AN INVESTIGATION OF COMMON GENETIC EXPRESSION PATHWAYS FOR TYPE 2 DIABETES AND ALZHEIMER'S IN AFRICAN AMERICANS
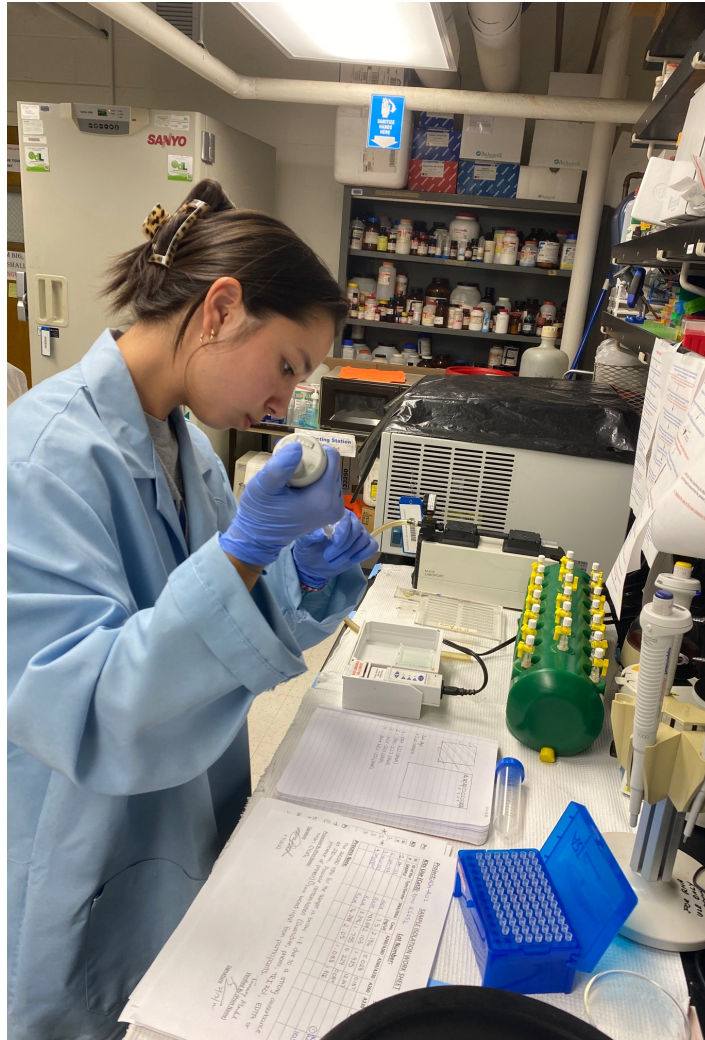
By Marika Clark

Format of my Internship
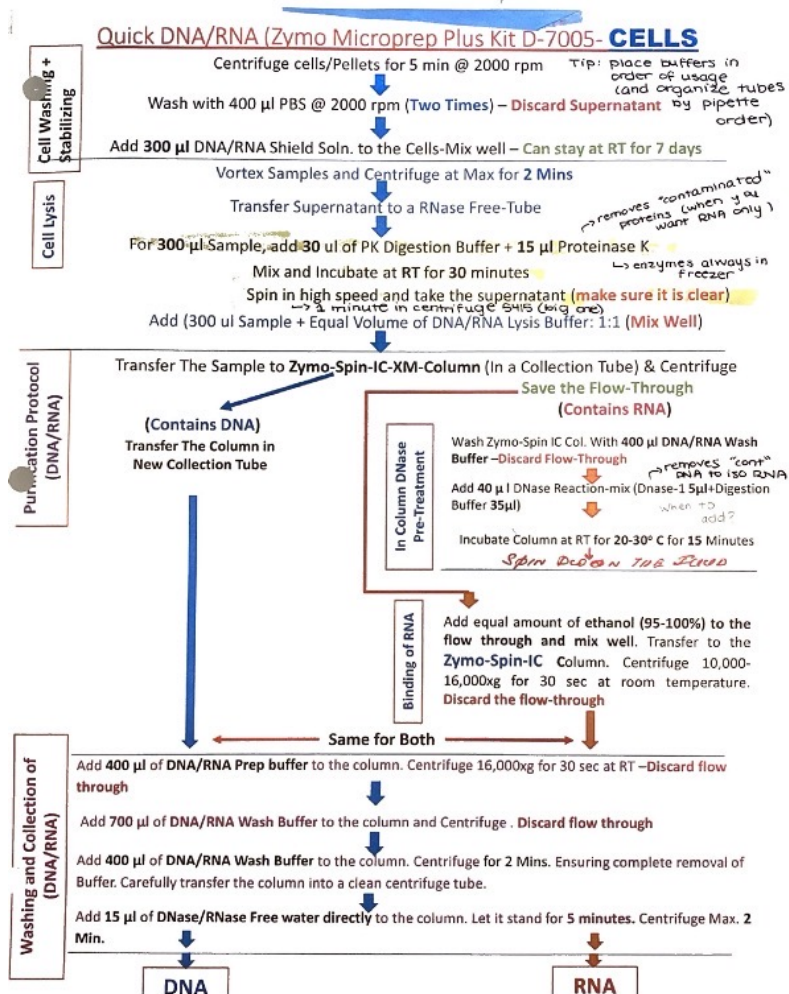
And this presentation!

