



BIONUMERICS Tutorial:

Importing antibiotics data

1 Aims

In BIONUMERICS a special *Antibiotics susceptibility plugin* is available. With this plugin, *zone diameter interpretive standards* and *equivalent minimal inhibitory concentration (MIC) breakpoints* can be imported from external files into BIONUMERICS. Based on these standards, imported antibiotics data is then automatically converted to categories. In this tutorial you will learn how to install the *Antibiotics susceptibility plugin* and how to import standards and antibiotics data in a BIONUMERICS database.

2 Sample data

Example data that will be used in this tutorial can be downloaded from the Applied Maths website (<https://www.applied-maths.com/download/sample-data>, click on "Antibiotics sample data").

3 Preparing the database

1. Create a new database (see tutorial "Creating a new database") or open an existing database.
2. Call the *Plugins* dialog box from the *Main* window by selecting **File > Install / remove plugins...** (⌘P).
3. Select the *Antibiotics susceptibility plugin* from the list in the *Applications tab* and press the **<Activate>** button.
4. The program will ask to confirm the installation of the plugin. Press **<Yes>** and **<OK>** twice.
5. Press **<Exit>** to close the *Plugins* dialog box.
6. Close and reopen the database to activate the features of the *Antibiotics susceptibility plugin*.

The *Antibiotics susceptibility plugin* installs itself in the main menu of BIONUMERICS (see Figure 1) and import routines are activated in the *Import* dialog box (see Figure 2).

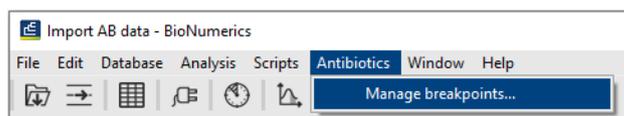


Figure 1: Menu-item in the *Main* window.

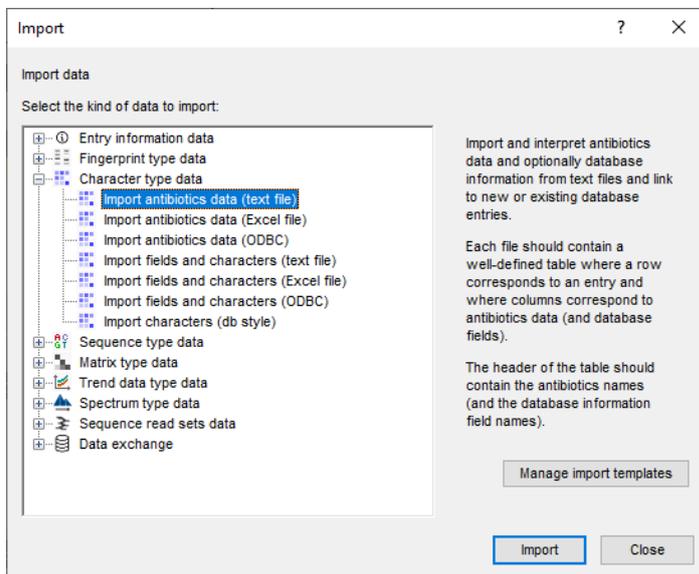


Figure 2: The *Import* dialog box.

4 Managing breakpoints

In order to convert antibiotic resistance data to categories (S, I, and R), we first need to define the cut-off values for the *disk diffusion* experiment (zone diameter standards) or the *MIC* experiment (minimum inhibitory concentration breakpoints) for the bacteria under study. These values can be added either manually or can be imported from a text file.

As an example, we will import (fictitious) MIC breakpoints for *Enterobacteriaceae*. These breakpoints are stored in the `MIC breakpoints.txt` file.

1. Select **Antibiotics** > **Manage breakpoints...** in the *Main* window.
2. Press the <**Add new**> button.
3. In the *Add ABx breakpoints* dialog box, check the **Minimum Inhibitory Concentration (MIC)** option.
4. Enter "Ecoli" in the **Organism** text box and select the <**Create new**> option for **Experiment** (see Figure 3).
5. Press <**Browse...**>, navigate to the downloaded and unzipped `MIC breakpoints.txt` file.
6. Press <**OK**> in the *Add ABx breakpoints* dialog box.

As a result, the *Edit ABx breakpoints* dialog box pops up, displaying the breakpoints from the `MIC breakpoints.txt` text file (see Figure 4).

