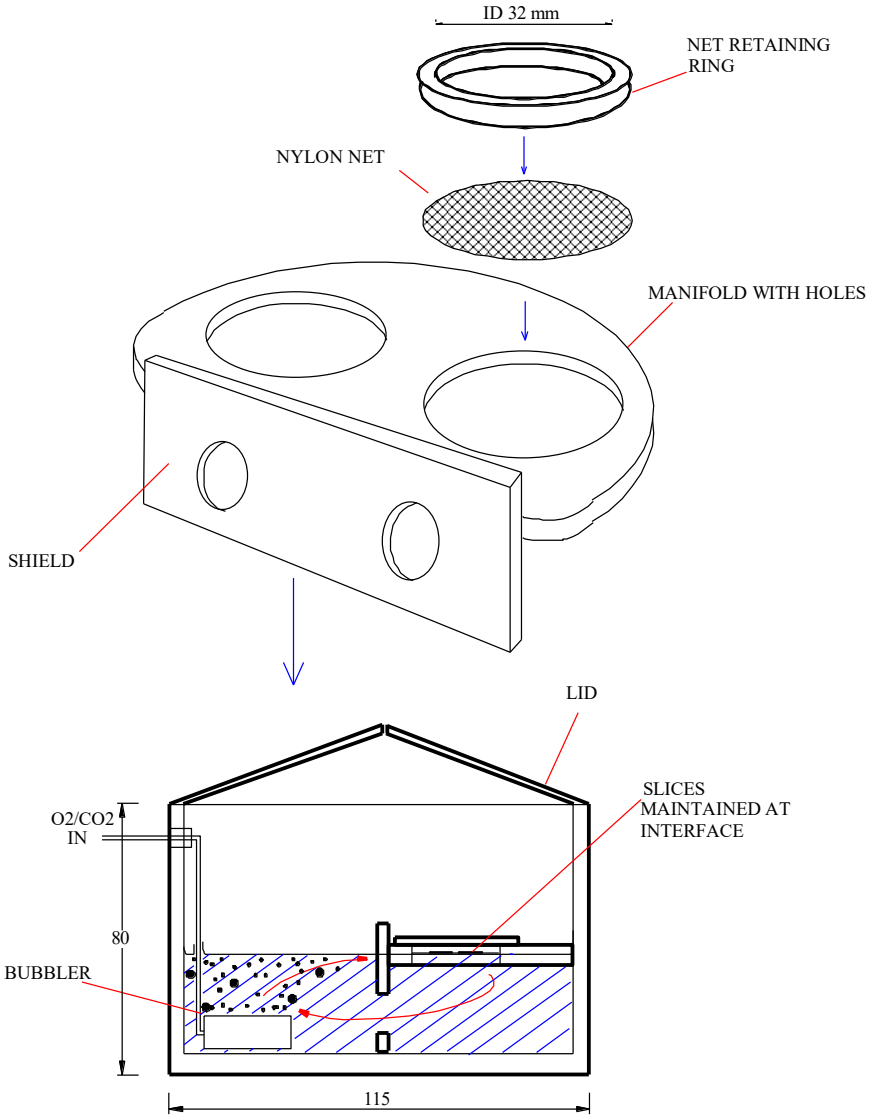

Schematic arrangement of BSK5-2 and location of manifold in purpose designed trough with conical profiled lid. Red arrows indicate circulation from the top corners and below with return towards bubbler. Approximate dimensions are in mm.

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Innovative Engineering for Science

BRAIN SLICE KEEPER

BSK5-2

BRAIN SLICE KEEPER

CAUTION !

YOUR BRAIN SLICE KEEPER IS A PRECISION ENGINEERED TOOL FOR SCIENTIFIC RESEARCH. PLEASE TAKE A FEW MINUTES TO FAMILIARISE YOURSELF WITH THE KEEPER AND READ THROUGH THIS SHORT MANUAL BEFORE ATTEMPTING TO USE IT.

