H0W TO MAKE EASILY VEGETAL SLICES

This may not necessarily be the first activity to do when starting on microscopy, because, traditionally, you must thave a microtome. The simplest model is the Ranvier hand microtome but the success of the results depends heavily on the razor, the medium used to hold the object to be cut (elderberry pith or, more modern, high density polystyrene) ... and also your ability! But there is another simple and inexpensive technique,

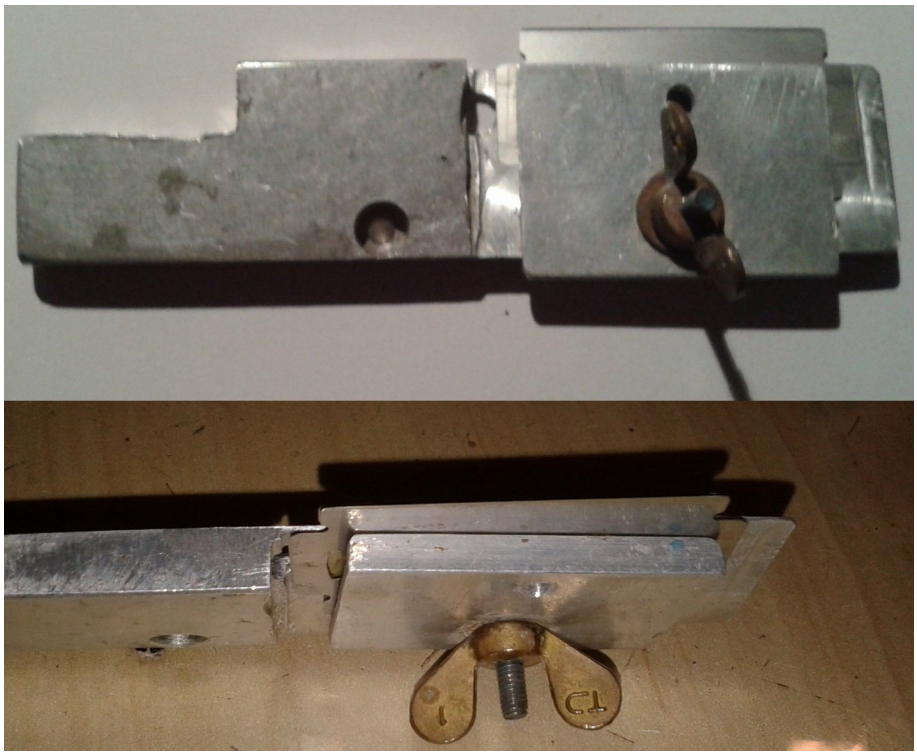
Re-reading old articles (written 17 years ago!) from *Micscape* I was pleased to see those of our late friend Walter Dioni. Here are the links :

www.microscopy-uk.org.uk/mag/artapr04/wdslicera.html

www.microscopy-uk.org.uk/mag/artapr04/wdslicerb.html

Walter described a simple way to make vegetal slices: a tool called "double razor blade" or "mesotome". I will therefore not repeat the description of the device, very well detailed by Walter in these two articles with his remarks on its optimization. To pay tribute to him, because he spent a lot of time developing simple techniques, please go read the aforementioned articles but also his other articles (fixing methods, coloring,….)!

So, at the time, I had made a variant of the tool to apply this principle:



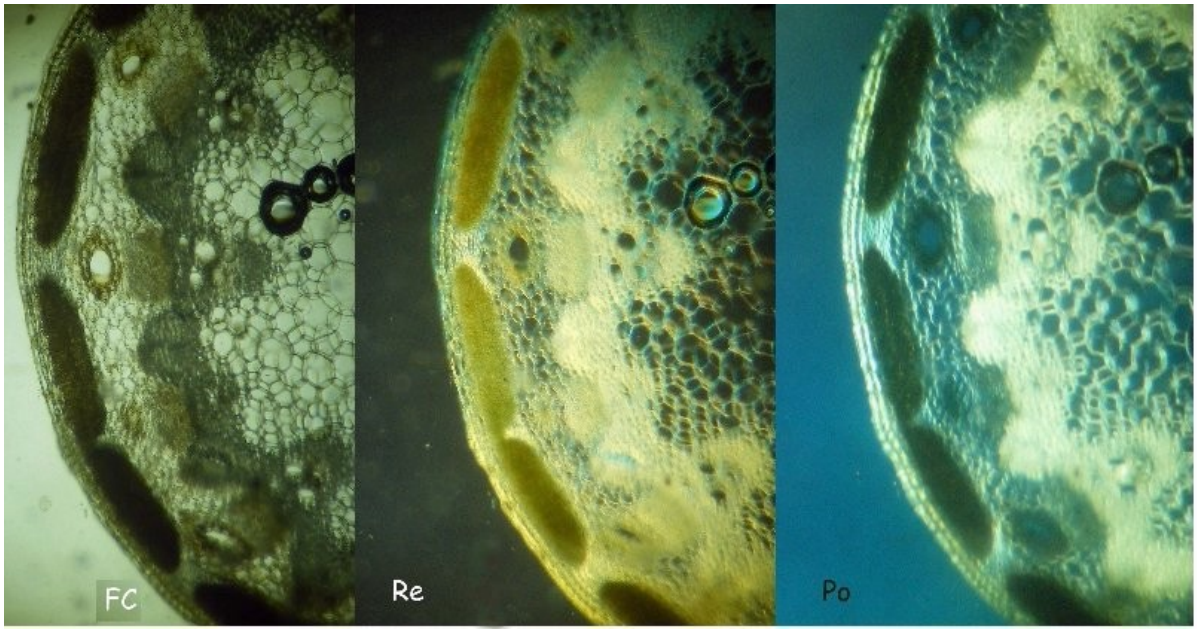
Here are the parts of the tool, on the right, one of the two blades with two layers of adhesive tape which determine the thickness of the cuts (only one blade is equipped as well).

Note the plate "P" carrying an edge "r" which serves to apply pressure on the blades at the level of this shim. This arrangement firmly holds the blades and prevents them from spreading apart during the cutting effort. In this configuration, the thickness of the sections will be between 0.2 and 0.3 mm, which is sufficient to allow the lighting to pass through. Very important: See in Walter's articles how to cut underwater and on a semi-hard support (polystyrene).

I did not have any patience to process the slices in the traditional way (removal of cell content with a chlorine product, specific stains, etc.) but even with this rapid method the results are interesting and the natural color retained.

Regarding "coloring", various modes of illumination of sections can be invaluable in highlighting specific structures ... (dark field, Rheinberg, polarization ...). I will not comment on the identification of structures, but readers can have fun taking the images and documenting them! !

Since we are talking about lighting, here is a first example on a plant stem very abundant at the edges of the roads in this summer period, namely the fennel, easy to recognize by the aniseed smell when the leaves are crumpled: (FC: Brightfield, RE: Rheinberg, PO: polarization) Below is the disk I have used.



