

# **MEA Amplifier with Blanking Circuit for Inverse Microscopes**

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Printed: 07. 04. 2016

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# 1 Introduction

## 1.1 About this Manual

This manual comprises all important information help the first installation of the hardware and software, and help the daily work with the instrument. It is assumed that you have already a basic understanding of technical and software terms. No special skills are required to read this manual.

If you are using the device for the first time, please read the **important safety advice** before **installing the hardware** and **software**, where you will find important information help the installation and first steps.

The **printed manual** and **help** are basically the same, so it is up to you which one you will use. The help offers you the advantage of scrolling through the text in a non-linear fashion, picking up all information you need, especially if you use the **Index**, and the **Search** function. If you are going to read larger text passages, however, you may prefer the printed manual.

The device and the software are part of an ongoing developmental process. Please understand that the provided documentation is not always up to date. The **latest information** can be found in the **help**. Check also the MCS web site ([www.multichannelsystems.com](http://www.multichannelsystems.com)) for downloading up-to-date manuals and help files.

## 1.2 Terms of Use for the Program

You are free to use the program for its intended purpose. You agree that you will not decompile, reverse engineer, or otherwise attempt to discover the source code of the software.

## 1.3 Limitation of Liability

Multi Channel Systems MCS GmbH makes no guarantee as to the accuracy of any and all tests and data generated by the use the software. It is up to the user to use good laboratory practice to establish the validity of his findings.

To the maximum extent permitted by applicable law, in no event shall Multi Channel Systems MCS GmbH or its suppliers be liable for any special, incidental, indirect, or consequential damages whatsoever (including, without limitation, injuries, damages for data loss, loss of business profits, business interruption, loss of business information, or any other pecuniary loss) arising out of the use of or inability to use the program or the provision of or failure to provide Support Services, even if Multi Channel Systems MCS GmbH has been advised of the possibility of such damages.



## 2 Important Information and Instructions

### 2.1 Operator's Obligations

The operator is obliged to allow only persons to work on the device, who

- are familiar with the safety at work and accident prevention regulations and have been instructed how to use the device;
- are professionally qualified or have specialist knowledge and training and have received instruction in the use of the device;
- have read and understood the chapter on safety and the warning instructions in this manual and confirmed this with their signature.

It must be monitored at regular intervals that the operating personnel are working safely.

Personnel still undergoing training may only work on the device under the supervision of an experienced person.

### 2.2 Guarantee and Liability

The *General conditions of sale and delivery* of Multi Channel Systems MCS GmbH always apply. The operator will receive these no later than on conclusion of the contract.

Multi Channel Systems MCS GmbH makes no guarantee as to the accuracy of any and all tests and data generated by the use of the device or the software. It is up to the user to use good laboratory practice to establish the validity of his findings.

Guarantee and liability claims in the event of injury or material damage are excluded when they are the result of one of the following.

- Improper use of the device.
- Improper installation, commissioning, operation or maintenance of the device.
- Operating the device when the safety and protective devices are defective and/or inoperable.
- Non-observance of the instructions in the manual with regard to transport, storage, installation, commissioning, operation or maintenance of the device.
- Unauthorized structural alterations to the device.
- Unauthorized modifications to the system settings.
- Inadequate monitoring of device components subject to wear.
- Improperly executed and unauthorized repairs.
- Unauthorized opening of the device or its components.
- Catastrophic events due to the effect of foreign bodies or acts of God.

### 2.3 Important Safety Advice



Warning: Make sure to read the following advice prior to install or to use the device and the software. If you do not fulfill all requirements stated below, this may lead to malfunctions or breakage of connected hardware, or even fatal injuries.



Warning: Obey always the rules of local regulations and laws. Only qualified personnel should be allowed to perform laboratory work. Work according to good laboratory practice to obtain best results and to minimize risks.

The product has been built to the state of the art and in accordance with recognized safety engineering rules. The device may only

- be used for its **intended purpose**;
- be used when in a **perfect condition**.
- Improper use could lead to serious, even fatal injuries to the user or third parties and damage to the device itself or other material damage.



Warning: The device and the software are **not** intended for medical uses and **must not** be used on humans.

Malfunctions which could impair safety should be rectified immediately.

#### High Voltage

Electrical cords must be properly laid and installed. The length and quality of the cords must be in accordance with local provisions.

Only qualified technicians may work on the electrical system. It is essential that the accident prevention regulations and those of the employers' liability associations are observed.

- Each time before starting up, make sure that the **mains supply** agrees with the specifications of the product.
- Check the **power cord** for damage each time the site is changed. Damaged power cords should be replaced immediately and may never be reused.
- Check the **leads** for damage. Damaged leads should be replaced immediately and may never be reused.
- Do not try to insert anything sharp or metallic into the vents or the case.
- Liquids may cause short circuits or other damage. Keep the device and the power cords always **dry**. Do **not** handle it with wet hands.

#### Requirements for the installation

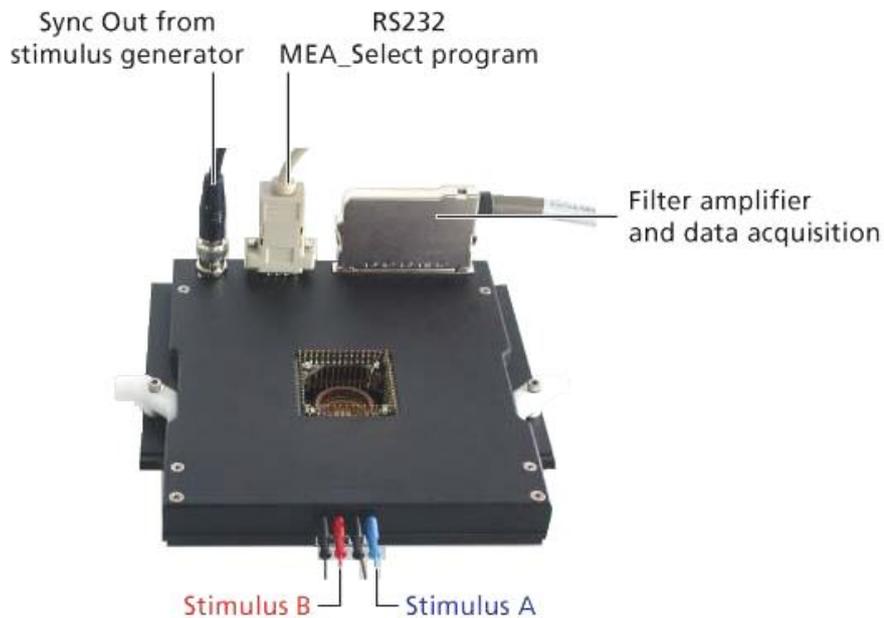
- Make sure that the device is not exposed to direct sunlight. Do not place anything on top of the device, and do not place it on top of another heat producing device. Never cover the device, not even partially, so that the air can circulate freely. Otherwise, the device may overheat.
- Use and keep the device only in a dry environment. Fluids or damp air may damage or destroy the device. Spilled liquid can damage or even completely destroy the electronics of the MEA amplifier. Avoid it by all means.



Warning: The device must not get in contact with fluids! Spilled liquid can damage or even completely destroy the electronics of the amplifier! This is eminently important when using a perfusion system. Take care that the flow rates of the inlet and outlet flow match so that flooding of the amplifier is efficiently prevented.

## 3 First Use of the MEA Amplifier

### 3.1 Welcome to the MEA Amplifier



Raw data from up to 60 electrodes of a **microelectrode array (MEA)** is amplified by 60 channels of **pre-** and **filter amplifiers**.

The **MEA sensor** is placed directly into the small-sized MEA **preamplifier** with **blanking circuit (MEA1060-BC-PA)**. When the amplifier is closed, the **contact pins** in the lid of the amplifier are pressed onto the MEA **contact pads**. The very close location of the amplifier to the MEA sensor is very favorable concerning a high signal-to-noise ratio.

MEA amplifiers can be ordered with different **gain** and **bandwidth** configurations by the user's choice. For example, typical pass bands would be 1 to 300 Hz for recording field potentials and 300 to 3000 Hz for recording action potentials. It is also possible to use a broadband amplifier and filter the data with the digital filter of the free MC\_Rack program. Gain settings from 100 to 5000 are possible.

The amplifier is connected to the **data acquisition** computer via a single 68-pin MCS standard cable. The analog output signals of the MEA amplifier are then acquired and digitized by the MC\_Card (MEA-System) or an USB based data acquisition device (USB-MEA-System) or your custom data acquisition system.

MEA amplifiers have an **integrated heating system** for controlling the **MEA's temperature**. The desired temperature can be easily programmed with a temperature controller. You will not need an incubator during recording, all environmental conditions are reliably controlled directly in the MEA dish.

## 3.2 Installing the Software

### System requirements

Software: One of the following Microsoft Windows ® operating systems is required: **Windows 10, 8.1, Windows 7** (32 or 64 Bit), **Vista** or **Windows XP** (**English** and **German** versions supported). Other language versions may lead to software errors.

Hardware: Free RS232 port

### Installing the software MEA\_Select, Version 1.3.0

Please check the system requirements before you install the software. MCS cannot guarantee that the software works properly if these requirements are not fulfilled.

**Important:** Please make sure that you are logged in as an **administrator** before installation. Otherwise, is possible that the installed software does not work properly.

1. Double-click **Setup.exe** on the installation volume. The installation assistant will show up and guide you through the installation procedure.
2. Follow the instructions of the installation assistant.

## 3.3 MEA\_Select Program

### MEA Amplifier with blanking circuit (MEA1060-BC)

<p>Electrode button array: Select MEA layout from drop down menu</p>	<p>Electrode mode for stimulation, recording or ground</p>	<p>Blanking: ON / OFF</p>	<p>Load and save electrode configuration</p>	<p>Online "Help"</p>
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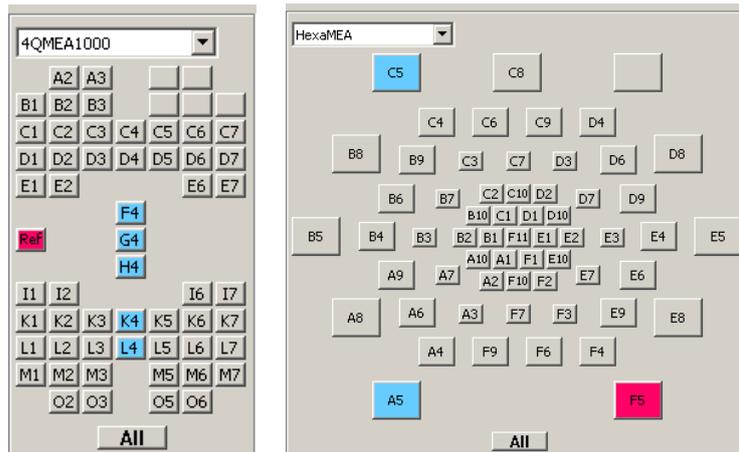
The screenshot shows the MEA\_Select software interface. Key elements include:
 

- Electrode button array:** A grid of buttons numbered 21 to 88, with an 'All' button below it.
- ME layout selection:** A dropdown menu currently showing 'MEA 8x8 200/30'.
- Stimulation options:** Radio buttons for 'Not Stim.', 'Ground', 'Stim. A', and 'Stim. B'.
- Blanking control:** A checkbox for 'Blanking' which is checked.
- Configuration:** 'Load' and 'Save' buttons.
- Wait time:** A numeric input field set to '60' with the unit '[µs]'.
- Port selection:** A dropdown menu set to 'COM1'.
- Buttons:** 'Download', 'Change MEA', 'Test Conn.', 'Apply', and 'List Mode'.

<p>Click any or "All" button to select electrodes for recording, stimulation or grounding</p>	<p>Connection to MEA amplifier and "Download"</p>	<p>"Change MEA": Sets all electrodes to ground</p>	<p>Delay between end of stimulation and start of recording</p>	<p>Toggle "List Mode"</p>
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When starting the software program **MEA\_Select** for controlling the blanking circuit of the MEA amplifier the main window appears. You see the **electrode button array** on the left side. Select the MEA layout from the drop down menu and the electrode button array will be adapted. Click any or "All" button to select any or all electrodes for recording, stimulation or grounding.

Electrode button array adapted for 4QMEA1000 layout and HexaMEA layout.



The **Stimulation** window provides four options: **Not Stim.** = Recording (the button in MEA layout is grey and labeled with the number of the electrode). **Ground** (the button in MEA layout is grey without number of electrode). **Stim. A** = Stimulation A (the button in MEA layout is blue and labeled with the number of the electrode). **Stim. B** = Stimulation B (the button in MEA layout is red and labeled with the number of the electrode). The software allows to select any electrode on a MEA for recording, grounding, and stimulation. You can apply two different stimulus protocols A and B.

A **blanking signal** (TTL pulse) transiently switch off the input stage of the amplifier during the stimulus, thus avoiding the stimulus artifacts on non-stimulating electrodes. Amplifier saturation is effectively prevented and the recovery time is greatly reduced. It is also possible to record from stimulation electrodes shortly after stimulation.

With the MEA\_Select program, it is easy to change the electrode selection during the experiment, for example, to use stimulation electrodes for recording and vice versa. You can also ground unwanted or bad electrodes by mouse click. Do not forget to send modified settings to the MEA amplifier with **Download** command.

Without blanking technology, recovery time depends on the filter amplifier type. It is roughly the time constant of the high pass filter multiplied by ten. During this time, the data output is distorted. With the blanking circuit, the voltage outputs of the preamplifier are held constant during the time when the blanking input is active. Thus, the following filter amplifier does not get saturated by the stimulus artifact. When the blanking signal has stopped, the channel output follows the input signal again.

By pressing the button **Download** you send the setting information to the MEA amplifier.

**Port** shows the connection to the serial port RS232 to which the MEA amplifier is connected. With the option **Test Conn.** = test connection you are able to check whether you connected the MEA amplifier to the fitting serial port. If not there is an error message "Connection not Ok." and you have to change the port and check the cable. If the connection is perfect the message "Connection Ok" and the version of the firmware of the MEA\_Select program are displayed.

