**Role of Voltage Dependent Anion Channels in the HBV life cycle and associated pathologies**

Pierre-Louis SCHMIT-VERGELY\*(1,2), Jennifer MOLLE(1,2), Ahmed GABALLAH(3), Kathrin WEBER(1,2), Fabien ZOULIM(1,2,4), Birke BARTOSCH(1,2)

1: INSERM U1052, CNRS UMR-5286, Cancer Research Center of Lyon (CRCL), Lyon, France.

2: University of Lyon, Université Claude-Bernard (UCBL), Lyon, France.

3: Microbiology Department, Medical Research Institute, Alexandria University, Egypt.

4: Hospices Civils de Lyon, France.

\*This author is the speaker

E-mail address of the speaker : [pierre-louis.schmit-vergely@inserm.fr](mailto:pierre-louis.schmit-vergely@inserm.fr)

**Background**

Hepatitis B virus (HBV) chronic infection affects about 250 million patients worldwide, and is a major cause of cirrhosis and hepatocellular carcinoma. The viral protein HBx has been shown to interact with Voltage Dependent Anion Channel 3 (VDAC3)1 and is suspected to interact with its isoform VDAC1, a porin spanning the outer mitochondrial membrane (OMM). Moreover, it has been demonstrated that HBx affects the mitochondrial metabolism and apoptotic function2,3,4. Here, we investigate the interaction of HBx protein with VDAC1 and the consequences on mitochondrial roles in metabolism and apoptosis.

**Material and Methods**

Immuno-precipitation techniques were used to purify either VDAC1 and its interactome, or whole mitochondria from HepG2 cells engineered to inducibly express HBx. The effects of HBx on VDAC1-mediated apoptosis were explored with cell viability assays based on Neutral Red, SRB stains and cytochrome c release assay in the presence of cytotoxic drugs.

**Results**

We validated an interaction between HBx and VDAC1 by immuno-precipitation using an anti-VDAC1 antibody. HBx was also detected in isolated whole mitochondria. Finally, Neutral Red, SRB stain and cytochrome c release assays showed a significant cytoprotective effect of HBx against various cytotoxic drugs acting potentially via VDAC1.

**Conclusion**

These results suggest that HBx localizes to mitochondria, where it interacts with VDAC1 and potentially alters its apoptotic function. We are working to expand these results to a physiologically more relevant infection system (HBV virus / Primary Human Hepatocytes). Moreover, we developed a fast immuno-precipitation method of whole mitochondria, suitable for mass spectrometry analysis of metabolites, to determine how HBx affects the proteome and metabolome of mitochondria. Ultimately, these discoveries will help us understand the role of mitochondria in the pathobiology of HBV.

**References**

1. Rahmani Z, Huh KW, Lasher R, Siddiqui A. Hepatitis B virus X protein colocalizes to mitochondria with a human voltage-dependent anion channel, HVDAC3, and alters its transmembrane potential. J Virol. 2000 Mar;74(6):2840-6. doi: 10.1128/jvi.74.6.2840-2846.2000.
2. Clippinger AJ, Bouchard MJ. Hepatitis B virus HBx protein localizes to mitochondria in primary rat hepatocytes and modulates mitochondrial membrane potential. J Virol. 2008 Jul;82(14):6798-811. doi: 10.1128/JVI.00154-08.
3. Hepatitis B virus X protein reduces starvation-induced cell death through activation of autophagy and inhibition of mitochondrial apoptotic pathway
4. Association of hepatitis B virus X protein with mitochondria causes mitochondrial aggregation at the nuclear periphery, leading to cell death

**Graphical abstract**