- Many small projects/skill building
- Mix of Wet-lab and Downstream Analysis (computer work)
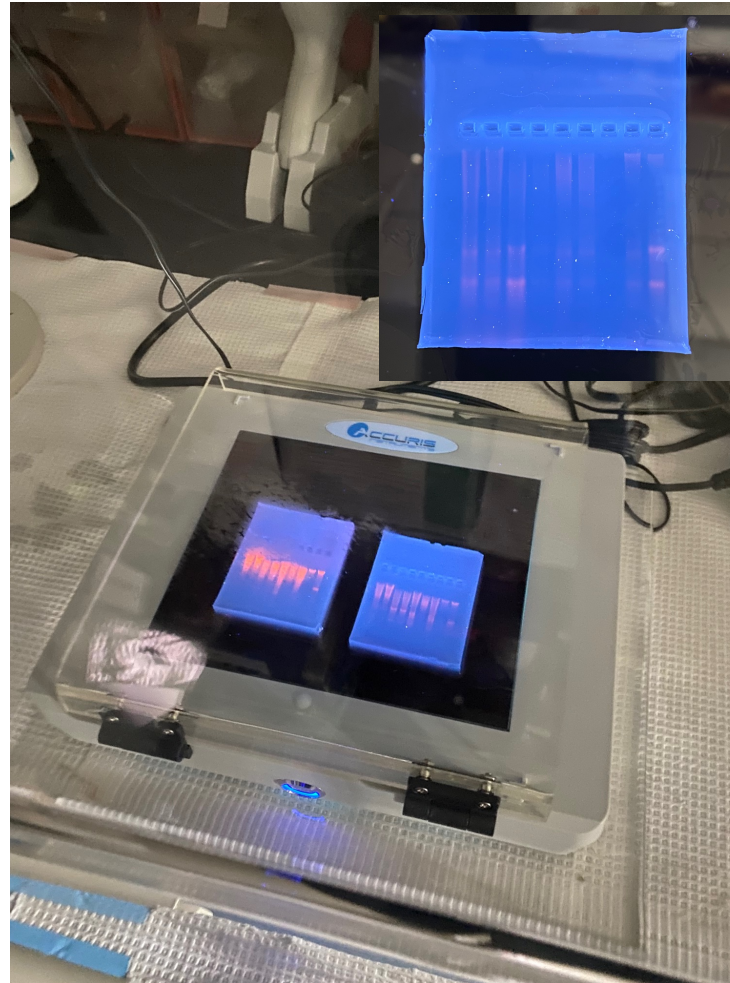- Context in the bigger picture

Bench Work

Quality Control Testing of Nucleic Acids (DNA and RNA from Cell Samples)
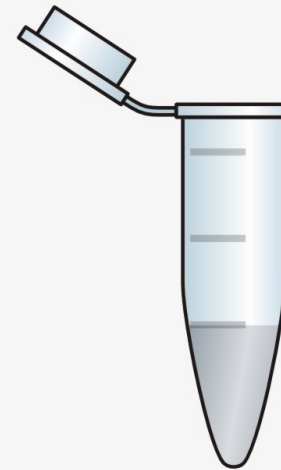

