A novel vasorelaxing peptide derived from the G Protein-coupled Estrogen Receptor 1

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Store-operated Ca^{2+} entry is required for many endothelial functions, such as the production of nitric oxide. In this mechanism, depletion of ER Ca^{2+} triggers conformational changes in the stromal interaction molecule 1 (STIM1) to promote its interaction with plasma membrane Ca^{2+} channels. Sustained ER Ca^{2+} depletion, however, is associated with ER stress. We recently developed a unique peptide (G2) based on the G protein-coupled estrogen receptor 1 (GPER) that strongly binds the Ca^{2+}-binding loop of STIM1 leading to large conformational changes.

Here, we conjugated G2 with an endothelium-specific leader sequence, aiming to promote endothelial Ca^{2+}-dependent functions without causing ER stress. In *in vitro* testing using a novel STIM1 biosensor, the conjugated peptide (EFG2) directly interacts with the Ca^{2+}-binding loop of STIM1 with 500-fold higher affinity and causes 10-fold greater conformational change therein compared to saturating Ca^{2+}. *In-cell* testing showed that EFG2 triggers Ca^{2+} entry but does not affect ER Ca^{2+} in endothelial cells. EFG2 has no effect in human aortic smooth muscle cells, demonstrating its specificity for the endothelium. In anesthetized rats, intravenous infusion of EFG2, but not a scrambled peptide, causes dose-dependent vasorelaxation. However, when G2 is conjugated to the universal cell penetration sequence TAT, the resultant peptide TFG2 triggers an initial vasorelaxation response followed by rapid vasoconstriction. TFG2 treatment does not increase endothelial ER stress markers BiP or IRE1α or affect voltage-dependent Ca^{2+} current in excitable cells. Thus, EFG2 is a novel vasorelaxing peptide with promising actions to protect endothelial functions.