

X-ray microtomography

David Haberthür

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X-ray microtomography

Hello!

- Office B311 | haberthuer@ana.unibe.ch
- Master in Physics
- PhD in high resolution imaging of the lung at the Institute of Anatomy
- Post-Doc
 - TOMCAT, Swiss Light Source, Paul Scherrer Institute
 - μ CT-group, Institute of Anatomy (Ruslan Hlushchuk, David Haberthür, Oleksiy-Zakhar Khoma, Fluri Wieland, Carlos Correa Shokiche)
- Biomedical research
 - microangioCT [**Hlushchuk2018**]: Tumor vasculature, angiogenesis in the heart, musculature and bones
 - Cancer research: Melanoma
 - Lung imaging: Tumor detection and classification
 - Physiology: Zebrafish musculature and gills [**Messerli2019**]
 - SkyScan 1172 & 1272

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X-ray microtomography

Contents

Biomedical imaging

Imaging

- Tomography
- X-ray production
- Interaction of x-rays with matter
- History
- A scan, from start to finish
- Example
- Imaging performance

Image processing

- Image display

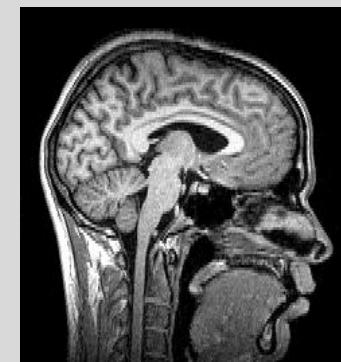
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X-ray microtomography

Biomedical imaging

- Medical research
- Non-destructive insights into the samples
- (Small) Biological samples

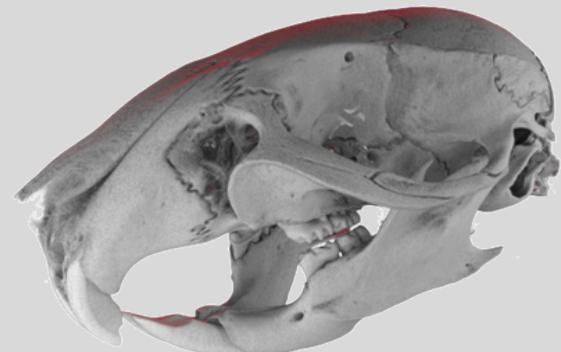


w.wiki/7g4

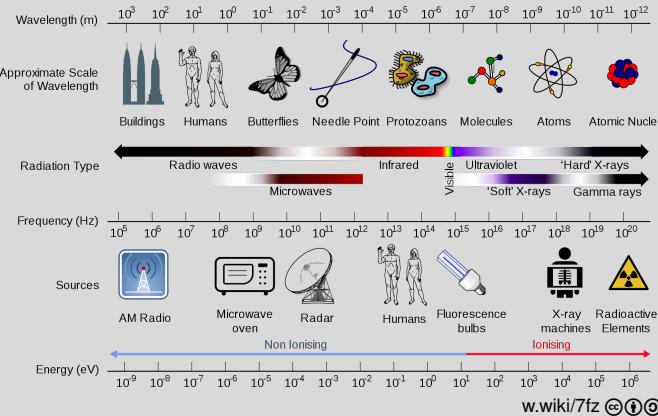
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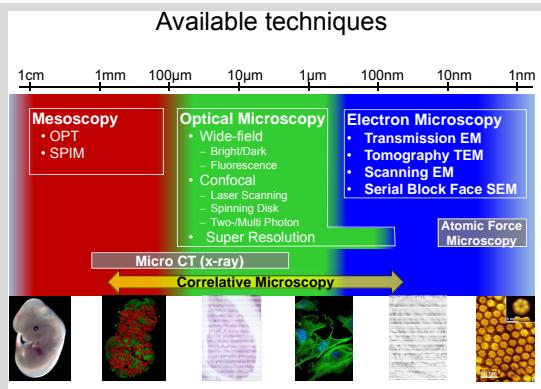
Biomedical imaging

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Wavelength & Scale

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Wavelength & Scale

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Imaging methods

- Light microscopy: see lecture of Nadia Mercader Huber
- X-ray imaging
- Electron microscopy: see lectures *Transmission Electron Microscopy* by Dimitri Vanhecke, *Scanning Electron Microscopy* by Michael Stoffel and *Cryo-electron Microscopy & Serial Block Face SEM* by Ioan Iacovache.

