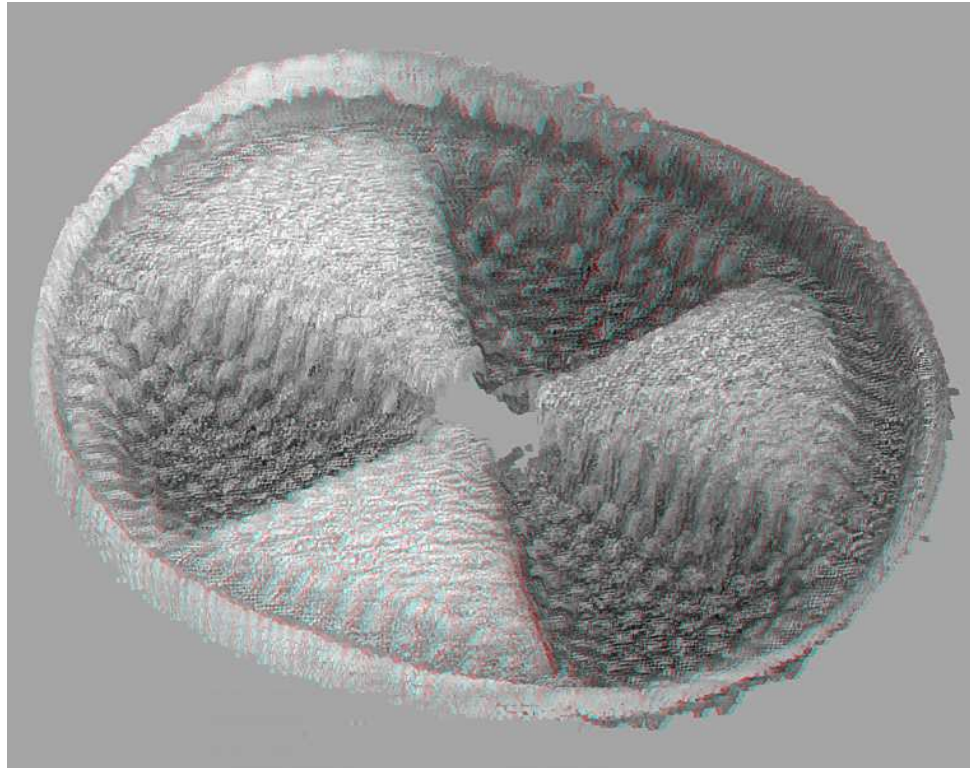


# Zwei- und dreidimensionale Stapelverarbeitung von Mikrobildern mit PICOLAY



Vortrag auf  
der IME am  
18.9.2010

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Institut für Chemie und Biologie des Meeres  
Universität Oldenburg



[www.pmbio.icbm.de](http://www.pmbio.icbm.de)

# Themen

Theorie (wenig) und Anwendung

3D-Visualisierung:

→ Tiefenkarten-basiert

→ Hologramm-Verfahren

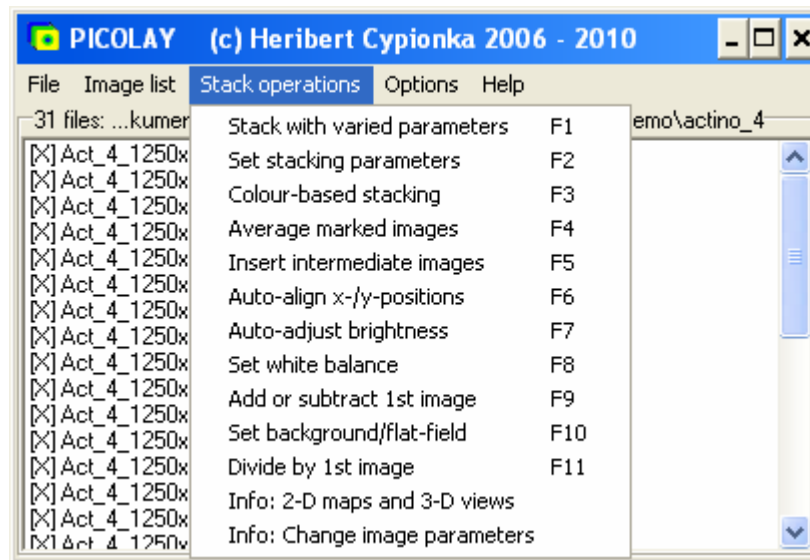
# Mikrobiologischer Garten



[www.mikrobiologischer-garten.de](http://www.mikrobiologischer-garten.de)

[www.microbiological-garden.net](http://www.microbiological-garden.net)

[www.picolay.de](http://www.picolay.de)



**Das kleinste und schnellste Stacking-Programm**

**Erzeugt 2D- und 3D-Bilder**

**Erledigt nötige Bildbearbeitung**

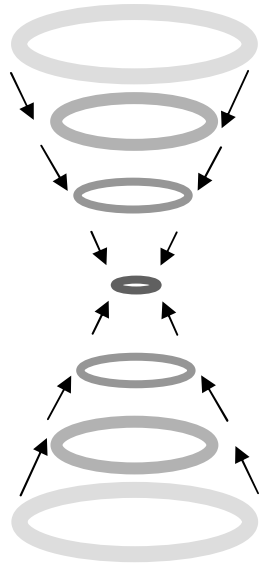
**Braucht keine Installation**

**Freeware**

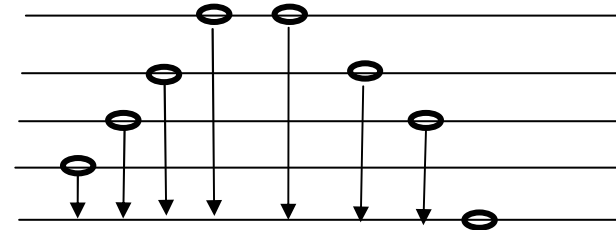
## Things you can do with PICOLAY

- 1. Browse images of a list at constant position and size
- 2. Display images of a list as slide show
- 3. Generate an animated gif file
- 4. Cut the edges of one or all images in the list
- 5. Resize one or all images of the list
- 6. Rename images
- 7. Change the file format of images
- 8. Delete selected images from disk
- 9. Add or subtract images from one another
- 10. Convert coloured images to grey scale
- 11. Increase sharpness
- 12. Increase contrast
- 13. Increase colour saturation
- 14. Change gamma value
- 15. Change brightness
- 16. Add or subtract offset or percentage to the red, green or blue channel
- 17. Run a median filter over one or all images of your list
- 18. Rotate images by any angle
- 19. Flip images horizontally or vertically
- 20. Generate negatives of images
- 21. Select sharp areas from an image stack (focus stacking)
- 22. Select areas with a target colour from your stack
- 23. Average images
- 24. Generate and insert intermediate images between the images of your stack
- 25. Align displaced images of your stack with respect to x- and y-positions
- 26. Harmonise brightness of the images in your stack
- 27. Set white balance of one or all images in your stack
- 28. Set a smooth background (flat field) by removal of disturbing items from your stack
- 29. Divide RGB values of images by those of the first image in the list
- 30. Generate a depth map of your stack
- 31. Generate 3D images from depth map and stacked image.
- 32. Generate red-cyan images for anaglyph goggles
- 33. Generate (2 or 4) separate images for a 3D observation with parallel or crossed eyes
- 34. Generate spatial projections of stacked objects from any angle
- 35. Generate 3D hologram views that allow to display covered structural details
- 36. Generate rocking and rotating animations in 3D
- 37. Generate 3D images for defined observation distances (viewing angles)
- 38. Let 3D images appear in front of or behind the screen
- 39. Paint on images
- 40. Blur disturbing details
- 41. Clone (= copy) parts of an image to other areas
- 42. Clone areas of original images to the stacked result (including depth map information)
- 43. Write text on images horizontally or vertically with any font and size
- 44. Draw a scale bar (rectangle with any colour and size)

