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**UNIVERSITY
OF BERN**

u^b X-ray microtomography

485018-HS2025-0: Advanced Course II Ultraprecision Engineering

David Haberthür

Institute of Anatomy, September 16, 2025

u^b Grüessech mitenang!

- David Haberthür
 - Physicist by trade
 - PhD in high resolution imaging of the lung, Institute of Anatomy, University of Bern, Switzerland
 - Post-Doc I: TOMCAT, Swiss Light Source, Paul Scherrer Institute, Switzerland
 - Post-Doc II: µCT group, Institute of Anatomy, University of Bern, Switzerland

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Grüessech from the µCT group



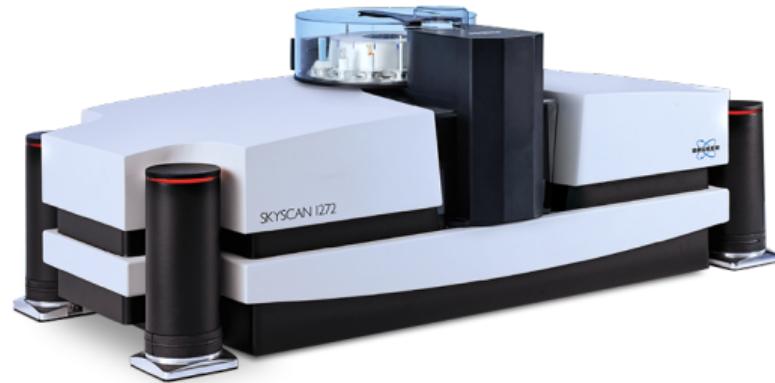
David.Haberthuer@unibe.ch

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Oleksiy.Khoma@unibe.ch

μ CT-group

- microangioCT [1]
 - Angiogenesis: heart, musculature [2] and bones
 - Vasculature: (mouse) brain [3], (human) nerve scaffolds [4], (human) skin flaps [5] and tumors
- Zebrafish musculature and gills [6]
- (Lung) tumor detection and metastasis classification [7]
- Collaborations with museums [8] and scientist at UniBe [9] to scan a wide range of specimens
- Automate *all* the things! [10, 11]



bruker.com/skyscan1272

Contents

Overview & Imaging methods

Tomography

History

Interaction of x-rays with matter

Tomography today

A scan, from *getting started* to *nice image*

Examples

A study about teeth

Overview

Materials & Methods

Results

Metal foam analysis

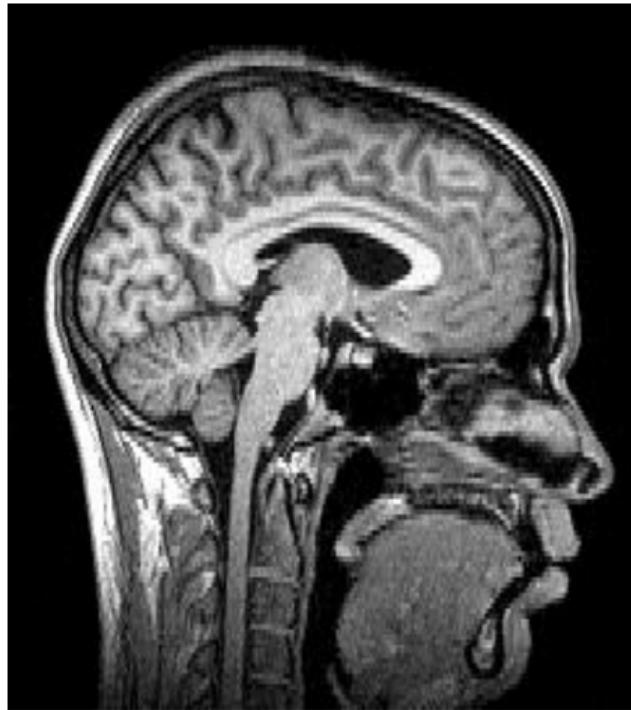
A study on fish

- Dense and/or non-transparent samples
- Calibrated & isotropic 3D images at micron resolutions
- Covers a very large range of sample sizes
- Gives information at different length scales
- Nondestructive imaging, thus compatible with routine sample preparation.
Enables correlative imaging pipelines, scanning of museum & collection material

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Biomedical imaging

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



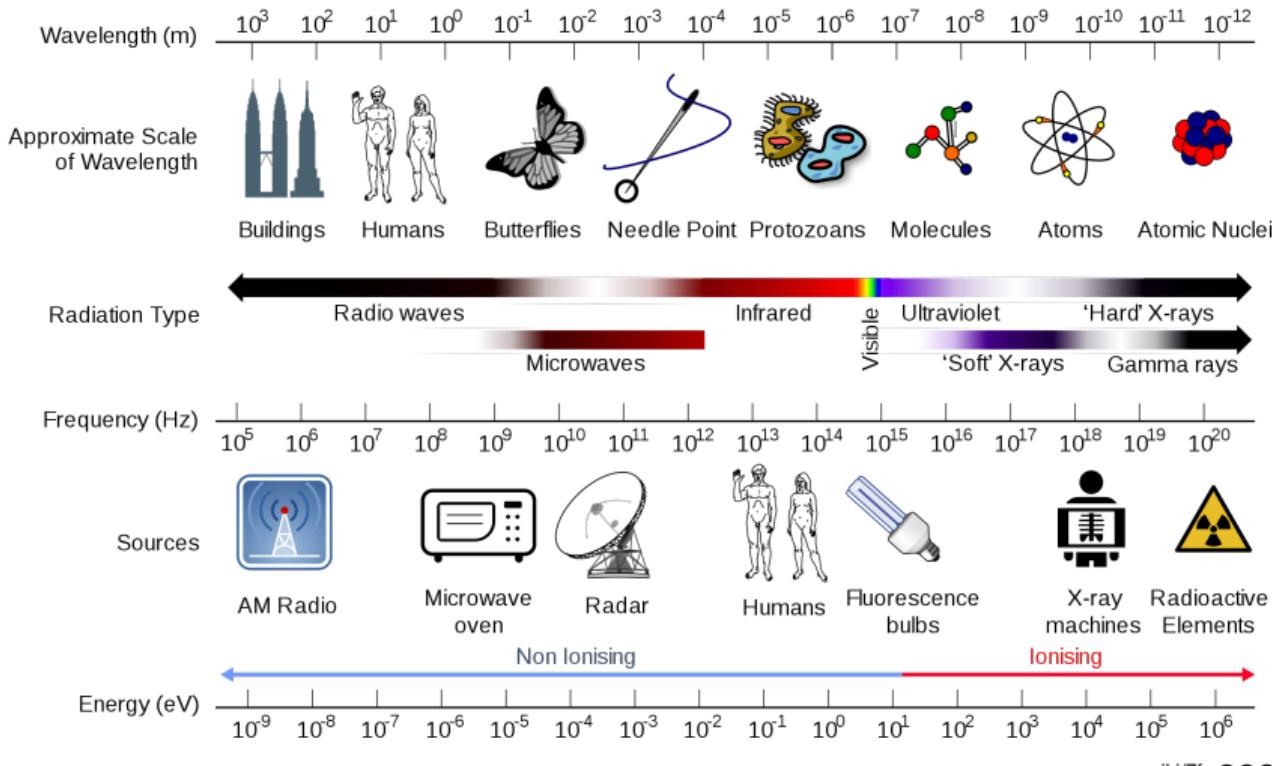
w.wiki/7g4 CC BY-NC-SA

Biomedical imaging

- Medical research
- Non-destructive
- (Small) Biology

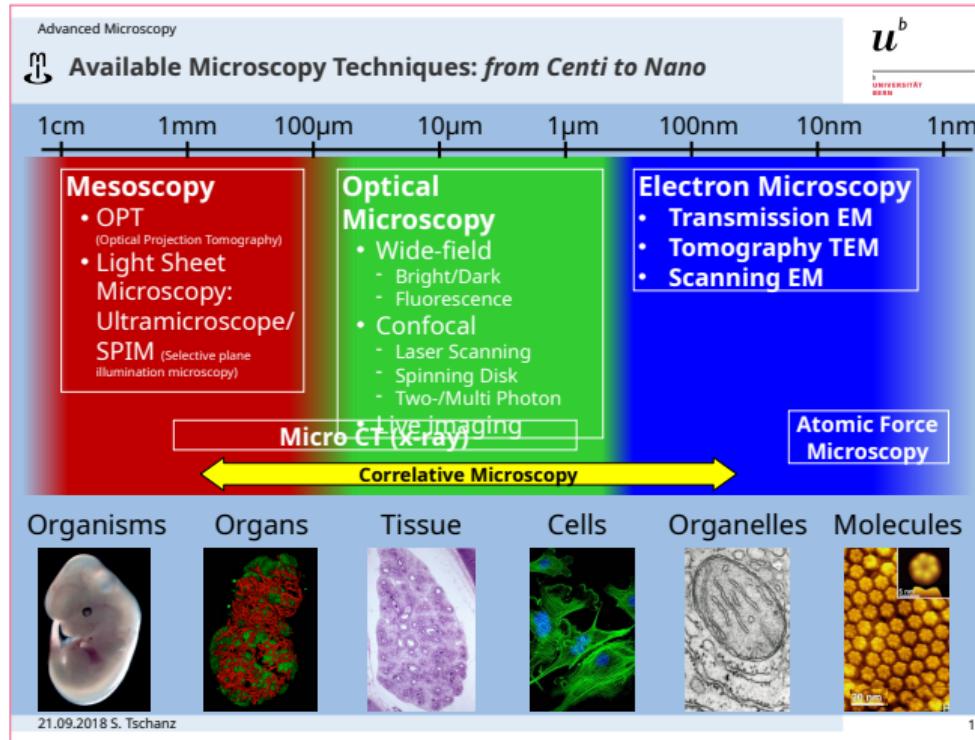


u^b Wavelengths & Scales



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u^b Wavelengths & Scales



Introduction to Microscopy by Stefan Tschanz, Slide 1

Imaging methods

- Light (sheet) microscopy: see lecture of Nadia Mercader Huber
- X-ray imaging
- Electron microscopy
 - *Analytical electron microscopy* by Dimitri
 - *SEM Grundlagen* by Sabine Kässmeyer and Ivana Jaric
 - *Cryoelectron Microscopy & Serial Block Face SEM* by Ioan