7. Close the dialogs.

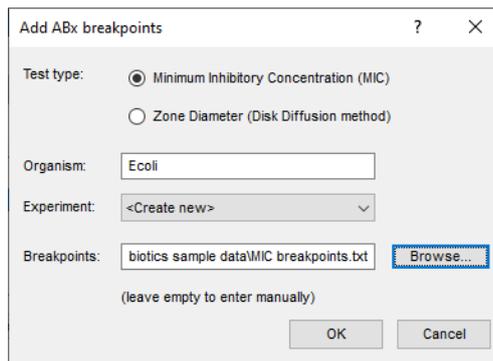


Figure 3: The *Add ABx breakpoints* dialog box.

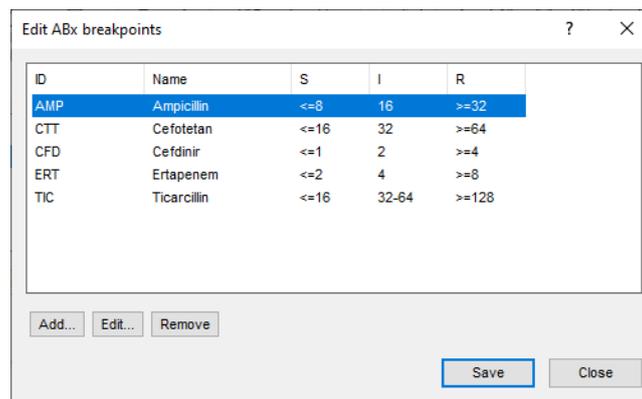


Figure 4: The *Edit ABx breakpoints* dialog box.

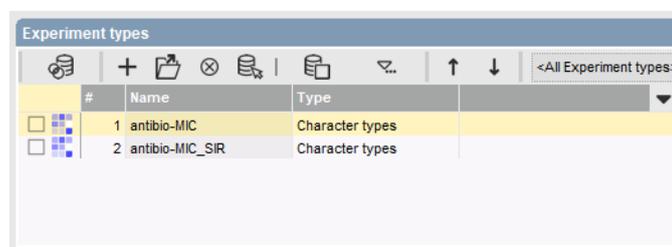


Figure 5: Two new character types.

In the *Experiment types* panel, two new character types are listed (see Figure 5).

8. Double-click on the **antibio-MIC_SIR** experiment type in the *Experiment types* panel.

9. Make sure the *Characters* panel is displayed in the *Character type* window (see Figure 6).

The abbreviations of the antibiotics names are listed in the first column. The **Name** column lists the full names of the antibiotics. This column is used as the default field in the *Comparison* window (column highlighted in pale green; see Figure 6). The 'Min.' and 'Max.' columns list the minimum and maximum categorical values for each category. The default color scale ranges from white (category 0), over green (category 1), over yellow (category 2), to red (category 3).

10. Select the *Mapping* panel in the *Character type* window (see Figure 7).

Four mapping names are defined for the four different categories: N/A (not available) if the categorical value is 0 (absent value); S (susceptible) if the categorical value is 1; I (intermediate) if the

