MED64 System

(MED64-Basic/Quad II/Allegro)

Manual

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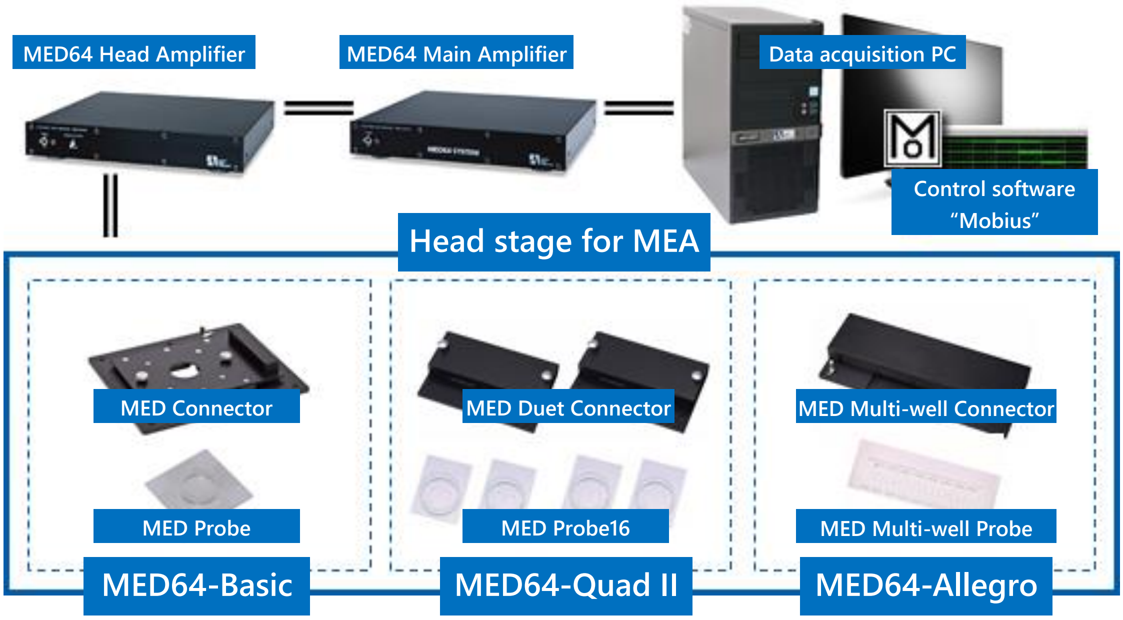
1. Introduction

- The MED64 System is a micro electrode array (MEA) system commercialized for the first time in the world in 1997. Since then, the MEA system has been widely used in the United States and the EU for basic research in the fields of the central nervous system and the cardiovascular system. Nowadays, its application to screening for drug discovery is rapidly spreading, thanks to its highly evaluated excellent operability and efficiency.
- The system use a MEA with 64 planar electrodes arrayed in a pattern (MED probe) on a glass substrate. Users can acquire extracellular potential simply by placing a tissue slice or by culturing cells on these electrodes. Current stimulation can be applied through any electrode selected by software. Unlike a glass capillary electrode, the MED64 system requires no troublesome operations to measure spontaneous activity or evoked response and no special training for electrophysiological experiment is necessary. The system allows for accurate and easy measurements even by unexperienced researchers.
- The electrodes are made of platinum black or carbon nanotube with impedance 7 to 10 kΩ, the lowest impedance among similar products on the market. The system is substantially free from external noise, thanks to its low impedance electrodes. It needs no special environment/facility for electrophysiological experiment, such as a shield box, and it can be placed on a laboratory desk for stable, daily measurement with no need for noise elimination.

2. Configuration of the MED64 System

The MED64 System consists of the following units. The number of biological specimens from which simultaneous measurements can be made depends on the type of the Connector onto which the MED Probe is mounted.



The MED64 Head Amplifier has connection terminals dedicated for all types of Connector, which allows the user to switch the system easily by changing the type of Connector to be connected. The extracellular potentials detected by the recording electrodes of the MED Probe are amplified 10 times by the MED64 Head Amplifier and then further amplified (varied) by the MED64 Main Amplifier before analog to digital conversion. The obtained digital signals are sent through the USB cable to the data acquisition PC system, where data recording and analysis are performed online using the MED64 Mobius, which is the dedicated MED64 System control software. (Recorded data can also be analyzed offline.)

2.1. Part names and functions

2.1.1. MED64 Main Amplifier

【Front】

❶POWER ..... To switch ON/OFF of the power.

【Back】

❶INPUT ..... An analog input terminal to input signals from the MED64 Head Amplifier. It is connected to the OUTPUT of the MED64 Head Amplifier via the 68-pin SCSI cable.

❷OUTPUT ..... An analog output terminal to connect an external device (MED Feedback Stimulator etc.)

❸DIO 1、2、3 ..... Digital input/output terminal to connect MED64 Multiplexer.

❹F1 STIMULUS OUTPUT ..... An output terminal of stimulus pattern signal from F1 stimulator. It is connected to F1 STIMULUS INPUT of the MED64 Head Amplifier via the BNC cable.

❺F2 STIMULUS OUTPUT ..... An output terminal of stimulus pattern signal from F2 stimulator. It is connected to F2 STIMULUS INPUT of the MED64 Head Amplifier via the BNC cable.

❻CONTROL OUTPUT ..... An output terminal of stimulation channel signal. It is connected to CONTROL INPUT of the MED64 Head Amplifier via the round-pin cable.

❼SIGNAL GND ..... A ground terminal to connect a lead for grounding.

❽USB ..... It is connected to a USB terminal of the data acquisition PC via the USB cable.

❾DC INPUT ..... To connect to a power adaptor cord.

2.1.2. MED64 Head Amplifier

【Front】

❶Power ..... To switch ON/OFF of the power.

❷STIMULUS CURRENT ..... The output current is doubled by moving the lever toward x2.

【Back】

❶INPUT 1-16CH、17-32CH、33-48CH、49-64CH

..... Analog input terminals to input signal from the MED Duet Connectors or the MED Multi-well Connector. Those are connected to the output terminals of the either via the 20-pin SCSI cable. Keep ❷INPUT opened (unconnected) when connecting the cables (Do not connect at the same time.)

❷INPUT ..... An analog input terminal to input signal from the MED Connector. It is connected to the output terminal of the MED Connector via the 68-pin SCSI cable. Keep the all ❶INPUT opened (unconnected) when connecting the cables (Do not connect at the same time.)

❸OUTPUT ..... An analog output terminal to connect the INPUT terminal of the MED64 Main Amplifier via the 68-pin SCSI cable.

❹F1 STIMULUS INPUT ..... An input terminal of stimulus pattern signal to connect the F1 STIMULUS OUTPUT of the MED64 Main Amplifier via the BNC cable.

❺F2 STIMULUS INPUT ..... An input terminal of stimulus pattern signal to connect the F2 STIMULUS OUTPUT of the MED64 Main Amplifier via the BNC cable.

❻CONTROL INPUT ..... An input terminal of stimulation channel signal. It is connected to CONTROL OUTPUT of the MED64 Main Amplifier via the round-pin cable.

❼SIGNAL GND ..... A ground terminal to connect a lead for grounding.

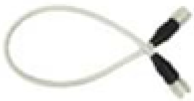
❽DC INPUT ..... To connect to a power adaptor cord.

2.1.3. Accessories of the MED64 Main Amplifier


❶Power adaptor

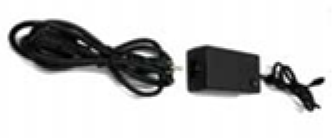

❷68-pin SCSI cable (50cm)


❸BNC cable


❹Round-pin cable


❺USB cable

2.1.4. Accessories of the MED64 Head Amplifier


❶Power adaptor

3. Setup of the MED64 System - procedure common to Basic, Quad II, and Allegro

Thanks to the excellent electrode impedance (impedance of 50 μm electrode is 10 kΩ in frequency range 1 kHz) of the MED Probe, which is the sensor of the MED64 System, the MED64 System offers several technological advantages:

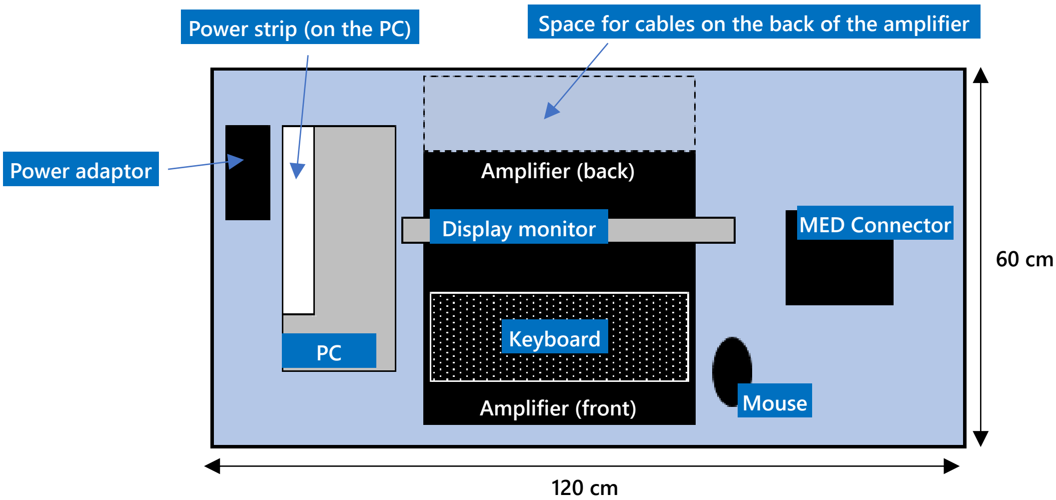
1) Less influence by external noise (hum noise, etc.)
2) Very low Johnson noise (baseline noise) at about several μV
3) The MED Probe/Connector can be installed at a place physically distant from the amplifier (e.g., inside of an incubator with 100% humidity) via a 2 m connection cable, without being affected by noise or attenuation of the signal acquired.

A faraday cage or vibration isolation table that is typically necessary for an electrophysiological experiment is not necessary for the MED64 System, but the MED64 System should be set up on a stable table without vibrations, such as a laboratory desk. A desk of about 100 cm width and 75 cm depth is necessary. For the user’s convenience, a larger desk is preferable. The power adaptor connected to the amplifier, etc., serves as a magnetic field-derived noise source. When the MED64 System is set up on a desk against a wall, make space for the cord between the wall and the desk and place the adaptor on the floor in order to place the power adaptor at a distance from the amplifier and other components (the desk must not be fixed to the wall). Alternatively, a space is necessary to ensure sufficient distance between the MED64 System and the power adaptor.

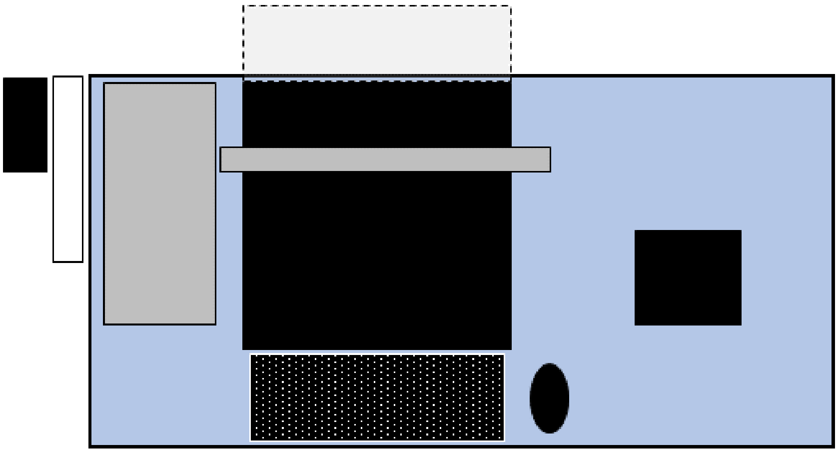
3.1. Considerations for the position of devices

Considering the position of the SCSI cable that connects the Input Terminals of the MED64 Head Amplifier and the MED Connector, place the MED64 Main Amplifier on the MED64 Head Amplifier (not essential). Place the display monitor on the MED64 Main Amplifier with the PC and the power strip arranged on the right or left side and the MED Connector on the opposite side to provide sufficient space on the MED Connector side so that the user can perform an experiment easily on the MED Connector side. Do not place the power adaptor or other electronic devices close to the connection terminals on the back of the amplifiers.

- <u>Layout sample 1</u>



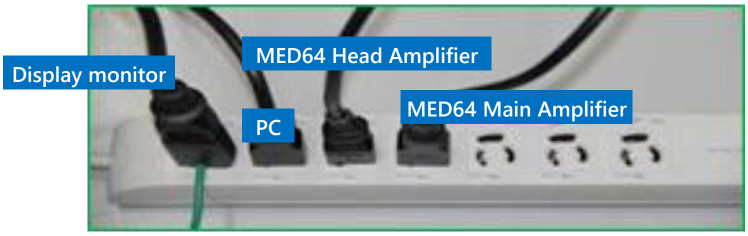
- <u>Layout sample 2</u>



Above is an image of the ideal positions of the components. The user can perform an experiment easily when there is a sufficient space next to or at the back of the laboratory desk with the power strip on the floor (as shown in Layout Sample 2). If the MED64-Quad II is to be used in combination with four perfusion systems, a larger space will be necessary.

3.2. Power source

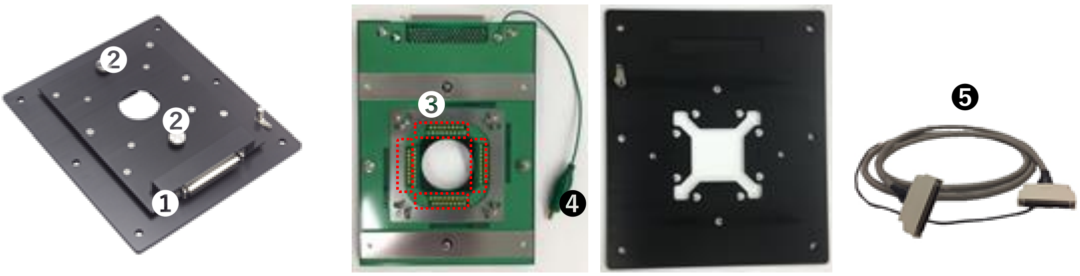
Connect all the 3-terminal power plug (MED64 Main Amplifier, MED64 Head Amplifier, data acquisition PC, etc.) to one power strip connected to a wall outlet with the ground terminal (do not use an outlet on the desktop rack because it is often not grounded properly). Do not connect devices that are not necessary for the MED64 System to the power strip or to the wall outlet to which the MED64 System and other components are connected. Place the power cable and the power adaptor at a distance from the amplifier, the MED Connector and the 68-pin SCSI cable.



