P20. Mitoception, the transfer of mitochondria from colon cancer cells to normal colonic cells, reverses remodeling of store-operated channels

Verónica Feijóo[1,2*], Sendoa Tajada[1,2], Lucía Núñez[1,2] and Carlos Villalobos[1]

[1] Unidad de Excelencia Instituto de Biología y Genética Molecular (IBGM), Universidad de Valladolid and Consejo Superior de Investigaciones Científicas (CSIC), Valladolid, Spain
[2] Departamento de Bioquímica y Biología Molecular y Fisiología, Facultad de Medicina, Universidad de Valladolid. Valladolid, Spain.
* Correspondence to VF, veronica.feijoo@uva.es

During cancer process there is a metabolic reprograming which provides survival advantages to tumor cells. The cornerstone of this reprograming is the Warburg effect consisting in the rewiring of aerobic metabolism to glycolisis due to defective mitochondrial synthesis of ATP. Due to this effect, most tumor cells display enhanced mitochondrial potential (ΔΨ), the driving force for mitochondrial Ca^{2+} uptake. Mitochondria are critical players in intracellular Ca^{2+} homeostasis. For instance, they control the Ca^{2+}-dependent inactivation of store-operated channels involved in cell proliferation and other cancer hallmarks. In addition to metabolic reprogramming, cancer cells undergo a deep remodeling of intracellular Ca^{2+} homeostasis. To learn about the contribution of cancer mitochondria to this remodeling we asked whether transfer of mitochondria from normal cells may influence Ca^{2+} remodeling in cancer cells. For this end we isolated mitochondria from normal, human colonic NCM460 cells and labelled them with a fluorescent marker. Then we adapted a protocol of mitoception and transfer of exogenous, normal mitochondria to human colon cancer HT29 cells before investigating intracellular Ca^{2+} homeostasis in mitocepted cells. Our preliminary results showed that HT29 cells with normal mitochondria (HT29 mitocepted) show a lower store-operated Ca^{2+} entry (SOCE) than control HT29. In contrast, when colon cancer cells are self-mitocepted with mitochondria isolated from colon cancer cells, SOCE is enhanced. These results suggest that transformed mitochondria may modulate dramatically CRAC channels involved in store-operated Ca^{2+} entry likely actin on the slow Ca^{2+}-dependent inactivation of these channels.

This work has been supported by grant RTI2018-099298-B-100 from Ministry of Science and Innovation, Spain and grants CCVC8485 and VA294P18 from Junta de Castilla y León, Spain. VF is supported by a predoctoral fellowship from Junta de Castilla y León, Spain.

P21. New approaches for the identification of KChIP2 ligands to study the K_V4.3 channelosome in atrial fibrillation

C. Viedma-Barba,[1] P. Martinez-Salas[1], A. de Benito-Bueno[2,3], PG Socuéllamos[2,3], I. Marín-Olivero[4], M. Daniel-Mozo[5], A. Perez-Lara[4,6], J. A. Gonzalez-Vera[4], A. Orte[4], A. Albert[5], Y. Rodríguez[7], M. Martín-Martínez[1,8], C. Valenzuela[2,3] and M. Gutiérrez-Rodríguez[1,8]

1 Instituto de Química Médica (IQM-CSIC), Spain.
2 Instituto de Investigaciones Biomédicas "Alberto Sols", CSIC-UAM, Spain.
3 CIBERCV Instituto de Salud Carlos III, Spain.
4 Nanoscopy-UGR Laboratory, Dept. de Fisicoquímica, Unidad de Excelencia de Química Aplicada a Biomedicina y Medioambiente, Fac. de Farmacia, Univ. de Granada, Campus Cartuja, Spain.
5 Instituto de Química Física Rocasolano, (IQFR-CSIC), Spain.
6 Dept. of Neurobiology, Max Planck Institute for Multidisciplinary Sciences, Germany. 7Dept. of Natural Sciences, Hostos Community College of CUNY, NY, USA.8PTI-Global Health, CSIC.
carmen.viedma@iqm.csic.es

Ion channels are macromolecular complexes present in the plasma membrane and in intracellular organelles of the cells, where they play important functions. The dysfunction of these channels

results in several disorders named channelopathies, which represent a challenge for study and treatment.[1]

We are focused on voltage-gated potassium channels, specifically on K_V4.3. Kv4.3 is expressed in smooth muscle, heart and brain. Within the heart, Kv4.3 channels generate the transient outward potassium current (I_{TO}). However, I_{TO} characteristics are only observed when Kv4.3 assemble with accessory subunits as KChIP2 and DPP6.