The box **Blanking** must be enabled in most cases, but you can disable the blanking for testing your amplifier, for example.

The button **Change MEA** sets all electrodes to ground and is used for manipulations on the MEA amplifier. When replacing a MEA chip, the stabilizing time can be greatly reduced by connecting all electrodes to ground during the opening of the amplifier and change of the MEA. The connection to ground results in lower electrode offsets. Therefore, you should always use the "Change MEA" command when replacing a MEA.

**Important:** Click **Change MEA** in the MEA\_Select program before you remove the shielding or open the amplifier. Deactivate the **Change MEA** mode only after having completely set up the amplifier, including grounding the bath and shielding the amplifier. Otherwise, it can take very long (several seconds to minutes) until the amplifier has recovered and is ready for operation.

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On the right side of the main window the **Configuration** box gives you the possibility to reuse electrode configurations which are saved before. The configuration file has the extension ".scl". This feature is important for the reproducibility of experiments. You can also save the list of blanking patterns in **List Mode** together with a configuration file or just the list.

You can use the **Wait** for optimizing the electrode's behavior. The **Wait** parameter is the delay between switching the electrodes from stimulation back to recording, that is, the stimulating electrodes are disconnected from the stimulus input at the end of the trigger signal, and all electrodes are connected to the amplifier input at the end of the **Wait** period. The **Wait** helps avoiding cross-talk between stimulating and non-stimulating electrodes as well as preventing switch artifacts.

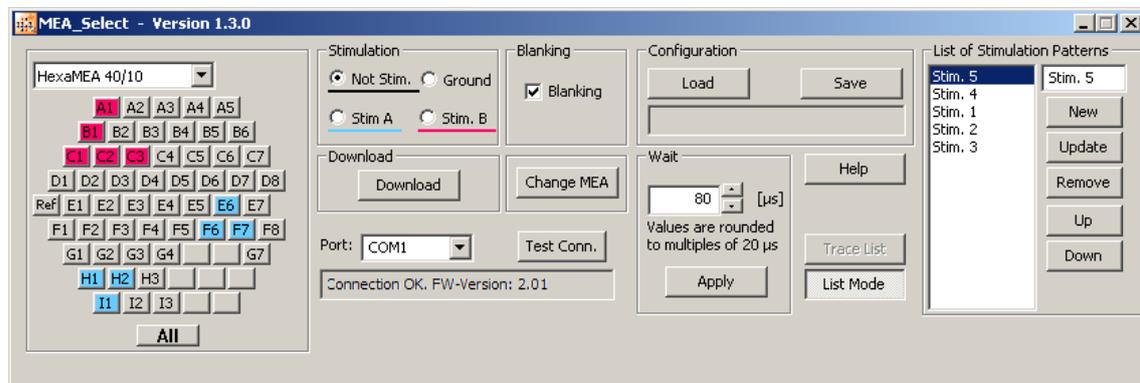
The length of the **Wait** depends on several factors (electrode performance, stimulus amplitude, for example) and has to be optimized empirically for each experimental setup. To test this, enter increasing **Wait** values and monitor the performance of the stimulus artifact suppression. Generally, the lower the amplitude of your stimulus is, the shorter can be the blanking signal. For strong stimuli, a **Wait** of up to 400  $\mu\text{s}$  can be necessary.

A factory-set **Wait** of 40  $\mu\text{s}$  is added to all user defined **Wait** values. For example, a user defined **Wait** of 0 results in a total delay of 40  $\mu\text{s}$ .

The command **List Mode** opens an additional window: **List of Stimulation Patterns**. In **List Mode** you can download a list of up to 50 user defined stimulation patterns. The quantity of 50 patterns is hardware controlled. A stimulation pattern can contain single or multiple electrodes and must get an user defined name. Click "New", and it appears in the small upper window pane. Change "New" into your specific name for the actual pattern, click "Update" to confirm the name and add the pattern to the list. By clicking the button **Trace List**, the stimulation patterns in this list are applied one after the other as stimulation signals.

**Note:** A Firmware update is necessary to use this feature.

Please read chapter "Stimulation" for more information.



It is very convenient to use a stimulus generator and MC\_Stimulus to set up both your stimuli and the blanking signal. You can then use the digital Sync Out output channel for controlling the blanking of the MEA preamplifier and triggering the recording with the MC\_Card or an USB based data acquisition device.

### 3.4 Setting Up and Connecting the MEA Amplifier and the STG



**Warning:** Spilled liquid can damage or even completely destroy the electronics of the MEA amplifier. Please be extremely careful when setting up your perfusion system and when starting the perfusion. Take care that the flow rates of the inlet and outlet flow match so that flooding of the amplifier is efficiently prevented.

**Note:** The housing of the MEA amplifiers was optimized for Zeiss Axiovert microscopes. MEA Amplifiers are compatible to most standard microscopes if the following prerequisites are met. This MEA amplifier type has been developed for standard inverse microscopes with a rectangular microscope table. The focal plane of the microscope has to be in a distance of 3.5 mm to the microscope table.

**Note:** If you use a complete MEA-System, the MEA amplifier will usually be powered by the isolated power supply (IPS10W) that is integrated into the data acquisition computer. The power is distributed along the MCS 68-pin MCS high grade cable. If you use your MEA amplifiers together with a MEA Switch, or if you use a custom data acquisition system **without** IPS10W, you will need an **external power supply** for operating the amplifier(s). Please ask your local retailer for more information on setup options.

**Note:** If you use an USB-ME-System you need an **external power supply**, for example a PS40W, for operating the amplifier(s). Please ask your local retailer for more information on setup options.

Connect the MEA preamplifier with blanking circuit to the filter amplifier's input with a 68-pin MCS standard cable. Using an USB-ME data acquisition device: Connect the filter amplifier's output to the USB-ME input via 68-pin MCS standard cable. If you are using a MEA Switch, connect the amplifiers to the MEA Switch inputs, and the MEA Switch output to the USB-ME device.

Place the MEA amplifier onto the microscope table.

Connect the internal heating element to a Temperature Controller's output channel (D-Sub9 socket) with the black cable. (The Temperature Controller is not part of the standard scope of delivery, but is included in the MEA-System or USB-ME-System, and can also be ordered separately.) Do not connect the black heating element cable to the data acquisition computer.

Connect the serial port of the preamplifier to a free RS232 serial port of the computer, from which you like to control the MEA amplifier (usually the data acquisition computer) with the provided RS232 cable. Note that you need a separate Com port for each amplifier in use.

Place the STG on a stable surface, where the air can circulate freely and the STG is not exposed to direct sunlight.

Connect the power supply unit to the STG and to the power outlet.

Connect the USB connector to the USB port of the computer. The computer connection is necessary for programming the STG, but not for operating it (except of Batch or streaming mode).

Connect a BNC cable with BNC-BNC heads from one of the Sync Out connectors on the rear panel of the STG (channel 1 or 2) to the T-fitting mounted on the BNC connector of the MEA1060-INV-BC amplifier.

Connect a BNC-Lemo cable from the T-connector to the Digital Trigger IN at the front panel of the USB-ME device.

Connect the red stimulator cable to the plus (+) connector of channel 1 on the front panel of the STG and to channel A of the MEA1060-INV-BC amplifier.

Connect the black cables to the ground (GND) connectors of the STG and to one of the connectors labeled "G" of the MEA1060-INV-BC amplifier.

Connect the blue cable to the (+) connector of channel 2 of the STG and to channel B of the MEA1060-INV-BC. You can change the red and the blue cable, it does not matter. But please connect the black cables from the ground connector of the STG to the ground connectors of the MEA1060-INV-BC amplifier.

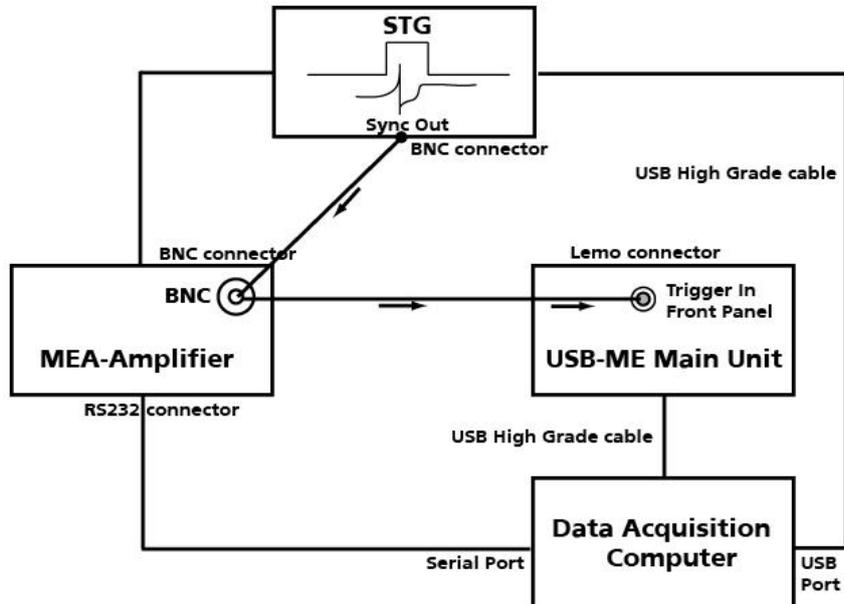
## MEA Amplifier with Blanking Circuit for Inverse Microscopes

Switch the STG on by pressing the toggle switch on the rear panel.

Note: It is recommended to start the STG ten minutes before use to allow warming up!

Please see also the separate handout "MEA Microelectrode Systems" for setup suggestions with detailed illustrations.

The following illustration shows a suggested setup for an USB-ME-System with a MEA1060 amplifier with blanking circuit.



## 4 First Tests and Tutorial

### 4.1 First Functional Tests

Each MEA amplifier has been thoroughly tested at the factory site before delivery. However, you may want to perform some tests yourself before you begin your experiment to exclude any damage that might have occurred during transportation, or to fulfill your own guidelines, for instance.

Some of the tests will also help you to get to know the basic functions of the hard- and software, like a short tutorial. It will take only a few minutes time and can save time and trouble in the long run. Multi Channel Systems MCS GmbH recommends running these tests after the setup of your system before you start your real experiments.

### 4.2 General Performance / Noise Level

Please use the provided test model probe to test the amplifier immediately after installation. The test model probe is already mounted on the amplifier. It simulates a MEA with a resistor of 220 k $\Omega$  and a 1 nF capacitor between bath and electrode. Use MC\_Rack or your custom data acquisition program to record from the test model probe and to check the amplifier.

#### Setting up MC\_Rack

Please refer to the MC\_Rack manual for more information.

1. Start MC\_Rack.
2. Open the file **MEA\_Display.rck** on the installation volume. This basic rack contains the virtual **MC\_Card** instrument with appropriate gain settings and a continuous raw data display.

- OR -

Set up the rack on your own:

1. Click **Data Source Setup** on the **Edit** menu. Select **USB-MEA or MC\_Card** in **Data Source** drop down menu. Select a 2-dimensional (MEA) layout or Configuration in **Source Layout**. Close the dialog with OK.
2. Add the **Data Source** to your virtual rack.
3. Click the **Hardware** tab and enter the appropriate amplifier gain (standard: 1100).
4. On the **Edit** menu, click **Add Data Display** to add a raw data display to your virtual rack.

## MEA Amplifier with Blanking Circuit for Inverse Microscopes

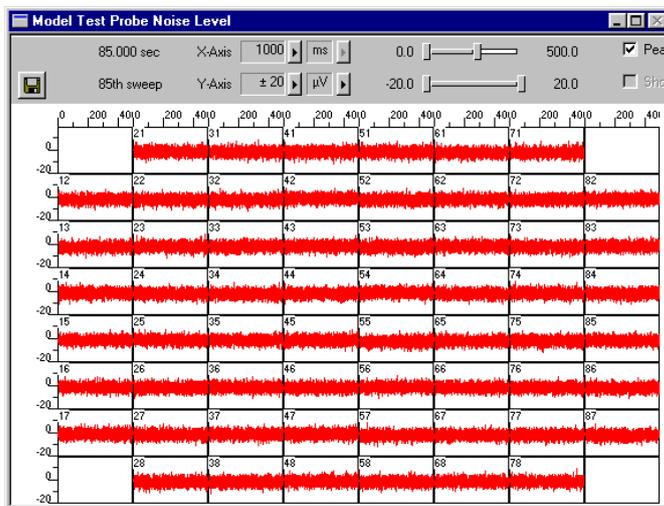
### Starting the recording

1. On the **Measurement** menu, click **Start** to start the recording. You see the raw data streams of all 60 channels in the typical MEA layout.
2. You may have to **adjust** the **position** and **span** of the **axes** until you can clearly see the noise level.

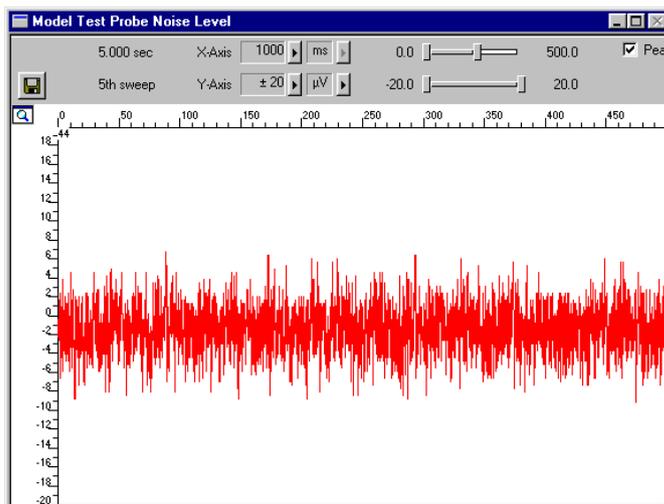
During the first minute, you will see the baseline on all channels, because the filter amplifier is still saturated. All electrodes have a different DC offset at the beginning of an experiment. The internal baseline restore routine restores the offset to the ground level, that is, to zero. During this time, you will not see any "true" signals or noise. See also the chapter "Operating the MEA Amplifier", "Signal Amplification and Filters".

