PathFX: Applications on Breast Cancer

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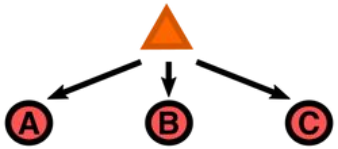
C&S BIO 185

Thesis Research in Computational and Systems Biology

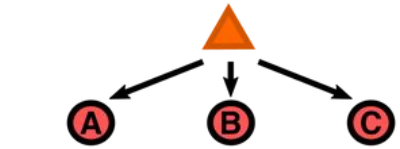
What is PathFX?

- Developed by Dr. Jennifer Wilson, professor of Bioengineering, at UCLA
- Non-commercial, cost-effective software and algorithm
- Identifies drug pathways and drug-related phenotypes by compiling and analyzing protein-protein and drug-protein interaction networks



} user specifies a drug

} (optional) user specifies drug targets



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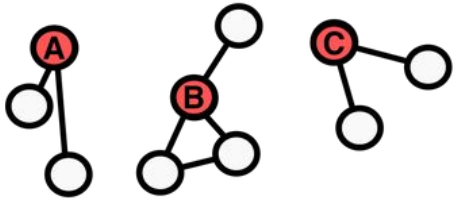
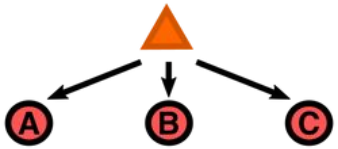


} PathFX discovers the relevant network around each target



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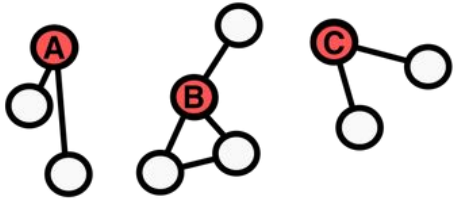

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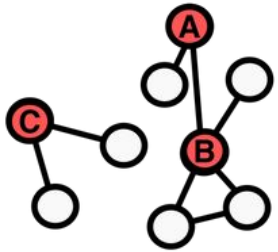

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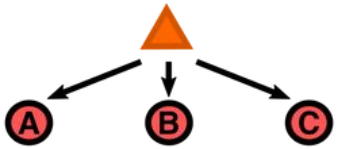

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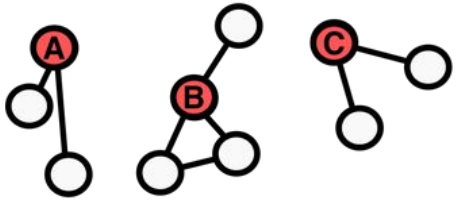

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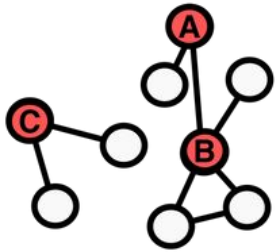

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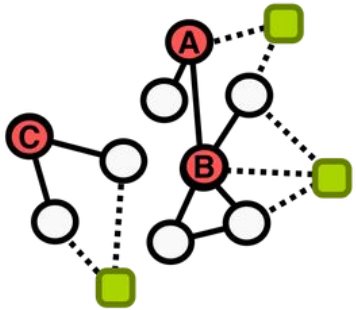


} PathFX identifies phenotypes associated with the genes in the merged network

Hypothetical Situation:

- Patient A has a rare life threatening disease.
- There are two drugs that are known to treat this disease.
- However, Patient A cannot use either of these drugs because they have another condition that will worsen if these drugs are used.
- How can we find a treatment option for this patient?
  - **Option 1:** Conduct expensive and time consuming clinical trials to find a new treatment for this patient.
  - **Option 2:** Find out if other already made drugs can potentially be repurposed to treat the patient to some extent.

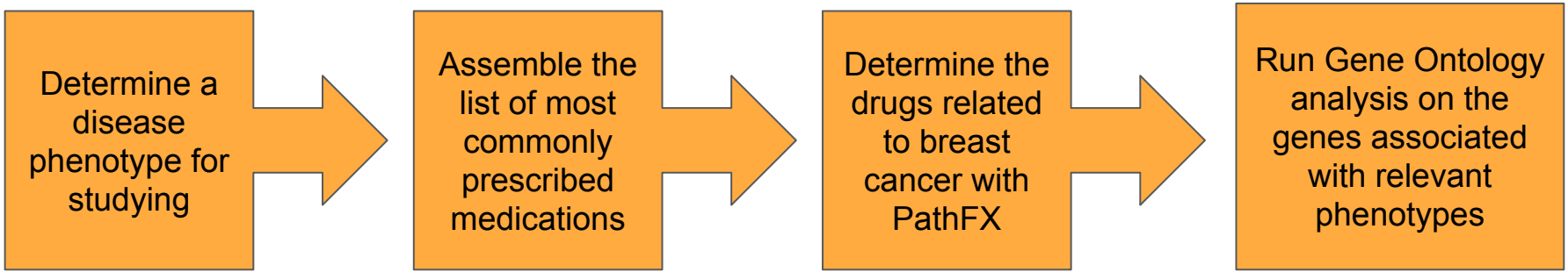
Question and Goals

Question: Can we repurpose the top 5 most commonly prescribed cardiovascular, endocrine, respiratory, and gastrointestinal drugs for breast cancer related purposes?

