

MC_Rack Tutorial: Long Term Potentiation (LTP)



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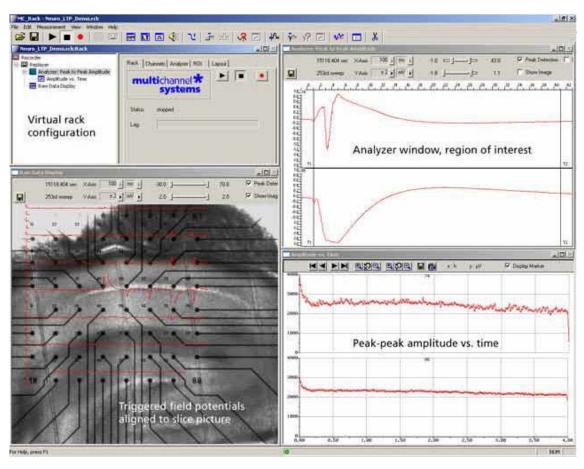
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1 Long Term Potentiation (LTP)

1.1 Aim of Experiment



This experiment was performed on an **acute hippocampal slice** on a microelectrode array (MEA) with 64 channels with the aim to analyze **long-term potentiation** (**LTP**) following electrical stimulation.

The 300 μ m thick acute hippocampus slice was prepared from a 21 days old rat. The slice was stimulated every 60 s with the **test stimulus**, a single biphasic pulse with 1.5 V amplitude and 100 μ s duration per phase, by using one of the embedded MEA electrodes (No. 75).

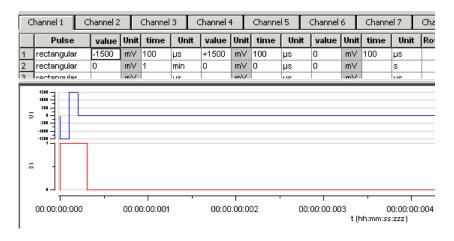


Fig. 1 Test stimulus for LTP experiment

The screen shot from the MC_Stimulus program shows the biphasic test pulse (amplitude: 1.5 V, duration: 100 μ s per phase, inter-stimulus-interval: 60 s). The red trace is the Sync Out (TTL) pulse that was used for triggering the recording.

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LTP was induced by an **LTP train** with **100 pulses** of same amplitude and duration as the test stimulus, and a frequency of 100 Hz. The Sync Out signal of the STG (stimulus generator) has been used to trigger the recording, with a pre-trigger of 30 ms and a time window of 100 ms. Data was recorded before and after the LTP induction; the LTP induction itself was not recorded.

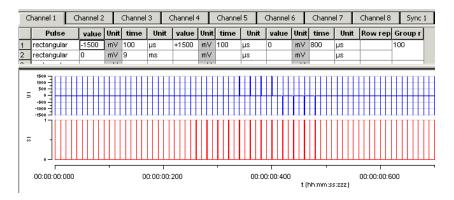


Fig. 2 Pulse train of 100 biphasic pulses for LTP induction

Amplitude: 1.5 V, duration: 100 µs per phase, inter-stimulus-interval/frequency: 10 ms / 100 Hz)

As the data was acquired with a MEA1060-BC amplifier with **blanking circuit**, all MEA electrodes were disconnected from the amplifier shortly during stimulation, in this case for **540 \mus** (300 μ s Sync Out pulse of the STG, plus intrinsic **Wait** of 40 μ s, plus user-defined **Wait** of **200 \mus**). This efficiently prevented stimulus artifacts.

The demo data used in this part of the tutorial was kindly provided by Dr. Frank Hofmann, University of Heidelberg, Germany.

Task

Show the impact of an LTP train on the LTP peak-to-peak amplitude by an offline analysis; graph, print and save the results.

Note: In this chapter, you will set up an offline analysis rack (for the demo data file) step by step. You can set up a virtual rack for online recordings and analysis likewise, simply add the **MC_Card** as the data source to the rack instead of the **Replayer**.

- Rack file: Neuro_LTP_Demo.rck
- Data file: LTP-Demo.mcd (recorded after LTP induction), preLTP-Demo.mcd (recorded before LTP induction)
- Image file: LTP_slice.jpg

Please see also the MEA Application Notes for more information on the preparation techniques and experiments.

