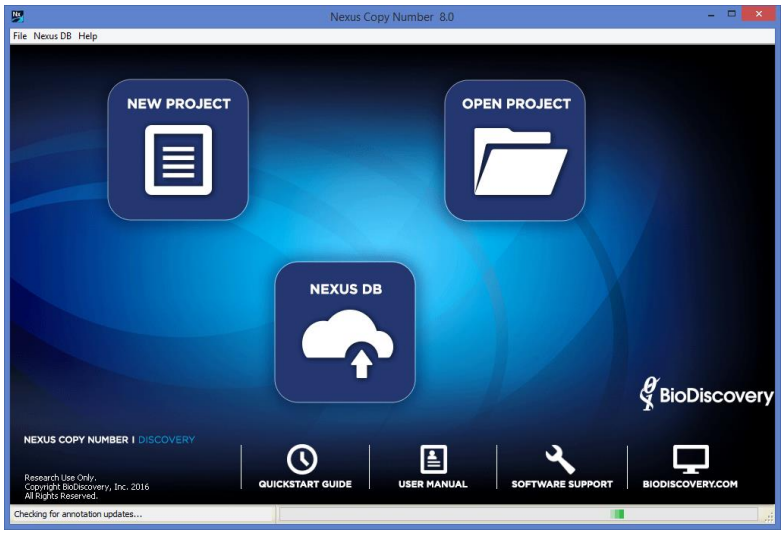
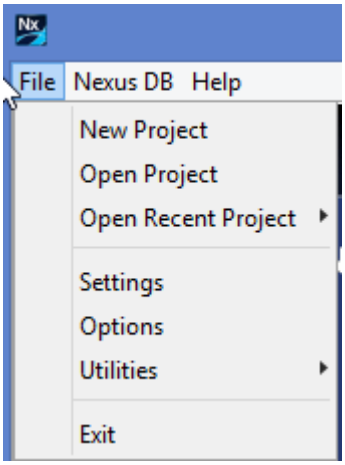
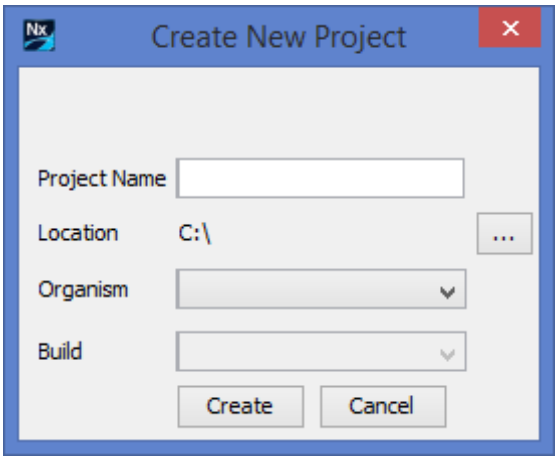
To Process Agilent (CGH+SNP array) Data Generated with Feature Extraction:

1. Launch Nexus Copy Number. Create a new project or open an existing project from the start page. You can also open and create projects from the **File** menu.