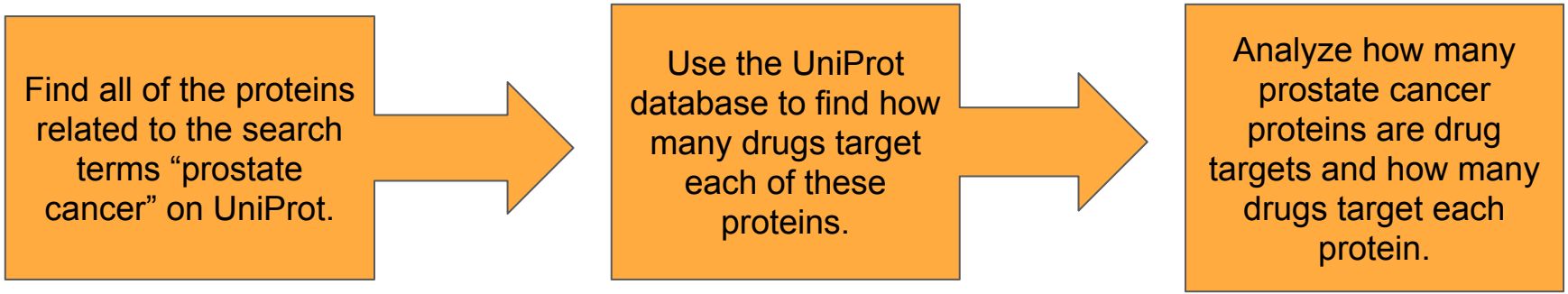
Goals:

(1) **Identify** factors that may influence the number of drugs that will target a phenotype.
(2) **Discover** which of the top 5 commonly prescribed cardiovascular, endocrine, respiratory, and gastrointestinal drugs can be repurposed for breast cancer related purposes according to PathFX.
(3) **Understand** and **validate** why certain drugs are repurposable for breast cancer purposes through an ontological analysis.

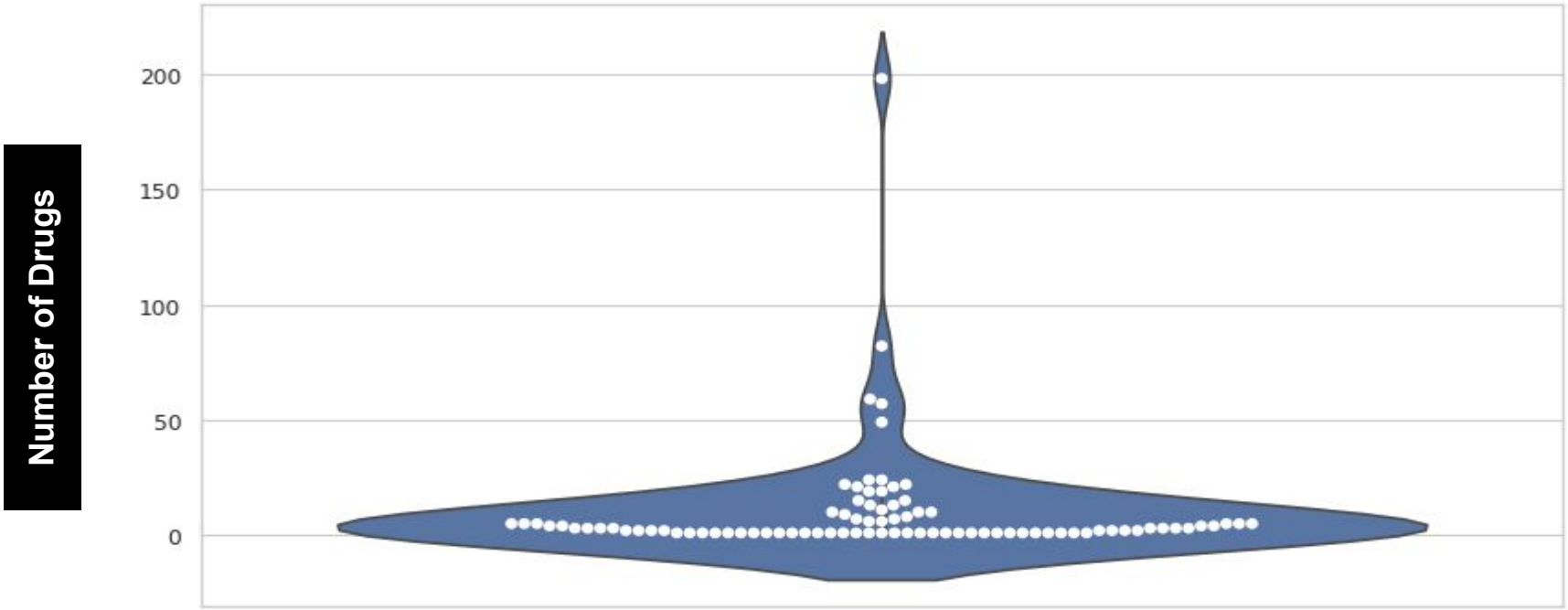
Workflow


Determine a disease phenotype for studying
Assemble the list of most commonly prescribed medications
Determine the drugs related to breast cancer with PathFX
Run Gene Ontology analysis on the genes associated with relevant phenotypes

We had initially wanted to look for drugs that could be repurposed for prostate cancer…