You will learn in this chapter ...

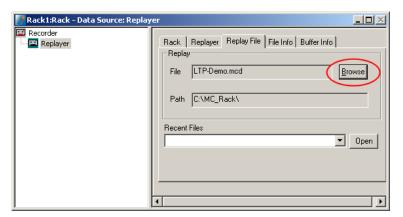
- How to define basic functions in your rack configuration:
 - Record data (Recorder)
 - Analyze field potentials, for example, extract peak to peak amplitudes (Analyzer)
 - Plot the amplitude vs. time (Parameter Display)
- How to **customize** the rack:
 - Modify the display layout (all display types)
 - Rename virtual instruments for a better overview (all virtual instruments)

- More details:
 - Align the data traces to a slice picture (Analyzer, Data, or Averager display)
 - Customize the colors of the traces and axes in the display (all display types)

1.2 Preparations

We recommend that you take some time for rebuilding the virtual rack for this application step by step in this tutorial, but if you prefer having a look at the completed rack or if you get stuck during the tutorial, you can also open the rack file "Neuro_LTP_Demo.rck". Click **Open** on the **File** menu to open the rack file.

- 1. Copy the **complete** MC_Rack **Tutorial** folder from the installation volume into the MC_Rack program directory with the following path "c:\Program Files\Multi Channel Systems\MC_Rack\"
- 2. Start MC Rack or click **New** on the **File** menu to generate a new virtual rack file configuration.
- 3. Click on the toolbar to add a **Replayer** to your virtual rack.
- 4. In the tree view pane of the virtual rack, select the **Replayer** and click the **Replay File** tab. Click the **Browse** button and browse to the **Offline** subfolder of the **Tutorial** folder, and load the data file **LTP-Demo.mcd** into the **Replayer**.



1.3 Analyzing Field Potentials

You can use a virtual **Analyzer** for **extracting parameters** of interest, for example, the **minimum** or **maximum**, the **amplitude**, or the **slope** from a **region of interest (ROI)** of the field potentials.

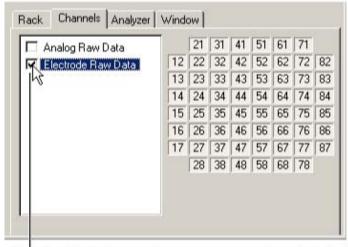
As the data was recorded on a **trigger** (the Sync Out pulse of the stimulator) and the region of interest needs to be specified relative to a trigger event, the **Analyzer** will also operate based on this trigger. In an online experiment, you would now have to define the trigger with a **Trigger Detector**. In the data file, the trigger was already recorded together with the raw data, and you can use the existing trigger for triggering the **Analyzer**. (In this experiment, more than one trigger were used, but trigger 2 was used for recording the data, trigger 1 was not saved. Therefore, there is only trigger 2 available in the data file.) You can see this information on the **File Info** page of the **Replayer**.

- 1. On the toolbar, click to add an **Analyzer** to the virtual rack configuration.
- 2. Select now the channels you are interested in. Start with all channels, you can then later decide to deselect channels with no LFP responses (for optimizing the computer performance). For example, you can deselect the stimulating electrode No. 75, or electrodes that are not covered by the slice.
- 3. In the tree view pane of the virtual rack, select the **Analyzer** and click the **Channels** tab of the **Analyzer**.

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- 4. Click the data stream **Electrode Raw Data**. On the right half of the **Channels** page, the channels appear in the standard 8 x 8 MEA grid.
- 5. Select the check box next to the stream name to select **all** channels or click the buttons with the electrode numbers to pick **single** channels. The check box next to the stream name appears shaded if some, but not all channels have been selected.

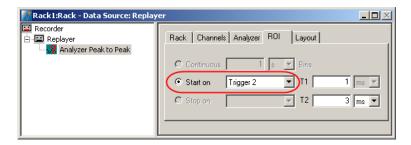
Selecting all channels of a data stream



The channels of the selected data stream appear as toggle buttons in the designated channel layout. You can select single channels by clicking the corresponding buttons. Selected channels appear pressed in.

Click the check box next to a stream name to select all channels.

6. Click the **ROI** tabbed page, which stands for **region of interest**. **Start on Trigger 2** is preselected, because the recording of this data file was triggered on trigger 2. The option **Continuous** is not available, because the sample data is not a continuously recorded file.