# Dekonvolution and Fokus-Stacking

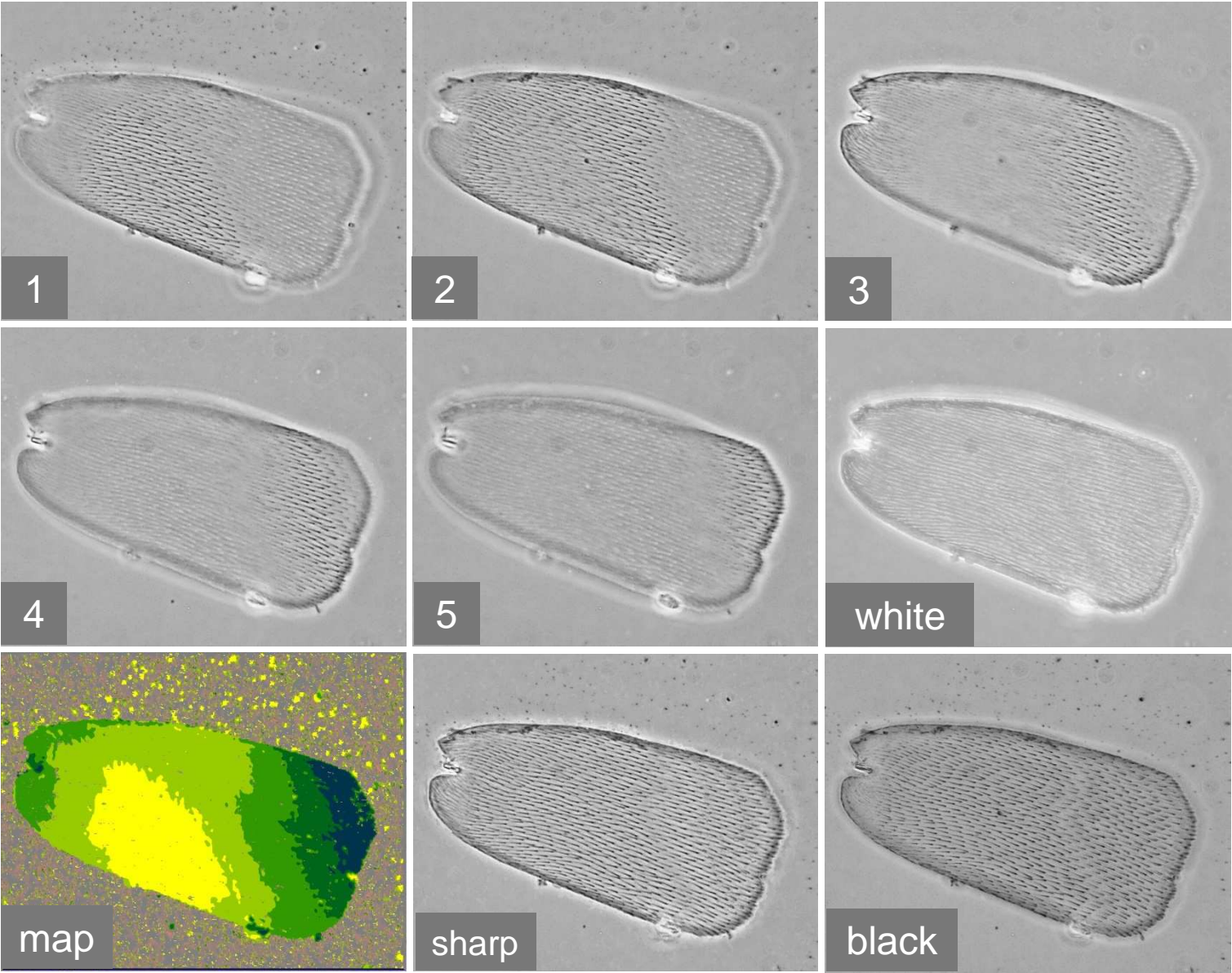


**Dekonvolution:** Rekonstruktion basierend auf Information aus allen Schichten



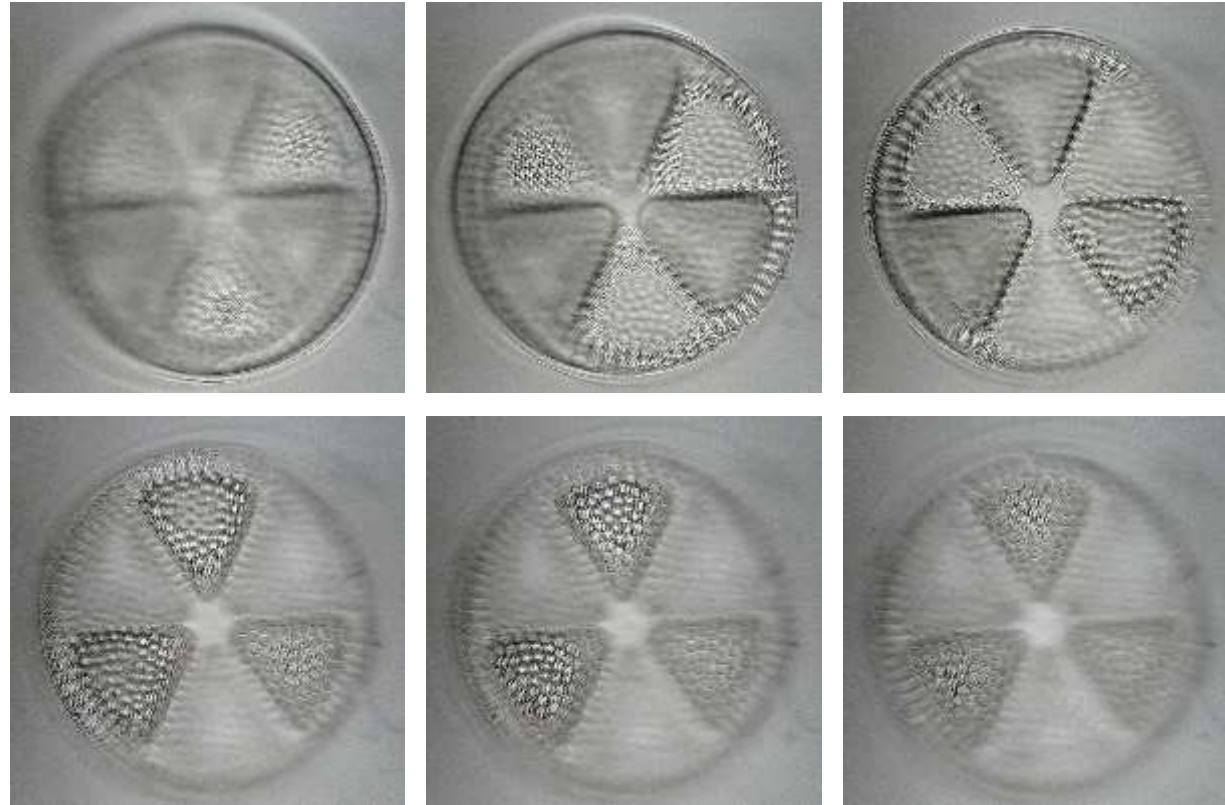
**Stacking:** Selektion einer einzelnen Ebene (nach Schärfe oder Farbe)