RNA/DNA Isolation

Gel Electrophoresis

NanoDrop Analysis

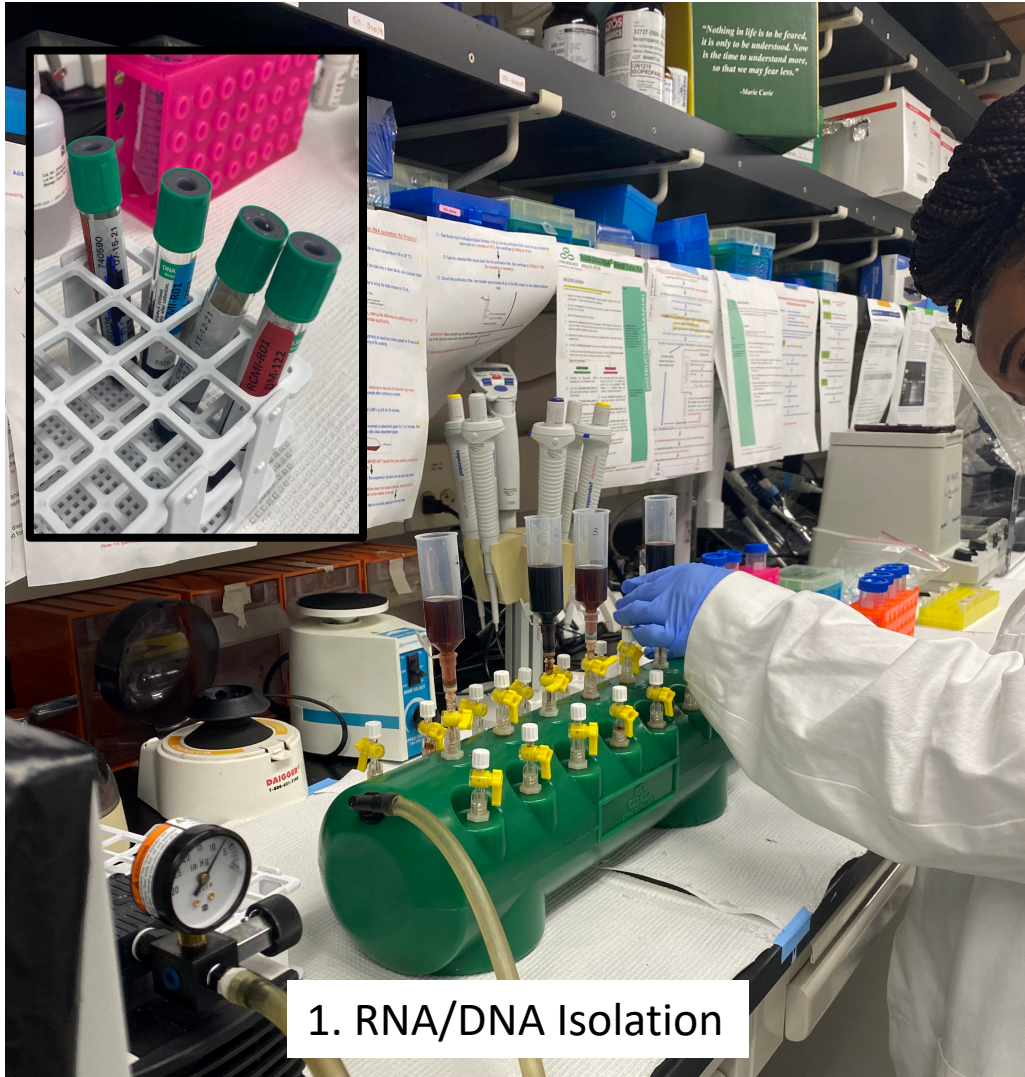# Preparation and Analysis of Blood Samples