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CT-Scanner



youtu.be/2CWpZKuy-NE

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CT History

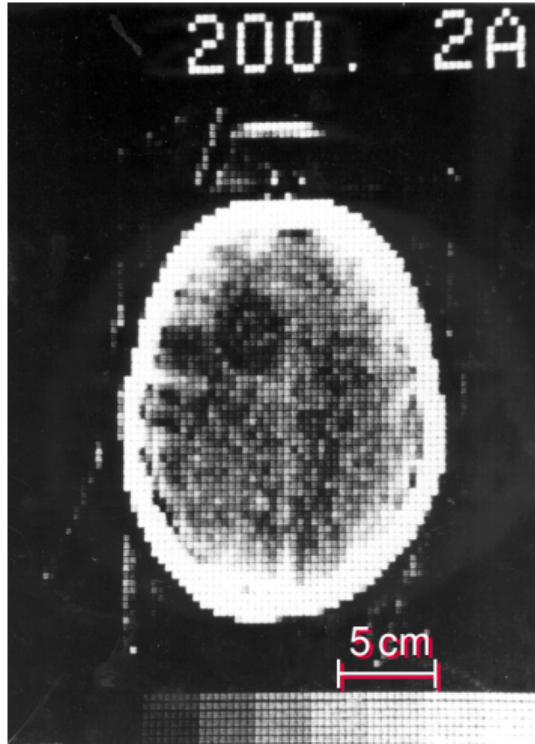
- 1895: Wilhelm Conrad Röntgen discovers X-rays



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CT History

- 1895: Wilhelm Conrad Röntgen discovers X-rays
- 1963: Cormack used a collimated ^{60}Co source and a Geiger counter as a detector [12]
- 1976: Hounsfield worked on first clinical scanner [13]



From [14], Figure 5

CT History

- 1895: Wilhelm Conrad Röntgen discovers X-rays
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- Nobel Prize in 1979, jointly for Allan Cormack and Godfrey Hounsfield

U.S. Patent Feb. 24, 1976 Sheet 1 of 2 3,940,625

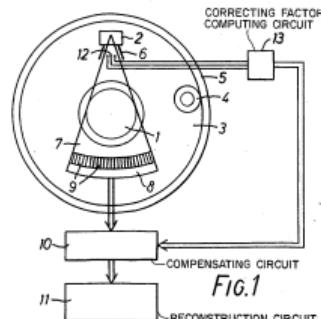


Fig.1

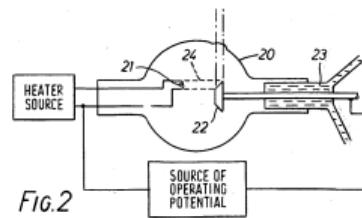
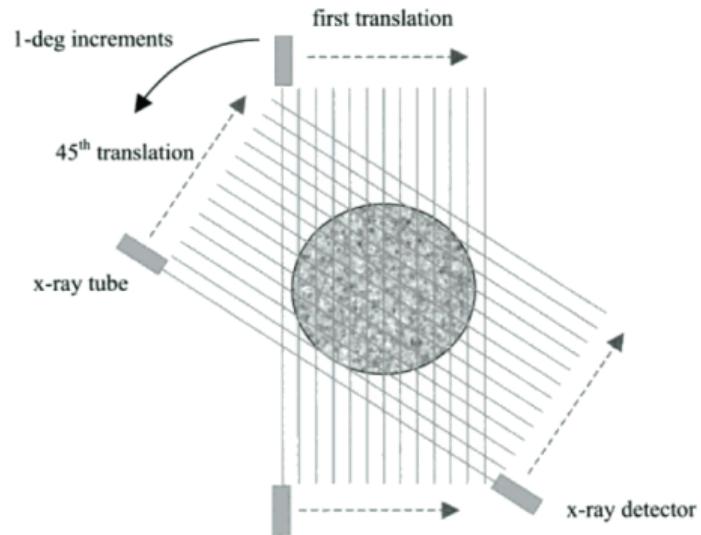


Fig.2

CT History

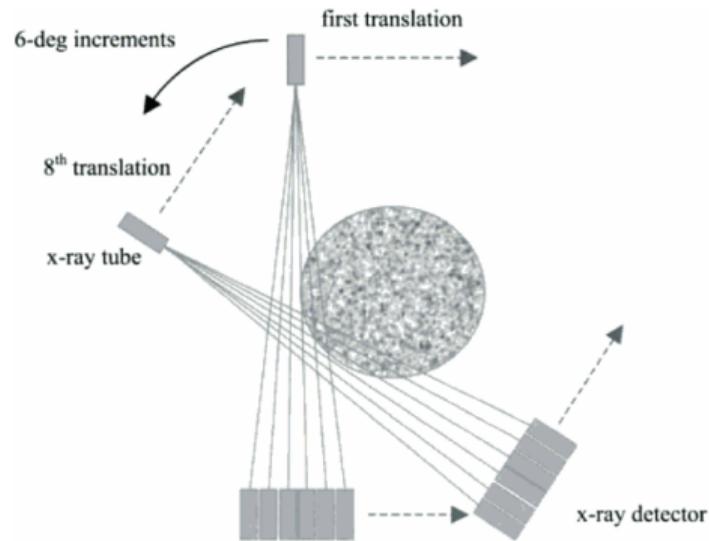
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- CT scanner generations
 - First generation



From [16], Figure 1.12

CT History

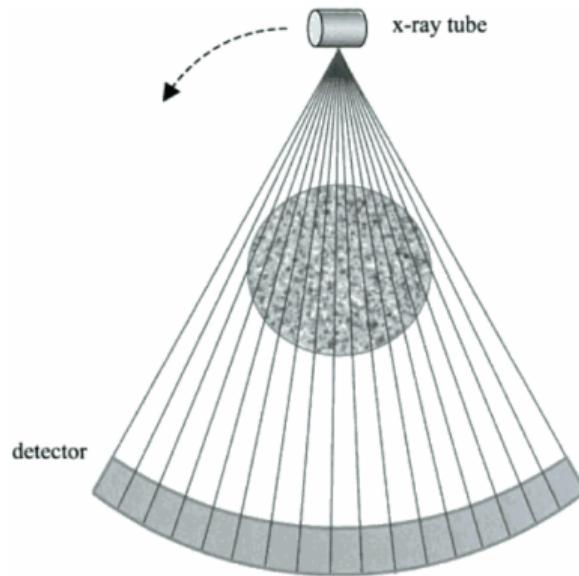
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 - Second generation



From [16], Figure 1.13

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- CT scanner generations
 - First generation
 - Second generation
 - Third generation



From [16], Figure 1.14

μ CT History I

- X-ray computed tomography began to replace analog focal plane tomography in the early 1970s [17]
- Non-medical use in the late 1970s, for detection of internal defects in fabricated parts and equipment
- Lee Feldkamp [18] developed an early laboratory microCT system by assembling a micro-focus cone beam x-ray source, specimen holder and stages, and an image intensifier at Ford Motor Company's Scientific Research Laboratory to nondestructively detect damage in ceramic manufactured automobile parts
- Feldkamp met with scientists at Henry Ford Hospital and University of Michigan interested in understanding the relationship between the microstructure and biomechanical function of trabecular bone to study osteoporotic fractures [19]

μ CT History II

- μ CT was first reported in the 1980s, for scanning gemstones
- Early 1990s: Manufacturers like SkyScan and Scanco Medical made μ CT systems commercially available
- Today: Nondestructive imaging for quantifying the (micro)structure of (organic) materials
 - Mineralized bone tissue and the relationships between the mechanical behavior of bone to its structural and compositional properties
 - Teeth and their internal details
 - Tissues, small animals, and medical devices like stents and implants
 - Soft tissues and vasculature using radio-opaque contrast agents
 - Characterization of anatomical details in high resolution
- \approx 2500 μ CT systems are in use worldwide with over 1000 publications annually

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.” ([20])
 - 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 [21, i. e. Beer-Lambers law]: $I(t) = I_0 e^{-\alpha z}$

Composition of biological tissues

Tissue: content by mass percentage

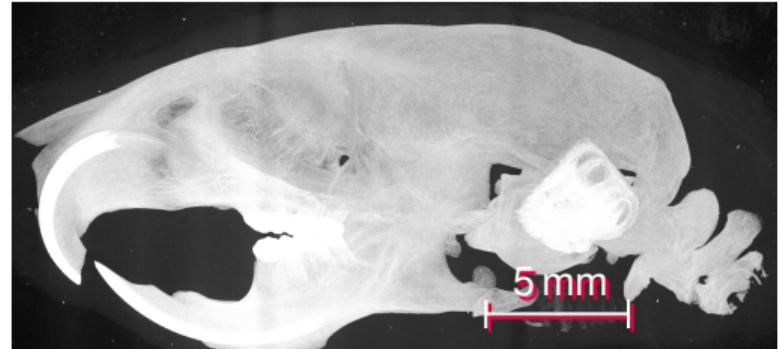
Element Atomic number	H 1	C 6	N 7	O 8	Na 11	P 15	S 16	Cl 17	K 19	Ca 20
Fat	11.4	59.8	0.7	27.8	0.1		0.1	0.1		
Water	11.2			88.8						
Blood	10.2	11	3.3	74.5	0.1	0.1	0.2	0.3	0.2	
Liver	10.2	13.9	3	71.6	0.3	0.2	0.3	0.2	0.3	
Brain	10.7	14.5	2.2	71.2	0.2	0.4	0.2	0.3	0.3	
Bone	3.4	15.5	4.2	43.5	0.1	10.3	0.3			22.5

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Why μ CT?