3.3. Setup of the MED64-Basic

3.3.1. Part names and functions of the MED Connector

【Top unit】 【Base plate】 【Accessory】



❶Output terminal ….. To connect to INPUT of the MED64 Head Amplifier via the accessory, the 68-pin SCSI cable.

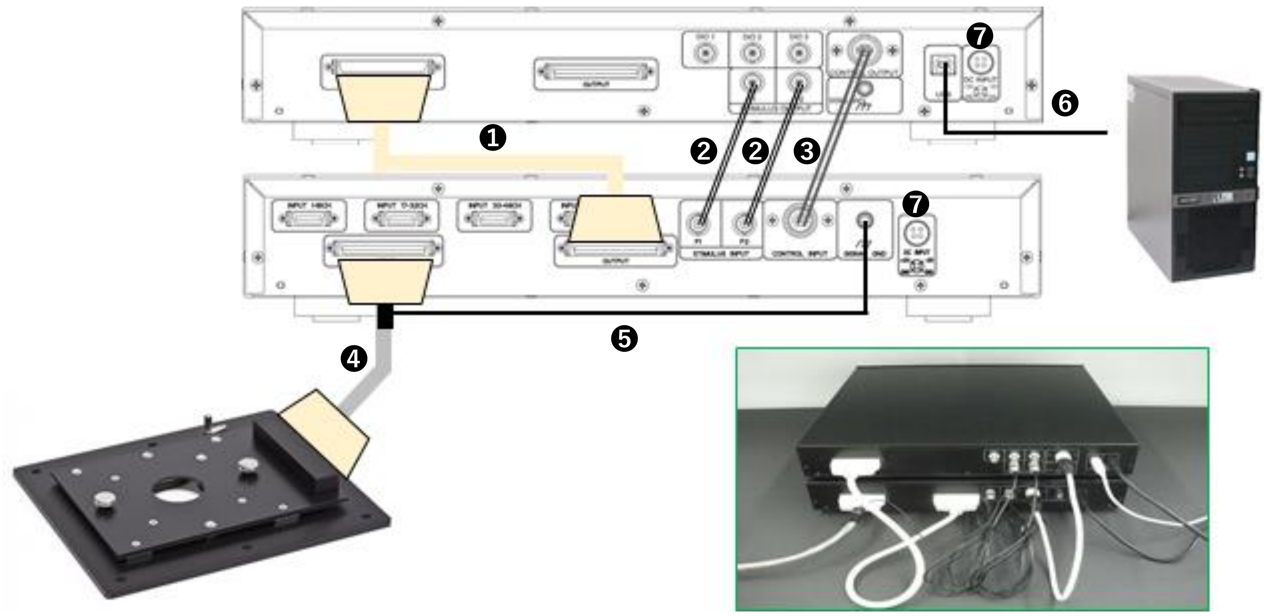
❷Fixation screw ….. To fix the fitted top unit and the base plate with a MED Probe attached.

❸Contact pin ….. To contact with the MED Probe terminal to read signals.

❹Ground wire with an bagworm clip ….. To connect to a platinum wire with a perfusion cap when a perfusion system is used.

❺68-pin SCSI cable (2 m) ….. A conductive cloth tape and ground wire are wound on the cable.

3.3.2. Connections among devices



❶Connect the INPUT terminal of the MED Main Amplifier and the OUTPUT terminal of the MED Head Amplifier, using the 68-pin SCSI cable (50 cm).

❷Connect the F1/F2 STIMULUS OUTPUT of the MED64 Main Amplifier and F1/F2 STIMULUS IMPUT of the MED64 Head Amplifier, using the BNC cables.

❸Connect the CONTROL OUTPUT terminal of the MED64 Main Amplifier and the INPUT terminal of the MED64 Head Amplifier, using the round-pin cable.

❹Connect the output terminal of the MED Connector and the INPUT terminal of the MED64 Head Amplifier, using 68-pin SCSI cable.

❺Connect the ground wire wound on the 68-pin SCSI cable to the SIGNAL GND terminal of the MED64 Head Amplifier.

❻Connect the USB port (type-B) of the MED64 Head Amplifier and the USB port (type-A) of the data acquisition PC (USB2.0 is preferable), using the USB cable

❼Connect the power adaptor cables to both amplifiers.

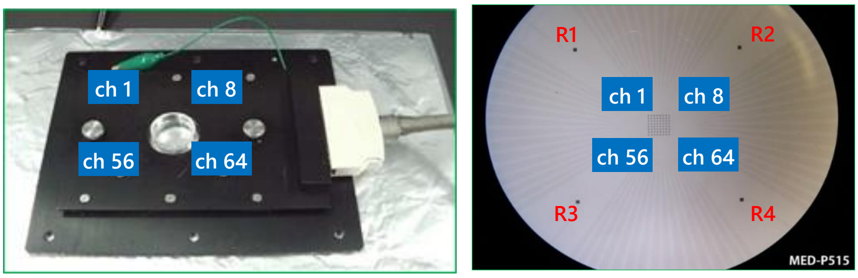
Note1: The 68-pin SCSI cable is wound with a conductive cloth tape and an uncoated lead is wound on it. If the other side of the lead is free, the conductive cloth tape may serve as an antenna that collects electric noise and may cause noise. Make sure that the other side of the lead is connected to the SIGNAL GND terminal of the MED64 Head Amplifier (to use the conductive cloth tape as an electric shield).

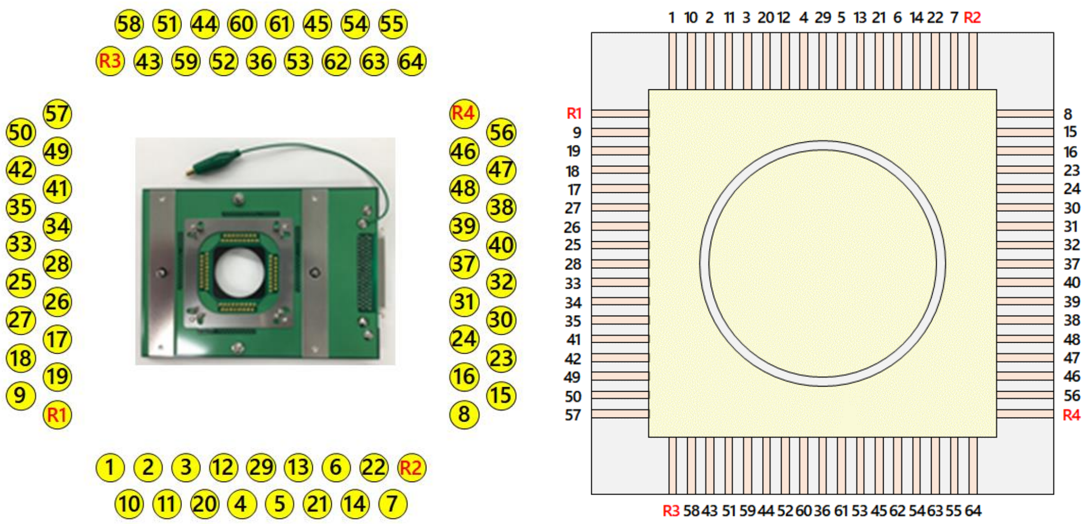
Note2: In order to prevent noise, use the SIGNAL GND terminal of the MED64 Head Amplifier for single-point grounding when the peripheral devices are grounded.

Loosen the screw of SIGNAL GND terminal to lead the ground wire into the hole (left), then thighten to fix it (right) 。

3.3.3. Positioning of the terminal of the MED Connector

Place the MED Connector with its output terminal and the 68-pin SCSI cable on the right side. In this position, the left top of the MED Probe is assigned to ch 1 and the right bottom is assigned to ch 64. The orientation of the MED Probe is important because the electrode number of the MED Probe is defined by the contact pins of the MED Connector.





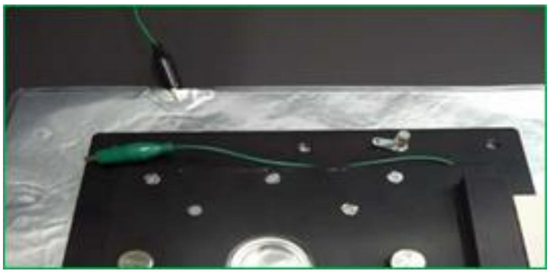
Correspondence between the contact pin of the MED Connector and the electrode number: correspondence from the contact surface side with the output terminal on the right (left panel) and correspondence with the MED probe terminal (right panel). R indicates the reference electrode.

29	5	21	6	22	7	50	49	41	35	33	28	26	27	18	19	8	15	23	24	31	32	40	39	48	47	56	64	63	54	45	53	36	60
4	13	12	14	20	57	3	42	11	34	2	25	10	17	1	9	R2	16	R4	30	R1	37	R3	38	58	46	43	55	51	62	59	61	44	52

Correspondence between the MED Connector output terminal and the electrode number. R indicates the reference electrode.

3.3.4. Electric shield with aluminum foil

If a noise check reveals that external electric noise is affecting the MED Connector, the noise may be avoided by placing a sheet of aluminum foil under the MED Connector and using a lead to connect the aluminum foil sheet and the SIGNAL GND terminal of the MED64 Head Amplifier for grounding.



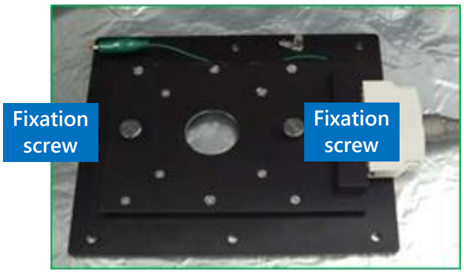
A sheet of aluminum foil serves as an electric shield by grounding. Magnetic field-derived noise cannot be prevented by an electric shield.

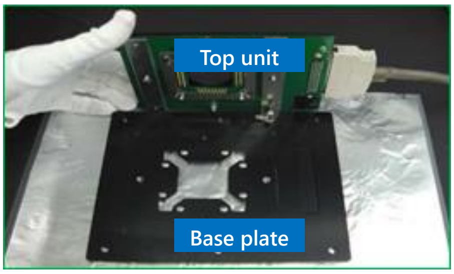
Note: Although the MED Connector is made from aluminum, its surface is coated and insulated from the aluminum foil. Make sure that the aluminum foil is grounded to the SIGNAL GND terminal of the MED64 Head Amplifier when a sheet of aluminum foil is placed under the MED Connector, even if the baseline noise level is within an acceptable range. Without grounding, the aluminum foil serves as an antenna to collect electric noise and may generate unexpected noise.

MED64-Basic after setup. In this example, the laptop PC is used for a data acquisition PC system.

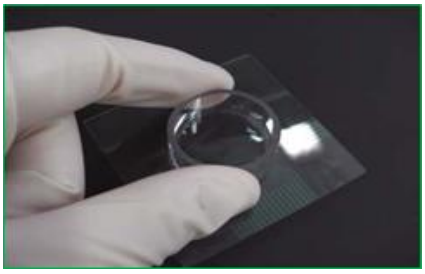
3.3.5. Preparation for the noise check – attaching the MED Probe

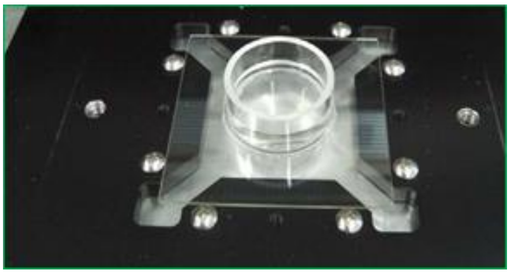
Loosen the fixation screw of the MED Connector to remove the top unit.





On the base plate, place the MED Probe filled with saline, such as aCSF or PBS holding the ring chamber of the MED probe or the end of the glass substrate. Pay attention so as not to touch the terminals. If any waterdrop, medium or smudge is on the terminal, wipe the terminal with a Kim wipe before setting the MED Probe. Subsequently, place the top unit on the MED Probe and fix with a fixation screw.





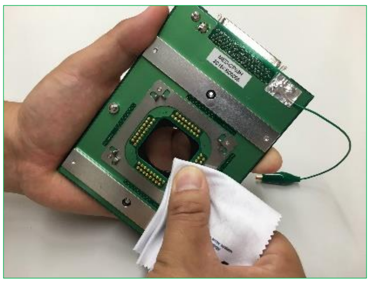
Note1: Loosen/tighten both screws little by little at the same time. When the screws are sufficiently loosened/tightened, they can be completely loosened/tightened one by one. If only one screw is completely tightened/loosened without tightening/loosening the other screw, the second screw cannot be inserted perpendicular to the hole.

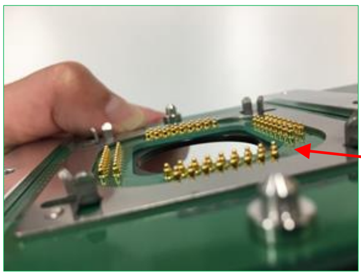
Note2: Do not touch the contact pin of the MED Connector with bare hands. If sebum of the hand attaches to the pin, contact impedance between the contact pin and the terminal of the MED Probe will increase and may cause noise. Pay attention so as not to spill liquid (saline, medium etc.) on the MED Probe after it is set. Prevent liquids from coming into contact with the contact pin.

About the details of noise check, refer to p.19 “Noise check by operating the Mobius - procedure common to Basic, Quad II, and Allegro”. Also, for the details of operating the Mobius refer to “MED64 Mobius Tutorial.” Refer to the relevant application note for the preparation of specimens used for the experiment.