1. RNA/DNA Isolation

2. cDNA Prep

3 Main Parts:
1. RNA/DNA Isolation
2. cDNA Preparation
3. qPCR Analysis
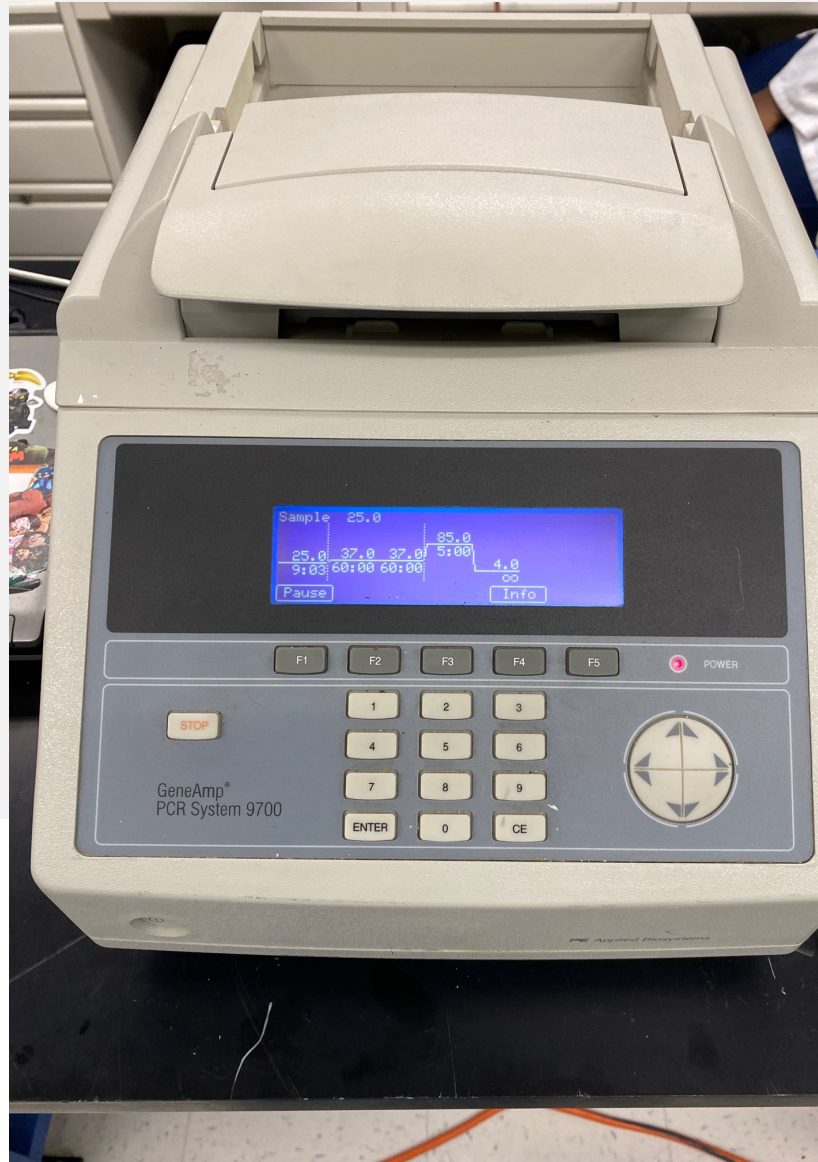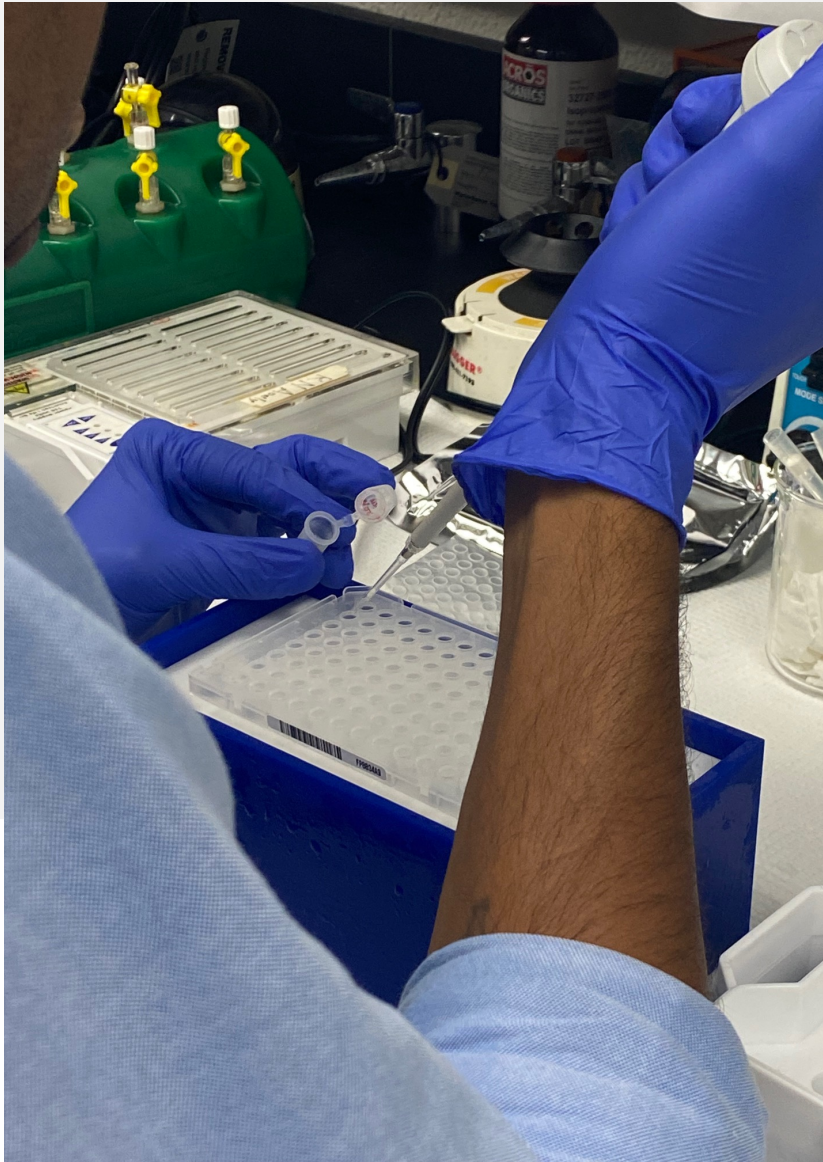
qPCR Analysis

Blood Samples Continued

Gene Expression Data Analysis (in Silico/Computer Work)

Ingenuity Pathway Analysis

- Creates renderings of the genetic relationships and expressions of samples

- Includes information about…
    - Diseases and functions
    - Canonical Pathways
    - Upregulation/Downregulation of genes

  And much more!

