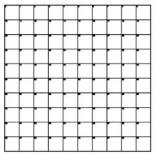
Correlative Microscopy Coverslips

A unique film reticle for use in Correlative Microscopy. Designed specifically to allow identification and location of a particular area of interest under brightfield or fluorescence microscopes and then sectioning for electron microscopy.

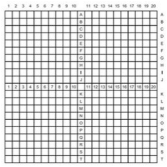
All Correlative Microscopy Coverslips are produced on a polymer based film, 22 x 22 x 0.18mm thick. 25pcs per box. Other patterns, grid sizes and film sizes are available to special order.

CMC35



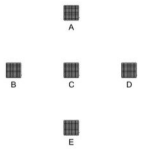
Order Code: 01ACMC35
10 x 10 grid of 1mm squares, each square individually identified 0 to 99

CMC71



Order Code: 01ACMC71
20 x 20 grid of 0.5mm squares, indexed 1-20 along top, A-T down the side and on central cross

CMC34A



Order Code: 01ACMC34A
10 x 10 grid of 0.1mm squares at 5 positions identified as A-E, Indexed 1-10 along top. A-J down the side

CMC1

Order Code: 01ACMC01
10mm scale in 0.1mm divisions

Physical and Chemical Characteristics

- Resistant to normal chemicals used in electron microscopy
- No oxygen retention and compatible resin LR White
- Good optical quality in brightfield & UV fluorescence
- Excellent transparency
- Does not deform at temperatures (positive 100°C and negative liquid N2)
- Rigid, does not float in the middle of culture
- Easy to handle and cut with a knife or micro-punch
- Simple sterilisation using alcohol or UV
- Detaches easily from resin after polymerisation
- Low cost

How to use the Correlative Microscopy Coverslips

1- Sterilise the coverslip with alcohol, then dry and add the culture.

2- Ensure that the grid is positioned correctly so that the text is readable.

3- Observe your cell culture using light microscopy (transmitted and/or fluorescence) and identify the area of interest (Fig. 1 and 2).

4- Record the images needed, and note the co-ordinates of the squares where there are cells of interest (Fig. 1 and 2 show co-ordinate 8C).

5- Fix, dehydrate and embed with resin for examination by transmission electron microscopy.

6- At the end of the embedding procedure, invert a BEEM type capsule filled with resin onto the coverslip covering the selected cells of interest (Fig. 3).

7- Cure and detach the coverslip (Fig. 4), the footprint of the grid (Fig. 5) allows location of the position. Trim the block (Fig. 6) in the selected area then make cuts using an ultra microtome.

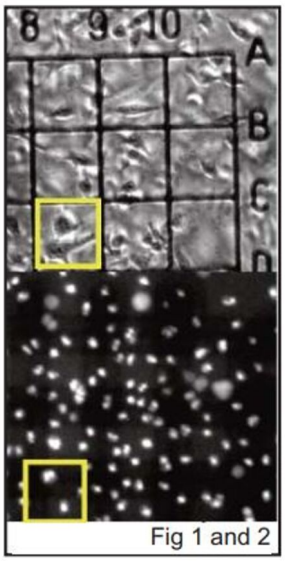


Fig 1 and 2


Fig 3

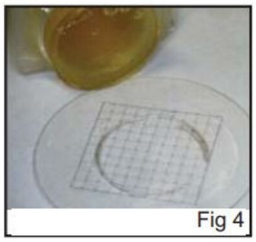


Fig 4

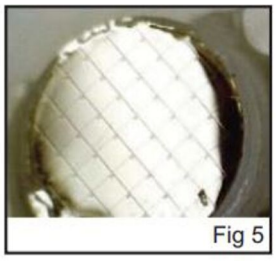


Fig 5

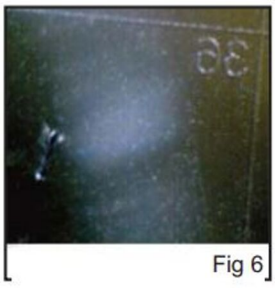


Fig 6

Punching the area of interest for cryofixation

Technical Advise

- Use with Fluorescent Microscopes. These new correlative microscopy coverslips (CMC's) are not designed to replace the glass or quartz coverslips,which have superior optical properties, and so are far better for fluorescent microscopy. The film itself has a structure which is visible under certain light conditions, especially with fluorescence. The CMC's are designed to satisfy the needs of correlative microscopy where an initial, general analysis is performed using light or fluorescent microscopy and then the specimen is further processed for analysis by SEM, TEM or cryofixation.

  If you need to perform detailed analysis using fluorescent microscopy then the CMC's are not the solution. If however you want to perform a comprehensive analysis using different analysis techniques then the CMC's are the only solution. The key advantage of the CMC's is that they are made on special film that can be cut or punched (for cryo applications), something that is not possible with glass coverslips. The grid image is also transferred to any embedded specimen making cell location far easier.

- Best Types of Resin to Use. Because this is new technology it often requires a change in the preparation method. The method that technicians have traditionally used for glass coverslips may not be suitable for these film ones. The key thing to successful use of these CMC's seems to be the type of resin and the preparation. We have already mentioned about the need to ensure a hermetic seal between the Beem capsule and the CMC film. The preparation works perfectly with low viscosity epoxy resins (Spurr, EPON). Sufficient polymerisation is obtained in 24 hours at 60 degrees Celsius – it does not normally require 48 hours.

- Cleaning Procedure. Always clean the coverslips before applying the specimen. This can be done either with UV light or alcohol. After using alcohol it is best to immerse the coverslip in polysyline to give better adhesion.

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