Reprocessing Bruderer Data With Skyline Batch

This tutorial covers how to use Skyline Batch to import and run two configurations using the Bruderer 2015 data.

If you have not already downloaded Skyline, Skyline Batch, the Skyline Batch configuration file (Bruderer.bcfg), and the R script (install_R_packages.R), you can find and download them here:

https://panoramaweb.org/dia-bruderer15.url

Setting Up R

Check if you have a "C:\Program Files\R\ R-4.0.3" folder. If you do not, go to:

Index of /bin/windows/base/old/4.0.3 (r-project.org)

> *Note: You can use an R-4.0.3 installation located somewhere else on your computer. However, you will have to add your R directory to Skyline Batch.

Install R-4.0.3 from the R-4.0.3-win.exe link. When it is done downloading, the setup installer will open.

- Click the **Next** button until you see a green Installing progress bar.
- Click the **Finish** button.

Go to the Rtools website:

Building R for Windows (r-project.org)

- Click on "Rtools35.exe" to begin downloading the correct version of Rtools.
- Click the **Next** button until you reach **Install** to accept the default settings.
- Click the **Install** button.
- Click the **Finish** button.

If you do not have Rstudio, download it here:

Download the RStudio IDE – Rstudio

- Install it, accepting the default settings.
- Open RStudio.
- Go to **Tools** and click **Global Options**.
- Select the **General** tab on the left-hand side.
- Check if **R version** is showing R-4.0.3
  - If it is:
    - Click **OK**
  - If it is not:
    - Click **Change**
    - Select **R-4.0.3**

- Click **OK** twice
- Restart RStudio

You are now ready to start downloading packages in R-4.0.3

- In RStudio, click **File** > **Open File**
- Navigate to your downloads folder and double-click on "install_R_packages.R" to open it.
- Select line 2, then click the **Run** toolbar button. (Ctrl-Enter)

You should see this output if the download was successful:

```
package 'gplots' successfully unpacked and MD5 sums checked
package 'dplyr' successfully unpacked and MD5 sums checked
```

- Select lines 4-5, then click the **Run** toolbar button. (Ctrl-Enter)

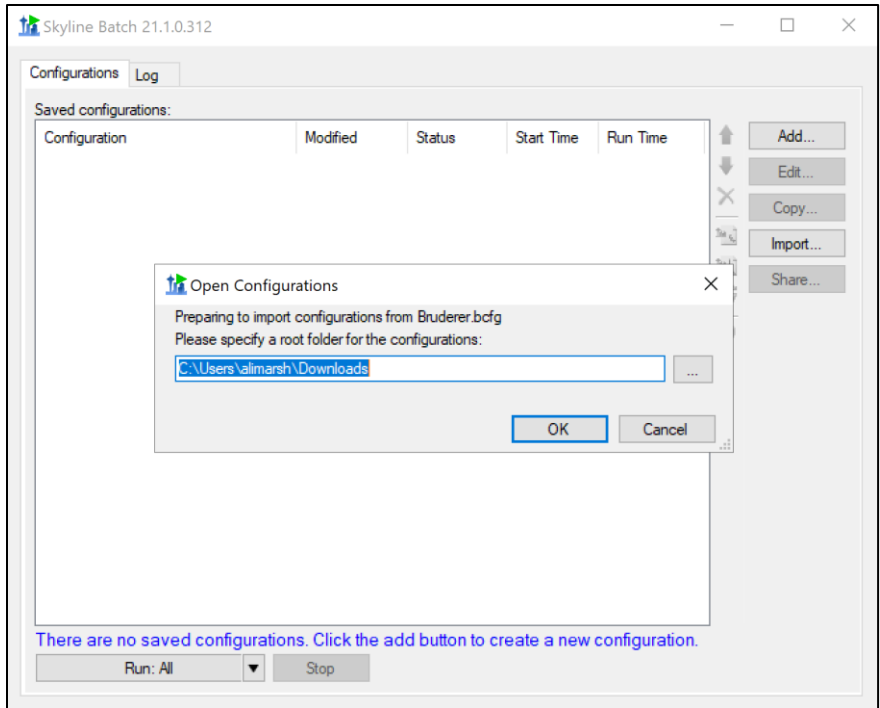You should see this output if the download was successful:

```
package 'BiocManager' successfully unpacked and MD5 sums checked

package 'MSstats' successfully unpacked and MD5 sums checked
```