Analysis: T2H50PvsCONTROL_DEG.xls for IPA - 2022-07-19 11:48 AM

·T2H50PvsCONTROL_DEG.xls for IPA - 2022-07-19 11:48 AM

Hani-Beebe at Al. 2022
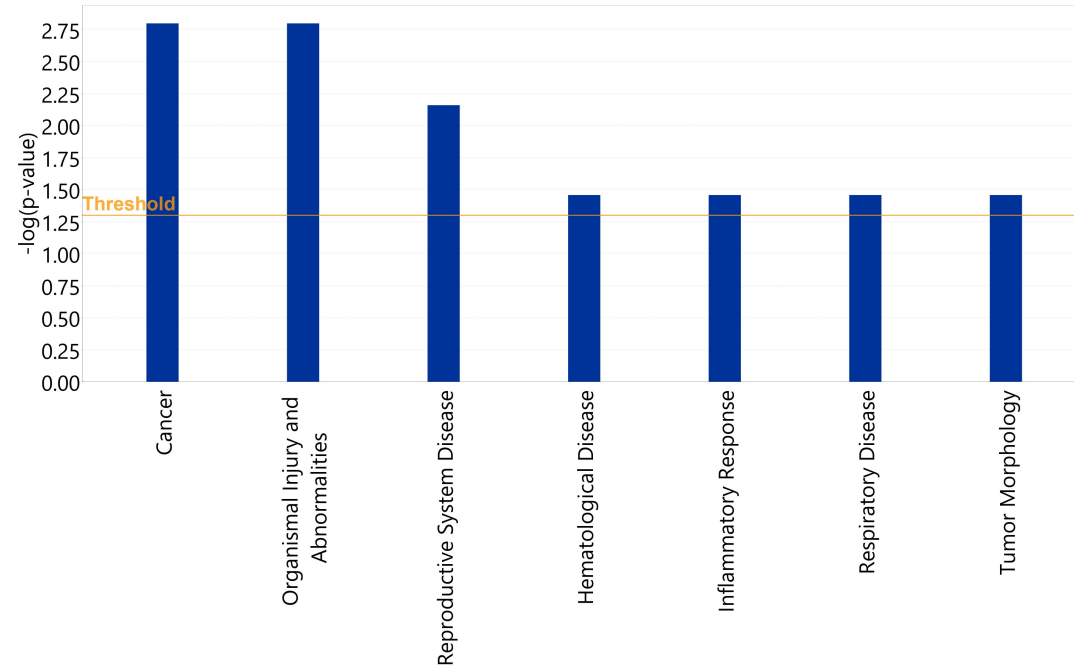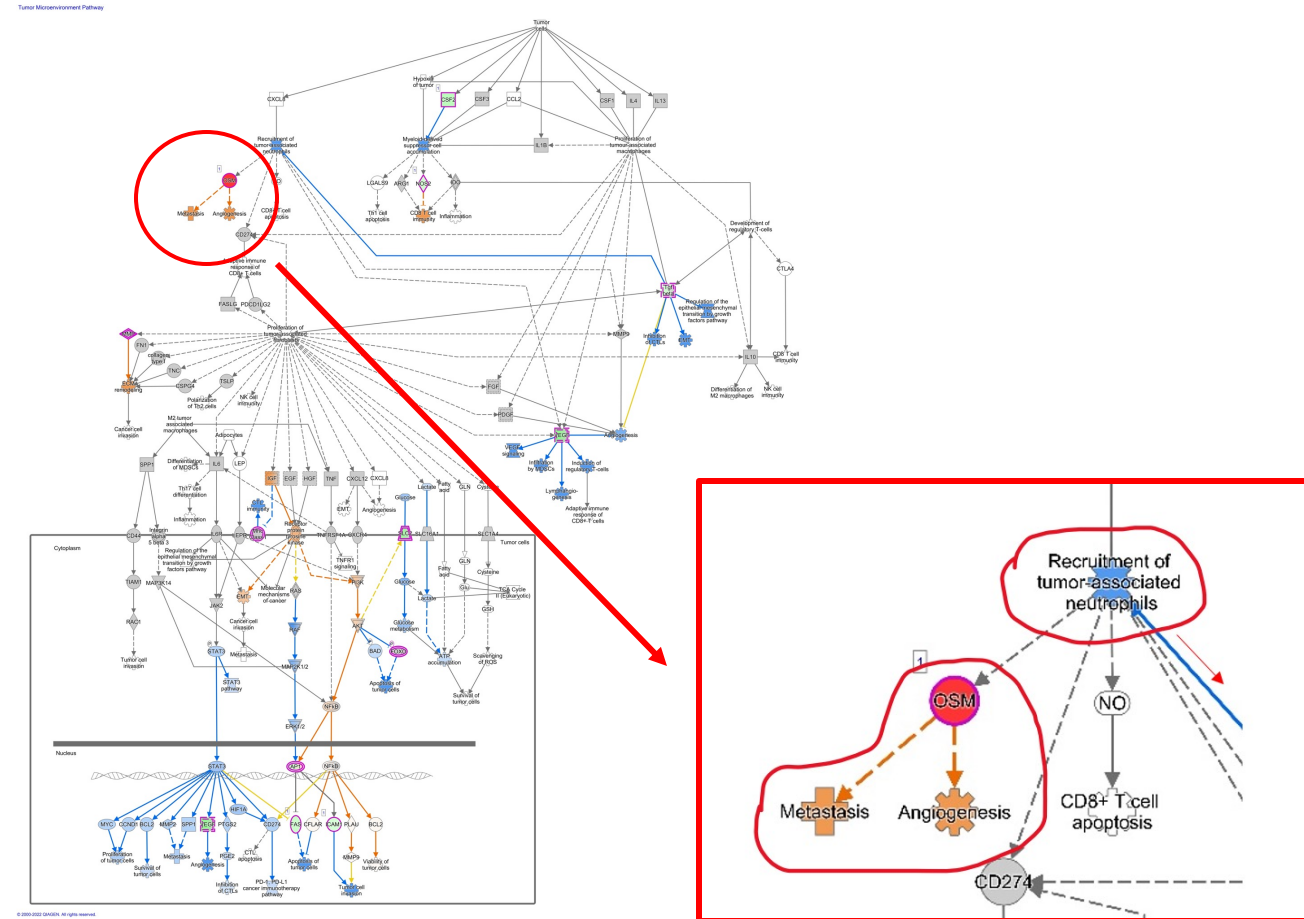
Overlay of Canonical Pathways of this Data Set (Gene Network)