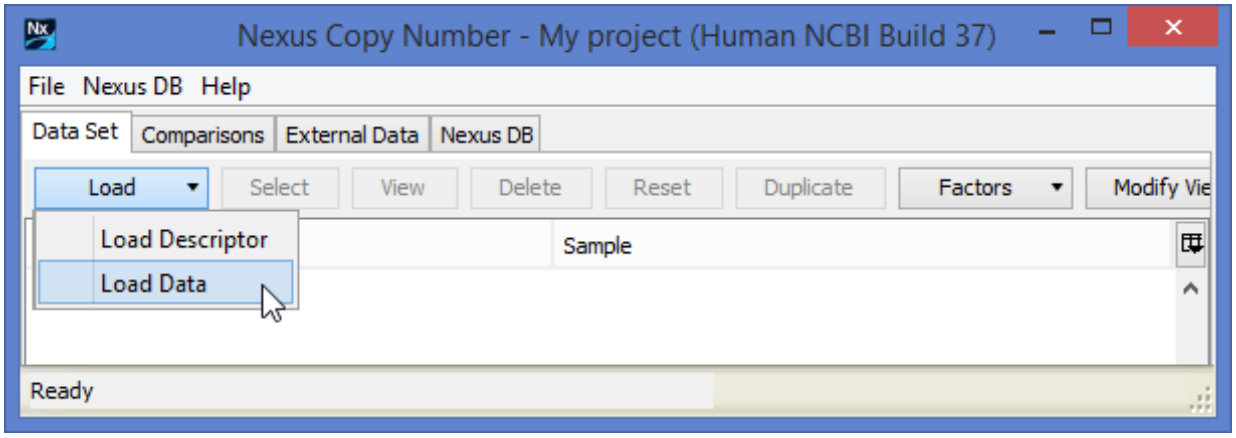




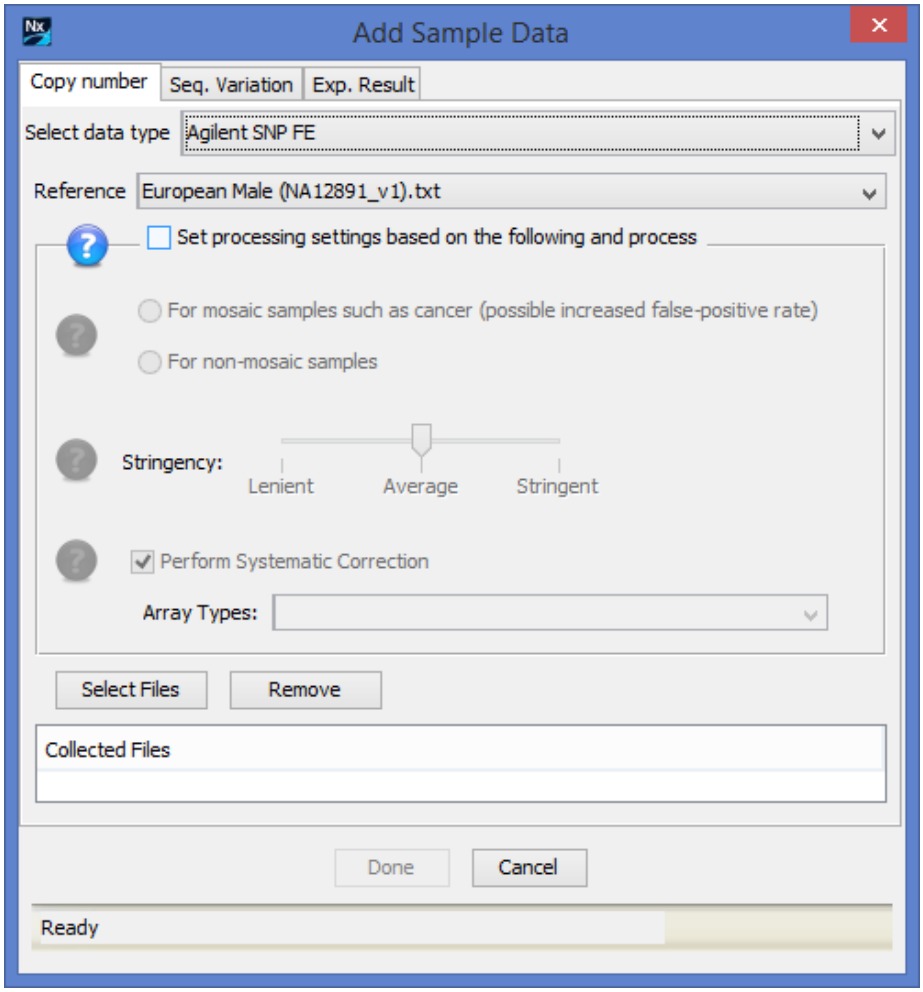
2. Specify a name for your project, a place to save it on your computer, a genome and then the appropriate genome build based on the mapping information for your probes.



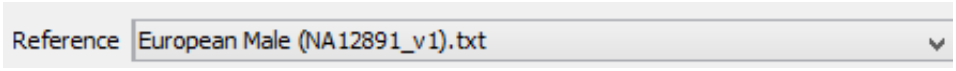
3. Select **Load -> Load Data** from the Data Set tab.



4. In the **Add Sample Data** window, select **Agilent SNP FE** from the "Select data type" drop down in the "Copy Number" tab. A number of options will be displayed. Clicking on the  icon will provide more details on the parameters.

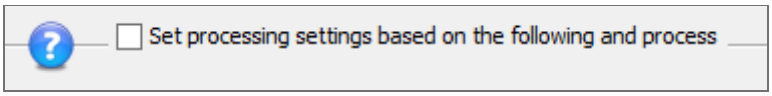


a. Select the reference file you wish to use. If the reference file you require is not listed, contact BioDiscovery Support (support@biodiscovery.com) to obtain the necessary files.



b. The option **Set processing settings based on the following and process** uses default parameters for the type of data you are loading and immediately begins processing after data loading is complete.

