Do the differentially expressed genes in pancreatic cancer before treatment match those after CAR T Cell treatment.

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INTRODUCTION

This project approaches the task of gene selection and structuring through mathematical optimization methods in microarray experiments involving CAR T-Cells. To achieve this, we utilized Multiple Criteria Optimization (MCO) in gene selection to analyze microarray datasets and conducted the individual analysis of single datasets.

The objective of this project is to evaluate the effectiveness of CAR T Cell treatment through a comparison between the differentially expressed genes (DEG) after treatment and the independently obtained potential biomarkers in particular types of cancer. To enable proof-of concept, three aims are defined in the context of pancreatic cancer (PC):

- Identify the DEGs in comparative microarray studies between samples with pancreatic cancer and samples with pancreatic cancer after treatment with CAR T Cells
- Identify the DEGs in comparative microarray studies between samples free of pancreatic cancer and samples with pancreatic cancer.
- Determine the matching level between the two sets of DEGs previously identified.

METHODS

The microarray experiments were analyzed using a methodology developed by our research group, the Applied Optimization group (AOG). Using our analysis, it is possible to find those genes that changed their expression the most, without the adjustment of any parameter. Our methodology makes use of Multiple Criteria Optimization (MCO). We emphasize that the application of these methods has the possibility of analyzing multiple experiments simultaneously with deterministic repeatability and user-independent objectivity.

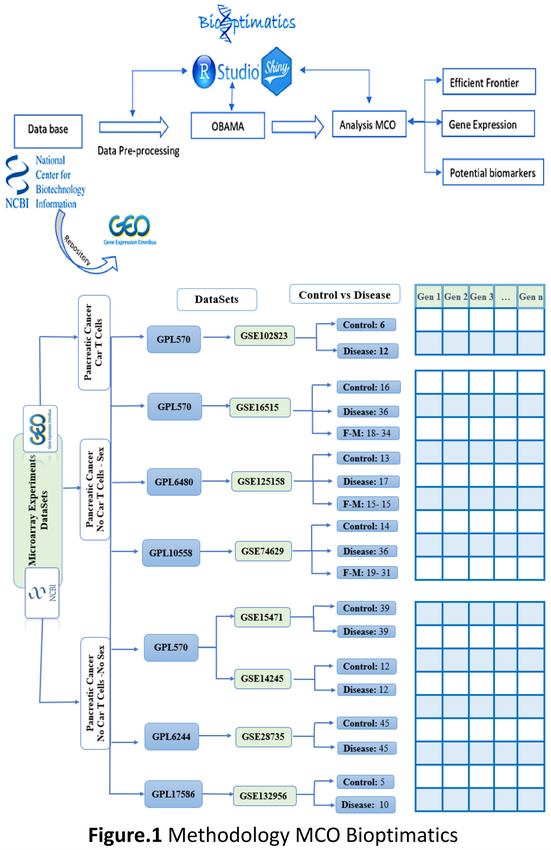


Figure.1 Methodology MCO Bioptimatics

RESULTS

N°	Genes	N°	Genes
1	TMSB10	16	RPL23A
2	RPL23A /// SNORD42A	17	XCL1 /// XCL2
3	RPL41	18	HILPDA
4	LOC101928826 /// TPT1	19	HLA-C
5	ND4	20	RPL39
6	TMSB4X	21	RPS18
7	B2M	22	ACTB
8	COX1	23	GNLY
9	EEF1A1	24	GAPDH
10	GZMB	25	HLA-A
11	RP5-882O7.1	26	HNRNPA1 /// HNRNPA1L2 /// HNRNPA1P10 /// HNRNPA1P33
12	RPS3A /// SNORD73A	27	RPL30
13	ACTB /// ACTG1	28	HNRNPA1 /// HNRNPA1P10
14	LOC100506248 /// LOC728026 /// MIR1244-1 ///MIR1244-2 /// MIR1244-3 /// PTMA	29	RPL9
15	MT2A	30	RPS10

Table 1. MCO Results- Genes of interest from individual analysis in CAR T cells – PC datasets.

Date Sets	Cell Line	Tissue	Genes	Samples: Total	Samples: Control	Samples: Disease	Genes MCO Ind Analysis	
GSE102823 Pancreatic Cancer CAR T Cells	HPAC	Pancreatic Adenocarcinoma	23522	18	6	12	30	
GSE16515 Pancreatic Cancer (sex info) No CAR T-Cells.	(1) AsPC-1, (2)BxPC-3, (3) CFPAC-1, (4)PANC-1, (5) PANC0403, (6)SU86, (7)HupT3, (8)MIApaca-2	Pancreatic tumor and normal tissue	23520	52 18 Female 34 Male	16 12 Female 4 Male	36 14 Female 22 Male	24 Female Genes 27 Male Genes	61 Control Genes 35 Disease Genes
GSE125158 Pancreatic Cancer (sex info) No CAR T-Cells.		Whole blood cells	19527	30 15 Female 15 Male	13	17	48 Female Genes 118 Male Genes	28 Control Genes 14 Disease Genes
GSE74629 Pancreatic Cancer (sex info) No CAR T-Cells.		Peripheral blood	31328	50 19 Female 31 Male	14	36	39 Female Genes 30 Male Genes	52 Control Genes 38 Disease Genes
GSE28735 Pancreatic Cancer No CAR T-Cells.	(1) hTERT-HPNE (CRL-4023™) (2)MIApaca2 (CRL-1420™), (3)Panc1 (CRL-1469™)	Pancreatic tumor and non-tumor tissue	23309	90	45	45	40	
GSE15471 Pancreatic Cancer No CAR T-Cells.	(1) PANC-1 (2)SW 1990	Pancreatic tumor and non-tumor tissue	23522	78	39	39	42	
GSE14245 Pancreatic Cancer No CAR T-Cells.		Saliva	23522	24	12	12	109	
GSE132956 Pancreatic Cancer No CAR T-Cells.		Pancreatic Ductal Adenocarcinoma tissue and normal	30908	15	5	10	29	