# Farbe, Schärfe, Höhe



Schmetterlingsschuppe

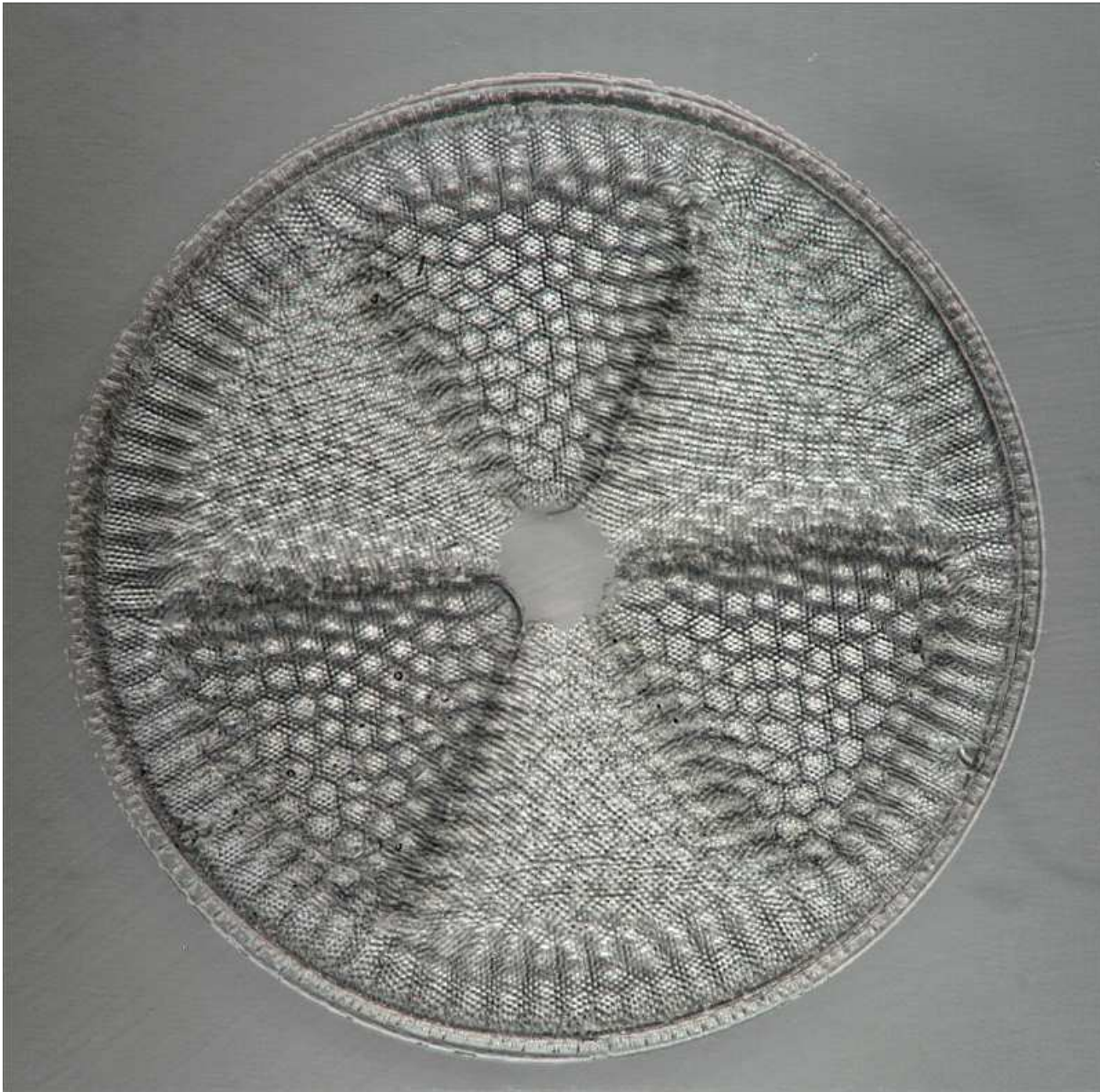
# *Actinoptychus*



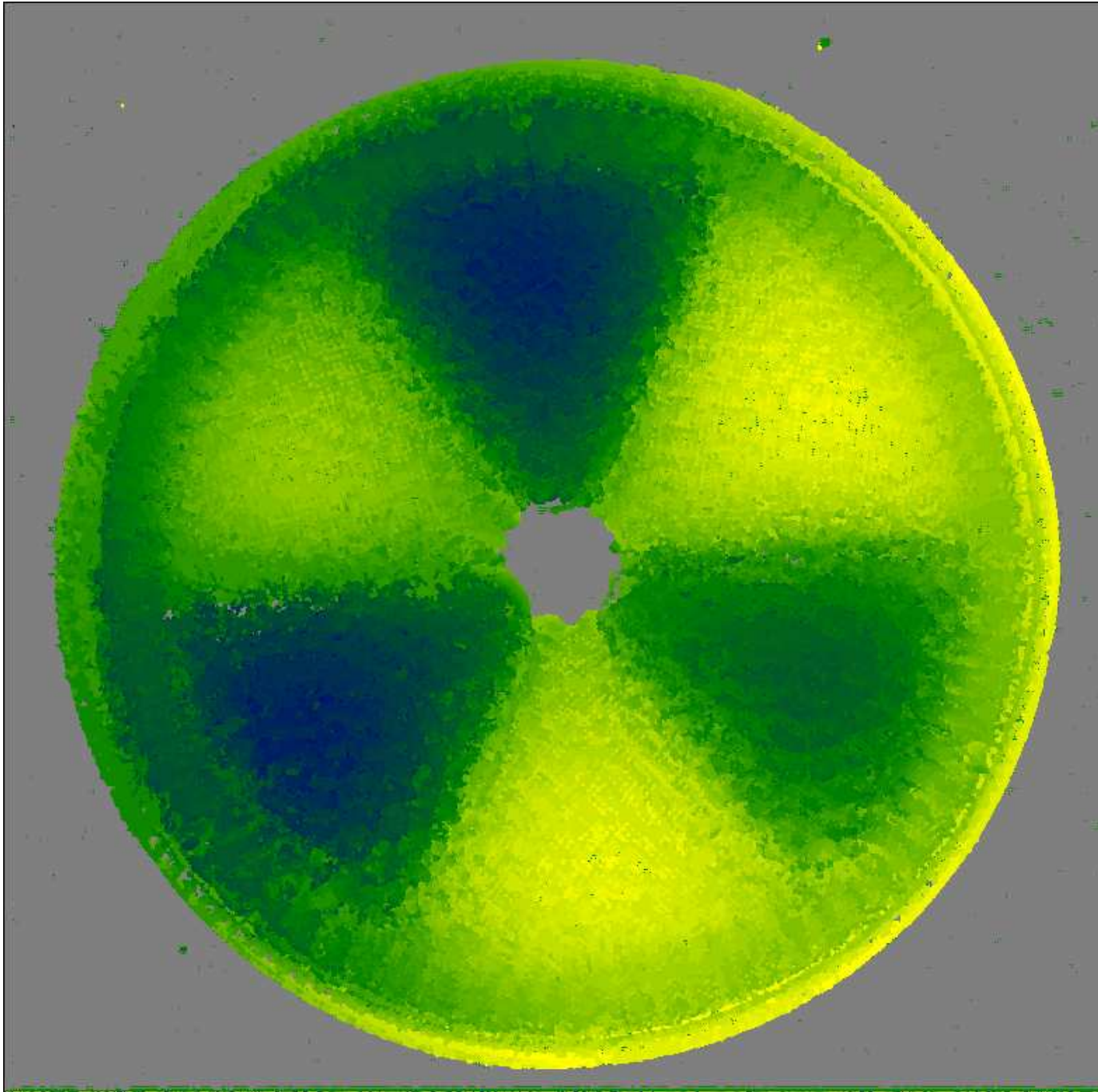
6 von 31 Bilder aus dem Stapel



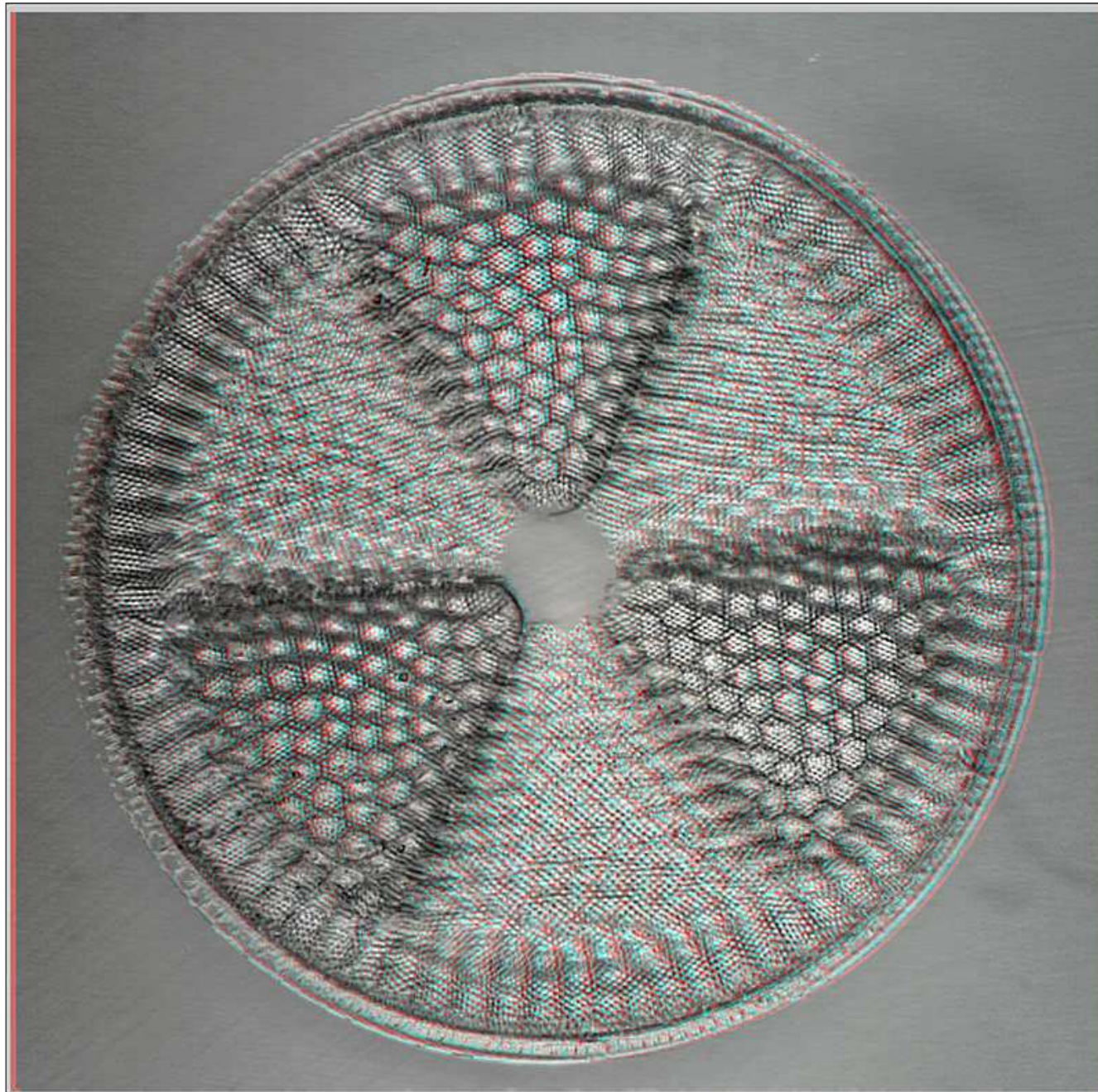
*gestackt*




# *Tiefenkarte*



# 3D



**PICOLAY 3-D view**

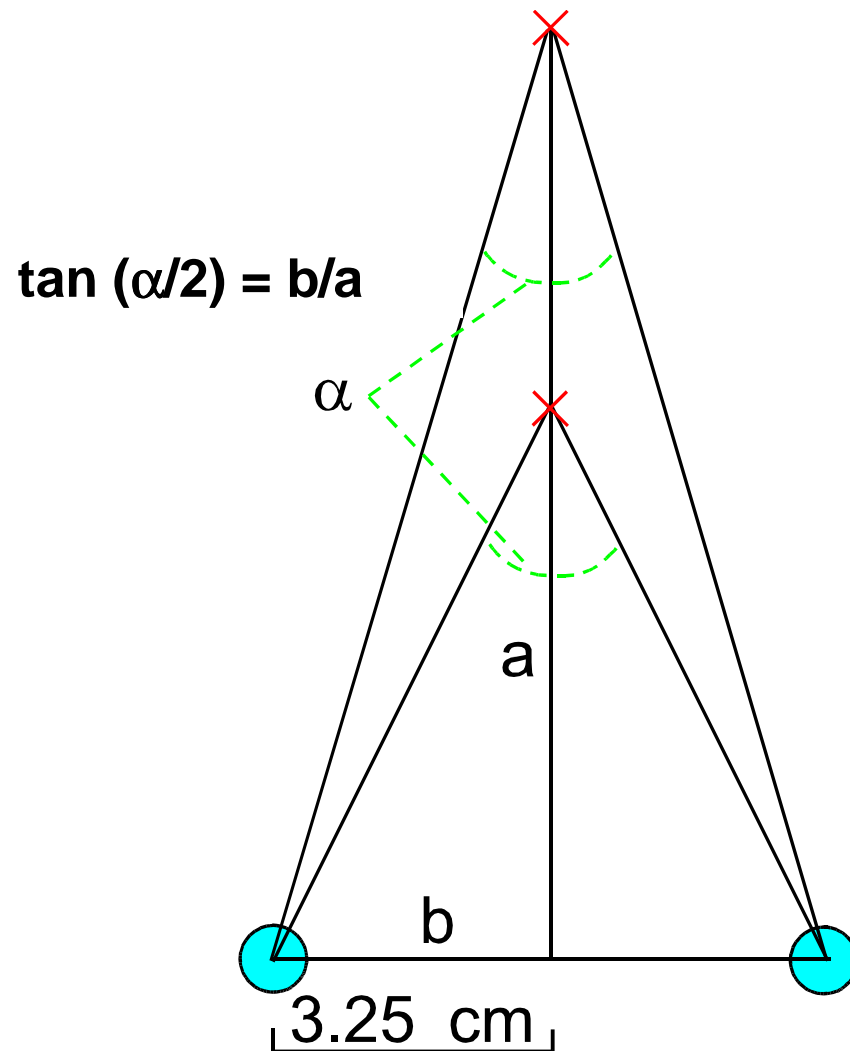
 Length of Z axis (% of image height)   
Perspective (%)   
Cyan-red shift   
Seam around 3-D pixels

Projection based on 2-D map  
 Hologram stacking

Images to be generated  
 Original  Map  Overlay  
 Black & white  Grid  
 3-D view Viewing angle  °  
 Cyan-red  [RL]  [RLR]  
 [R] + [L]  [LR]  [LRL]  
 [R - L]  [L - R]  Rocking gif

Auf der Tiefenkarte  
basierend

## Blickwinkel aus verschiedenen Abständen



Augenabstand:  
Basis = 6.5 cm

Objekt- abstand	Betrachtungs- winkel
--------------------	-------------------------

$a$	$\alpha$
-----	----------

6.5 cm →	53°
----------	-----

0.3 m →	12°
---------	-----

0.5 m →	7.4°
---------	------

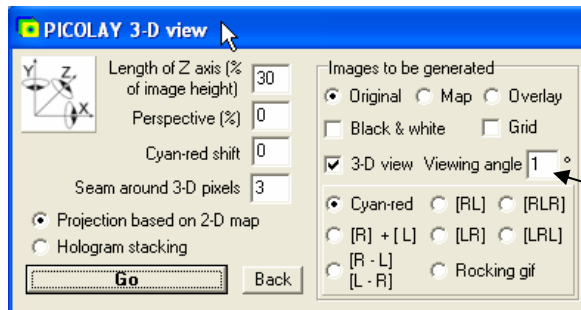
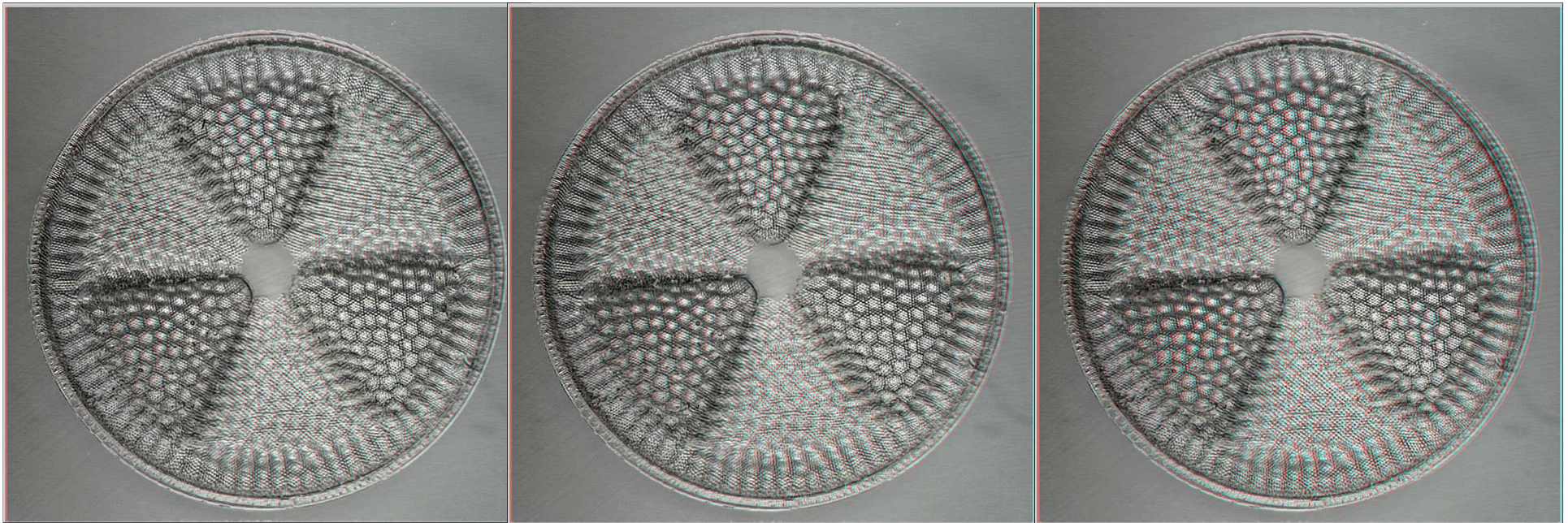
1 m →	3.7°
-------	------