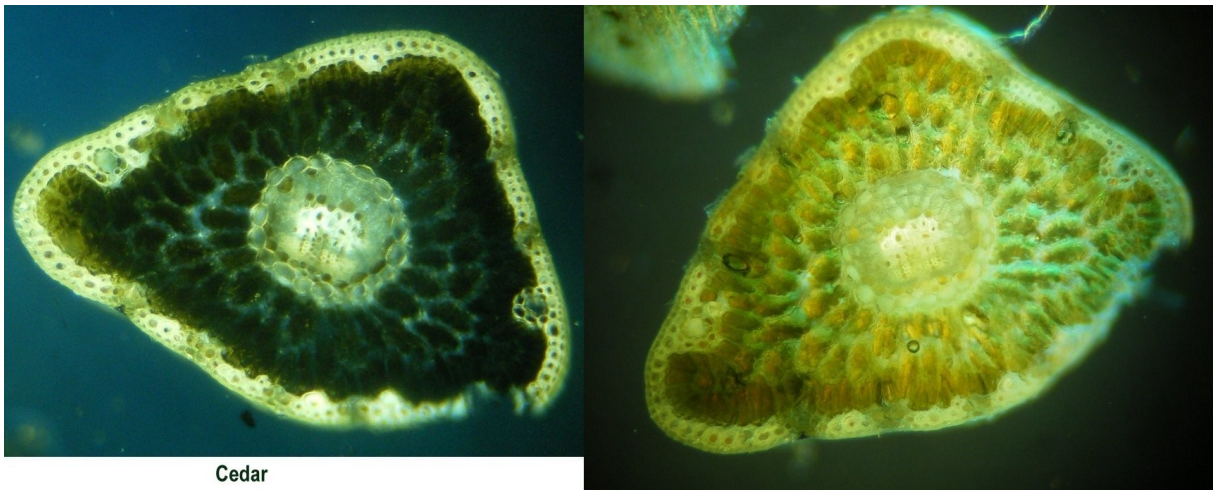
Another plant that can easily be found on the edges of beaches and dunes: oatmeal. The leaf is well adapted to drought and closes on itself in the event of a lack of water to limit dehydration:

Colored image on the right (neutral red, Rheinberg lighting) and on the right in bright field: note the smiling "face" at the top of the right image which is the endoderm with two bubbles trapped in the holes, simulating eyes!

It is not the same leaf in the two images

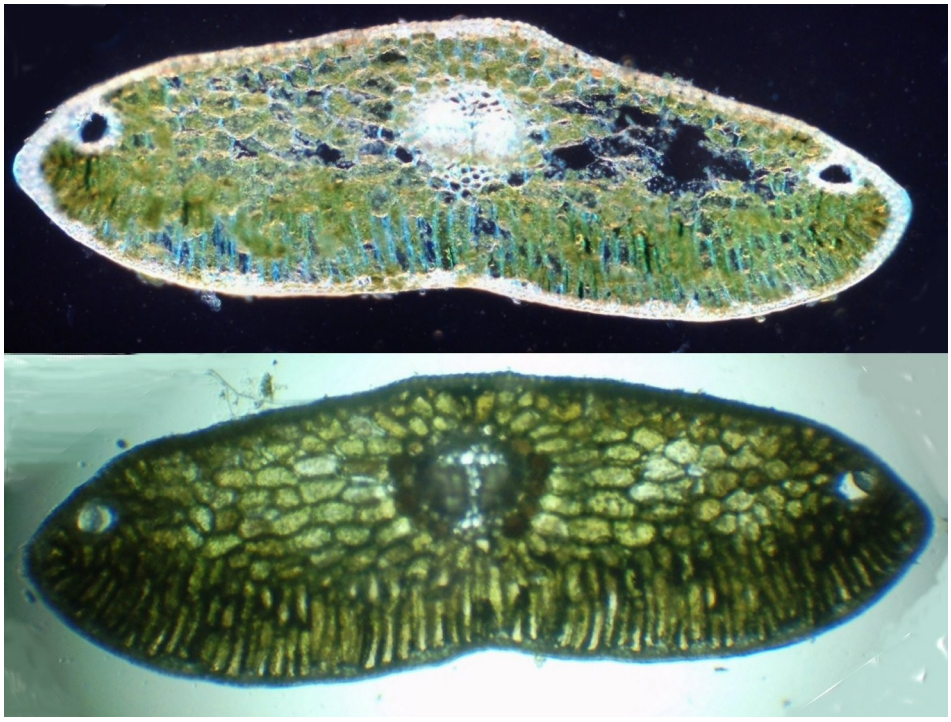
The leaves (needles) of conifers are a good subject because their consistency neither too hard nor too soft, allows easy cuts.

Cedar for example: image in polarization on the left and Rheinberg/dark field on the right: (I find that the dark field gives good results on these types of slices).



Cedar

Spruce: dark field image above, bright field below.