Meta-Analysis Research

Investigating published literature for common genes and pathways between Alzheimer's and Type 2 Diabetes in efforts to reaffirm or find contradictions to our lab's data

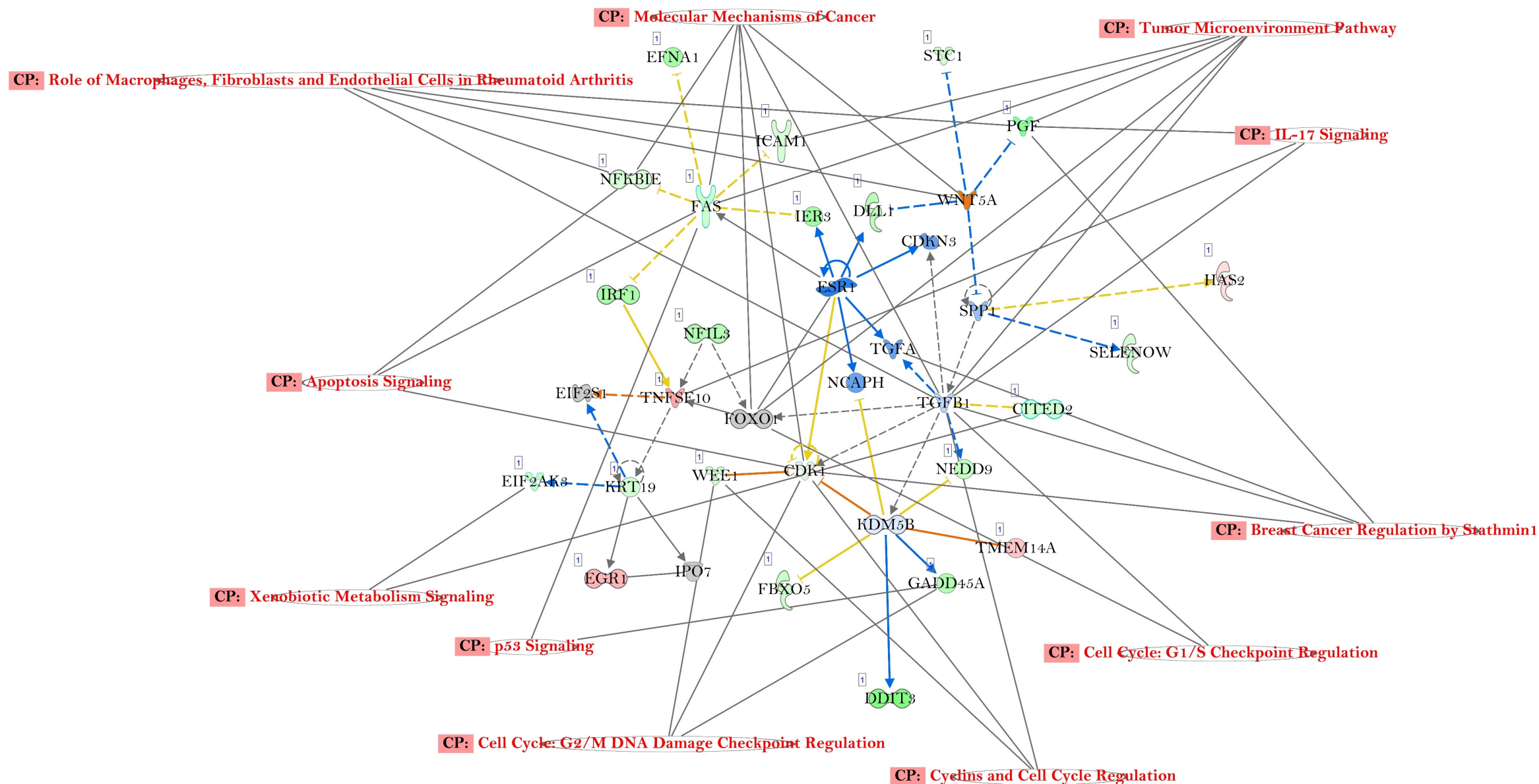| Publication Name	Gene of interest/meas urement	# of Citations	Citation	Link (URL)	Full Text	Study Population	Study location	Study type	Keywords	Date of Publication	Main findings	Notes
RAGE and AGEs in Mild Cognitive Impairment of Diabetic Patients: A Cross-Sectional Study	amyloid-β	8	Wang, P., Huang, R., Lu, S., Xia, W., Cai, R., Sun, H., & Wang, S. (2016). RAGE and AGEs in Mild Cognitive Impairment of Diabetic Patients: A Cross-Sectional Study. PloS one, 11(1), e0145521. https://doi.org/10.1371/journal.pone.0145521	https://pubmed.r	https://www.ncbi	All type 2 diabetes patients - 82 fit the diagostic criteria for MCI (mild cognitive impairment), and 85 were non-MCI controls. All participants were right-handed Han Chinese individuals who had at least eight years of education	China	case/control	Amyloid beta pathway + alzheimers + type 2 diabetes	2016	The RAGE (Receptor for advanced glycation end products) pathway partially mediates AGE(advanced glycation end products)-induced MCI in diabetic patients. The serum AGE-P level may serve as a serum biomarker of MCI in these individuals, and sRAGE represents a predictor and even a potential intervention target of early cognitive decline in type 2 diabetes patients.	RAGE bind enough of a
Association studies of several cholesterol-related genes (ABCA1, CETP and LIPC) with serum lipids and risk of Alzheimer's disease	ABCA1	12	Xiao, Z., Wang, J., Chen, W., Wang, P., Zeng, H., & Chen, W. (2012). Association studies of several cholesterol-related genes (ABCA1, CETP and LIPC) with serum lipids and risk of Alzheimer's disease. Lipids in health and disease, 11, 163. https://doi.org/10.1186/1476-511X-11-163	https://pubmed.r	https://www.ncbi	208 Han Chinese (104 AD patients and 104 non-demented controls) from Changsha area in Hunan Province	China	case/control	Cholesterol biosynthesis + alzheimers + ABCA1	2012	This preliminary study showed that the gene variants of ABCA1R219K and LIPC-250 G/A might influence AD susceptibility in South Chinese Han population, but the polymorphism of CETPTaq1B didn't show any association in despite of being a significant determinant of HDL-C.	seems con rows 16 an