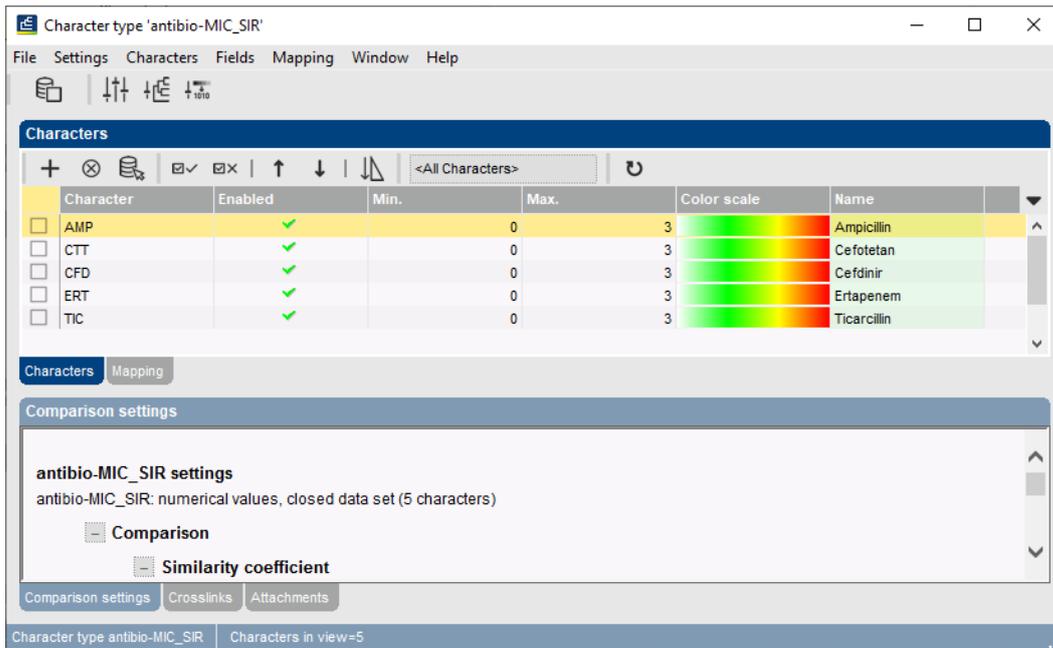


Figure 6: The *Characters* panel.

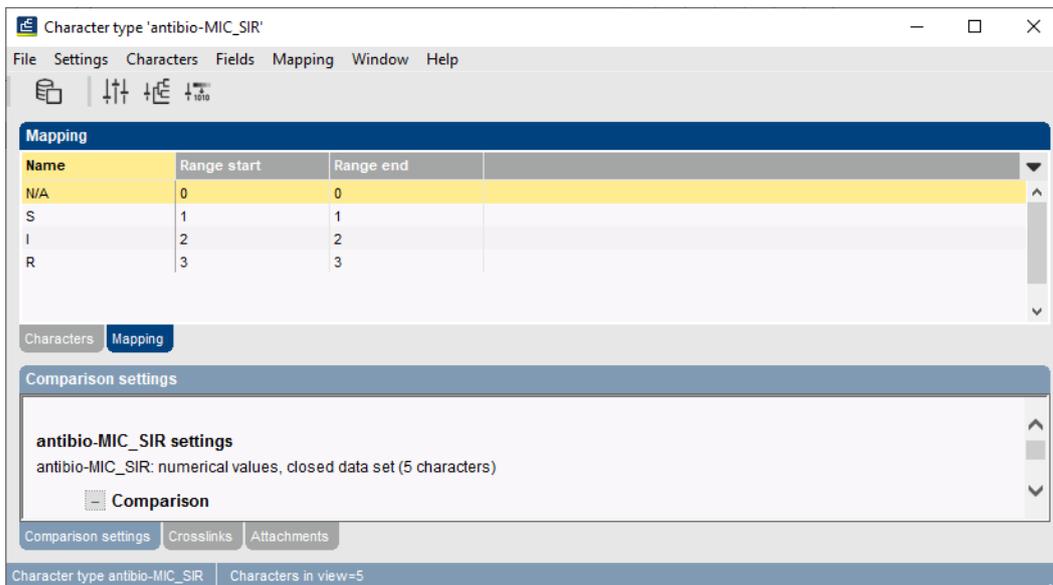


Figure 7: The *Mapping* panel.

categorical value is 2; R (resistant) if the categorical value is 3.

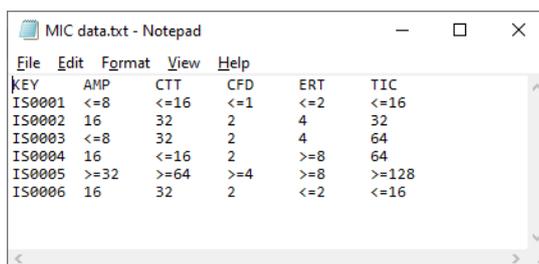
11. Close the *Character type* window with **File > Exit**.

5 Importing antibiotics data

5.1 Compatible data formats

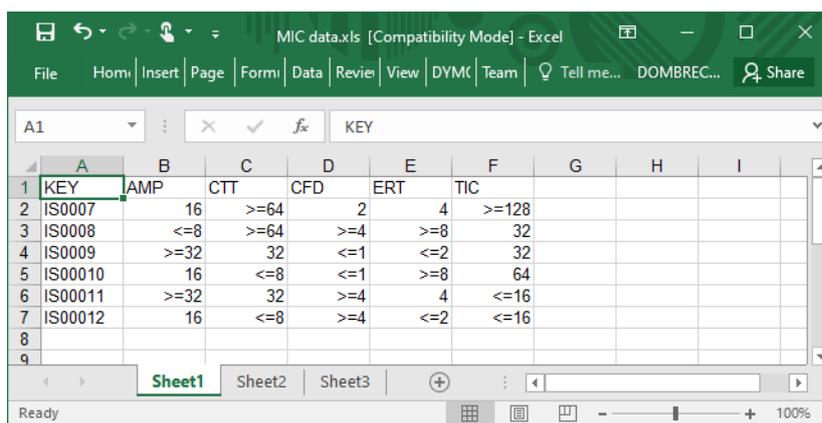
Once the breakpoints are defined in the database, antibiotics data can be imported. During import the antibiotics data is linked to the **antibio-MIC** character type and the data is automatically converted to the correct category (SIR) based on the settings defined in the **antibio-MIC_SIR** character type.

