Custom Module: Flat File Export

Version: 1.2.0

Description: Export Flat files used in Seurat's LoadNanostring function

countMatrix, cellMetadata, transcripts, & polygons are required for the LoadNanostring function

[Vignette](https://satijalab.org/seurat/articles/spatial_vignette_2#human-lung-nanostring-cosmx-spatial-molecular-imager) to analyze data. **Flat File Export v1.2.0 works on Seurat v4.3+**

Setup:

|  |  |  |  |
| --- | --- | --- | --- |
| R version | CPU cores | RAM (GB) | Max run (h) |
| Any | 16 | 128+ | 4+ |

\* RAM and Max run time listed above are suggested minimums. Values may be increased (note that increasing RAM or Max run time may increase costs). If the module exceeds either value, it will immediately fail.

Variables:

Name must be spelled exactly as shown for script to work.

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Type | Name | Display name | Range | Default | Required |
| BOOL | countMatrix | Generate count matrix file | 0-1 | 1 | Yes |
| BOOL | cellMetadata | Generate cell metadata file | 0-1 | 1 | Yes |
| BOOL | transcripts | Generate transcripts file | 0-1 | 1 | Yes |
| BOOL | polygons | Generate cell boundaries | 0-1 | 1 | Yes |
| BOOL | fovPositions | Generate FOV position file | 0-1 | 0 | Yes |
| BOOL | gzip | gzip files, does not affect LoadNanostring function | 0-1 | 1 | Yes |
| STRING | studyName | Output Folder Name | 1-100 |  | Yes |
| STRING | outPath | Destination S3 file path | 5-300 |  | Yes |
| PRIVATE | access\_key | Destination AWS access key | 16-128 |  | Yes |
| PRIVATE | secret\_key | Destination AWS secret key | 1-300 |  | Yes |
| STRING | s3Region | Destination AWS region | 8-16 |  | Yes |
| PRIVATE | session\_token | Destination AWS session token, if configured | 1-1200 |  | No |

In AtoMx™ v1.3.2, PRIVATE parameters have been introduced. PRIVATE parameters are protected string variables and will not be displayed in the user interface (UI) or logged in any system logs.

If you do not see PRIVATE as a parameter type, it indicates that your AtoMx version has not yet been upgraded to v1.3.2. For older releases where PRIVATE parameters are unavailable as an option, please select STRING instead.

Packages: None

Outputs:

* Count matrix flat csv file
* Cell metadata flat csv file
* Global transcripts flat csv file
* Global cell boundaries flat csv file
* Global FOV position file

Gzip:

If gzip was selected in the module parameters, exported files will be in a compressed format with the suffix .gz and must be unzipped by running from a command line (gunzip is not a natively installed package on Windows cmd). Windows explorer might remove the .gz suffix on file but this does not mean they have been decompressed. Files in a gzip format cannot be opened in Excel.

A screenshot of a computer

Description automatically generated

On a Windows computer:

If you want to view the data in Excel, it is recommended to unselect gzip in the flat file export settings. If you are reading into R/Seurat, it is recommended to select gzip to compress the files.

On a Mac/Linux computer:

It is recommended to select gzip to compress the files. If you want to view the data in Excel, use on the command line before viewing.

FOV Images for packages like Giotto -

If you are using a package like Giotto that require FOV images, the images can be retrieved using the [normal export custom module](https://github.com/Nanostring-Biostats/CosMxDACustomModules/tree/main/Export) with the and options selected. After exporting, run the following cmd commands to get the files in the expected folder structure. Variables in **[**brackets**]** indicate the variable will change based on your setup.

**cd** **[**flatFile\_folder**]**

**mkdir** CellLabels CompartmentLabels

**cp** **[**exported\_flowcell\_folder**]/**CellStatsDir**/**Cell**\*/ .** -r

**cp** **[**exported\_flowcell\_folder**]/**CellStatsDir**/**FOV**\*/**CellLabels**\*** CellLabels

**cp** **[**exported\_flowcell\_folder**]/**CellStatsDir**/**FOV**\*/**CompartmentLabels**\*** CompartmentLabels