Supervisor: Meike Vogler

Project title: The potential of BCL2-inhibitors for the treatment of lung cancer

Introduction:

Due to their central role in apoptosis regulation the anti-apoptotic BCL2 proteins are attractive targets for the development of novel cancer therapeutics. In haematological malignancies, BCL2-inhibitors like ABT-199 or ABT-263 display clinical activity as single agents. Thus, research and development of these compounds by pharmaceutical companies is currently focussed entirely on lymphoid malignancies. However, a phase II clinical study in advanced SCLC indicated that patients with gene amplification of BCL2 may in fact respond well to ABT-263, highlighting the potential for stratified clinical trials with BCL2-inhibitors in solid tumors, in particular in lung cancer (Rudin et al. 2012, Clin Cancer Res 18, 3163-69). Gene amplification of the BCL2-proteins MCL1 and BCL-XL have been described in a subset of tumors including non-small cell lung cancer (NSCLC) (Beroukhim et al. 2010, Nature 463, 899-905).

In contrast to NSCLC, the most amplified BCL2-protein in Small Cell Lung Cancer (SCLC) is BCL2 itself. Of all solid tumours, SCLC cell lines display the highest sensitivity to ABT-7373, indicating the potential of BCL2-inhibitors to induce apoptosis as single agents. The role of BCL-XL or MCL1 are less clear in SCLC. Preliminary data obtained in SCLC cell lines indicate that BCL-XL and MCL1 may both be relevant targets for anti-cancer therapy.

In this very translational project the addiction of lung cancer cells to BCL2 proteins will be investigated. The student would be integrated in a highly skilled team of researchers at different career levels and would benefit from the collegial environment at the forefront of apoptosis research.

Project outline:

Explant cultures: Targeting of anti-apoptotic BCL2-proteins will be investigated in primary NSCLC tissues acquired at surgical resection. These tissues will be cultured as explants to directly assess the efficacy of BCL2-inhibitors. BCL2-inhibitors are commercially available (ABT-737, ABT-199) or will be obtained through collaboration with Pharma industry (e.g. Servier, Abbvie). Apoptosis will be assessed by immunohistochemistry and staining with cleaved caspase-3 and PARP. Immunohistochemistry will be used to investigate protein expression of BCL2-proteins and correlate with responses.

Primary SCLC cells: In contrast to NSCLC, surgery is rarely performed in SCLC. However, primary cells can be obtained from those patients presenting with
malignant cells and fluid in the pleura and undergoing pleural effusion. In addition, SCLC patients have been shown to contain high numbers of circulating tumour cells (CTCs) which may be purified from the peripheral blood. Any primary material obtained from SCLC patients will be investigated for sensitivity to BCL2-inhibitors.

**BH3-profiling:** Addiction to specific anti-apoptotic BCL2-proteins will be investigated using a technique known as BH3-profiling (Ni Chonghaile et al 2011, Science 334:1129-33), employing sensitivity of permeabilised tumor cells to a panel of pro-apoptotic BH3-peptides that neutralise specific anti-apoptotic BCL2-proteins as determined by mitochondrial outer membrane permeabilisation (MOMP).

**Gene amplification:** To determine whether 1q21.2 (MCL1) or 20q11.21 (BCL-XL) amplifications correlate with BCL2-inhibitor sensitivity, genomic qPCR (duplex reaction test) and fluorescence in situ hybridisation will be conducted in primary tissues. To investigate whether amplifications of anti-apoptotic BCL2-proteins are truncal (ubiquitous) and therefore early driver mutations during NSCLC evolution, we will interrogate the TRACERx database for spatial copy number variations in anti-apoptotic BCL2 genes.