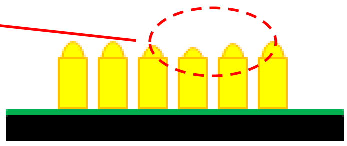
3.3.6. Maintenance of the MED Connector

A printed circuit board onto which 68 contact pins contacting the terminal of the MED probe are soldered is fixed by screws to the top unit of the MED Connector. Spring-loaded pins are used as contact pins and keeping the surface of the pins clean is important for the maintenance of the MED Connector. If any abnormal noise is found at a particular electrode and noise persists on that electrode even when the orientation of the MED Probe is changed, the noise may be caused by deformation (crushing) due to the wear of the contact pins associated with everyday use or dust or sebum on the contact pins. In the latter case, the noise may be reduced by wiping the contact pin by pushing the eyeglass cleaner cloth on the pin. Do not use ethanol because the contact pin is filled with lubricant for the spring. The printed circuit board needs to be replaced (paid repair) in the case of malfunction due to wear of the contact pin associated with everyday use or malfunction due to salt (perfusion solution or media) attached on the pin.



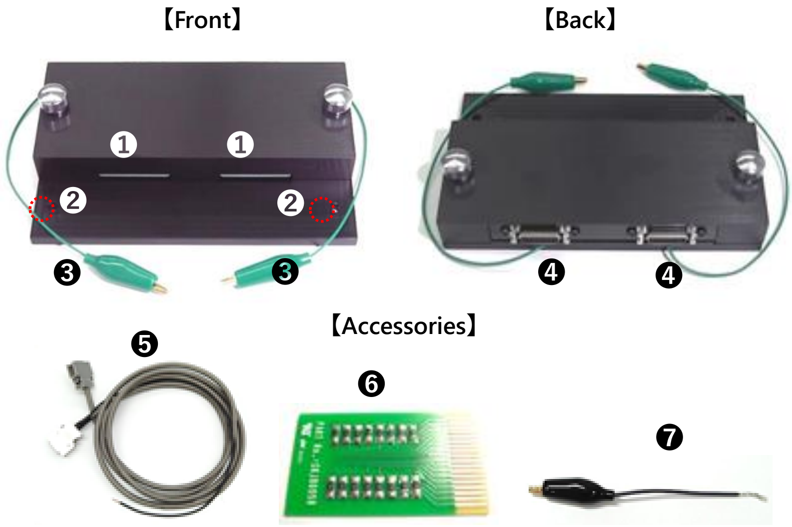


When the elasticity of the pin decreases, irreversible contact failure occurs.



3.4. Setup of the MED64-Quad II

3.4.1. Part names and functions of the MED Duet Connector



❶Insertion slot of the MED Probe16

❷Screw hole ..... To fix the MED Duet Connector onto the MED Thermo Base (MED-CPB02).

❸Ground wire with an bagworm clip ..... To connect to a platinum wire with a perfusion cap when a perfusion system is used.

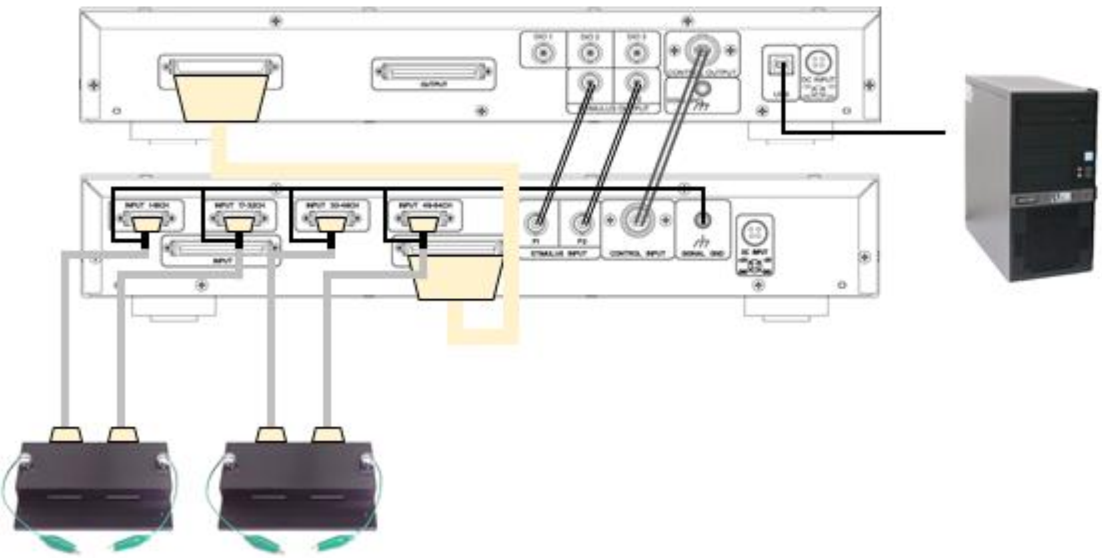
❹Output terminal ..... To connect to the INPUT terminal of the MED64 Head Amplifier via the 20-pin SCSI cable.

❺20-pin SCSI cable (2 m x 4) ..... A conductive cloth tape and ground wire are wound on the cable (the specification is different from the 20-pin SCSI cable for the MED Multi-well Connector).

❻Test board ..... A PCB with multiple resistors to check the baseline noise level of the whole system and its environment.

❼Ground wire with an bagworm clip ..... To hold four ground wires from four 20-pin SCSI cables together and connect them to the SIGNAL GND terminal of the MED64 Head Amplifier.

3.4.2. Connections among devices



❶Connect the INPUT terminal of the MED Main Amplifier and the OUTPUT terminal of the MED Head Amplifier, using the 68-pin SCSI cable (50 cm).

❷Connect the F1/F2 STIMULUS OUTPUT of the MED64 Main Amplifier and F1/F2 STIMULUS IMPUT of the MED64 Head Amplifier, using the BNC cables.

❸Connect the CONTROL OUTPUT terminal of the MED64 Main Amplifier and the INPUT terminal of the MED64 Head Amplifier, using the round-pin cable.

❹Connect the output terminals of the MED Duet Connectors and the INPUT terminals of the MED64 Head Amplifier, using 20-pin SCSI cable.

❺Connect the ground wire wound on the 20-pin SCSI cable to the SIGNAL GND terminal of the MED64 Head Amplifier.

❻Connect the USB port (type-B) of the MED64 Head Amplifier and the USB port (type-A) of the data acquisition PC (USB2.0 is preferable), using the USB cable

❼Connect the power adaptor cables to both amplifiers.

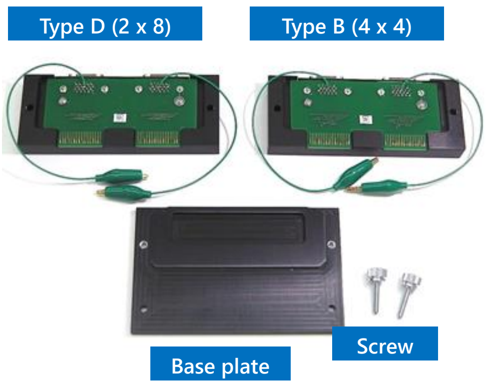
Note1: The 20-pin SCSI cable is wound with a conductive cloth tape and an uncoated lead is wound on it. If the other side of the lead is free, the conductive cloth tape may serve as an antenna that collects electric noise and may cause noise. Make sure that the other side of the lead is connected to the SIGNAL GND terminal of the MED64 Head Amplifier (to use the conductive cloth tape as an electric shield).

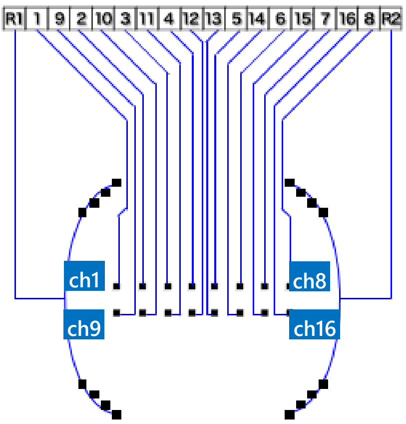
Loosen the screw of SIGNAL GND terminal to lead the ground wire into the hole (left), then tighten to fix it (right). Because it is difficult to put four ground wires through the hole of the SIGNAL GND terminal, use the ground wire with a bagworm clip to hold them together

Note 2: In order to prevent noise, use the SIGNAL GND terminal of the MED64 Head Amplifier for single-point grounding when the peripheral devices are grounded.

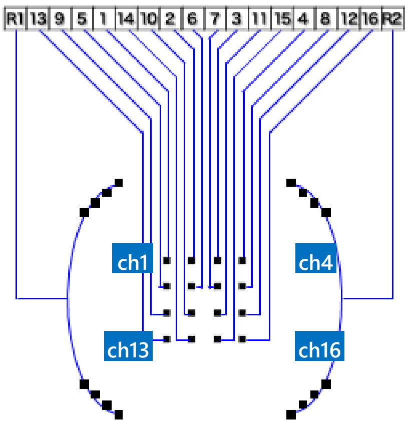
3.4.3. Positioning of the terminal of the MED Duet Connector

A circuit board is incorporated into the top unit of the MED Duet Connector of two types, with one type corresponding to the 2 x 8 array MED Mini Probe and the other to the 4 x 4 array MED Mini Probe. Replace the top unit with the one compatible with the MED Mini Probe to be used and place and secure it on the base plate with the fixation screws.

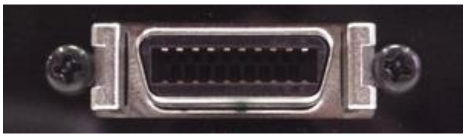




2 x 8 array



4 x 4 array

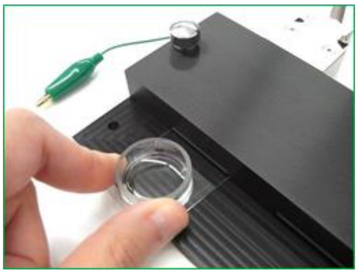


12	11	10	9	N	R	4	3	2	1
16	15	14	13	R	N	8	7	6	5

Correspondence between the MED Duet Connector output terminal and the electrode number. R indicates the reference electrode; N is not assigned.

3.4.4. Preparation for the noise check – attaching the MED Probe16

Insert the terminal of the MED Mini Probe filled with saline, such as aCSF or PBS into the insertion slot gently and horizontally, holding the ring chamber of the MED Mini probe or the end of the glass substrate. If any waterdrop, medium or smudge is on the terminal, wipe the terminal with a Kim wipe before setting the MED Mini Probe.

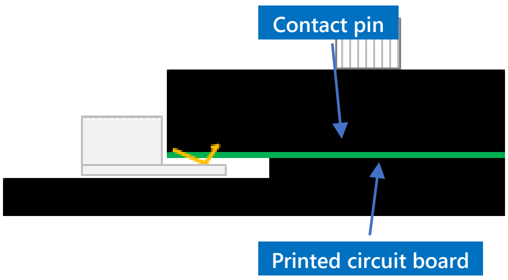
Note1: Pay attention so as not to spill liquid (especially saline) on the MED Mini Probe after it is set. Prevent liquids from coming into contact with the contact pins.

Note2: The test boards for a noise check are supplied with the MED Duet Connector as accessories, but it is recommended to perform a noise check using an unused MED Mini Probe (with no electrode detachment) filled with saline to check the influence of external noise from the installation environment. This is because the test board, which is designed to act as a resistor, is less susceptible to external noise with different frequency characteristics.

About the details of noise check, refer to p.19 “Noise check by operating the Mobius - procedure common to Basic, Quad II, and Allegro”. Also, for the details of operating the Mobius refer to “MED64 Mobius Tutorial.” Refer to the relevant application note for the preparation of specimens used for the experiment.

3.4.5. Maintenance of the MED Duet Connector

The contact pins, of 18 pins per well, constitute the area where the MED Duet Connector comes into contact (reads signals from) the MED Mini Probe terminal. If abnormal noise is observed at a particular electrode even when a noise test is performed with the test board, the noise may be caused by a contact pin being depressed onto the printed circuit board side or by dirt being attached to the pins. In the latter case, remove the top unit from the base plate and wipe the contact pins with a Kim wipe moistened with 70% ethanol by gently wiping from front to back. If any contact pins are rusted, damaged, or detached from the printed circuit board for whatever reasons, the printed circuit board needs to be replaced (a paid-for repair).



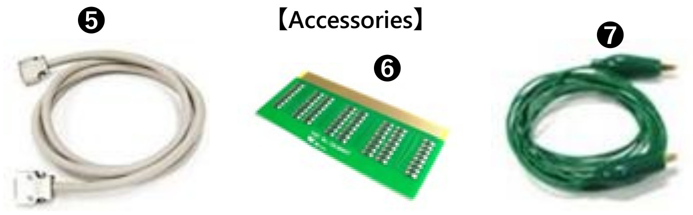

3.5. Setup of the MED64-Allegro

3.5.1. Part names and functions of the MED Multi-well Connector

【Front】

【Back】

【Accessories】



❶ Insertion slot of the MED Multi-well Probe

❷Ground terminal ….. To connect the SIGNAL GND terminal of the MED Head Amplifier using the accessory ground wire with a bagworm clip.

❸Screw hole ….. To fix the MED Multi-well Connector onto the MED Thermo Base (MED-CPB02).

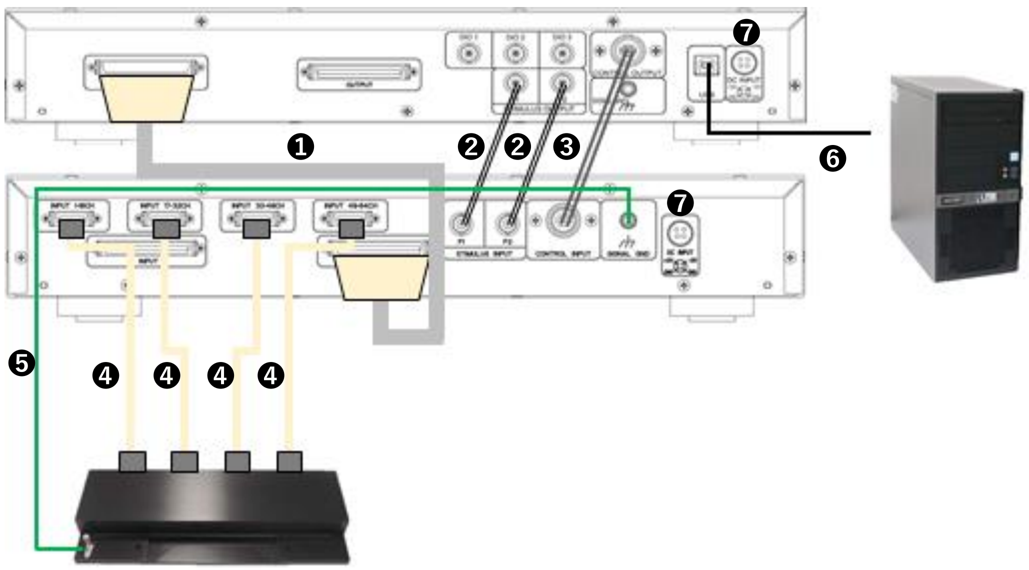
❹Output terminal ….. To connect to the INPUT terminal of the MED64 Head Amplifier via the 20-pin SCSI cable.