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CT-Scanner



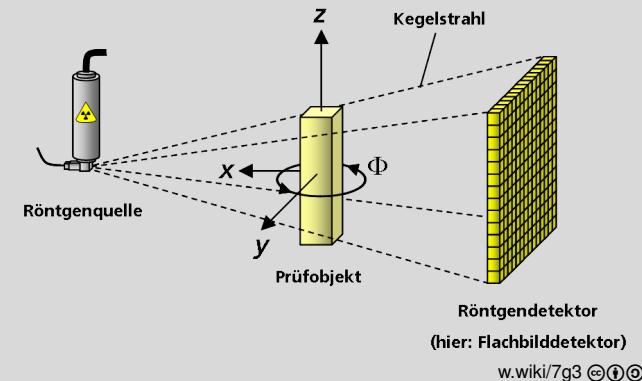
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What is happening?



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X-ray generation

- How are x-rays generated
- Why do we need them

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X-ray interaction

- "X-rays interact with tissue in 2 main ways: photoelectric effect and Compton scatter. To a first approximation, the photoelectric effect contributes to contrast while the Compton effect contributes to noise. Both contribute to dose." ([\[xrayphysics\]](#))
 - Photoelectric absorption (τ) is strongly dependent on the atomic number Z of the absorbing material: $\tau \propto \frac{Z^4}{E^{3.5}}$
 - Compton scattering is one of the principle forms of photon interaction and is directly proportional to the (electron & physical) density of the material. It does *not* depend on the atomic number: $\lambda' - \lambda = \frac{h}{m_e c} (1 - \cos \theta)$
 - Lowering x-ray energy increases contrast
 - X-ray penetration decreases exponentially with sample thickness ([\[wiki:beer-lambert\]](#))
 $I(t) = I_0 e^{-\alpha z}$

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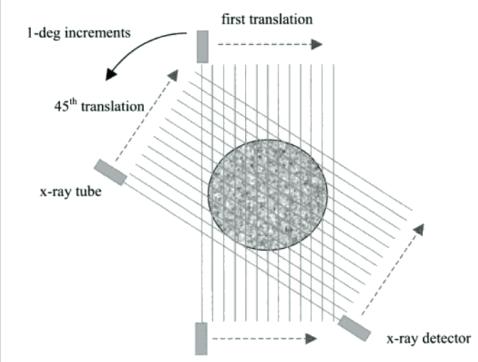
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Composition of biological tissues

- table with read csv
- booktabs

History

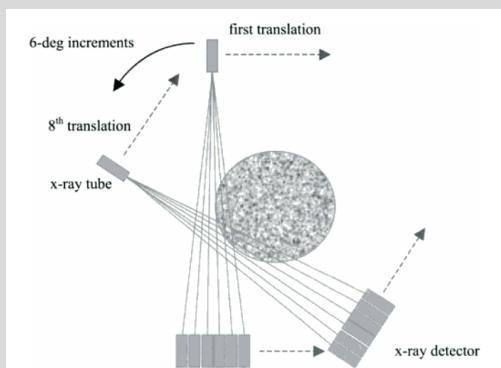
- Extensive history is found in [Cormack1963, Hsieh2003]
- First, second and third generation of scanners



