

## From the photography to the digitalized outline suitable for MORPHOMATICA

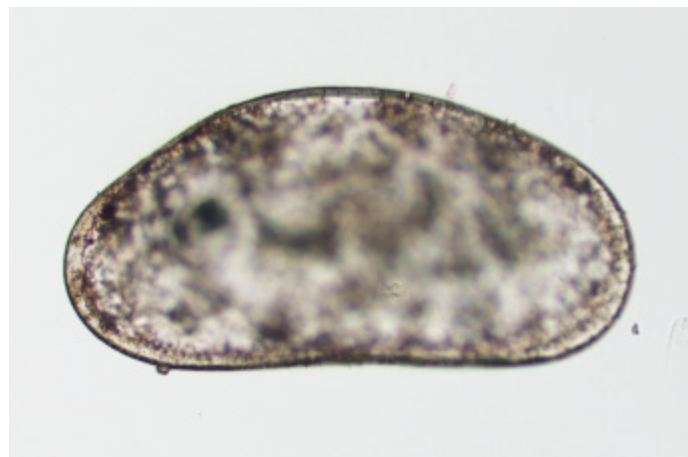
Anika Stracke<sup>1</sup>

<sup>1</sup>Heinrichstrasse 55, A-8010 Graz (E-Mail: anika\_boriss@yahoo.com).

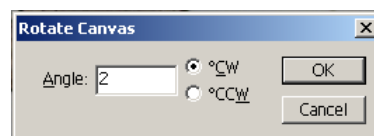
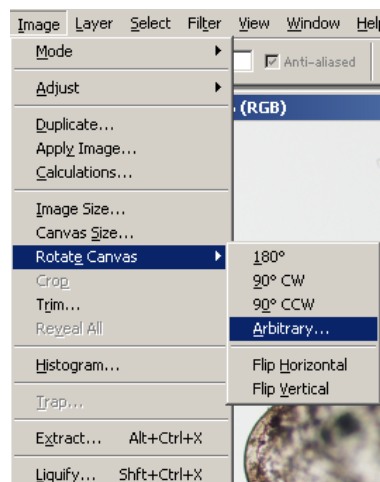
Below is presented a technique that we used successfully at the Limnological Institute in Mondsee during the last years. Various amendments to this procedure can be applied.


For digitising the valve we first insert the microphotograph in the computer programme “Adobe Photoshop” as jpg file.

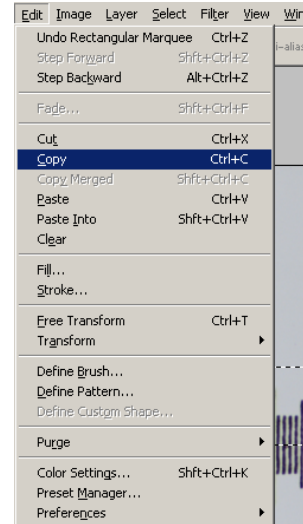
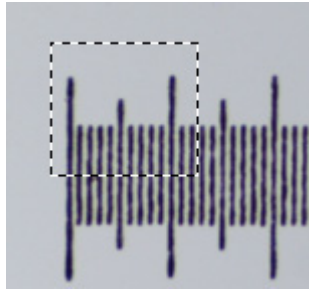
It is necessary to correct the outline of the valves with Photoshop in order to digitalize it with tps.dig. If the outline has a sharp edge on a clean background very few modifications are necessary, but many outlines need to be ‘cleaned’.



First the picture is opened and the valve righted, so that the ventral contour is vertical.  
*Menubar – Image – Rotate Canvas – Arbitrary.*



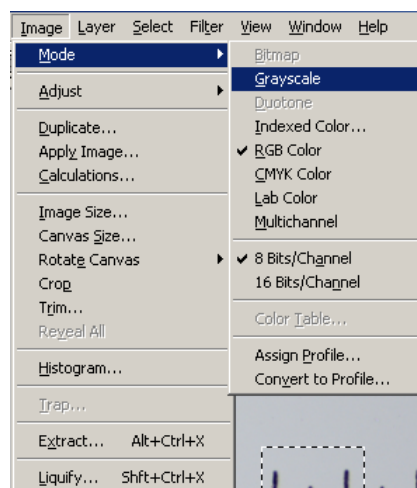
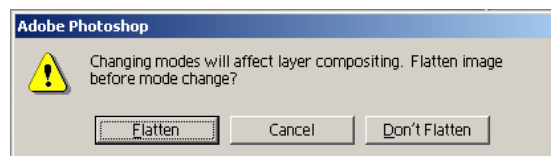
Next the scale, if not there, is inserted. If it is necessary to use a scale from another picture, open it and use the Rectangular Marquee tool  of the toolbar on the left side of the screen to cut out 0.1 mm of the measure (below, left).