After about one minute, the baseline restorer has corrected the DC offset, and you see true signals. You should see the baseline with a maximum noise level of  $\pm 8 \mu\text{V}$ .

The following screen shot shows a recording from a typical MEA amplifier with a test model probe and a sampling rate of 25 kHz.



→ Double-click a channel in the display to have a closer look.



## 4.3 Computer Connection

### Testing the computer connection

The MEA\_Select program automatically detects and lists all serial ports of your computer.

1. Start MEA\_Select.

Select the serial port to which you have connected the MEA preamplifier (COM1, COM2, and so on) from the drop down list.



1. Click **Test Connection**. A valid computer connection should be confirmed by **OK**. If not, please check all cables and connections.

**Note:** You can control multiple MEA amplifiers from the same computer by starting several instances of the MEA\_Select program. Connect the amplifiers to different serial ports and select the appropriate serial port for each amplifier in the MEA\_Select program.

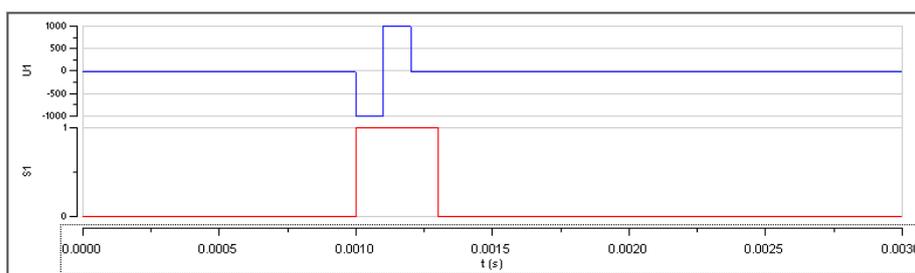
## 4.4 Stimulation and Recording

In this chapter, we provide a step by step instruction for first tests that we think useful for learning more about the features and behavior of the amplifier. It is assumed that you will use MCS components (MEA-System and stimulus generator) for the complete setup. Please make sure that you have set up the system as described in the chapter "Setting Up and Connecting the MEA Amplifier" (or likewise, if you use other components). You can use the provided **test model probe** for your first experiment.

First, you have to set up your stimulus generator and MC\_Rack. Then you can start MEA\_Select to operate the MEA amplifier. Of course, you can also change your MC\_Stimulus or MC\_Rack settings during amplifier operation.

### Setting up the Stimulus Generator of 4000 series

1. Start the stimulation software MC\_Stimulus II.
2. Open the demo stimulus file on the installation volume. You can choose between a monophasic and biphasic stimulus (biphasic stimuli with the negative phase first are recommended). The voltage demo stimulus is a biphasic or monophasic pulse with a duration of 100  $\mu$ s for each phase and an amplitude of 1 V / +1 V. The pulse is repeated continuously each 24 ms (with the **Repeat** feature of MC\_Stimulus). The Sync Out trigger pulse for triggering the MEA amplifier and the MC\_Card is 100  $\mu$ s longer than the stimulus pulse.



3. On the **STG** menu, click **Download (all)** to download the file onto the stimulus generator.

4.4.1 Operating multiple MEA Amplifiers with one STG

It is possible to operate more than one MEA amplifier **independently** with a single stimulus generator of the 4000 series. To do this, activate the **Independent Channel Download** feature in the **Settings** menu of MC\_Stimulus II.



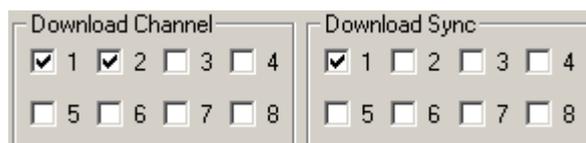
This feature will allow to download new stimulation paradigms to individual channels without interrupting the ongoing stimulation on other channels. See chapter "Downloading Stimulus Files" for more information.

It is mandatory that all stimulation and Sync Out channels that are associated with one amplifier are assigned to the same trigger in the **Trigger Settings** menu. For example, if **four MEA1060-BC** amplifiers are connected to **one STG4008**, usually two stimulation channels and one Sync Out are associated to each amplifier. In that case, the trigger settings should be as shown:



Amplifier A is connected to stimulation channels 1 and 2 and Sync Out 1, Amplifier B is connected to stimulation channels 3 and 4 and Sync Out 2, and so on. All channels connected to one amplifier are assigned to the **same trigger**.

To download a stimulation paradigm to individual channels assigned to one amplifier only, select all channels connected to this amplifier in the "Download Channel" check box (1 and 2 in this example) and Sync Out 1 in "Download Sync" check box, whether you changed them all or not!



Only these channels will be downloaded, **ongoing stimulation** on other channels will not be interrupted.

To **start and stop** only the channels associated with **one amplifier**, use the Start / Stop buttons for individual triggers. Each button controls one trigger and hence all assigned channels. The orange LED indicates **ongoing stimulation**.

In our example, the configuration shown below would mean that amplifiers A, B and D are receiving a stimulation signal, while stimulation for amplifier C is stopped.



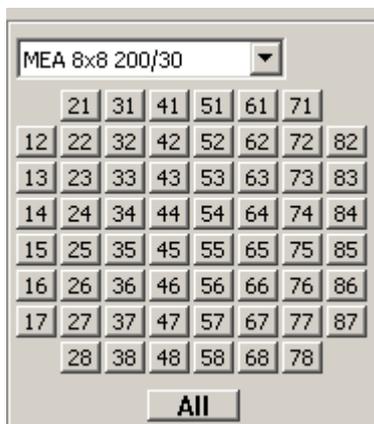
Triggers 5 to 8 have no function in this example. You could download a new paradigm for amplifier D by downloading channels 5 and 6 and Sync Out 3, and then start it with the start button of Trigger 3. This would not interfere with the ongoing stimulation to the other three amplifiers, but only if the **Independent Channel Download** function is activated. If the STG is in default download mode, any download will stop all ongoing stimulation and empty the memory of the STG. See chapter "Downloading Stimulus Files" for more information.

### Data acquisition with MC\_Rack

1. Start MC\_Rack.
2. Open the demo rack file **MEA1060BC\_demo.rck** on the installation volume. It is a simple rack with only a **Trigger Detector**, one continuous and one triggered **Data Display**. You will need a trigger for stimulation and blanking, and for triggering the triggered display.
3. Bring the continuous display to front.
4. Start the recording by clicking **Start**.

### Operating the MEA amplifier with blanking circuit

- Start MEA\_Select.
- On the left, you see the MEA layout button array. Here, you can select any electrodes for stimulation or connect them to ground. The default state of all electrodes is selected for Recording = **Not Stim.**. This is indicated by the neutral grey background of the electrode buttons. Accordingly, you will see the standard noise level on all channels in the continuous display in the MC\_Rack program. The triggered display is still empty, because you have not started triggering the stimulation and the MC\_Card yet.

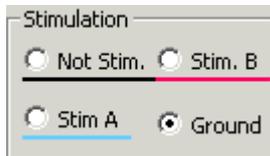


## MEA Amplifier with Blanking Circuit for Inverse Microscopes

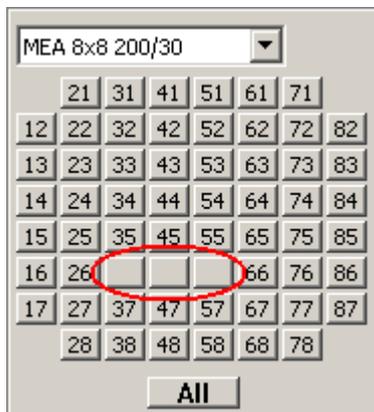
### Grounding electrodes

You can connect any electrode to ground. Usually, you will ground defective electrodes. Now, we are trying out this feature even if it makes no sense with the MEA test model probe.

1. Under **Stimulation**, select **Ground**.

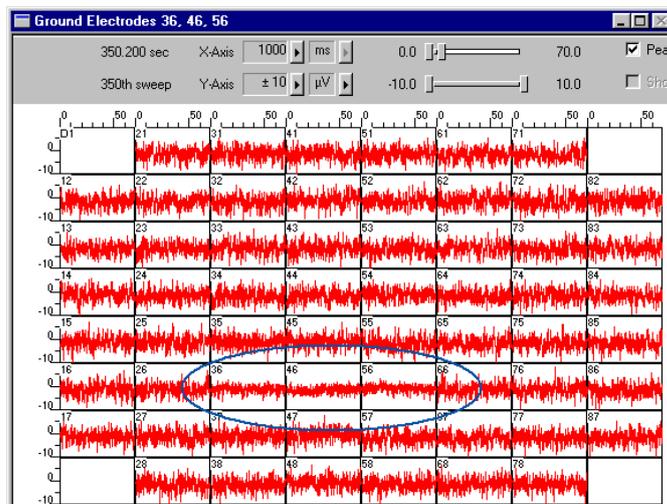


2. Click any electrodes that you like to ground, for example electrodes 36, 46, and 56. Grounded electrodes show no electrode number, only a blank button. By clicking on one of the grounded electrodes again you can toggle back to recording mode. If you click, for example in Stimulation mode B onto a grounded electrode, you overwrite the grounded mode with Stim. B mode. That means, the color will turn to red and the number of the electrode appears again. If you click for the third time, the selected electrode toggles back to recording (= default) mode.



3. Click **Download** to download this information on the MEA amplifier.
4. Switch to the MC\_Rack display.

You can see that the noise level of the grounded electrodes is reduced. The effect is quite small because the model test probe has no defective electrodes and shows a fine low noise level on all "electrodes".



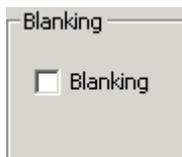
## Stimulation without blanking

Usually, you will always use the blanking when you stimulate. But it is possible to switch off the blanking for testing your setup.

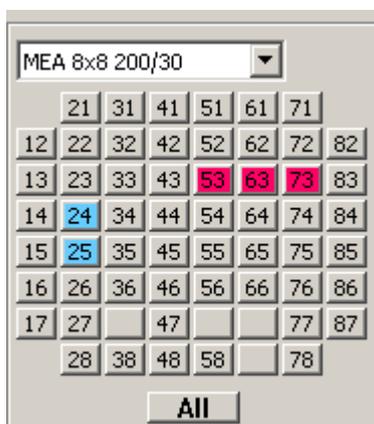
1. In MC\_Rack, bring the triggered display to the front. Make sure you have connected the digital Sync Out output of the stimulus generator to the digital channel 1 (bit 0) of the MC\_Card. Otherwise, you have to select the appropriate bit in the **Trigger Detector**.



2. In MEA\_Select, under **Blanking**, deselect **Blanking**.



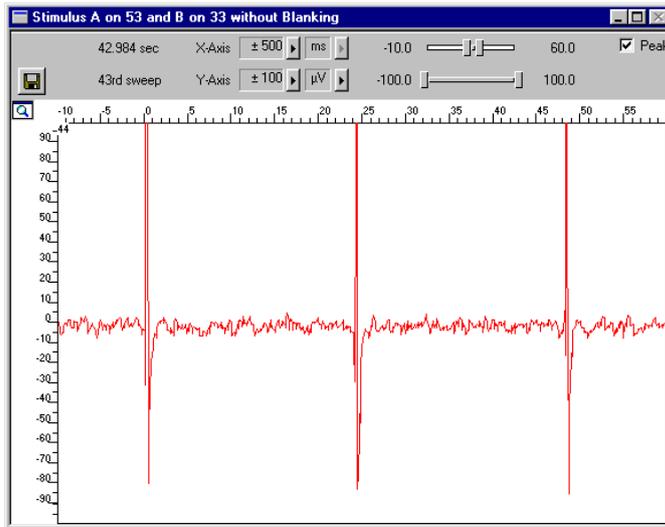
3. Under **Stimulation**, select the appropriate stimulus input (**Stim A** or **Stim B**).
4. Click any electrode that you like to use for stimulation, for example 53. The stimulation electrodes are marked by a color, that is, blue for stimulus **A** and red for stimulus **B**. By clicking on one of the selected electrodes again you can toggle back to recording mode. If you click, for example in Stimulation mode B onto a blue (Stim. A) or grounded electrode, you overwrite the mode with Stim. B mode. That means, the color will turn to red and the number of the grounded electrode appears again. If you click for the third time, the selected electrode toggles back to recording (= default) mode.



5. Click **Download** to download this information on the MEA amplifier.
6. Start the stimulus generator.

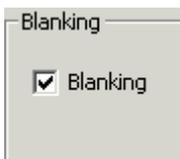
## MEA Amplifier with Blanking Circuit for Inverse Microscopes

The stimuli are sent to the selected stimulation electrodes. You see stimulus artifacts on all channels. The display operates now in a triggered mode, so that the stimuli appear aligned to the trigger event.

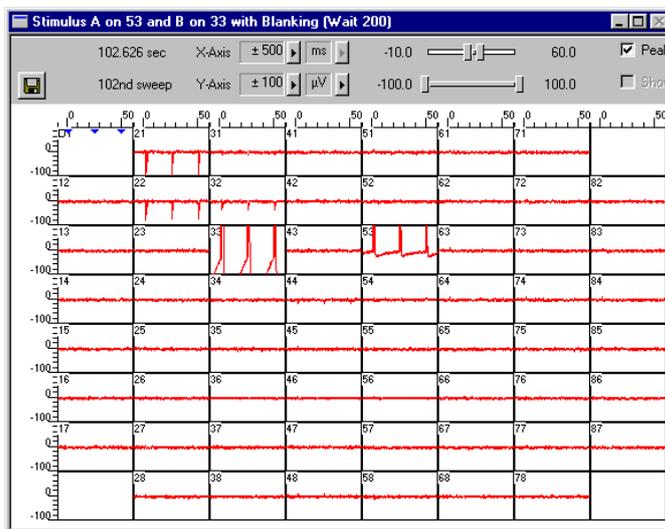


### Stimulation with blanking

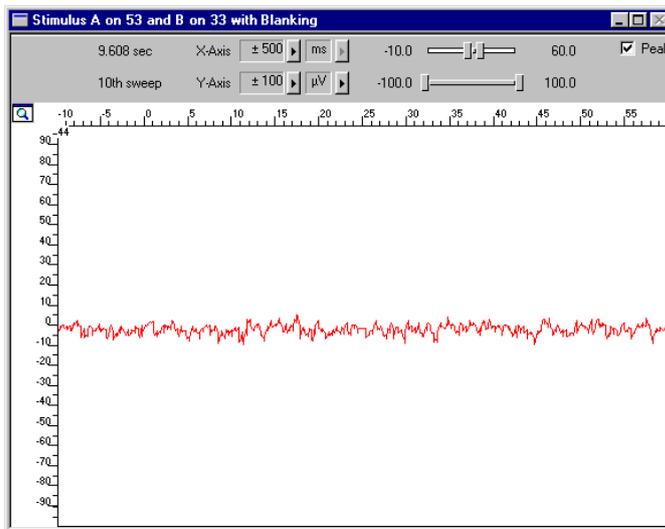
1. Select the **Blanking** in the MEA\_Select program.