❺20-pin SCSI cable (2 m x 4) ….. To connect the output terminal and the INPUT terminal of the MED64 Head Amplifier (the specification is different from the 20-pin SCSI cable for the MED Duet Connector).

❻Test board ….. A PCB with multiple resistors to check the baseline noise level of the whole system and its environment.

❼Ground wire with a bagworm clip ….. To connect the ground terminal of the MED Multi-well Connector and the SIGNAL GND terminal of the MED64 Head Amplifier.

3.5.2. Connections among devices



❶Connect the INPUT terminal of the MED Main Amplifier and the OUTPUT terminal of the MED Head Amplifier, using the 68-pin SCSI cable (50 cm).

❷Connect the F1/F2 STIMULUS OUTPUT of the MED64 Main Amplifier and F1/F2 STIMULUS IMPUT of the MED64 Head Amplifier, using the BNC cables.

❸Connect the CONTROL OUTPUT terminal of the MED64 Main Amplifier and the INPUT terminal of the MED64 Head Amplifier, using the round-pin cable.

❹Connect the output terminals of the MED Multi-well Connector and the INPUT terminals of the MED64 Head Amplifier, using 20-pin SCSI cable.

❺Connect the ground terminal of the MED Multi-well Connector and the SIGNAL GND terminal of the MED64 Head Amplifier, using the ground wire with a bagworm clip.

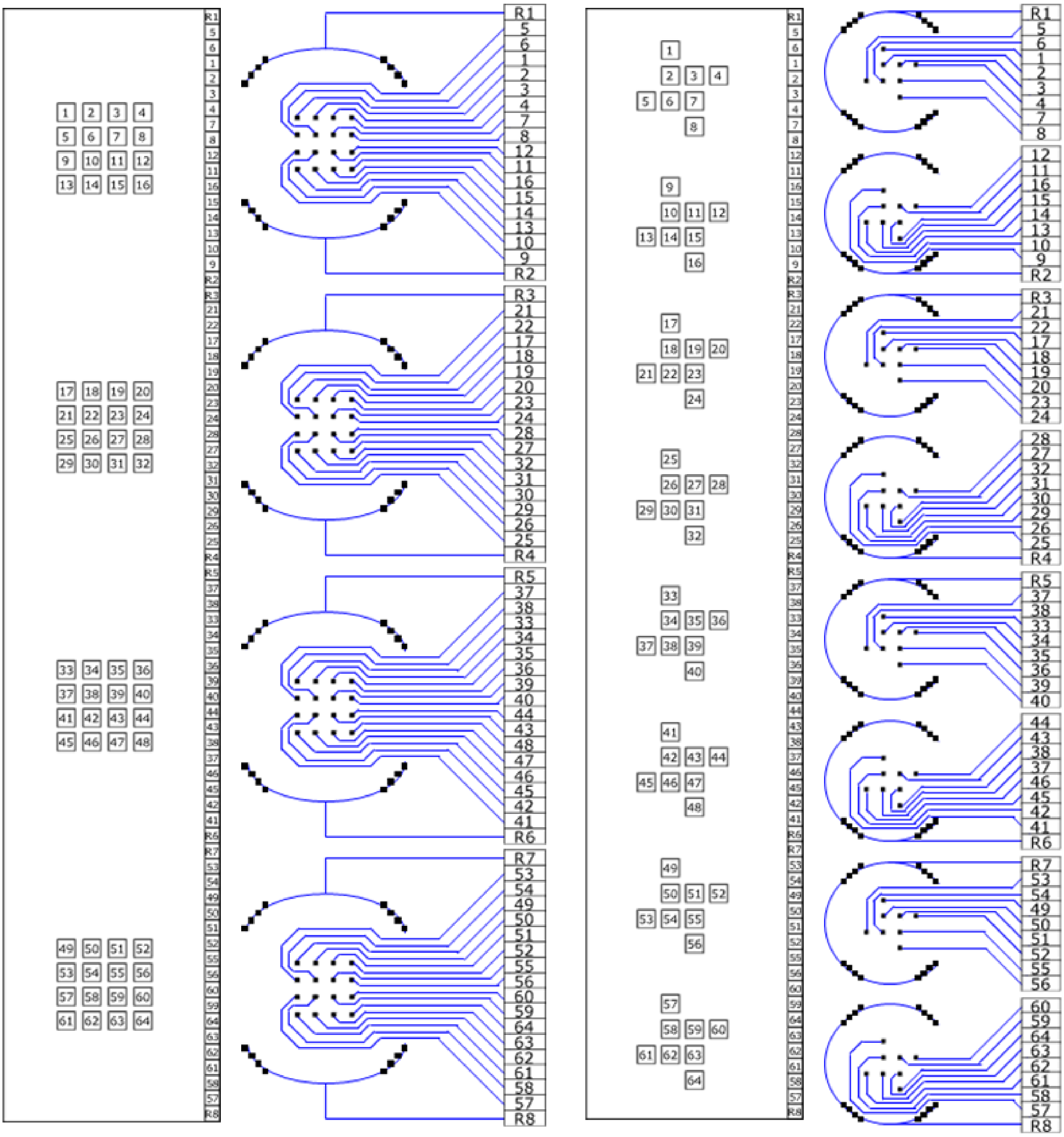
❻Connect the USB port (type-B) of the MED64 Head Amplifier and the USB port (type-A) of the data acquisition PC (USB2.0 is preferable), using the USB cable

❼Connect the power adaptor cables to both amplifiers.

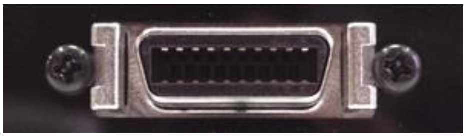
Note: The ground terminal of the MED Multi-well connector must be grounded to the SIGNAL GND terminal of the MED64 head amplifier. If not grounded, it will be affected by external noise.

3.5.3. Positioning of the terminal of the MED Multi-well Connector

The electrode number will be assigned as the below when place the MED Multi-well Connector with its output terminal and the 20-pin SCSI cable on the right side.



MED-P5NF30 MED-P5N811

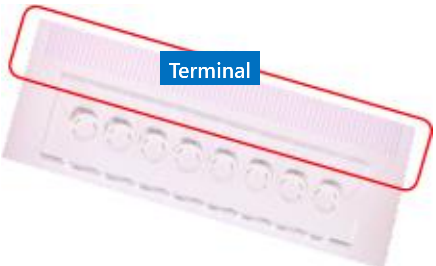


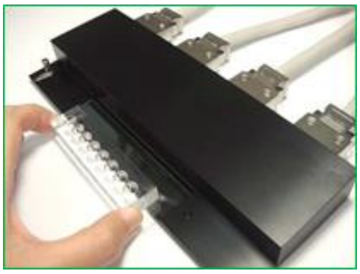
12	11	10	9	N	R	4	3	2	1
16	15	14	13	R	N	8	7	6	5

Correspondence between the MED Duet Connector output terminal and the electrode number. R indicates the reference electrode; N is not assigned. The pin alignment is divided in the center to correspond the adjacent two well while using the MED-P5N811.

3.4.4. Preparation for the noise check – attaching the MED Multi-well Probe

Insert the terminal of the MED Multi-well Probe filled with saline, such as aCSF or PBS into the insertion slot gently and horizontally, holding the well chamber of the MED Multi-well probe or the end of the glass substrate. If any waterdrop, medium or smudge is on the terminal, wipe the terminal with a Kim wipe before setting the MED Multi-well Probe.





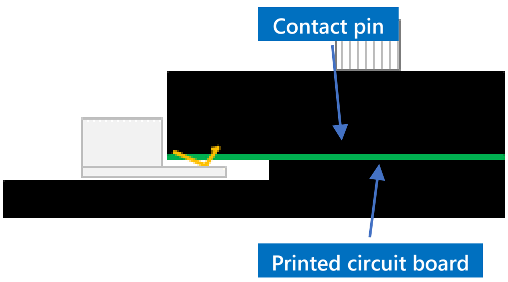
Note1: The terminal comes into contact with the contact pins of the MED Multi-well Connector. Attachment of chloride or a fingerprint may cause deterioration of the contact pins.

Note2: If liquid is spilled and comes into contact with the contact pins, contact failure may occur. In particular, pay attention so as not to spill saline.

About the details of noise check, refer to p.19 "Noise check by operating the Mobius - procedure common to Basic, Quad II, and Allegro". Also, for the details of operating the Mobius refer to "MED64 Mobius Tutorial." Refer to the relevant application note for the preparation of specimens used for the experiment.

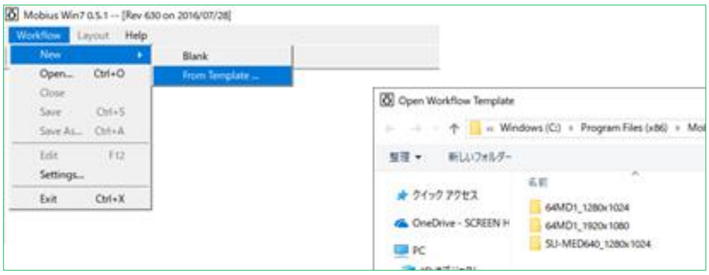
3.4.5. Maintenance of the MED Multi-well Connector

The contact pins, of 72 pins per well, constitute the area where the MED Multi-well Connector comes into contact (reads signals from) the MED Multi-well Probe terminal. If abnormal noise is observed at a particular electrode even when a noise test is performed with the test board, the noise may be caused by a contact pin being depressed onto the printed circuit board side or by dirt being attached to the pins. In the latter case, remove the top unit from the base plate and wipe the contact pins with a Kim wipe moistened with 70% ethanol by gently wiping from front to back. If any contact pins are rusted, damaged, or detached from the printed circuit board for whatever reasons, the printed circuit board needs to be replaced (a paid-for repair).



4. Noise check by using the Mobius - procedure common to Basic, Quad II, and Allegro

Tun on the power of the amplifiers, the data acquisition PC, and the display monitor. Start the Mobius by double clicking the Mobius icon on the desktop. Select Workflow > New > From Template on the menu bar and then select 64MD1_1280x1024 or 64MD1_1920x1080, depending on the resolution of the display monitor.

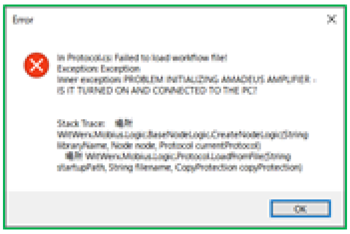


Note: Do not start other software during the data acquisition because it may place an unexpected load on OS and cause it to freeze or to force a shutdown. Especially, you need to pay attention to software running background, such as anti-virus software. Do not connect to the Internet, because it may place an unexpected load due to the communication and cause it to freeze or to force a shutdown.

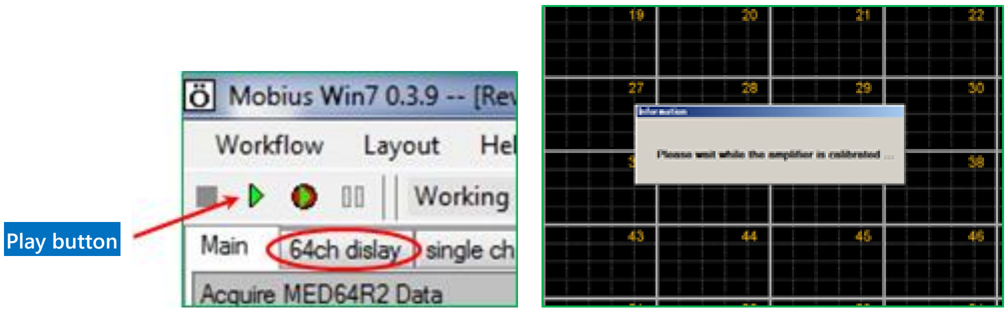
In addition, open the Basic_recording folder and select and call up “Noise_check.moflo” as the template workflow. The default conditions for data acquisition are shown below. Do not change these setting conditions as they serve as reference for the noise level check of the MED64 System.

Input Range (mV): 5.0、Low cut freq (Hz): 1、High cut freq (Hz): 10000

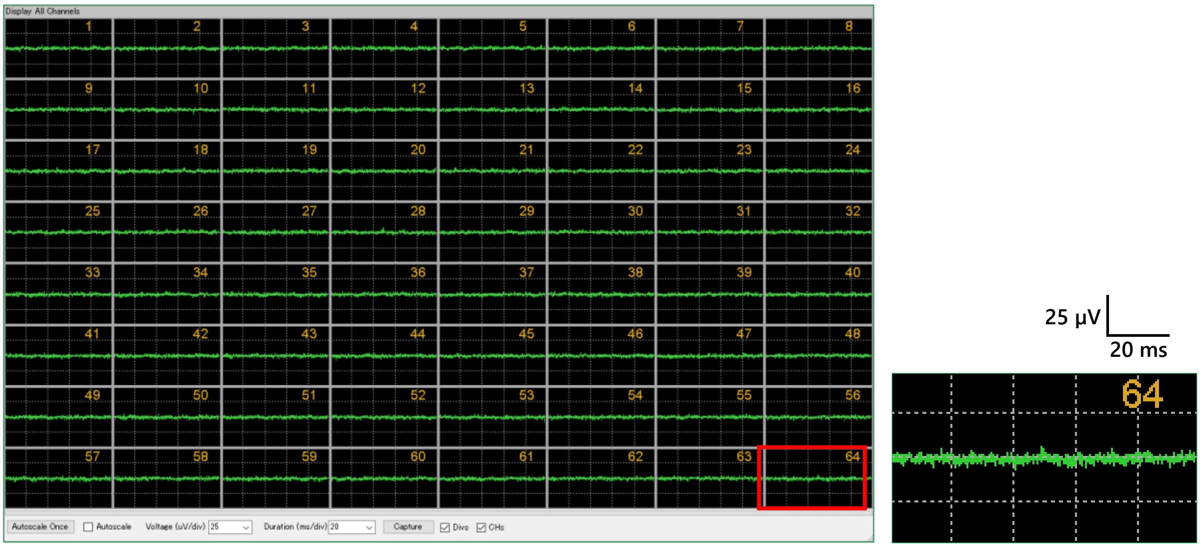
Note: To call up Noise_check.moflo, the workflow communicates with the MED64 Main Amplifier via the USB cable. If the Amplifier is not turned on, a pop-up window appears with an error message, as shown in the right figure.



