

BioNumerics Tutorial:

Creating an MLVA typing scheme

1 Preparing the database

1. Create a new database, install the *MLVA plugin* and create an MLVA scheme as described in the tutorial: "Setup of an MLVA scheme".
2. Import and pre-process the VNTR sequencer sample trace files as described in the tutorial "Importing and processing VNTR capillary electrophoresis data".
3. Calculate and assign VNTR copy numbers for the imported data based on the steps described in the tutorial "Calculating and assigning VNTR copy numbers".

2 Setting up an MLVA typing

1. Click somewhere in the *Database entries* panel to make it the active panel and select **Edit > Select all (Ctrl+A)** to select the 20 entries in the database.
2. In the *Main* window, select **MLVA > MLVA management window...** (🔗) to call the *MLVA management* window.
3. Select **Edit > Typing schemas > Add typing schema...** to call the *Add typing schema* dialog box.
4. Specify a *Name*, e.g. **My Typing Scheme**, select all (or a subset of) VNTRs using the **Ctrl-** and **Shift-** keys (see Figure 1) and press **<OK>**.

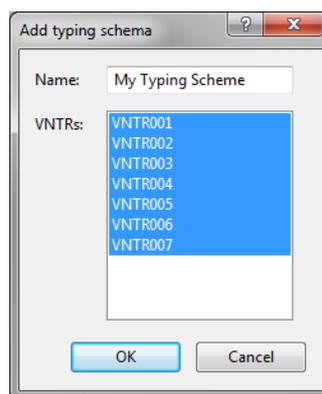


Figure 1: Add a typing scheme.

The MLVA typing scheme is created in the database but no actual typing information is available yet.

5. Select **File > Typing...** (🔗) to call the *Select typing* dialog box and press **<Edit>**.

In this tutorial we will let BioNumerics assign types based on the copy numbers linked to the selected entries in our database.

6. Leave all settings at their default for this exercise and press **<OK>**.

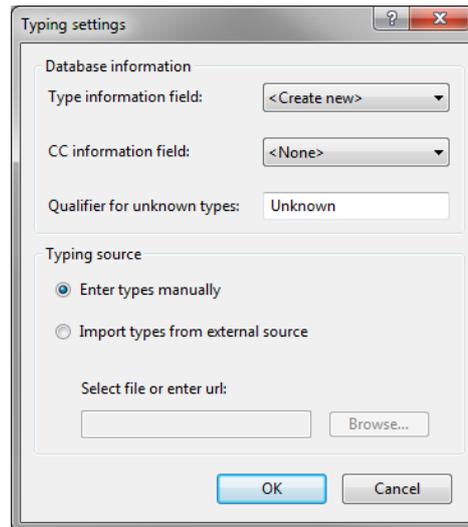


Figure 2: MLVA typing settings.

7. Specify a name (e.g. **MLVA Type**) for the new information field that will hold the MLVA type for each entry (see Figure 3) and press **<OK>**.

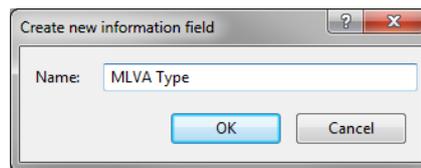


Figure 3: New information field for the storage of the MLVA type.

BioNumerics will ask "Do you want to scan the selected entries for types?".

8. Press **<Yes>**.

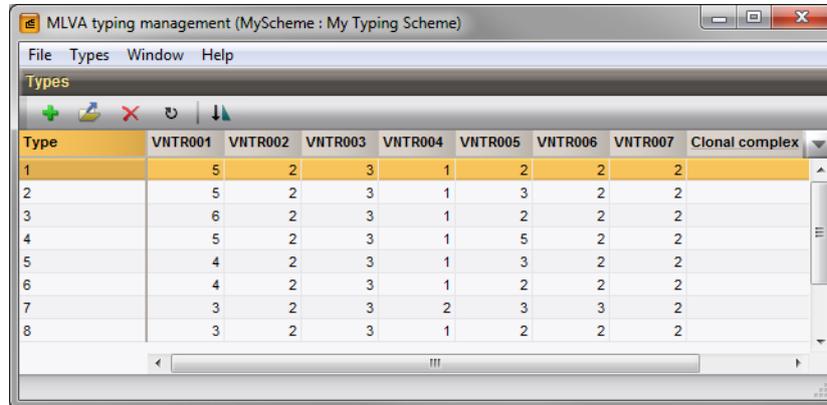
The new types detected in the selected entries are listed in the *Update* dialog box (see Figure 4). In our database, 8 new types are detected.