2 m →	1.9°
-------	------

4 m →	0.9°
-------	------

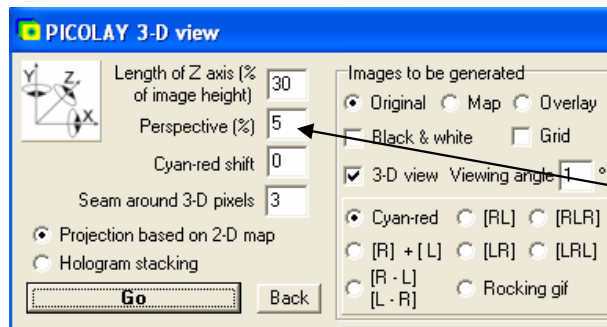
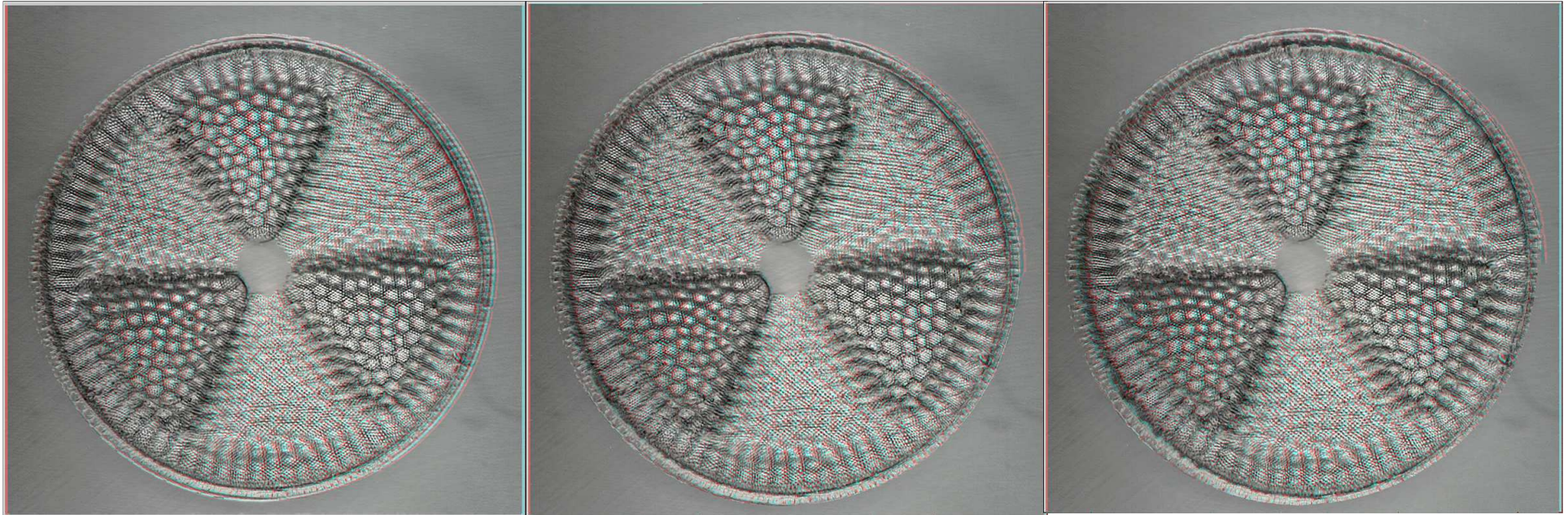
8 m →	0.5°
-------	------

100 m →	0.04°
---------	-------



3D

Betrachtungswinkel 1, 2 und 4 °

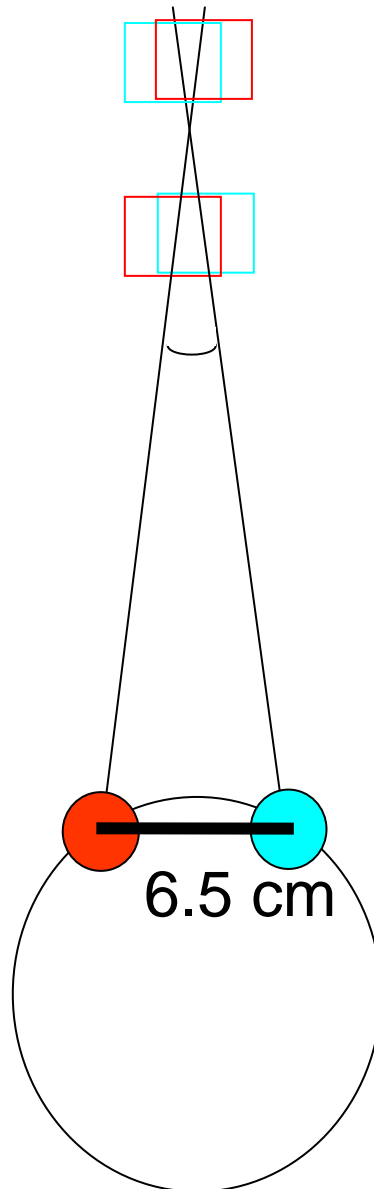


3D

0, 5 und 10 %  
Perspektive

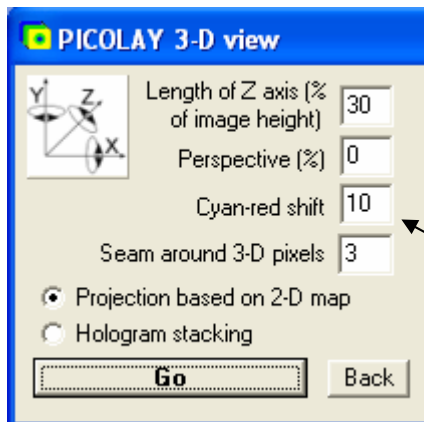
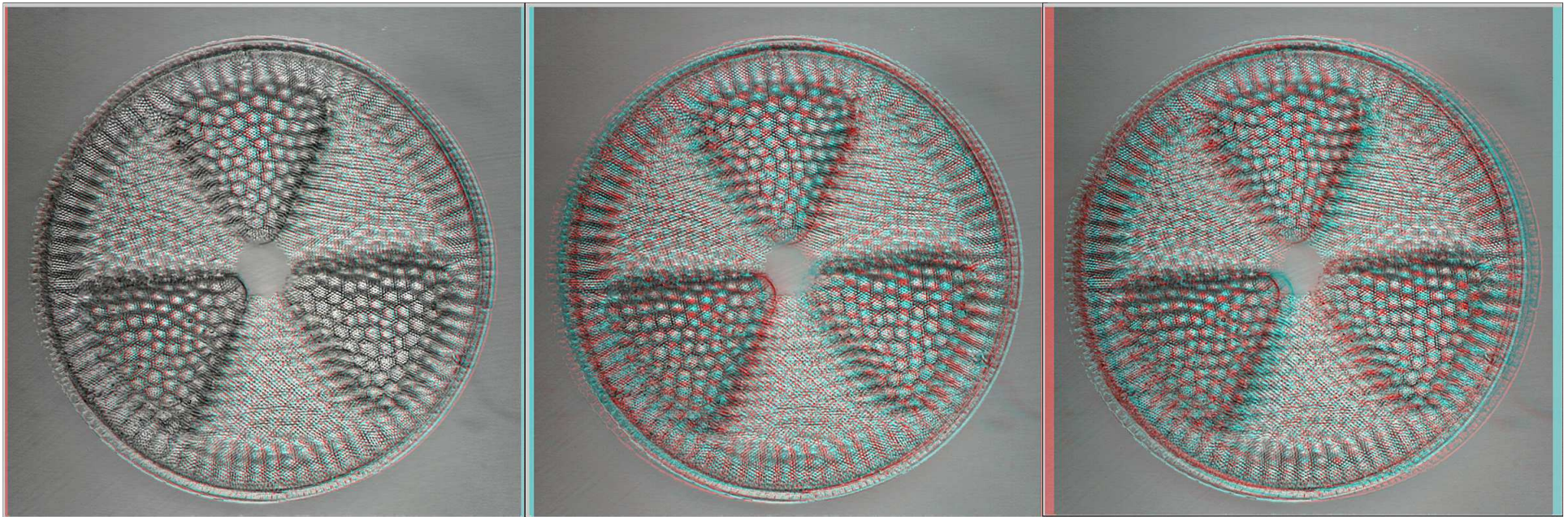
# Veränderungen der Betrachtungsweise

Anaglyphenbrillen (z.B. Rot-Cyan) erlauben es, zwei farbseparierte 3D-Bilder übereinander zu projizieren und das Ergebnis mit normalem Kreuzblick zu betrachten.



Änderungen im Versatz der Bilder lässt das Objekt nach vorn oder hinten wandern

Vertauschen der Bilder (oder Gläser der Anaglyphenbrille) vertauscht hinten und vorne und lässt das Objekt konkav statt konvex erscheinen.

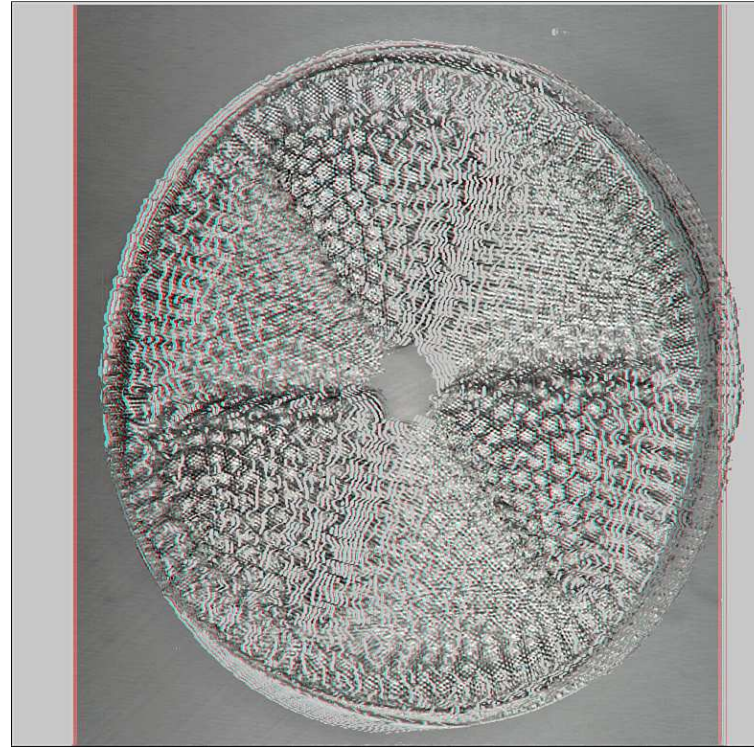
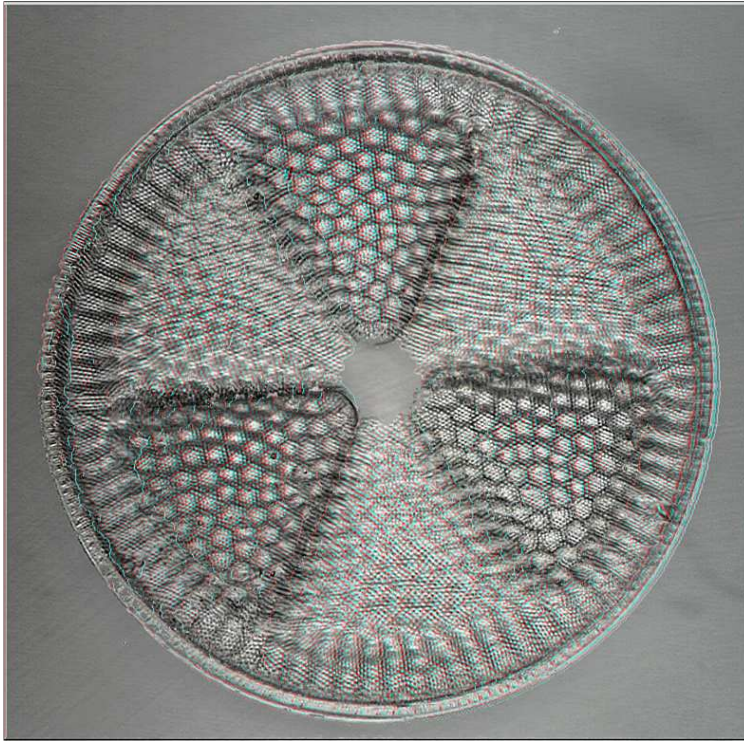