The role of ABCA1 gene sequence variants on risk of Alzheimer's disease	ABCA1	10	Lupton, M. K., Proitsi, P., Lin, K., Hamilton, G., Daniilidou, M., Tsolaki, M., & Powell, J. F. (2014). The role of ABCA1 gene sequence variants on risk of Alzheimer's disease. Journal of Alzheimer's disease : JAD, 38(4), 897–906. https://doi.org/10.3233/JAD-131121	https://pubmed.r	https://content.io	311 LOAD (Late Onset Alzheimers Disease) cases and 360 control individuals drawn from the Greek population	Greece	case/control	Cholesterol biosynthesis + alzheimers + ABCA1	2014	Although recent large scale genome wide association studies (GWAS) have failed to find significant associations with common genetic variants in this gene and LOAD, rare variants in ABCA1 have been shown to influence plasma high-density lipoprotein cholesterol levels....There were a significantly higher proportion of rare non-synonymous variants in control individuals compared to AD cases, suggestive of a protective effect. These findings provide new evidence of an effect of ABCA1 variants on AD risk	red marks a studies look studies four genetic var looked sole connection: without a si
Parahippocampal gyrus expression of endothelial and insulin receptor signaling pathway genes is modulated by Alzheimer's disease and normalized by treatment with anti-diabetic agents	None from our keywords - but study highlights the effects of antidiabetic treatments (insulin or oral agents) on AKT3 and GLUT4 (SLC2A)	5	Katsel, P., Roussos, P., Beeri, M. S., Gama-Sosa, M. A., Gandy, S., Khan, S., & Haroutunian, V. (2018). Parahippocampal gyrus expression of endothelial and insulin receptor signaling pathway genes is modulated by Alzheimer's disease and normalized by treatment with anti-diabetic agents. PloS one, 13(11), e0206547. https://doi.org/10.1371/journal.pone.0206547	https://pubmed.r	https://www.ncbi	Study population consisted of controls (N = 30: 26 white, 3 black, 1 hispanic), of persons with AD (N = 19: 17 white, 2 black, 1 hispanic), and of persons with AD and T2D, who, in turn, had been treated with anti-diabetic drugs (insulin and/or oral agents; N = 34: 26 white, 5 black, 3 hispanic)	Mount Sinai NIH Neurobiobank --> linked address says New York	case/control	Amyloid beta pathway + alzheimers + type 2 diabetes	2018	Our results also showed that the numbers of abnormally expressed microvasculature and IRSP associated genes in diabetic AD donors who had been treated with anti-diabetic agents were reduced significantly. These findings suggest that anti-diabetic treatments may reduce or normalize compromised microvascular and IRSP functions in AD	analysis of