From [22], Subject C3L-02465



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Why μ CT?



From [22], Subject C3L-02465

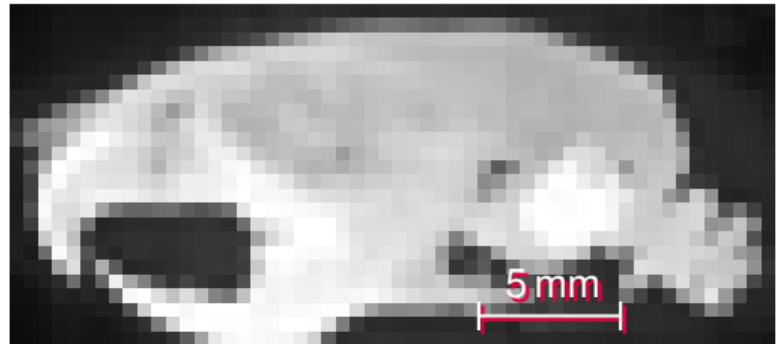


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Why μ CT?



From [22], Subject C3L-02465

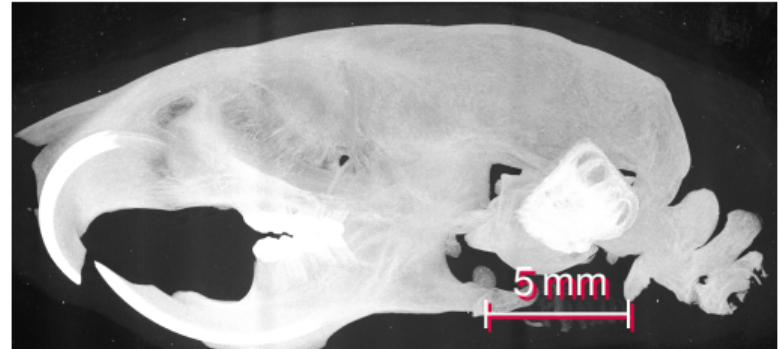


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Why μ CT?

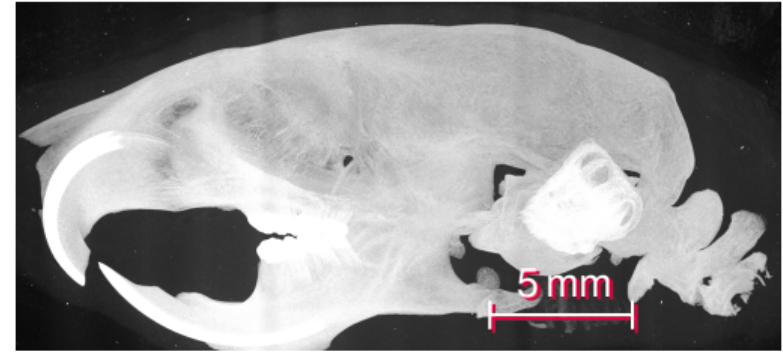
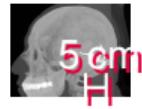


From [22], Subject C3L-02465



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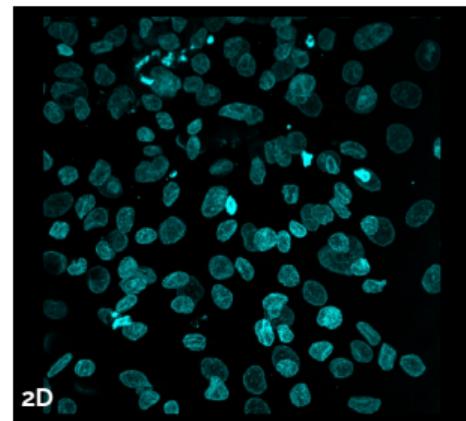
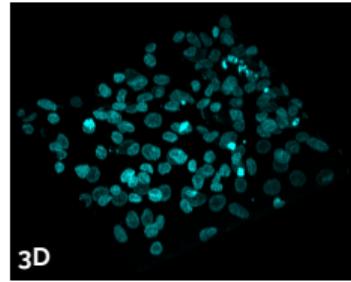
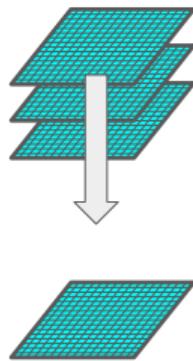
Why μ CT?



Maximum intensity projection

Projections

Reducing the dimensions of a dataset. For example projecting a volume (3D) to a surface by taking the maximum value across planes for each pixel.



Machinery

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



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Machinery

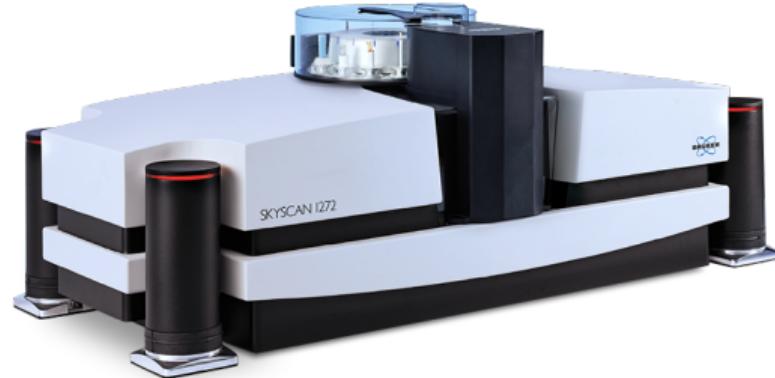
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Machinery

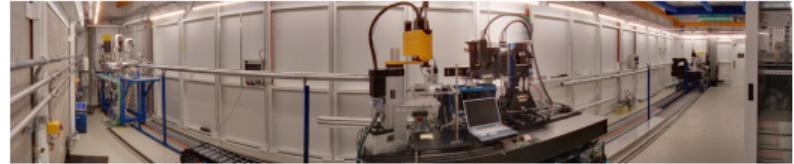
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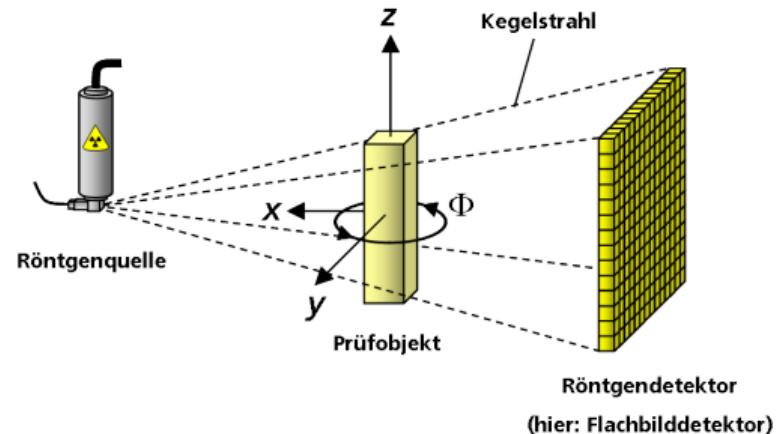
flic.kr/p/7Xhk2Y @CC BY-NC-SA

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What is happening?