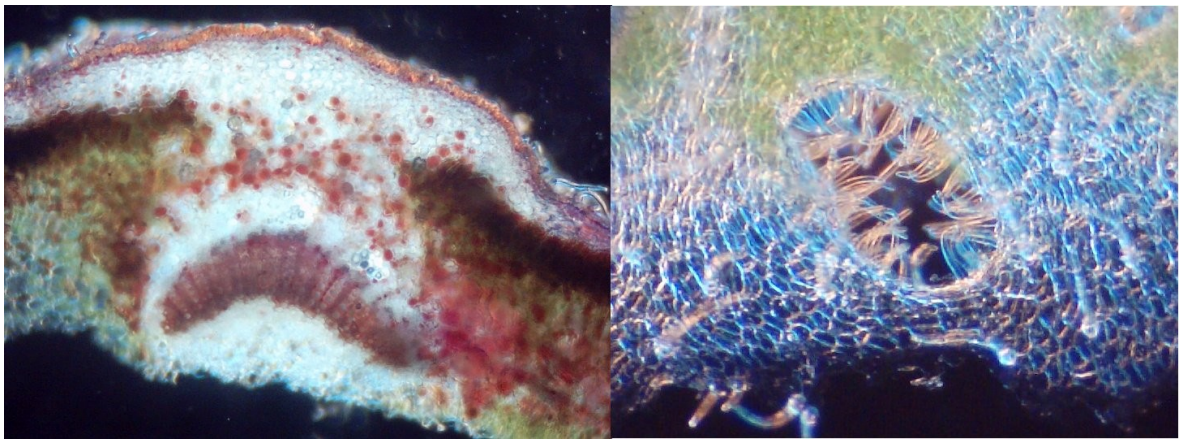


Pine: colored cross section on the right, on the left, cut is too thick, but part of the epidermis has been torn off, so you can see the stomata lined up!

Pine needle

Of course you can cut anything you find!

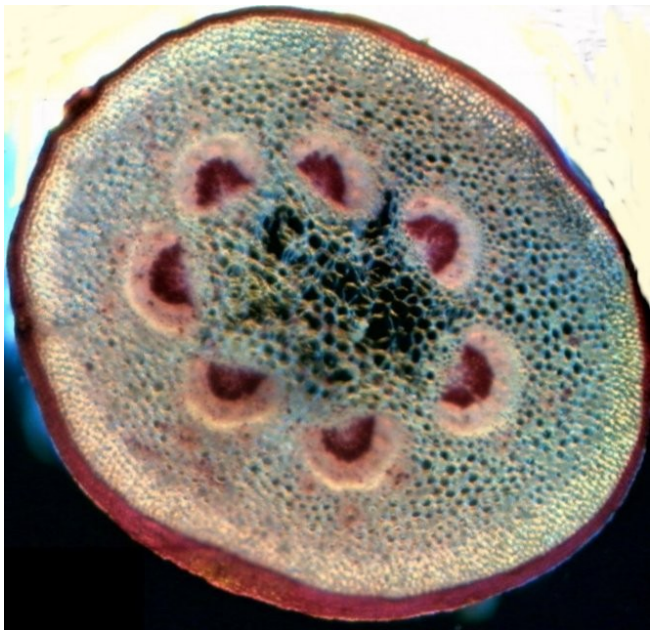
Section of oleander leaf vein in the center of the leaf: right image showing one of the hairy cracks in the epidermis ... (Rheinberg lighting).
.



