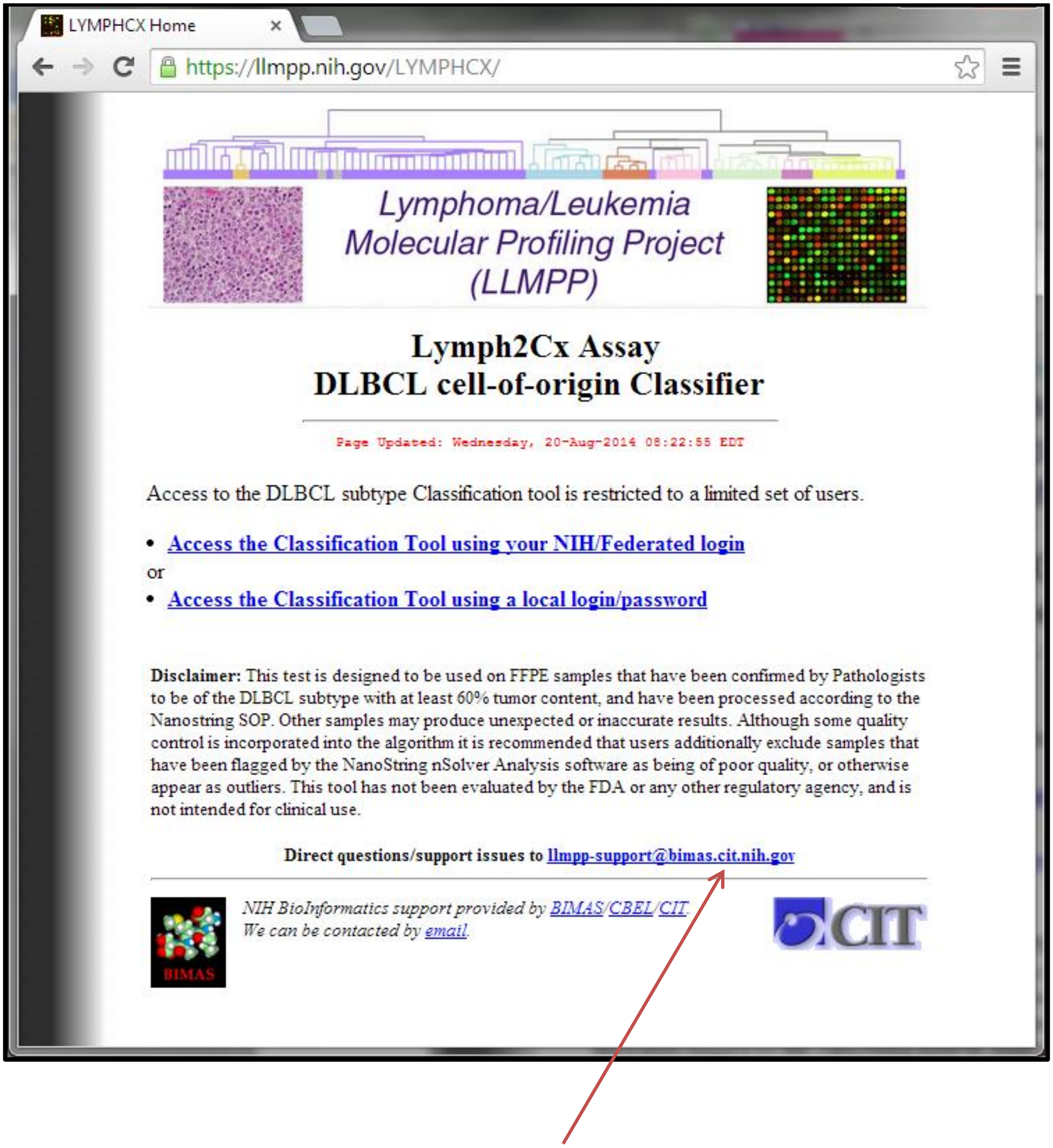
# Quick Start Guide: Lymph2Cx Assay DLBCL cell-of-origin Classifier Web Site

Home page: https://llmpp.nih.gov/LYMPHCX/index.shtml



Lymphoma/Leukemia
Molecular Profiling Project
(LLMPP)

**Lymph2Cx Assay**
**DLBCL cell-of-origin Classifier**

Page Updated: Wednesday, 20-Aug-2014 08:22:55 EDT

Access to the DLBCL subtype Classification tool is restricted to a limited set of users.