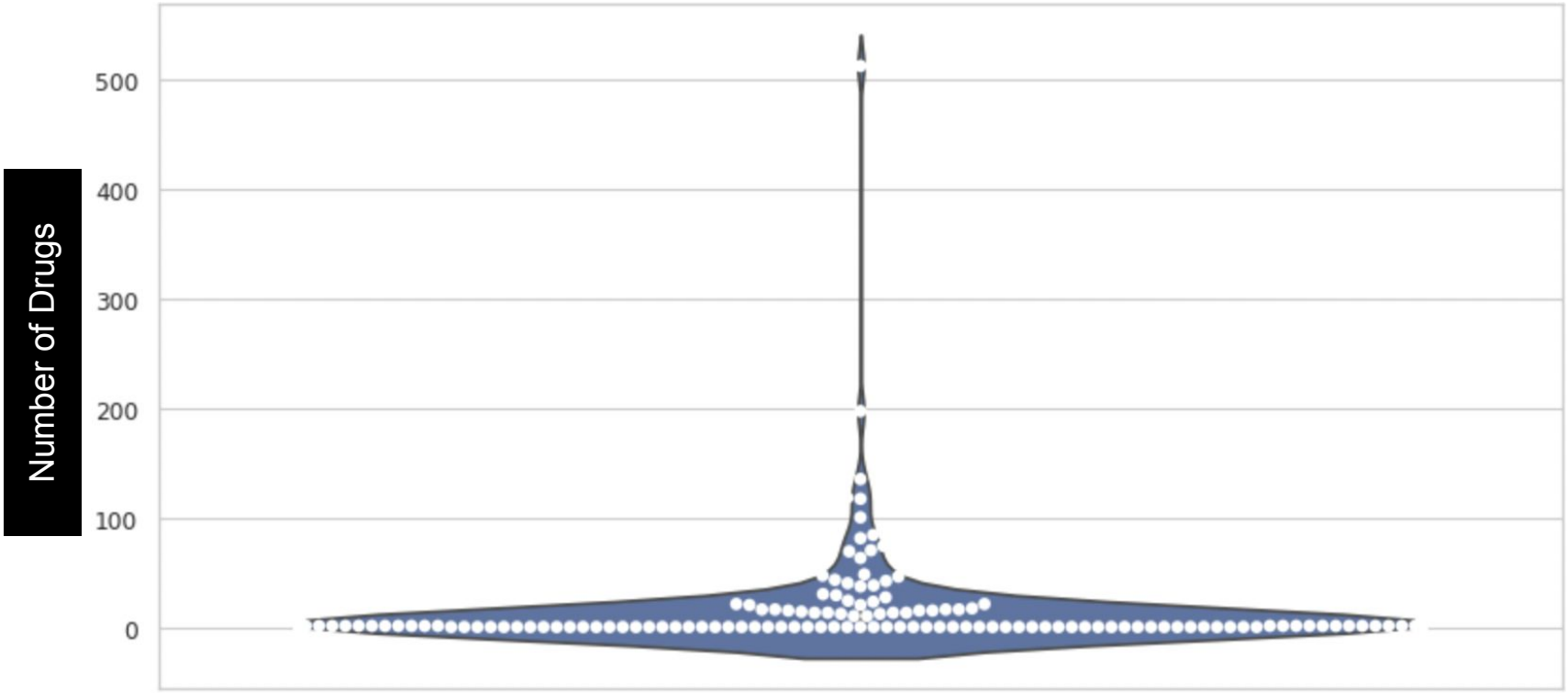


Prostate Cancer Proteins that are Drug Targets and the Number of Drugs that Target Each of them



(Every white point represents a protein that was found to be targeted by at least one drug. The y axis helps to demonstrate how many drugs actually target the protein.)

Breast Cancer Proteins that are Drug Targets and the the Number of Drugs Associated with Each of Them



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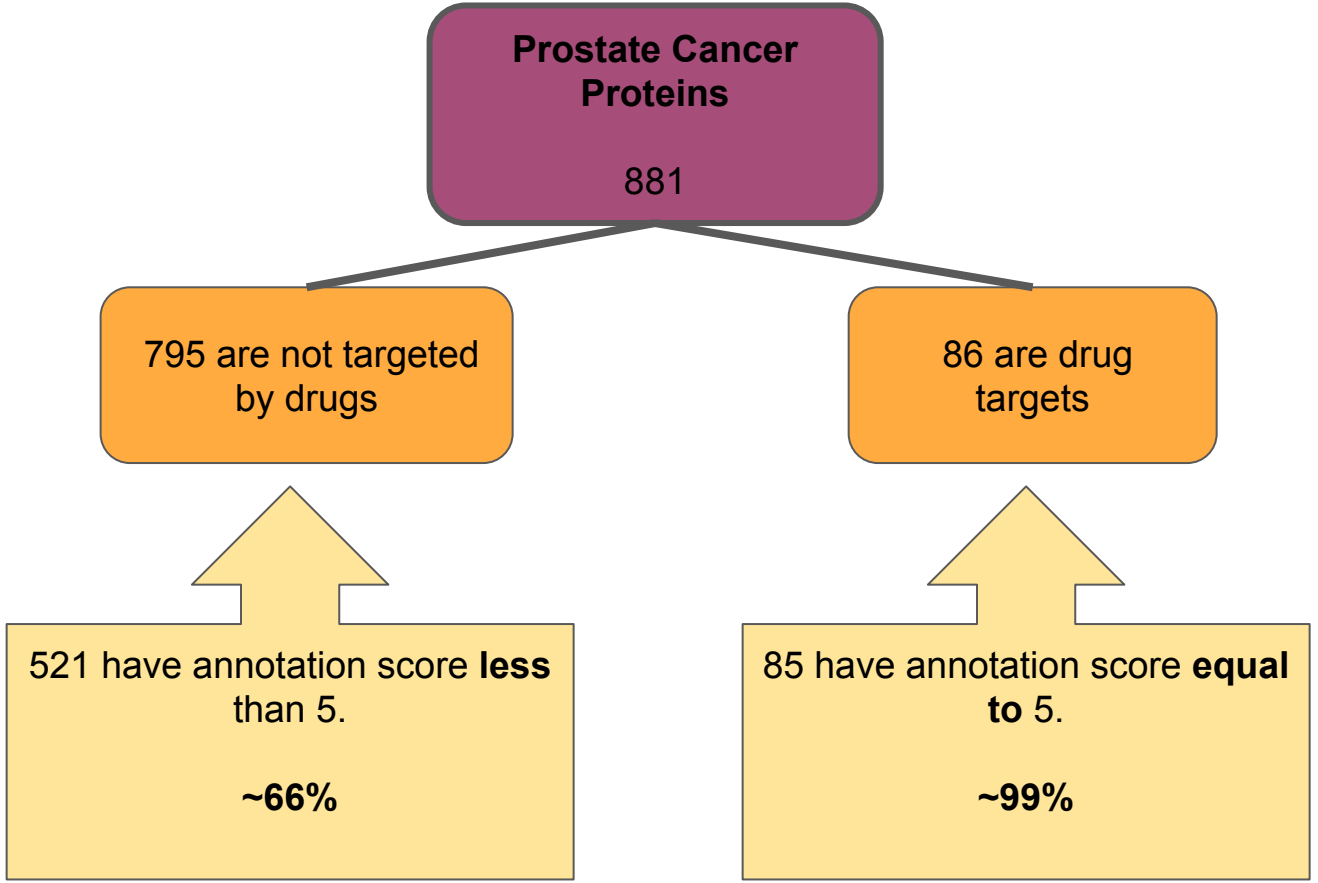
Annotation Score vs. Drug Target Status