Click the 64ch display tab to display the 64ch display window. Click the play button (playback button) to start data acquisition without outputting a data file. Once the button is clicked, a pop-up window appears saying “Please wait while the amplifier is calibrated,” but the pop-up window disappears when a few minutes of the calibration is completed and the Mobius automatically starts data acquisition with the waveform of the baseline noise displayed on the 64ch window.



The ideal baseline noise level is about ±5-8 μV peak-to-peak. If the baseline noise shows an amplitude greater than the value, take measures against the noise, with reference to p.19 “6. Abnormal noise.”



Normal baseline noise.

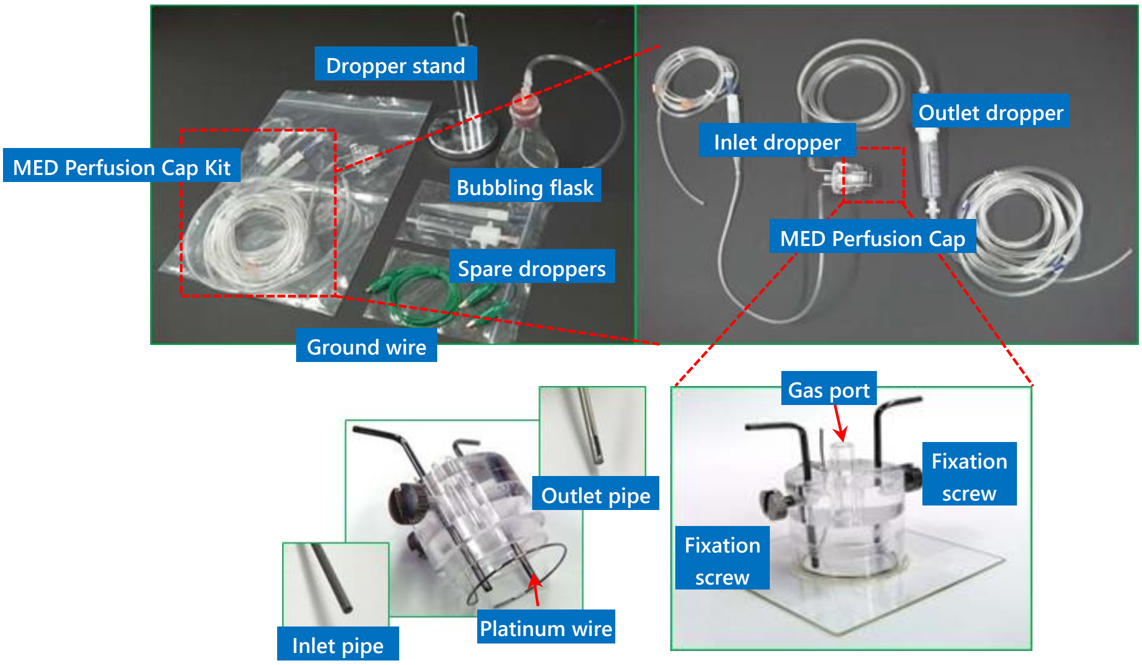
5. Combining with a perfusion system

This section explains the procedure for setting up a perfusion system when the MED64 System is used in combination with it. The perfusion system recommended to be used in combination with the MED64 System consists of the following:

1) MED Perfusion Cap Kit【MED-KCAP01TU】
2) Peristaltic Pump【Mini Pulse III Model 312 / MP-2, Gilson】

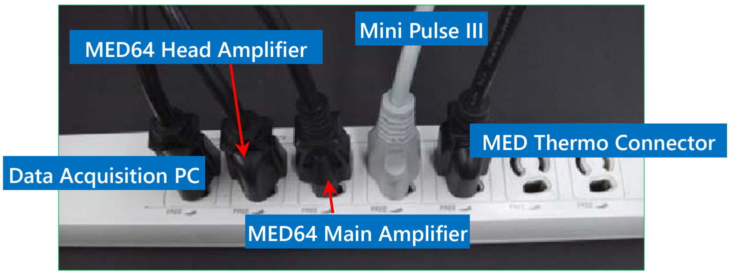
Note: When a perfusion pump is used, read the pump's product manual thoroughly and follow the instructions. Set up the MED64 System properly before setting up the perfusion system.

The MED Perfusion Cap Kit is designed to be used with Minipuls 3. If a perfusion pump other than Minipuls 3 is used, change the tubing configuration to one that is suited to the perfusion pump before use. (Please note that the operation of the MED64 System cannot be guaranteed if it is used in combination with pumps other than Minipuls 3 for the MED Perfusion Cap Kit.) Extra droppers and leads for grounding are supplied as accessories for the MED Perfusion Cap Kit, as shown in the figure on the next page.



5.1. Position of the Minipuls 3 and power supply

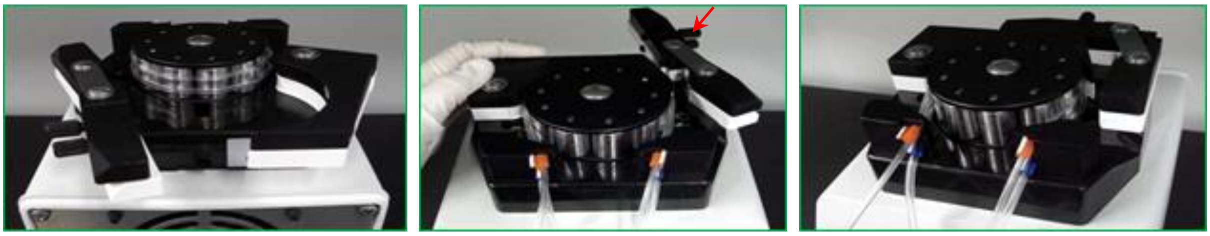
The Minipuls 3 itself serves as a magnetic noise source. If it is suspected that it is affecting the baseline nose level, place it away from the amplifier, the MED Thermo Connector, and the 68-pin SCSI cable, considering its distance from the devices. Connect the three-terminal plug of the power cable of the Minipuls 3 to the same power strip into which the amplifier is plugged.



5.2. Installing MED Perfusion Cap Kit to the Minipuls 3

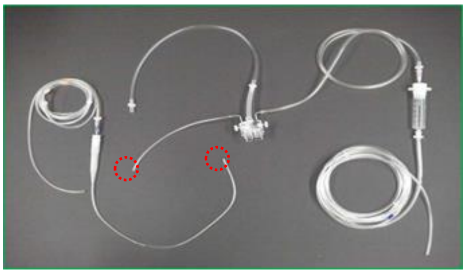
Unlock the 2-channel (upper and lower) locking bars of the Minipuls 3 and loosen the compression cams. Wind the flow tubes of the MED Perfusion Cap Kit around the rollers and fix the tubes by catching the tube hook with the plastic part. Pay attention to the orientation of the flow tubes so that the perfusion solution properly runs from the liquid supply side to the drainage side along the rotation direction of the rollers (clockwise or counterclockwise). The example in the figure below shows that the flow tubes are wound so that the lower (white) compression cam is on the drainage side, while the upper (black) compression cam is on the liquid supply side.

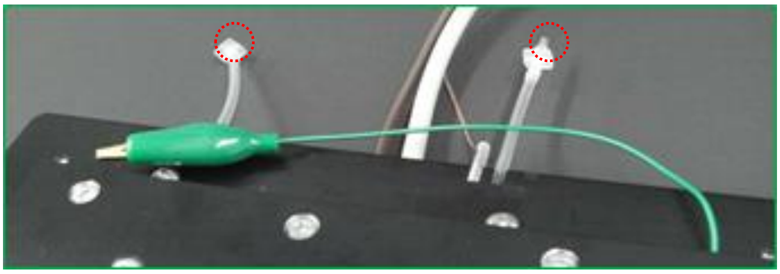
In the area of the flow tubes to which the compression cams apply pressure, adjust the winding so that both the flow tubes are horizontal around the center of the upper and lower compression cams and then press the compression cams onto the tubes and fix them with the locking bars. Make sure that the adjustment screws of the Adjustment screw completely loosened when fixing the tubes.



5.3. Connection between MED Thermo Connector and MED Perfusion Cap Kit

The MED Thermo Connector contains an in-line tube (or pipe). Connecting the inlet tube (pipe) to the in-line tube (or pipe) enables the perfusion solution to flow through and to be warmed.



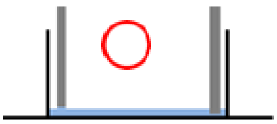


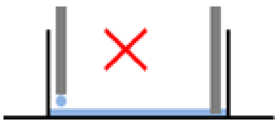
5.4. Fixing the droppers to the dropper stand

Temporarily disconnect the fitting (connection part) of the droppers so that they go through the ring in the dropper stand and position the droppers vertically. Keep the outlet dropper upright and fix it so that the ring holds the rubber stopper. Slightly slant the inlet dropper, because positioning it upright may cause air entrainment of droplets at the connection part with the tube.

5.5. Adjusting the height for the inlet/outlet pipes in the MED Perfusion Cap

Adjust the height for the inlet/outlet pipes from the bottom of the MED Probe with the fixation screws. Position the inlet pipe so that its bottom tip is slightly above the bottom of the MED Probe. This is because, depending on the relationship with the position of the outlet pipe, the inlet pipe positioned higher than the solution surface level may cause noise every time a droplet falls on the surface. Fix the outlet pipe at a certain height that serves the purpose.

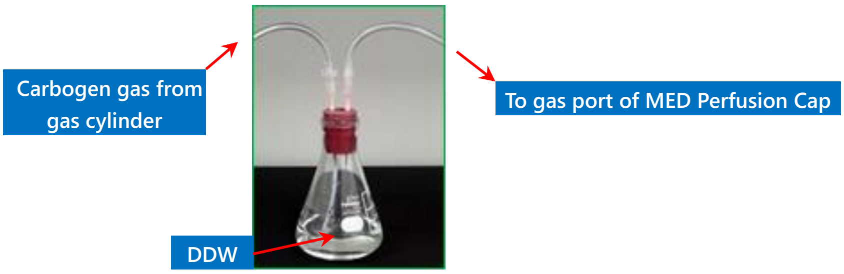




Note: The experiment condition in which the solution surface is as high as the surface of the brain slice (the solution barely covers the surface of the brain slice) is called the interface condition, while the experiment condition in which the solution is deep enough to submerge the brain slice is called the submerge condition. The interface condition that enables oxygen to be supplied from the gas port provides a greater response than the submerge condition, when compared at the same perfusion rate.

5.6. Connecting the gas tubing to the gas port of MED Perfusion Cap

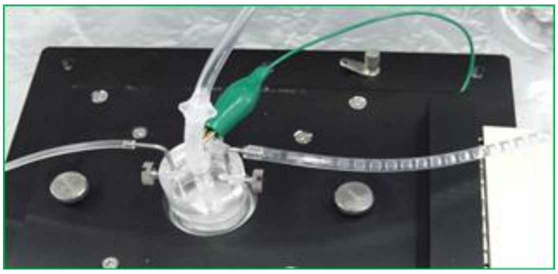
To connect the gas tubing to the gas port of the MED Perfusion Cap, use a conical flask for aeration to humidify the carbogen gas (95% O_2, -5% CO_2) without directly connecting the gas tubing to the gas port. Fill the flask with distilled water before use and supply the carbogen gas humidified by bubbling to the MED Probe chamber through the gas port.



5.7. Grounding platinum wire of the MED Perfusion Cap

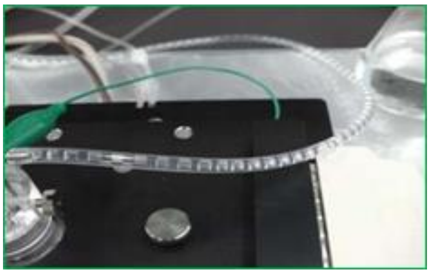
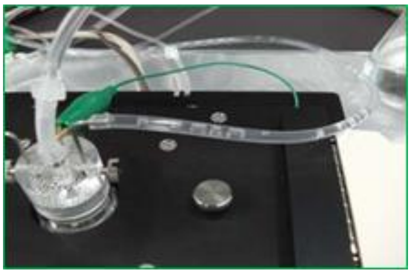
When platinum wire are gripped with a ground wire with a bagworm clip of the MED Connector, the platinum wires function as additional reference electrode. The reference electrodes of the MED Probe are positioned at 4 points, and these additional reference electrode of the platinum wire further reduce the total impedance of the reference electrodes and reduce the noise from the perfusion pump and the stimulation artifact.

Note: Make sure that the platinum wire is fully immersed in the perfusion solution (electrolyte) to conduct them with the reference electrodes of the MED Probe, and that they are gripped by the ground wire with a bagworm clip. If the platinum wire are open, they serve as antennas that collect electric noise and cause noise.




5.8. Running the Minipuls 3

Turn on the power of the Minipuls 3 to activate it. The inner diameter of the flow tube is larger on the drainage side than on the liquid supply side, and the drainage pipe has holes with slits. This design allows the drainage pipe to suck the perfusion solution and air, thereby producing layers of air inside the perfusion tube at a constant speed, which indicates the stability of perfusion. On the other hand, if layers of air are randomly produced, such as when the solution stops flowing at a certain period and then flows out suddenly, the surface level of the solution in the chamber may abruptly change, resulting in the occurrence of noise caused by fluctuation at the baseline.

5.9. Noise check by operating the Mobius

Check the noise level while performing perfusion, referring to p. 15 "4. Noise check by operating the Mobius." When the Minipuls 3 is running with no problems, a flat and stable baseline noise (±5 to 8 μV) is obtained in the same manner as when it is operated without perfusion. If constant noise is observed every hundred milliseconds, the noise may be reduced by grounding the inlet pipe to the platinum wire.

