The primary focus of our work is to develop new methods to non-invasively image cancer metabolism and then apply these techniques to investigate the causes and consequences of metabolic heterogeneity in high-risk mouse models of cancer. Our research has two main themes, first we develop novel technologies such as new metabolic radiotracers and new quantitative methods. Second, we exploit PET as a biological imaging modality and investigate the molecular mechanisms and vulnerabilities underlying regional tumour metabolism. The goal of our work is to validate imaging biomarkers for visualising in vivo metabolic phenotypes and, by investigating the liabilities of these phenotypes, determine if we can use metabolic imaging to identify susceptibilities that we can use to guide therapy in individual patients.

Visualising metabolic heterogeneity and plasticity in lung cancer

Metabolic heterogeneity presents both a challenge and an opportunity to imaging. Due to heterogeneity, it’s unlikely that a single imaging test will detect cancer in all cases. However, if we could develop a complementary panel of PET tracers and develop a better understanding of how PET imaging signatures relate to underlying metabolic and molecular features of cancer, we could potentially identify metabolic differences between or within patients and use this information to stratify treatment.

Lung cancer has large regional variations in glucose uptake, hypoxia and blood flow; regions of high and low perfusion within the same lung tumour have striking differences in metabolism. To understand the significance of these imaging signatures we need to relate them to the underlying genetics and metabolism of tumour sub-regions.

Our lab develops new ways to visualise cancer – we create novel molecular tracers that image metabolic reprogramming, a hallmark of cancer, and use state-of-the-art methods such as PET/CT to non-invasively detect and characterise tumour development. This year, we have been developing technologies to image metabolic responses to cancer treatment. Our goal is to develop a better understanding of how cancer drugs work, identifying when those drugs succeed or fail, and supporting the use of more effective therapeutics.

Our goal is to understand the role that metabolism plays in cancer drug development and drug resistance. Our research has two main themes.

1. **Understanding of how cancer drugs work, identifying when those drugs succeed or fail, and supporting the use of more effective therapeutics.**

2. **Developing new ways to visualise cancer – we create novel molecular tracers that image metabolic reprogramming, a hallmark of cancer.**

### Figures

#### Distinct metabolic subtypes identified in oncogenic Kras-driven lung cancer.

(a) and (b) Dual radiotracer PET imaging with sequential [11C]acetate and [18F]FDG injections indicating respective tumour regions with elevated fatty acid synthesis and glucose uptake. (c) Principle component analysis from dual-isotope positron emission tomography (PET) imaging reveals distinct metabolic phenotypes, showing distinct separation. (d) Gene set enrichment analysis highlighting elevated fatty acid metabolism as a key differentially expressed pathway.


#### Tomographic imaging of spontaneous tumorigenesis and treatment response.

Genetically engineered mouse models (GEMMs) of cancer recapitulate the genetic and microenvironmental heterogeneity characteristic of clinical tumours. There is evidence that GEMMs are more predictive of patient response than subcutaneous tumours. However, visualising tumour formation and treatment response is difficult as tumours develop with varying latency and at autochthonous sites.

We are therefore further developing radionuclide imaging of NIS as a method for visualising oncogenesis and drug response in transgenic mouse models. NIS imaging is an improvement on current optical methods, as it does not suffer from the same photon scatter and absorbance as light, for example, bioluminescence. We have taken two approaches to the gene delivery of NIS: somatic induction and germ-line transgenesis. Somatic induction has the advantage that the vector can be readily customised at the bench to modify reporter readout or tumour genotype without additional mouse development or breeding. While germ-line transgenesis results in more reliable gene expression at predetermined genomic loci but requires extensive breeding.

We built three novel lentiviral vectors (LV-PKCh-415NIS, LV-PKCh-415Liu-LV-PKCh-Lini), which we used to stably deliver multiple transgenic elements to somatic cells of adult mice with fixed oncogenic Kras (LSL-KrasG12D) and p53loxP/loxP (lps) mice. The vectors contain CRE recombinase for tumour induction driven by a constitutively active P21 promoter and reporter genes (luciferase/ mStrawberry, sodium-iodide symporter in mNIS/mStrawberry and luciferase/ sodium-iodide symporter in mNIS) driven by the high activity promoter Ephi.

Mice were longitudinally imaged using [11C]acetate and [18F]FDG, and lesions down to the nanocarriers could be repeatedly imaged. Imaging after drug therapy identified single injection lesions with significantly reduced [11C]acetate uptake and elevated cleaved caspase 3 (CC3) staining, indicating that imaging identified true responders 24 hours after treatment. This is the first example of a radionuclide reporter gene used to monitor spontaneous tumour development in a live mouse. This is important, as unlike bioluminescence techniques, it enables sensitive three-dimensional imaging of tumour development in vivo that is unaffected by overlying tissue-depth or tissue pigmentation.

We are exploiting the tomography of radionuclide imaging to track single lesions at nanoscale resolution during cancer therapy and further to monitor inter- and intra-tumoural heterogeneity in drug response.

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