Differentiate Colorectal carcinoma and para-carcinoma tissue based on expression profile

Title: Hypoxia-induced cysteine metabolism reprogramming are crucial for the tumorigenesis of colorectal cancer

Gene Expression Omnibus: GSE223119

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Q1: Main objective of the analysis that specifies whether your model will be focused on prediction or interpretation and the benefits that your analysis provides to the business or stakeholders of this data.

Answer:
This analysis wil be focused on prediction
By determining whether the gene expresison profiles capable of classifying the sample's origin, future expression profiles of unknown origin may be classified accordingly
In addition, the analysis will compare and contrast the performance of classificaiton tools on low sample, high-dimension data set.
ie. each gene is assumed to be independent feature for this analysis

Q2: Brief description of the data set you chose, a summary of its attributes, and an outline of what you are trying to accomplish with this analysis.

Answer:
Metabolic reprogramming is a hallmark of human cancer and cancer-specific metabolism provide opportunities for cancer diagnosis, prognosis, and treatment. However, how metabolic pathways affect the initiation and progression of colorectal cancer remain largely unknown.
This data set includes 40 gene expression profiles of cancer and para-carcinoma tissue (tissue surrounding cancer).
This analysis attempt to build a model for predicting the tissue origin of unknown gene profiles

In [1]:
```
#No warning output
def warn(*args, **kwargs):
    pass
import warnings
warnings.warn = warn
```

Q3: Brief summary of data exploration and actions taken for data cleaning and feature engineering.

Answer:
Data Exploration:

- Raw data is composed of gene ID, gene expression on each sample, gene names, and database and gene network category
- Gene expression has been preprocessed by the data provider, with no missing values
- This data set contain extremely high amount of features (ie. 61,700 genes)

Data Engineering:

- For the project purpose, only gene ID (feature), gene expression, and tissue origin will be considered:
- Removing features that are not expressing or have very low variability among all profiles
- For simplification in this analysis, I randomly selected 3000 features for analysis
- Gene expression may or may not be skewed, and expression levels vary from gene to gene, standardization is applied

Detail analysis see below

In [2]:
```
#Import Data processing libraries
import numpy as np
import pandas as pd
```

In [3]:
```
#Load Data, data is preprocessed
rawData=pd.read_csv(r'C:\Users\kai-w\Desktop\03_Supervised Machine Learning Classification\GSE223119_MJ20190424016-gene.tpm.matrix.annot.txt',sep='\t',header=0)
```

In [4]:
```
#Data overview
rawData.head()
```

Out[4]:

	Unnamed: 0	C10	C11	C12	C13	C14	C15	C18	C1	C20	...	cog	cog_description	KO_id	KO_name	paths	pfam	go	n
0	ENSG00000000003	61.65	37.27	85.58	68.76	32.17	49.63	64.18	10.96	36.86	...	ENOG4111IRY(S:Function unknown)	ENOG4111IRY(Tenomodulin)	K17295	TSPAN6	NaN	PF00335.17(Tetraspannin:Tetraspanin family)	GO:0039532(biological_process:negative regulat...	NP_003261.1(tetraspanin-(isoform a [Homo sapi.
1	ENSG00000000005	0.31	3.39	1.34	2.24	0.25	4.48	1.64	0.15	0.89	...	ENOG410YB96(S:Function unknown)	ENOG410YB96(Tenomodulin)	NaN	NaN	NaN	PF04089.11(BRICHOS:BRICHOS domain)	GO:0005737(cellular_component:cytoplasm); GO:0...	XP_006986474.1(PREDICTED tenomodulin [Peromys.
2	ENSG00000000419	81.96	34.78	56.60	81.13	49.78	121.31	91.79	39.23	41.02	...	COG0463(M:Cell wall/membrane/envelope biogenesis)	COG0463(Glycosyl transferase, family 2)	K00721	DPM1	map00510(N-Glycan biosynthesis)	PF00535.23(Glycos_transf_2:Glycosyl transferas...	GO:0019673(biological_process:GDP-mannose meta...	NP_001303964.1(dolichol phosphate mannosyltran.
3	ENSG00000000457	4.98	1.42	2.84	4.01	4.90	3.86	3.63	4.08	3.95	...	ENOG410XQTG(S:Function unknown)	ENOG410XQTG(S. cerevisiae)	K17542	SCYL3	NaN	PF00069.22(Pkinase:Protein kinase domain); PF0...	GO:0005794(cellular_component:Golgi apparatus)...	XP_003893590.2(protein associating with the ca.
4	ENSG00000000460	7.14	1.99	2.80	9.40	5.90	4.13	9.92	4.05	4.21	...	ENOG4110VTC(S:Function unknown)	ENOG4110VTC(Chromosome 1 open reading frame 112)	NaN	NaN	NaN	PF14868.3(DUF4487:Domain of unknown function (...	NaN	XP_005245374.1(uncharacterizec protein C1orf11.

