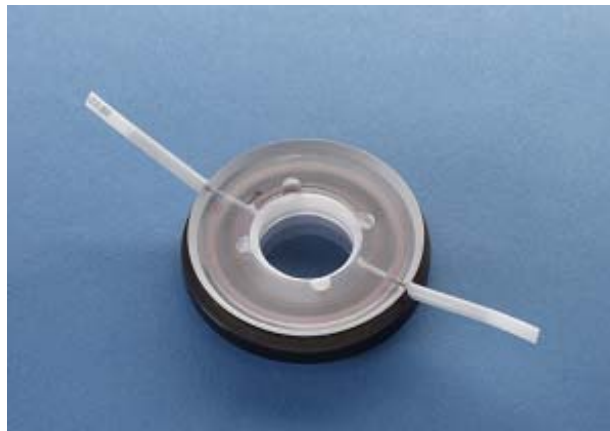


The family of **Series 40** Quick Change Imaging Chambers have several features in common. These include the use of a #1 coverslip to form the floor of the chamber. In most cases, this same coverslip contains the imaging sample. When viewed with inverted microscopes, images are visualized through a single thickness of glass, usually 0.13-0.17 mm.

Other features include quick and easy disassembly and assembly for replacement of coverslips. Since bath volumes are generally small, exchange times are measured in seconds even when flow rates are less than 1 ml/min. **Series 40** Chambers are optimized to be used with Warner's **QE-1** Quick Exchange platform.



THE RC-43C

The **RC-43C** is a medium volume imaging chamber featuring rapid solution exchange, short working distances and a closed bath. This chamber is designed to be inserted into Warner's **QE-1** allowing a variety of assays to be quickly performed on cultured cells. The **RC-43C** features a 213 μ l working volume with a 15.8 mm aperture on the chamber bottom. This chamber accommodates a 25 mm #1 coverslip. The top of the chamber also has a 15.8 mm aperture and accepts a 18 mm coverslip. The working distance between the top and bottom coverslips is 1 mm.



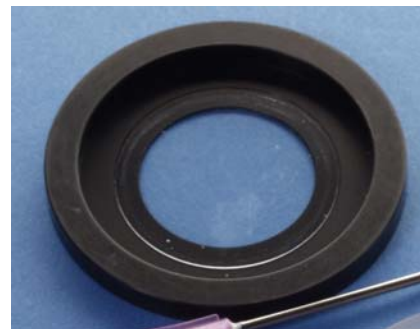
The **RC-43C** is shipped with an anodized base, the chamber body and a retaining ring to hold the top coverslip in place. Also included are 10 ft of **PE-160** tubing, 1 ft of **PE-90** tubing and 100 each of 25 and 18 mm round #1 coverslips. The package is rounded out by the inclusion of two (2) replacement gaskets and two (2) tools for use in removing the retaining ring from the chamber body.

ASSEMBLY

The general procedure for the assembly of the **RC-43C** is to first prepare the base to accept a sample containing coverslip, then to mount the bottom coverslip and chamber body, in that order. The top coverslip is then inserted and the retaining ring is secured into place. The assembled chamber can then be placed into the **QE-1** and mounted onto your microscope in the usual manner. Prior to beginning assembly make sure all required components are available and thoroughly cleaned.

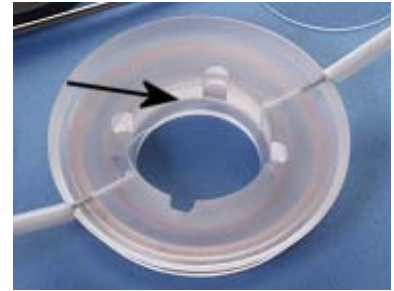
1. Apply a small layer of vacuum grease around the recess in the bottom of the base.

NOTE: Vacuum grease can be most simply applied 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 is also available. (Catalog # 64-0378.)

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 around the periphery of the recess..
4. Place your sample containing coverslip, sample side up, into the pre-greased recess in the base.
5. Examine the polycarbonate chamber body to verify that the two gaskets are in place.
6. Paint a small amount of vacuum grease to the upper side of the shelf in the chamber body. (See arrow in photo to the right.) Insert and remove a clean 18 mm round coverslip into the recess and clean up any excess grease from the seating.
7. Gently press the chamber body into place in the coverslip containing base. Be sure that the lower, innermost o-ring seats securely to the bottom coverslip.
8. Place a new, clean 18 mm round coverslip into the recess and secure it into place using the round retaining ring.
9. The completed assembly can then be placed into Warner's **QE-1** and mounted onto your microscope.



PERFUSION

Attachment of fluid lines

Input and output perfusion lines are attached to the **RC-43C** via 20 gauge stainless steel perfusion ports.

A short length of **PE-90** tubing is supplied to facilitate coupling of the **PE-160** perfusion tubing to the chamber. Cut a short length of **PE-90** (approximately $\frac{3}{4}$ of an inch is sufficient) and attach to the stainless steel ports. The OD of the **PE-90** conveniently matches the ID of the **PE-160** to make a leak-free seal.

NOTE: We recommend pre-filling all perfusion lines before connection to reduce the occurrence of bubbles in the bath.

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**) all of which 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 Flow Regulator**). 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/techref.html>).

Multiple perfusion solutions

Warner Instruments multi-port manifolds (**MM** or **ML Series**) can be used to connect up to 8 solution lines to the **Series 40** chamber. Connect the manifold output tube to the input port feeding the chamber. Air should be removed from each feed line by pre-filling with its appropriate solution. We recommend making the connection between the manifold and chamber as short as possible to minimize exchange times.

MAINTENANCE

Cleaning of the **Series 40** chamber should be performed using a dilute 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.