From [Hsieh2003], Figure 1.12

History

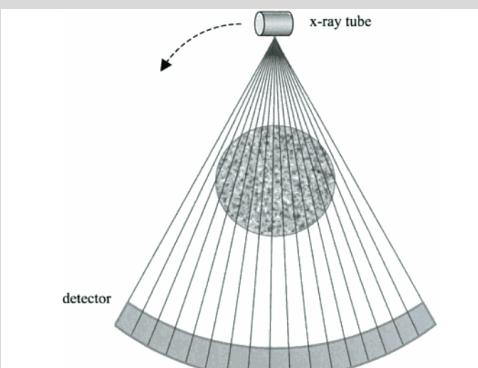
- Extensive history is found in [Cormack1963, Hsieh2003]
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From [Hsieh2003], Figure 1.13

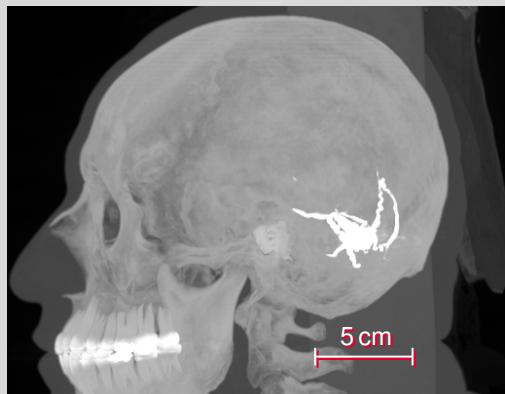
History

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From [Hsieh2003], Figure 1.14

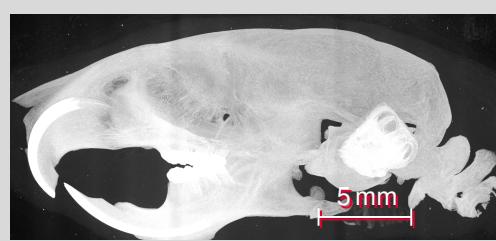
Why µCT?



From [Clark2013], Subject C3L-02465

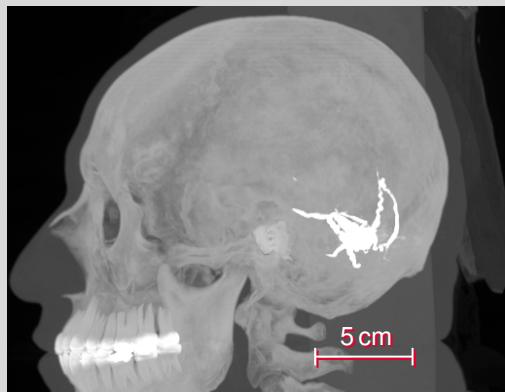
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Why µCT?



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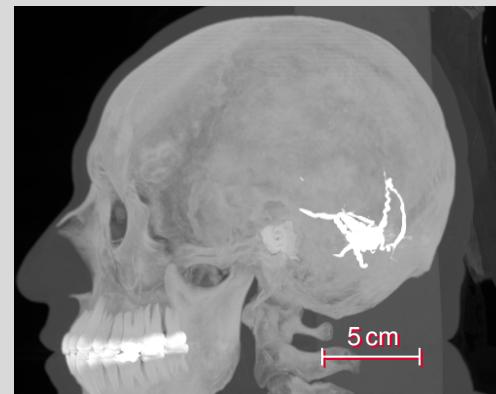
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Why µCT?



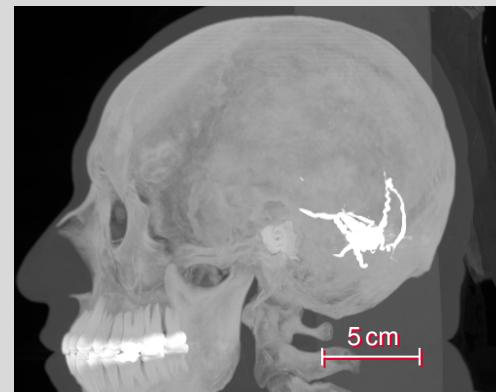
From [Clark2013], Subject C3L-02465

5 cm

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Why µCT?