3D

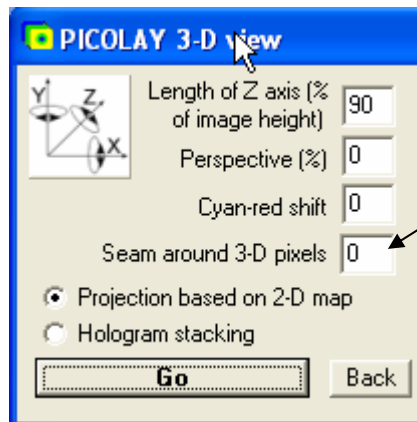
Cyan-red shift

0, 10, -10



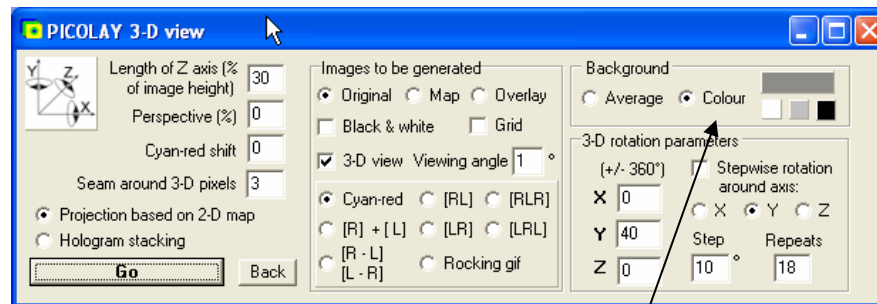
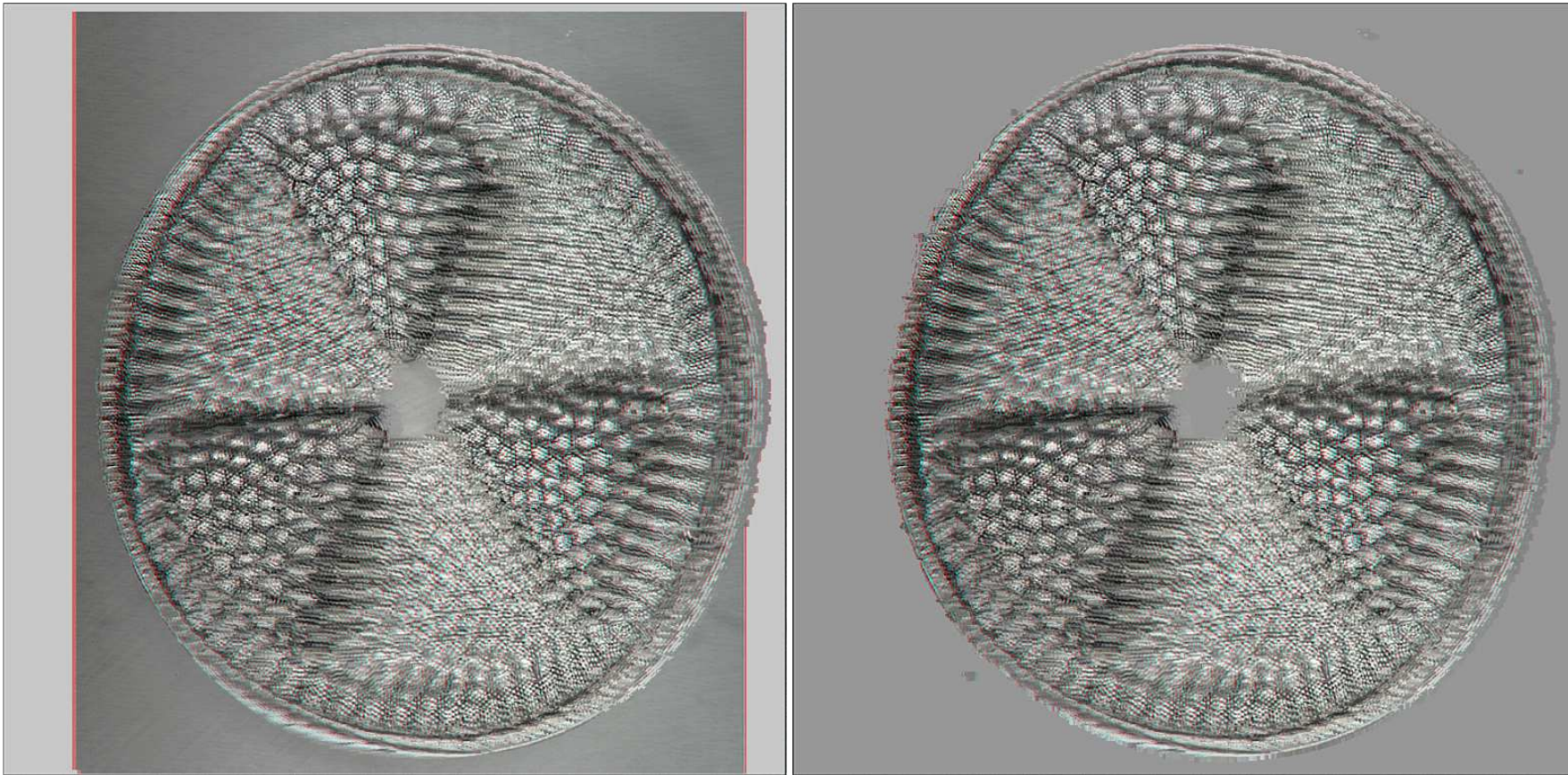


Objekt 40° um y-Achse gedreht



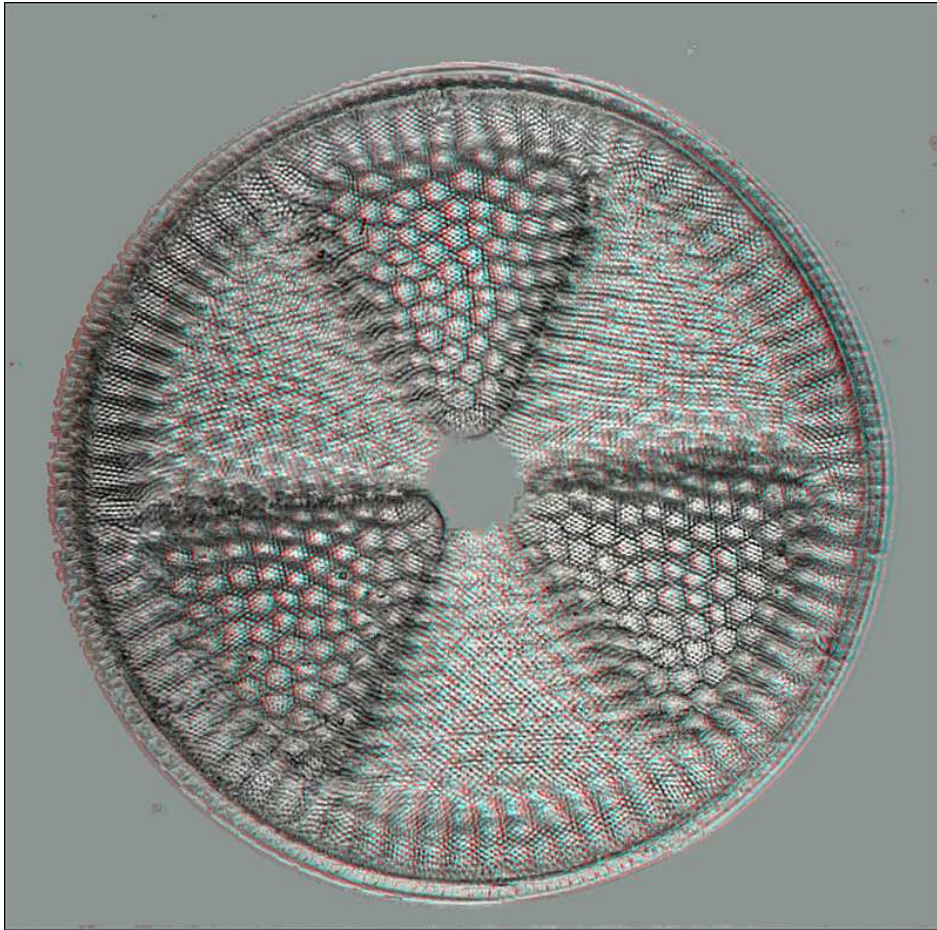
Saum

Ohne Saum ,zerreißen'  
Tiefenkarten-basierte  
3D-Bilder (Karte nicht  
größer als Grundfläche)