No matter what kind of machine, the basic principle is always

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



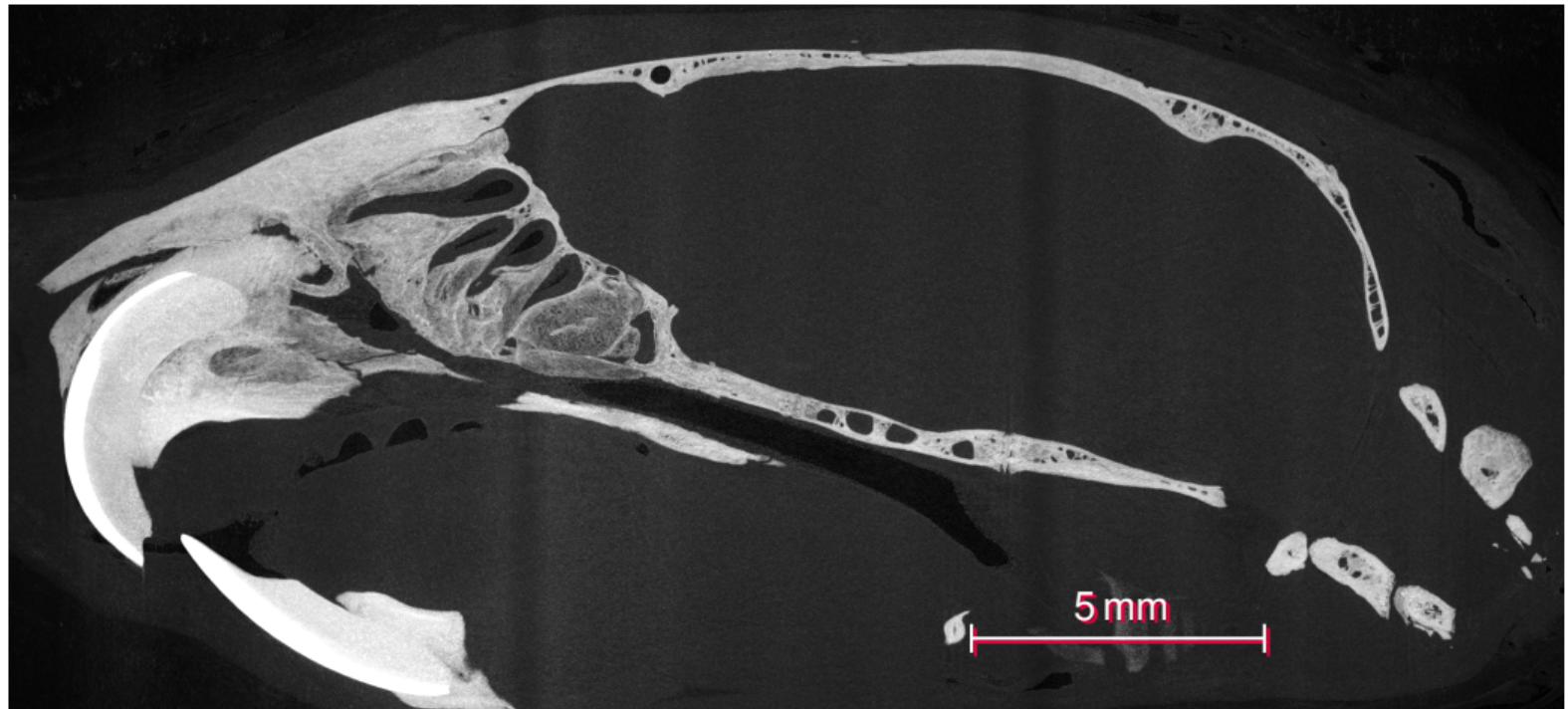
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Machinery

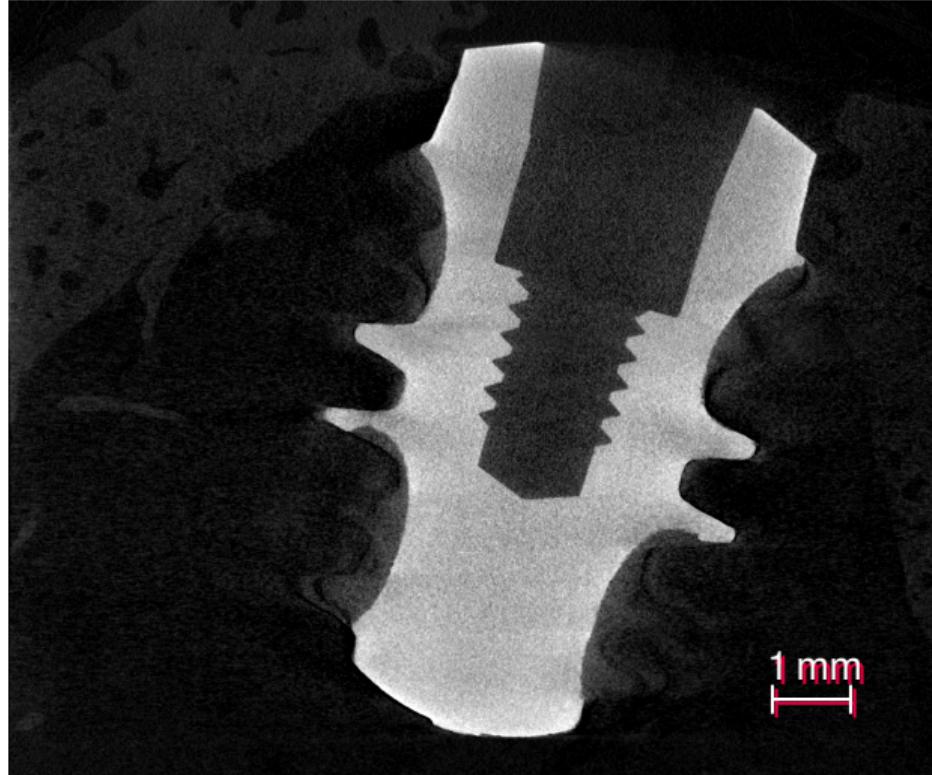
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Examples



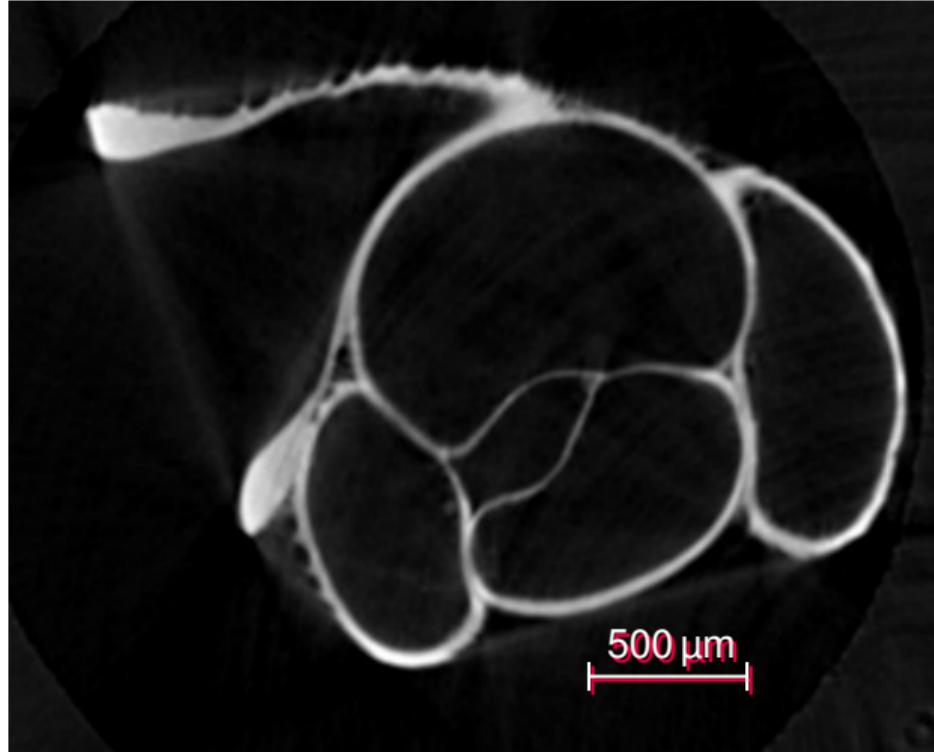
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Examples



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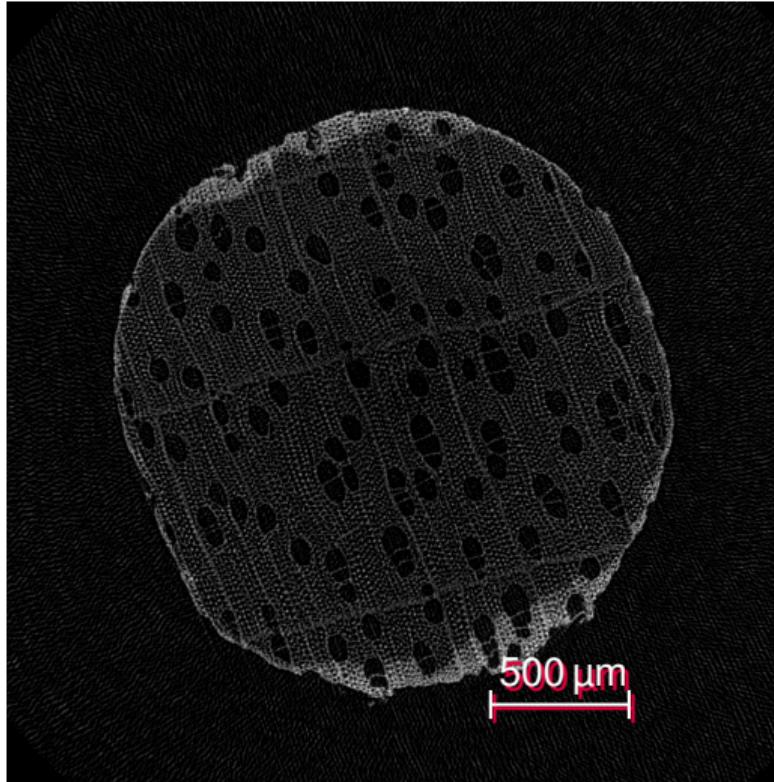
Examples



From [8], *Diancta phoenix*

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Examples



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Examples



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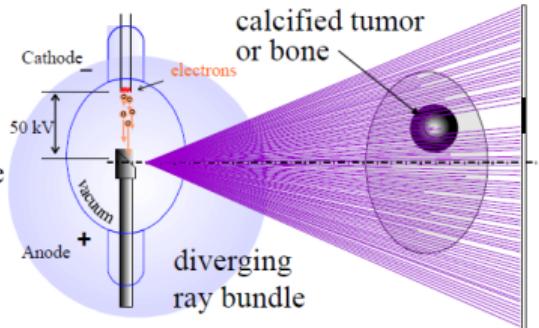
Preparation