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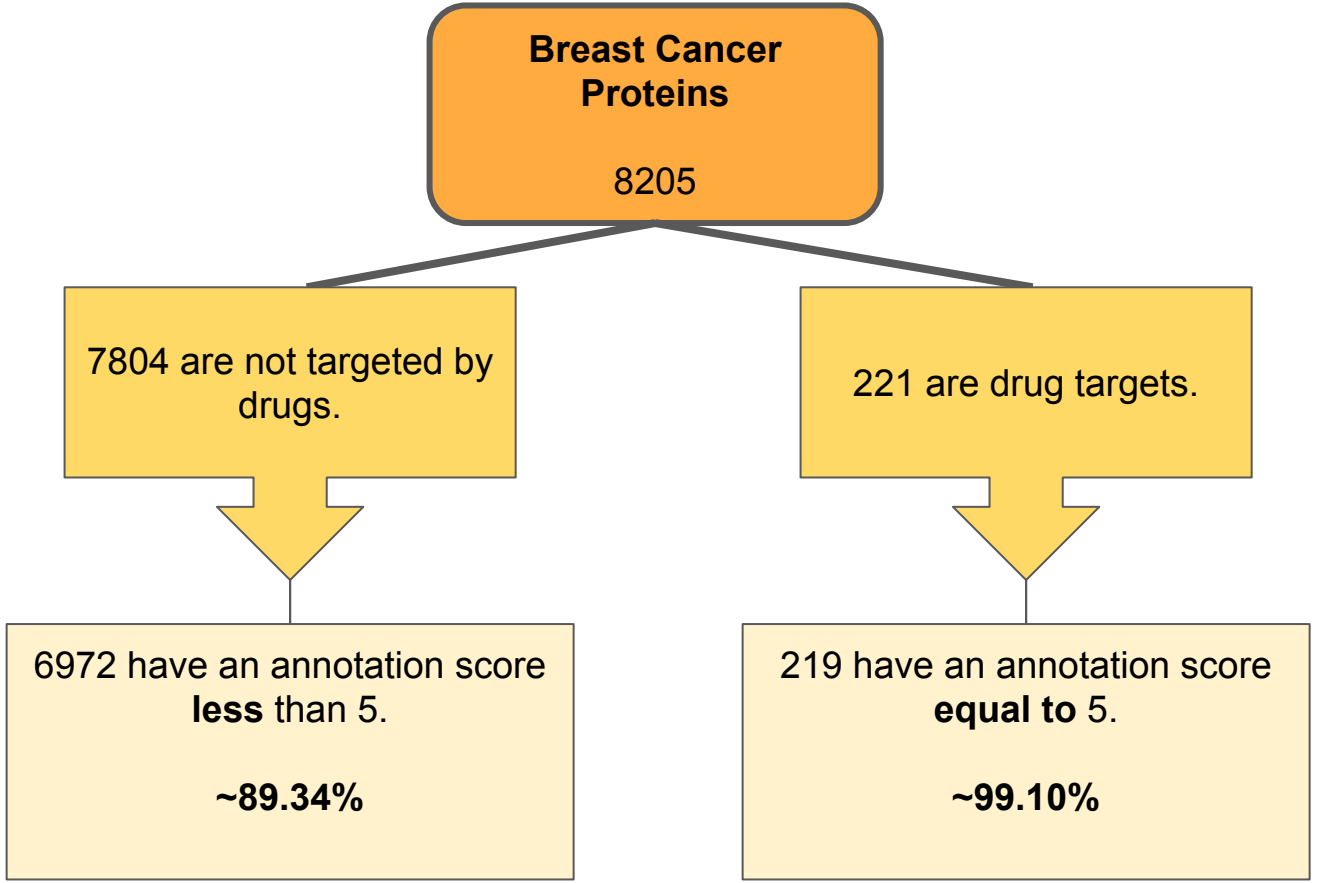


Why did we switch our focus to breast cancer?

Out of the **881** proteins that are related to prostate cancer (source: Uniprot), only **86** of those are targeted by drugs

On the other hand, we found that out of the **8025** proteins that are related to breast cancer, **221** of those are targeted by drugs.

Thus, with a larger number of proteins that are drug targets, we felt that it would be more likely that breast cancer would show up as a phenotype during our analysis.

Additionally…

Breast cancer became the most common cancer globally as of 2021, accounting for 12% of all new annual cancer cases worldwide, according to the World Health Organization.

About 1 in 8 U.S. women (about 13%) will develop invasive breast cancer over the course of her lifetime.

This information is provided by Breastcancer.org.

Most Commonly Prescribed Drugs

Gastrointestinal:

Omeprazole

Pantoprazol

Ranitidine

Famotidine

Esomeprazole

Respiratory:

Albuterol

Montelukast

Fluticasone

Budesonide

Formoterol

Endocrine:

Levothyroxine

Metformin

Prednisone

Finasteride

Prednisolone

Cardiovascular:

Lisinopril

Metoprolol

Amlodipine

Losartan

Hydrochlorothiazide

Breast Cancer Phenotypes

Luminal A Breast Carcinoma

Neoplasm of the Breast

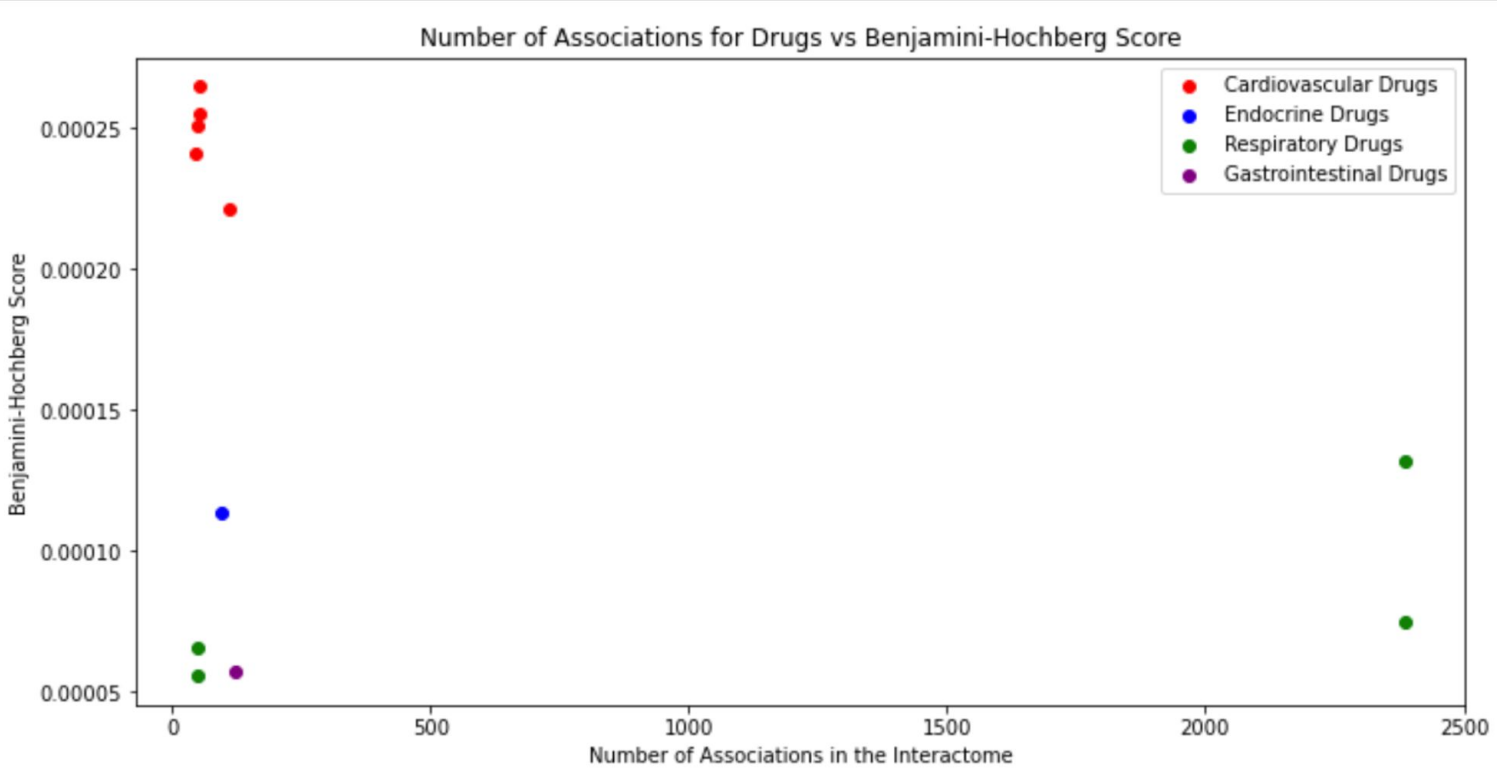
Female Breast Carcinoma

Hereditary Breast and Ovarian Cancer Syndrome

Ductal Breast Carcinoma

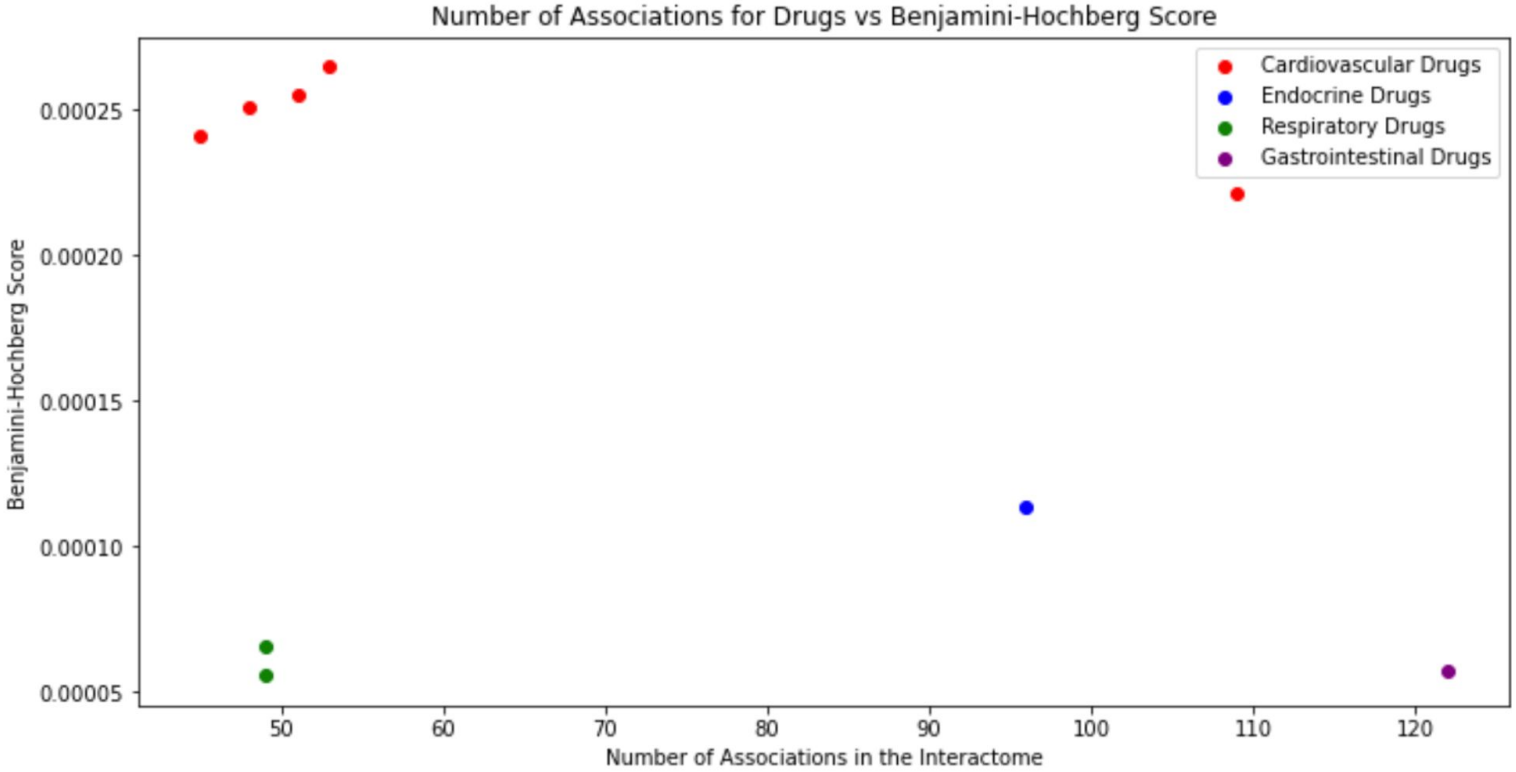
Atypical Ductal Breast Hyperplasia

Number of Associations vs Benjamini-Hochberg Score



Benjamini-Hochberg Score: a method for scoring false discovery rate to help adjust incorrectly small p-values
Number of Associations in Interactome: determining how many interactions are in the network

