GCIG Translational Committee – **November 2013, London, UK**

MINUTES

1. GCIG tissue MTA - use TCGA/IRCI as a template, send to harmonisation group for their thoughts. Try to get in place in time for the Chicago meeting. Need to liaise with Harmonisation to achieve this. This is particularly important in collaboration with the phase II working group and the potential trial in carcino-sarcoma.

2. David Bowtell (ANZGOG – Walter and Eliza Hall Institute, Melbourne). Presented data on Cyclin E1 amplification as a potential therapeutic in high grade serous ovarian cancer. Data suggesting that cdk2 inhibitors and/or proteasome inhibitors may have therapeutic use in CCNE1 amplified disease - phase II trial in set up dinaciclib.

3. Suzy Scholl (GINECO – Institut Curie, Paris). Presented background on the RAIDS (Rational Assessment Innovative Drug Selection) project, looking at evaluation of biomarkers in cervix cancer, including sequencing of primary tumours and establishment of new cervix cancer lines. Looking for more centres to participate - 700 required in total.

4. Paul Speiser (AGO-Austria – Medical University, Vienna). Update on uterine lavage as a method of detecting Fallopian tube tissues - two separate studies, LUDOC - in women with suspected pelvic serous cancer and LUSTIC - in women with germline BRCA1/2 mutations undergoing risk-reducing BSO.

5. James Brenton (SGCTG – CRUK Cambridge Institute, Cambridge). Intra-tumoral heterogeneity and techniques to identify key driver mutations as low frequency alleles and ability of circulating tumour DNA to detect these.

ADDENDUM NOTES:

David Bowtell. Primer driver of 19q12 amplicon is CCNE1. There are also a group with gain rather than amplification - their outcome is halfway between normal and amplification. CCNE1 amp and BRCA1 germline are mutually exclusive. Bowtell data suggest that they are synthetic lethal to each other, so it's not possible to have both in a cell and for that cell to survive. Amplified 19q12 lines are dependent upon CCNE1 - if you siRNA, they die and they are also sensitive to cdk2 inhibition.

The BRCA1:CCNE1 synthetic lethal concept came from siRNA screen in CCNE1 amplified cells. There isn't a link with BRCA2, which is interesting. There are other genes that come up in the screen that produce greater additivity to CCNE1 gain - several of these genes were in the protein degradation/proteasome pathway. Hence the idea of bortezomib. Alan D'Andrea's group had shown that bortezomib can non-specifically inhibit Fanconi pathway and sensitise cells to cisplatin. The idea is that the HR pathway does rely upon multiple ubiquitylation processes for adequate function.

DB data show that CCNE1 amplified cells are sensitive to bortezomib even if they are resistant to cdk2 inhibitor.

Trial plan: patients with HGS with short PFS, screen for CCNE1 amplification by IHC and CISH, then treat with dinaciclib (Cdk2 inhibitor) and/or bortezomib or ANother proteasome inhibitor.

Cells can be made resistant to cdk2 inhibitor by classic cooking in the lab. They don't change copy number or new point mutations, nor is there cross resistant to MDR1 pumped drugs.

OVCAR3 cells contain two populations - one is hypotriploid and the other is hyperpentaploid. The resistant cells contain the pentaploid population. CCNE1 amplified tumours seem to contain naturally occurring tetraploid/polyploid populations

Suzy Scholl
Evaluation of biomarkers in advanced cervix cancer. RAIDS - 15 partners in 8 EU with 4 SME. 8 workpackages, most important one of which is prospective assessment of active pathways and genetic mutations in 700 cervix cancer patients, followed by RPPA, sequencing etc etc. Two clinical trials biult in - one vaccine trial with chemorad and second trial with cidovovir plus CCRT. Still looking for new bona fide Cervix cancer cell lines. Full exome sequencing has been done on 6 cell lines so far with 6 recurrent mutations emerging so far.

Interesting questions as to where HPV inserts and which genes it might sit next to/activate/inhibit etc.

Samples are take both FFPE and fresh/frozen. Looking for more centres - only 6 patients recruited and need 700 in the next 2 years.....

Paul Speiser.
Uterine lavage update.
STICs exfoliate cells very readily, so it might be possible to find them in the uterus by lavage.
Two studies - LUDOC - in women suspected as having serous pelvic cancer, and LUSTIC - in women with known germline mutations undergoing risk-reducing BSO.

Technique is to inject and simultaneously aspirate 10ml N/Saline into the uterine cavity. Initial analysis - droplet PCR for TP53 and KRAS and then NGS for 25 genes.
In first 8 patients who were shown to have either p53 or KRAS mutation in droplet PCR 7 were shown to have other mutation by NGS in first study. Then did further 23, in which 23 had mutations identified, with the inevitable variability in allele frequency. V good correlation between droplet PCR and NGS (Illumina)

LUSTIC will require 200 germline carriers in the expectation of finding 20 STICs.

James Brenton
First description of ITH in breast cancer came in 1978.
See Mittermeyer et al PLoS One 2013. Crowley Nat Rev Clin Oncol 2013 10:472
Question: can you quantitate ITH? Shorter, wider branches on phylogenetic trees are associated with worse outcome than long, narrow branches.
Mutant p53 allele fraction can reach 30% in ctDNA