*Edit – Copy* – click the picture where you want to insert – *Edit – Paste* (above, right).

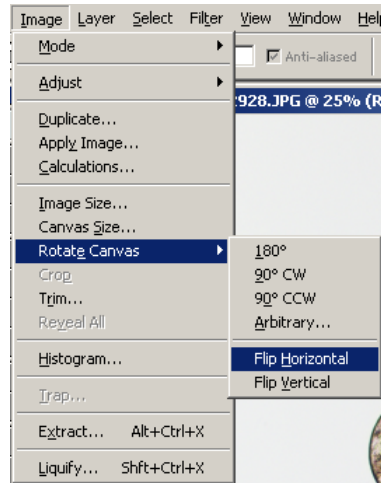
With the Move tool the measure is positioned. 


Next all colour information is discarded. *Image – Mode – Grayscale* – a window asking you to first flatten opens – *Flatten*. (Prior to flattening, the picture and the measure are independent layers, after flattening the layers are permanently joined).

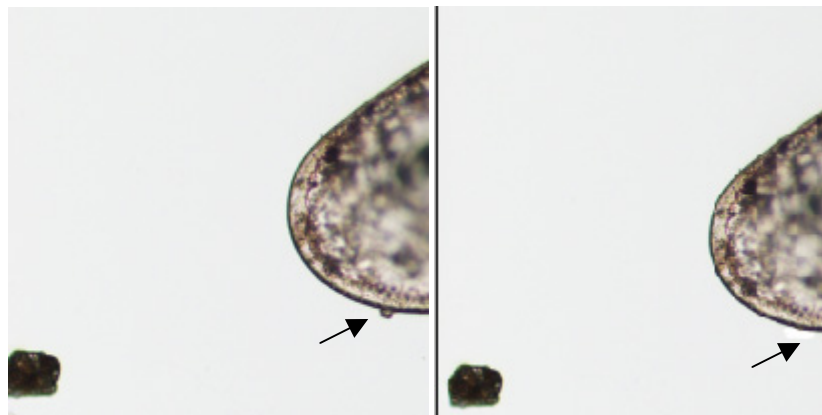



All valves need to be oriented in the same way to compare the outlines; right valves need to be flipped horizontally to be comparable to left valves.

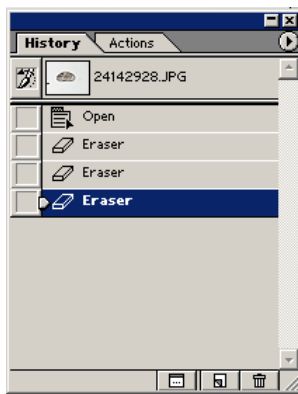
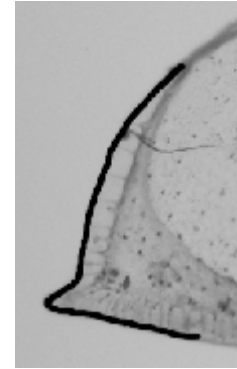
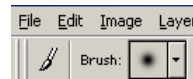
*Image – Rotate Canvas – Flip Horizontal.*



The outline is checked; any pieces of dirt that are connected to the valve are erased with the Eraser tool  (dirt somewhere in the background is ignored).

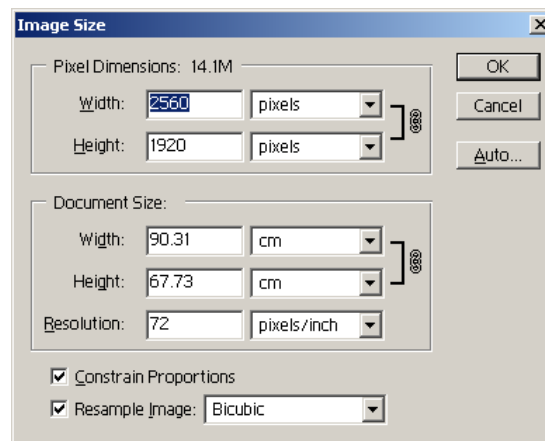


If the outline is very translucent there is too little contrast and the edge needs to be outlined with the Paintbrush Tool , select the thickness of the stroke in the toolbar below the menubar.



In the menubar under Window there is a heading Show History, if selected a window is shown where the modification steps are listed, it is possible to undo the last 20 changes.

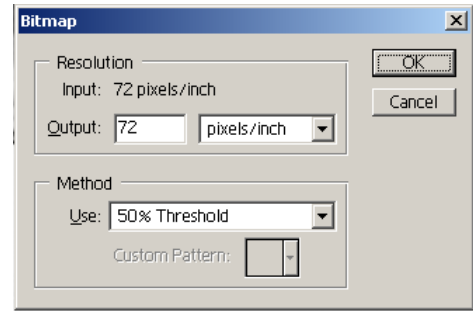
The original is shown in the header bar of the history window and can always be gone back to.



