SR Ca2+-Handling Proteins and PKCε: A Role in Low Molecular Weight FGF2-Mediated Cardioprotection?

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Fibroblast growth factor-2 (FGF2) plays a cardioprotective role against post-ischemic cardiac dysfunction. Both an FGF2-overexpressing mouse model and a high molecular weight knockout (HMWKO) model, expressing only the low molecular weight isoform, have demonstrated increased cardiac function following ischemia-reperfusion (I/R) injury as compared to the wildtype (WT), while an FGF knockout (FGFKO) model, expressing neither isoform, exhibits decreased function. The low molecular weight (LMW) isoform has been shown to be the cardioprotective protein. Protein kinase C (PKC), a modulator of Ca2+ homeostasis via phosphorylation of sarcoplasmic reticulum (SR) Ca2+-handling proteins, is a critical component of LMW FGF2-induced cardioprotection, although upon inhibition of PKCε, only partial abrogation of increased cardiac function is observed. The role of such SR Ca2+-handling proteins in LMW FGF2-mediated cardioprotection is unknown, however, given the central role of these proteins in cardiac muscle function, it is hypothesized that the LMW isoform-mediated cardioprotection against dysfunction involves modulation of SR protein levels and/or activity. In the current study, we measured total protein levels of multiple SR proteins in non-ischemic WT, FGFKO and HMWKO mouse models to elucidate any role that these proteins may play in priming the heart against I/R injury. Phospho-Thr17 PLB and total PLB levels were measured, both in the presence and absence of a PKCε inhibitor for the HMWKO model, to determine what role PLB may play in LMW FGF2-mediated cardioprotection, and if PKCε is involved. Using an isolated work-performing heart model, WT, FGFKO and HMWKO mouse hearts were exposed to varying durations of ischemia and reperfusion. Both non-ischemic and I/R-exposed heart tissue were homogenized and Western blot analysis was performed to evaluate alterations in protein levels or activity. Phospho-Thr17 PLB/total PLB ratio levels were found to be decreased in both FGFKO and HMWKO non-ischemic hearts in comparison to those in WT non-ischemic tissue. Total PLB, SERCA and calsequestrin levels were similar across all non-ischemic models. Phospho-Thr17 PLB/total PLB ratio levels did not differ among models upon I/R stress, while PKCε inhibition failed to modulate these levels. These findings suggest that neither modulation of key SR protein levels nor their activity is critical for LMW isoform-mediated cardioprotection against dysfunction.