IJAE
Vol. 117, n. 2 (Supplement): 40, 2012
ITALIAN JOURNAL OF ANATOMY AND EMBRYOLOGY

Role-shifting PKCζ fosters its own proapoptotic destruction by complexing with Bcl10 protein at the nuclear envelope of human cervical carcinoma cells

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Many features of deadly human cervical cancers (HCCs) still require elucidation. Among HCC-derived cell lines, here we used the C4-I one since its quantitative gene expression pattern most closely mimics invasive HCCs, including protein kinase-Cζ (PKCζ) overexpression. Via proteomic, bioinformatic, and biochemical approaches (see for technical details [1,2]) we identified 31 and 33 proteins coimmunoprecipitating with PKCζ from nuclear membranes (NMs) of, respectively, untreated or VP-16-exposed C4-I cells. Such proteins belonged to eight functional groups, whose compositions and relative sizes changed with either context. Of the 56 proteins identified, only eight were shared between the two subproteomes, including Bcl10. Surprisingly, proteins known to associate with Bcl10, like Carma1/3 and Malt1 in so called CBM signalosomes were absent. Notably, in VP-16-treated C4-I cells, PKCζ•Bcl10 complexes increasingly accrued at NMs, where PKCζ phosphorylated Bcl10—as PKCζ also did in vitro and in cell-free systems—both processes being thwarted by interfering RNA (iRNA) PKCζ depletion. Caspase-3 was associated with PKCζ•Bcl10 complexes and proteolyzed PKCζ leading to its inactiv-ation/destruction—both events were prevented by Bcl10 iRNA suppression. Thus, PKCζ's molecular interactions and functional roles changed strikingly according to the untreated or apoptogen-treated cells context, and by complexing with Bcl10, PKCζ surprisingly favored its own demise, which suggests both proteins as HCCs therapeutic targets.

References

[1] A. Chiarini et al. (2009) Proteomic analysis of GTP cyclohydrolase I in multi-protein complexes in cultured normal adult human astrocytes under both basal and cytokine-activated conditions. Proteomics 9: 1850-1860.

[2] A. Chiarini et al. (2008). Photoexcited calphostin C selectively destroys nuclear lamin B1 in neoplastic human and rat cells–A novel mechanism of action of a photodynamic tumor therapy agent. Biochim Biophys Acta–Mol Cell Res 1783: 1642-1653.

Keywords: Proteomics, bioinformatics, MALDI-TOF/MS, iRNA suppression, human cervical carcinoma, protein kinase Cζ, BCL10 protein, nuclear envelope, apoptosis, etoposide (VP-16).