- Study design
- Sample preparation

Projections

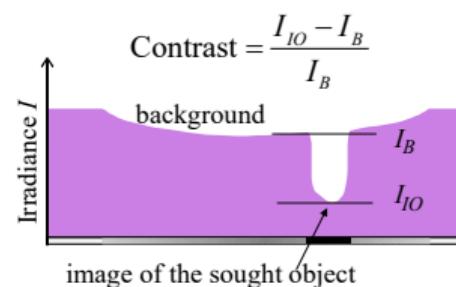
X-ray generation and contrast

X-ray tube:
nearly point like
photon source



Contrast is given by
absorption of intensity I

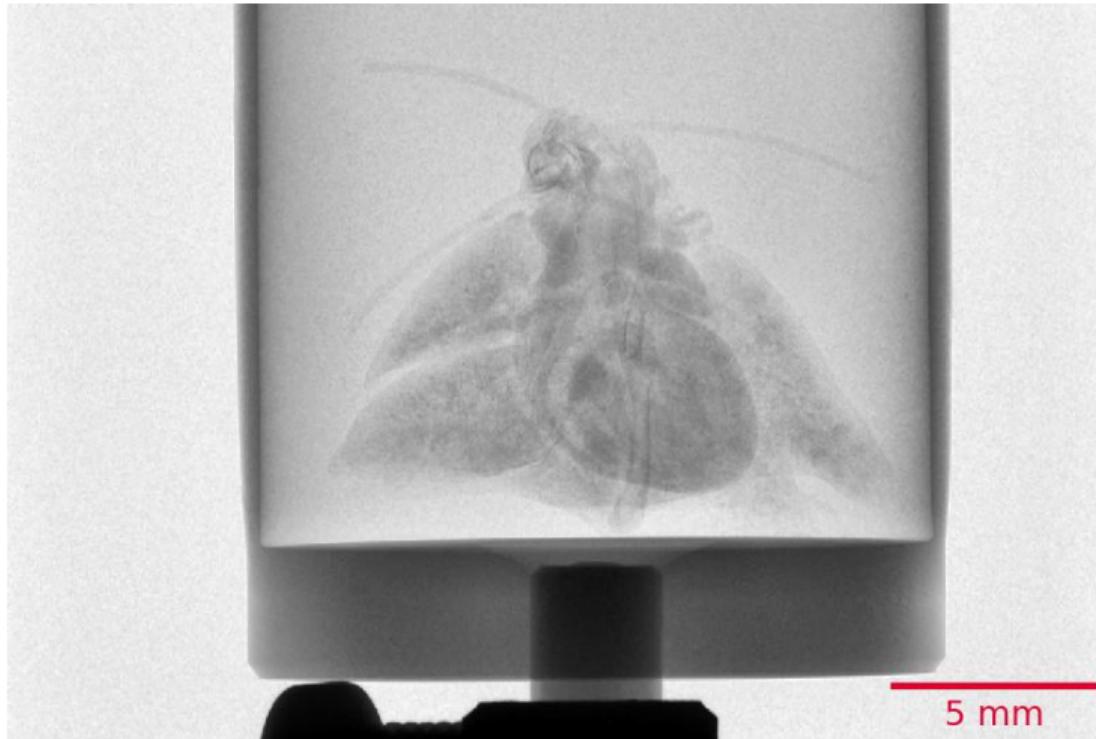
Note that contrast is negative
X-ray shadowgraphy
is a bright field technique



Contrast, Magnification and Resolution—Laws of Physics for Microscopists (1, 2022) by Martin Frenz, Slide 21

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Projections



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



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
- Cone beam reconstruction [18]
- Corrections (beam hardening, etc.)
- Writing to stack

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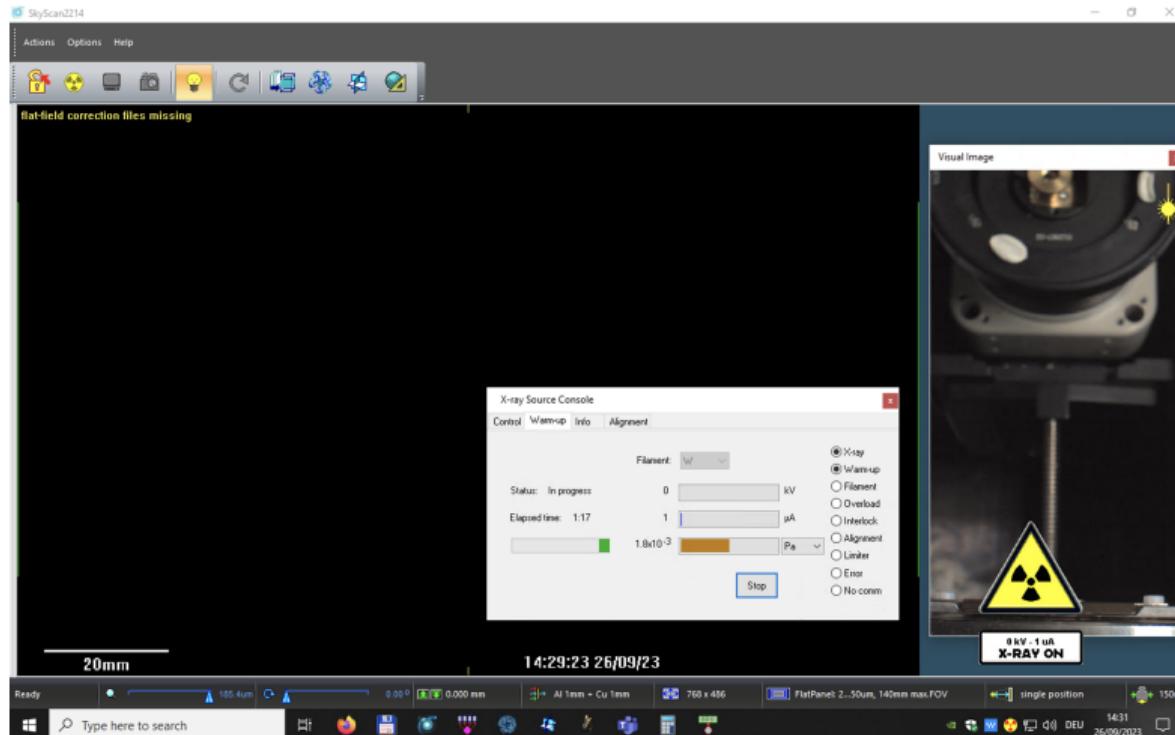
Visualization



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

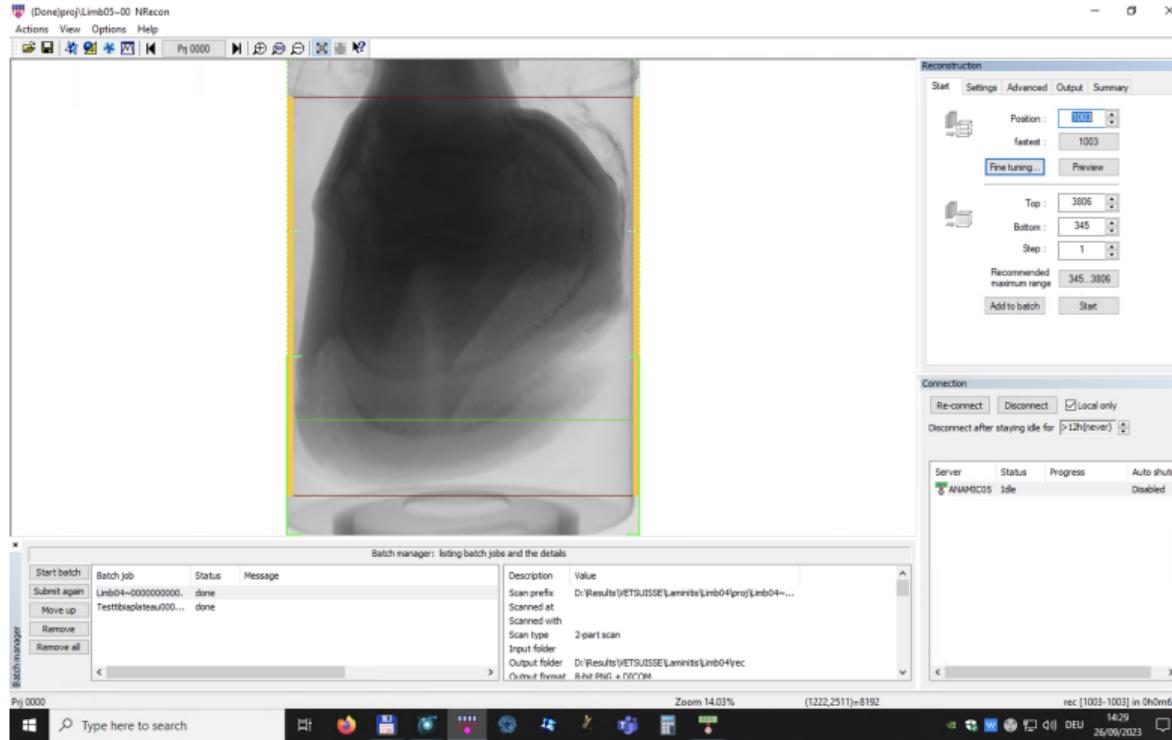
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Demo: Acquisition