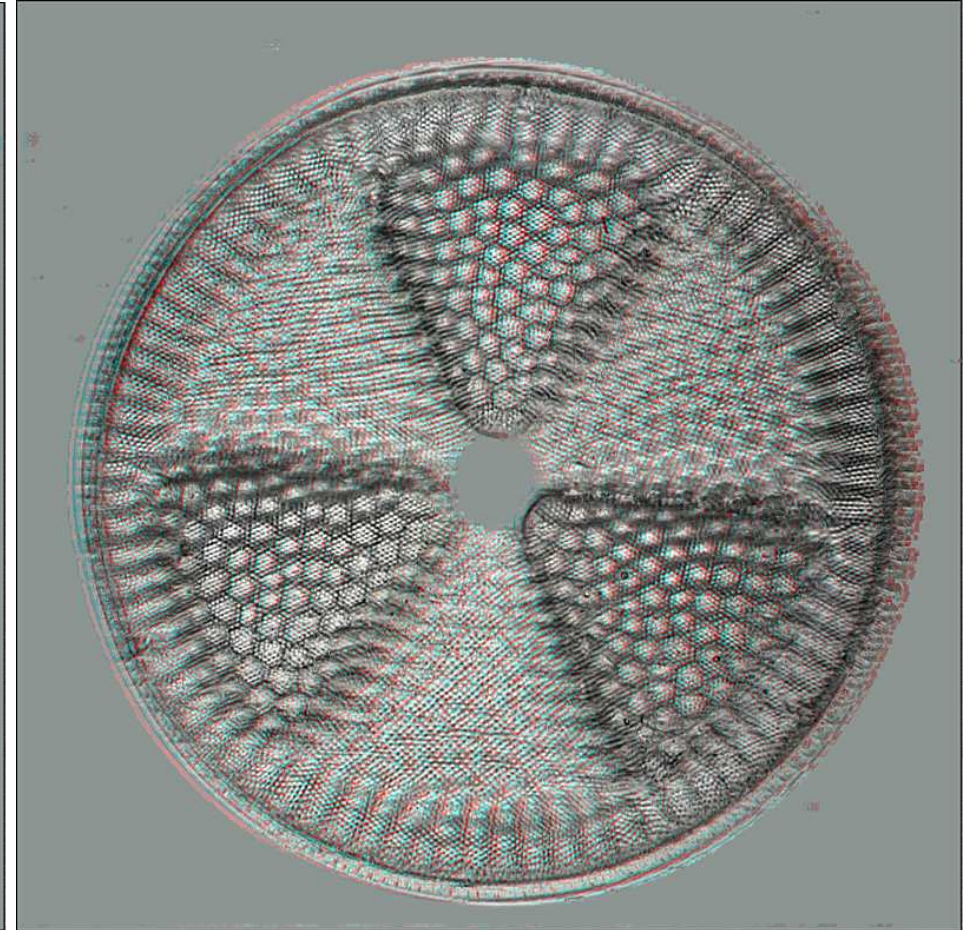
Hintergrund:  
Farbe statt  
Mittelwert

## Tiefenkarten-basiert



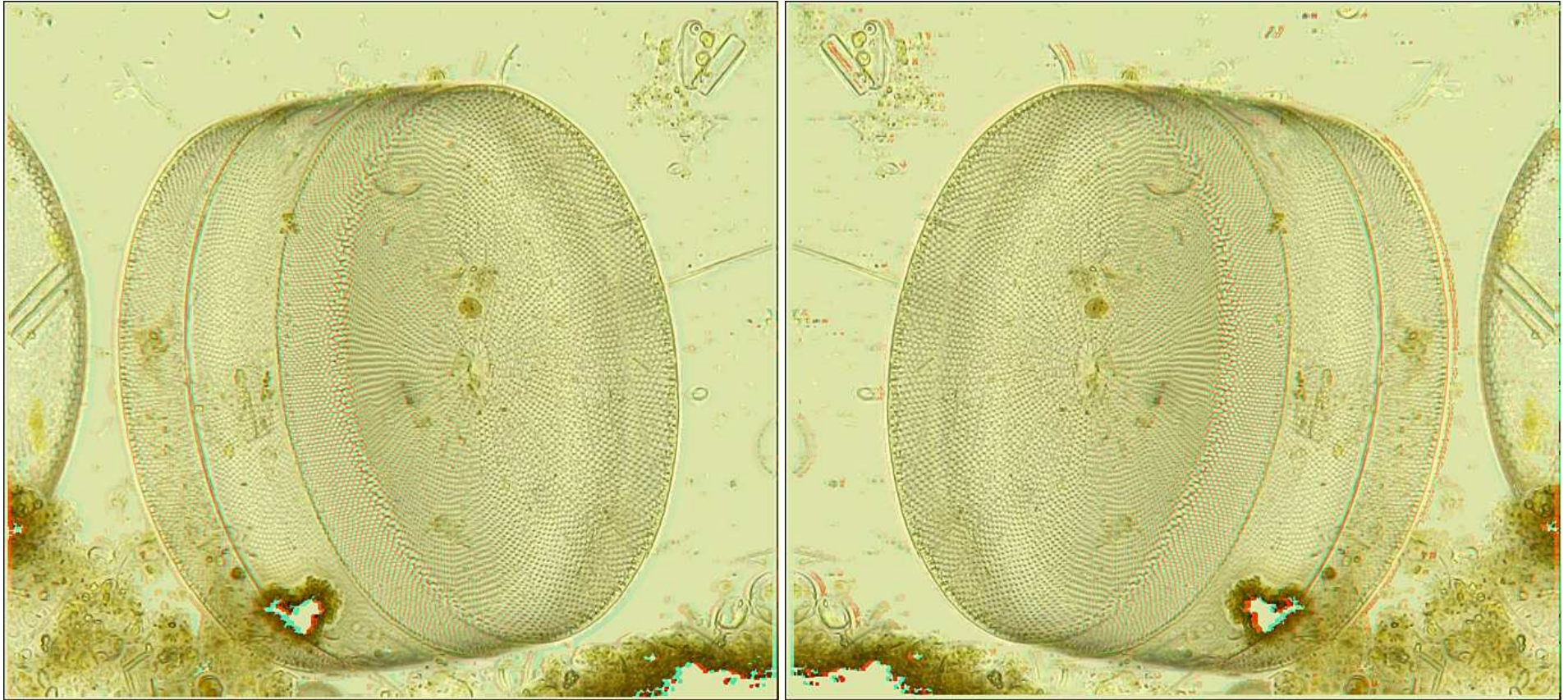
0°

*Actinoptychus*



y 180°

# Karten-basierte Darstellung

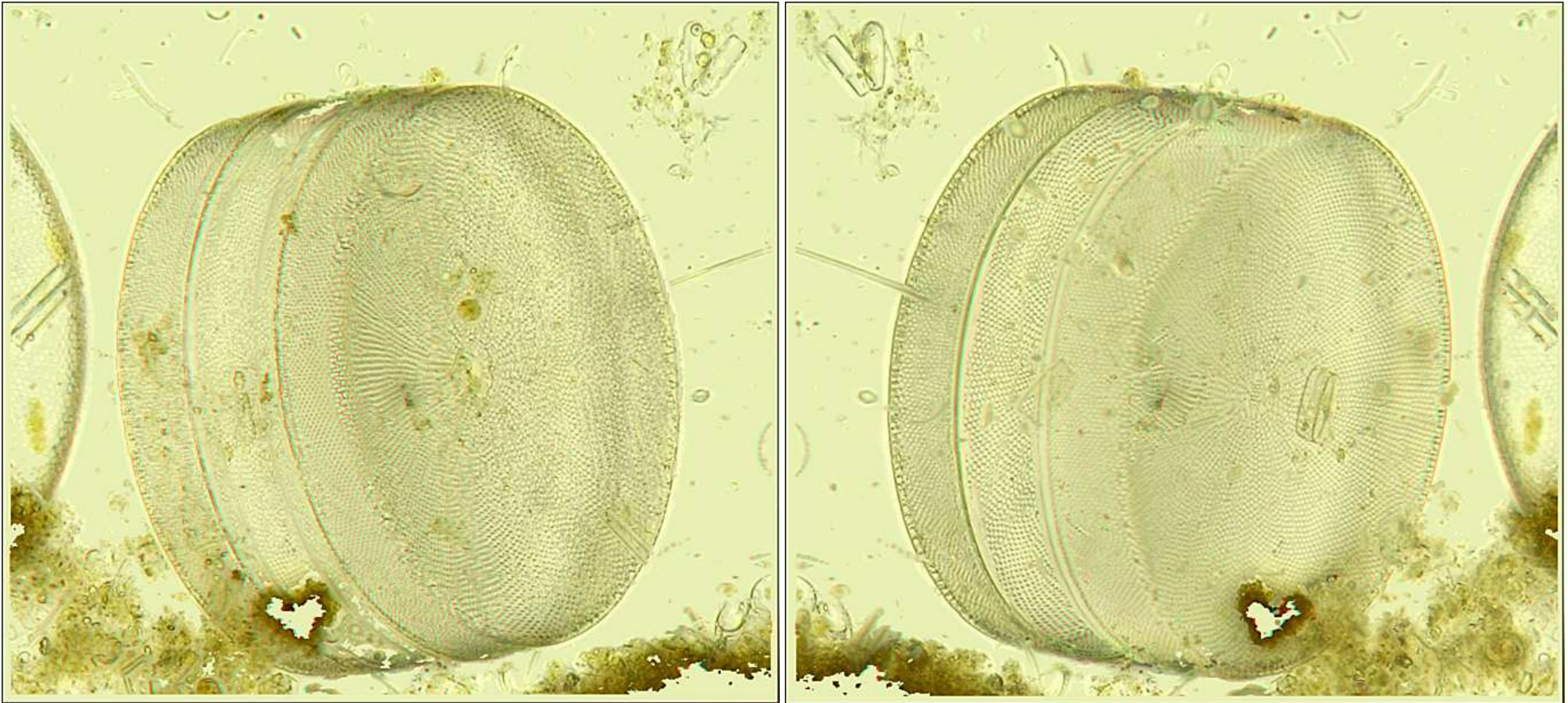


0°

***Coscinodiscus***

y 180°

# Hologramm

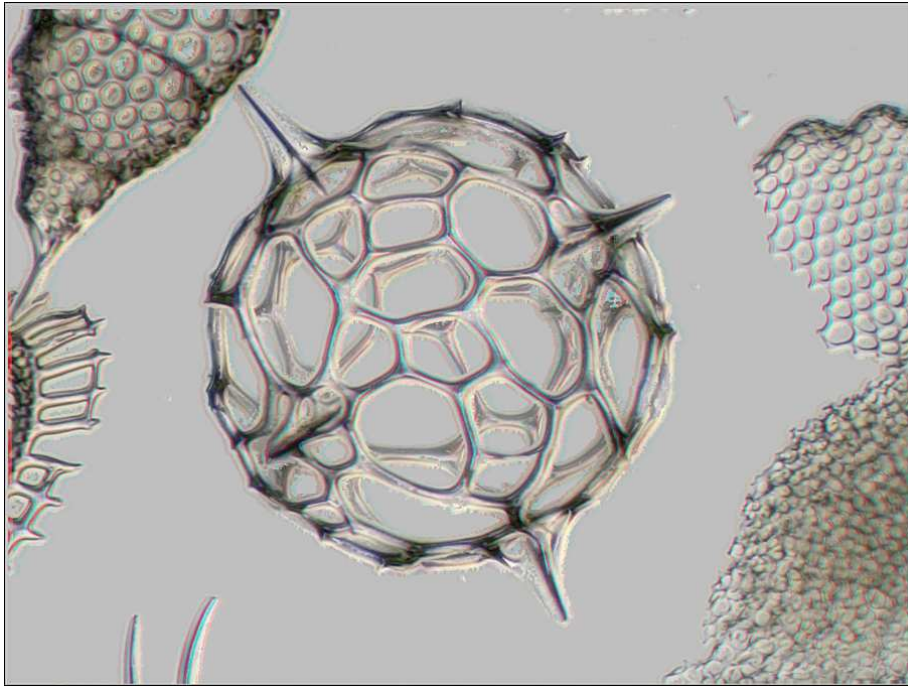


0°

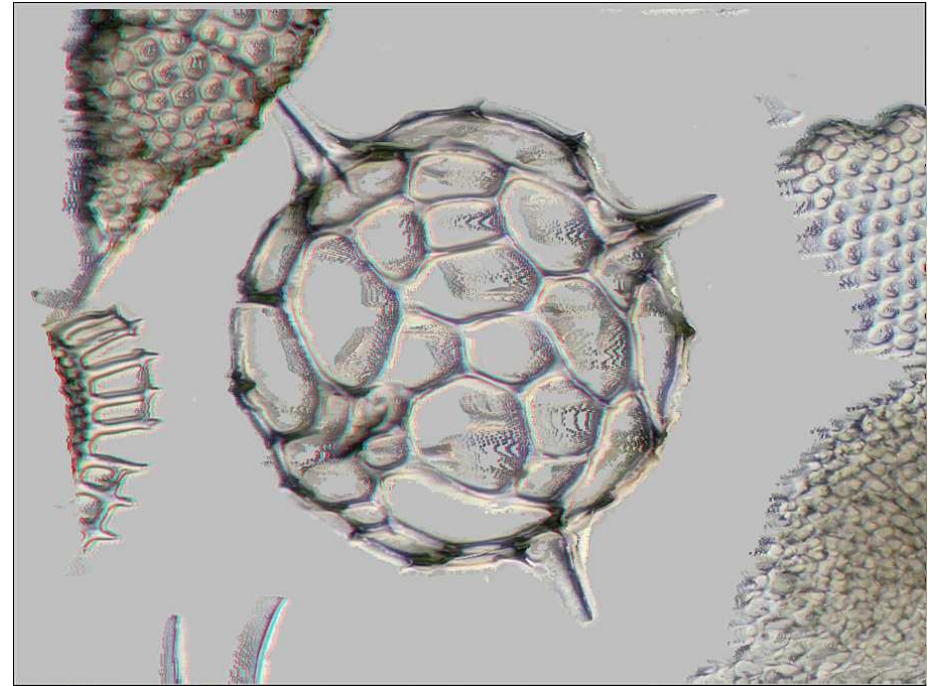
***Coscinodiscus***

y 180°

# Karten-basierte Darstellung



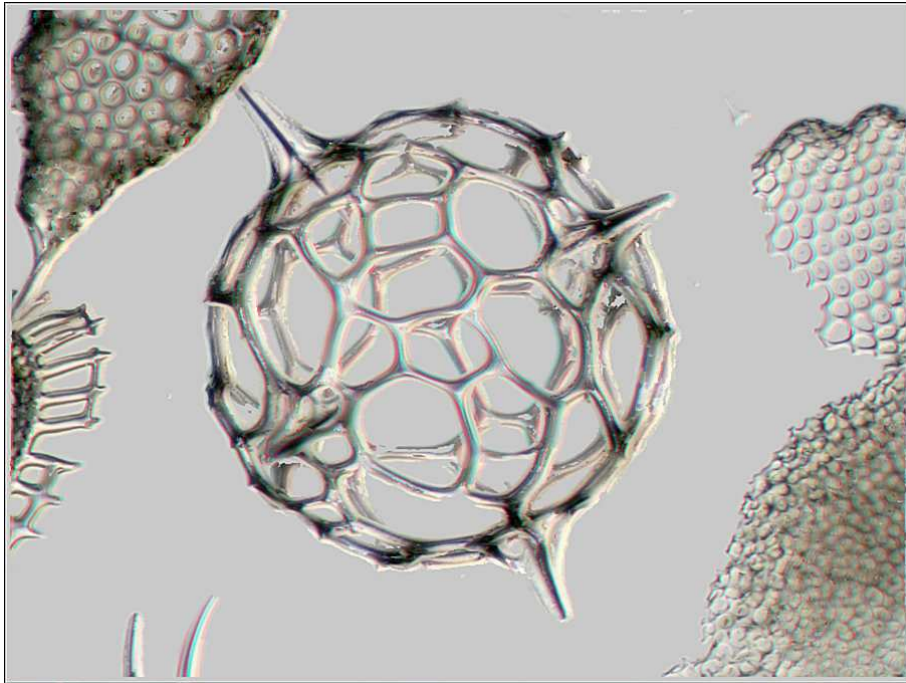
0°