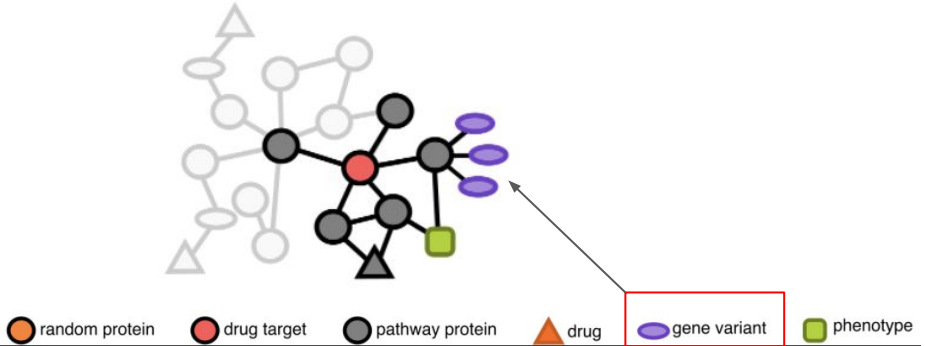
Number of Associations vs Benjamini-Hochberg Score

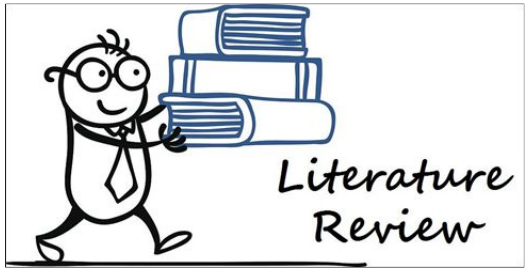


Benjamini-Hochberg Score: a method for scoring false discovery rate to help adjust incorrectly small p-values
Number of Associations in Interactome: determining how many interactions are in the network

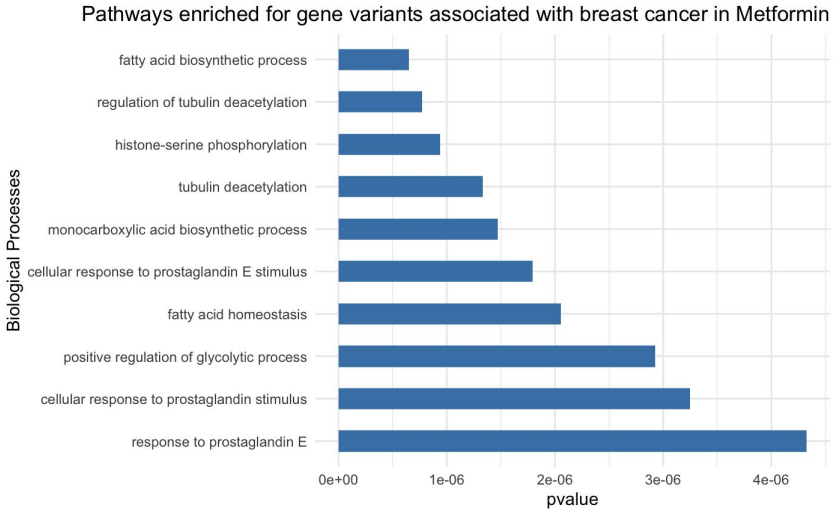
From the PathFX results, we conduct gene ontology analysis on genes associated with breast cancer phenotypes for each drug category.

GO analysis → Literature search on top enriched pathways/functions → Summarize evidence that would support our initial goal of repurposing

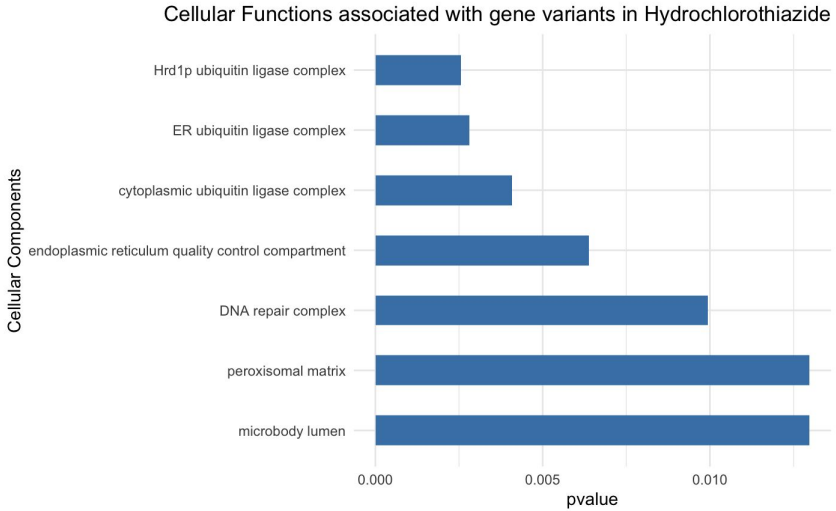




From the GO result, we were able to identify pathways and cellular components that show up repetitively in the analysis…

