganese addition and manganese chelation had no significant effect on HSP60 or transketolase protein levels. These studies suggest that SPCA1 silencing reduces the ability of MDA-MB-231 to remove $Mn^{2+}$ ions, but this effect does not appear to be the mechanism for SPCA1 silencing-mediated changes in HSP60 and transketolase expression.

Grice DM, Vetter I, Faddy HM, Kenny PA, Roberts-Thomson SJ, Monteith GR. (2010). Golgi calcium pump secretory pathway calcium ATPase 1 (SPCA1) is a key regulator of insulin-like growth factor receptor (IGF1R) processing in the basal-like breast cancer cell line MDA-MB-231. *J Biol Chem*. **285**, 37458-37466.