DO NOT USE ALCOHOL OR SIMILAR SOLVENTS IN ANY CONCENTRATION ON ANY PART OF THE KEEPER SINCE AS WITH MOST ACRYLICS, IT MAY FRAGMENT OR DEVELOP HAIR-LINE CRACKS. DO NOT AUTOCLAVE AS HEAT CYCLING CAN PRODUCE STRESS CRACKS.

OPERATION

The BSK5-2 Brain Slice Keeper has been designed to pre-incubate a large number of brain slices prior to transfer into recording chambers. Based on the BSK4, it consists of two closely fitting acrylic rings, located in a half cylinder manifold. The close fitting rings allow a removable sheet of nylon netting to be wedged and held in place. In this slice keeper, the slices are maintained at an interface between the incubation medium and a humidified, high oxygen atmosphere within the holding vessel. Furthermore the slices can be maintained on a flat porous membrane (eg *Millicell-cm, 0.4um Cat.# PICMORG50 from Millipore) attached to the nylon netting in order to maintain the underside of slices perfectly flat for subsequent experiments utilizing microscope techniques.

FEATURES

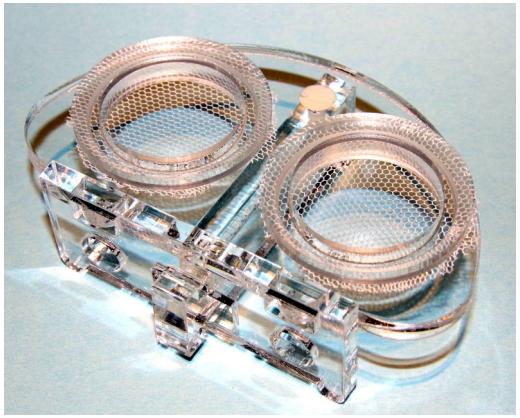
** Slices maintained for many hours at an interface with incubating media*
** Two separate rings allow separation of different types of preparations*
** Slices supported on a quick change nylon net with optional attached porous membrane*
** Modular design simple to set up and maintain*

In operation, the manifold carrying two rings are located in the base of the holding vessel and are partially immersed with ACSF by filling to the level of the netting. The purpose-designed holding vessel is supplied with a 95% O2, 5% CO2 gas mixture via an air-stone bubbler. The air-stone is located to the side of the manifold. The bubbles rising from the base saturate the ACSF and provide constant circulation of medium to the slices from the underside by causing a flow from the top edges towards the underside and out through port holes in the base of a shield. Typically slices are placed on lens tissue which rests on the nylon nets or optionally on porous membrane discs attached to the netting. Slices remain viable for many hours in these conditions. The BSK5-2 together with the holding vessel can be easily placed into a water bath for regulating the incubating temperature as desired. A lid with a conical profile ensures that condensation drops do not fall directly on top of the slices. The typical fluid volume with BSK5-2 is around 250ml.

TEMPERATURE CONTROL

Temperature can be maintained by submerging the trough into a water bath set to the desired temperature. The heated water bath fill level should be about 15mm below the fill level of the BSK5 to prevent floating and instability. Bubbling within the BSK5 ensures circulation and uniform temperature of the incubating media around the slices.

Above: BSK4 Brain slice keeper with rings and net in place and with lid in place (inset). A needle valve is used to control the flow of oxygen mixture to the ceramic air-stone. Below: View of two-ring manifold.



MAINTENANCE

Alcohol should never be used on the acrylic manifold and rings of the slice keeper for cleaning purposes even at low concentrations because it de-hydrates and produces hair-line cracks in acrylic plastics. Only the plastic trough can be cleaned with alcohols. The acrylic component can be cleaned with special laboratory detergents such as *Micro-90™ which completely rinses out. The ceramic air stone must be removed when using such detergents. Heavy deposits of salts should be washed out with distilled water overnight and carbonate salts treated with mild acids such as citric acid. The most common contaminant is fungal growth, this can be avoided by washing out with distilled water and drying out completely at the end of each day. Hydrogen peroxide solution 30 Vols, 1/10 dilution is also an effective cleaning agent followed by overnight soaking in distilled water.