5 rows × 54 columns

In [5]:
```
#Data types for each column
rawData.dtypes
```

Out[5]:

```
Unnamed: 0          object
C10                float64
C11                float64
C12                float64
C13                float64
C14                float64
C15                float64
C18                float64
C1                 float64
C20                float64
C22                float64
C23                float64
C24                float64
C25                float64
C3                 float64
C4                 float64
C5                 float64
C6                 float64
C7                 float64
C8                 float64
C9                 float64
P10                float64
P11                float64
P12                float64
P13                float64
P14                float64
P15                float64
P18                float64
P1                 float64
P20                float64
P22                float64
P23                float64
P24                float64
P25                float64
P3                 float64
P4                 float64
P5                 float64
P6                 float64
P7                 float64
P8                 float64
P9                 float64
gene_name           object
length               int64
description         object
cog                 object
cog_description     object
KO_id               object
KO_name             object
paths               object
pfam                object
go                  object
nr                  object
swissprot           object
entrez             float64
dtype: object
```

In [6]:

```
#Transpose so treat genes as feature, and each profile as independent record
data=rawData.loc[:,rawData.columns.str.match('^C|^P')].rename(index=rawData['Unnamed: 0']).T
data.head()
```

Out[6]:

	ENSG00000000003	ENSG00000000005	ENSG00000000419	ENSG00000000457	ENSG00000000460	ENSG00000000938	ENSG00000000971	ENSG00000001036	ENSG00000001084	ENSG00000001167	...	MSTRG.9662	MSTRG.9664	MSTRG.9666	MSTRG.9667	MSTRG.9925
C10	61.65	0.31	81.96	4.98	7.14	4.40	15.56	38.45	18.87	12.65	...	0.02	0.00	0.0	0.13	0.37
C11	37.27	3.39	34.78	1.42	1.99	3.10	5.15	34.72	11.98	3.12	...	0.10	6.57	0.0	1.17	0.61
C12	85.58	1.34	56.60	2.84	2.80	5.31	8.48	29.41	25.83	11.12	...	0.07	0.00	0.0	0.20	0.17
C13	68.76	2.24	81.13	4.01	9.40	4.46	14.99	45.74	25.93	13.76	...	0.04	0.00	0.0	0.33	0.75
C14	32.17	0.25	49.78	4.90	5.90	2.46	10.40	33.28	17.18	11.91	...	0.27	7.07	0.0	0.36	0.54

5 rows × 61700 columns

In [7]:

```
#Assign tissue types for classification
data['Tissue']=data.index.str[0:1]
data.head()
```

Out[7]:

	ENSG00000000003	ENSG00000000005	ENSG00000000419	ENSG00000000457	ENSG00000000460	ENSG00000000938	ENSG00000000971	ENSG00000001036	ENSG00000001084	ENSG00000001167	...	MSTRG.9664	MSTRG.9666	MSTRG.9667	MSTRG.9925	MSTRG.9927
C10	61.65	0.31	81.96	4.98	7.14	4.40	15.56	38.45	18.87	12.65	...	0.00	0.0	0.13	0.37	0.00
C11	37.27	3.39	34.78	1.42	1.99	3.10	5.15	34.72	11.98	3.12	...	6.57	0.0	1.17	0.61	1.78
C12	85.58	1.34	56.60	2.84	2.80	5.31	8.48	29.41	25.83	11.12	...	0.00	0.0	0.20	0.17	0.50
C13	68.76	2.24	81.13	4.01	9.40	4.46	14.99	45.74	25.93	13.76	...	0.00	0.0	0.33	0.75	0.23
C14	32.17	0.25	49.78	4.90	5.90	2.46	10.40	33.28	17.18	11.91	...	7.07	0.0	0.36	0.54	0.20