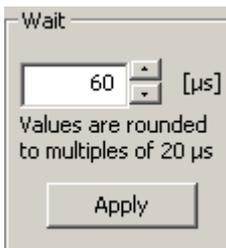
2. The Sync Out signal of the stimulus generator triggers not only the stimulation, but the blanking as well.



You see that there are no stimulus artifacts visible on almost all non-stimulating electrodes, with some residual artifacts on the neighbor electrodes of the stimulating electrode 33. In this example, we have used a **Wait** of 200  $\mu\text{s}$ .



### Wait settings



You can use the **Wait** for optimizing the electrode's behavior. The **Wait** parameter is the delay between switching the electrodes from stimulation back to recording, that is, the stimulating electrodes are disconnected from the stimulus input at the end of the trigger signal, and all electrodes are connected to the amplifier input at the end of the **Wait** period.

The **Wait** helps avoiding cross-talk between stimulating and non-stimulating electrodes as well as preventing switch artifacts.

The length of the **Wait** depends on several factors (electrode performance, stimulus amplitude, for example) and has to be optimized empirically for each experimental setup.

- Enter increasing **Wait** values and monitor the performance of the stimulus artifact suppression.



## 5 Operating the MEA Amplifier

### 5.1 Signal Amplification and Filters

The MEA amplifier with **blanking circuit** is a 60-channel preamplifier with a broad bandwidth. Filter specifications and gain are defined by the following filter amplifier.

Different filter settings are used for different applications to enhance the signal-to-noise ratio. The pass band of the filter amplifier should be chosen according to the signal type. It is generally useful to set the upper limit of the amplifier near the highest expected signal frequency, but also at a safe distance to make sure that the full gain is used for signal amplification.

For slow signals like field potentials, a bandwidth of 1 to 300 Hz is appropriate. If you like to record fast signals like spikes, a passband of 300 Hz to 3 kHz is suitable. Cardiac signals have fast and slow components; therefore, you usually need a wider bandwidth of 1 Hz to 3 kHz.

Multi Channel Systems provides custom amplifiers with a bandwidth of your choice, from 0.1 Hz to 10 kHz. Please note that it is often useful to acquire the data with a broadband amplifier and use the digital filter of the free MC\_Rack program to change the pass band and filter the raw data. This way, you are much more flexible in designing your experiments. As a further advantage, you can see the original (not filtered) data as well. This is especially important because all filters are known to distort signals. On the other hand, you may need a higher sampling rate to avoid aliasing, and you will have a lower signal to noise ratio.

The standard gain of a MEA amplifier is 1200 (1100 in case of a MEA amplifier with blanking circuit), which is fine for most applications, but MCS can also provide amplifiers with a gain of your choice (from 100 to 5000) as well. For large signals (for example, from whole-heart preparations), you need a lower gain to prevent a saturation of the amplifier. Please note that the gain is a fixed hardware property; and that you cannot change the gain of the amplifier by software controls. For more information on the technical specifications of your amplifier, please see the separate data sheet that is delivered with each amplifier.

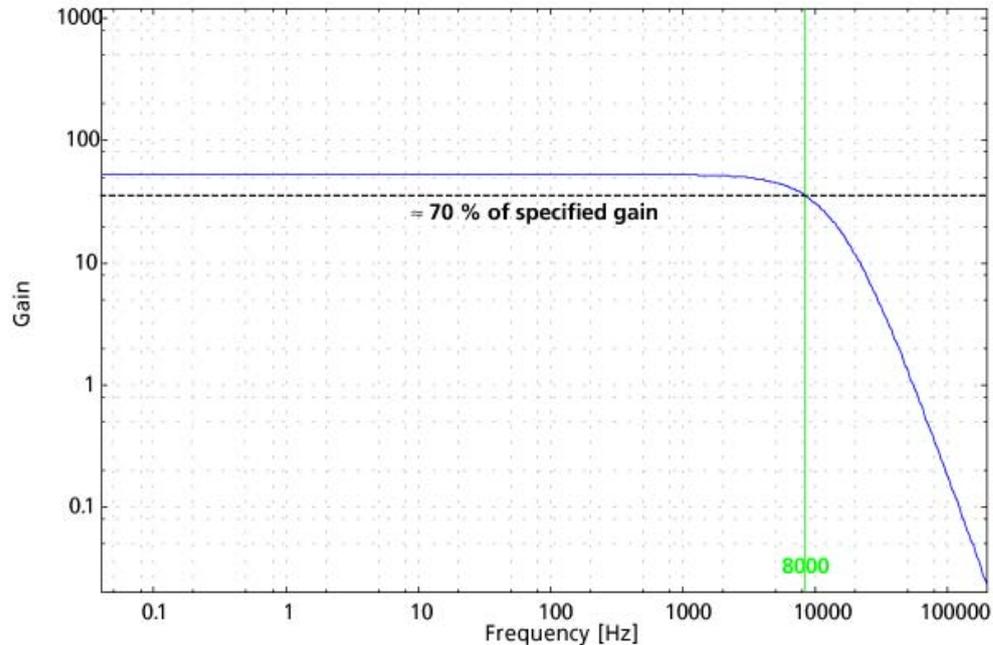
Please note that the ratio of the output signal to the input signal, that is, the gain, is not a fixed parameter for the complete bandwidth. The gain that was specified for the amplifier, for example, 1200, is not fully reached at the borders of the amplifier's pass band. The general rule is, that at the lower and upper limit of the frequency band, the gain is  $\sqrt{2}/2$ , that is approximately 70 %, of the full gain. Therefore, you should use a bandwidth that is at a safe distance of the signals of interest. Outside the pass band, the gain decreases with the frequency and finally approaches zero.

The first illustration shows the broad frequency band of the MEA1060-BC amplifier alone (without Filter Amplifier); the following illustration shows a typical frequency band of the MEA amplifier with standard gain and bandwidth (in blue). Output signal amplitudes were divided by the input signal amplitude and the resulting gain was plotted versus the input signal frequency on a logarithmic scale. A straight line at approximately 70 % of the specified gain intersects the plotted curve at the lower and upper limit of the pass band. The low pass filter property of the combined MEA1060 amplifier and MC\_Card (in black) is only slightly different.

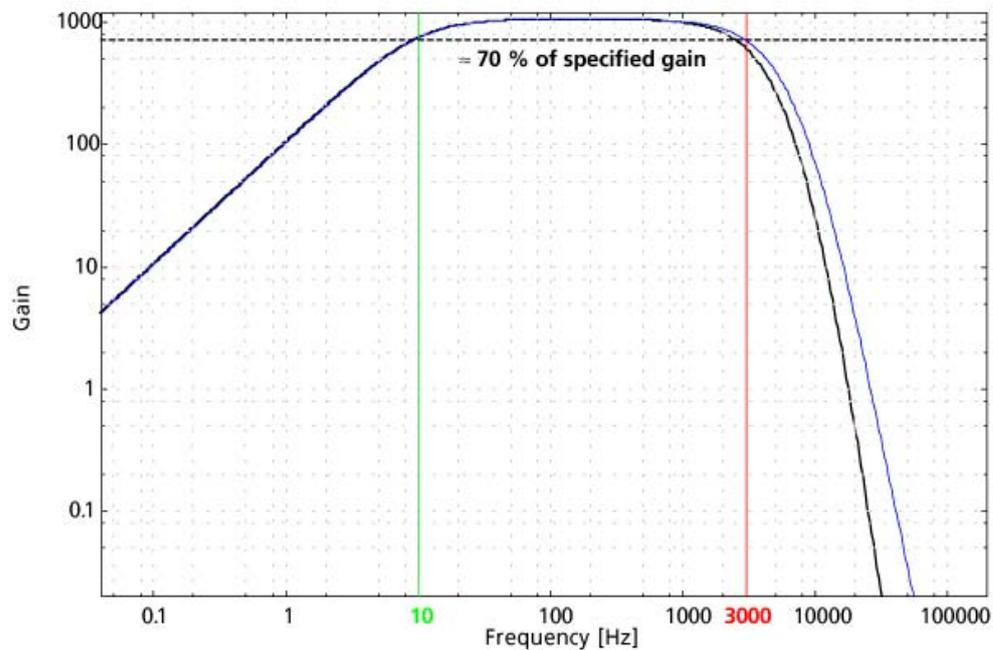
## MEA Amplifier with Blanking Circuit for Inverse Microscopes

For information on the gain and filters of the MC\_Card data acquisition system, please see the ME- / MEA-System manual. For more information on gain and filters in general, please refer to standard literature or contact your local retailer.

### MEA1060-BC-Standard frequency band (without FA60S)



### MEA1060-BC-Standard frequency band including FA60S (+ MC\_Card)



If you use a MEA-System, the isolated power supply IPS10W that is integrated in the data acquisition computer supplies the power for operating the MEA amplifier. The power is distributed along the MCS high grade cable. Please consider that the amplifier can only operate properly if the supply voltage and current specifications are fulfilled, especially if you use a custom power supply. See also the Technical Specifications and the Troubleshooting section.

## DC offset correction

The internal baseline restore routine of the preamplifier restores the offset to the ground level, that is, to zero. This has to be a slow process. If the routine corrected too fast, slow signals would be eliminated as well.

All electrodes on an array have different DC offsets at the beginning of an experiment. The DC offsets are amplified by the preamplifier, and the amplified output signal of the preamplifier is likely to exceed the input range of the following filter amplifier. In order to eliminate this DC offset, a so-called baseline restorer is integrated in the preamplifier. For signals in the usual measurement range, the baseline restore routine acts like a high pass filter with a very low cut-off frequency of roughly 0.01 Hz, but unnaturally high signals like stimulus artifacts are limited to a negligible value.

Due to the low cut-off frequency, the baseline restorer takes some time to stabilize after switching on the amplifier or after replacing the MEA. One to two minutes are quite normal. During this time, the filter amplifier input will be overloaded and you will see only a straight line with very low noise. This is considered a normal behavior and should not trouble you. Please wait until the baseline restore routine has been completed.

When replacing a MEA, the stabilizing time can be greatly reduced by connecting all electrodes to ground during the opening of the amplifier and change of the MEA. The connection to ground results in lower electrode offsets. Therefore, you should always use the **Change MEA** command when replacing a MEA. See also the chapters First Tests and Tutorial, General Performance / Noise Level and Operating the MEA Amplifier, Mounting the MEA Probe.

## 5.2 Temperature Control

The biological sample is cultured directly on the MEA. You can record without needing an incubator, because a heating element is integrated into the amplifier and the perfusion cannula and are both controlled by a Temperature Controller (Temperature Controller and perfusion cannula with heating element and sensor are either included in the MEA-System or can be ordered separately). You have several options regarding the culture chamber, such as a semipermeable seal that guarantees stable environmental conditions.

For using the integrated heating element of the MEA amplifier, connect it with the black cable to the temperature controller. The heating is active, but the cooling is passive. Therefore, the minimum temperature is limited by the room temperature. Please refer to the temperature controller manual for more information.

**Important:** Please make sure that you have selected the appropriate PID coefficients for the amplifier type (for inverted or upright microscopes) in use.

## 5.3 Mounting the MEA Probe and Grounding the Bath



Warning: Do not use too much force. Otherwise you could damage the delicate MEA or contact pins of the amplifier. Put the lid of the amplifier only onto a dry and clean area with the bottom side down. Otherwise, you can easily damage the contact pins.

Important: Click **Change MEA** in the MEA\_Select program before you remove the shielding or open the amplifier. Deactivate the **Change MEA** mode only after having completely set up the amplifier, including grounding the bath and shielding the amplifier. Otherwise, it can take very long (several seconds to minutes) until the amplifier has recovered and is ready for operation.

### Mounting the MEA Probe



1. In the MEA\_Select program, click **Change MEA**. All electrode inputs are connected with the amplifier's ground. This helps avoiding a large DC offset and reduces the baseline restoring time significantly. See also the chapters "Operating the MEA Amplifier", and "Signal Amplification and Filters".
2. Place the MEA amplifier as shown on the photo. Open the lid of the MEA amplifier.
3. Place the MEA probe inside.

Important: MEA chips are not symmetrical! That is, why the writing (NMI) on the MEA chip should be on the right side. Otherwise, the MEA layout will not match with the pin layout of the channel map in MC\_Rack. This is very important for MEAs with internal reference electrode, for example.

4. Replace the lid and close it carefully. Do **not** unpress the **Change MEA** button until you have grounded the bath and replaced the shielding, that is, until the setup has been completely finished.



Warning: Spilled liquid can damage or even completely destroy the electronics of the MEA amplifier. Please be careful when setting up your perfusion system and when starting the perfusion. Take care that the flow rates of the inlet and outlet flow match so that flooding of the amplifier is efficiently prevented.

