The role of different isoforms of IP_3 receptor in activation of store-operated Ca^{2+} channels in liver cells

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In most animal cells a decrease in the concentration of Ca^{2+} in the endoplasmic reticulum (ER) and possibly other intracellular stores results in activation of store-operated Ca^{2+} channels (SOCs) in the plasma membrane. Physiologically, the empting of Ca^{2+} stores occurs through inositol 1,4,5-trisphosphate (IP_3) receptor (IP_3R)-operated Ca^{2+} channels. In addition to being a major component in the mechanism responsible for Ca^{2+} release from the ER in non-excitable cells, it has been suggested that IP_3Rs play a direct role in activation of SOCs by conformational coupling. To elucidate the role of IP_3Rs in activation of SOCs in liver, we used short interfering RNA (siRNA) to reduce specifically the expression of the genes encoding each type of IP_3R in H4IIE liver cells. Whole cell patch clamping was used to measure the SOC current (I_{SOC}) initiated by the SERCA inhibitor thapsigargin or IP_3. Immunofluorescence and Western blotting were employed to verify the effectiveness of siRNA and the time course of the knock down of IP_3Rs. We found that all 3 types of IP_3Rs are expressed in H4IIE cells, and were able to knock each type down using specific siRNAs. The amplitude of thapsigargin-initiated I_{SOC} in cells transfected with siRNA against each type of IP_3R was the same as that in control cells. These results indicate that, in contrast to considerable published data for other cell types, IP_3Rs are unlikely to activate SOCs through a conformational coupling mechanism. Using IP_3 in the pipette revealed that knocking down IP_3 R1, but not the other types of IP_3R, is sufficient to prevent activation of the I_{SOC} by IP_3. This indicates that Ca^{2+} stores linked to SOCs predominantly express type 1 IP_3R.