PluriTest™ Analysis

HELP

Publication Number MAN0018056

Revision B.0

Affymetrix Inc. | 3450 Central Expressway | Santa Clara, CA 95051 | USA

The information in this guide is subject to change without notice.

DISCLAIMER: TO THE EXTENT ALLOWED BY LAW, THERMO FISHER SCIENTIFIC INC. AND/OR ITS AFFILIATE(S) WILL NOT BE LIABLE FOR SPECIAL, INCIDENTAL, INDIRECT, PUNITIVE, MULTIPLE, OR CONSEQUENTIAL DAMAGES IN CONNECTION WITH OR ARISING FROM THIS DOCUMENT, INCLUDING YOUR USE OF IT.

Revision history: Pub. No. MAN0018056

Revision	Date	Description
B.0	06 February 2019	Add Delete analysis group workflow
A.0	06 September 2018	New help system

Important Licensing Information: This product may be covered by one or more Limited Use Label Licenses. By use of this product, you accept the terms and conditions of all applicable Limited Use Label Licenses.

TRADEMARKS: All trademarks are the property of Thermo Fisher Scientific and its subsidiaries unless otherwise specified.

Contents

Welcome to PluriTest™ Analysis

PluriTest™ Analysis is a bioinformatics tool for pluripotency assessment that provides an alternative to the teratoma assay based on global gene expression data. The PluriTest™ Analysis compares the transcriptional profile of a sample to an extensive reference set of >450 cell/tissues types, including 223 hESC lines, 41 iPSC lines, somatic cells, and tissues.

PluriTest™ Analysis confirms pluripotency marker expression using two separate scores: pluripotency and novelty. The pluripotency score indicates how strongly a model-base pluripotency signature is expressed in the samples analyzed. The novelty score indicates the general model fit for a given sample.

Create an analysis group

Note: At this time, only `.cel` files can be processed.

1. Click **Create an analysis group**.

2. Type the name of the analysis group, then select the **File/Array type** from the dropdown menu.

3. Click **Ok**.

Import the data files and run analysis

Files can be uploaded into analysis group from local drive or from Thermo Fisher Cloud Data Manager.

1. After entering into an analysis group, click on **Import Data**

2. To select files from your local drive:
   a. In the **Import Panel**, click **My Computer** and click **Import**.

   b. Choose the destination folder and click **Select File**.

   c. In the file browser that launches, select the input files and click **Open**.

      Note: The imported files will also be automatically saved in your Thermo Fisher Cloud Data Manager.

3. To select files from your Thermo Fisher Cloud Data Manager.
   a. In the **Import Panel**, click **Thermo Fisher Cloud** icon and click **Import**.

b. In the **Import Data** panel that appears, expand the relevant directory to view the cel files. Click **Add** for each file to move it to the **Items to import** section.

c. Click **Import** to view the files in the analysis group.

4. After the files are imported, click **Analyze** to run the analysis.

5. Once analysis is run successfully, click **View Results**.

Analysis details

The analysis results are displayed in a table showing for each data file: **PluriTest Results** as **Pass**, **Further Evaluate**, or **Fail** (see Table 1), **PluriCor** (Pluripotency Score), and **NovelCor** (Novelty Score).

Table 1 PluriTest™ Analysis Results

PluriTest Results[1]	PluriCor (Pluripotency Score)	NovelCor (Novelty Score)
Pass	>20	<1.67
Further Evaluate	>10	<1.67
Fail	<10	>1.67

[1] For a **Pass** or a **Fail** results, both scores must be within the indicated ranges. For a **Further Evaluate** result, only one score must be within the indicated ranges.

You can also view the following graph plots by clicking the corresponding button:

Button	Description
Relative Log Expression	The relative log expression values are computed by calculating for each probe-set the ratio between the expression of a probe-set and the median expression of this probe-set across all arrays analyzed. Most probe-sets are not changed across the arrays, therefore these ratios are around 0 on a log scale. The box plots presenting the distribution of these log-ratios should be centered near 0 and have similar spread. Other results would indicate low quality.

Button	Description
Normalized Unscaled Standard Errors	The normalized unscaled standard error is the individual probe error fitting the Probe-Level Model (the PLM models expression measures using a M-estimator robust regression). The NUSE values are standardized at the probe-set level across the arrays: median values for each probe-set are set to 1.
	Use the box plots to check for low quality. Examples of low quality are if all distributions are centered near 1 (typically an array with a boxplot centered around 1.1 shows bad quality), or if one array has a globally higher spread of normalized unscaled standard errors distribution than others.
Box Plots	A box plot is generated after the samples are transformed with a variance stabilizing transformation (VST) and before robust spline normalization (RSN). Use this quality control plot to spot outlier arrays with too much technical variation, for example, if they show a different probe intensity distribution pattern in the box plots when compared to probes on the other arrays on the same chip or when compared to arrays on other chips.
Clustering	Hierarchical clustering is a quality control plot generated after the samples were transformed with a variance stabilizing transformation (VST) and before robust spline normalization (RSN). Outlier arrays with too much technical variation might be spotted if they do not cluster with their respective technical or biological replicates from the same sample or sample type.
Pluripotency Plot	The pluripotency plot and novelty x/y scatter plot combines the pluripotency score on the y-axis with the novelty score on the x-axis. The red and blue background hint to the empirical distribution of the pluripotent (red) and nonpluripotent samples (blue) in the reference data set.

Delete analysis group(s)

One or more analysis groups can be deleted from the application.

1. Select analysis group then click **Delete an analysis group**.
   A confirmation prompt will be displayed.

2. Click **Yes** to remove the analysis group(s) permanently from the list.

   Note: If analysis group that is in **Completed** status is deleted, then it's corresponding analysis results folder available on Thermo Fisher Cloud Data Manager will also be deleted.

   Note: Analysis groups that are in progress status cannot be deleted because analysis is running.

Limited product warranty

Life Technologies Corporation and/or its affiliate(s) warrant their products as set forth in the Life Technologies' General Terms and Conditions of Sale at **www.thermofisher.com/us/en/home/global/terms-and-conditions.html**. If you have any questions, please contact Life Technologies at **www.thermofisher.com/support**.