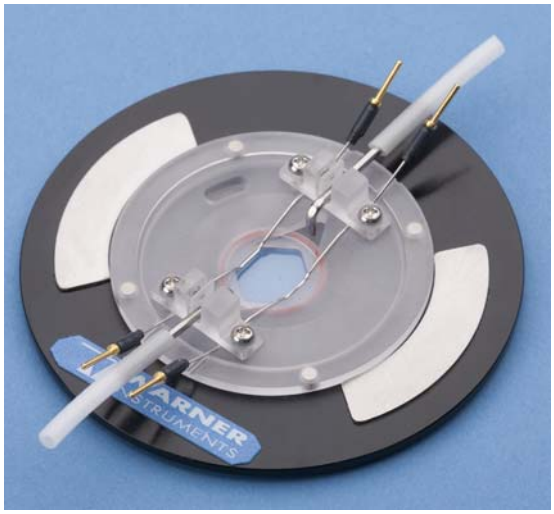
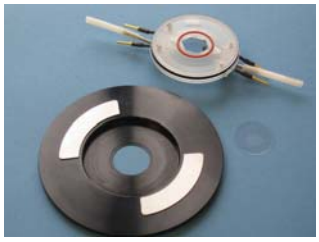
WARNER INSTRUMENTS
A Harvard Apparatus Company

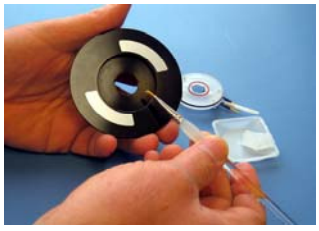
RC-49MFS/RC-49MFSH
Quick-Release Low-Profile Field-Stimulation Chamber
(Heating Elements optional)



The **RC-49MFS** field stimulation perfusion chamber features replaceable platinum stimulating electrodes and a low profile. This configuration provides for maximum flexibility in electrode access to the bath. The chamber uses 18mm-diameter, #1.5-thickness coverslips. In many cases, this same coverslip contains the biological sample. The **RC-49MFS** is a small volume imaging chamber featuring rapid solution exchange, short working distances and an open bath.

Other features include quick and easy magnetic closures for disassembly and assembly and replacement of the coverslip. The design of these **RC-49MFS** chambers incorporates a diamond bath which has been shown to produce a laminar flow across the chamber. Since bath volumes are generally small, exchange times are measured in seconds even when flow rates are less than 1 ml/min. **RC-49MFS** chambers are designed to be used with Warner's Series 20 stage adapters. The **RC-49MFSH** has the same features and is provided with heating elements.





ASSEMBLY

The general procedure for the assembly of the **RC-49MFS** Quick Release Imaging Chamber is to prepare the base to accept a coverslip containing a sample, then to mount the sample and bath ring, in that order. The chamber can then be inserted into a Series 20 stage adapter and further mounted onto your microscope in the usual manner. Prior to beginning assembly make sure all required components are available and thoroughly cleaned.

NOTE: Vacuum grease can be easily applied to the **RC-49MFS** chambers by use of a small, #1 or #2 artist's dotting brush. Brushes can be found in your local art shop, university bookstore, or can be purchased from Warner. (A silicone grease kit available from Warner has all necessary components; KIT-111, #64-0378.)

1. Apply several small patches of a thin layer of vacuum grease around the recess in the base of the chamber bottom.
2. Evenly distribute the grease by placing a spare coverslip into the recess and gently pressing it into position.
3. Remove and discard the spare coverslip. Clean away any grease which may have entered the bath area.

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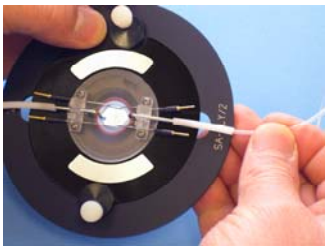
4. Place your sample-containing coverslip, sample side up into the pre-greased recess in the chamber bottom.
5. Complete the formation of the chamber by gently pressing the polycarbonate ring into place over the coverslip-containing chamber base. Be sure that the lower, innermost o-ring seats securely to the coverslip and that the magnets are aligned for maximum clamping.
6. Assemble the **RC-49MFS** into a Series 20 stage adapter, and position the assembly on the microscope stage.
7. The perfusion tubes and platinum stimulating electrodes can be repositioned as needed. Use the provided hex key to loosen the perfusion tube holders and reposition them if desired. The stimulating electrodes can be adjusted or replaced by loosening or removing the mounting screws. Upon reassembly, the stimulating electrodes are affixed into the chamber by tightening the mounting screws.

PERFUSION

Fluid control

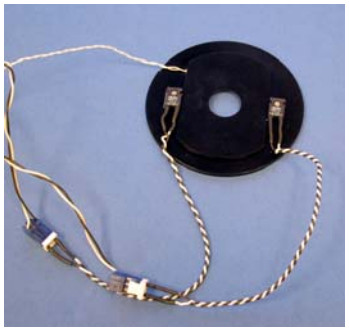


The selection of solution source and rate of delivery can be of either manual or automatic design and is left to the user. However, Warner Instruments manufactures several perfusion control systems (such as the valve-driven VC-8 and VC-8M Control Systems) that can be used with this application. The rate of solution delivery can be established either by pump or gravity feed. While these approaches allow good control of the flow rate, Warner Instruments also offers a dedicated solution flow regulator (FR-50). A reference by Trese Leinders-Zufall describing the advantages of different perfusion control systems is available for download from the Support section of our website (http://www.warneronline.com).

Suction/Level control

Removal of solution from the **RC-49MFS** chamber is performed by aspiration. We recommend the use of a vacuum trap to avoid introduction of aspirant into your house vacuum lines.

CABLE ATTACHMENT



The **CC-28** cable has two blue connectors for attachment to heater blocks, a small red-orange monitor thermistor, and a large white feedback thermistor. The blue connectors make direct attachment to the pins on the associated heater block cable. The monitor thermistor is placed in the bath to monitor the solution temperature at any point of interest (usually in the vicinity the sample). The white feedback thermistor is inserted into the hole in the bottom of the **RC-49MFSH** assembly.

MAINTENANCE

Cleaning of the **RC-49MFS** chamber should be performed using a diluted mild detergent solution. Alternatively, Warner Instruments has developed a trisodium phosphate (TSP) wash protocol which is effective in cleaning plastic parts. Contact our Technical Support staff or download the protocol in PDF format from our website (http://www.warneronline.com). **NOTE**: Do not use alcohol, ether or other solvents on plastic parts.