5 rows × 61701 columns

In [8]:

```
#check for presence of empty values
data.isna().values.sum()
```

Out[8]:

```
0
```

In [9]:

```
#huge feature size, reduce by taking randomly 5000 for the purpose of the project
data.shape
```

Out[9]:

```
(40, 61701)
```

In [10]:

```
#some features/genes have low variability, remove those
feature_var=pd.DataFrame([[i, data[i].std()] for i in data.columns[:-1]],
                         columns=['feature', 'std']).set_index('feature')
print(feature_var[feature_var['std']>0].sort_values('std'))
```

```
                          std
feature
ENSG00000232264      0.001581
ENSG00000228856      0.001581
ENSG00000231051      0.001581
ENSG00000133105      0.001581
ENSG00000156925      0.001581
...                       ...
ENSG00000211890   6428.959034
ENSG00000240040   6780.842254
ENSG00000212907   7435.193295
ENSG00000198804   8216.179258
ENSG00000228253  18410.117553

[41928 rows x 1 columns]
```

In [11]:

```
#random select 3000 features/genes that are variable for the project's purpose
features=feature_var[feature_var['std']>0.5].index.to_series().sample(3000).to_list()
data[features].head()
```

Out[11]:

	ENSG00000283050	ENSG00000178971	ENSG00000267530	ENSG00000181274	ENSG00000137414	ENSG00000268621	ENSG00000175309	ENSG00000103047	ENSG00000129226	ENSG00000133818	...	ENSG00000181754	MSTRG.35998	ENSG00000054654	ENSG000001120
C10	3.51	7.54	0.81	33.93	12.00	0.08	38.41	1.90	61.67	11.94	...	2.31	1.91	22.00	5
C11	0.82	8.99	1.85	18.74	4.18	129.29	12.25	2.26	111.31	9.39	...	0.65	12.09	13.21	3
C12	2.89	8.23	0.99	36.07	5.21	0.42	19.09	2.71	62.22	12.51	...	2.01	5.18	36.34	2
C13	3.47	5.18	0.41	18.03	8.45	0.00	30.69	4.56	64.74	19.50	...	1.16	4.44	20.36	5
C14	2.82	10.46	0.75	25.51	14.27	5.83	23.80	3.72	43.10	16.90	...	1.82	8.47	24.61	4

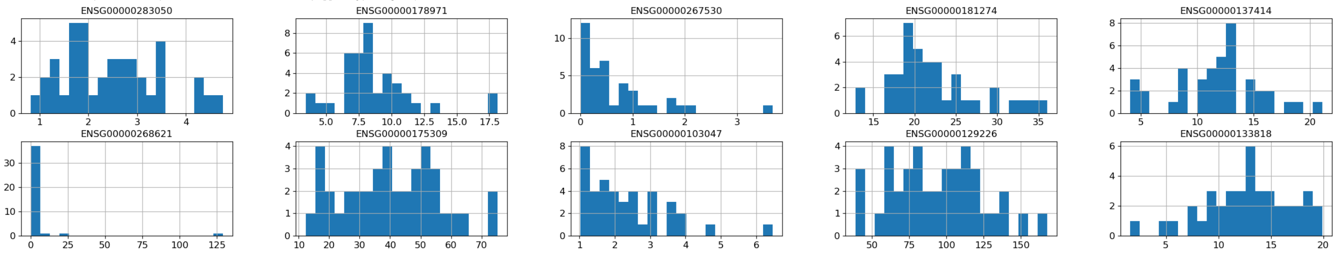
5 rows × 3000 columns

In [12]:

```
#some features are skwed and some are not, vary dependent on genes
import matplotlib

params = {'axes.titlesize':'12',
          'xtick.labelsize':'12',
          'ytick.labelsize':'12'}
matplotlib.rcParams.update(params)
data[features[0:10]].hist(bins=20,figsize=(30, 5),layout=(2,5))
```

```
Out[12]: array([[<AxesSubplot:title={'center':'ENSG00000283050'}>,
                 <AxesSubplot:title={'center':'ENSG00000178971'}>,
                 <AxesSubplot:title={'center':'ENSG00000267530'}>,
                 <AxesSubplot:title={'center':'ENSG00000181274'}>,
                 <AxesSubplot:title={'center':'ENSG00000137414'}>],
                [<AxesSubplot:title={'center':'ENSG00000268621'}>,
                 <AxesSubplot:title={'center':'ENSG00000175309'}>,
                 <AxesSubplot:title={'center':'ENSG00000103047'}>,
                 <AxesSubplot:title={'center':'ENSG00000129226'}>,
                 <AxesSubplot:title={'center':'ENSG00000133818'}>]], dtype=object)
```



Q4: Summary of training at least three different classifier models, preferably of different nature in explainability and predictability. For example, you can start with a simple logistic regression as a baseline, adding other models or ensemble models. Preferably, all your models use the same training and test splits, or the same cross-validation method.

