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Instructions

KCl Bridge Electrode Holder

The KCl Bridge Electrode holder is just like a standard holder with an extra section added in the middle. This new section links the amplifier input with the section that actually holds the electrode. The middle section holds a small amount of KCl solution where it can form a circuit with the silver chloride pellet and the quartz capillary tube.

The front part of the holder features the usual silicone seal for the electrode. The glass electrode is inserted in this part until it can go no further. Then the securing nut is twisted to form a tight seal. On patch electrodes this section will feature a port for attachment of a suction tube.

Where the front section of the holder meets the middle section, there is another thread that connects the two parts. Here a silicone seal is compressed against a black plastic backing plate. At the center of the seal and the plate is a tiny hole to accommodate the quartz capillary tube. The capillary is pushed through this hole so that its end is securely seated in the middle compartment of the pipette holder. Tightening the front section into the middle section compresses the rubber seal on the quartz capillary and holds it in place.

The quartz capillary is the conduit down into the recording pipette. The capillaries supplied are 100µm and 200µm ID. Try to use the 200µm capillary if you can since it is better (if your pipette is big enough), otherwise the 100µm can be used. The 200µm ID capillary should have a 100µm silver wire (supplied) inserted to lower resistance. The silver wire must be chlorided and should be cut just a little shorter than the tubing and sit in the tubing so that the wire does not protrude from either end. The 100µm ID tubing is used for patch recording and the 200µm ID tubing is used for whole cell recording. In either case, the capillary is filled with warm KCl by using the luer-lock compression fitting (supplied) which must be mounted on a syringe. The quartz capillary is inserted into the compression fitting, the

fitting is tightened, and the KCl is injected into the quartz capillary until small droplet forms at the open end. (1 molar KCl is recommended.)

Once the capillary is full it can be placed in the holder as described above. The capillary can also be filled if it is installed in the holder properly, and if the KCl reservoir is filled, by simply applying pressure to the KCl filling port on the middle section of the pipette holder. Pressure should be applied until a small droplet forms on the end of the capillary tube. (Remove this droplet before installing your recording pipette.). Do not insert the quartz capillary into your recording pipette if it is dripping. You do not want to push KCl into your recording pipette so be careful when tightening fittings on the holder that might increase pressure and push KCl out. After adjustments, wait until the unit stabilizes before use. In either filling procedure, if the capillary does not fill, it is most likely clogged and in need of replacement.

The middle section of the pipette holder, where the KCl is stored, is filled by removing the red cap and depositing the KCl with a needle. The section should always be full and the cap should always be in place when in use. This section should be filled before the quartz capillary is inserted.

It is very important to prevent air bubbles from entering the circuit pathway. This means that precautions must be taken to keep air bubbles from forming in the quartz capillary tube. The best way to do that is to fill it with warm KCl. On a daily basis one can open the filling port on the KCl module, and inject some warm KCl into the quartz capillary from the front end using the luer compression fitting and a syringe. Close the filling port and the holder is ready for the pipette.

Never open up the back section of the holder unless you need to change the silver chloride pellet. Replacing this section is tricky since the short silver wire of the pellet must be inserted through the back of the middle section and then into the connector section where it must pass through another seal. If you need to disassemble this part for any reason, observe how it is set up as you take it apart. Simply put it back the same way. The use of forceps is recommended.

Things to note about the use of the KCl bridge electrode holder:

The KCl Bridge electrode holder allows the user to record from cells without the need for chloride containing solutions in their recording pipette. KCl will be in contact with the recording solution via the quartz capillary tube. Since the tube has such a small ID, very little KCl will diffuse out even over several hours. Also, the small ID of the tube creates a wire with an OD of 0.1mm which is less than ½ the diameter of most silver wires. The noise that a wire adds to a recording is proportional to the Log of its OD, so a smaller wire will give a quieter recording. In addition, there is no need to fill the recording pipette all the way to the back. It only needs to be filled up enough to make contact with the quartz capillary—further lowering noise! After usage, the holder does not have to be emptied, but it should be stored with the tip of the quartz capillary submerged in KCl so crystals don't form in the quartz capillary tube. So say good-by to baseline shift and enjoy the next generation of pipette holder for electrophysiology!

For reference please see: Snyder, Kriegstein and Sachs, A convenient electrode holder for glass pipettes to stabilize electrode potentials, Pflugers Arch. 438: 405-411 (1999)