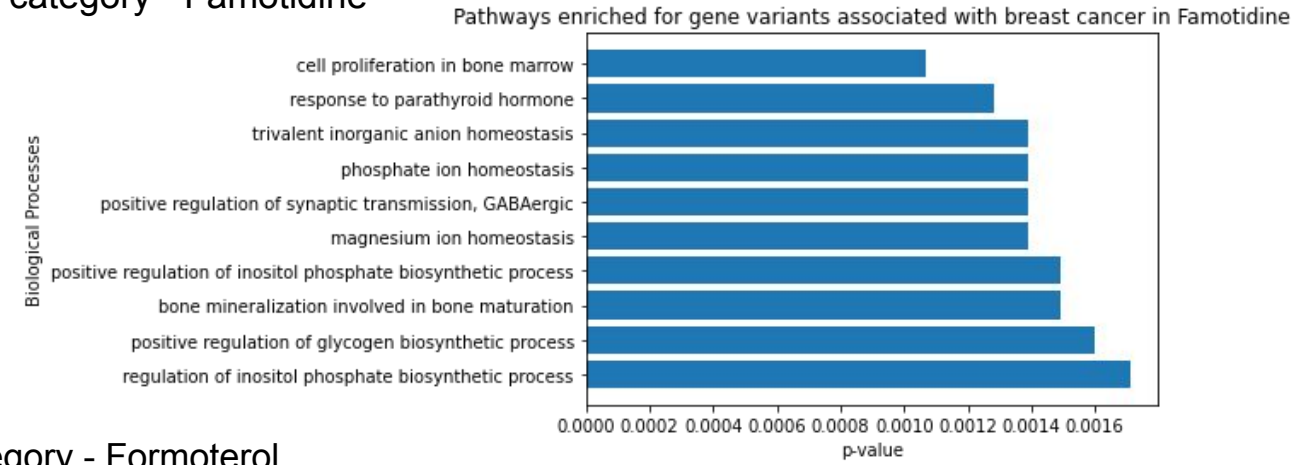


Endocrine category – Metformin

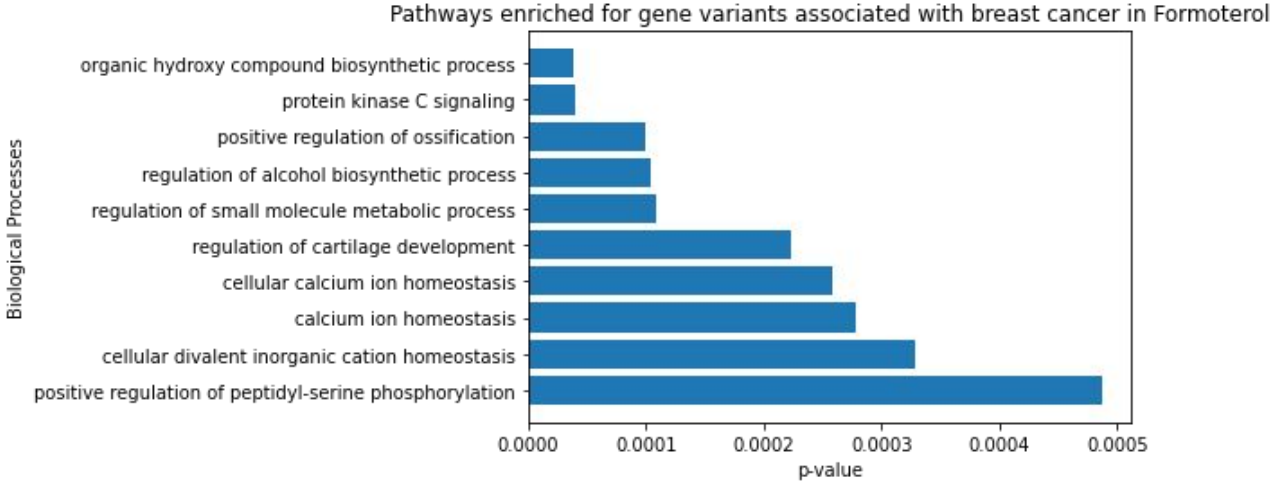


Cardiovascular category – Hydrochlorothiazide

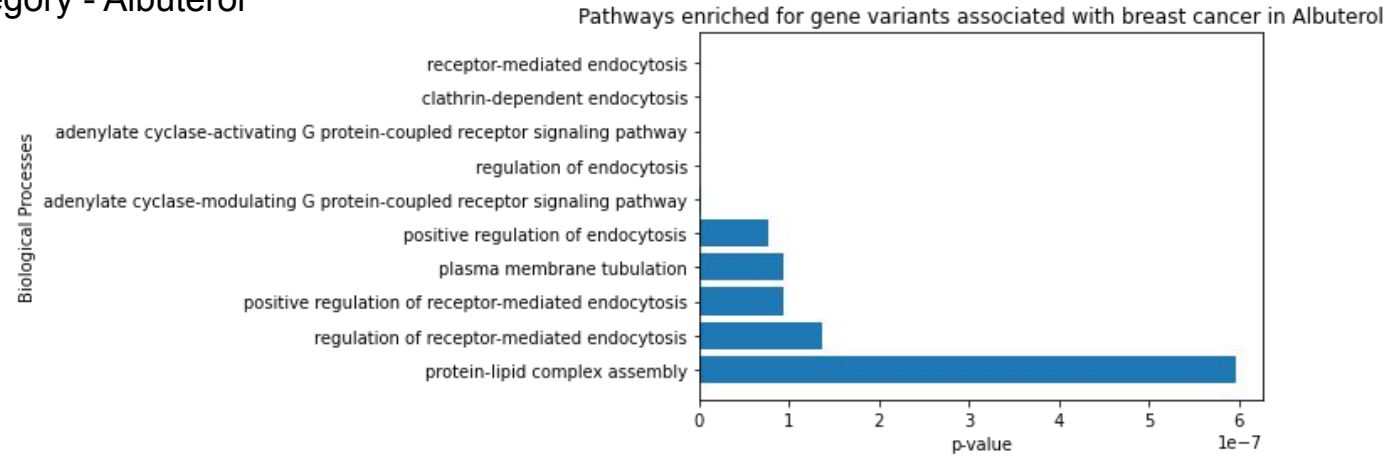
Gastrointestinal category - Famotidine



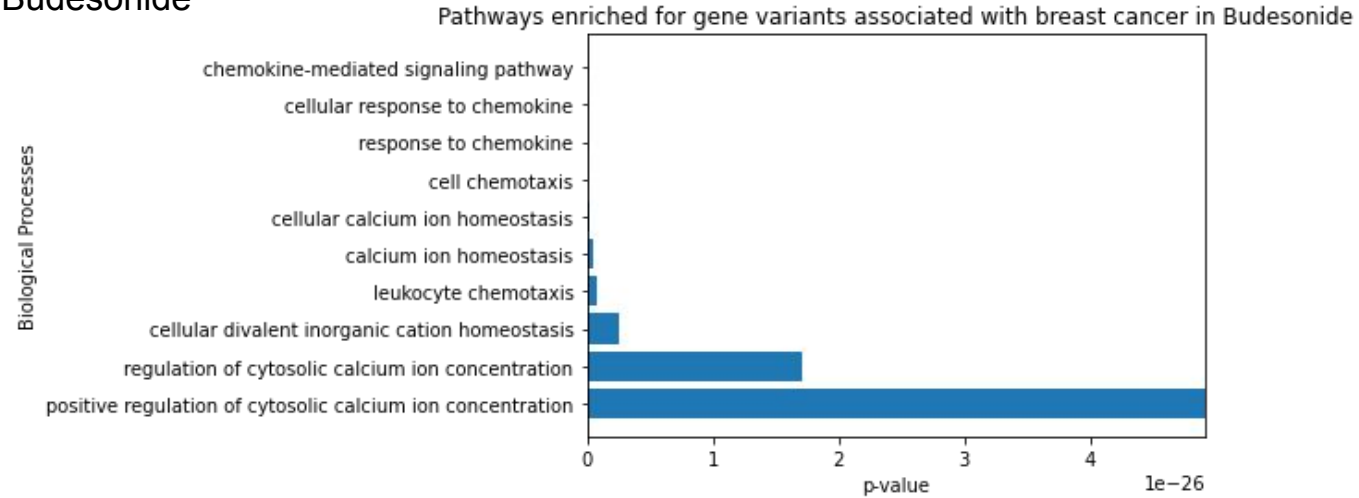
Respiratory category - Formoterol



Respiratory category - Albuterol



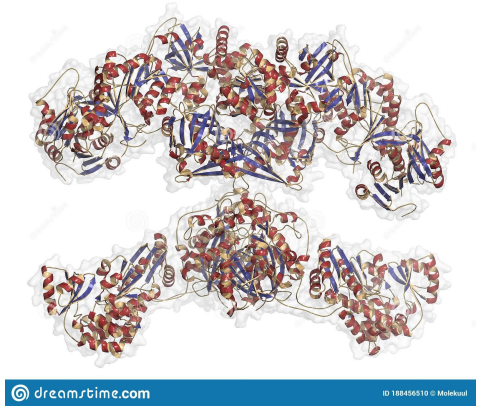
Respiratory category - Budesonide



Further literature search validates the association between identified biological pathways/cellular functions and breast cancer.

Significant pathways in Metformin:

- Fatty acid biosynthesis



[fatty acid synthase 3D structure]

Significant cellular component in Hydrochlorothiazide:

- Ubiquitin ligase complex

many proteins targeted by clinical breast cancer researchers are involved in ubiquitin pathways, including the famous BRCA1-BARD1.

Further literature search validates the association between identified biological pathways/cellular functions and breast cancer.

Significant pathways in Formoterol:

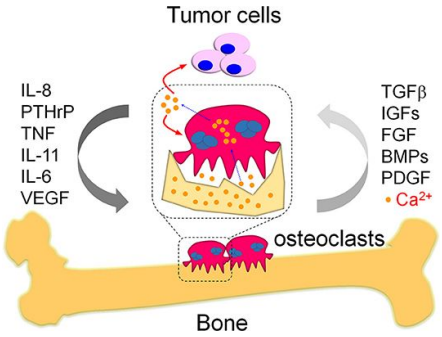
- Protein kinase C signaling

Literature reference:

- Relationship found between PKCα expression and cancer metastasis

Significant pathways in Formoterol and Budesonide:

- Calcium ion homeostasis



Literature reference:

- Relationship found between calcium ion homeostasis and bone metastasis/tumor progression

Limitations

- PathFX is unable to detect the directionality between relationships
  - Manual verification is needed
- PathFX does not consider tissue specificity and is biased to selecting phenotypes with fewer gene annotations
- We only conducted GO analysis on genes found to be associated with breast cancer related phenotypes
- Some literature did not exist for the pathways of interest