MIC values and inhibition zone diameters can be imported from **text files**, **MS Excel files** or from any **ODBC-compatible source**. In any case, the data should be arranged in a grid containing a header row. One column should contain a strain identifier, which could correspond to the key or a unique database field such as an isolate or strain number. Optionally, one or more columns can contain strain descriptive information. Additional columns should represent MIC values or inhibition zone diameters of individual antibiotics. Greater-or-equal (\geq) or smaller-or-equal signs (\leq) preceding the values are optional. The header row should contain only unique column names, i.e. information field names or antibiotics names or abbreviations. See Figure 8 and Figure 9 for an example of the expected format.



KEY	AMP	CTT	CFD	ERT	TIC
IS0001	<=8	<=16	<=1	<=2	<=16
IS0002	16	32	2	4	32
IS0003	<=8	32	2	4	64
IS0004	16	<=16	2	>=8	64
IS0005	>=32	>=64	>=4	>=8	>=128
IS0006	16	32	2	<=2	<=16

Figure 8: MIC values for *E. coli* entries with key IS0001-IS0006: text file.



KEY	AMP	CTT	CFD	ERT	TIC
IS0007	16	>=64	2	4	>=128
IS0008	<=8	>=64	>=4	>=8	32
IS0009	>=32	32	<=1	<=2	32
IS00010	16	<=8	<=1	>=8	64
IS00011	>=32	32	>=4	4	<=16
IS00012	16	<=8	>=4	<=2	<=16

Figure 9: MIC values for *E. coli* entries with key IS0007-IS00012 : Excel file.

5.2 Import routine

In this section we will import MIC values stored in the text file MIC_data.txt (see Figure 8).

1. Select **File > Import...** (📁, **Ctrl+I**) in the *Main* window to call the *Import* dialog box.

2. Select **Import antibiotics data (text file)** from the Import tree under **Character type data** and press **<Import>**.
3. Press the **<Browse...>** button, browse for the MIC data.txt text file (see Figure 10) and press **<Open>**.

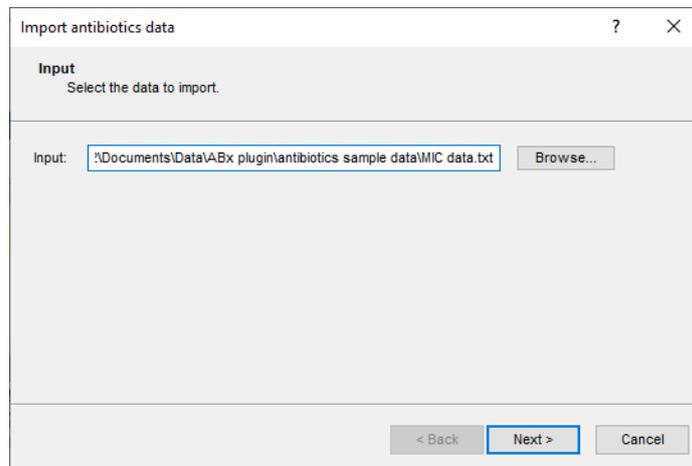


Figure 10: Browse for the text file containing the data.

4. Press **<Next>** in the *Input* dialog box.

Since no import template for antibiotics data is present yet, the *Import rules* dialog box will open.

5. Double-click on the "KEY" row, select **Key** as data destination and press **<OK>**.
6. Click on the "AMP" row, hold the **Shift**-key and click on the "TIC" row to select all antibiotics rows simultaneously.
7. Press **<Edit destination...>**, select **ABX.MIC_Ecoli_antibio-MIC** under **ABx value** (see Figure 11) and press **<OK>**.

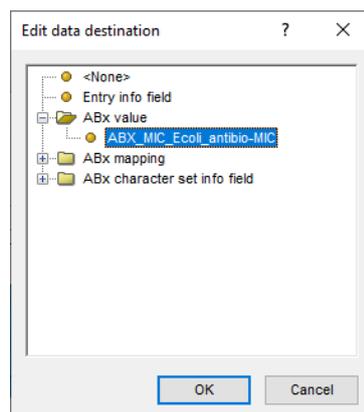


Figure 11: Edit data destination.