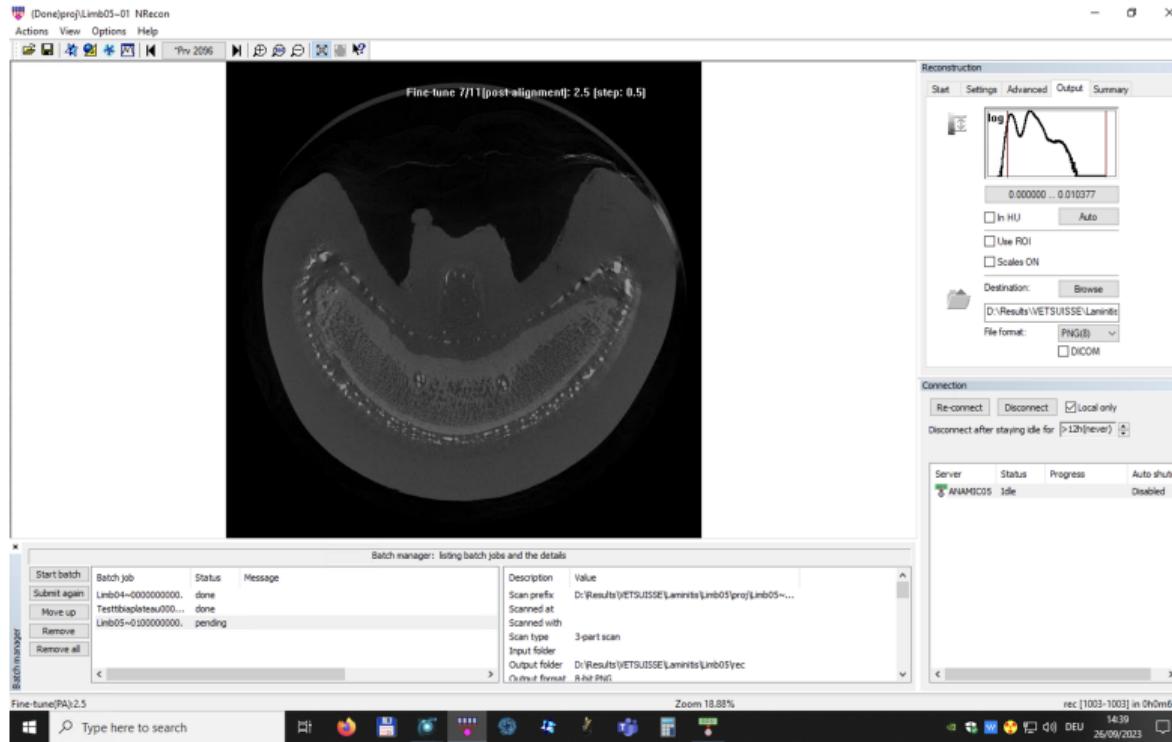
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Demo: Reconstruction



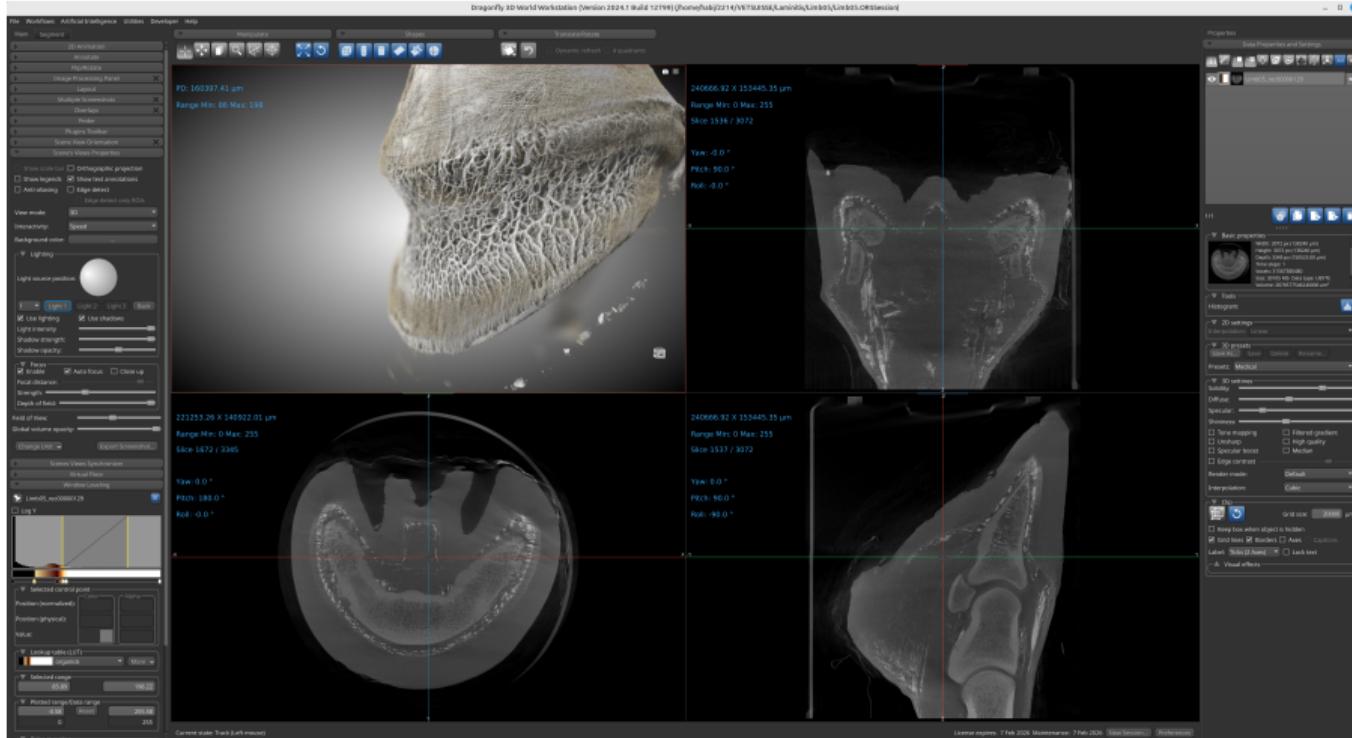
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Demo: Reconstruction



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Demo: Visualization



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What to use?

- ImageJ/Fiji [**Schindelin2012**]
- Also see *Fundamentals of Digital Image Processing* by Guillaume Witz
- Reproducible research
 -  in Jupyter [**Kluyver2016**]
 - **git**
 - Script all your things!
 - Data repositories; i. e. sharing is caring!

Quantitative data

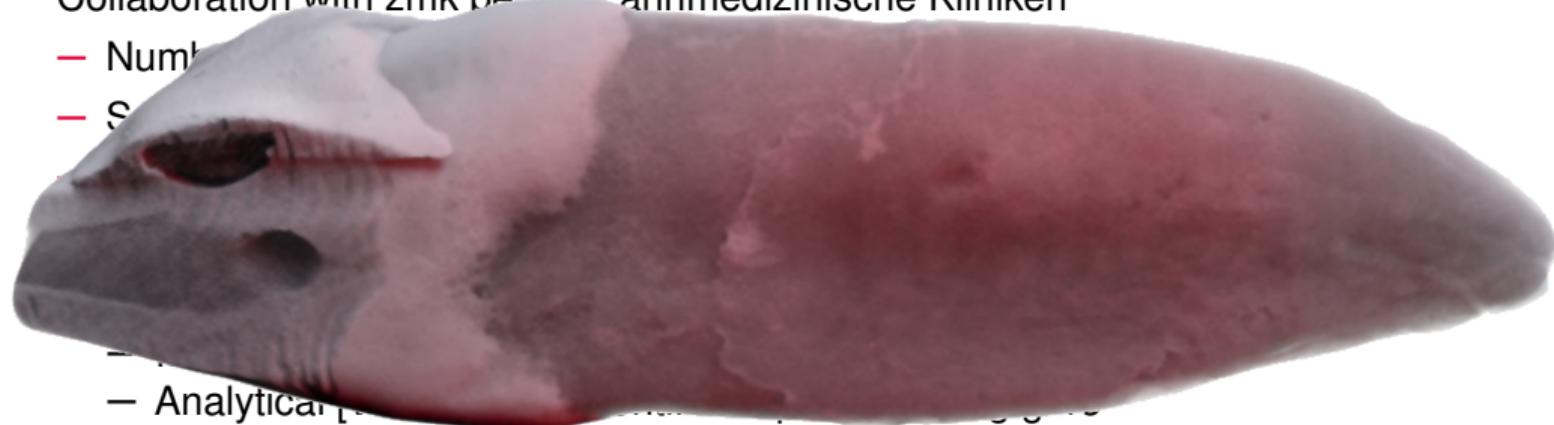
- Pretty images are nice to have, but science is built on quantitative data.
- Segmentation
- Characterization

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Internal morphology of human teeth

Collaboration with zmk bern – Zahnmedizinische Kliniken

- Number
- Structure
- Function
- Analytical



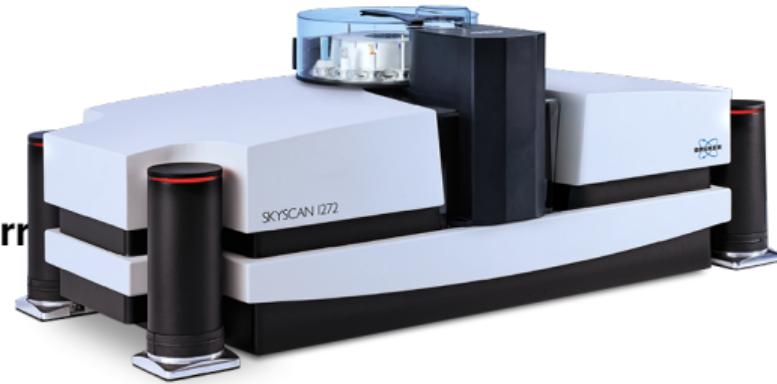
How?

- 104 extracted human permanent mandibular canines
- µCT imaging
- Root canal configuration, according to **Briseno-Marroquin2015 [Briseno-Marroquin2015]**
- *Reproducible* analysis [**Haberthuer2020a**], e. g. you can click a button to double-check or recalculate the results yourself!