From [Clark2013], Subject C3L-02465

5 mm

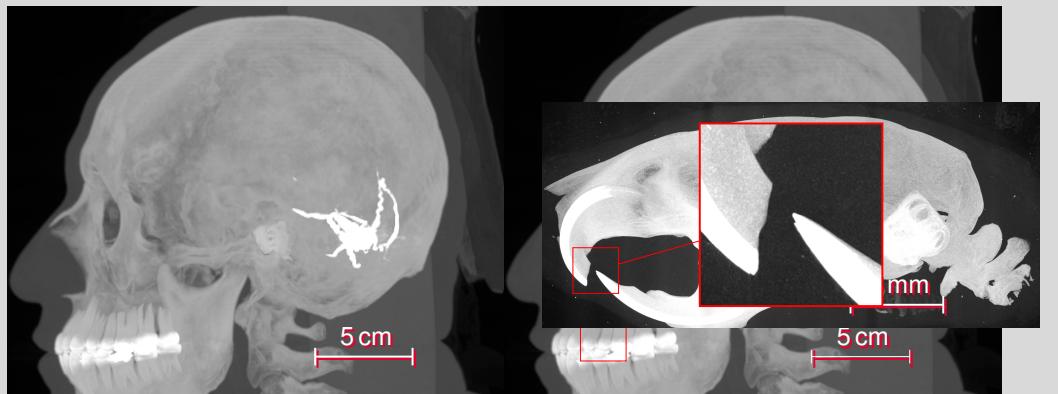
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Why µCT?



Machinery

- Hospital CT
 - Voxel size around 0.5 mm
- Lab/Desktop CT
 - Voxel size around 7 µm (*in vivo*) or 0.5 µm (*ex vivo*)
- Synchrotron CT
 - Voxel size down to 160 nm



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flic.kr/p/7Xhk2Y

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Machinery I

Independent on the machine, technically they are all a simple combination of

- an x-ray source
- a sample
- a detector

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Machinery II

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Preparation

- Study design
- Sample preparation

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Projections

Image formation: shadow projection

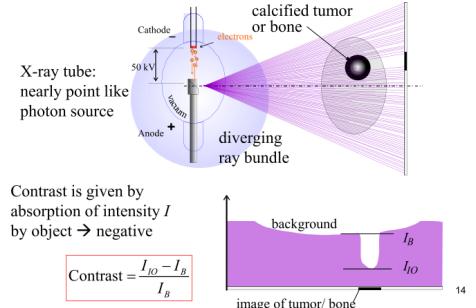
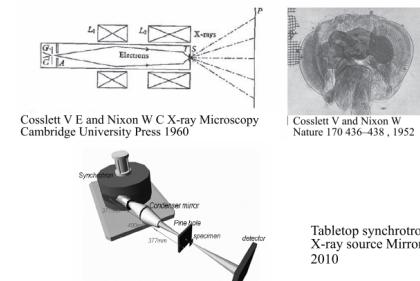


Image formation: shadow projection

X-ray projection microscopy



Laws of Physics for Microscopists by Michael Jaeger, Slide 14 Laws of Physics for Microscopists by Michael Jaeger, Slide 15

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Projections

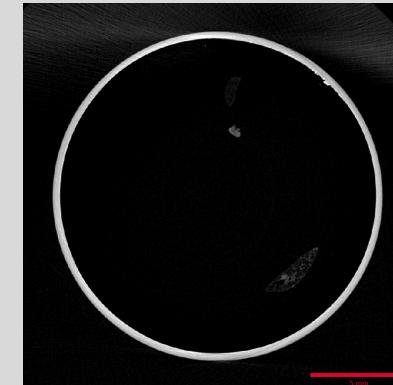
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Projections

