Introduction

Materials and Methods

The Cancer Genome Atlas (TCGA) is a collaboration between the National Cancer Institute (NCI) and the National Human Genome Research Institute (NHGRI) that has generated comprehensive, multi-dimensional maps of the key genomic changes in 33 types of cancer. It is an open source data repository that holds genomic data (mutations, expression quantification, copy number variation) helps the cancer research community to improve the prevention, diagnosis, and treatment of cancer. In 2012, TCGA and other similar NIH funded open source projects were migrated to a single portal, the GDC data portal.

The NCI's Genomic Data Commons (GDC) provides the cancer research community with a unified data repository that enables data sharing across cancer genomic studies in support of precision medicine.

A cancer is called estrogen-receptor-positive (or ER+) if it has receptors for estrogen. This suggests that the cancer cells, like normal breast cells, may receive signals from estrogen that could promote their growth. Testing for hormone receptors is important because the results help you and your doctor decide whether the cancer is likely to respond to hormonal therapy or other treatments. Hormonal therapy includes medications that either (1) lower the amount of estrogen in your body or (2) block estrogen from supporting the growth and function of breast cells. If the breast cancer cells have hormone receptors, then these medications could help to slow or even stop their growth. If the cancer is hormone-receptor-negative (no receptors are present), then hormonal therapy is unlikely to work.

Cytokines are any of a number of substances, such as interferon, interleukin, and growth factors, that are secreted by certain cells of the immune system and have an effect on other cells.

Using various tools (Firehose, TCGABiolinks, RTCGA Toolbox) we downloaded breast cancer datasets with clinical and genetic information from the TCGA/GDC portal into R.

How we processed

Our analysis tool was R studio

Understanding the TCGA dataset (1098 rows, 1948 variables)

TCGA Barcodes, Patients, Samples, Aliquotes

Normal Tissue vs Primary Tumor

Estrogen receptor positive vs negative

Cytokines involved in Breast Cancer

Microarray vs RNASeq gene expression quantification data

We sepa

How we analyzed- Analysis tools in R

Limma

TCGA

Deseq

String

How we got our results

Er + tumor vs normal

Er – tumor vs normal

Normal vs tumor

Er + vs Er -

Matched Er + tumor vs normal

Matched Er - tumor vs normal

Microarray vs RNAseq

Top Genes by logFC, p-value

**Results**

Tables

|  |  |  |
| --- | --- | --- |
| **GLOBAL** | **Up-regulated** | **Down-regulated** |
| ER-positive Tumor vs Normal |  |  |
| ER-negative Tumor vs Normal | 2261 | 2410 |
| Normal vs Tumor |  |  |
| ER positive and ER negative | 7493 | 6572 |
|  |  |  |
| **CYTOKINES** |  |  |
| ER-positive Tumor vs Normal |  |  |
| ER-negative Tumor vs Normal |  |  |
| Normal vs Tumor |  |  |
| ER positive and ER negative |  |  |
|  |  |  |

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Global** |  | **Up-Reg** |  | **Down-Reg** |
| **Top Tables** | **Gene Names** | **LogFC** | **Gene Names** | **LogFC** |
| ER-positive: Tumor VS normal: | GPI  GRM4  ITPK1  ITPKB  TMEM206 | 9.170819  8.708092  8.349087  8.319999  8.262449 | GZMB  AGFG2  AHCY  C2CD3  ITIH4 | -7.101645  -6.507704  -6.452276  -6.214164  -5.924074 |
| ER-Negative:  Tumor vs Normal: | IQCF3  PHKG2  PLEKHB1  PLEKHH1  AGT | 9.470106  9.276100  8.782796  8.501724  8.174082 | EPHB2  PRSS48  TYMS  CHST12  RAB11FIP4 | -6.995358  -6.951212  -6.618847  -6.220915  -6.149925 |
| RNASeq:  Matched  Tumor vs Normal | TMEM206  IGDCC3  FAM13B  ITPK1  FAM161A  HOXC8  SLC6A19  ITLN1  SFRS3  GRM4 | 8.958426  8.686416  8.276534  8.267515  8.108979  7.358470  7.267708  6.957867  6.891656  6.835357 | ISLR2  ISL1  ITIH4  CDC42BPG  GZMB  ERMAP  USP31  SUDS3  EFEMP2  SBF1 | -9.898563  -9.481294  -8.896361  -8.678778  -8.095539  -7.569973  -7.555336  -7.364178  -7.283555  -7.020664 |
| Microarray:  Matched Tumor vs Normal | COL11A1  COL10A1  GJB2  PPAPDC1A  MMP13  MMP11  NEK2  CEACAM6  CEP55  S100P | 5.154717  4.529933  3.934013  3.814097  3.758533  3.467785  3.288261  3.268321  3.202401  3.073844 | ANGPTL7  ATP1A2  PCOLCE2  SFRP1  CA4  FIGF  SDPR  CXCL2  NPY2R  G0S2 | -4.335967  -4.173679  -4.139050  -4.132469  -4.003679  -3.957979  -3.879663  -3.875834  -3.769950  -3.728704 |
| RNASeq  ER+ vs ER- | MYL1  CARTPT  CYP2A7  CSRP3  MYH2  CYP2A6  MYL2  XIRP2  UCN3  CPB1 | 8.907592  8.400998  8.100730  7.994015  7.724777  7.645191  7.625013  7.403499  7.186851  7.185329 | SMR3B  SMR3A  HTN1  STATH  PRM1  REG1A  NOBOX  C20orf185  C4orf7  TAAR6 | -9.248781  -8.864729  -7.663618  -6.703072  -6.546426  -6.258131  -6.127830  -6.106026  -6.005504  -5.979640 |

