MICROSCOPY 101

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Glass Knives verses Diamond Knives

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In regards to the purchase of a histology diamond knife for preparing light microscopy sections, I was one of those folks, in possession of a nice glass knife maker, who was very comfortable with breaking, making and using glass knives. So the question was—did I really need a costly diamond knife for making light microscopy sections?

Once I finally tried a diamond histo knife, I never went back to glass. There are some very good incentives for this: long lasting cutting surface, much time saved in sample preparation, larger block faces can be prepared, and more consistent cutting results obtained. A histo diamond knife is also a big aid in preparing specimens for diagnostic or biological microscopy. These very important bonuses make a good list to have in hand when justifying the purchase to your organization. However, are they worth the price? Yes, I think so—even so far as providing each tech with their own diamond histo knife.

The knife-edge can last a long time, though all of us know that with certain materials, or an accident, the edge of a diamond knife can have an incredibly short life span. With glass, one might get from zero to 10 or 12 good sections from small block faces. With a diamond knife the specimen collection boat can be filled many times over, and one can even slowly work through an entire specimen. As with thin sectioning for the TEM, particles made of glass, silicone, bone etc, can scratch a diamond knife easily. Often we use an older, already scratched, knife for such work. However, for many biological samples, the knife-edge of a diamond histology knife can cut many sections from hundreds or even thousands of blocks.

To extend the life of a histo-diamond edge, one can use old, thin-sectioning diamond knives or eventually, old diamond histology knives, for first facing a trimmed block and then swapping in the good diamond histology knife for the final cutting. One learns quickly how to correct for any slight change of cutting angle and how to realign the new knife for any changes between the two knives. Good cleaning of the edge can also prolong its life. I use water and a styrofoam stick after each use, and with a deep sink the knife can be held at a distance for a little bit of controlled water force to the edge. The knife is then dried with a filtered air stream from the room's "house" air outlet, so that nothing dries down in a water ring on the edge.

With a "thick section" work load of around 500-600 blocks peryear, consisting of samples of skin, nasal biopsies, and pelleted cell cultures, as well as "safe" soft tissue, the knives have a good lifetime. One knife I finally stopped using lasted seven years—this exceeds the useful life for an average knife but, with care, it is possible for knives to last that long. This knife now, at last, acts as a facer knife for my new 14-month-old diamond knife. The seemingly large price of a diamond histo knife, now looks much more affordable. By prefacing the block face with an old diamond knife, one can find the hidden surprises in a block, which may damage your knife if they are not caught during hand trimming. In blocks with potentially damaging inclusions in them, a lower ranking, or less well-conditioned knife,

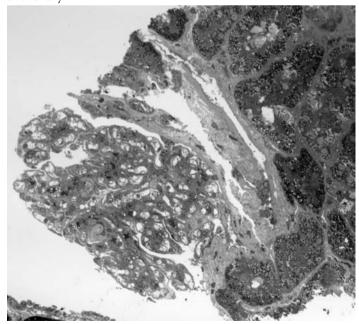
can be used to save your good histo knife.

Time is saved in that your knife is always ready to use, no knife making is needed, and one knife-edge for the final cuts is all that is needed. The knife surface is straight; so far, less continuous facing of the block is needed. Glass knives often have a slight curve to them, moving along the knife-edge as it wears out, often incurs the cost of having to take extra sections into the block to reface it. A good reminder in histo knife purchasing, is that the money saved is not only in glass, and glass breakers, but mostly in technologist time over a very long period of time. The histo knife suddenly seems very affordable.

Being able to work with larger block faces helps greatly, especially in diagnostic work. A needle kidney biopsy is small, but the sample can be cut lengthwise as well as along the long axis, this exposes multiple larger surface areas to catch glomeruli when they are rare and one can have as many as four to seven of these surfaces in one single block that has been embedded with a flat face. This gives more viewing with only one or two blocks to cut and speeds up the sample turnaround time as well as increasing the viewing area, as long as one can keep the face to the size of the thin sectioning diamond knife. With a straight surface, there is far less risk of losing these precious tissue fragments.

Because of the straight edge, and long term using of the knife, results that are more consistent can be obtained. The edge remains constant in quality for some time unless damaged, and the operator can learn and remember or chart where the usage flaws appear. Because the edge is straight, less slicing needs to be done. The thickness of the sections can be less than with glass knives, our lab sections at 0.33 μ m, though some labs have good results sectioning down to 1 μ m for light microscopy viewing. Thinner sections are good for resolution, can create less wear, and can preserve the presence of depth sensitive and small samples for future sectioning.

Therefore, if you are on the verge of considering a diamond histo knife, and the money is there, a diamond knife is not so much of a luxury



Human kidney glomerulus 20x magnification, 0.35µm thin. Representing a small portion of 1 of the 5 pieces of tissue in one block face and section. Tissue is stained with Toluidine Blue and Basic Fuchsin.



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