Answer:

Logistic regression, K-nearest neighbours, SVM, and decision tree are applied

Given only few data record, bagging and boosting may be less suit for this analysis

Preprocessing

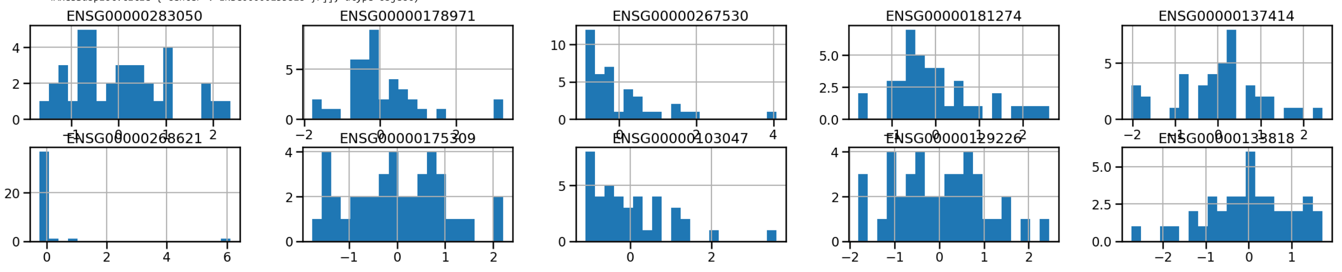
```
In [13]: #Load preprocessing libraries
         from sklearn.preprocessing import StandardScaler
         from sklearn.preprocessing import LabelBinarizer
```

```
In [14]: #subset of data for standardize and binarize labels
         X=pd.DataFrame(StandardScaler().fit_transform(data[features]),columns=features)
         Y=pd.DataFrame(LabelBinarizer().fit_transform(data['Tissue']),columns=['Tissue'])
         print(X.iloc[:5,:4])
         print(Y.head())  #C:0 ; P:1
```

```
   ENSG00000283050  ENSG00000178971  ENSG00000267530  ENSG00000181274
0         1.081370        -0.381823         0.206446         2.204380
1        -1.679139         0.113386         1.607449        -0.659688
2         0.445119        -0.146172         0.448927         2.607876
3         1.040322        -1.187818        -0.332401        -0.793558
4         0.373284         0.615425         0.125619         0.616793
   Tissue
0       0
1       0
2       0
3       0
4       0
```

```
In [90]: #data distribution after standard scaling, some skewness presist
         X.iloc[:,:10].hist(bins=20,figsize=(30, 5),layout=(2,5))
```

```
Out[90]: array([[<AxesSubplot:title={'center':'ENSG00000283050'}>,
                 <AxesSubplot:title={'center':'ENSG00000178971'}>,
                 <AxesSubplot:title={'center':'ENSG00000267530'}>,
                 <AxesSubplot:title={'center':'ENSG00000181274'}>,
                 <AxesSubplot:title={'center':'ENSG00000137414'}>],
                [<AxesSubplot:title={'center':'ENSG00000268621'}>,
                 <AxesSubplot:title={'center':'ENSG00000175309'}>,
                 <AxesSubplot:title={'center':'ENSG00000103047'}>,
                 <AxesSubplot:title={'center':'ENSG00000129226'}>,
                 <AxesSubplot:title={'center':'ENSG00000133818'}>]], dtype=object)
```



```
In [27]: #Load libraries for classifier and reports
         from sklearn.model_selection import train_test_split
         from sklearn.linear_model import LogisticRegressionCV
         from sklearn.neighbors import KNeighborsClassifier
         from sklearn.svm import SVC
         from sklearn.tree import DecisionTreeClassifier

         #load report libraries
         from sklearn.metrics import confusion_matrix, classification_report, accuracy_score
         from sklearn.metrics import precision_score, recall_score, f1_score, roc_auc_score

         #grid search for best parameters
         from sklearn.model_selection import GridSearchCV

         import time
```

```
In [28]: #train-test split, double check for splitted data set
         X_train, X_test, Y_train, Y_test = train_test_split(X, Y, test_size=0.4, random_state=5)
         print("X_train: {}, Y_train: {}, X_test: {}, Y_test: {},".format(X_train.shape,Y_train.shape,X_test.shape,Y_test.shape) )
```

```
X_train: (24, 3000), Y_train: (24, 1), X_test: (16, 3000), Y _test: (16, 1),
```