- A (micro-focus) x-ray source illuminates the object
- The x-rays penetrate the sample and are attenuated
- A scintillator converts the x-rays to visible light
- A (planar) x-ray detector collects (magnified) projection images.
- The projections are recorded on disk

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Reconstructions

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Reconstructions

- Based on hundreds of angular views acquired while the object rotates, a computer synthesizes a stack of virtual cross section slices through the object.
- Radon Transformation
- Filtered back projection
- Fan beam reconstruction
- Corrections (beam hardening, etc.)
- Writing to stack

Visualization



Visualization

- Based on reconstructions, a computer synthesizes a three-dimensional view of the scanned sample

What to use?

- ImageJ [1]
- in Jupyter [[Kluyver2016](#)]
- Also see *Fundamentals of Digital Image Processing* by Guillaume Witz

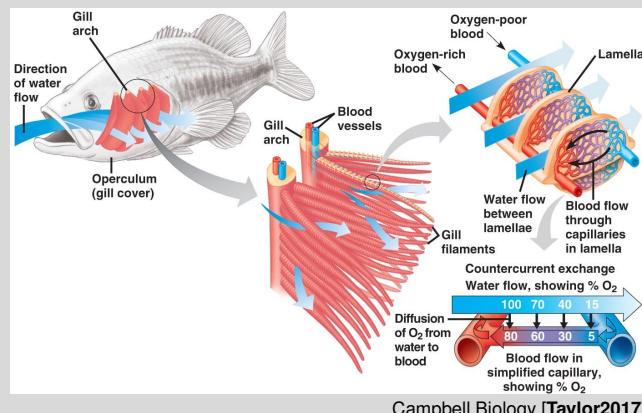
Quantitative data

- Raw numbers instead of just pretty images
- Segmentation
- Characterization

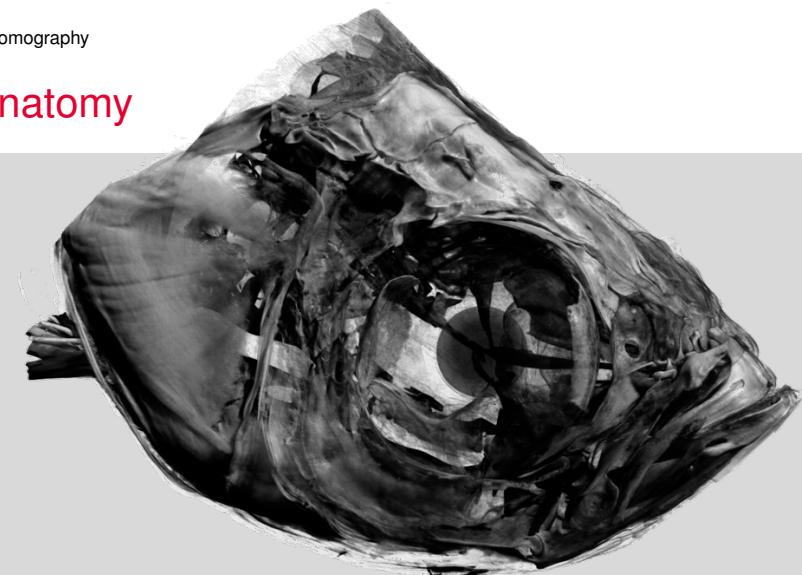
Reproducible research

- git
- Jupyter
- Script all your things!
- Data repositories → Sharing is caring!

An example: Do gills change with training?



Gill anatomy

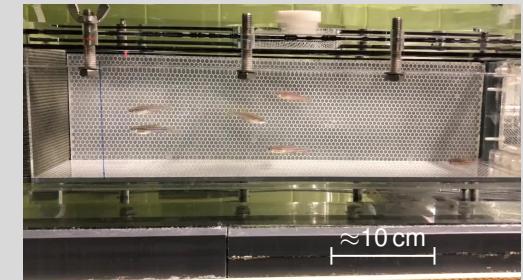


Gill anatomy

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How?