- Access the Classification Tool using your NIH/Federated login

or

- Access the Classification Tool using a local login/password

**Disclaimer:** This test is designed to be used on FFPE samples that have been confirmed by Pathologists to be of the DLBCL subtype with at least 60% tumor content, and have been processed according to the Nanostring SOP. Other samples may produce unexpected or inaccurate results. Although some quality control is incorporated into the algorithm it is recommended that users additionally exclude samples that have been flagged by the NanoString nSolver Analysis software as being of poor quality, or otherwise appear as outliers. This tool has not been evaluated by the FDA or any other regulatory agency, and is not intended for clinical use.

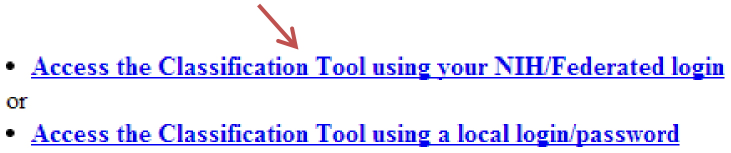
**Direct questions/support issues to llmpp-support@bimas.cit.nih.gov**

*NIH BioInformatics support provided by BIMAS/CBEL/CIT.*
*We can be contacted by email.*

At the bottom of the every page you will find an email link to llmpp-support@bimas.cit.nih.gov . Use this email for any questions, problems or requests related to this Web Site.

**- Accessing the Classification Tool with an NIH Login and Password**

Users who have an NIH Login and Password should click the “Access the Classification Tool using your NIH/Federated login” link.



- Access the Classification Tool using your NIH/Federated login

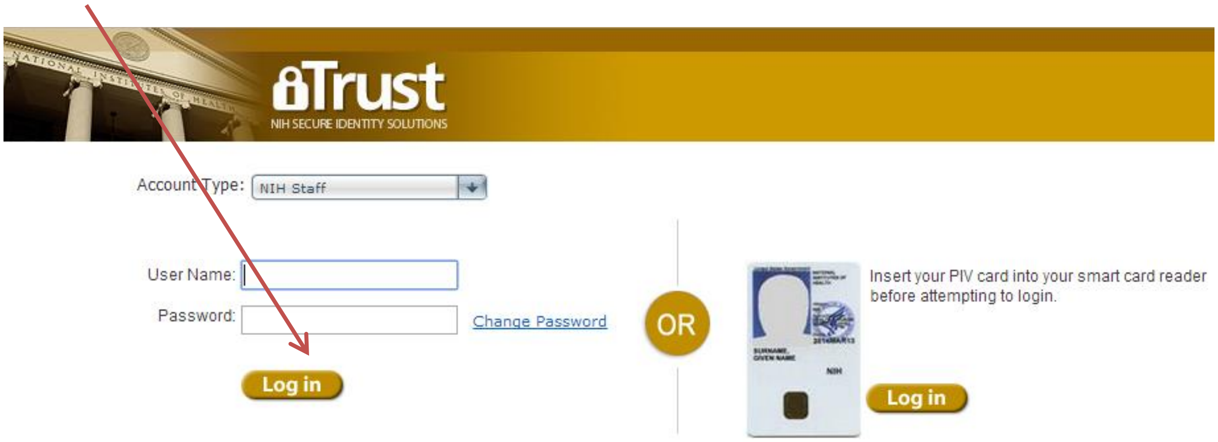
or

- Access the Classification Tool using a local login/password

You will then be directed to the NIH Single Sign On site to authenticate. For the Account Type value, select “NIH Staff”.

Account Type: -- Select Value --

You may authenticate using either your NIH User Name/Password or with your PIV card/PIN.  To authenticate with you NIH User Name and Password, enter your NIH User Name and Password and click the “Log in” button.



Account Type: NIH Staff

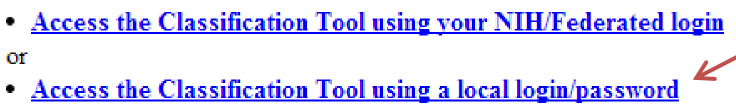
User Name:

Password:

Insert your PIV card into your smart card reader before attempting to login.

### - Accessing the Classification Tool with a provided login and password

Users without an NIH Login and Password have been provided with a user name and password and should click on the “Access the Classification Tool using a local login/password” link.