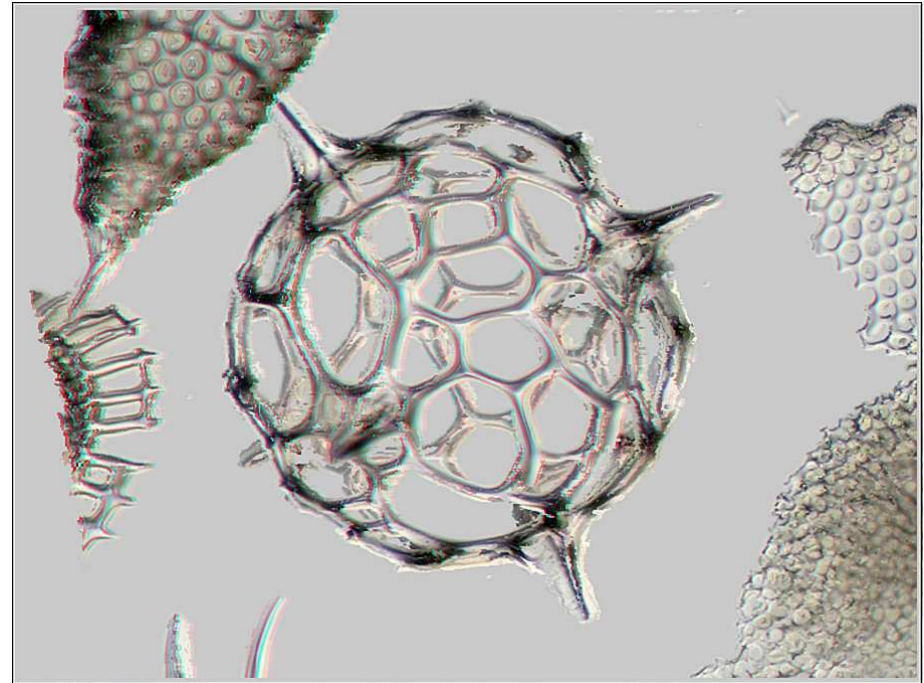
y 15°

Bilderstapel Eberhard Raap

# Hologramm



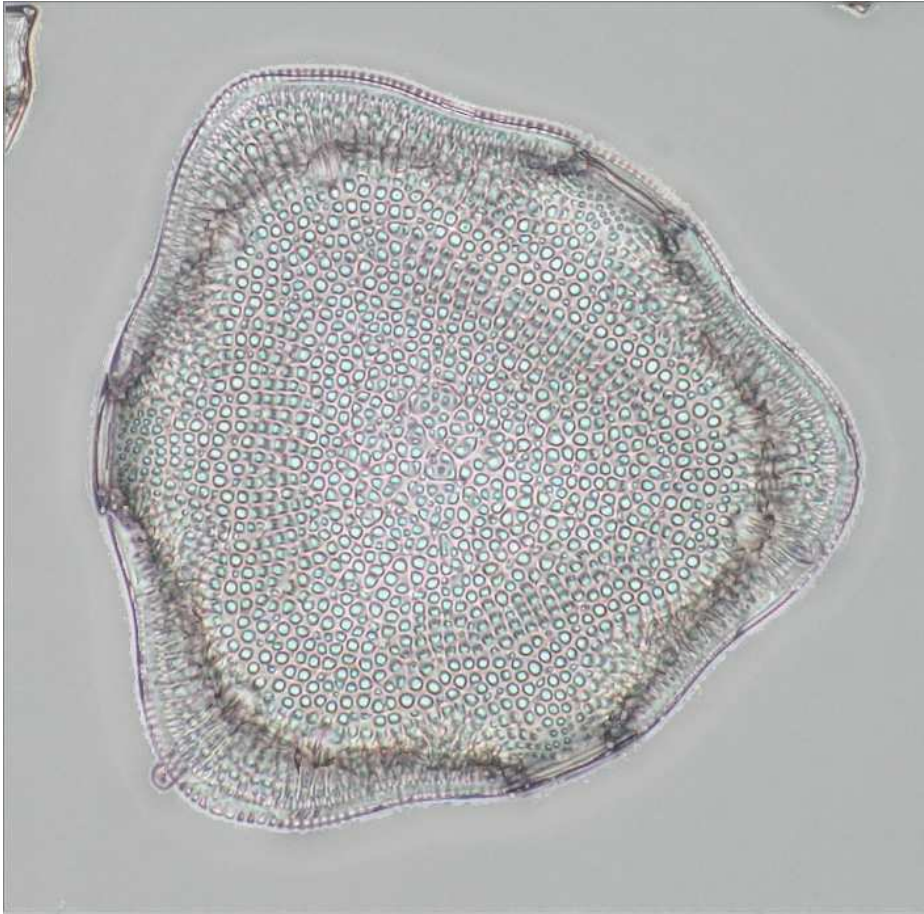
0°



15°

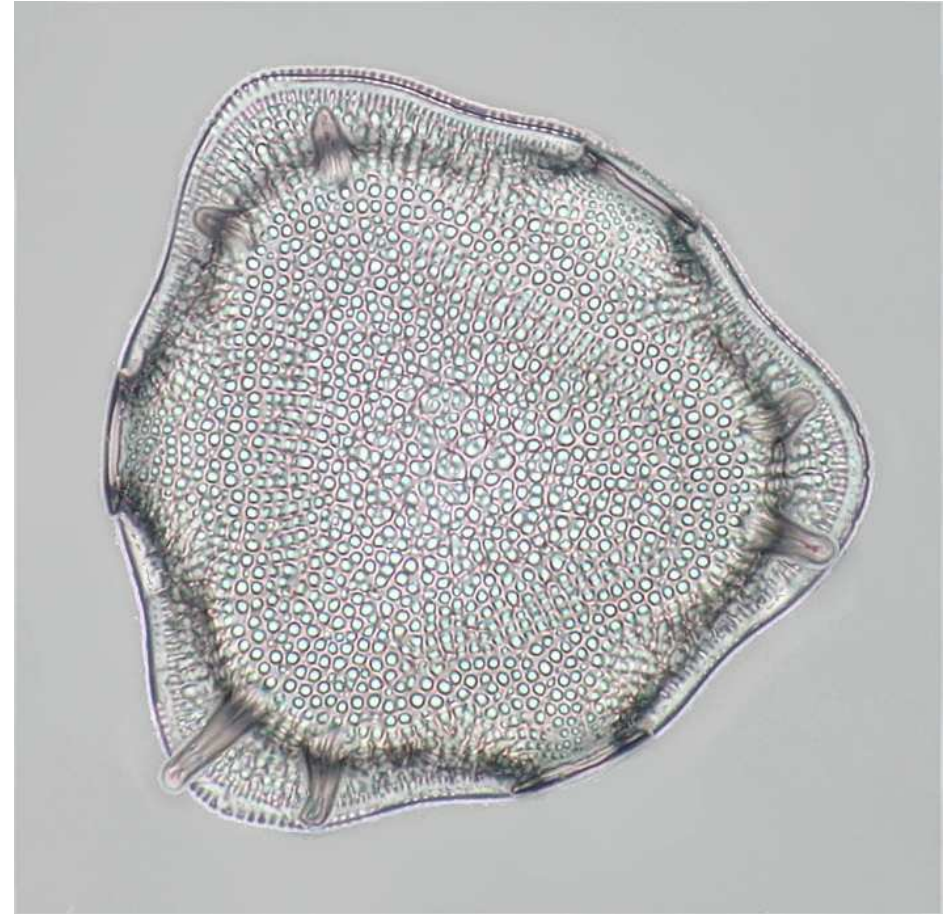
Bilderstapel Eberhard Raap

# Cerataulus



Erster Stack-Versuch

Bilderstapel Eberhard Raap

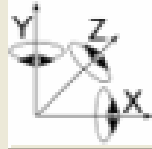


Nach Bearbeitung mit PICOLAY:  
Umkehrung der Reihenfolge,  
Bevorzugung oberer Strukturen,  
ein wenig Klonen, Reinigung des Hintergrunds  
und Verstärkung von Kontrast und Schärfe