### Grounding the bath

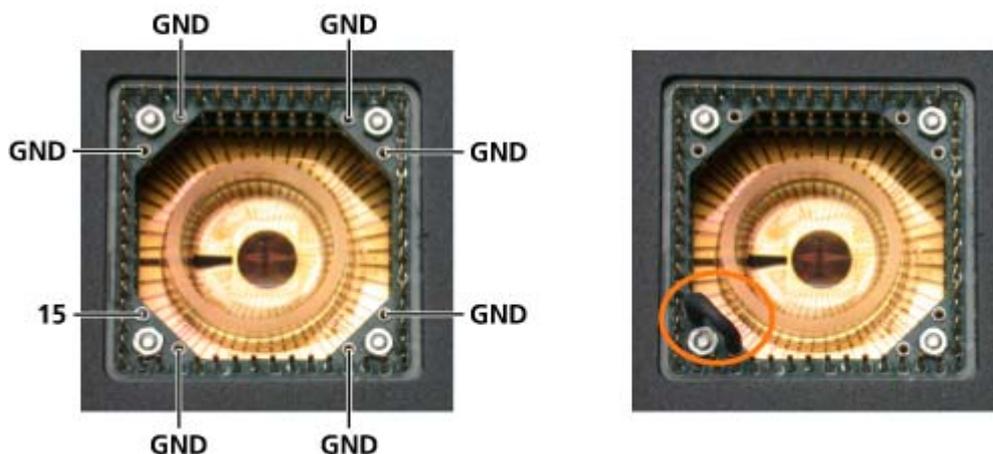
You can ground the bath with a plain silver wire or with an Ag/AgCl pellet. Only the Ag/AgCl electrode provides a stable intrinsic potential. In practice, the plain silver wire will be ok for 90 % of applications. If you do not achieve satisfying results with the plain silver wire, try an Ag/AgCl electrode.

If you use MEAs with internal reference electrode, you can use the reference electrode for grounding the bath. This has the advantage that you can keep the culture chamber closed and sterile (for example, with MEA-MEM semipermeable membranes).

**Important:** If you use a MEA with internal reference electrode, you have to connect pin 15 to ground with the provided cable. Setting electrode 15 to ground in the MEA\_Select program is not sufficient for grounding the bath.

1. Attach the provided **silver wire** or **pellet** to the amplifier's **ground** and place it into the **bath**, near the rim of the culture chamber. If you use a MEA with **internal reference electrode**, connect the ground to the **reference electrode socket (pin 15)** with the provided connector. Make sure that the orientation of the MEA inside the amplifier is appropriate.

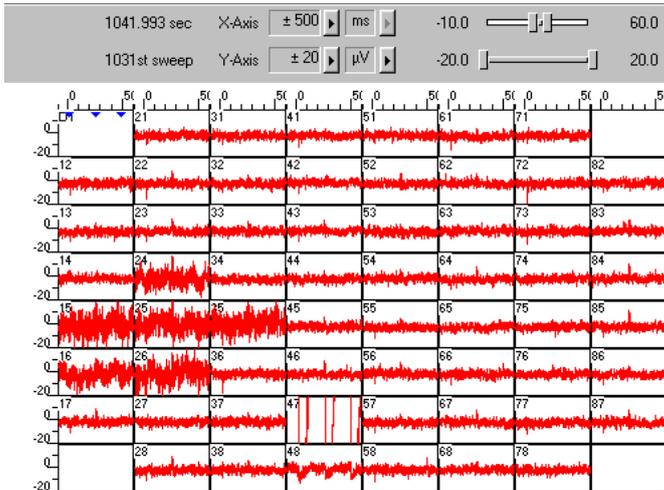
### Connecting electrode 15 to ground



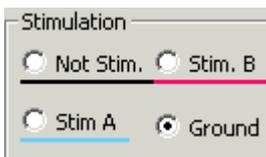
2. Replace the shielding. Make sure the setup is complete.
3. Click **Change MEA** again to reconnect the electrodes to the amplifier's input stage.
4. Wait until the amplifier has recovered and you see the standard noise level on all channels before starting your experiment. The recovery time depends on the electrodes used and usually takes about one or two minutes.

## 5.4 Grounding Defective Electrodes

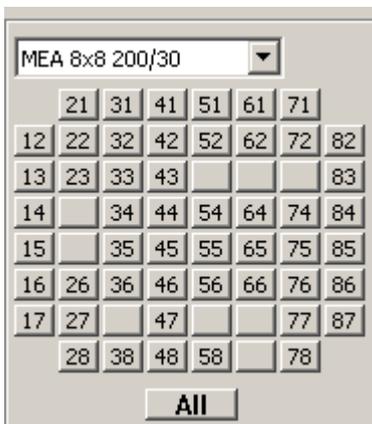
You can ground defective electrodes with the MEA\_Select program. In the following example, electrodes No. 15, 16, 24, 25, 26, 35, and 47 show a very high noise level.



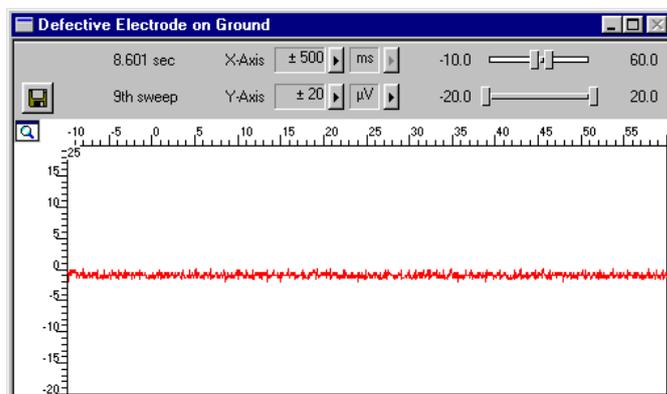
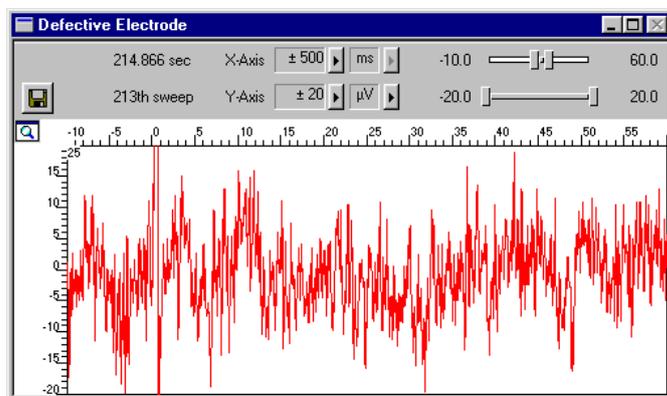
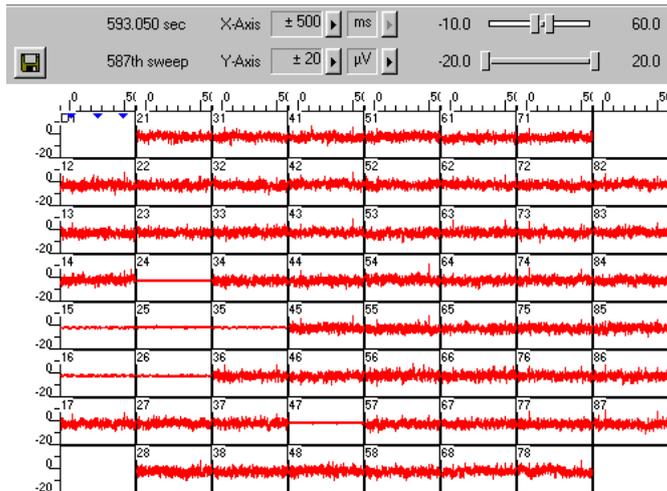
1. Under **Stimulation**, select **Ground**.



2. Click any electrodes that you like to ground. Grounded electrodes show no electrode number, only a blank button.



- Click **Download** to download this information on the MEA amplifier. You can clearly see the difference between the high noise level and the zero line of the grounded electrode.



## 5.5 Stimulation

### Recommended stimulus protocols for MEA electrodes

MCS recommends to use only **biphasic voltage** pulses with the negative phase first. The duration of the pulse should be 100  $\mu$ s for each phase and the amplitude between 100 mV and 3000 mV. Make sure that the voltage level of the stimulating electrode is zero at the end of the pulse.

(See also Potter, S. M., Wagenaar, D. A. and DeMarse, T. B. (2005). Closing the Loop: Stimulation Feedback Systems for Embodied MEA Cultures. Advances in Network Electrophysiology Using Multi-Electrode Arrays. M. Taketani and M. Baudry, Springer; Wagenaar, D. A., Madhavan, R., Pine, J. and Potter, S. M. (2005). "Controlling bursting in cortical cultures with closed-loop multi-electrode stimulation." J Neurosci 25(3): 680-8.)

### Stimulation with MEA electrodes

You can apply two different stimulus protocols to any selection of electrodes. Use the MEA\_Select program to select electrodes for recording and stimulation. The stimulation is triggered with a TTL pulse, the so-called **blanking signal**. Please see "Blanking" for more information how to set up and time the blanking signal.

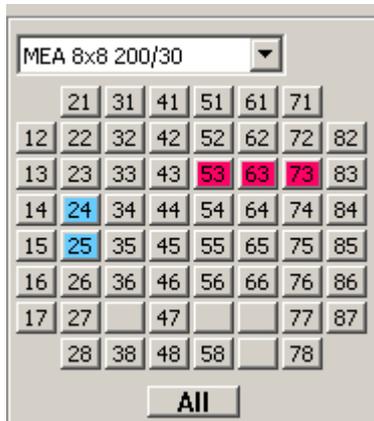
You can also stimulate without blanking, for example, for testing your setup. Under **Blanking**, deselect the **Blanking** option for disabling the blanking function.

**Important:** You can **only stimulate during an active TTL pulse**, which is used for triggering the stimulation **and** the blanking (see illustration of switch positions below). You can switch off the blanking for test purposes, but this is not considered a useful mode of operation, because the amplifier will get saturated by the high stimulus input without blanking.

**Important:** The **input voltage** of the MEA1060-BC amplifier is **limited to +/- 5 V**. Exceeding the voltage input range can result in unpredictable noise and artifacts. In rare cases, even damage to the amplifier electronics might occur. For current driven stimulation, the voltage amplitude depends on the electrode impedance. As the electrode impedance depends on several parameters, for example, the electrode's degree of wear and its hydrophilicity, and therefore cannot be predicted, it is generally recommended to use **only voltage driven stimulation**. Current driven stimulation may work with larger (30  $\mu$ m) TiN electrodes that have a low impedance, but this cannot be guaranteed by the manufacturer.

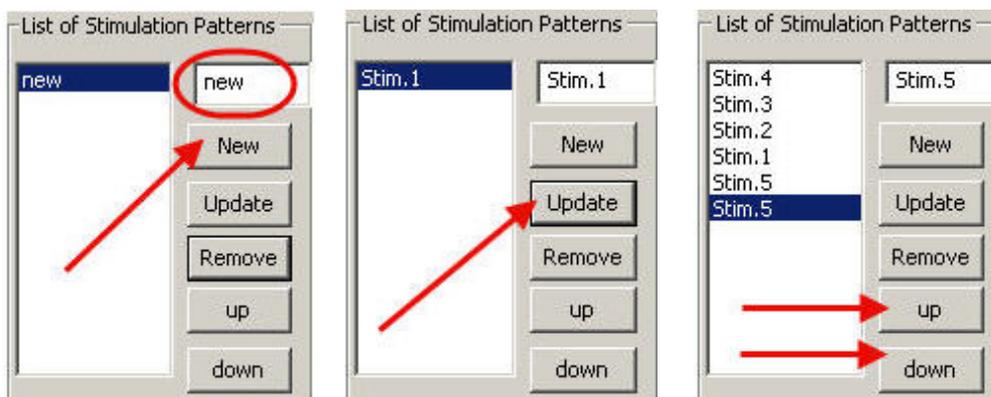
For more information on the setup, please see the chapter "Setting Up and Connecting the MEA Amplifier".

Under **Stimulation**, select a stimulus input (**Stim A** or **Stim B**) and click any buttons on the button array in the MEA layout. Click any electrode that you like to use for stimulation, for example 23 or 73. The stimulation electrodes are marked by a color, that is, blue for stimulus **A** and red for stimulus **B**. By clicking on one of the selected electrodes again you can toggle back to recording mode. If you click, for example in Stimulation mode B onto a blue (Stim. A) or grounded electrode, you overwrite the mode with Stim. B mode. That means, the color will turn to red and the number of the grounded electrode appears again. If you click for the third time, the selected electrode toggles back to recording (= default) mode.



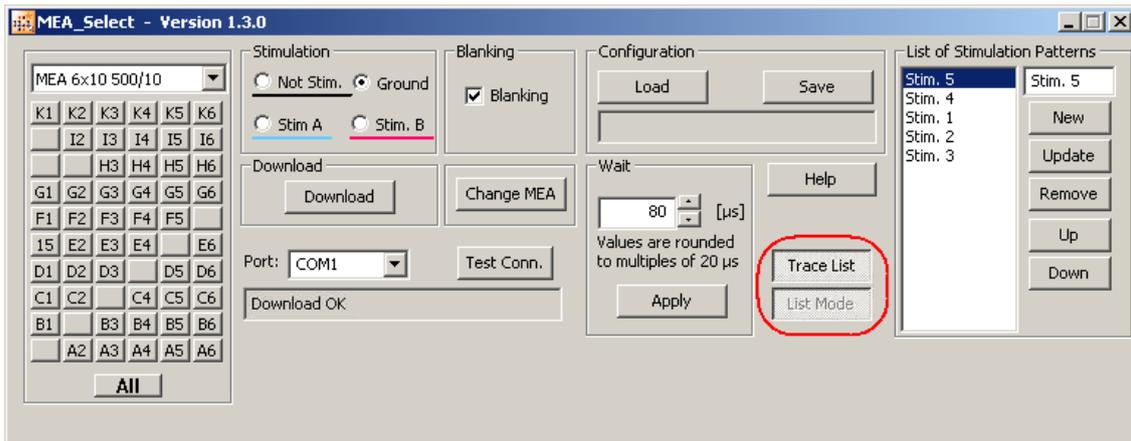
1. Click **Download** to apply the settings. This single stimulation pattern will be repeated as stimulation pulse until you download a different one.