- Training for 5 w, 5 d/w, 6 h/d, at 66 % of critical speed [Palstra2010]
 - Endurance



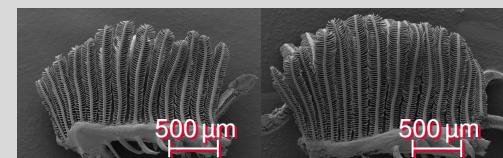
How?

- Training for 5 w, 5 d/w, 6 h/d, at 66 % of critical speed [Palstra2010]
 - Endurance
- Morphology & Physiology
 - Body size & weight
 - O₂ consumption

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How?

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- Scanning electron microscopy
 - Gill structure



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- Critical point drying



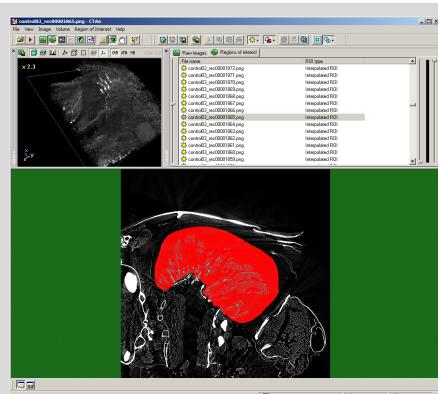
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- Scanning electron microscopy
 - Gill structure
- Critical point drying, µCT imaging

```
Scanner=Skyscan1172
Instrument S/N=12001199
Hardware version=F
Software=Version 1. 5 (build 23)
Filename Prefix=Control05
Number of Files= 3979
Source Voltage (kV)= 49
Source Current (uA)= 167
Number of Rows= 2672
Number of Columns= 4000
Image Pixel Size (um)= 1.66
Object to Source (mm)=40.030
Camera to Source (mm)=212.399
Filter=No Filter
Exposure (ms)= 890
Rotation Step (deg)=0.050
Frame Averaging-ON (6)
Scan duration=08:55:28
Reconstruction Program=NRecon
Program Version=Version: 1.7.1.0
```

How?

- Training for 5 w, 5 d/w, 6 h/d, at 66 % of critical speed [Palstra2010]
 - Endurance
- Morphology & Physiology
 - Body size & weight
 - O₂ consumption
- Scanning electron microscopy
 - Gill structure
- Critical point drying, µCT imaging, delineation in CTAn



How?

- Training for 5 w, 5 d/w, 6 h/d, at 66 % of critical speed [Palstra2010]
 - Endurance
- Morphology & Physiology
 - Body size & weight
 - O₂ consumption
- Scanning electron microscopy
 - Gill structure
- Critical point drying, µCT imaging, delineation in CTAn and analysis
 - Gill volume, structure and complexity



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Gill volume

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X-ray microtomography

Gill volume

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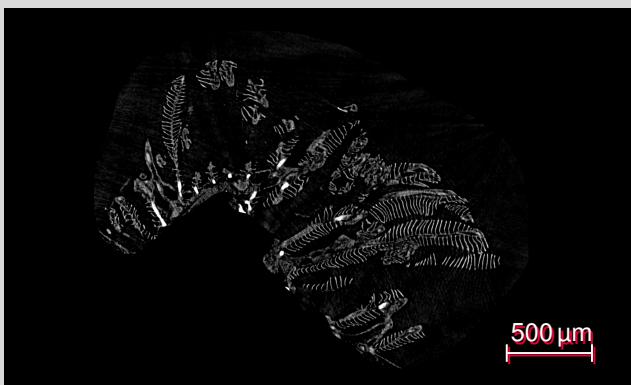
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X-ray microtomography

