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MEETING ABSTRACT

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TRPC1 impact on calcium release mechanisms from the endoplasmic reticulum via interactions with inositol trisphosphate receptors

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Background: Transient receptor potential canonical proteins (TRPC1–7) are a group of calcium-permeable cation channels that exhibit a significant presence in the brain. Among the seven isoforms, TRPC1 stands out for its role in hippocampal physiology and pathophysiology via the regulation of cellular calcium homeostasis. A peculiar feature of TRPC1 is its targeting to the endoplasmic reticulum (ER) membrane, where it can potentially physically associate with other proteins involved in calcium release mechanisms. These interactions might result in functional implications and the engagement of TRPC1 could potentially affect the extent of calcium signaling within the cell, influencing various cellular processes. Based on the intracellular localization of TRPC1, we hypothesize that the channel has the ability to engage with the primary calcium efflux channel in the ER membrane, namely inositol trisphosphate receptors (IP_3R) and, consequently, impact the release of calcium.
Methods: To investigate a potential interaction between TRPC1 and IP_3R, we utilized fluorescence resonance energy transfer (FRET) and Ca^{2+} imaging techniques as well as total internal reflection fluorescence (TIRF) microscopy in transiently transfected HEK 293 cells.
Results: We confirmed via TIRF microscopy that TRPC1 channels target to the ER membrane. FRET experiments, using D1ER as a calcium sensor, showed a significantly lower calcium efflux via IP_3R after application of carbachol (CCh) when TRPC1 was present as opposed to control cells without TRPC1. When introducing the TRPC1 pore dead mutant D582K instead, calcium release levels were restored to match those observed in control cells. Ca^{2+} imaging measurements, using R-GECO as a cytosolic calcium sensor, validated these findings. We detected significantly lower cytosolic calcium levels after perfusion with CCh in cells expressing TRPC1 compared to control cells. Furthermore, cells transfected with D582K displayed increases of cytosolic calcium levels similar to the control.
Discussion: Due to the association of TPC1 with neurodegenerative disorders, we have a vast interest in gaining a deeper understanding of the role of TRPC1 in cellular calcium regulation and the function it plays in the ER. Our findings strongly suggest that TRPC1 engages in regulating IP_3R since its presence led to alterations in calcium release mechanisms. On one hand, we hypothesize that TRPC1 acts as a channel regulator for IP_3R, restricting its function in calcium release. On the other hand, we propose an alternative speculation that TRPC1 serves as a calcium leak channel in the ER since its presence led us to observe a reduced calcium release via IP_3R. This could be attributed to the fact that calcium stores in the ER were already partially depleted through the actions of TRPC1.
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Keywords: TRPC channels – TRPC1 – calcium dynamics – inositol trisphosphate receptors (IP_3R)

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