# CNIO - LILLY CELL SIGNALLING THERAPIES SECTION

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**Titulado Superior* (Advanced Degree)

## SCOPE OF THE ELI LILLY - CNIO PARTNERSHIP

Eli Lilly and CNIO are collaborating on the identification and validation of novel targets in cancer immunometabolism. Our Section is funded through a research contract with Eli Lilly and focuses on the identification of small molecular weight molecules that regulate the metabolism of malignant cells, with the objective of killing them, either directly, acting synergistically with other anti-tumour agents, or activating the anti-tumour immune response. Exploring how to better target these mechanisms would lead to better and more efficient therapeutic options.

A combination of *in vitro* and *in vivo* approaches is being utilised to obtain a complete understanding of the metabolic reprogramming regulated by oncogenes like *RAS*, as well as the characterisation of the metabolic status of tumours (Cerezo A. *et al*, February 2016; Keystone symposium meeting on 'New Frontiers in Understanding Tumor Metabolism' in Banff, Canada). For this purpose, we have developed a series of biochemical and cell-based assays exploiting advanced techniques such as extracellular flux analysis (Seahorse technology), NMR and metabolomics. Finally, each target goes through an *in vivo* validation process using xenografts, allografts and mouse models developed at the CNIO; this process includes the use of non-invasive *in vivo* imaging technologies, as well as the immunohistochemical characterisation of tumours for different metabolic, immune and tumour markers.





## SCIENTIFIC CONTEXT

The observation of an altered metabolic state in cancer cells dates back to the early 20th century when Otto Warburg observed that cancer cells preferentially utilise glycolysis over oxidative phosphorylation for growth, even in the presence of normal oxygen levels (Warburg 1956), a phenomenon known as the 'Warburg effect'. Warburg argued that, 'this altered metabolic state was the underlying cause for cancer'.

The molecular mechanisms driving an altered tumour metabolism have only recently begun to be understood as a result of large-scale genomic sequencing as well as advances in metabolomic profiling technologies. Recent studies have shown that many oncogenes, including Myc and Ras, impart an altered metabolic phenotype in cancer cells through the regulation of genes involved in central metabolic pathways such as glycolysis, fatty acid metabolism, oxidative phosphorylation, and the one carbon pool.

Cellular metabolism is a fine tuned process; tumours may rely heavily on specific metabolic pathways to obtain their energy while using other pathways to grow in order to give tumour cells a growth advantage. This situation may leave tumour cells in a frail position under certain treatments or circumstances, while normal cells may be able to compensate and survive (FIGURE, C). Furthermore, the high requirements of nutrients and other soluble factors, and the release of metabolites with immunosuppressive properties, together with the hypoxic conditions found in tumours, creates a 'non-friendly' microenvironment for an anti-tumour immune surveillance, while facilitating the growth of other tumour-promoting cells such as the stroma and myeloid cells (FIGURE A,B). Thus, the mechanistic understanding of cancer metabolism has led to renewed interest in developing therapeutics that target key enzymes involved in this process. Checkpoint-blockade immunotherapy has been one of the most exciting advances made in cancer treatment in recent years. Metabolic interplay in the local microenvironment can mediate T cell differentiation and function. 'Checkpoint-blockade' antibodies can also influence cellular metabolism. Finally, recent clinical trials have shown that combination immunotherapy based on immune checkpoints blockade, provides even higher response rates than either approach alone. ■

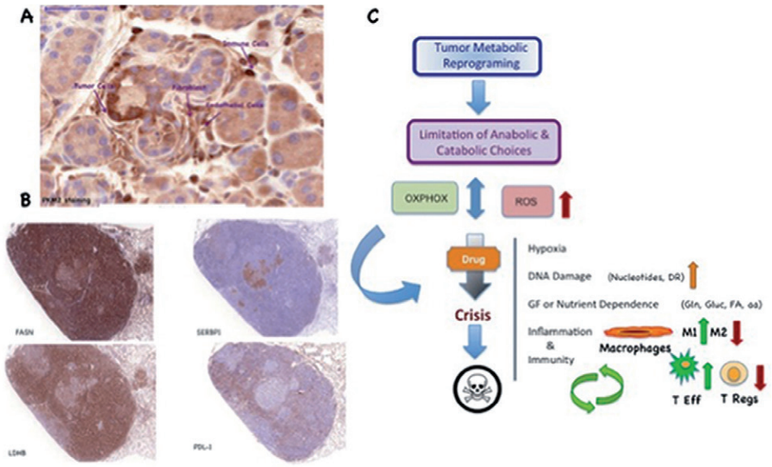


**Figure** (**A**) Immunohistochemical analysis of metabolic enzyme PKM2, showing the tumour microenvironment in a PANIN-3 from a Pancreatic Adenocarcinoma model *Elas-tTA/tetO-Cre;K-Ras(+/LSLG12Vgeo);P53(lox/lox):* PDAC (Carmen Guerra & Mariano Barbacid). In addition to the tumour cells, there are fibroblasts, endothelial, myeloid cells and immune cells that may be facilitating tumour growth, while the immune cells involved in the anti-tumour immune response are absent or inhibited. (**B**) Immunohistochemical analysis showing that the expression of the immune checkpoint ligand PDL1 correlates with the expression of specific tumour profiling markers (SERBP1, Ambrogio *et al.*, 2016) and certain metabolic markers (LDHB and FASN) in a lung adenocarcinoma derived from a *KrasLSLG12Vgeo;Trp53lox/lox* (Chiara Ambrogio & Mariano Barbacid). (**C**) Cartoon depicting a strategy to control tumour growth through the regulation of specific metabolic targets. Tumours may rely heavily on specific metabolic pathways to grow and evade immune surveillance, as well as to obtain energy. This rapid growth also results in increased DNA damage, either through an increase in the production of ROS or due to replication stress. This situation leaves tumour cells more vulnerable to certain metabolic interventions as well as increasing the anti-tumour response of the immune system.