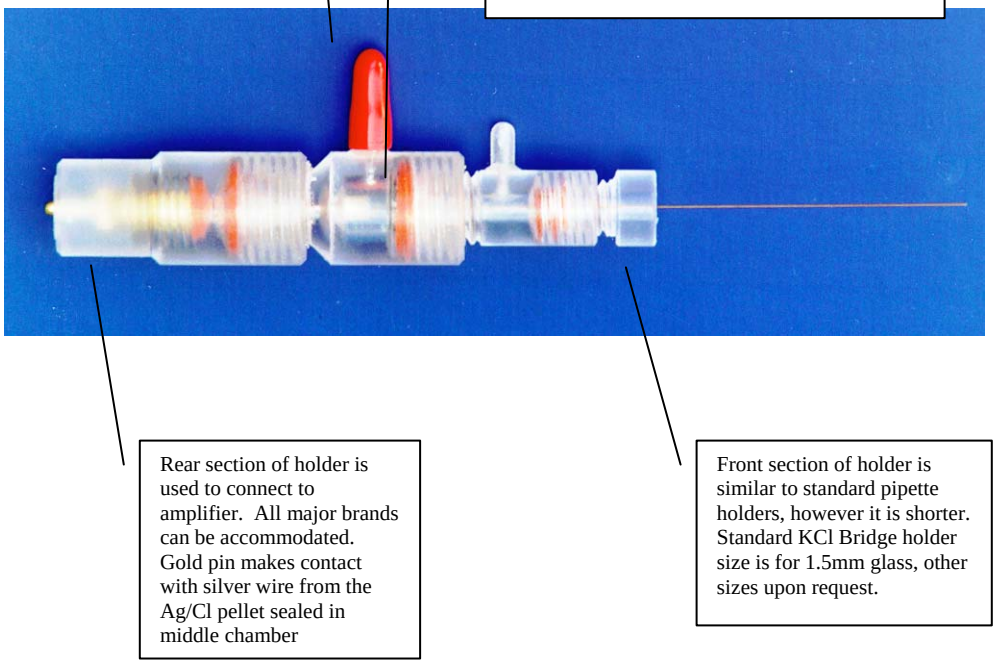
Cleaning the Holder

The holder should be cleaned every few weeks. If excessive noise is seen, then the holder should be emptied, disassembled, flushed out with distilled water followed by methanol and then dried thoroughly. Again, do not remove the silver chloride pellet or open this section, unless the pellet or wire needs replacement.

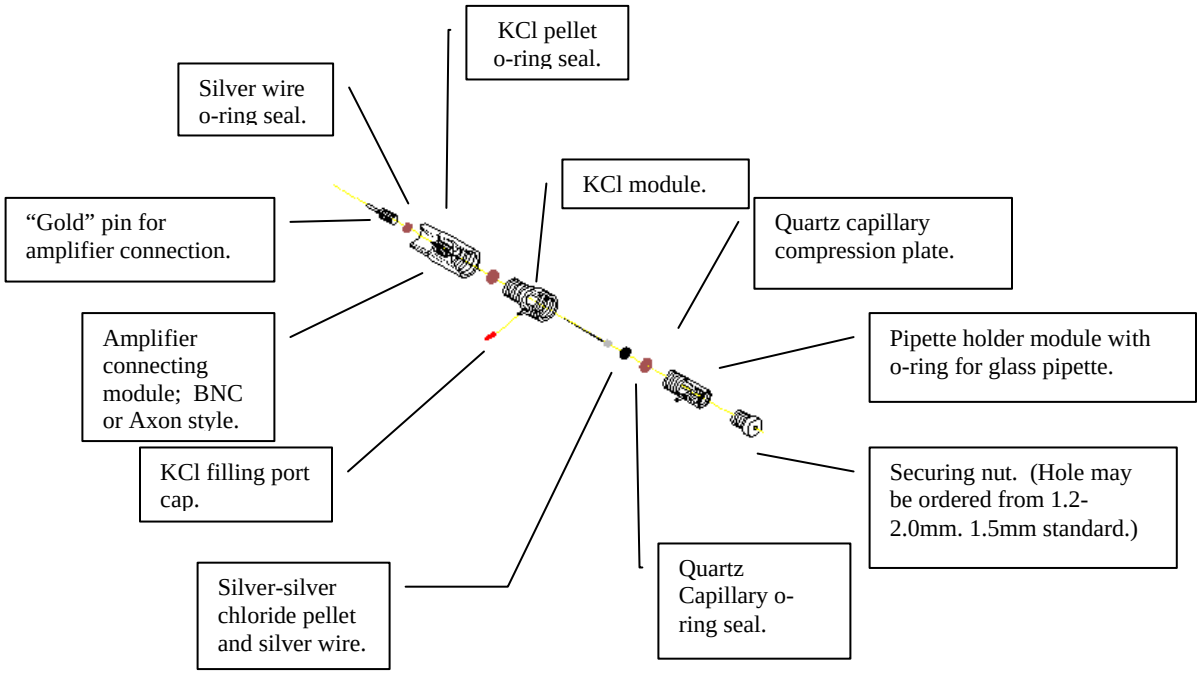
Storage for use

When the holder is being used for experiments, and it is filled with KCl with the quartz capillary installed, it is important not to let the capillary dry out. If the KCl dries out then the capillary will become clogged and will need to be replaced. The best thing to do is store the holder overnight in a way that the tip of the KCl filled capillary is submerged in KCl solution. Don't forget each day to replace the KCl in the quartz capillary with fresh warm KCl to prevent air bubbles!

KCl Bridge Pipette Holder



Assembly Diagram of KCl Bridge Pipette Holder



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