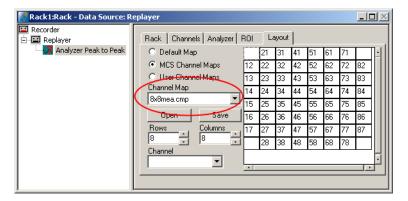
The **Analyzer** displays the electrode channels in the layout of the **Channel Map** you have created or loaded.

1.4 Customizing the Display Layout

The following information applies not only to the **Analyzer** display, but also to other displays, such as the **Raw Data** display or the **Parameter** display.

Hint: Channel maps are provided for all MEA types available from Multi Channel Systems. You can download channel maps for all MEAs available from **Ayanda Biosystems** from the Ayanda web site (http://www.ayanda-biosys.com/download.html). You can also set up custom channel maps with a preselected number of electrodes and save them for later use.

- 1. In the tree view pane of the virtual rack, select the **Analyzer** and click **Layout**.
- 2. To load a **Channel Map** in the MEA layout, click **Open**.
- 3. Browse your folders and open the MC_Rack program folder.
- 4. Select the appropriate MEA layout, for example, the **8x8mea.cmp** file for standard 8 x 8 MEAs, and click **Open**. The MEA layout appears on the right side of the dialog box, and the corresponding display is updated accordingly.



5. Switch to the **Analyzer** display. You can choose the appropriate range of the y-axis from the drop-down list. The x-range and the display refresh rate are defined by the sweep time. You can see on the **File Info** page of the **Replayer** that the data is triggered and that one sweep is 100 ms long. Therefore, the maximum range of the x-axis is 100 ms. You can fine-tune the ranges with the sliders.

1.5 Renaming Virtual Instruments

You can rename instruments for a better overview. This is especially useful if you use the same type of virtual instrument several times, for different purposes, in the same rack.

1. In the tree view pane of the virtual rack, select the **Analyzer 1**, and then click its name (do not double-click it). An insertion point appears.



2. Type in the new name, for example **Peak-Peak Amplitude**.

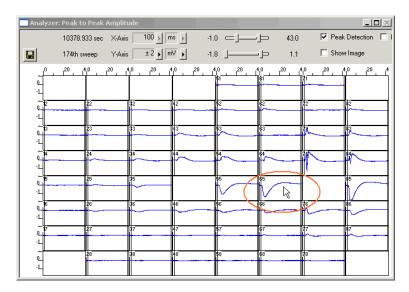
1.6 Zooming into a Single Channel

In all displays, you can switch to a single channel by double-clicking the channel in the display.

1. Click **Start** (either on the **Measurement** menu, the toolbar, or the **Rack** tabbed page) to start the replaying of the data.

Each virtual instrument in the rack configuration starts to process the channels and data streams that were assigned to it. For example, data is graphed in the displays, and so on. You can adjust the replaying **speed** on the **Replayer** tabbed page of the **Replayer**.

2. Double-click channel No. 65 in the **Analyzer** display. As the pathway that is going to be analyzed in this experiment is directed from the right to the left in the slice, the electrodes of interest are left of the stimulating electrode.

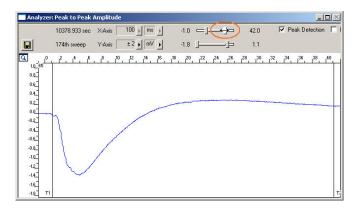


The magnifying-glass icon on the top left indicates that you have zoomed to a single channel.

3. **Zoom in** the y-axis by moving the two sliders **to each other**.



- 4. **Move the range** by clicking in the **middle** between the two sliders and dragging with your mouse or pressing the LEFT ARROW and the RIGHT ARROW keys. For larger steps, you can also use the PAGE UP / PAGE DOWN keys.
- 5. **Zoom the x-axis** as well until you have centered on the response following the stimulus (at time point 0).



