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Mechanisms of type 1 diabetic serum-induced hyperactivation of \( \text{Ca}_\text{V}1 \) channels in the pancreatic \( \beta \) cell

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ABSTRACT

The pancreatic β cell relies on appropriate Ca\(^{2+}\) entry through voltage-gated calcium (Ca\(_V\)) channels to accomplish its unique function insulin secretion and to guarantee its viability. Well-regulated β cell Ca\(_V\) channels are critical to ensure adequate functional β cell mass, thereby maintaining adequate insulin release and glucose homeostasis in the body. When β cell Ca\(_V\) channels mediate insufficient or excessive Ca\(^{2+}\) influx due to either inherited or acquired defects, β cell becomes malfunctioning and even dies. Type 1 diabetic (T1D) serum hyperactivates β cell Ca\(_V\)1 channels driving Ca\(^{2+}\)-dependent β cell apoptosis via previously unappreciated mechanisms. The present PhD work has mechanistically dissected T1D serum-induced hyperactivation of Ca\(_V\)1 channels in the β cell by combining patch-clamp techniques, confocal microscopy, as well as molecular and cellular approaches. It reveals the following findings:

Functional Ca\(_V\)1.3 channels reside in 20 % of mouse islet Ca\(_V\)1.2\(^{-/-}\) β cells. They characteristically show a large unitary Ba\(^{2+}\) conductance with long-lasting openings in plasma membrane patches of islet cells endowed with undetectable voltage-gated Na\(^+\) currents, larger cell capacitance (> 7 pF) and insulin mRNA. These observations pinpoint β cell-specific Ca\(_V\)1.2\(^{-/-}\) mice as a convenient small animal model for investigation of human β cell Ca\(_V\)1.3 channel-related disorders such as T1D serum-induced hyperactivation of β cell Ca\(_V\)1.3 channels.

T1D serum hyperactivates both Ca\(_V\)1.2 and Ca\(_V\)1.3 channels by elevating their conductivity and number in the β cell plasma membrane. This finding emphasizes that both Ca\(_V\)1.2 and Ca\(_V\)1.3 channels are potential druggable targets for prevention of Ca\(^{2+}\) overload-induced β cell death.

Apolipoprotein CIII (ApoCIII) in T1D serum is electrophysiologically validated to be the actual factor enhancing Ca\(_V\) channel currents in the β cell. This validation opens up the possibility to deplete or neutralize ApoCIII in T1D serum for medical intervention of Ca\(_V\) channel hyperactivation-driven β cell destruction.

ApoCIII activates both PKA and Src kinase in a scavenger receptor class B type I/β1 integrin-dependent fashion to selectively hyperactivate β cell Ca\(_V\)1 channels without altering β cell Ca\(_V\)1 channel expression. ApoCIII-induced hyperactivation of β cell Ca\(_V\)1 channels results from the enriched density and increased activity of functional Ca\(_V\)1 channels in the β cell plasma membrane. This newly-identified signaling pathway shows great potential as a set of novel druggable targets for prevention of Ca\(^{2+}\)-dependent β cell death in association with diabetes.

The key endocytic protein syndapin I/PACSIN 1 (PCS1) is richly expressed in β cells to govern endocytic activity. PCS1-mediated endocytosis acts as a homeostatic control system to fine-tune the Ca\(_V\)1 channel density in the β cell plasma membrane. These findings add a new layer of complexity to the mechanisms of β cell Ca\(_V\)1 channel regulation.

ApoCIII impairs both constitutive and regulated β cell endocytosis with no influence on PCS1 expression. Consequently, ApoCIII abrogates PCS1-dependent endocytic trafficking, thereby accumulating excessive Ca\(_V\)1 channels in the β cell plasma membrane. These results delineate a novel mechanism of Ca\(^{2+}\)-dependent β cell destruction in diabetes development and reveal a promising and attractive option to counteract the critical diabetogenic process of Ca\(^{2+}\)-dependent β cell death.

Overall, the aforementioned findings depict a mechanistic picture of how ApoCIII renders Ca\(_V\)1 channels highly enriched and excessively activated in the β cell plasma membrane, thereby resulting in pathologically exaggerated Ca\(^{2+}\) influx and Ca\(^{2+}\)-dependent β death. These findings lay the foundation for novel treatment strategies for diabetes.