If you like to download several (up to 50) stimulation patterns, click **List Mode**. Now you can create a list of different stimulation patterns. In the small upper window pane the term "**New**" will appear and you can change it into the specific name you will give the current stimulation pattern, for example, Stimulation Pattern 1 = Stim. 1, Stim. 2, Stim. 3. With **Update** you can confirm the name and the pattern will be added to the list of stimulation patterns in the left window pane. **Remove** eliminates the pattern, which is highlighted in blue. With **up** or **down** you can move the saved patterns in the list to arrange them in that sequence you like to apply them. To duplicate a saved pattern you have to click the selected pattern, so that it is highlighted in blue and confirm the same name with Update again. Click **Download** to send the list of stimulation patterns to the MEA amplifier.



## MEA Amplifier with Blanking Circuit for Inverse Microscopes

If the **List Mode** is open, there is an additional button **Trace List**.

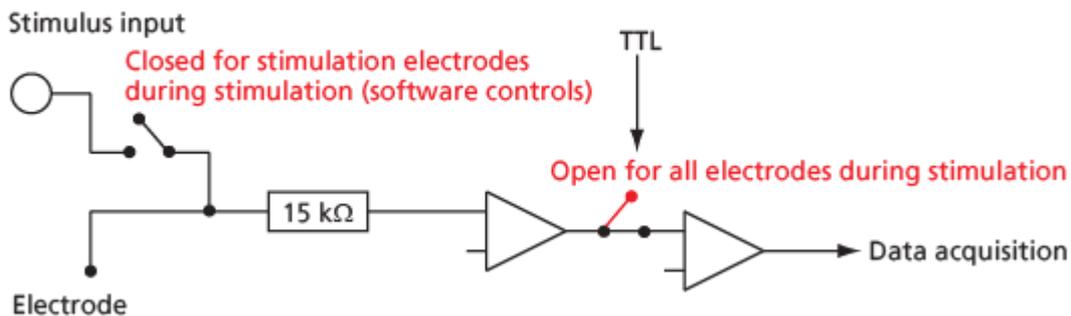


If the **Trace List** button is active all other functions are automatically disabled. The stimulation patterns of the list, created in **List Mode**, are applied one after the other as stimulation pulse. To control which pattern of the list is reached the name of the stimulus pattern will be highlighted in blue. Reaching the end of the list, the program reruns the list of the stimulation patterns, starting at the top of the list again. It is not recommended to change the settings of stimulation patterns during experiment. Please stop recording data, change settings and send the new information to the amplifier with **Download**. Then start recording again. It is possible to save the list of stimulation patterns by clicking **Save** under **Configuration** together with a previously loaded configuration file or you can save the list only. The file for just the list has the same extension as the configuration file ".scl".

**Note:** A Firmware update is needed to use this feature.

During the blanking signal, the **stimulus inputs** are **connected** to the software-selected **stimulation electrodes**, and **all MEA electrodes** are **disconnected** from the **amplifier**. The last signal value on all electrode channels before the blanking pulse is applied will be saved and then kept until the channels are reconnected to the amplifier.

### Switch position during recording and stimulation





### Wait settings

A **delay** (up to 5000  $\mu$ s) between **stopping stimulation** (end of TTL pulse) and **restarting the recording** can be programmed with the **Wait** property in the MEA\_Select program in order to optimize the blanking. The **stimulation** electrodes are **disconnected** from the stimulus input at the end of the **trigger signal**, and **all** electrodes are **connected** to the amplifier input at the end of the **Wait period**.

The **Wait** helps avoiding cross-talk between stimulating and non-stimulating electrodes as well as preventing switch artifacts of the amplifier.

The length of the **Wait** depends on several factors (electrode performance, stimulus amplitude, for example) and has to be optimized empirically for each experimental setup. Generally, the lower the amplitude of your stimulus is, the shorter can be the blanking signal. For strong stimuli, a **Wait** of up to 400  $\mu$ s can be necessary.

A factory-set **Wait** of 40  $\mu$ s is added to all user defined **Wait** values. For example, a user-defined **Wait** of 0 results in a total delay of 40  $\mu$ s.

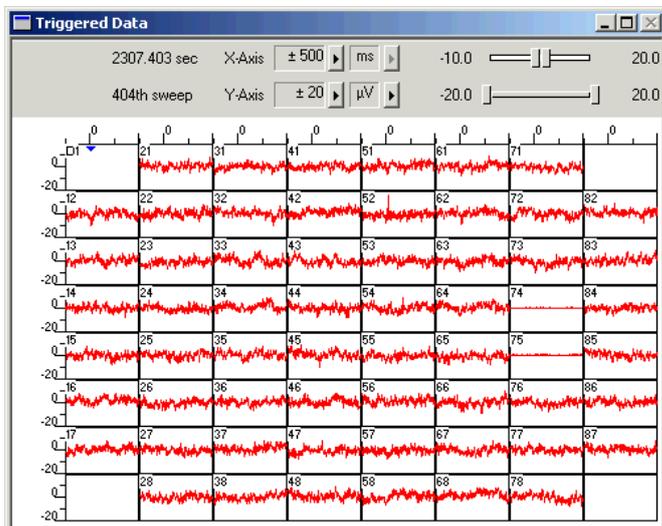
### Electrode impedance

A parameter that was found to be quite important for the stimulus artifact suppression performance is the **electrode impedance**. Electrodes that are bigger (30  $\mu$ m) or **TiN electrodes** show a **better** blanking performance compared to very small (10  $\mu$ m) or PT electrodes (3D-MEAs) with a higher impedance. Please note that a **hydrophilic** electrode has a lower impedance and therefore also shows a better blanking performance. New MEAs are generally very hydrophobic and should be **hydrophilized before use** (please see the MEA manual for more information).

Note: When using current driven stimulation, the combination of electrode impedance and current amplitude may lead to high voltages. Please regard the maximum input voltage of 5 V of the MEA1060-BC amplifier. Otherwise, an unpredictable behavior of the amplifier can be expected. In rare cases, even damage to the amplifier electronics might occur. MCS recommends to use voltage driven stimulation only.

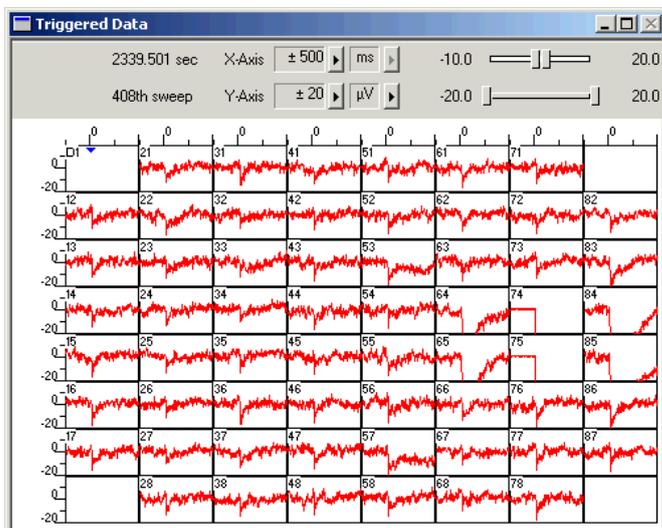
### Single pulses versus repeated pulses

Interestingly, the behavior of MEA electrodes when applying pulses that are **repeated** after some milliseconds, is **much better** than when a single pulse is applied. You can observe that the kinetics improve, that is, you need a shorter **Wait**, following the application of a few pulses. This is generally not a problem, as stimulation in regular intervals is the standard case for MEA experiments, but should be considered when testing out your experimental settings.



Repeated bipolar biphasic voltage stimulation on electrodes 74 and 75.

The figure illustrates a stimulation with biphasic pulses at a 50 ms interval: 200 mV stimulus amplitude, 60 μs duration, 3D-MEA filled with PBS, silver pellet as bath electrode. The non-stimulating electrodes show an excellent behavior without stimulus artifacts.



Single shot bipolar biphasic voltage stimulation on electrodes 74 and 75.

The figure illustrates a stimulation with single biphasic pulses. The experimental settings are identical to the preceding figure. The stimulus artifact performance is not as good as with the repeated stimulation.

### Behavior of stimulation electrodes

Due to the high electric charge that is applied by the stimulus and the general slow kinetics of electrode discharge, the recording from **stimulation** electrodes is usually **not possible shortly after** stimulation. You might also observe a **minor crosstalk** on the **two channels** that have **neighbor tracks** to the stimulation electrodes on the MEA. This is considered a normal behavior of the amplifier.

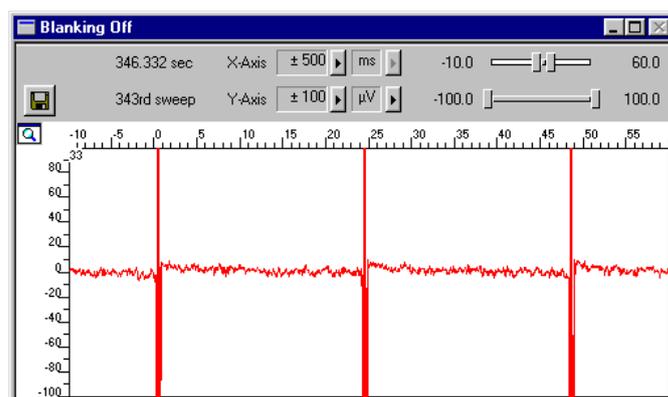
The switch artifacts of the internal switches between stimulus input and stimulation electrode of the MEA1060-BC-PA also contribute to the artifacts on the stimulation channels. The switch artifacts are generally not important for the performance of the stimulus artifact suppression, because the charge applied by the stimulus is much higher, but the information in this paragraph might be useful for you as a background information. The switches generate artifacts that are low but still higher than the noise level. As the MEA type and the bath affect the kinetics of the electrode discharge following the switch artifact, the amplitude and duration of the switch artifact depend on the experimental set up. The switch between stimulus input and stimulation electrode is moved only when the TTL blanking pulse is connected and active, and only on stimulation channels (selected in MEA\_Select). Therefore, you will see a switch artifact on the stimulation channels during the TTL pulse even if you do not send a stimulus pulse to the stimulation electrodes at all.

## 5.7 Examples with Different MEA Types

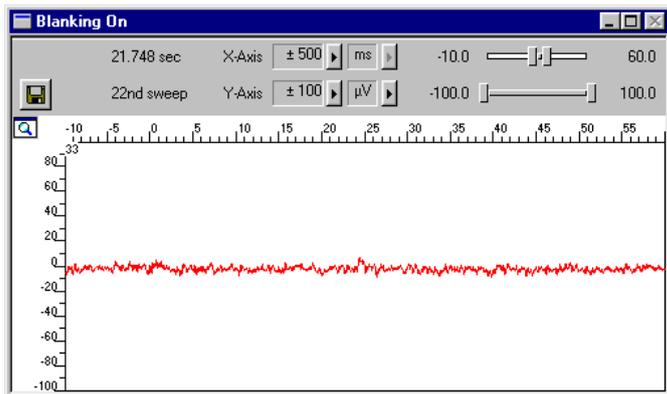
In the following, you will find typical examples for stimulus artifact suppression with standard MEA and electrode types under standard experimental conditions: Monopolar voltage driven stimulation, **1 V** (monophasic), **100  $\mu$ s** duration at a **24 ms interval**, **Wait 0**, MEA filled with PBS, silver pellet or internal reference electrode as bath electrode. Under these conditions, the stimulus artifact suppression performance showed no difference whether the internal reference electrode or a silver pellet was used (except for EcoMEAs, where the performance was sometimes much better with the silver pellet). The data acquisition was performed with MC\_Rack, at a sampling rate of 25 kHz.

These examples are intended to give you an impression of the results that you can expect with the MEA1060-BC amplifier, but please note that the results may vary depending on your experimental settings, please see also the recommendations under "Stimulus Artifact Suppression (Blanking)". Please see also the step-by-step tutorial on Stimulation and Recording under "First Tests and Tutorial".

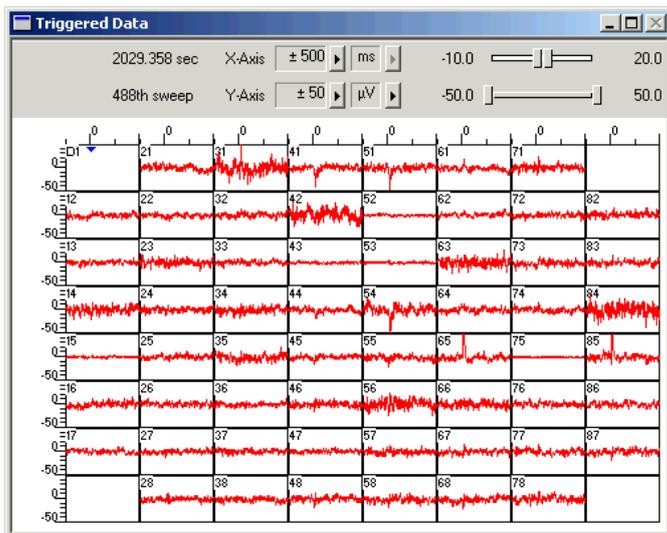
### (Used) 60MEA200/30iR-Ti



Stimulus artifacts with switched off blanking on a typical non-stimulating 30  $\mu\text{m}$  electrode.



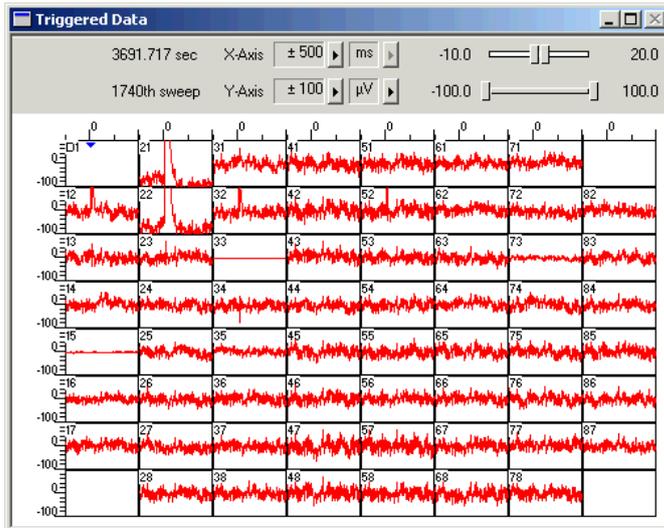
The same electrode and pulse protocol, but with blanking.