K_V4.3 *channelosome* play a key role in atrial fibrillation (AF), the most common cardiac arrhythmia, with an estimated prevalence in the general population of 1.5–2%. However, current antiarrhythmic drugs for AF prevention have limited efficacy and considerable potential for adverse effects.[2]

KChIP2 (Potassium Channel Interacting Protein 2) belongs to the calcium binding protein superfamily. It is the KChIP member predominantly expressed in heart and a key regulator of cardiac action potential duration.

The identification of novel KChIP2 ligands could be useful to understand the role of K_V4.3 channelosome in AF and it could help to discover new treatments for AF. [3]

In this regard, structure-based virtual screening could be an important tool to accelerate the identification of novel KChIP2 ligands.

In this communication, we will describe a multidisciplinary approach that, starting with a structure-based virtual screening, followed by an iterative process of synthesis/biological evaluation/docking studies, has led to the identification of new KChIP2 ligands.

References

[1] Curran, J. and Mohler, P. J. *Annu. Rev. Physiol.* **2015**, 77, 505.

[2] Birnbaum, S.G. *et al. Physiol. Rev.* **2004**, *84*, 803.

[3] Cercós, P. *et al. Int. J. Mol. Sci.* **2021**, *22*, 1419.

Acknowledgements. PID2019-104366RB-C21, PID2019-104366RB-C22, PID2020-114256RB-I00 and PID2020-119805RB-I00 grants funded by MCIN/AEI/10.13039/501100011033; and PIE202180E073 and 2019AEP148 funded by CSIC. C.V.B. holds PRE2020-093542 FPI grant funded by MCIN/AEI/10.13039/501100011033. PGS was recipient of an FPU grant (FPU17/02731). AB-B holds BES-2017-080184 FPI grant and A.P-L.holds RYC2018-023837-I grant both funded by MCIN/AEI/ 10.13039/501100011033 and by "ESF Investing in your future".

P22. Different modes of synaptic and extrasynaptic NMDA receptor alteration in the hippocampus of P301S tau transgenic mice

Rocío Alfaro-Ruiz[1], Carolina Aguado[1], Alejandro Martín-Belmonte[1], Ana Esther Moreno-Martínez[1], Jesús Merchán-Rubira[3], Félix Hernández[2,3], Jesús Ávila[2,3], Yugo Fukazawa[4] and Rafael Luján[1]

[1] Synaptic Structure Laboratory, Instituto de Investigación en Discapacidades Neurológicas (IDINE), Departamento de Ciencias Médicas, Facultad de Medicina, Universidad Castilla-La Mancha, Campus Biosanitario, C/ Almansa 14, 02006 Albacete, Spain.

[2] Centro de Investigación Biomédica en Red sobre Enfermedades Neurodegenerativas, ISCIII, Madrid, Spain,

[3] Centro de Biología Molecular Severo Ochoa, CSIC-UAM, Madrid, Spain

[4] Department of Brain Structure & Function, Faculty of Medical Science, University of Fukui, Fukui, Japan.

N-methyl-*D*-aspartate receptors (NMDARs) are pivotal players in the synaptic transmission and synaptic plasticity underlying learning and memory. Accordingly, dysfunction of NMDARs has been implicated in the pathophysiology of Alzheimer's disease (AD).

Aims: to investigate the expression and subcellular localization of GluN1, the obligatory subunit of NMDARs, in the hippocampus of P301S mice.