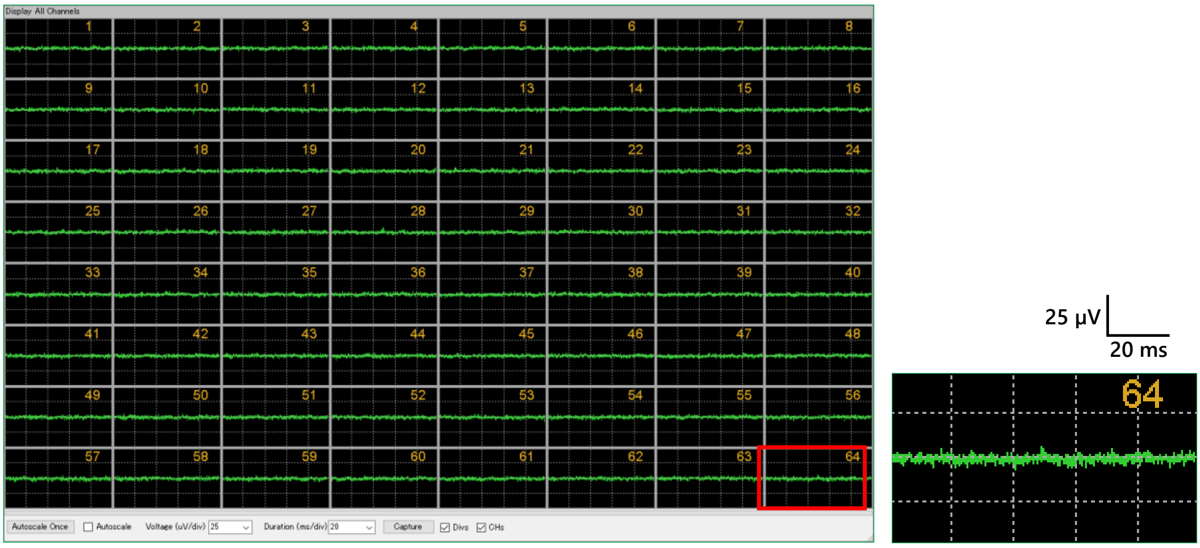
Note1: Grounding the inlet pipe to the platinum wire forms a grounding loop, causing noise.

Note2: After use, clean by perfusing distilled water in the perfusion tube, and replace the perfusion tube when necessary if the dropper chips and the inside of the syringe is completely hydrophilic or if too many blemishes are observed on the perfusion tubes.

6. Abnormal noise

6.1. Noise check point

- The amplitude of normal baseline noise has about ±5-8 μV peak-peak value in bandwidth between Low cut freq 1 Hz and High cut freq 10,000 Hz for signal acquisition. Visually check whether the baseline noise is flat on the display with 20 ms/div on the horizontal axis and a display 500 ms/div on the horizontal axis.



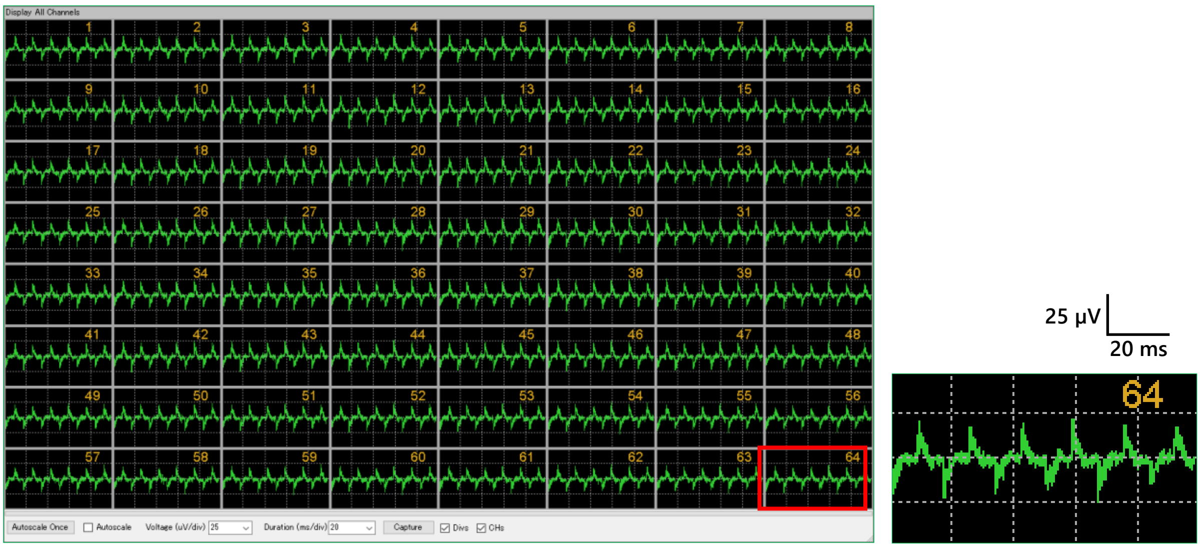
Normal baseline noise.

- For electrodes with noise, check whether noise occurs in a particular subset of electrodes or in all 16 electrodes and whether there is any bias in the noise.
- For the shape of noise, set the horizontal axis at 20 ms/div and check whether the noise is a sinusoidal-like noise repeated 5 or 6 times or a high-frequency noise disproportionately observed in particular electrodes.
- There are two types of extrinsic noise; noise that can be shielded by an electric shield regardless of distance, and noise for which an electric shield is not effective but can be reduced by moving the noise source to a distant position. The latter noise may be caused by a magnetic field generated by a device connected to the power cable (power adaptor, perfusion pump, and water bath, etc.). Move the responsible device to a position where the noise disappears or unplug the power cable of that device when MED64 System is being used.

Note: To contact our help desk, take a screenshot of 64 electrodes display (Display All Channels module panel), paste the image of 64 electrodes on an image editor such as paint, store in jpg or tif format, and send it to the help desk as an e-mail attachment. Do not use bmp format because it has a large file size. Prepare images with 20 ms/div and 500 ms/div horizontal axis and 25 µV/div vertical axis, or a vertical axis automatically adjusted by Autoscale.

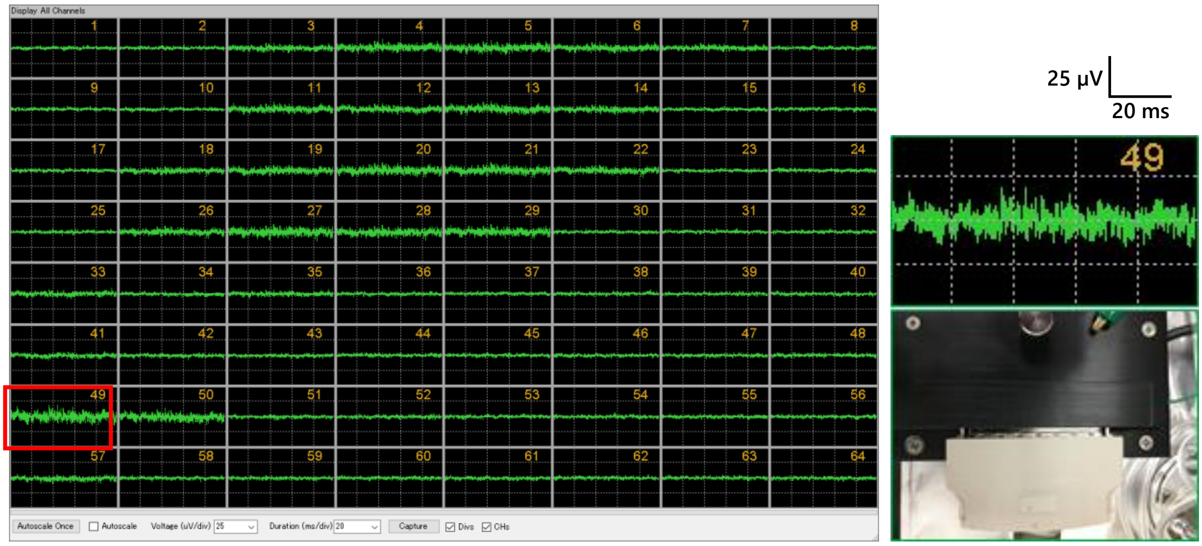
6.2. Noise relating to installation

- A fixation screw of the MED connector is loose.



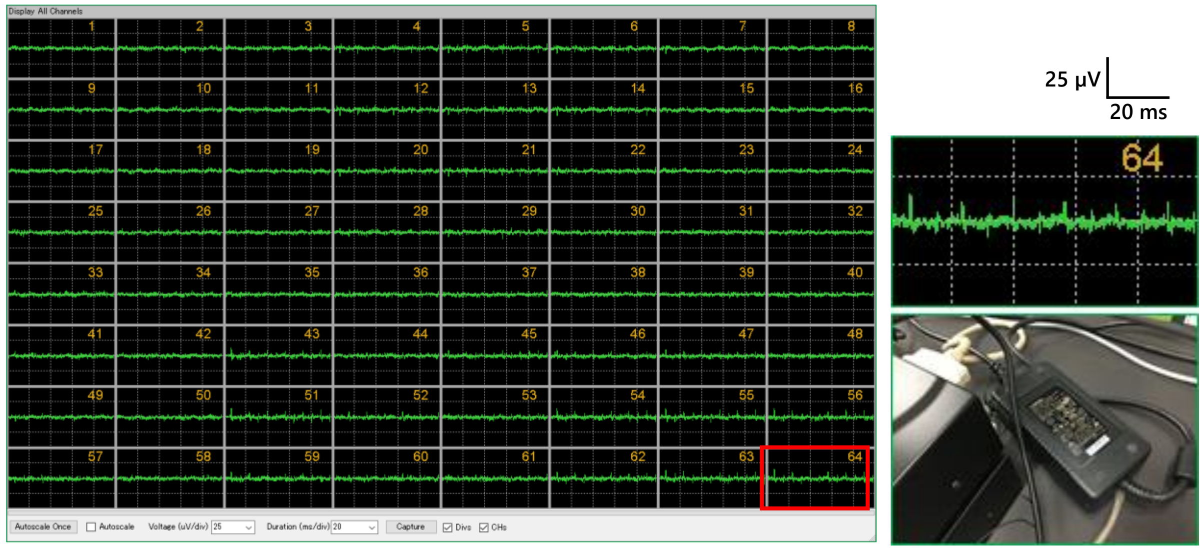
An example of noise when a fixation screw is loose. Noise is likely to occur in all 64 electrodes.

- Poor contact between the SCSI cable and the analog input terminal.



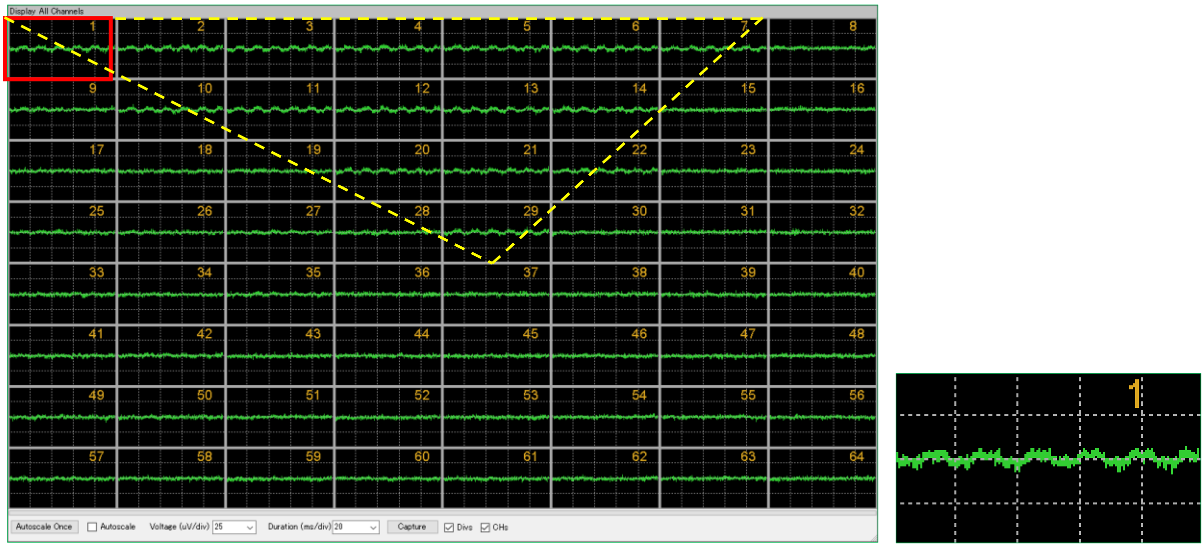
An example of noise when the contact of the SCSI cable is poor. It might be likely occur when switching the connection between the MED Connector and MED Duet Connector.

- The power adaptor or other electronic device (incubator, etc.) is placed close (about 30 to 100 cm) to the terminal of the amplifiers or the 68 pin SCSI cable.



Magnetic field noise caused by a nearby power adaptor. Beard-shaped characteristic noise. Interference (an effect of the magnetic field) occurs in particular electrodes.

- A heater circuit or electromagnetic valve of the incubator operates and a magnetic field is generated inside the incubator.

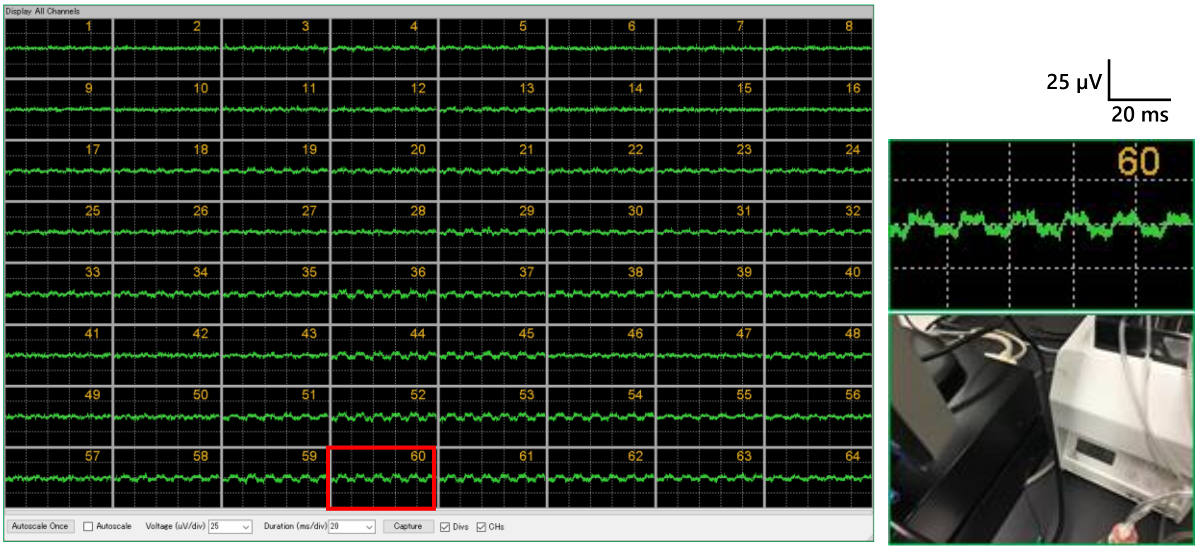


An example of magnetic field noise caused by an incubator. Interference occurs in particular electrodes, the magnitude of effect depends on the position in the incubator, and the noise disappears when the temperature is stabilized (an example screen of the MED64-Basic to clearly show the disproportionate effect).