Hits	Type	VNTR001	VNTR002	VNTR003	VNTR004	VNTR005	VNTR006	VNT
5	1	5	2	3	1	2	2	2
2	2	5	2	3	1	3	2	2
1	3	6	2	3	1	2	2	2
1	4	5	2	3	1	5	2	2
1	5	4	2	3	1	3	2	2
1	6	4	2	3	1	2	2	2
1	7	3	2	3	2	3	3	2
1	8	3	2	3	1	2	2	2

Figure 4: New MLVA types.

9. Press <OK> to add the types to the MLVA typing schema.

The *Typing management* window opens, providing an overview of the types that are currently present in the MLVA typing schema, and the corresponding copy numbers for each of the VNTRs that are included in the typing schema.



Type	VNTR001	VNTR002	VNTR003	VNTR004	VNTR005	VNTR006	VNTR007	Clonal complex
1	5	2	3	1	2	2	2	
2	5	2	3	1	3	2	2	
3	6	2	3	1	2	2	2	
4	5	2	3	1	5	2	2	
5	4	2	3	1	3	2	2	
6	4	2	3	1	2	2	2	
7	3	2	3	2	3	3	2	
8	3	2	3	1	2	2	2	

Figure 5: The *Typing management* window.

10. Close the *Typing management* window and *MLVA management* window.

3 Assigning types

1. In the *Main* window make sure all entries are selected in the *Database entries* panel.
2. Select *MLVA > Perform MLVA typing...* (🔍) in the *Main* window. This action opens the *Perform typing* dialog box (see Figure 6).

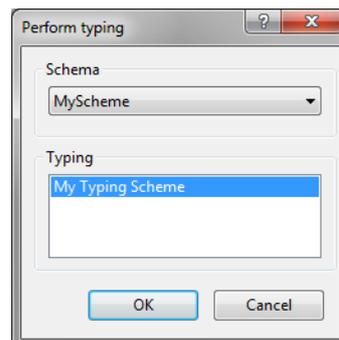


Figure 6: Perform MLVA typing.

3. In this database, only one MLVA scheme and associated MLVA typing scheme is available, so press <OK> to start the typing.

BioNumerics will determine the MLVA types and will add these to the entry information field that was specified for the typing information (in this exercise: **MLVA Type**) (see Figure 7). The text "Incomplete profile" will be filled in when copy numbers are not available for all VNTRs as defined in the MLVA typing schema. For new profiles, i.e. VNTR copy number combinations that are not in the database yet, the text "Unknown" is filled in.

The screenshot displays the BioNumerics software interface. The main window is titled "BioNumerics" and contains several panes:

- Database entries:** A table with columns for Key, Modified date, MLVA Type, and 13 lanes (1-13). The MLVA Type column shows values like "Incomplete profile", "2", "4", "7", "1", "1", "Incomplete profile", "6", "1", "8", "1", "Incomplete profile", "Incomplete profile", "1", "3", "Incomplete profile", "2", "5".
- Experiment types:** A table with columns for #, Name, and Type. It lists 13 experiment types, including "MyScheme_vals", "MyScheme_frags", "MLVA", and various "MLVAMP" types (e.g., MLVAMP16-FAM, MLVAMP1LZ, MLVAMP1NED, MLVAMP1PET, MLVAMP1VIC, MLVAMP26-FAM, MLVAMP2LZ, MLVAMP2NED, MLVAMP2PET, MLVAMP2VIC).
- Fingerprint files:** A table with columns for File name, Experiment type, Link, Modified date, and Source. It lists files like "Batch1_LIZ", "Batch1_6-FAM", "Batch1_VIC", "Batch1_NED", and "Batch1_PET", all with "MLVA" as the experiment type and "Curve" as the source.
- Comparisons:** A section with a table for Name, Modified date, Level, and Number of entries. Below it are tabs for "Comparisons", "Decision networks", "Alignments", "Chromosome comparisons", "Annotations", and "BLAST projects".

The status bar at the bottom indicates: "Database: MLVA tutorial 2 (_DefaultUser_) Entries: Loaded=20, View=20, Selected=0 13 experiments C:\Users\Public\Documents\BioNumerics\Data 75\MLVA tutorial 2 This is a time limited package valid until 2015-12-30".

Figure 7: The Main window after MLVA typing.