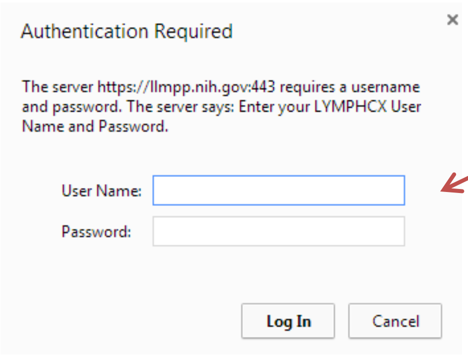


- Access the Classification Tool using your NIH/Federated login

or

- Access the Classification Tool using a local login/password

Depending on your browser, you will be prompted to “Log in” or “Authenticate”. The example below is using the Chrome Browser. The first time you access the site, enter the user name and password information that was provided to you and click the “Log In” button.



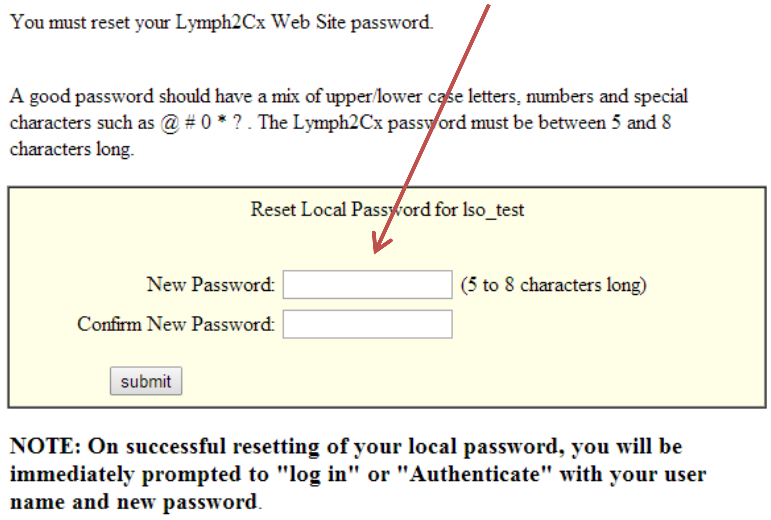
Authentication Required

The server https://llmpp.nih.gov:443 requires a username and password. The server says: Enter your LYMPHCX User Name and Password.

User Name:

Password:

You will then be required to change the password



You must reset your Lymph2Cx Web Site password.

A good password should have a mix of upper/lower case letters, numbers and special characters such as @ # 0 * ? . The Lymph2Cx password must be between 5 and 8 characters long.

Reset Local Password for lso_test

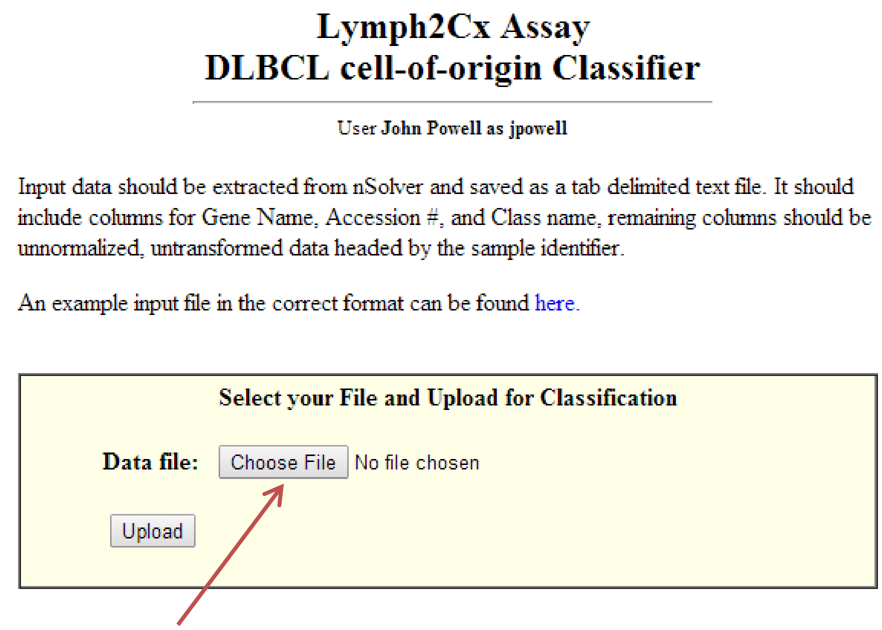
New Password: (5 to 8 characters long)

Confirm New Password:

**NOTE: On successful resetting of your local password, you will be immediately prompted to "log in" or "Authenticate" with your user name and new password.**

After submitting your new password, you will be required to “Log in” or “Authenticate” again with your user name and new password.