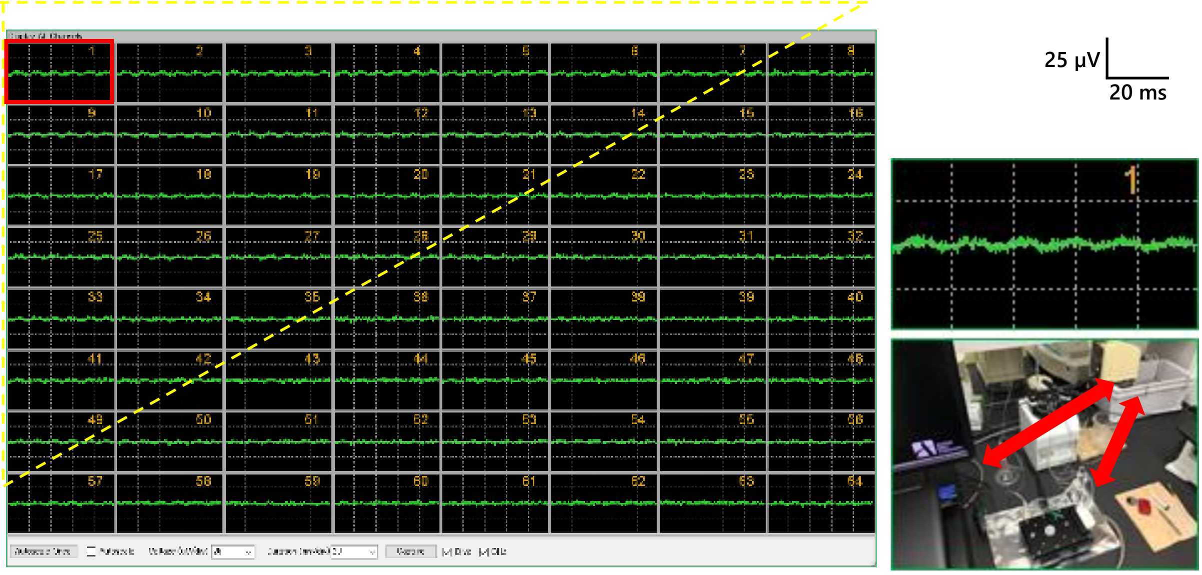
- A power cable of an electronic device not relating to the MED64 system is connected to the power strip, and the noise disappears when the device is disconnected.

6.3. Noise relating to perfusion

- A perfusion pump or its power adaptor is placed close to the amplifier, the MED Connector, or the 68 pin SCSI cable.



An example of magnetic field noise caused by a nearby perfusion pump. Interference occurs in particular electrodes (an example screen of the MED64-Basic to clearly show the disproportionate effect).



An example of magnetic field noise caused by a water bath placed close to the MED64 system. A relatively large area is affected (an example screen of the MED64-Basic to clearly show the disproportionate effect).

- The electrodes' surfaces of the MED Probe or platinum wire is not completely immersed in perfusion solution. Some parts are not exposed to the atmosphere.
- The platinum wire is not gripped by a ground wire with a bagworm clip.
- The platinum wire is not stably fixed with a fixation screw (it is prone to staggering).
- A spike-like noise occurs at several seconds interval. A flat baseline noise is obtained when the peristaltic pump is turned off.
  * The inside of the dropper is completely hydrophilic with a lot of water dropping. It is likely that the inside insulation is destroyed every time the perfusion solution drops, and the noise from the pump is conducted. Connect the inlet pipe and the platinum wire using a ground wire with a bagworm clip. When the noise disappears, the inlet dropper is identified as the cause (this procedure cannot be used for the outlet dropper because an air layer is mixed).
  * Check whether the tip of the inlet pipe is too close the platinum wire to not putting hydraulic pressure when the perfusion solution drops.
  * Check whether air entrainment occurs in the inlet dropper and bubbles enter the MED Probe chamber periodically to compress the platinum wire.

6.4. Identification of cause when a malfunction of the device is suspected

If the noise persists in a particular electrode even after setting an electric shield, moving the possible noise source or unplugging the power cable, a malfunction may have occurred in the device. Identify the cause by the procedures below:

Procedure 1: MED Probe

Turn the orientation of the MED Probe by 90° and connect it to the MED Connector. Check whether the electrode where the noise is generated moves to a position according to the orientation of the MED probe (e.g., whether the position where the noise occurs moves from Electrode 1 to Electrode 8). If the position moves in line with the orientation of the MED probe, the coating of the relevant electrode may have come off or the insulation layer on the ITO lead pattern to the electrode may have been damaged (if it is detected in the first time use within 6 months after buying, it has an early failure and will be replaced with a new one). If the position does not move, proceed to Procedure 2 and the subsequent procedure.

Procedure 2: poor contact of cable or lead

If poor contact is detected in the output terminal of the MED Connector or the input/output terminal of the amplifiers, disconnect the SCSI cable completely and connect it again. Check whether the lead or alligator clip connected to the GND terminal has come off or not.

Procedure 3: MED Connector

Check whether poor contact with the MED Probe terminal occurs or not because of sebum or dust on the contact pin. Wipe the contact pin by pushing it with an eyeglass cleaner cloth and attach it the MED Probe again. If the noise persists, visually check whether the levels of some contact pins are lower than those of other contact pins. Contact pins are filled with oil. If the oil comes off due to age deterioration and a pin becomes less elastic, the pin stays in a depressed position. This causes unreversible poor contact between the pin and the MED Probe terminal, which necessitates repair by replacement of the substrate. If the noise disappears after replacement with a normal MED connector, the cause is identified as wear of the contact pin.

Procedure 4: MED64 Head Amplifier and MED64 Main Amplifier

If the noise is reduced by turning off the MED64 Head Amplifier and only the MED64 Main Amplifier is active, a malfunction of the MED64 Head Amplifier is suspected. If the noise persists, a malfunction of the MED64 Main Amplifier is suspected.

7. Appendix

7.1. Specification

7.1.1. MED64 Main Amplifier [MED-A64MD1A]

General information	
Power	DC ±12V
Weight	5.9 kg
Size	W430 x D437 x H74 mm
Input terminal	68-pin x 1
Output terminal	68-pin x 1
Amplifer	
Number of channels	64
Gain *1	×20 - 217
Bandwidth *2	0.1 Hz - 10 kHz (+0 dB - 3 dB)
Analog high-cut filter	1 / 2.5 / 5 / 7.5 / 10 kHz (-12 dB/oct)
Analog low-cut filter	0.1 / 1 / 10 / 100 Hz (-12 dB/oct)
Input impedance	100 MΩ
Digitizer	
Resolution	16 bit
Sampling rate	20 kHz
Output	USB
Power adaptor	
Input	AC 100 - 240 V (50 / 60 Hz)
Output	DC ±12V

※1 Bandwidth

The MED64 Main Amplifier enables the acquisition of signals for a wide range of bandwidths between 0.1 Hz and 10 kHz. It also has an analog low-cut filter (high pass filter) for cutoff frequencies of 0.1, 1, 10, and 100 Hz and an analog high-cut filter (lowpass filter) for cutoff frequencies of 1, 2.5, 5, 7.5, and 10 kHz. Frequencies can be set on the Mobius for both the filters. However, please note that applying analog filtering may cause a distortion of the original waveforms of the signals written in the data file. Therefore, it is recommended to record signals for a wide range of bandwidths and to apply digital filtering as necessary for the data analysis. The MED64 System shows quite low noise levels thanks to the low impedance electrodes of the MED Probe. Even if the analog high-cut filter is set to 10 kHz, the system provides superior signal to noise ratios. In the analog low-cut filter, on the other hand, setting the lowest frequency of 0.1 Hz is not required except when it is used to obtain signals that contain slow frequency components, such as the pacemaker potential of a gastrointestinal tract muscle layer tissue specimen or the second peak of a cardiac culture. The recommended setting is as follows:

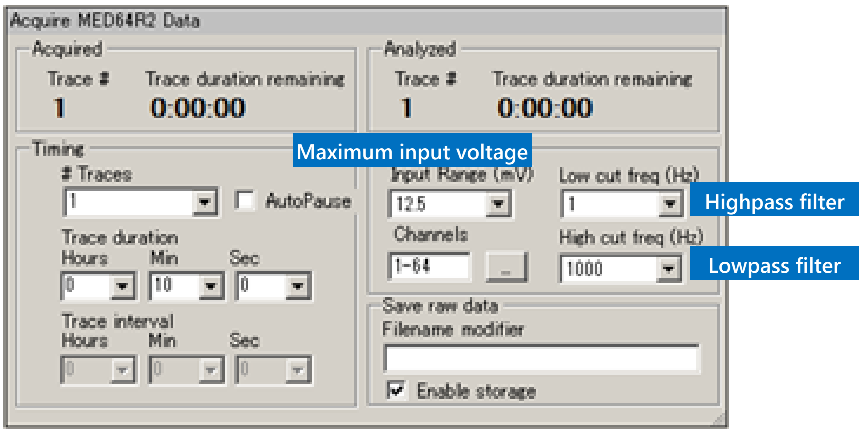
- Evoked potentials in a brain slice: 1 Hz - 10 kHz
- Spontaneous firing in a dissociated neural culture (spike detection): 1 Hz - 10 kHz
- Spontaneous beating signal in a cardiac culture (FPD assay): 0.1 Hz - 10 kHz

※2 Correlation between gain and voltage resolution

The acquired signal is amplified tenfold by the MED64 Head Amplifier, and then to transmitted to the MED64 Amplifier. The gain of the MED64 Main Amplifier is selected by changing “Input Range (maximum input voltage)” on Acquire MED64R2 Data or MED64R2 Data w/Stim module panel of the Mobius. The maximum input voltage selected represents the upper limit of the amplitude of the potential acquired. When 5 mV is selected for example, a signal exceeding 5 mV is acquired as a 5 mV signal. The low level maximum input voltage means high resolution. The resolution of amplitude obtained by the maximum allowable input is shown in the table below.

Input Range (mV)	Gain	Resolution
2.3 mV	×217	0.07 V
2.9 mV	×172	0.09 V
5.0 mV	×100	0.15 V
12.5 mV	×40	0.38 V
25 mV	×20	0.76 V

The recommended Input Range is as below.
- Evoked potentials in a brain slice: 5.0 mV
- Spontaneous firing in a dissociated neural culture (spike detection): 2.3 mV
- Spontaneous beating signal in a cardiac culture (FPD assay): 12.5 mV



Setting of maximum input voltage and high/low pass filter on Acquire MED64R2 Data module panel.

7.1.2. MED64 Head Amplifier [MED-A64HE1S]

General information	
Power	DC ±12V
Weight	6.6 kg
Size	W430 x D437 x H74 mm
Input terminal	68-pin x 1, 20-pin x 4
Output terminal	68-pin x 1
Amplifier	
Number of channels	64
Gain	x 10
Bandwidth	0.1 Hz - 10 kHz (+0 dB - 3 dB)
Input impedance	100 MΩ
Output impedance	10 kΩ
RMS noise (typical value) Input short-circuit: MED-P515A input:	 14 nV/$\sqrt{f}$ 2.0 μV (f < 10 kHz)、1.3 μV (f < 5 kHz)、1.0 μV (f < 3 kHz)
Digitizer	
Number of channels	2
Output format	Current-driven
Maximum input voltage	±4 V
Maximum output current	±200 μV
Power adaptor	
Input	AC 100 - 240 V (50 / 60 Hz)
Output	DC ±12V

7.1.3. MED Connector

General information	
MED Probe securing mechanism	Screw down
Material	Aluminum, Gold plate for contact pins
Contact impedance	< 30 mΩ
Weight	480 g
Size	W174 x D150 x H21 mm

7.1.4. MED Duet Connector

General information	
MED Probe securing mechanism	Slide in
Material	Aluminum, Gold plate for contact pins
Contact impedance	< 30 mΩ
Weight	381 g
Size	W130 x D85 x H30 mm

7.1.5. MED Multi-well Connector

General information	
MED Probe securing mechanism	Slide in
Material	Aluminum, Gold plate for contact pins
Contact impedance	< 30 mΩ
Weight	700 g
Size	W210 x D83 x H34 mm

7.1.6. Data Acquisition PC System

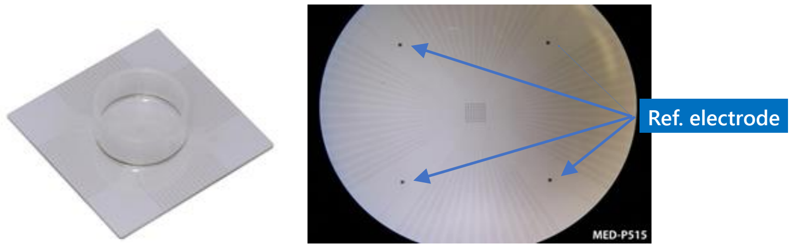
Any PC model with equivalent performance can be used.