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Cytokines** |  | **Up-Reg** |  | **Down-Reg** |
|  | **Gene Names** | **LogFC** | **Gene Names** | **LogFC** |
| ER-positive: Tumor VS normal: | IFNB1  IL12B  IFNG  IL18 | 2.77619627  1.26491838  1.05085123  0.79361053 | IFNA1  IL2  IL12A  IL13  IL6 | -1.19165485  -1.31264485  -1.44218279  -1.96254142  -3.11672893 |
| ER-Negative:  Tumor vs Normal: | IFNB1  IL12RB2  IL8  IFNG  IL2RA  IL12B  IL18 | 4.3691031  3.0976924  1.9669441  1.9607590  1.5673409  0.8629049  0.7919899 | IL2  IL13  IL4  IL4.1  IL6 | -0.9080108  -1.0751707  -1.5729367  -1.5729367  -2.7294322 |
| RNASeq:  Matched  Tumor vs Normal | IFNB1  IL12RB2  IFNG  IL12B  IL18  IL2RA  IL8 | 3.1737071  1.4720836  1.4633940  1.2285418  0.8687162  0.7188611  0.5734657 | IL1B  IL12A  IL4  IFNA1  IL2  IL13  IL6 | -0.6593007  -0.8978855  -0.8984633  -1.0650591  -1.1344595  -1.6109529  -2.9609044 |
| ER positive vs ER negative | IL4  IL13RA1  IRF2 | 0.5988642  0.3140835  0.1530753 | IL13  IFNB1  IL12A  IL2RA  IFNG  IL8  IL13RA2  IL12RB2 | -0.7274415  -0.9892813  -1.2394836  -1.4542249  -1.4639946  -1.7635835  -1.8570144  -2.9661638 |

**Discussion**

Global

ER-positive: Tumor VS normal: Comparing normal as reference to ER+ tumors, thousands of genes were differentially expressed. Top positive and negative are presented.

ER-Negative: Tumor vs Normal: Comparing normal as reference to ER- tumors, thousands of genes were differentially expressed. Top positive and negative are presented.

Further analysis of this group of genes should identify genes responsible for breast cancer.

**Matched samples**

RNASeq: Matched Tumor vs Normal: Comparing tissue samples from the same women, we found 97 matches who had RNAseq data and 60 matches with Microarray data. Using their normal as reference to Tumors, thousands of genes were differentially expressed. Top positive and negative are presented.

Results gotten from RNASeq were a bit different, maybe better.

ER + vs ER –

Using ER- as reference against ER+ve tumor cells, we found top up/down regulated genes. Further analysis of this group may reveal differences between ER+ and ER- breast cancers

Cytokines

We narrowed down our analysis to the investigate the influence of Cytokines on breast cancer

ER-positive/ER-negative: Tumor VS normal:

The striking similarity between both groups show consistently that these cytokines are definitely involved in breast cancer. Some are consistently up/down regulated irrespective of breast cancer receptor type

RNASeq: Matched Tumor vs Normal:

Analysis of matched samples from the same woman were also consistent with our initial findings of cytokines involved in the body’s response to breast cancer.

ER+ vs ER -:

This gives a slightly different picture and shows what cytokines might be more active in ER+ or ER- breast cancers.

**Conclusions**

Our study showed top up/down regulated in breast cancer patients and differential expression between 4 sub-groups of breast cancer patients. From the global group, further analysis is necessary to filter genes up/down regulated due to other factors. The cytokines subset shows clearly which cytokines are more active in ER+/ER- breast cancer. Further research is necessary to determine if these could be useful in the management or treatment of ER+/ER- breast cancer patients.