Hypothesis: **Centrosome amplification leads to changes in the expression pattern of tight junction protein and adhesion molecule genes.**

Centrosome amplification (cell containing supernumerary or ≥3 centrosomes/cell) has long been known as a hallmark of solid tumours in human and as being associated with high- grade tumours with high levels of genomic instability. Addressing the proposed objectives would provide more information on the signalling pathways stimulated by CA and possible links with the pathways which result in a localised (in situ) tumour becoming invasive and travelling outside of the original tumour site (metastasis).

The aim of the proposed study is to find an association between the gene expression of centrosomal genes (focusing on genes associated with centrosome amplification, including a validated CA20 panel) and tight junction (TJ) protein and adhesion molecule genes (and signalling pathways) in breast cancer.

**Research proposal:**

Define the genes involved in each pathway using validated, accepted gene signatures:

The GO sets can be found at Gene Ontology (GO): <http://geneontology.org/page/go-enrichment-analysis>

* These will need to be defined for both CA and TJ pathways prior to any analysis/association study.

i.e <http://amigo.geneontology.org/amigo/search/ontology?q=centrosome>

Select previously completed gene expression experiments in cell lines (make sure to include multiple cancer cell lines, controls and possible relevant treatments):

* EMBL: <https://www.ebi.ac.uk/services/gene-expression>
* cBioPortal may also be useful, http://www.cbioportal.org/

Once a specific defined set of associated CA and TJ genes is identified in cell lines, validate using expression of the genes in human breast cancer samples:

* TCGA: https://portal.gdc.cancer.gov/

Main Research aims:

1. To define/refine a set of CA and TJ gene expression profiles.
2. To discover any association between the CA and TJ gene expression sets in breast cancer cell lines, creating a CA/TJ profile.
3. To validate the associated CA/TJ expression profile in clinical breast cancer expression profiles.

Secondary aims:

1. Determine if there is any association of the CA/TJ profile in clinical samples with:
   1. Subtype
   2. Overall survival
   3. Relapse rate
2. To link the expression profile associated with CA (CA20) with alterations in gene expression patterns of Tight junction/Adhesion Molecule genes and signalling associated pathways in breast cancer.
3. To identify the possible signalling pathways connecting the genes from both CA and TJ gene sets.

A preliminary data set search used a panel of 30 CA genes [**CA30**] (selected by Anu Prakash based alteration in expression in ≥3% of patients from cBioportal database) and a **TJ15** profile (Tight junction /Adhesion molecule genes selected as showing an alteration in expression in ≥3% of patients from cBioportal database). This identified some interacting gene partners and possible pathways by STRING and Reactome pathway analysis showing some potential connection with CA30 and TJ15. Lack of bioinformatics expertise meant this analysis was put on hold.

There also exists a CA20 gene signature which was validated in *Ogden et al., 2017 PMID:28325915*.

**Gene expression profiles for reference:**

|  |  |
| --- | --- |
| **Centrosome genes (CA30 panel)** | |
|  | |
| PLK4 |  |
| SASS6 |  |
| STIL |  |
| AURKA |  |
| NEK2 |  |
| NEK7 |  |
| PLK1 |  |
| CCP110 |  |
| CCSAP |  |
| MCPH1 |  |
| CEP112 |  |
| CEP131 |  |
| CEP170 |  |
| CEP19 |  |
| CEP250 |  |
| NUMA1 |  |
| PTEN |  |
| PIK3CA |  |
| KIF2B |  |
| CCND1   |  | | --- | | CENPF | | TUBD1 | | NUF2 | | CSPP1 | | DCTN6 | | MAP10 | | ASPM | | NDRG1 | | AXIN2 | | MDM2 | |  |

|  |  |
| --- | --- |
| **Tight junction and associated protein (TJ15 panel)** | |
|
| OCLN | (Occludin) |
| MPDZ | Multiple PDZ Domain Crumbs Cell Polarity Complex Component |
| TJP1 | (ZO-1) |
| CGN | (Cingulin) |
| CTTN | (Cortactin) |
| ACTN2 | (actinin alpha 2) |
| CTNNB1 | ( Catenin Beta 1) |
| CRB1 | (crumbs 1) |
| F11R | (JAM-A) |
| ICAM2 | (Cell Adhesion Molecules2) |
| CLDN9 | (Claudin 9) |
| CLDN6 | (Claudin 6) |
| CLDN11 | (Claudin 11) |
| PECAM1 | Platelet And Endothelial Cell Adhesion Molecule 1 |
| PARD3 | Par-3 Family Cell Polarity Regulator |

|  |  |
| --- | --- |
| **Centrosome genes (CA20 panel)** | |
|  | |
| *AURKA* | Aurora Kinase A |
| *CCNA2* | Cyclin A2 |
| *CCND1* | Cyclin D1 |
| *CCNE2* | Cyclin E2 |
| *CDK1* | Cyclin Dependent Kinase 1 |
| *CEP63* | Centrosomal Protein 63 |
| *CEP152* | Centrosomal Protein 152 |
| *E2F1* | E2F Transcription Factor 1 |
| *E2F2* | E2F Transcription Factor 2 |
| *LMO4* | LIM Domain Only 4 |
| *MDM2* | MDM2 Proto-Oncogene |
| *MYCN* | MYCN Proto-Oncogene, BHLH Transcription Factor |
| *NDRG1* | N-Myc Downstream Regulated 1 |
| *NEK2* | NIMA Related Kinase 2 |
| *PIN1* | Peptidylprolyl Cis/Trans Isomerase, NIMA-Interacting 1 |
| *PLK1* | Polo Like Kinase 1 |
| *PLK4* | Polo Like Kinase 4 |
| *SASS6* | SAS-6 Centriolar Assembly Protein |
| *STIL* | STIL, Centriolar Assembly Protein |
| *TUBG1* | Tubulin Gamma 1 |