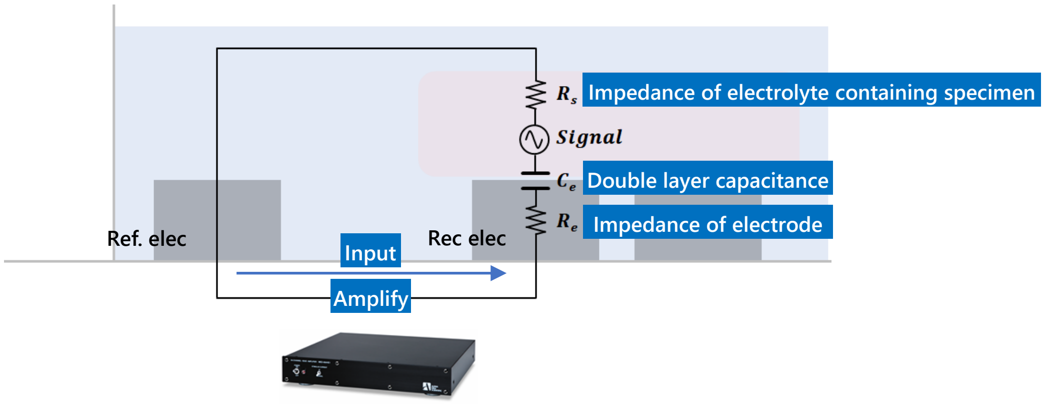
Computer	
Case type	Mini-tower desktop
Operating system	Windows 10 Pro 64bit
CPU	Intel Core i7-6700K (4 core/4.00GHz/8MB cache)
RAM	16GB (8GB×2)
Main disk drive (for OS)	512GB SSD
2nd disk drive (for data storage)	2TB HDD (7200rpm/6Gbs)
Optical drive	DVD super-multi drive
USB terminal	More than 1 (USB2.0 is preferable)
Accessories	Keyboard, mouse
Weight	6.9 kg
Size	W175 x D385 x H365 mm
Display monitor	
Resolution	23 inches with 1920×1080 resolution
Weight	4.0 kg
Size	W533 x D180 x H388 mm

7.2. Explanation of the technology

7.2.1. Signal acquisition by the MED64 system

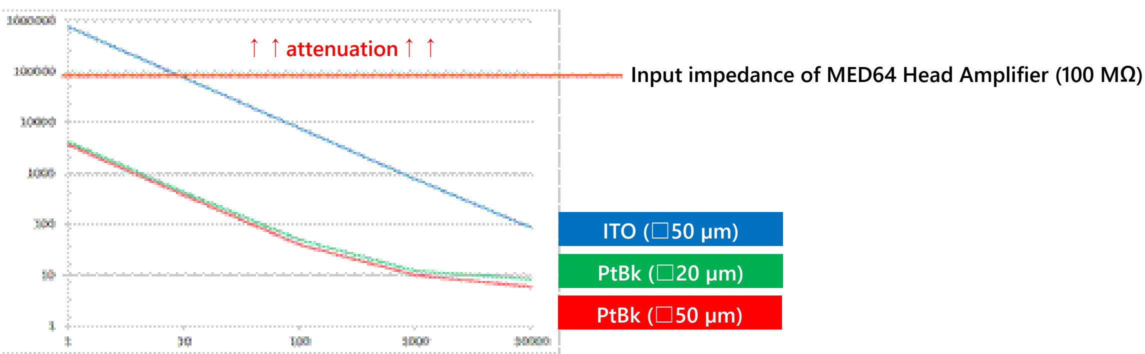
In the MED Probe, 64 recording electrodes and 4 (or 8, 16) reference electrodes are arrayed in a pattern. The MED64 System acquires the potential between the recording electrodes and the reference electrodes. The figure below illustrates an equivalent circuit between the recording electrodes and the reference electrodes.





<u>Input impedance necessary to avoid attenuation of the signal</u>

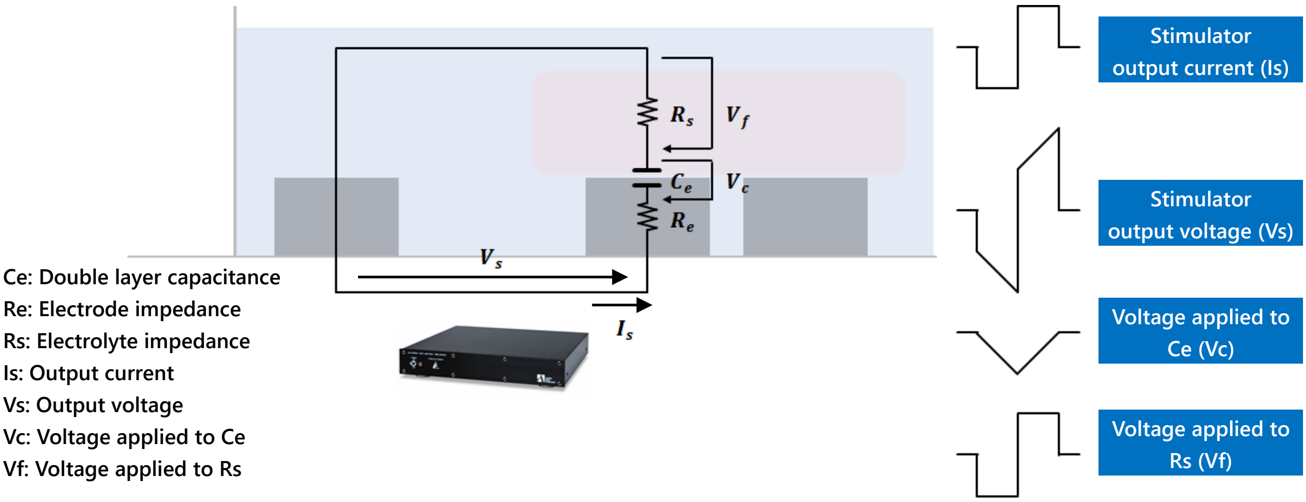
One of unique characteristics of a MEA an "electrical double layer capacitance formed between the interface of the electrodes and the electrolyte. An electrical double layer capacitance increases impedance in the low frequency area of the electrode (see the figure below). When the electrode impedance is higher than the input impedance of the amplifier, attenuation occurs in the signal acquired. The platinum black electrode on the MED probe has very low impedance (at signal frequency 1 kHz, 10 kΩ for 50 µm×50 µm electrode, and 15 kΩ for 20 µm x 20 µm electrode), and impedance is low in the low frequency area. Therefore, the MED64 System has sufficiently high input impedance of 100 MΩ and a low frequency signal can be acquired without attenuation, such as the evoked potential of a brain slice or the pacemaker potential of a gastrointestinal tract muscle layer tissue. When 1 Hz signal is acquired at an ITO electrode without a coating, the signal is attenuated because its electrode impedance is higher than 100 MΩ (blue line in the figure below). On the other hand, a platinum black electrode can acquire the signal without attenuation because its impedance is lower than 100 MΩ.



Impedance relationship between electrode and MED64 Head Amplifier.

7.2.2. Electric stimulation by the MED64 system – Applying current to the micro electrode array

The MED64 Head amplifier has a 2-channel current drive stimulator that can apply electric stimulation to any of the 64 electrodes. The figure below illustrates an equivalent circuit of electric stimulation by the MED64 system. The stimulation current flows from the electrode selected to the reference electrode, and then it is stored in the electrical double layer capacity on the electrode surface and changes the electrode potential. This change in potential changes the extracellular potential and induces depolarization of the cell membrane. As shown on the right side of the figure below, the extracellular potential (Vf) changes according to output current (Is).



Electrical stimulation via micro electrode.

7.2.3. Stimulus artifact and biphasic stimulation

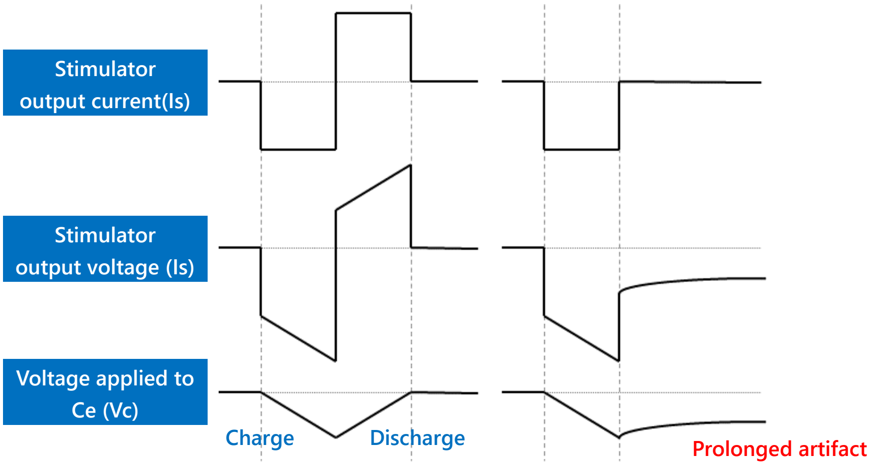
Because of cross-talk between electrodes, the stimulation pulse waveform is received as a "stimulation artifact" by all recording electrodes. A stimulation artifact remains after the end of the stimulation pulse and interferes with the signal waveform from the biological specimen. Mainly the factors listed below influence this process.

1. Typically, the amplitude of the stimulation artifact exceeds the maximum input voltage of the amplifiers and causes tentative saturation of input. Time is necessary to recover from this condition. This time lag increases in proportion to the stimulator output voltage (Vs).
2. At the electrode, the time necessary for discharge is longer than that for charge. This time lag increases in proportion to voltage Ce (Vc).

Vs and Vc are low because of the low impedance (high capacitance) of the electrode. The MED64 system should be operated properly to minimize the duration of the stimulation artifact. Follow the instructions described below to minimize the width of the stimulation artifact.

1. Apply a current drive biphasic pulse stimulation consisting of a negative and positive pulse with the same width. When the biphasic pulse stimulation is applied, charge and discharge at the electrode occur quickly. On the other hand, when a monophasic pulse stimulation is applied, discharge takes longer than charge at the electrode, which markedly prolongs the stimulation artifact and damages the electrode.

Note: For the MED64 system, a current drive biphasic stimulation consisting of a negative and positive pulse of the same width is recommended. A monophasic pulse stimulation damages the electrodes.



Biphasic pulse stimulation and monophasic pulse stimulation.

2. A platinum wire is used as additional reference electrodes. When a platinum wire is gripped with a ground wire with a bagworm clip of the MED Connector, the platinum wire functions as an additional reference electrode. The reference electrodes of the MED Probe are positioned at 4 points, and this additional reference electrode of the platinum wire further reduce the total impedance of the reference electrodes and reduce the noise from the perfusion pump and the stimulation artifact.

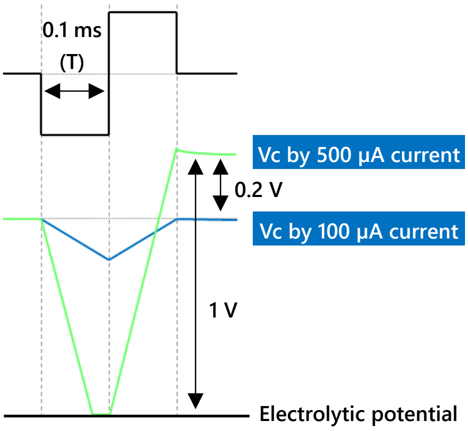
Note: Make sure that the platinum wire is fully immersed in the perfusion solution (electrolyte) to conduct with the reference electrodes of the MED Probe, and that it is gripped by a ground wire with a bagworm clip. If the platinum wire is open, it serves as antennas that collect electric noise and cause noise.

Reducing a stimulation artifact by a platinum wire. Examples without (left panel) and with (right panel) a MED perfusion cap

7.2.4. Stimulus intensity and electrolysis

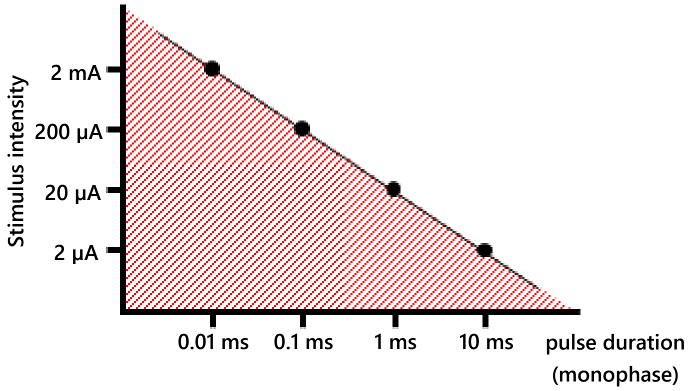
As described in the above section, the stimulation current is charged in the electrical double layer capacity and causes voltage Vc (see figure below). When the voltage Vc exceeds 1 V, the electrodes induce electrolysis and discharge H_2. This causes a marked decrease in stimulation efficiency and a marked expansion of the stimulation artifact, and it damages cells and the electrodes. Therefore, it is essential to prevent a decomposition voltage by stimulation current (Is).

Vc: Voltage applied to Ce
Is: Stimulator output current
T: Pulse duration
Ce: Double layer capacitance



Vc is defined as Vc = Is × T / Ce. A 50 x 50 μm electrode of the MED Probe has a 50,000 pF capacitance (Ce) (22,000 pF for 20×20 μm electrode). Up to 0.2 ms width (1 phase 0.1 ms) is acceptable for a 200 μA stimulation current.

Note: Do not apply a stimulation intensity and pulse duration width spanning to outside of the shaded area in the figure below. The numerical values in the graph are based on a "single pulse stimulation." When a multiple pulse stimulation is applied at a short inter-stimulation interval of less than 1 s, reduce the stimulation intensity and pulse duration to 80% of the value shown in the graph.

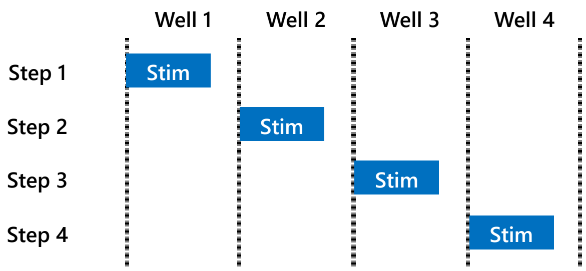


7.2.5. About the stimulus interval

The stimulus intensity and stimulation electrode are set in the Acquire MED64R2 Data w/Stim module panel of the Mobius (for details, refer to the Mobius Tutorial). Please note that in the amplifier, about 5 seconds is necessary to cancel mute at the stimulation electrode (recovery of baseline noise). Therefore, define an interval of not less than 5 seconds between traces.

Stimulation in the MED64-Quad II and the MED64-Allegro

When a stimulation pattern is composed, different steps can be used to vary the stimulation timing for each well and to switch the stimulation sequentially. Define an interval of not less than 5 seconds between traces.



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