- Annotation scores of proteins do not provide a full picture of the extent of research done on a protein, and thus the extent to which a phenotype has been studied

Conclusions and Future Directions

Conclusions:

- PathFX can be reliable for repurposing drugs based on pathway analyses
- Formoterol, metformin, and budesonide showed manually verified literature supporting our claim
- Phenotypes involving more proteins will have a greater chance of being targeted by other drugs

Future work:

- Expanding the number of drugs that were first initially searched
- Running GO analysis on genes associated with other disease phenotypes for each drug
- Finding/developing an even more informative metric to quantify the extent to which a protein or phenotype has been studied

Citations (for literature search)

Ohta, T., Fukuda, M. Ubiquitin and breast cancer. *Oncogene* 23, 2079–2088 (2004). https://doi.org/10.1038/sj.onc.1207371

Reader J, Holt D, Fulton A. Prostaglandin E2 EP receptors as therapeutic targets in breast cancer. Cancer Metastasis Rev. 2011 Dec;30(3-4):449-63. doi: 10.1007/s10555-011-9303-2. PMID: 22002714; PMCID: PMC3640271.

Xu S, Chen T, Dong L, Li T, Xue H, Gao B, Ding X, Wang H, Li H. Fatty acid synthase promotes breast cancer metastasis by mediating changes in fatty acid metabolism. Oncol Lett. 2021 Jan;21(1):27. doi: 10.3892/ol.2020.12288. Epub 2020 Nov 11. PMID: 33240433; PMCID: PMC7681230.

Pham, T.N.D., Perez White, B.E., Zhao, H. et al. Protein kinase C α enhances migration of breast cancer cells through FOXC2-mediated repression of p120-catenin. BMC Cancer 17, 832 (2017). https://doi.org/10.1186/s12885-017-3827-y

Yang Z, Yue Z, Ma X and Xu Z (2020) Calcium Homeostasis: A Potential Vicious Cycle of Bone Metastasis in Breast Cancers. Front. Oncol. 10:293. doi: 10.3389/fonc.2020.00293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