Journal Pre-proof

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PII: S0143-4160(19)30210-6
DOI: <https://doi.org/10.1016/j.ceca.2019.102141>
Reference: YCECA 102141

To appear in: *Cell Calcium*

Received Date: 6 December 2019
Revised Date: 7 December 2019
Accepted Date: 7 December 2019

Please cite this article as: Sneyers F, Rosa N, Bultynck G, Type 3 IP₃ receptors driving oncogenesis, *Cell Calcium* (2019), doi: <https://doi.org/10.1016/j.ceca.2019.102141>

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Type 3 IP₃ receptors driving oncogenesis

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Abstract

Type 3 Inositol 1,4,5-trisphosphate (IP₃) receptors (IP₃R3s) have been identified as anti-oncogenic channels by propelling pro-apoptotic Ca²⁺ signals to mitochondria. Yet, recent studies (Rezuchova et al, Cell Death Dis, 2019; Ueasilamongkol et al, Hepatology, 2019; Guerra et al, Gut, 2019) revealed that IP₃R3 upregulation drives oncogenesis *via* ER-mitochondrial Ca²⁺ crosstalk, adding complexity to IP₃R3's role in cancer.

Keywords: ITPR3; IP3R3 channels; calcium signaling; cancer; oncogenesis

IP₃Rs control cell death and survival processes, whose dysregulation contributes to oncogenesis. In part, this is due to IP₃Rs located at mitochondria/endoplasmic reticulum (ER) contact sites (mitochondria-associated ER membranes; MAMs), enabling ER-mitochondrial Ca²⁺ transfers. These can suppress oncogenesis by promoting cell death or promote oncogenesis by driving metabolism, anabolic processes, cell proliferation, metastasis and invasion.

Particularly, IP₃R3 has been implicated in pro-apoptotic mitochondrial Ca²⁺ transfers, preventing neoplasia. This was attributed to its preferential localization at MAMs, though recent work revealed that IP₃R2 isoform was the most effective IP₃R isoform in delivering Ca²⁺ to the mitochondria[1]. High IP₃R3-protein levels were found to be maintained by tumor suppressors BAP1 and PTEN, supporting cell death susceptibility. BAP1 deubiquitinates IP₃R3, whereas PTEN competes with FBXL2, an ubiquitin ligase for IP₃R3[2, 3]. Also, oncogenic K-Ras^{G13D}-expressing cancer cells that are highly resistant to apoptosis display IP₃R3 downregulation[4].

However, three recent studies nuance the “IP₃R3 = pro-apoptotic Ca²⁺ signal to mitochondria = anti-cancer” model. When cells can cope with an intrinsically pro-apoptotic IP₃R3 upregulation, they become dependent on IP₃R3 for their survival and cell death resistance, driving oncogenesis and malignant cell transformation (Fig. 1).

First, IP₃R3 levels were elevated in a cancerous region versus healthy region within a kidney biopsy from a clear cell renal cell carcinoma patient[5]. Functional studies in colorectal, ovarian and clear cell renal cell carcinoma cells revealed that IP₃R3 downregulation promoted apoptosis induction. IP₃R3-knockout colorectal cancer cells were severely impaired in *in vivo* tumorigenesis and in cell migration.

Second, IP₃R3 was upregulated in cholangiocarcinoma cancer biopsies and cancer cell lines in comparison to normal tissues and normal cholangiocyte cell models[6]. The mechanisms by which IP₃R3-protein levels are increased in these cancer models remain elusive, though activation of sphingosine-1-phosphate receptor, a factor underlying increased cholangiocyte proliferation, was excluded. IP₃R3 downregulation in cholangiocarcinoma cells abrogated nuclear Ca²⁺ signaling, reducing cell proliferation by prolonging their time in the S phase. Moreover, cancer cells lacking IP₃R3 displayed reduced cell motility and cell migration, potentially due to ~~upregulation~~ ^{downregulation} of occludin, a tight-junction protein involved in cell migration. Fascinatingly, excess IP₃R3 channels localized at MAMs, while in normal cholangiocytes IP₃R3s reside in the ER near the sub-apical pole. This ‘mislocalization’ of oncogenic IP₃R3 channels to MAMs augmented mitochondrial Ca²⁺ signaling, promoting basal respiration. Consistent with the role of IP₃R3 in specialized microdomains, IP₃R3 deletion in cholangiocarcinoma cells affected nuclear and mitochondrial, but not global cytosolic, Ca²⁺ signaling.