**- Uploading Lymph2cx Assay data**



Lymph2Cx Assay
DLBCL cell-of-origin Classifier

User **John Powell** as **jpowell**

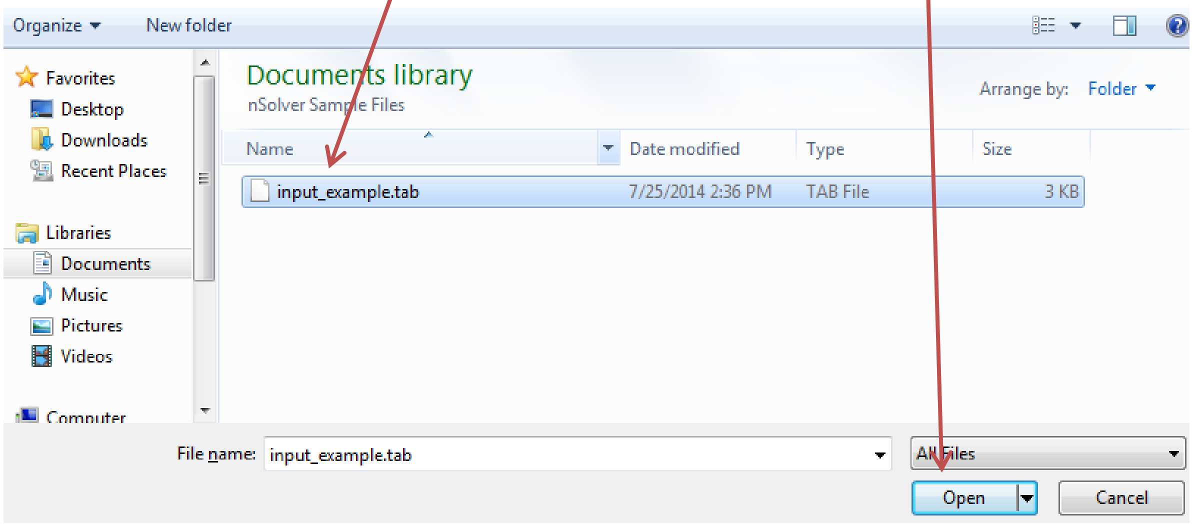
Input data should be extracted from nSolver and saved as a tab delimited text file. It should include columns for Gene Name, Accession #, and Class name, remaining columns should be unnormalized, untransformed data headed by the sample identifier.

An example input file in the correct format can be found here.

**Select your File and Upload for Classification**

**Data file:** Choose File No file chosen

Click on "Choose File" and navigate to and select the TAB delimited file extracted from NanoString's nSolver. In this example, "input_example.tab" has been selected. Click the "Open" button to complete the selection.

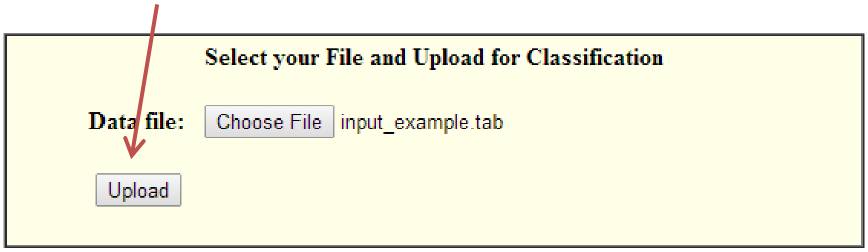


| Name | Date modified | Type | Size |
|---|---|---|---|
| input_example.tab | 7/25/2014 2:36 PM | TAB File | 3 KB |