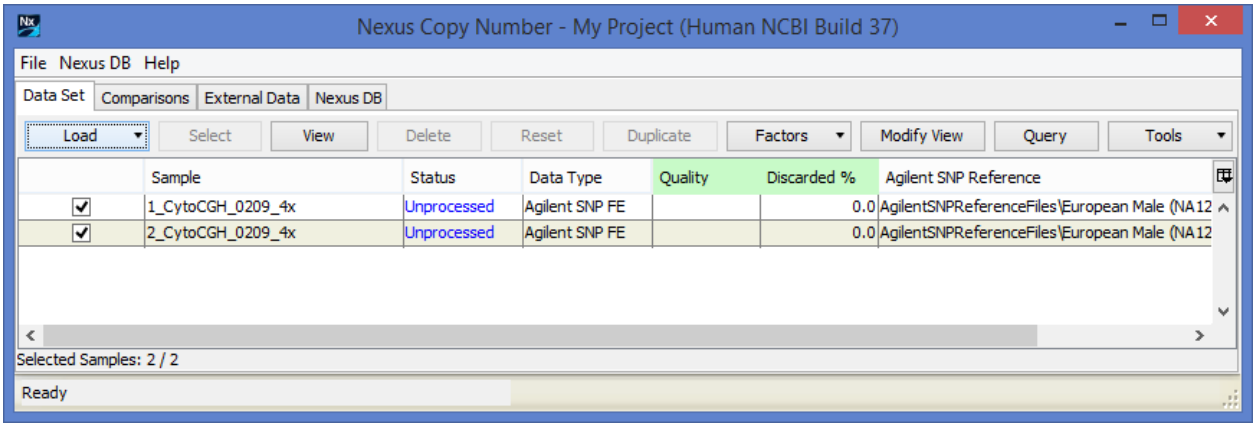
For the **Agilent SNP FE** data type, **you must uncheck the box** as you need to add a factor to the samples after data loading and before processing can commence.



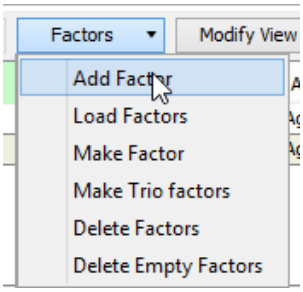
c. Click **Select Files** to select your Agilent Feature Extraction files to load into Nexus Copy Number. You can select any number of files but they must all be Agilent Feature Extraction files from Agilent CGH+SNP arrays. Once you have finished selecting files via the File Chooser, click **Open** and the sample file names will be listed in the **Collected Files** section:

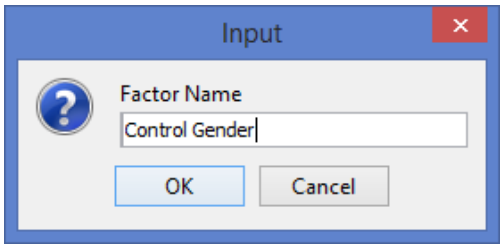
Click **Done** after you have selected all your files.