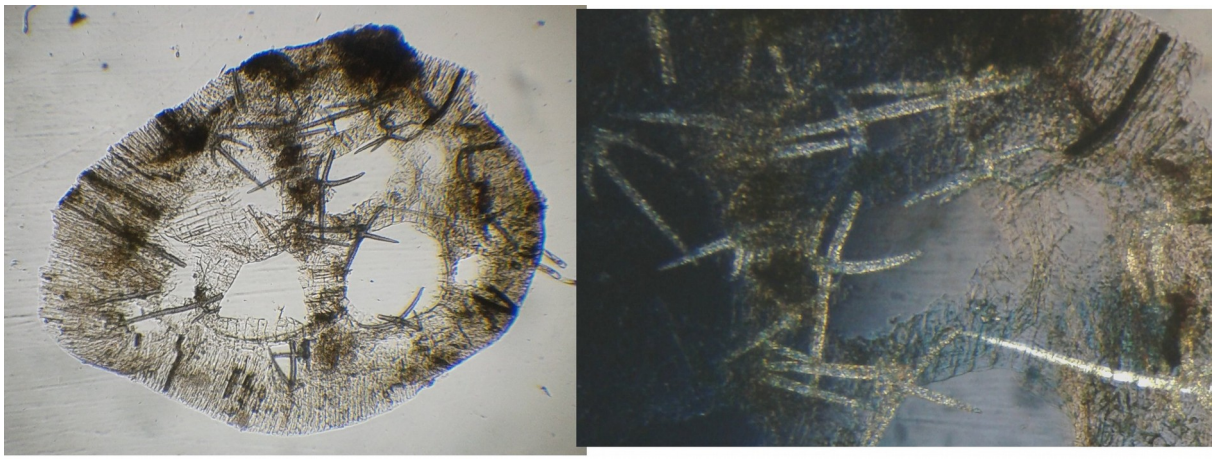
Lavender leaf (left) and stem (right) Dark field and polarization.

Lime tree little branch:

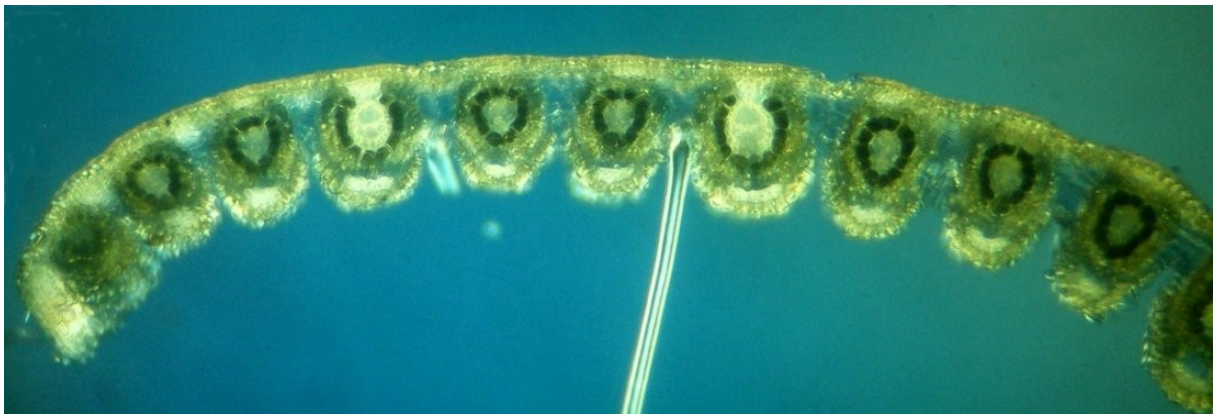
How about a cherry tail cut (colored neutral red)?



Water lily stem: not very convenient because soft stem crushes during cutting: we see the air-filled gaps which allow flotation; the right image, with x6.3 objective, shows the sclerites in polarization.



And to finish a cut of "idontknowthatis" from a very thin leaf but with interesting details (in polarization), note the hair in the center



To conclude: This technique makes it possible to obtain very honorable results with a minimal investment (especially in time!) ... Compare this with traditional processes including: fixative, inclusion, cutting, coloring ..., but slices are less thin than using a classical microtome.

Of course, you can use the most beautiful sections and to dissolve the cellular content and use specific stains to characterize the structures.

One last tip, the choice of blades is important, some wear out faster than others ... You can make about ten cuts before the edge becomes dull; all you have to do is turn over the 2 blades (the unused side - hidden under the presser - becomes the useful side) then replace the two blades when they no longer cut!

Jean-Marie Cavanihac, France, email – micromars AT orange DOT fr

Published in the October 2021 edition of *Micscape* magazine.

www.micscape.org