Documentation of Keyword Searches

List of Keywords

List of keywords
Type 2 Diabetes
Cholesterol biosynthesis off alzheimers
APP pathways
Amyloid
Alzheimers
APOE
ABCA1
ACHE
CYP46A1
SOAT1
LRPAP1
APOE4
APOC1
GSK3B
NAE1
ADAM10
ADAM17
ADAM9
APH1A
APH1B
BACE1
BACE2
LRP1
LRP2
PSEN1
PSEN2
PSENEN

Keywords	Database	Number of Results	Notes
amyloid processing + APOE + APOE4 + Cholesterol	PubMed	36 (16 when adjusted for past 10 years)	majority of publications are review articles
amyloid processing pathways + Cholesterol Biosynthesis + Type 2 Diabetes	PubMed	4	all studies that were not review articles were conducted on mice, and therefore could not be included
APOE + Alzheimers + APP pathways	PubMed	123 (75 when adjusting for past 10 years; ten 55 when adjusting for human studies; 53 when adjusting for English)	many studies don't mention an actual population - instead solely focused on human cells used (but don't note what population those samples came from, just which lab they bought or got them from) - either that or the cells used included a mix of mouse and human
Type 2 Diabetes + APP pathways + APOE + alzheimers	PubMed	2	
Cholesterol biosynthesis + alzheimers + APOE4	PubMed	325 (142 when adjusting for past 10 years, 122 when adjusting for human studies, 120 when adjusting for English)	
Cholesterol biosynthesis + alzheimers + ABCA1	PubMed	135 (75 when adjusting for past 1- years, 49 when adjusting for humans, stayed 40 when adjusting for English)	When applying the Human filter, several mice studies come up in the search results
Amyloid beta pathway + alzheimers + type 2 diabetes	PubMed	155 (122 when adjusting for the past 10 years, 77 when adjusting for Humans, 76 when adjusting for English)	significant number of studies based in China or involving Chinese population; majority review articles or simply mislabeled mouse models
APP pathways + Type 2 Diabetes + Alzheimers	PubMed	31 (29 when adjusting for past 10 years, 15 when adjusting for Humans, 15 when adjusting for English)	All usable publications are repeats from "Amyloid beta pathway + alzheimers + type 2 diabetes"
APOE4 + Type 2 Diabetes + Alzheimers	PubMed	60 (44 when adjusting for past 10 years, 42 when adjusting for humans, 41 when adjusting for English)	

…and final conclusions/results!

Mistakes Made & Lessons Learned

Mistakes Made

- Almost throwing away several hours of work!

- Pipette Specifics

Lessons Learned

- Scientists are not scary!

- Comprehension and good work is still very possible if you are a beginner/less educated

- Greater Independence and Responsibility

Acknowledgements

- All the members of Dr. Ghosh's Lab, but especially…
  - Dr. Somiranjan Ghosh
  - Dr. Tanmoy Mondal
  - Taina Cotin
  - Jheannelle Johnson
- Dr. Krug and all Holton Faculty who helped make this internship possible

Thank You!

Any Questions?