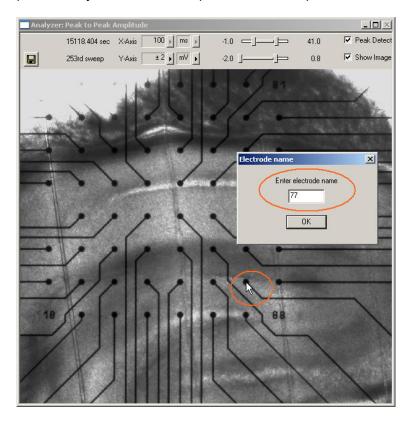
6. Double-click the display again to switch back to the full channel view. The settings have been applied to all channels.

1.7 Aligning the Data Traces to a Slice Picture

You can load a digital photograph of the acute slice as a background picture and align the signals to the electrodes and the regions of the slice.

Hint: Crop the picture in a graphics program so that only the electrode array is visible. This helps saving (unused) space on the screen.

- 1. On the **File** menu, click **Load Image**.
- 2. Browse your folders and open the **Tutorial** folder on the installation volume.
- 3. Select the LTP_slice.jpg file and click Open.
- 4. On the **Analyzer** or **Data Display**, click **Show Image**. You can now see the slice picture.
- 5. Now, you map the channels to the electrodes in the picture. Hold down the **SHIFT key**, point to any electrode in the picture, for example No. 77, and **double-click**.

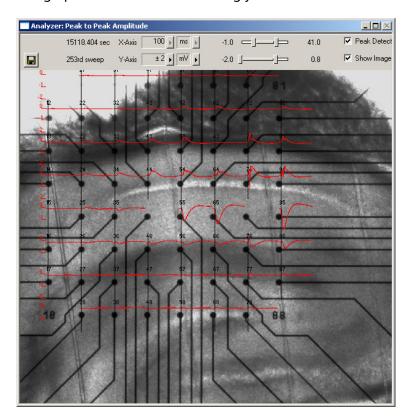


A text box opens.

- 6. Type in the number of the electrode according to the MEA layout, for example 77.
- 7. Hold down the **CTRL key**, point to any electrode in **another** row and column, for example No. 44, and **double-click**.
- 8. Type in the number of the electrode according to the MEA layout, for example 44. The data traces appear on top of the picture.

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9. You can resize the window by dragging the window frame with the mouse. The graphic will be scaled accordingly.



Note: If you have made a mistake, simply repeat the assignment from step 58.

Hint: If you **record** data with a background picture in your rack, the file path of the picture is saved together with the data, that is, it is opened automatically when you replay the file in a rack that has a **Display** with the **Show Image** option selected. (File paths are absolute, that means MC_Rack cannot find picture files if they have been moved to another folder or directory. You will be informed by an error message.)

1.8 Customizing the Display Colors

It may be difficult to see the traces or the axes because the contrast of the trace color to the background color is too low. You can define the colors of the traces and the axes to improve the view.

Point to any trace or to an axis and right-click.
 The **Color** dialog box opens.



2. Choose a color from the palette or set up a custom color. The color of all traces or of the axes is changed accordingly.

1.9 Extracting Parameters

In triggered mode, the **Analyzer** extracts the parameters of interest from a distinct **time window** relative to the **trigger event**, the region of interest (**ROI**). It is very important to define this time frame appropriately for **all** channels of interest, because otherwise you will not obtain any proper results from the **Analyzer**.

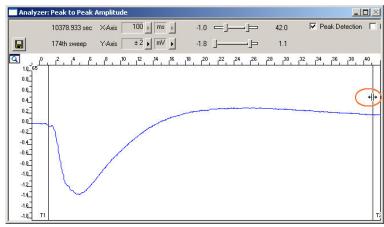
1. In the tree view pane of the virtual rack, select the **Analyzer**, and click the **Analyzer** tab. Select the parameters you like to extract, **Peak-Peak Ampl.** in this case.



2. Now, you have to define the ROI. Look at the Raw Data traces again in the Analyzer display and select a range that includes the biological responses on all channels. Shortly after the stimulus / trigger event (time = 0) will be fine. You can define the region of interest with by dragging the two vertical bars in the display, or by editing the T1 and T2 boxes on the ROI tabbed page of the Analyzer. For example, the start time (= T1) can be set to 1 ms (after the trigger event) and the stop time (= T2) 41 ms.

The selected parameters of interest will then be extracted from the region of interest, and can then be recorded to a data file, and / or plotted using a **Parameter** display.