File name: input_example.tab

All Files

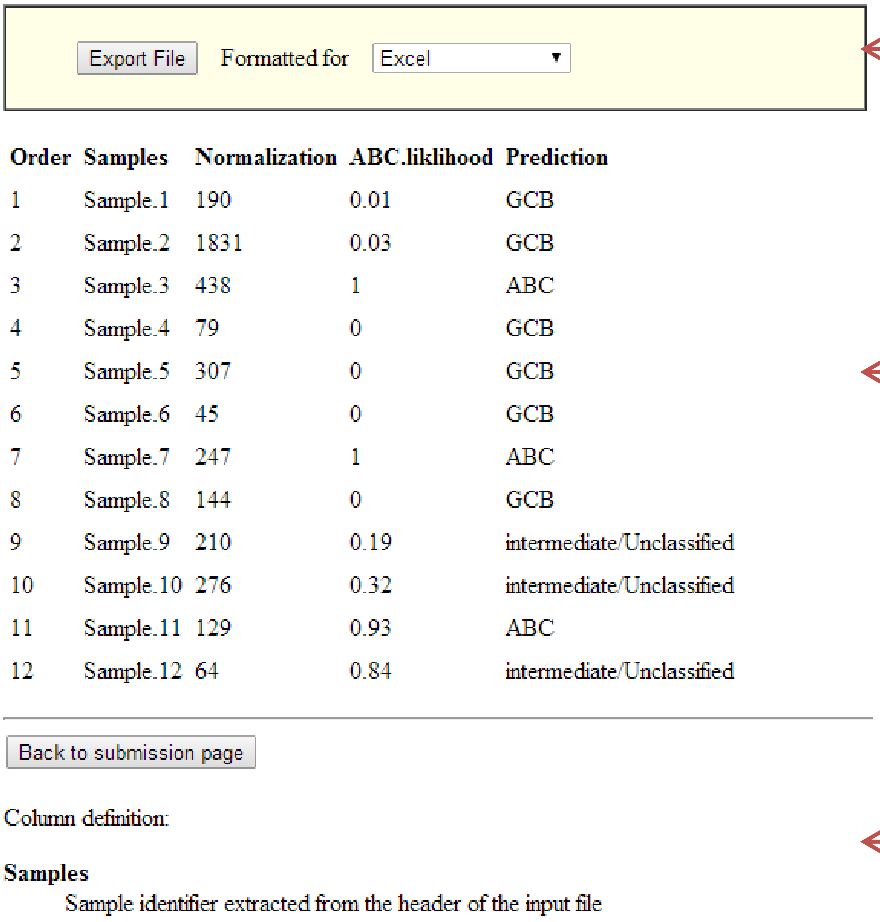
Click the "Upload" button to upload the selected file to the Lymph2Cx assay cell-of-origin classifier



**Select your File and Upload for Classification**

**Data file:** Choose File input_example.tab

**- Lymph2cx Assay cell-of-origin Classifier Results Display**



Form for Retrieving Results

| Order | Samples | Normalization | ABC.liklihood | Prediction |
|---|---|---|---|---|
| 1 | Sample.1 | 190 | 0.01 | GCB |
| 2 | Sample.2 | 1831 | 0.03 | GCB |
| 3 | Sample.3 | 438 | 1 | ABC |
| 4 | Sample.4 | 79 | 0 | GCB |
| 5 | Sample.5 | 307 | 0 | GCB |
| 6 | Sample.6 | 45 | 0 | GCB |
| 7 | Sample.7 | 247 | 1 | ABC |
| 8 | Sample.8 | 144 | 0 | GCB |
| 9 | Sample.9 | 210 | 0.19 | intermediate/Unclassified |
| 10 | Sample.10 | 276 | 0.32 | intermediate/Unclassified |
| 11 | Sample.11 | 129 | 0.93 | ABC |
| 12 | Sample.12 | 64 | 0.84 | intermediate/Unclassified |

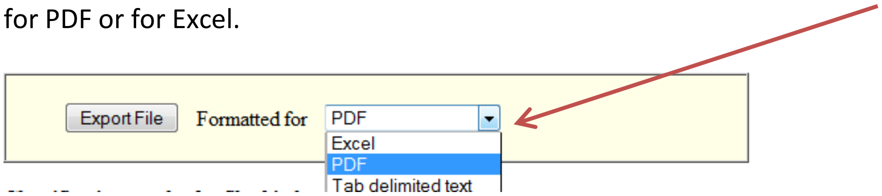
Display of Results

Column definition:

**Samples**
Sample identifier extracted from the header of the input file

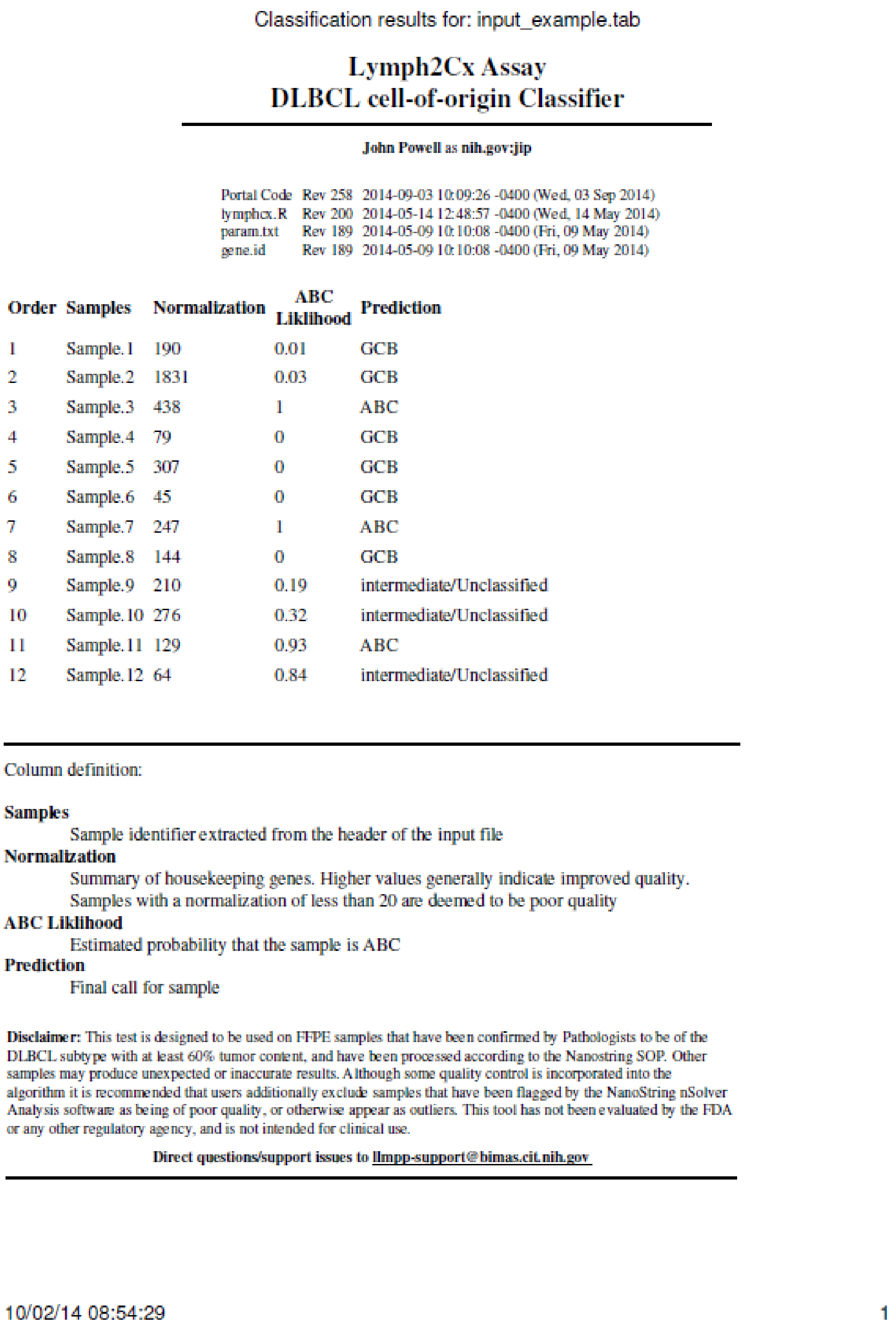
Column Definitions

Use the box at the top of the display to retrieve the prediction results formatted as TAB delimited text, for PDF or for Excel.



The lines in the TAB delimited retrieved files are terminated with a “linefeed” character.  If you use Notepad to view the TAB delimited text, the lines will display “run together”.  Use WordPad or Word to view the files.