5. Samples will be loaded but not processed and will appear in the Data Set tab with Status as “Unprocessed”:

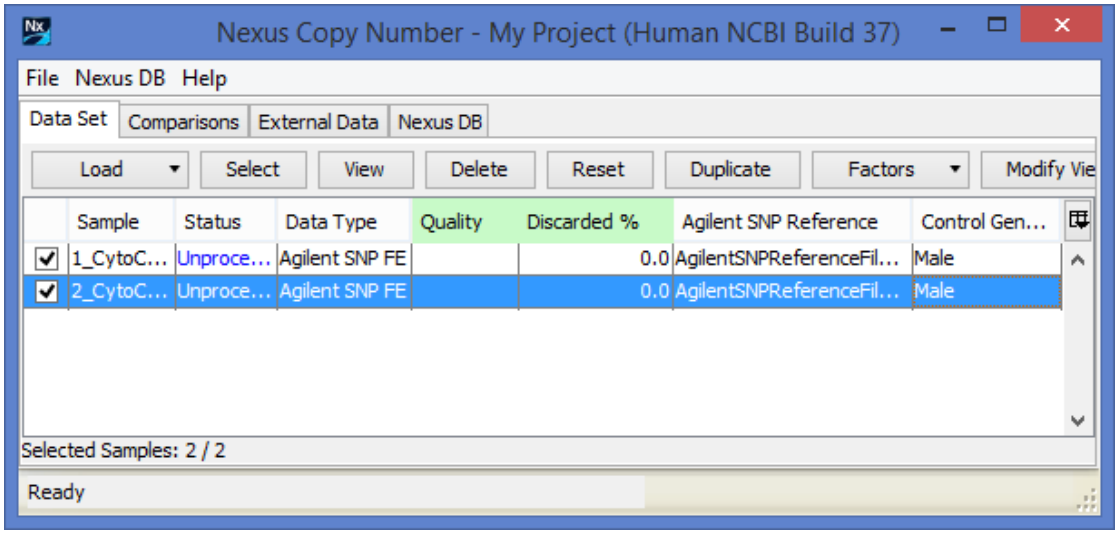


6. You must add a factor called **Control Gender** and specify the control gender for your samples. Without this value, samples will not be processed. From the Data Set tab, click on the **Factors** button, select **Add Factor** and an Input Dialog will open asking you to give the factor a name. Enter **Control Gender** and click **OK**.



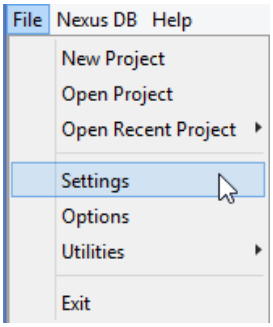


The new column shows up at the end of the table. You can either enter values manually by clicking on each cell and typing or you can copy and paste a column from a spreadsheet. See the User Manual for additional details on adding factors.

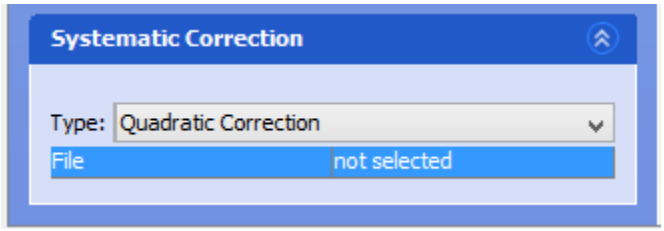


7. It is recommended to apply Systematic Correction (GC wave correction). **Systematic Correction** corrects the wavy pattern often present in the probe distribution. Since the correction is based on probes used, the specific array is needed.

   a. Open the Settings window via the **File** menu.

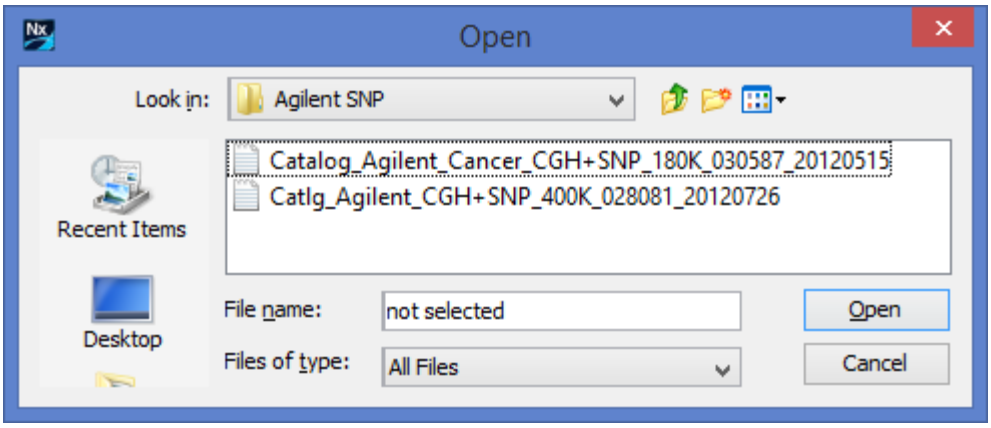


b. Scroll down to the Systematic Correction section. Choose the **Type** of correction (linear, quadratic, etc.) from the dropdown.



c. Select the correction file to use by clicking on the field displaying "not selected". A file chooser window will pop up. Navigate to the Systematic Correction folder (based on the organism and build for your project) in the Nexus Copy Number installation directory.

E.g. …\Nexus 8.0\Organisms\Human\NCBI Build 37\SystematicCorrection\Agilent SNP



Select the appropriate file and click **Open**. If a correction file for your array type is not here, please contact BioDiscovery Support (support@biodiscovery.com) to obtain the necessary files. You can continue to process without performing the correction; just uncheck the box. You can reprocess the samples later with systematic correction applied.

d. To adjust other settings from the default parameters, see the section **Settings** in the **User Manual** for more details. Once all settings are adjusted, click **Done** to apply those settings. Then click on **View** in the Data Set tab to process and see the results.