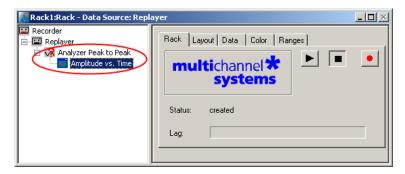




1.10 Plotting Amplitude versus Time

Add a display to graph the peak-to-peak parameter. As a parameter data stream is very different from raw data, MC_Rack features a specialized display type, the **Parameter Display**, which is optimized for parameter streams.

→ In the tree view pane of the virtual rack, select the **Analyzer** and click on the toolbar to add a **Parameter Display** in **series** with the **Analyzer** to the virtual rack. (It would not be possible to add the **Parameter Display** in parallel to the **Analyzer**, as the replayed data file does not contain any parameter streams as input streams for the display, and virtual instruments can only use the output streams of other virtual instruments that are upstream in the virtual rack tree.) Rename the display to **Amplitude vs. Time**.

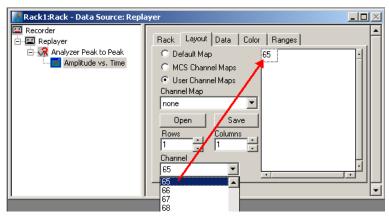


Customizing the display layout

You can define the layout for each display separately. If you have selected all channels as an input for the analyzer, you may want to see the plot for all channels as well, for example. On the other hand, you may be interested in **monitoring** only one or two representative channels online (for saving space on the screen or for saving computer performance). (You can still **save** the parameters of all channels in the **data file** to review it later or to analyze it further in your custom evaluation software.) Set up a custom layout with only **one** electrode of interest now.

Hint: Try typing the channel numbers instead of selecting them from the drop-down list with the mouse.

- 1. Click the **Layout** tab.
 The default layout is loaded. In the **Rows** and **Columns** boxes, you can now modify the layout.
- 2. Type or select the number of **Rows**, for example 1, and the number of **Columns**, for example 1. On the right, a custom layout with a single channel appears.
- 3. Assign the appropriate channel numbers to the empty slots. Click the empty slot to select it. The active slot is highlighted by a dotted line.
- Select 65 for the active slot from the **Channel** list, or type **65**.
 The **Parameter Display** is updated accordingly. Likewise, you can set up any custom configuration for all display types.

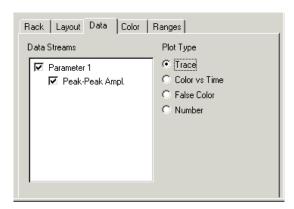


Selecting data streams

Now, select the data that you like to be displayed.

1. Click the **Data** tab.

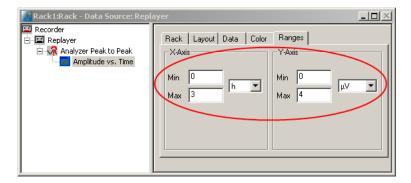
The **Peak-to-Peak Amplitude** is already preselected. The plot type is set to **Trace**, meaning that a curve is plotted. These settings are ok and do not have to be modified in the moment.



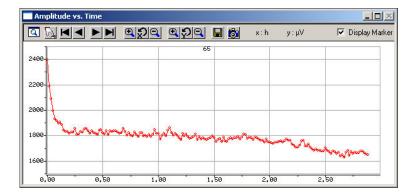
Setting the window extent

As you can see on the **File Info** page of the **Replayer**, the total recording time of the example file is about 3 hours. Adjust the ranges of the y-axis as well.

- 1. Click the Ranges tab.
- 2. Enter 3 h for the x-axis range, and 4 mV for the y-axis.



1. Click **Start** to graph the plots. The baseline and the LTP induction were not recorded. The high amplitudes at the beginning demonstrate the effect of short term potentiation. LTP starts after about 20 min following the LTP induction. The amplitude slowly decreases over time.



1.11 Zooming and Scrolling

Zooming to a single channel

To provide an overall view of the ongoing activity, normally **all channels** (depending on the layout) are shown in the parameter display.

→ Simply double-click a channel to have a closer look at it. The magnifying glass icon in the upper left corner indicates the zoom mode.

Double-click the zoomed channel again to switch back to the **overall view**.