Voltage driven stimulation (1 V, electrode 75) on 30  $\mu\text{m}$  TiN electrodes.

A used 60MEA200/30iR-Ti with some defective electrodes was used for this experiment. Defective electrodes 43, 52, 53 were switched to ground in MEA\_Select. You see only small artifacts on the channels 65 and 85 with tracks next to the stimulating electrode 75. Pin 15 of the 60MEA200/30iR-Ti was connected to ground. You see a flat line on the stimulating electrode 75, because the electrical state of the electrode after the stimulation results in a saturation of the amplifier.

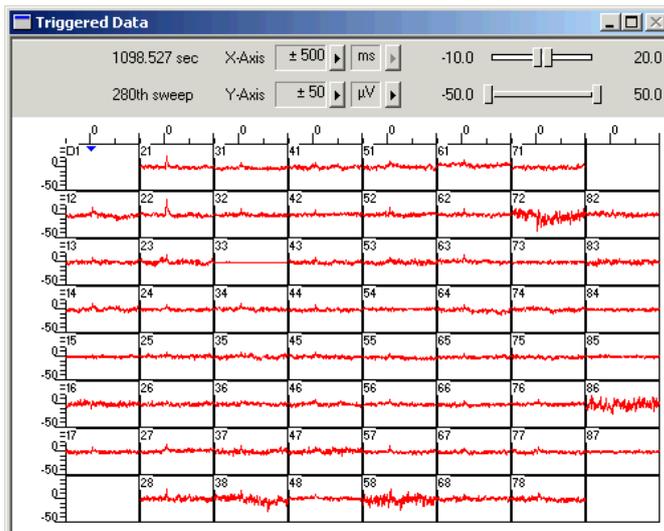
### 60MEA200/10iR-Ti



Voltage driven stimulation (1 V, electrode 33) on 10  $\mu\text{m}$  TiN electrodes.

The stimulus artifact suppression is slightly impaired by the higher impedance of the smaller electrodes, but still very good. You see only small artifacts on the channels 21 and 22 with tracks next to the stimulation electrode 33. Pin 15 of the 60MEA200/10iR-Ti was connected to ground.

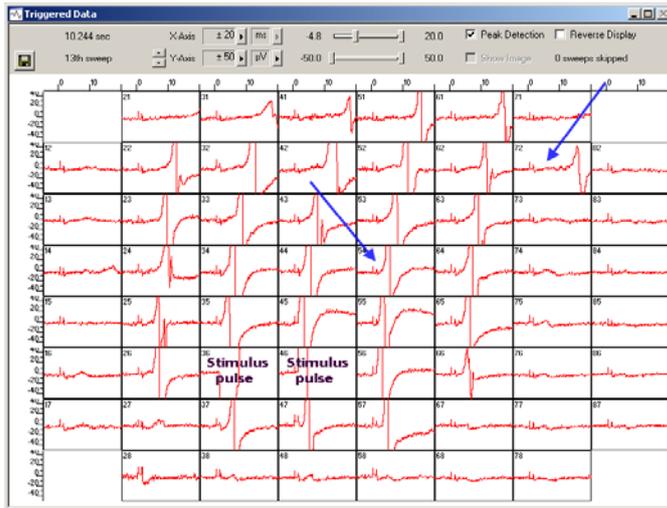
### 60EcoMEA



Voltage driven stimulation (1 V, electrode 33) on 100  $\mu\text{m}$  gold electrodes.

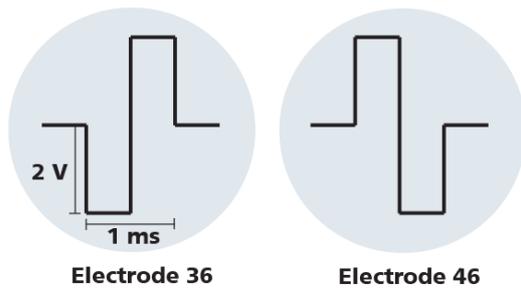
You see only small artifacts on the channels 21 and 22 with tracks next to the stimulation electrode 33. The blanking performance when using the silver pellet as a ground was reproducible. However, with the internal reference electrode, there could sometimes be observed an artifact with very slow kinetics. If you observe such problems, please try out a silver pellet for grounding the bath.

Standard 60MEA200/30iR-Ti



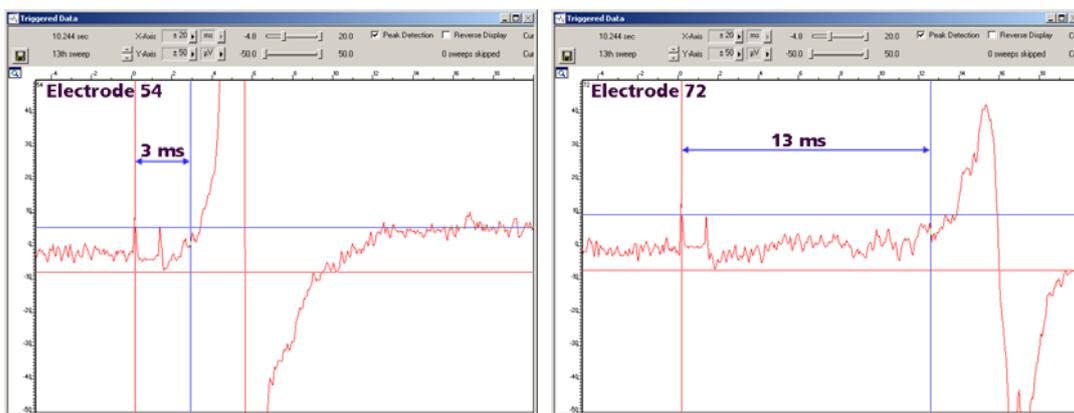
This example shows data from a syncytium of cardiac cells treated with a biphasic 2 V bipolar stimulus pulse field applied to electrodes No 36 and No 46. Please see the schema below.

Stimulation pattern



The blanking artefact is in the range of 10  $\mu\text{V}$  that is a negligible value in comparison to the amplitude of the cardiac field potentials ranging from a few hundred  $\mu\text{V}$  to several mV. You see the stimulation artefacts on all electrode channels.

The response of the cells to the stimulation is instantaneous in the cells near to the pulse. The cells recorded in the periphery of the syncytium need about 3 to 13 ms for their response to the stimulus pulse. This is the propagation time during which the electrical pulse is spreading in the cell syncytium.



## 5.8 Service and Maintenance

You should clean the contact pins of the amplifier with a soft tissue and pure (100 %) alcohol from time to time, especially if you have problems with the noise level.

It may be necessary after a longer time of operation to replace the contact pins. Replace gold connectors each year or if there are problems with the contacts. Please see the **Spare Parts** list under "Ordering Information".

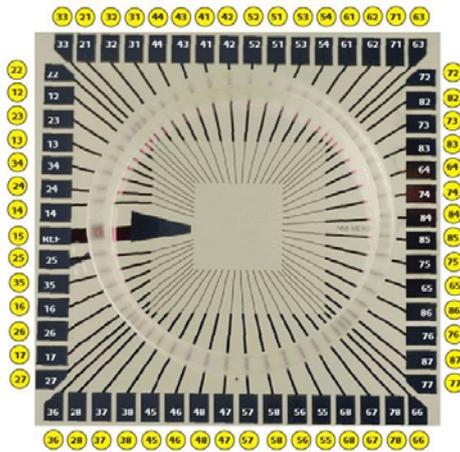


**Warning:** Please be very careful when handling the amplifier, cleaning the device, or replacing contact pins. The pins can be easily damaged. Do not damage the shafts of the pins, as they have to be replaced by the manufacturer.

### Replacing the gold spring contact pins

Define the pins which need to be changed via MC\_Rack software. Count the gold spring contacts in the lid of the amplifier to find the pin which should be changed.

Please use the pin layout of a standard 60MEA200/30iR-Ti for the MEA1060 amplifier. Please read also the respective data sheet of the MEA.



1. Disconnect the amplifier.
2. Open the housing and place the cover upside down on a soft and flat pad or cloth.
3. Grip the defective contact pin firmly with a pair of tweezers and carefully lift it up. Make sure that you pull only in a vertical direction, not sideways, as this may damage the hollow shaft of the pin in the housing.
4. Insert the new contact pin loosely into the shaft.
5. Grip the pin very firmly at its thinner part below the head, just above the thicker part, and push it carefully into the shaft until it snaps in. Again, make sure that you push only in a vertical direction. Do not push the pin too far into the shaft.

#### Replacing MEA1060-INV pins



## 6 Troubleshooting

### 6.1 About Troubleshooting

The following hints are provided to solve special problems that have been reported by users. Most problems occur seldom and only under specific circumstances. Please check the mentioned possible causes carefully when you have any trouble with the product. In most cases, it is only a minor problem that can be easily avoided or solved.

If the problem persists, please contact your local retailer. The highly qualified staff will be glad to help you. Please inform your local retailer as well, if other problems that are not mentioned in this documentation occur, even if you have solved the problem on your own. This helps other users, and it helps MCS to optimize the instrument and the documentation.

Please pay attention to the safety and service information (chapter "Important Safety Advice" and "Service and Maintenance" in the Manual / Help). Multi Channel Systems has put all effort into making the product fully stable and reliable, but like all high-performance products, it has to be handled with care.

### 6.2 Error Messages

MEA\_Select error messages are displayed in the box below the port settings. They show up if the computer fails to connect the MEA amplifier.



*Possible causes:*

- ? The wrong serial port is selected in the MEA\_Select program. The MEA amplifier is connected to another port.
  - Check to which COM port the MEA amplifier is currently connected and select the appropriate port in the program.
- ? The MEA amplifier is not connected properly to the computer.
  - Check the cables and all connections. Make sure all connectors fit tightly.

### 6.3 Noise on Single Electrodes

The noise level on single electrodes is significantly higher than +/- 8  $\mu$ V.

*Possible causes:*

- ? The electrode or the contact pin of the amplifier may be defective. To test this, do the following.
  1. Open the amplifier and turn the MEA by 90 degrees.
  2. Close the amplifier again and start the recording. If the same electrode in the MEA layout is affected, the amplifier's contact is not ok. If another electrode is now affected and the previously affected electrode is ok now, the MEA electrode is not ok, but the amplifier is fine.

- OR -

Use the test model probe to test the amplifier.

### MEA electrode is defective

See also the MEA manual.

*Possible causes:*

- ? The contact pads are contaminated.
- Clean the contact pads carefully with a swab and pure (100 %) alcohol.
- ? The contact pads or the electrodes are damaged.
- Use a new MEA. Try to handle and clean the MEA more carefully next time.

### Contact pin is defective

*Possible causes:*

- ? The contact pins are contaminated.
- Clean the contact pins carefully with a smooth and clean tissue and pure (100 %) alcohol.
- ? The contact pins are damaged.
- Replace the contact pins carefully. Please see the Spare Parts list under "Ordering Information". Try to handle and clean the contact pins more carefully next time.

## 6.4 Unsteady Baseline

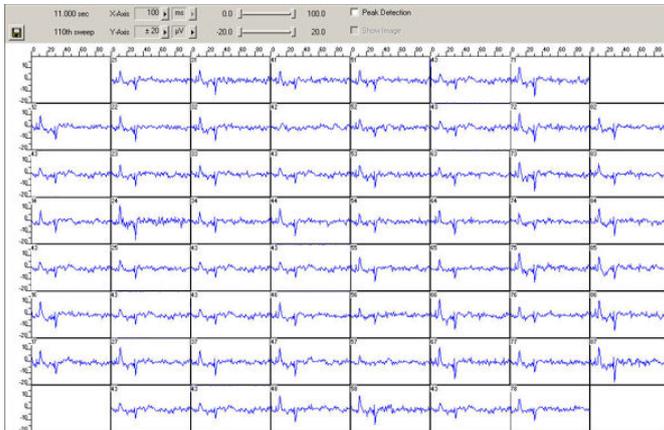
The baseline is unstable, signals are jumping or drifting.

*Possible causes:*

- ? Bath electrode is not connected to ground.
- Connect the internal or external bath electrode to one of the ground inputs of the amplifier as described in "Mounting the MEA Probe".
- ? AgCl bath electrode needs is not well-chlorided.
- Rechloride the electrode or use a new one.
- ? The MEA has an internal reference electrode and pin 15 is connected to ground appropriately, but the orientation of the MEA inside the amplifier is wrong. In this case, the wrong (standard size for recording) electrode is connected to ground, but not the reference electrode.
- Open the amplifier and check the orientation of the MEA and the reference electrodes described in "Mounting the MEA Probe".

## 6.5 Artifacts on All Channels

You see strange artifacts on all channels. This behavior can be caused by an insufficient supply power. Please see the Technical Specifications section. If the voltage drops beyond a critical level, the amplifier cannot operate properly, resulting in artifacts or in a saturation of the amplifier.