The Image Size (under *Image* in the menubar) is very important for the number of digitised points the outline is made up out of. Think about an appropriate number (> 1000 pts) before digitalizing, since MORPHOMATICA is sensible to variations of points used to calculate the outline. The number of points depends on the size of the picture and the resolution. Size and resolution should be adapted prior to producing the bitmap. No example for resolution and size can be given, because these parameters vary greatly, depending on the way the picture was produced.

The outline is digitalized from a bitmap of the valve, to get the bitmap go to *Image – Mode – Bitmap*.

Choose 50% Threshold for the bitmap. Check, if the valve has a continuous outline, wherever there is a small gap it will disturb the digitalizing of the outline. Make changes not in the bitmap, but go back in the history to the last point prior to bitmap to make modifications.



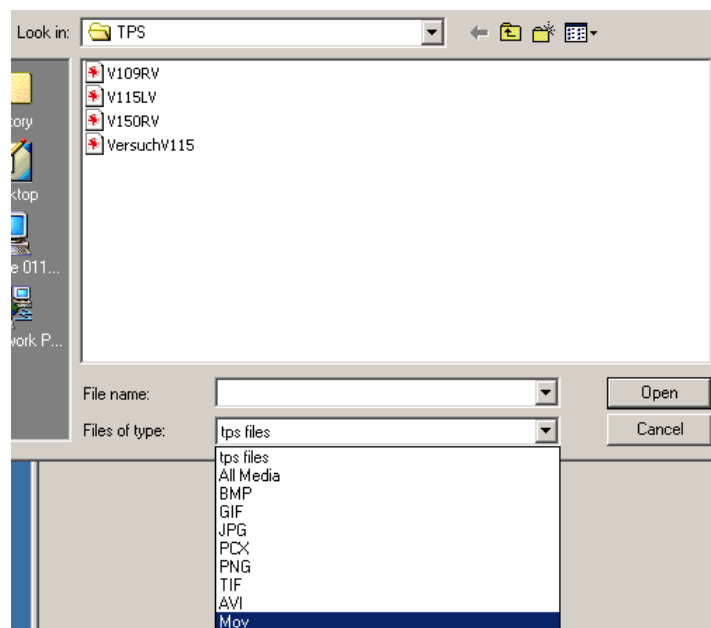
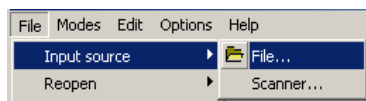
Finally the modified image is stored, go to *File – Save as* and store it either under a new name or a different format, do not simply *Save* since this way the original picture will be lost. This step can be performed first, depending on your personal preferences.

### The computer program “Tps.Dig”

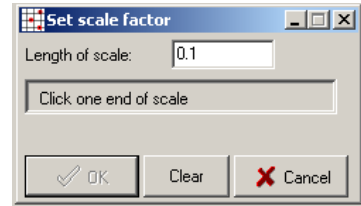
This program is a simple means to digitalize objects. If you have to download it from the internet choose:


Rohlf, F J. 2001. tpsDIG, Program version 1.43. Department of Ecology and Evolution, State University of New York, Stony Brook, NY.: <http://life.bio.sunysb.edu/morph/soft-dataacq.html> (1/17/04).

Open your picture under *File – Input Source – File*; select the format you used for storing the modified picture.



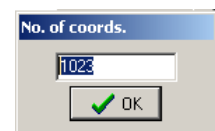
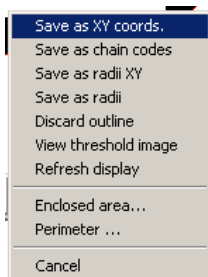
Go to *Options – Set Scale*, change the size to 0.1 (not comma, but point) and click the beginning and end of the measure on the picture.



The line must be parallel to that of the scale on the picture, be careful to work accurately since small mistakes here will result in artificial differences in size in MORPHOMATICA. Click the outline button  to activate the tool for digitalizing the contour. Place the tip of the arrow right of the valve, make sure that there are no pieces of dirt between the tip of the tool and the valve and click it. A red line marks the outline of the object, as recognized by the program.



If the line is partially on the inside of the valve, the contour was 'leaky' and has to be corrected in Photoshop (changes must be saved in Photoshop and the valve reopened in tps.dig to adopt the changes). Right click the mouse and choose *Save as XY coords.*, the number of points for the outline appears in a window and has to be accepted, an additional yellow outline appears.



If the number of digitised points is suitable (should be around 1000 - 1500 points), store the information under *File – Save data as*, if not go back to Photoshop to change the Image Size and Resolution to fit your ideas.