Scrolling back and forward

You can scroll the data forward and backward along the time axis. You are able to look for a region of the plot you are interested in and snapshot the displayed region.

- \rightarrow On the tool bar, click \square to scroll **backward** and \triangleright to scroll **forward**.
- → Click if you like to jump to the **beginning** of the plot.
- → Click if you like to jump to the **end** of the plot.

Scaling the x- and y-axis

You can zoom the x- and y-axis in and out separately. The left button array on the toolbar corresponds to the x-axis and the right button array to the y-axis.

- \rightarrow Decide which axis you like to scale and click the corresponding $\stackrel{\textcircled{\P}}{=}$ button to zoom the axis in and $\stackrel{\textcircled{\P}}{=}$ to zoom it out. The display will be updated accordingly.
- → To reset the scaling of the x-axis to default, click 🔽.
- \rightarrow Likewise, click \mathbf{r} to reset the scaling of the y-axis.
- → If you have zoomed to a single channel, you can click the Apply to All button to apply the settings to all channels. If you do not use this button and switch back to the overall view, any changes to the settings are lost.

1.12 Saving a Picture

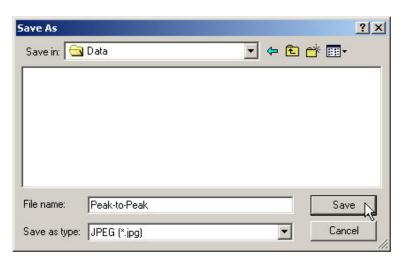
You can take a screen shot of the **Parameter Display**. This feature is especially useful for quickly setting up illustrations and presentations. Each visible channel on your display is saved as a separate image file. Each file has the chosen file name extended by channel number. If, for example, the entered file name is *Peak-to-Peak*, the file name of channel 41 will be *Peak-to-Peak_41.jpg*.

Note: Only the channels that are currently shown on the display are captured. Therefore, if you want to capture all channels, click the Snapshot button when all channels are visible. If you want to capture only a single channel, zoom this channel and then click the Snapshot button.

Hint: If you need more features for data screen shots, Multi Channel Systems recommends to use a commercially available screen shot program.

1. On the **Parameter Display** toolbar, click the **Snapshot** button to take screen shots of the **displayed** channel(s). The **Save As** dialog box opens.

- Select an image file format from the drop-down list: (*.emf), Enhanced Meta File format (vector graphics) (*.bmp), bitmap (*.jpg), compressed bitmap in JPEG format
- 3. Enter a file name.



The **file name** will automatically be **extended** by the **channel number**.

4. Confirm by clicking **Save**.

1.13 ASCII Export

You can save the displayed data as an **ASCII file** (text without formatting). You may then send this data to an editor or to your custom analyzing software, for example. This feature is also very convenient if you want to generate custom graphics, for example, with Microsoft Excel or Micrococcal Origin.

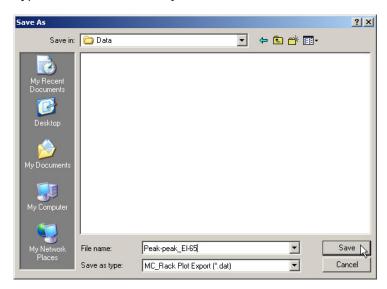
Saving data is only possible when you have stopped the recording. The **Save** button will reappear on the toolbar.

Note: Only the channels that are currently shown on the display are saved. In the example given below, only the Peak-to-Peak amplitude of the channel 65 will be saved. Therefore, if you want to save all channels **in a single file**, click the **Save** button when all channels are visible. If you want to save the parameter values of a single channel only, zoom this channel and then click the **Save** button.

Hint: If you need more features, for example, export of raw data, use the add-on program MC_DataTool that comes together with ME- / MEA-Systems.

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- 1. Click the **Save** button on the **Parameter Display** toolbar to open the **Save As** dialog. The **Save As** dialog box will be opened.
- 2. Type in a **file name** and **path** and click **Save**.



You can now open and process the file with any text editor, spreadsheet, or any other custom evaluation software. The following screen shot shows how it looks when the filet is opened with a standard spreadsheet. The first row contains the axes headers. The first column contains the x-values (in hours), the second column the y-values of channel 65 (in micro volts).