## - Lymph2cx Assay cell-of-origin Classifier exported PDF example



Classification results for: input_example.tab

### Lymph2Cx Assay
### DLBCL cell-of-origin Classifier

**John Powell** as **nih.gov:jip**

| | | |
|---|---|---|
| Portal Code | Rev 258 | 2014-09-03 10:09:26 -0400 (Wed, 03 Sep 2014) |
| lymphcx.R | Rev 200 | 2014-05-14 12:48:57 -0400 (Wed, 14 May 2014) |
| param.txt | Rev 189 | 2014-05-09 10:10:08 -0400 (Fri, 09 May 2014) |
| gene.id | Rev 189 | 2014-05-09 10:10:08 -0400 (Fri, 09 May 2014) |

| Order | Samples | Normalization | ABC Liklihood | Prediction |
|---|---|---|---|---|
| 1 | Sample.1 | 190 | 0.01 | GCB |
| 2 | Sample.2 | 1831 | 0.03 | GCB |
| 3 | Sample.3 | 438 | 1 | ABC |
| 4 | Sample.4 | 79 | 0 | GCB |
| 5 | Sample.5 | 307 | 0 | GCB |
| 6 | Sample.6 | 45 | 0 | GCB |
| 7 | Sample.7 | 247 | 1 | ABC |
| 8 | Sample.8 | 144 | 0 | GCB |
| 9 | Sample.9 | 210 | 0.19 | intermediate/Unclassified |
| 10 | Sample.10 | 276 | 0.32 | intermediate/Unclassified |
| 11 | Sample.11 | 129 | 0.93 | ABC |
| 12 | Sample.12 | 64 | 0.84 | intermediate/Unclassified |

Column definition:

**Samples**
Sample identifier extracted from the header of the input file
**Normalization**
Summary of housekeeping genes. Higher values generally indicate improved quality.
Samples with a normalization of less than 20 are deemed to be poor quality
**ABC Liklihood**
Estimated probability that the sample is ABC
**Prediction**
Final call for sample

**Disclaimer:** This test is designed to be used on FFPE samples that have been confirmed by Pathologists to be of the DLBCL subtype with at least 60% tumor content, and have been processed according to the Nanostring SOP. Other samples may produce unexpected or inaccurate results. Although some quality control is incorporated into the algorithm it is recommended that users additionally exclude samples that have been flagged by the NanoString nSolver Analysis software as being of poor quality, or otherwise appear as outliers. This tool has not been evaluated by the FDA or any other regulatory agency, and is not intended for clinical use.

**Direct questions/support issues to llmpp-support@bimas.cit.nih.gov**

## Appendix A

### - What ifs

What if I forget my local user name or password?

> You will need to send an email to llmpp-support@bimas.cit.nih.gov . Reset password information will be send to the email of record when the account was created.

What if I want to change my password?

> You will need to send an email to llmpp-support@bimas.cit.nih.gov . Reset password information will be send to the email of record when the account was created.

What if I want to log out of the Lymph2Cx Portal?

> If you logged in with an NIH/Federated login, you can click the “Log out” link near the top of the page. If you logged in with a local login, the only way to log out is to completely close your browser.

What if my uploaded Lymph2Cx Assay data file does not meet the requirements?

> The Lymph2Cx classified system does a thorough job of checking the input file for any issues before attempting to run the classification. Any issues detected with the file are reported back to you.

What if the classifier cannot determine the cell-of-origin (COO)?

> If the input was deemed to be to poor quality to attempt classification, the classifier prediction would be “Poor Quality”. If quality is Okay, the classifier returns “Intermediate/Unclassified” for those samples cannot be classified ABC or GCB.

## Appendix B

### - Required format of the Lymph2Cx Assay file extracted from nSolver Analysis Software

- The file must be a TAB-separated text file
- The first line of the file must be a header line. The first 3 header columns must be 'Gene Name', 'Accession #', and 'Class Name'. The remaining header columns are sample identifiers.
- The nSolver measurement values are expected to be zero or positive numbers. Negative values and missing measurements are not allowed.
- The following list of twenty Gene Name & Accession values must be present in the file:

| Gene Name | Accession # |
|---|---|
| ASB13 | NM_024701.3 |
| CCDC50 | NM_174908.3 |
| CREB3L2 | NM_194071.2 |
| CYB5R2 | NM_016229.3 |
| IRF4 | NM_002460.1 |
| ISY1 | NM_020701.2 |
| ITPKB | NM_002221.3 |
| LIMD1 | NM_014240.2 |
| MAML3 | NM_018717.4 |
| MME | NM_000902.2 |
| MYBL1 | XM_034274.14 |
| PIM2 | NM_006875.2 |
| R3HDM1 | NM_015361.2 |
| RAB7L1 | NM_001135664.1 |
| S1PR2 | NM_004230.2 |
| SERPINA9 | NM_001042518.1 |
| TNFRSF13B | NM_012452.2 |
| TRIM56 | NM_030961.1 |
| UBXN4 | NM_014607.3 |
| WDR55 | NM_017706.4 |