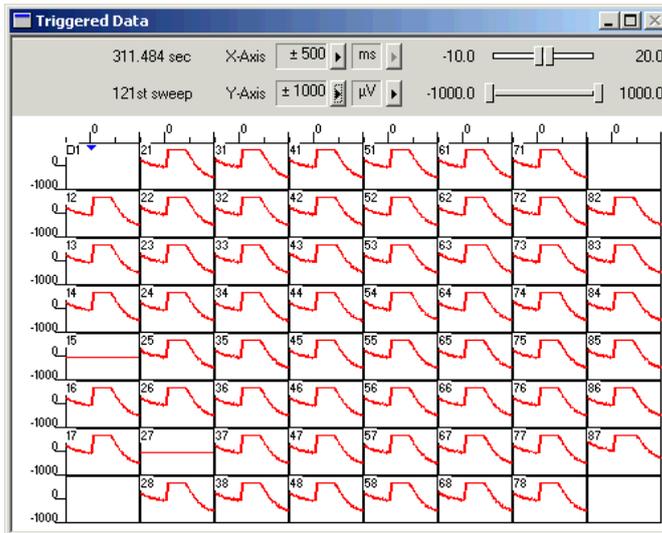
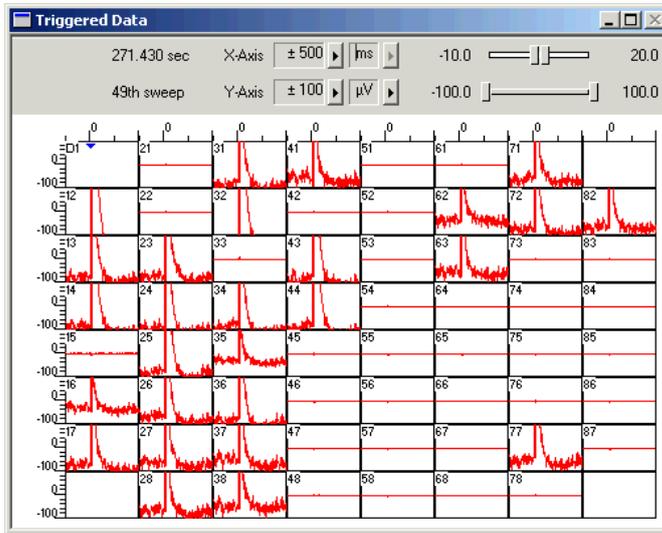


*Possible causes:*

- ? The cable connecting the amplifier to the power supply, that is, the MCS high grade cable leading from the amplifier to the data acquisition computer if you have a MEA-System, is too long. The applied voltage is not high enough for operating the amplifier.
  - Use a shorter cable, if possible, or try another power supply with a higher output power. MCS high grade cables with a total length of up to 3 m and the isolated power supply IPS10W are recommended for the MEA-System.
- ? You have connected too many amplifiers to your power supply. The isolated power supply IPS10W that is integrated in your data acquisition computer if you use a MEA-System can supply power for up to two MEA amplifiers with blanking circuit.
  - Use an additional power supply for operating the amplifiers or reduce the number of amplifiers.
- ? The supply power is too low for operating the amplifier. This is especially likely if you have upgraded your MEA-System from the standard amplifier to the MEA amplifier with blanking circuit or if you use a custom power supply. This generation of amplifiers needs an upgraded version of the isolated power supply IPS10W.
  - Contact your local retailer, describe the problem and your hardware configuration, and ask for a power supply that is suitable for your amplifier.

## 6.6 Unpredictable Noise and Artifacts

You use the amplifier for stimulation and you see unpredictable noise and / or artifacts on single or all channels.



Current driven stimulation (100  $\mu$ A, electrode 33) on 10  $\mu$ m TiN electrodes.

Due to the higher electrode impedance, the maximum input voltage of the MEA1060-BC amplifier is exceeded when applying a 100  $\mu$ A pulse, and a proper operation is not possible.

*Possible causes:*

- ? You use voltage stimuli with a higher amplitude than **+/- 5 V**.
- Multi Channel Systems recommends **not** to exceed the **maximum recommended stimulus input range** of **+/- 5 V** (see chapter Technical Specifications in the Appendix). Please adjust the amplitude of the stimuli used.
- ? You use current driven stimulation, and you have started the stimulation **before** selecting a stimulating electrode in MEA\_Select. In this case, the stimulus generator sends the current into the open circuit of the amplifier. This is not an allowed operation.
- Always start the stimulus generator **after** having selected a stimulating electrode in MEA\_Select. Make sure you have selected the appropriate stimulus input and **downloaded** the information onto the amplifier.
- ? You use current stimuli and electrodes with a smaller diameter than 30  $\mu\text{m}$ , or older electrodes (with a higher impedance).
- Multi Channel Systems recommends **not** to exceed the **maximum recommended stimulus input range** of **+/- 5 V** (see chapter Technical Specifications under Appendix). To avoid this, the stimulation with current is generally not recommended, especially not with electrodes that have a high impedance. The impedance of electrodes with a lower diameter (or the impedance of older deteriorated electrodes) is considerably higher, resulting in a high voltage exceeding the input range. MCS recommends to **use voltage stimuli with an amplitude in the recommended stimulus input range** to be on the safe side. For more information on stimulation via MEA electrodes, see also the MEA manual.

## 6.7 Liquid Spilled onto Amplifier



**Warning:** Spilled liquid can damage or even completely destroy the electronics of the MEA amplifier. Avoid it by all means.

Of course it is best to completely avoid this situation and consider the following instructions as an emergency measurement" only. Even with this measurement, a severe damage to the instrument cannot be excluded. It is very important that the amplifier is **not** connected or used again before repair to avoid further damage caused by corrosion. Even if the initial damage is negligible, the additional damage inflicted by corrosion over a longer period of time can completely destroy the electronics.

1. **Immediately** remove the amplifier from the power supply, that is, unplug the amplifier. Do not connect or use it again before repair.
2. Contact your local retailer and send the amplifier to Multi Channel Systems for cleaning and repair. Multi Channel Systems will check the device and prepare a quote for you, depending on the work and material costs needed for repair.

## 6.8 Technical Support

Please read the Troubleshooting part of the Manual / Help first. Most problems are caused by minor handling errors. Contact your local retailer immediately if the cause of trouble remains unclear. Please understand that information on your hardware and software configuration is necessary to analyze and finally solve the problem you encounter.

**Important:** If you need to send the amplifier back to the manufacturer for repair, use only the original case for packaging to avoid further damage inflicted during the shipment.

### Please keep information on the following at hand

- **Description** of the **error** (the error message text or any other useful information) and of the context in which the error occurred. Try to remember all steps you had performed immediately before the error occurred. The more information on the actual situation you can provide, the easier it is to track the problem.
- The **serial number** of the device. You will find it on the device.
- Description of the **data acquisition system**.
- The **operating system** and service pack number on the connected **computer**.
- The **hardware configuration** (Isolated Power Supply IPS10W, microprocessor, frequency, main memory, hard disk) of the connected **computer**. This information is especially important if you have modified the computer or installed new hard- or software recently.
- Other connected hardware.

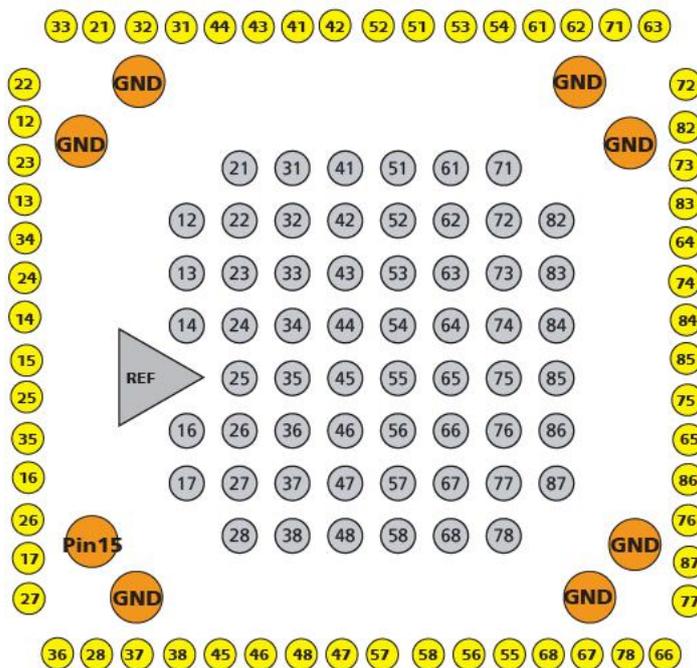
# 7 Appendix

## 7.1 Technical Specifications

### 7.1.1 Pin and MEA Layout

The layout of standard MEA electrodes follows the scheme of a standard grid: The first digit is the column number, and the second digit is the row number. For example, electrode 23 is positioned in the third row of the second column. The numbering follows the standard order from left to right, and from top to bottom.

MEAs are not symmetrical, please pay attention to the recommended orientation of the MEA chip inside the amplifier: The internal reference electrode should point to the left, the writing NMI should be on the right side. The connectors of the amplifier are in the back.



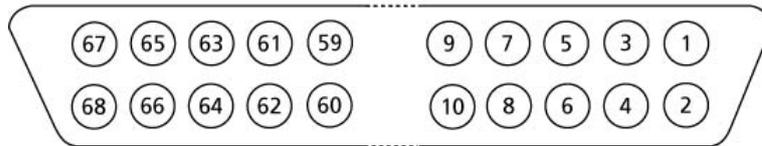
The following illustration shows the standard pin layout of the socket on the amplifier. The list below shows the assignment of the electrodes to the pins of the socket. Pin 1 is the ground of the power supply, pin 2 is the ground of the signal, that is, where the signal is referred to. Both pins are already **internally connected** inside the amplifier.

If you are planning to build a custom setup with a home-made filter amplifier, please be careful with the input of the filter amplifier. Since the MEA1060-BC is a quasi DC-coupled preamplifier, the input into the filter amplifier has to be AC-coupled, that is, there has to be a capacitor at the input, before the signal is led into the operational amplifier. Otherwise, you will have problems with the offset of the metal electrodes, and your filter amplifier will go into saturation.

Channels A1 to A4 (Pin 63 to 66) are the four additional analog inputs of the MCS Standard Connector that are not used by the MEA amplifier. You can access channels A1 to A3 via the BNC sockets labeled Analog IN on the rear side of the data acquisition computer of the MEA-System (see also MEA-System manual).

## MEA Amplifier with Blanking Circuit for Inverse Microscopes

68-Pin MCS standard socket



- Pin 1: GNDP (power ground)
- Pin 2: GNDS (signal ground)
- Pin 3-62: Electrode channels, see separate list
- Pin 63-66: Channel A1-A4
- Pin 67: Positive voltage supply
- Pin 68: Negative voltage supply

Assignment pin to electrode			
68-pin socket pin	MEA electrode	68-pin socket pin	MEA electrode
3	47	33	52
4	48	34	51
5	46	35	53
6	45	36	54
7	38	37	61
8	37	38	62
9	28	39	71
10	36	40	63
11	27	41	72
12	17	42	82
13	26	43	73
14	16	44	83
15	35	45	64
16	25	46	74
17	15	47	84
18	14	48	85
19	24	49	75
20	34	50	65
21	13	51	86
22	23	52	76
23	12	53	87
24	22	54	77
25	33	55	66
26	21	56	78
27	32	57	67
28	31	58	68
29	44	59	55
30	43	60	56
31	41	61	58
32	42	62	57

## 7.1.2 MEA1060-Inv-BC

Operating temperature	10 °C to 50 °C
Storage temperature	0 °C to 50 °C
Dimensions (W x D x H)	165 mm x 165 mm x 20 mm (29 mm with sockets)
Weight	800 g
Supply voltage (external power supply)	± 6.5 VDC to ± 9 VDC
Supply current	± 350 mA
Number of stimulus input channels	2
Maximum recommended stimulus input voltage	± 5 V
Number of analog input channels	60
Input voltage	-4 mV to 4 mV
Input impedance	10 <sup>11</sup> Ω parallel to 10 pF
Input noise	< 800 nV <sub>RMS</sub>
Noise density	$e_n = 15nV / \sqrt{Hz}$
Number of analog output channels	60
Output voltage	± 5 V
Output current	10 mA
Output impedance	300 Ω
Bandwidth	0.02 Hz to 8500 Hz (Standard total bandwidth MEA1060-Inv-BC and standard FA60S-BC = 1 Hz to 3000 Hz)
Gain	53 (Standard total gain MEA1060-Inv-BC and standard FA60S-BC = 1100)
<b>Microscope compatibility</b>	
	<b>Inverse microscopes</b>
Distance focal plane to microscope table	3.5 mm

**7.1.3 FA60S-BC**

Operating temperature	10 °C to 50 °C
Storage temperature	0 °C to 50 °C
Dimensions (W x D x H)	92 mm x 145 mm x 26 mm
Weight	450 g
Supply voltage (external power supply)	± 6.5 V to ± 9 V DC
Supply current	± 200 mA
Number of analog input channels	60
Input voltage	500 mV (with respect to 5 V supply voltage)
Input impedance	> 10 <sup>9</sup> Ω parallel to 10 pF
Input noise	< 1 μV RMS (full bandwidth, inputs short-circuited)
Noise density	$e_n = 9nV / \sqrt{Hz}$
Number of analog output channels	60
Output voltage	± 5 V
Output current	10 mA
Output impedance	300 Ω
Bandwidth	8 Hz to 3000 Hz (standard), 1 Hz to 5000 Hz (custom)
Gain	20 (standard), 10 to 100 (custom) (Standard total gain MEA1060-1BC and standard FA60S-BC = 1100)

## 7.2 Contact Information

### Local retailer

Please see the list of official MCS distributors on the MCS web site.

### User forum

The **Multi Channel Systems User Forum** provides the opportunity for you to exchange your experience or thoughts with other users worldwide.

### Mailing list

If you have subscribed to the Mailing List, you will be automatically informed about new software releases, upcoming events, and other news on the product line. You can subscribe to the list on the contact form of the MCS web site.

**[www.multichannelsystems.com](http://www.multichannelsystems.com)**

## 7.3 Ordering Information

Product information is subject to change without notice. Please contact your local retailer for pricing and ordering information.

### MEA Amplifiers

Product	Product Number	Description
MEA amplifier for inverted microscopes	MEA1060-Inv	Probe interface and 60 channel pre-and filter amplifier with custom gain and bandwidth.
MEA amplifier for upright microscopes	MEA1060-Up	
MEA amplifier with blanking circuit for inverted microscopes	MEA1060-Inv-BC	Probe interface and 60 channel pre-and filter amplifier with custom gain and bandwidth. The blanking circuit prevents the amplifier from getting saturated and thus prevents stimulus artifacts.
MEA amplifier with blanking circuit for upright microscopes	MEA1060-Up-BC	

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