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```
Scanner=SkyScan1272
Instrument S/N=15G09089-B
Software Version=1.1.19
Filename Prefix=Tooth045~00
Number Of Files= 482
Number Of Rows= 1092
Number Of Columns= 1632
Source Voltage (kV)= 80
Source Current (uA)= 125
Image Pixel Size (um)=9.999986
Exposure (ms)=950
Rotation Step (deg)=0.400
Frame Averaging=ON (3)
Filter=Al 1mm
Study Date and Time=02 Jul 2020 08h:23m:34
s
Scan duration=0h:39m:51s
```

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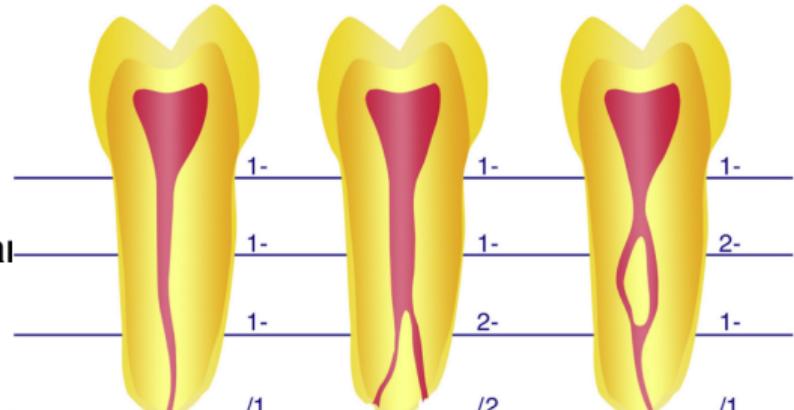
Sample changer on the SkyScan 1272

In total:

- 13 days of *continuous* μ CT scanning
- 819 GB of raw data
- 230 648 TIFF projections
- 326 GB data as input for analysis
- 282 062 PNG reconstructions

How?

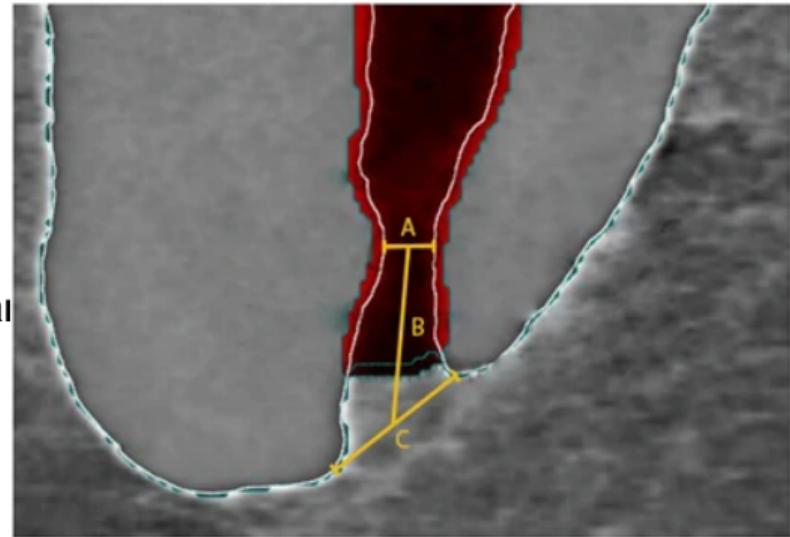
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From [Briseno-Marroquin2015], Fig. 2

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From [Wolf2017], Fig. 1

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gph.is/2nqkple

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The screenshot shows a GitHub repository interface. At the top, there are buttons for 'master', 'branch', 'tag', 'Go to file', 'Add file', and 'Code'. Below this is a list of files with their commit history:

File	Description	Last Commit
ghibli/actions.yml	Update actions file	20 days ago
ghigraze	Only 'model' changes	2 months ago
DownloadFromOSF.ipynb	Clean run of download script	22 days ago
README.md	Type in Binder badge & link to full repo on Binder	22 days ago
Tooth.Border.jpg	Only 'model' changes	2 months ago
Tooth.Characterization.jpg	Only 'model' changes	2 months ago