Table 2. Microarray Experiments- Datasets information and genes MCO individual analysis.

IMPACT ON CMaT

Vertically integrated in the 3 Plane Diagram

Project: Matching CAR T Cell treatment differentially expressed genes to potential biomarkers in particular types of cancer

Thrust 1: Cell-Omics: Cell Characterization and Computational Modeling, TB = Test-Bed 2: CAR-T cells.

- Fundamental Knowledge Plane:
  - New systems-driven multi-omics pipeline for cell characterization
- Enabling Technologies Plane:
  - Big data analytics tools for predicting cell function.
  - Computational tools for cross-platform multi-omics analyses
  - Multi-omics platform integration
- Systems Plane:
  - Predictive systems analyses of therapeutic cells
  - Education, outreach, inclusivity and workforce development.

Vertically and Horizontally Integrated on Workforce Training

- High School Trainees and Trainees mentored by UPRM and Grad Students
- Cross-institutional and cross-disciplinary training
- RET and REM trainees – Dissemination of knowledge

This project addresses the following systems specification for Engineered T cells:

- Develop robust and scalable manufacturing and supply chain platforms to reduce risk and cost, and improve access
- Develop in-line, nondestructive Process Analytical technologies (PATs), with more predictive potency and quality assays

RESULTS

Pancreatic Cancer - MCO Individual Analysis by Sex.

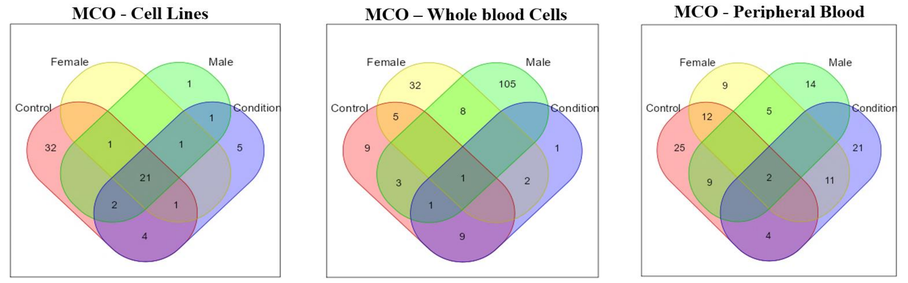
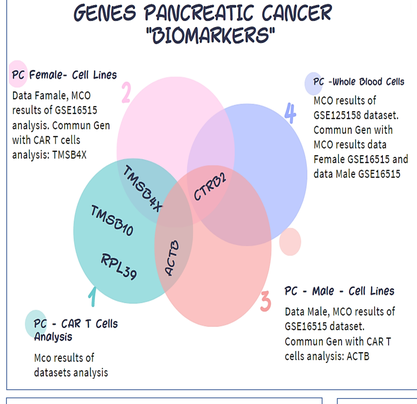


Figure 2. MCO Results by individual analysis for sex, datasets GSE16515, GSE125158 and GSE74629

	Common Genes										
MA1 - MPC	TMSB10	RPL39	TMSB4X	ACTB	B2M	EEF1A1	MT2A	RPL23A	RPS18	GNLY	GAPDH
MA1&MA2 - MPC	TMSB10	RPL39	RPS10								
MA2 - MPC	TMSB10	RPL39									

Table 3. Common Genes found between the following Meta-Analysis: MA1, the Meta-Analysis of pancreatic cancer datasets with non-CAR T cells and sex information; MA2, the Meta-Analysis of pancreatic cancer datasets with non-CAR T cells and without sex information, and MPC, the MCO individual analysis of dataset GSE102823.

CONCLUSION



This project allows the characterization of different genes with expression changes, their significance, and their role in cancer. The genes that demonstrated the largest expression changes and can be identified as potential biomarkers were TMSB4X, ACTB, RPL39, and TMSB10. TMSB4X has been shown to be overexpressed in pancreatic cancer, [1]. In contrast, ACTB is deregulated in pancreatic cancer, [2]. CTRB2 can increase the risk of pancreatic cancer, [3]. Additionally, RPL39 overexpression plays a vital role in the metastasis of pancreatic and breast cancer [4]. TMSB10 overexpression results in the development of renal cell carcinoma, non-small cell lung cancer, and papillary thyroid cancer, it is also upregulated in both pancreatic carcinoma tissues and cell lines. With this results, different alternatives for cell therapies that involve CAR T cells for cancer treatment can be thought of and developed.

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