Molecular dissection of colorectal cancer in pre-clinical models identifies patterns of intra-tumour heterogeneity and biomarkers predicting sensitivity to EGFR inhibitors
Methods for systematic next-generation oncology biomarker development

Preclinical models
- organoids
- xenografts

Oomics analysis
- GenomeSeq
- ExomeSeq
- RNASeq
- Methylome Array

Preclinical platforms
- EGFR blockade
- Chemotherapy
- MEK inhibitor
- mTOR inhibitor
- Angiogenesis inhibitors

Identification of biomarkers of drug sensitivity
NGS analysis of matching patients, patient-derived xenografts (PDX) and organoids (PDO) gives insight into intra-tumour heterogeneity.

1) Varying degree of concordance in somatic mutations
2) Discordances in cancer-relevant genes

* Validated by targeted seq. samples

OT cohort (101 samples)
Drug screening using 16 compounds

Drug sensitivity largely concordant between xenograft and organoid. Interesting exceptions with potential implications on personalized medicine.

**Drug Sensitivity**

<table>
<thead>
<tr>
<th>Drug</th>
<th>T/C (%)</th>
<th>Response categories</th>
</tr>
</thead>
<tbody>
<tr>
<td>SFU</td>
<td>0-100</td>
<td>Strong</td>
</tr>
<tr>
<td>Oxaliplatin</td>
<td>0-100</td>
<td>Moderate</td>
</tr>
<tr>
<td>Irinotecan</td>
<td>0-100</td>
<td>Minor</td>
</tr>
<tr>
<td>Selumetinib</td>
<td>0-100</td>
<td>Resistant</td>
</tr>
<tr>
<td>BI 860585</td>
<td>0-100</td>
<td>Strong</td>
</tr>
<tr>
<td>Linfatinib</td>
<td>0-100</td>
<td>Moderate</td>
</tr>
<tr>
<td>Nintedanib</td>
<td>0-100</td>
<td>Minor</td>
</tr>
<tr>
<td>Regorafenib</td>
<td>0-100</td>
<td>Resistant</td>
</tr>
<tr>
<td>Sorafenib</td>
<td>0-100</td>
<td>Strong</td>
</tr>
<tr>
<td>Vandetanib</td>
<td>0-100</td>
<td>Moderate</td>
</tr>
<tr>
<td>Atezolizumab</td>
<td>0-100</td>
<td>Minor</td>
</tr>
<tr>
<td>AZD98931</td>
<td>0-100</td>
<td>Resistant</td>
</tr>
<tr>
<td>Gefitinib</td>
<td>0-100</td>
<td>Strong</td>
</tr>
</tbody>
</table>

**Drug sensitivity: PDX vs PDO**

- **PDO**
  - Resistant
  - Minor response
  - Moderate response
  - Strong response

- **PDX**
  - Strong
  - Moderate
  - Minor
  - Resistant

**Interesting case for precision medicine:**
PDO-private EGFR mutation triggers drug response
New predictive gene expression signatures by combining molecular and drug sensitivity data

Targeted EGFR antibody cetuximab

Cetuximab: Validation on external cohort

Mutation status in KRAS/BRAF/NRAS Mutations status in KRAS codon 12/13 OT mini classifier
OT mini classifier OT mini classifier cases with mutation information (n=96) cases with mutation information (n=155) whole cohort (n=164)
WT KRAS codon 12/13 (n=96) WT KRAS/BRAF/NRAS mutations (n=36)