Cholangiocarcinoma cells became addicted to high IP₃R3 levels for their survival, as deleting IP₃R3 in these cells augmented spontaneous cell death, decreasing cell survival and 3D-spheroid formation. Importantly, abrupt IP₃R3 overexpression also augmented cell death in ~ $\frac{3}{4}$ th of the cell population. Hence, it seems that IP₃R3 upregulation promotes cell death but if cells are able to cope with this (e.g. through metabolic changes), it could promote cancer cell behavior and cancer cells become dependent on its expression. The factors that drive IP₃R3 upregulation to the MAMs in cholangiocarcinoma cells remain to be identified; though epigenetic factors demethylating the *ITPR3* promoter region could be at the heart of this mechanism (see below). The mechanisms by which excess IP₃R3 channels are recruited and stabilized at the MAMs in these models require further study.

Third, in hepatocellular carcinoma biopsies, IP₃R3-protein levels were elevated compared to healthy liver biopsies[7]. In hepatocellular carcinoma patients, IP₃R3 upregulation is associated with a severely reduced five-year disease-free survival. IP₃R3 contributed to oncogenesis, as *in vivo* tumor formation of IP₃R3-knockout HepG2 liver cancer cells was impaired. IP₃R3 upregulation was linked to a decreased methylation status of the promoter region of the *ITPR3* gene. This was supported by using 5'-aza, a demethylating agent that also demethylated the *ITPR3* promoter and increased IP₃R3 expression. Also, in this case, *in vivo* application of 5'-aza augmented IP₃R3 expression in hepatocytes that localized in microdomains, including perinuclear regions, increasing nuclear Ca²⁺ signaling driving cell proliferation and cell cycling. Additionally, excess IP₃R3 channels localized at MAMs. IP₃R3 deletion in HepG2 cells did not affect basal apoptosis, though augmented their susceptibility towards cell-death inducers. Fascinatingly, IP₃R3 overexpression in HepG2 cells promoted spontaneous apoptosis but rendered these cells more resistant to cell-death inducers. This demonstrates that IP₃R3 upregulation can promote spontaneous cell death, but over time, chronic IP₃R3 upregulation can render cells more resistant to exogenous cell death inducers, thus augmenting malignant cell survival. The underlying explanation for this phenomenon is not fully clear, but could involve a rewiring of the anti- and pro-apoptotic network.

Overall, the three studies shows that IP₃R3 channels can promote oncogenesis and malignant cell survival in different cancer types. While IP₃R3 intrinsically augments cell death, cells capable to cope with this can actually acquire survival benefits by accumulating IP₃R3 in specialized ER microdomains near mitochondria and/or perinuclear region. IP₃R3 at MAMs could increase mitochondrial metabolism and drive anabolic pathways. This higher mitochondrial output sustains enhanced cell proliferation. Additionally, IP₃R3 may cooperate with BK_{Ca} channels to promote cell proliferation, as observed in breast cancer cells[8]. Thus, cancer cells may become dependent on elevated IP₃R3 levels to maintain the survival of the metabolically rewired cancer cell and to acquire malignant properties such as increased motility, migration and invasion. It is fascinating that a pro-apoptotic factor such as IP₃R3 could become an oncogenic asset for certain cancers. This resembles observations for MCU, which antagonizes colon cancer cell survival by promoting cell death[9] but also promotes cancer progression in aggressive triple-negative breast cancers through ROS and HIF1 α signaling[10].

Acknowledgements

The work was supported by Grants from the Research Foundation—Flanders (FWO) (Grants 6.057.12, G.0819.13, G.0C91.14, G.0A34.16, and G.0901.18) and by the Research Council of the KU Leuven (Grants 14/101 and C14/19/099).

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Legend Figure 1.

Schematic overview of the pro-oncogenic impact of IP₃R3 channels upregulated in cancer.