	Α	В
1	T[h]	65-Height[µ∨]
2	0.000	2.396.364
3	0.017	2.189.091
4	0.033	2.090.909
5	0.050	1.994.545
6	0.067	1.930.909
7	0.083	1.918.182
8	0.100	1.898.182
9	0.117	1.905.455
10	0.133	1.887.273
11	0.450	1 0 40 000

1.14 Recording Analyzed Data

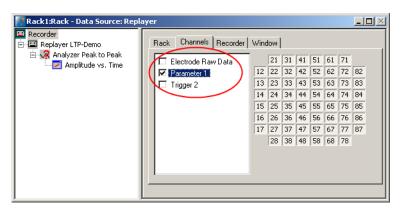
Now, the virtual rack for offline analysis of LTP experiments has been completed, and you can select the data that should be recorded, that is, saved in the generated data file. It is important to know that MC_Rack **never** modifies or overwrites an **existing** data file. That means, in an offline analysis, the extracted parameter streams are stored in a **new** data file. Therefore, you need to specify a file name and path in the **Recorder** for both online and offline analysis.

Hint: If you want to store raw data and extracted parameters in the same data file, you can select the already existing raw data traces in a replayed file for rerecording them together with the extracted parameter streams.

1. Select the **Recorder** and click the **Channels** tab.

You see all data streams on the left. You can now select any channels you like, independently from the channels you view on screen with MC_Rack. To select the **Electrode Raw Data** stream does not make much sense in this case, because the data file already exists and is only replayed in this tutorial. But if you performed an online analysis, you would probably want to save the raw data (cutout sweeps triggered on the stimulus) of the relevant channels.

- 2. In this case, we are interested in the **Peak to Peak** parameters. Select the **Parameter 1** data stream. All available channels appear in a button array on the right side. (Remember: Available are those channels that have been assigned to the **Analyzer** and thus have been processed by the **Analyzer**. If you change the **Analyzer** input, you will see that the recordable output will be changed accordingly.)
- 3. Select all channels by selecting the check box, or pick single channels. All channels selected on this page will be saved in the specified file when you start the recording. (If you have not already specified the file and path on the **Recorder** tabbed page, do so now.)



Click the **Record** button and then **Start** again.



4. The data will now be replayed and the selected data streams will now be saved to the specified location. You can then replay the newly generated file with the **Replayer** and a **Parameter Display**. You can also export the data to your custom graphics or analysis software.

Note: Do not forget to enable the recording by clicking the **Record** button !! Otherwise, no data will be stored.

2 Saving the Rack and Saving Data

2.1 Saving a Rack

Save the virtual rack configuration if you like to keep it for future use, for example for an offline analysis of identical data recorded in another experiment.

- 1. On the **File** menu, click **Save As**.
- 2. The **Save As** dialog box opens.
- 3. Browse your folders and select a path.
- 4. Enter a file name and confirm by clicking **Save**.

The file extension for the rack files is .rck.

2.2 Selecting Data Streams for Recording

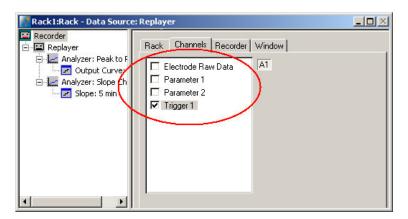
MC_Rack's philosophy is to **strictly separate** the actions of **all** virtual instruments in a rack. That means, that you could **record** to hard disk completely different data streams and channels than you **monitor** on the screen. This has the advantage that you can store exactly the channels you are interested in, but it also has the slight disadvantage that all virtual instruments have to be set up **separately**. Please be especially careful when configuring the **Recorder**, to avoid data loss.

When you have finished setting up the rack, you can select the data streams and channels that you want to save to the data file specified in the **Recorder**.

Selecting data streams and channels for recording

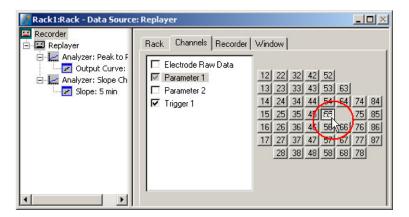
The fate of each single channel is **independent** from other channels. You can pick exactly the channels you like to save from all generated data streams. For example, you can decide to save only **one** channel of **raw data**, but the **peak-to-peak amplitude** results of **all**, or of a **specific selection** of channels.

