Data quantification with MAPIX Analyzer

PEPperCHIP[®] Peptide Microarrays are provided in combination with an electronic data package. The package includes the microarray grid file (*.gal*) that assigns the peptide sequences to the spot patterns, the Excel spreadsheet with the peptide map of your PEPperCHIP[®] Peptide Microarray and the MAPIX software license USB dongle (if purchased). You can download the MAPIX Analyzer software for <u>32-bit</u> <u>Windows and 64-bit Windows</u>. Please note that the software is only running if the USB dongle is connected to your computer. In the following section, you can find a quick guideline on how to quantify fluorescence intensities of microarray scans of your PEPperCHIP[®] Peptide Microarray using MAPIX Analyzer. For detailed information, please refer to the MAPIX software user manual.



1. Import raw scan images

- Open the MAPIX Analyzer software.
- Import the raw 16-bit greyscale .tiff image(s) of the microarray scanner for one or both fluorescence channels by selecting *File* → *Open Images*
- Assign colors to the respective wavelength depending on the applied fluorescence labels.



2. Image adjustment

- Assure that your image is in the correct orientation. If necessary, use the Zoom In and Zoom Out options, rotate or flip the image vertically or horizontally to obtain an appropriate image view.
- If necessary, increase brightness and contrast manually in the display options; any adjustment won't affect the real image data.



3. Import microarray grid file (.gal)

- Open the microarray grid file by selecting *Analyse → Open grid* and select the appropriate .gal file for your PEPperCHIP[®] Peptide Microarray.
- All peptide spots are grouped in blocks and visualized as grid according to the peptide microarray layout design and content.



4. Alignment of the microarray grid

- Select *block modification* in the tool bar or *Image → block mode*; now you can select either only the block of interest by clicking on it or all blocks with the Ctrl-A shortcut of your keyboard. Drag and drop the grid until the peptide spots of the microarray scan and the grid show an optimal overlap.
- If needed for an optimal overlay, please use the *Array Rotation* function in order to adjust the angle of the grid (Ctrl +/-).
- If further needed, you can also adjust the spot size by selecting *Image* → spot mode → Ctrl-A.



5. Image processing, data quantification and export

- Select the blocks of interest and initiate the data quantification with *Analyse* → *Photometric calculations.*
- Save the output data as .txt file on your computer.

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6. Data Analysis

- Open Microsoft EXCEL and Import the .txt file (Data → Import from text → Choose settings: "Delimited", "Tab", "General", "Existing Worksheet")
- Absolute spot intensities for further analysis are reflected as foreground median for each wavelength.
- For more information, please refer to the MAPIX user manual in the installation directory.