BIONUMERICS will automatically link the MIC values to the corresponding antibiotics in the antibiotics experiment via their abbreviations. The *Import rules* dialog box should now look like Figure 12.

8. Press **<Next>**.
9. Leave **Key** checked and press **<Finish>**.

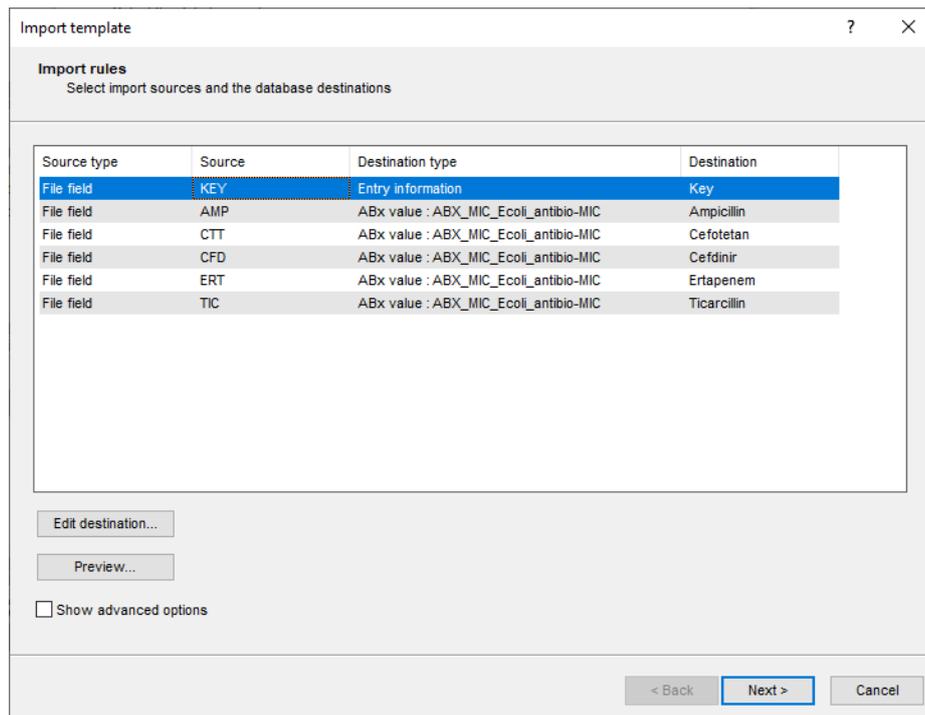


Figure 12: The *Import rules* dialog box, showing the import rules for the MIC data.txt example data file.

10. Enter a name for the import template (e.g. “Sample MIC data”) and press **<OK>**.

The new import template is now added to the database and is listed in the *Antibiotics data import template wizard* page (see Figure 13).

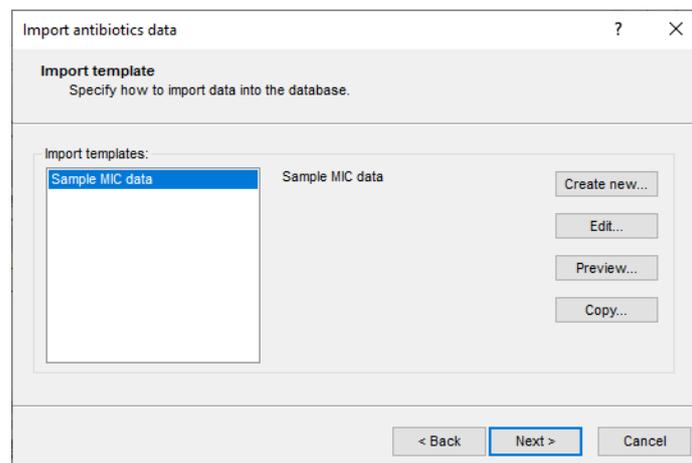


Figure 13: Import template.

11. Press **<Next>**.

The last dialog will indicate that 6 new entries will be added to the database (see Figure 14).