1. Select the **Recorder** in the virtual rack tree view pane and then click the **Channels** tabbed page. On the white pane on the left of the **Channels** page, you see the data streams that are available with your rack configuration. When replaying data, you are generally more interested in the parameter data streams, but you can rerecord the raw data as well, in case that you want to save the raw data and the extracted parameters to the same data file.



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2. Click the data stream that you are interested in, generally the parameter streams. The available electrode channels appear in a button array on the right side. Parameter stream 1 is generated by the first analyzer in the rack (here: for extracting the peak-peak amplitude), parameter 2 is generated by the second analyzer (here: for extracting the slope).



3. You can now either select all channels by clicking the check box next to the data stream name, or you can pick single channels by clicking the corresponding buttons. For more information, please see "Channel Selection" in the MC_Rack Features section. Only data from the selected channels will be saved to the hard disk.

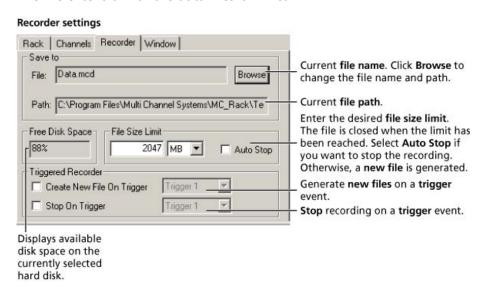
2.3 Creating a Data File

If you want to write the parameters that you will extract in an offline analysis to the hard disk, you have to specify the file name and path in the **Recorder**. (It is not possible to change or overwrite existing data files in MC_Rack, for example, adding the spike rate to an existing data file, but you can record the raw data together with the spike rate stream to the same file.)

Choosing the file name and path

- 1. Click the **Recorder** tab.
- 2. **Browse** your folders and select a path.
- 3. Type a file name into the text box.
- 4. Confirm by clicking **Save**.

The data file is then generated automatically when you start MC_Rack in **recording** mode. The file extension for the data files is *.mcd.



File size limit

When the maximum file size specified by the user has been reached, a new file is generated automatically. The file name is extended by four digits, counting up, for example LTP-Parameters0001.mcd, LTP-Parameters0002.mcd, and so on.

If you rather prefer that the recording is completely stopped when a file has reached the maximum size, please select the option **Auto Stop**.

For information on more options, please see "Generating Data Files" in the MC_Rack Features section.

Selecting data streams and channels

As has been said before, the fate of each single channel is **independent** from other channels. You can pick exactly the channels you like to save from all generated data streams. For example, you can decide to save only **one** channel of **raw data**, but the **peak-to-peak amplitude** results of **all**, or of a **specific selection** of channels.

→ Click the Channels tab.

As long as the rack is still empty, you see an empty box. There are no channels available at this point, because you have not chosen a data source yet (**MC_Card** for online data acquisition or **Replayer** for replaying data files). Without a data source, there are no data streams available for recording. Remember later, when you have completed the rack, to assign the channels that you like to save to the data file to the **Recorder**.

After you added a **data source**, you will see the electrode raw data streams provided by the data source (for example, electrode raw data, analog data, and digital data from the **MC_Card**, or the data streams included in the data file loaded into the **Replayer**). If the rack file contains virtual instruments that **generate** data streams such as Spikes from a **Spike Sorter** or Parameter streams from an **Analyzer**, these data streams will be available for recording as well.

2.4 Starting Data Acquisition and Recording

Now that you have completed the virtual rack, you are ready to start the rack.

- → Click **Start** (either on the **Measurement** menu, the toolbar, or the **Rack** tabbed page) to start the data acquisition. Each virtual instrument in your rack starts to process the channels and data streams that were assigned to it.
- → Click first **Record** and then **Start** to write data to the hard disk. The data from the electrodes selected in the **Recorder** is saved to the file and location specified in the **Recorder**.



→ Click **Stop** to stop the data acquisition.



Warning: **Only** data of the channels and data streams that were **selected** in the **Recorder** are saved in your data file when you start a recording. Data is **only** saved to the hard disk when the red **Record** button is pressed **in**. Make always sure that you have selected all channels of interest, and that the **Record** button is active before starting an experiment to avoid data loss.