Logistic Regression

```
In [81]: #Linear regresison model
         logR=GridSearchCV(estimator=LogisticRegressionCV(solver='saga',n_jobs=-1),
                          param_grid={'cv':[3,5], 'penalty':['l1', 'l2', 'elasticnet']})

         #train
         logR_time = time.time()
         logR.fit(X_train,Y_train)
         logR_time_train = time.time() - logR_time
         #test
         logR_time = time.time()
         Y_pred_logR = logR.predict(X_test)
         logR_time_pred = time.time() - logR_time
```

KNN

```
In [82]: #KNN model
         knn_time = time.time()
         knn=GridSearchCV(estimator=KNeighborsClassifier(n_jobs=-1),
                         param_grid={'n_neighbors':[3,8]})

         #train
         knn_time = time.time()
         knn.fit(X_train,Y_train)
         knn_time_train = time.time() - knn_time
         #test
         knn_time = time.time()
         Y_pred_knn = knn.predict(X_test)
         knn_time_pred = time.time() - knn_time
```

SVM

```
In [83]: #SVM model
         svm_time = time.time()
         svc=GridSearchCV(estimator=SVC(),
                          param_grid={'C':[1e-3,1e-2,1e-1,1], 'kernel':['linear', 'poly', 'rbf'], 'degree':range(2,5)})

         #train
         svm_time = time.time()
         svc.fit(X_train,Y_train)
         svm_time_train = time.time() - svm_time
         #test
         knn_time = time.time()
         Y_pred_svm = svc.predict(X_test)
         svm_time_pred = time.time() - svm_time
```

Decision Tree

```
In [84]: #Decision tree model
         dTree_time = time.time()
         dTree=GridSearchCV(estimator=DecisionTreeClassifier(random_state=13),
                           param_grid={'criterion':['gini', 'entropy', 'log_loss'], 'max_depth':range(5,50,5)})

         #train
         dTree_time = time.time()
         dTree.fit(X_train,Y_train)
         dTree_time_train = time.time() - dTree_time
         #test
         dTree_time = time.time()
         Y_pred_dTree = dTree.predict(X_test)
         dTree_time_pred = time.time() - dTree_time
```

Analysis of each classifiers

```
In [85]: score_df = pd.DataFrame()

         for i, j in enumerate([Y_pred_logR, Y_pred_knn, Y_pred_svm, Y_pred_dTree]):
             score_df[i] = [accuracy_score(Y_test,j),
                           precision_score(Y_test,j),
                           recall_score(Y_test,j),
                           f1_score(Y_test,j),
                           roc_auc_score(Y_test,j)]
         score_df = score_df.T
         score_df.columns=['accuracy','precision','recall','f1','auc']
         score_df.index=['Logistic Regression','KNN','SVM','Decision Tree']
         score_df['Training time']=[logR_time_train, knn_time_train, svm_time_train, dTree_time_train]
         score_df['Prediction time']=[logR_time_pred, knn_time_pred, svm_time_pred, dTree_time_pred]
         score_df
```

Out[85]:

	accuracy	precision	recall	f1	auc	Training time	Prediction time
Logistic Regression	1.0000	1.0	1.0	1.000000	1.00	21.612164	0.020988
KNN	1.0000	1.0	1.0	1.000000	1.00	0.835522	0.056967
SVM	0.9375	1.0	0.9	0.947368	0.95	8.081401	8.103387
Decision Tree	0.9375	1.0	0.9	0.947368	0.95	7.195887	0.020989

```
In [86]: # Display parameters for each estimators
         print("Logistic Rergression: ",logR.best_estimator_)
         print("KNN: ",knn.best_estimator_)
         print("SVM: ",svc.best_estimator_)
         print("Decision Tree: ",dTree.best_estimator_)

         Logistic Rergression:  LogisticRegressionCV(cv=3, n_jobs=-1, penalty='l1', solver='saga')
         KNN:  KNeighborsClassifier(n_jobs=-1, n_neighbors=3)
         SVM:  SVC(C=1, degree=2)
         Decision Tree:  DecisionTreeClassifier(max_depth=5, random_state=13)
```

```
In [87]: #visualization
         import matplotlib.pyplot as plt
         import seaborn as sns
         %matplotlib inline
```