## - Example Lymph2Cx Assay File containing 6 samples

| Gene Name | Accession # | Class Name | Sample.1 | Sample.2 | Sample.3 | Sample.4 | Sample.5 | Sample.6 |
|---|---|---|---|---|---|---|---|---|
| ASB13 | NM_024701.3 | Endogenous | 212 | 3639 | 111 | 876 | 617 | 42 |
| CCDC50 | NM_174908.3 | Endogenous | 634 | 4898 | 1360 | 242 | 345 | 205 |
| CREB3L2 | NM_194071.2 | Endogenous | 29 | 1401 | 805 | 19 | 91 | 180 |
| CYB5R2 | NM_016229.3 | Endogenous | 150 | 2236 | 34 | 189 | 37 | 8 |
| IRF4 | NM_002460.1 | Endogenous | 1349 | 11641 | 1934 | 291 | 794 | 831 |
| ISY1 | NM_020701.2 | Endogenous | 121 | 1280 | 208 | 71 | 230 | 22 |
| ITPKB | NM_002221.3 | Endogenous | 823 | 6800 | 690 | 227 | 1623 | 37 |
| LIMD1 | NM_014240.2 | Endogenous | 221 | 1275 | 403 | 22 | 105 | 112 |
| MAML3 | NM_018717.4 | Endogenous | 65 | 194 | 21 | 151 | 75 | 45 |
| MME | NM_000902.2 | Endogenous | 390 | 2699 | 5 | 99 | 36 | 536 |
| MYBL1 | XM_034274.14 | Endogenous | 190 | 3547 | 19 | 101 | 54 | 15 |
| PIM2 | NM_006875.2 | Endogenous | 7140 | 23675 | 2191 | 483 | 1228 | 3721 |
| R3HDM1 | NM_015361.2 | Endogenous | 310 | 2558 | 578 | 99 | 449 | 63 |
| RAB7L1 | NM_001135664.1 | Endogenous | 493 | 5748 | 4646 | 319 | 385 | 163 |
| S1PR2 | NM_004230.2 | Endogenous | 168 | 2059 | 144 | 255 | 540 | 341 |
| SERPINA9 | NM_001042518.1 | Endogenous | 326 | 998 | 1 | 330 | 76 | 1528 |
| TNFRSF13B | NM_012452.2 | Endogenous | 150 | 2653 | 1490 | 21 | 192 | 1 |
| TRIM56 | NM_030961.1 | Endogenous | 49 | 767 | 280 | 16 | 110 | 9 |
| UBXN4 | NM_014607.3 | Endogenous | 898 | 4412 | 986 | 208 | 823 | 294 |
| WDR55 | NM_017706.4 | Endogenous | 150 | 1857 | 484 | 130 | 290 | 52 |
| NEG_A | ERCC_00096.1 | Negative | 0 | 0 | 0 | 0 | 1 | 1 |
| NEG_B | ERCC_00041.1 | Negative | 0 | 1 | 1 | 1 | 2 | 0 |
| NEG_C | ERCC_00019.1 | Negative | 1 | 4 | 1 | 1 | 0 | 0 |
| NEG_D | ERCC_00076.1 | Negative | 0 | 7 | 1 | 1 | 0 | 1 |
| NEG_E | ERCC_00098.1 | Negative | 1 | 0 | 2 | 2 | 1 | 2 |
| NEG_F | ERCC_00126.1 | Negative | 0 | 2 | 0 | 0 | 0 | 0 |
| POS_A | ERCC_00117.1 | Positive | 26180 | 30754 | 31312 | 34199 | 30995 | 20442 |
| POS_B | ERCC_00112.1 | Positive | 6221 | 7901 | 7041 | 7836 | 7998 | 5102 |
| POS_C | ERCC_00002.1 | Positive | 1538 | 1708 | 2155 | 2320 | 2068 | 1257 |
| POS_D | ERCC_00092.1 | Positive | 535 | 536 | 558 | 498 | 572 | 325 |
| POS_E | ERCC_00035.1 | Positive | 99 | 192 | 144 | 165 | 138 | 81 |
| POS_F | ERCC_00034.1 | Positive | 22 | 34 | 35 | 31 | 28 | 18 |