Gill volume

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X-ray microtomography

Gill volume

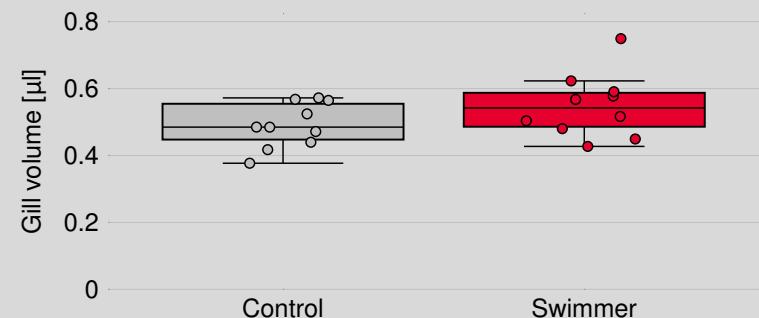
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Gill volume



Gill complexity

500 μm

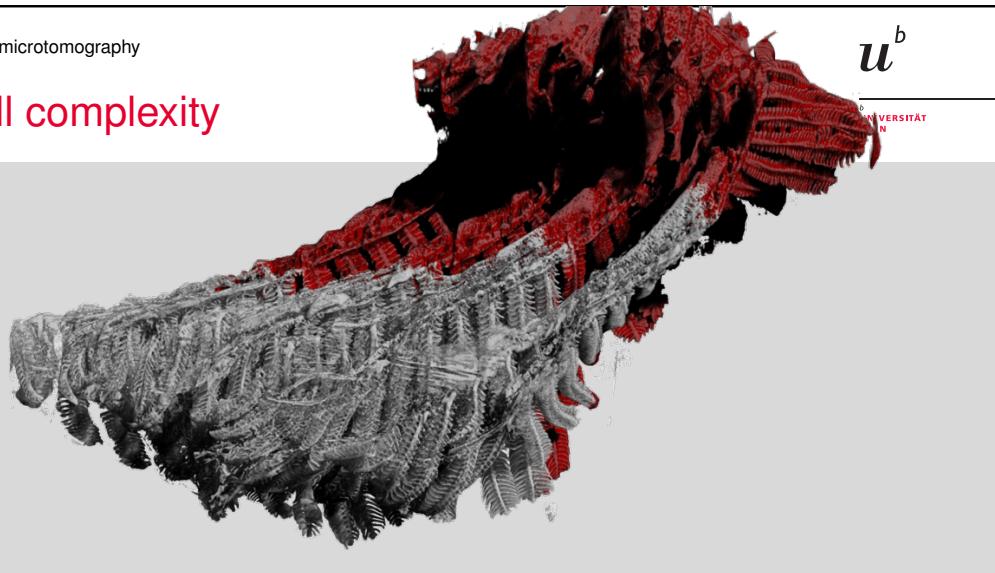
Gill complexity

500 μm

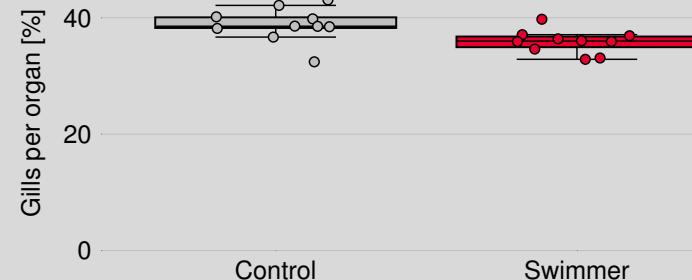
Gill complexity

500 μm

Gill complexity



Gill complexity



Colophon

- This BEAMER presentation was crafted in \LaTeX with the (slightly adapted) template from *Corporate Design und Vorlagen* of the University of Bern.
 - Full source code: git.io/fjpP7
 - The \LaTeX code is automatically compiled with a GitHub action [2] to a PDF which you can access here: git.io/JeMjP
 - Spotted an error?
Then please file an issue (git.io/fjpPb) or (even better) submit a pull request (git.io/fjpPN).

[2] Details on how this works are specified in a small test repository here: git.io/JeOOj

References