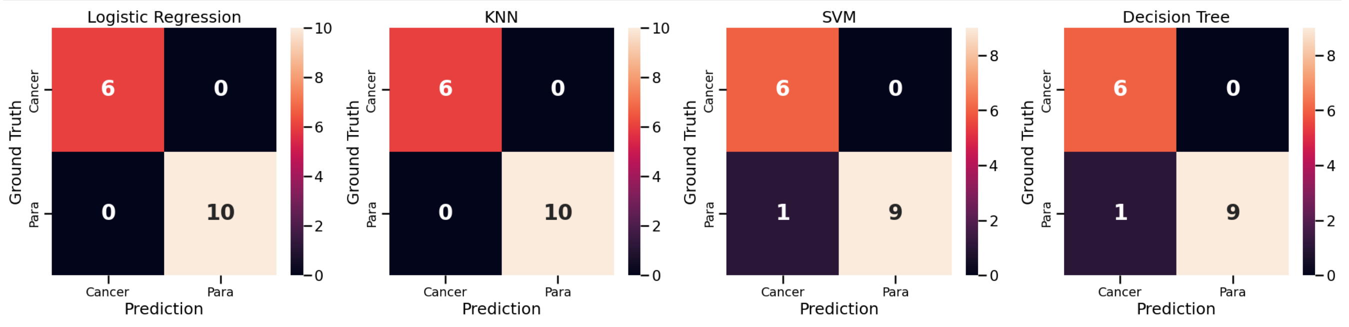
```
In [88]: #Y_test data
         Test_Data=pd.DataFrame(Y_test.value_counts().to_list(),index=['Para','Cancer'],columns=['Counts'])
         Test_Data
```

Out[88]:

	Counts
Para	10
Cancer	6

```
In [89]: #plot curves for each classification methods
         from sklearn.metrics import confusion_matrix
         sns.set_context('talk')

         fig, axes = plt.subplots(1, 4, figsize=(26,5))
         for i, j in enumerate([Y_pred_logR, Y_pred_knn, Y_pred_svm, Y_pred_dTree]):
             cm = confusion_matrix(Y_test, j)
             sns.heatmap(cm, ax=axes[i], annot=True, fmt='d', annot_kws={"size": 24, "weight": "bold"})
             axes[i].set_title(score_df.index[i])
             axes[i].set_xticklabels(['Cancer','Para'], fontsize=14);
             axes[i].set_yticklabels(['Cancer','Para'], fontsize=14);
             axes[i].set_ylabel('Ground Truth', fontsize=18)
             axes[i].set_xlabel('Prediction', fontsize=18);
```



Q5: A paragraph explaining which of your classifier models you recommend as a final model that best fits your needs in terms of accuracy and explainability.

Answer:

For this data set, all 4 tested classifiers showed strong predictability.
Among which, Logistic Regression and KNN perform best (highest accuracy, precision, and recall)
Comparatively, KNN with 3 nearest neighbours perform fast in both training and prediction, therefore may best suit for this data set, which contain large number of features (genes)

Q6: Summary Key Findings and Insights, which walks your reader through the main drivers of your model and insights from your data derived from your classifier model.

Answer:

Given large amount of features granted the stronger predicrbaility for all tested models: Logistic Regression, KNN, SVM, and Decision Tree.
Several runs were performed, and number of random selected features/genes from 500 to 5000 were tested. The outcomes turned out to be almost identical. This implies that given high amount of features, individual feature may not play a decisive role in classification models. These also suggest the robustness of this model.

Q7: Suggestions for next steps in analyzing this data, which may include suggesting revisiting this model after adding specific data features that may help you achieve a better explanation or a better prediction.

Answer:

Given the KNN model is highly predictable for this data set, more data sets of similar processing steps may be applied for classification. Given the robustness(100% predictability) of this model, more gene expression profiles may be used to test the predictability of this model.