12. Press **<Finish>**.

The entries are imported in BIONUMERICS. Their values (stored in the **antibio-MIC** experiment

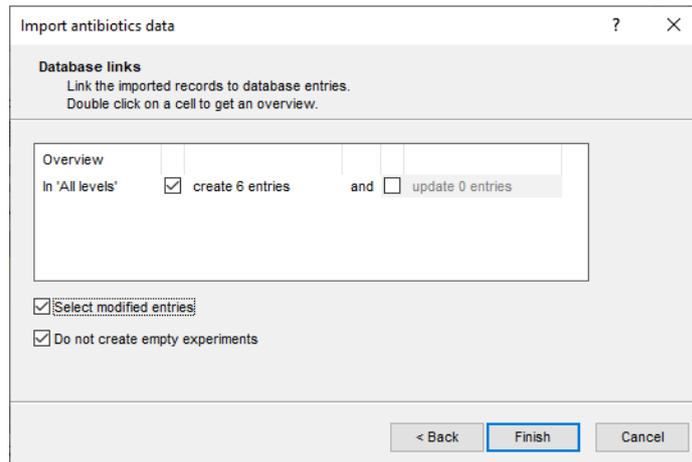


Figure 14: Database links.

type) are automatically converted to categories (stored in the **antibio-MIC_SIR** experiment type).

13. Click on a colored dot for **antibio-MIC_SIR** in the *Experiment presence* panel of the *Main* window.

The *Experiment card* window pops up (see Figure 15).



Figure 15: The *Experiment card* window for the **antibio-MIC_SIR** character experiment.

By default, the tests are shown on a panel using colors, corresponding to the color range set in the *Character type* window: white: absent; green: susceptible; yellow: intermediate; red: resistant.

14. Click on the upper left triangle button to close the *Experiment card* window.
15. In the *Database entries* panel of the *Main* window, make sure all six entries are selected. To select all entries at once, make sure the *Database entries* panel is the active panel and use the **Ctrl+A** shortcut.
16. Highlight the *Comparisons* panel in the *Main* window and select **Edit > Create new object...** (+) to create a new comparison for the selected entries.
17. Click on the  next to the experiment name **antibio-MIC_SIR** in the *Experiments* panel and select **Characters > Show mappings+colors** to display the categories and the corresponding colors in the *Experiment data* panel.
18. Make sure **antibio-MIC_SIR** is selected in the *Experiments* panel and select **Clustering > Calculate > Cluster analysis (similarity matrix)...**
19. Select **Categorical (mappings)** from the list and press **<Next>** and **<Finish>** to start the cluster analysis.

When finished, the dendrogram and the similarity matrix are displayed in their corresponding panels (see Figure 16).

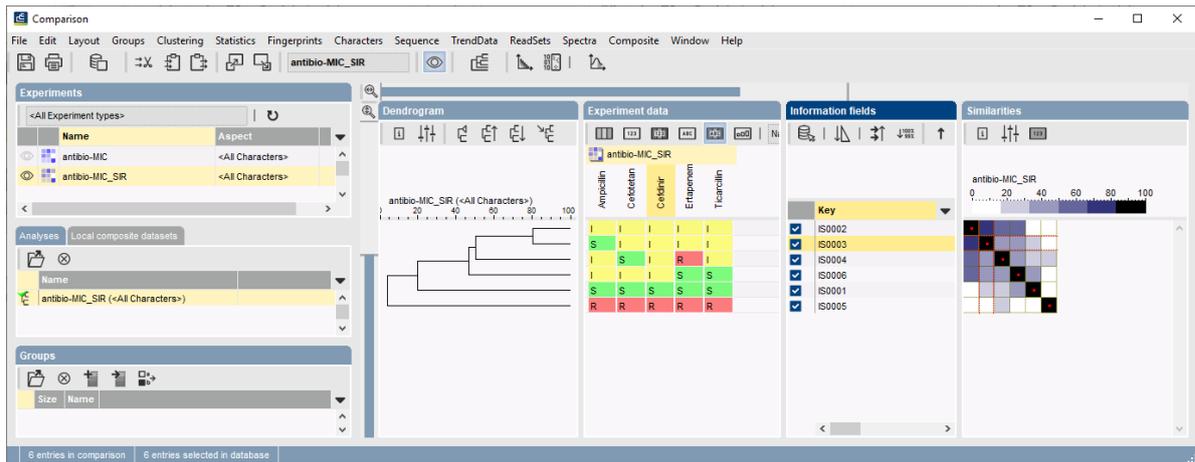


Figure 16: The *Comparison* window.

20. Save the comparison with the dendrogram by selecting **File > Save** (📁, **Ctrl+S**). Specify a name and press **<OK>**.

More information about the *Antibiotics susceptibility plugin* can be found in the manual of this plugin. Detailed information about the *Comparison* window can be found in the reference manual and in the analysis tutorials on our website.