# Verschiedene 3D-Darstellungsweisen

**PICOLAY 3-D view**

 Length of Z axis (% of image height)   
Perspective (%)

Seam around 3-D pixels

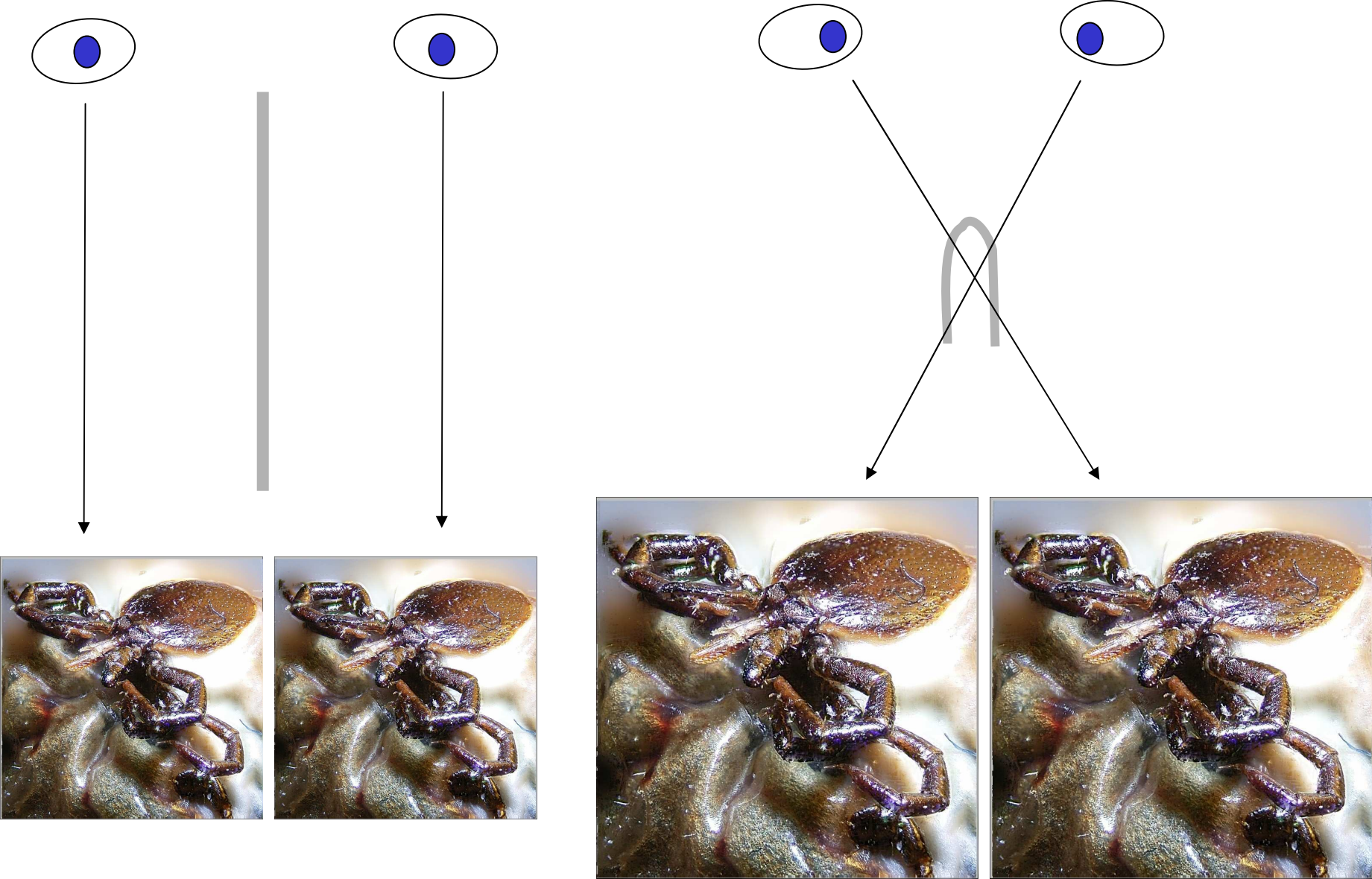
Projection based on 2-D map  
 Hologram stacking

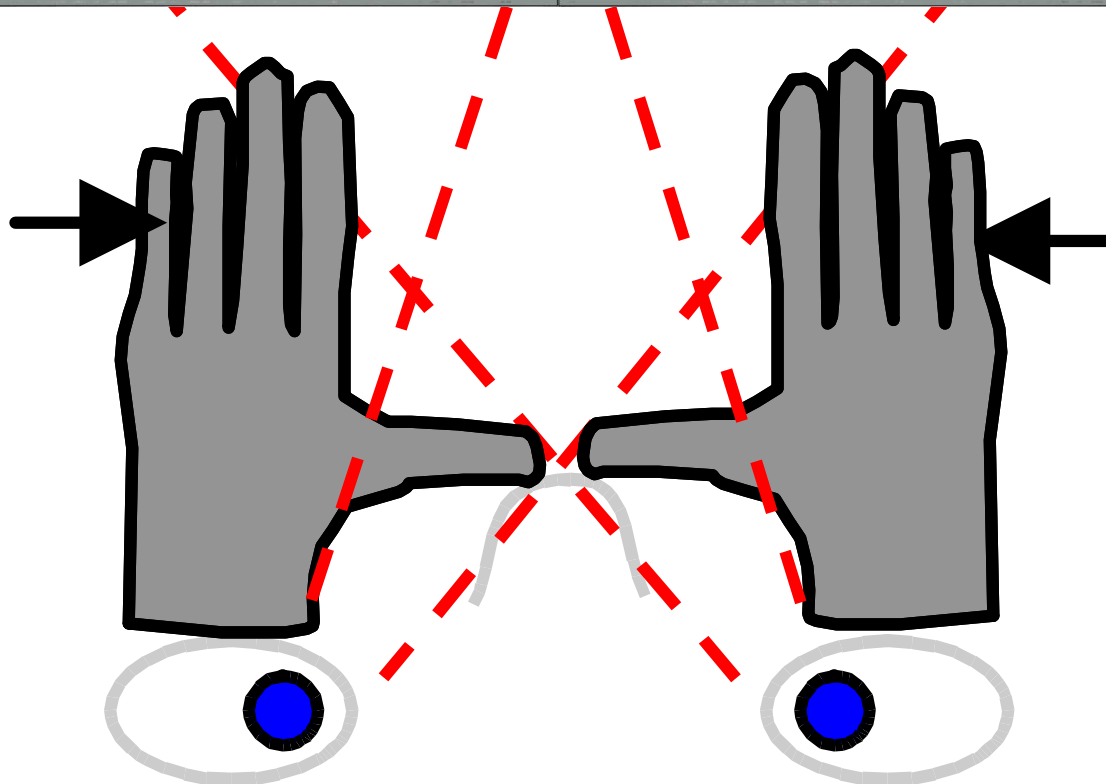
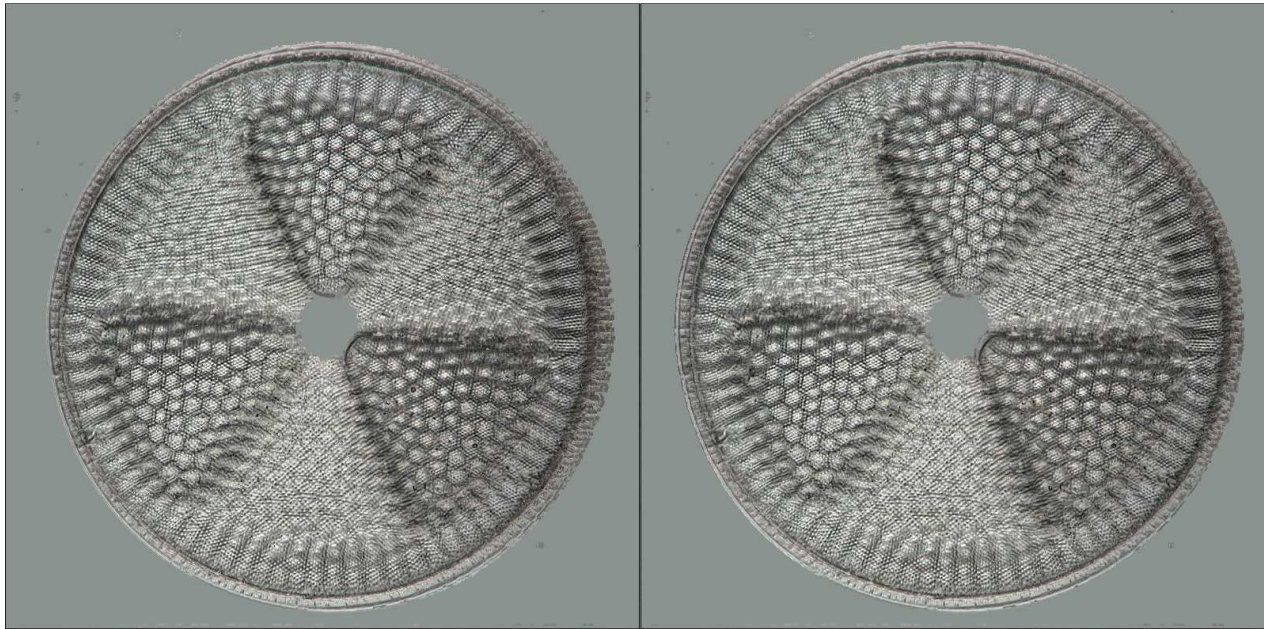
Images to be generated

Original  Map  Overlay  
 Black & white  Grid  
 3-D view Viewing angle  °

Cyan-red  [RL]  [RLR]  
 [R] + [L]  [LR]  [LRL]  
 [R - L]  **Rocking gif**  
 [L - R]

# Parallel- und Kreuzblick



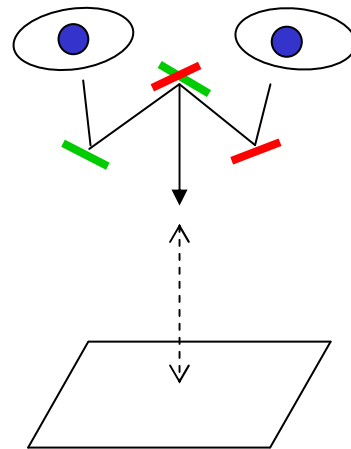


## Kreuzblick mit manueller Hilfe

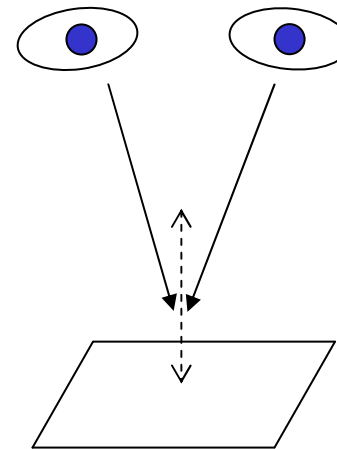
# Vergleich von normalem und Stereo-Mikroskop



Wenn man bei fixierter Optik ausschließlich die Distanz verändert, braucht man keine Größenkorrektur



Im Stereo-Mikroskop wandern Mittelpunkt und Blickwinkel mit dem Fokus





Stereo-Mikroskop,  
ohne Korrektur gestackt



x-/y-korrigiert  
& gestackt



Normales  
Mikroskop,  
gestackt