Opening Configurations

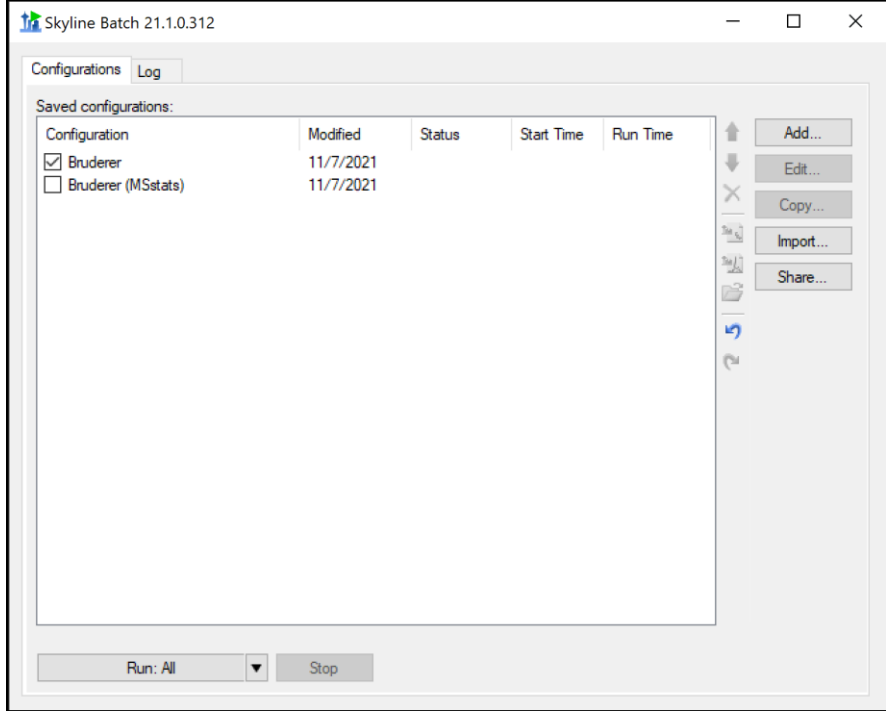- Navigate to your Downloads folder and double-click on "Bruderer.bcfg" to open it.

*Note: If Skyline Batch is already open, you will need to close the program before double-clicking the "Bruderer.bcfg" file.

This will open Skyline Batch and start importing the configurations. You will see a dialog asking for a root folder:

- Click the folder explorer (**…**) button and create a "Bruderer" folder on a drive with at least 70GB of free space.
- Click **OK**.

This will save the configuration file inside the Bruderer folder and import the configurations. You will see the imported configurations in the configuration list:

Running the Configurations

The first configuration is checked, meaning it will be run when the run button is clicked.
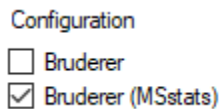
- Click the **Run All Steps** button.

Skyline Batch brings you to the **Log** tab to show the output of running the configurations. It will download files, import data, and export reports for the configuration.

When the configuration has finished running, it will display a **Completed** status in the main window. Wait until the Skyline Batch run has completed.
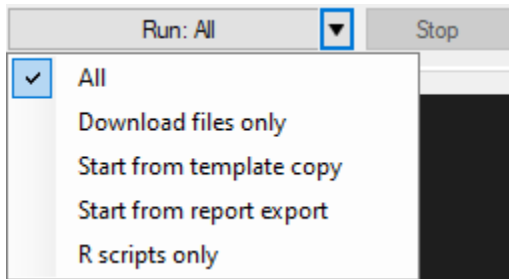
To run the MSstats processing, we can run the MSstats configuration from the "R scripts only" step.

- Uncheck the Bruderer configuration.
- Check the Bruderer (MSstats) configuration.

The configurations should now look like this:

- Click the downward arrow next to **Run all steps** to expand the run options.

- Click **R scripts only**.

This will run the MSstats script for Bruderer (MSstats).

When the Skyline Batch has finished running, view the output files by clicking on a configuration to select it, and using the following buttons:

 Opens the Skyline template document

 Opens the Skyline results document

 Opens the analysis folder

If you want to learn more about Skyline Batch, visit the documentation at:

SkylineBatchDocumentation.pdf