ToothAnalysis.ipynb	Only select a subset if we actually have data now	22 days ago
ToothDatabase.sqlite	Clean run of notebook	22 days ago
ToothDisplay.ipynb	Display Tooth048 for manuscript	22 days ago
requirements.txt	We also need this	2 months ago
treeboard.yaml	Add treeboard configuration	20 days ago

Below the file list is a 'README.md' page with the following content:

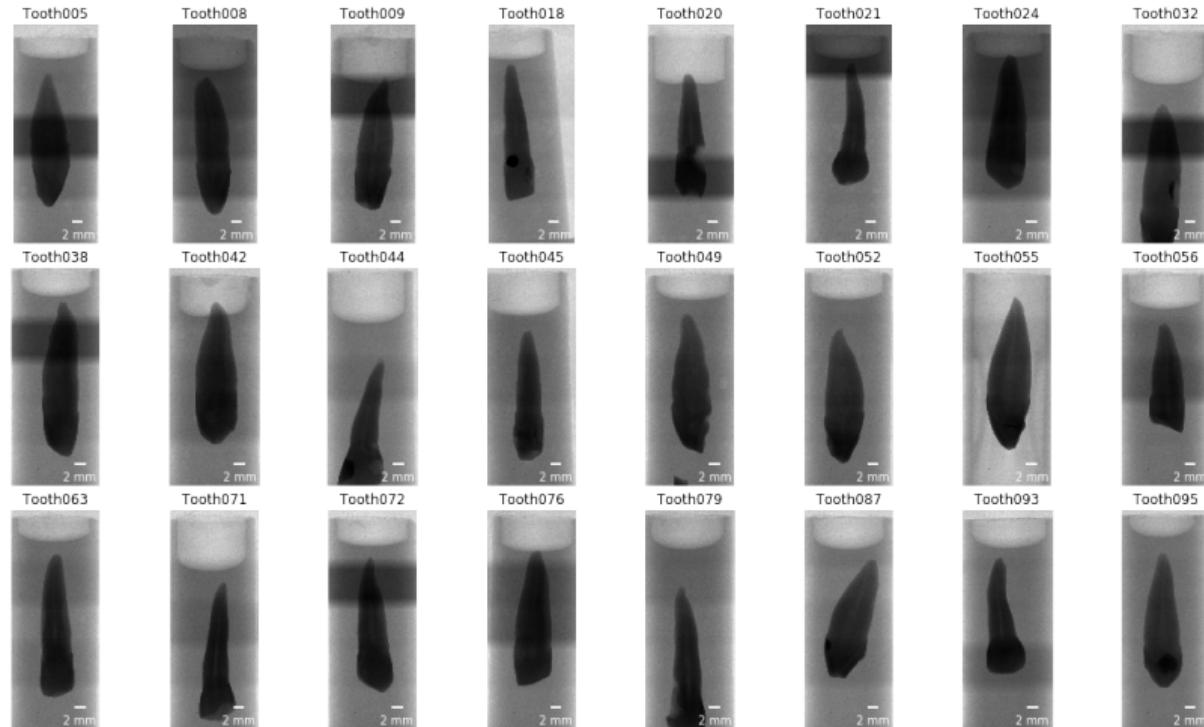
A big tooth cohort

We scanned a big bunch of teeth for a team of the dental clinic of the University of Bern.

To get an overview of the samples while we scanned the whole tooth cohort we generated a [preview](#) and [analysis notebook](#). The analysis notebook (with download possibility for two of the +100 teeth) can be started in your browser by clicking on the 'Binder' badge above, without installation of any software. If you'd like to start a Binder Instance with the full repository, you can click [here](#).

μ b

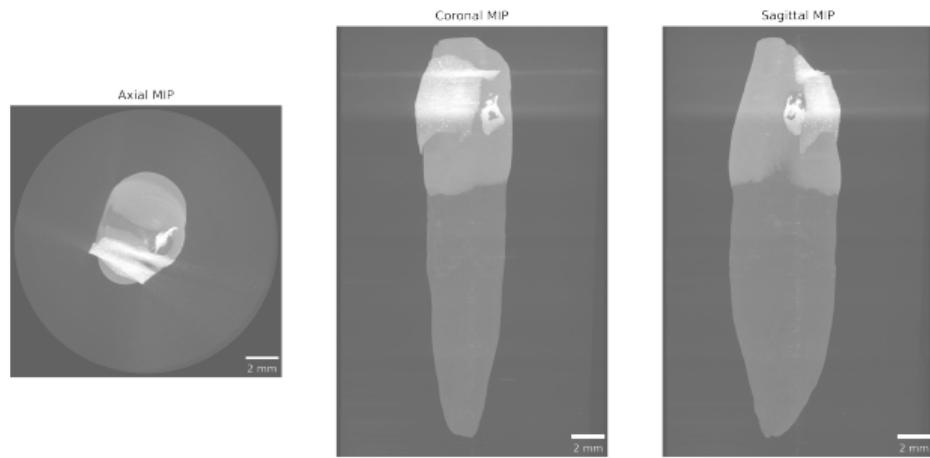
μ CT imaging



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Dataset cropping

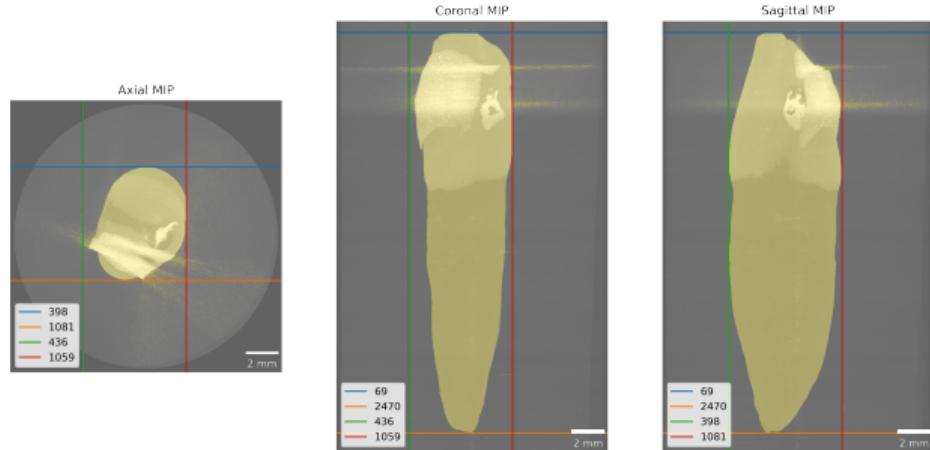
- Full datasets: 326 GB
- Cropped datasets: 115 GB



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Dataset cropping

- Full datasets: 326 GB
- Cropped datasets: 115 GB



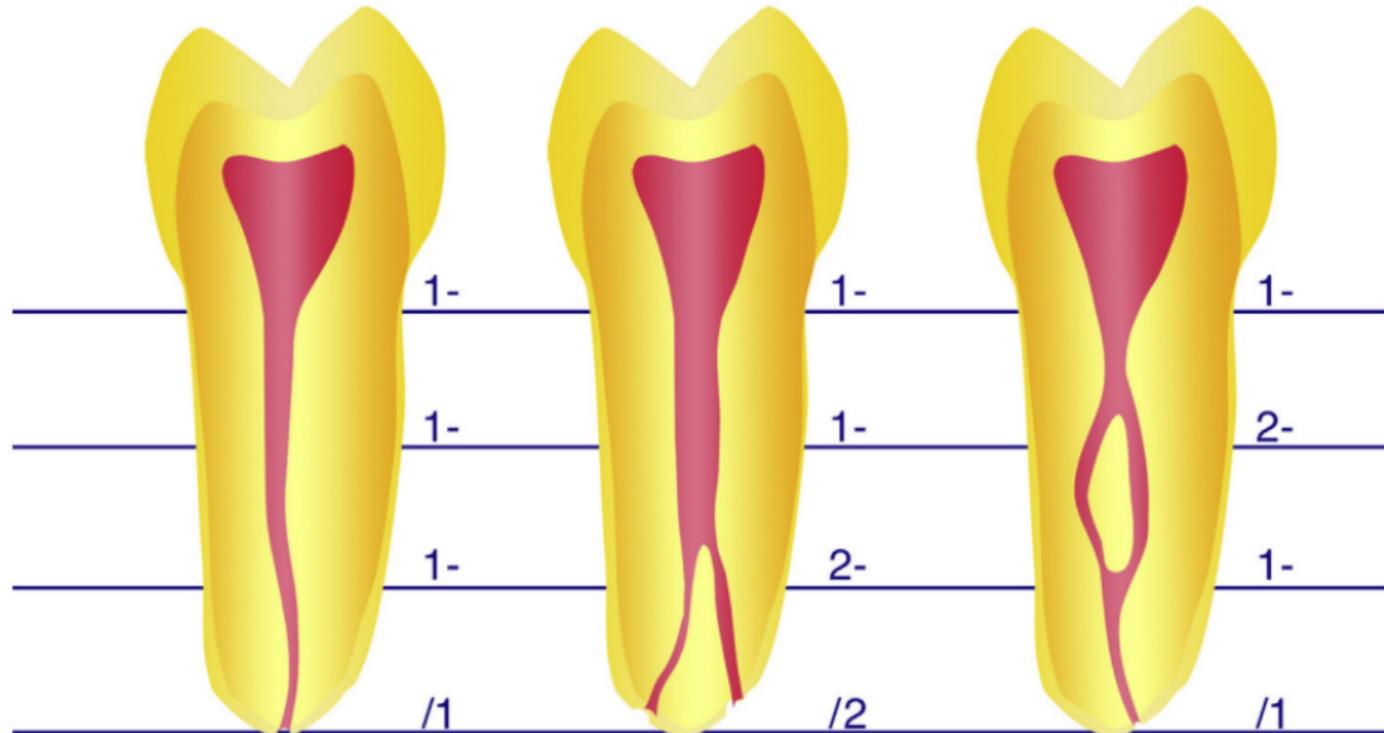
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Tooth morphology



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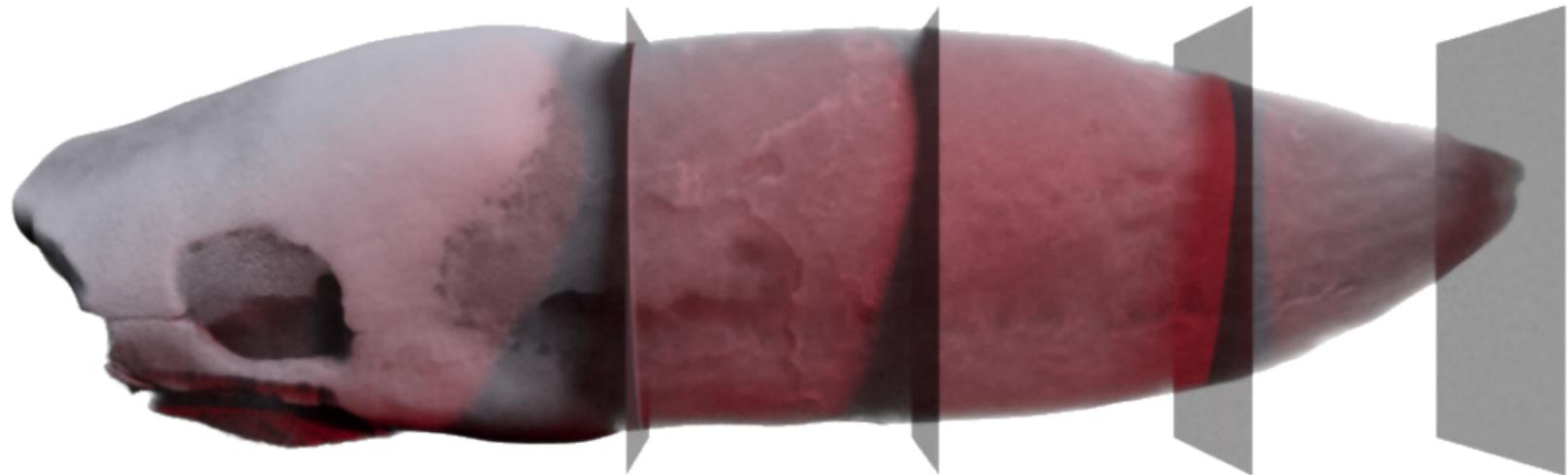
Tooth morphology



From [Briseno-Marroquin2015], Fig. 2

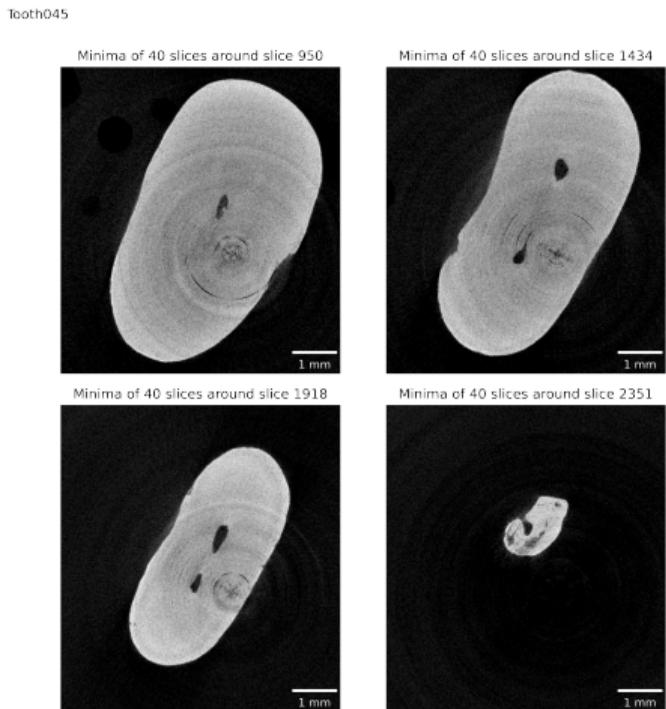
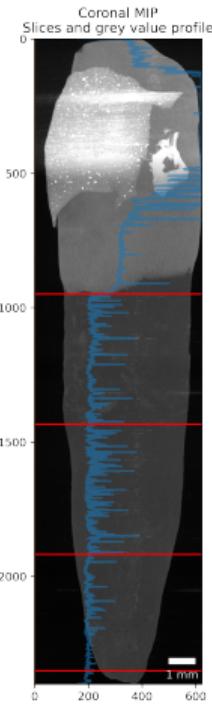
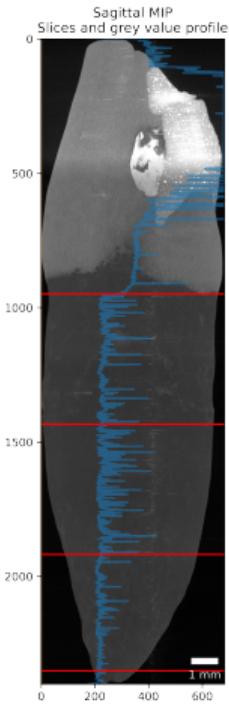
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Tooth morphology



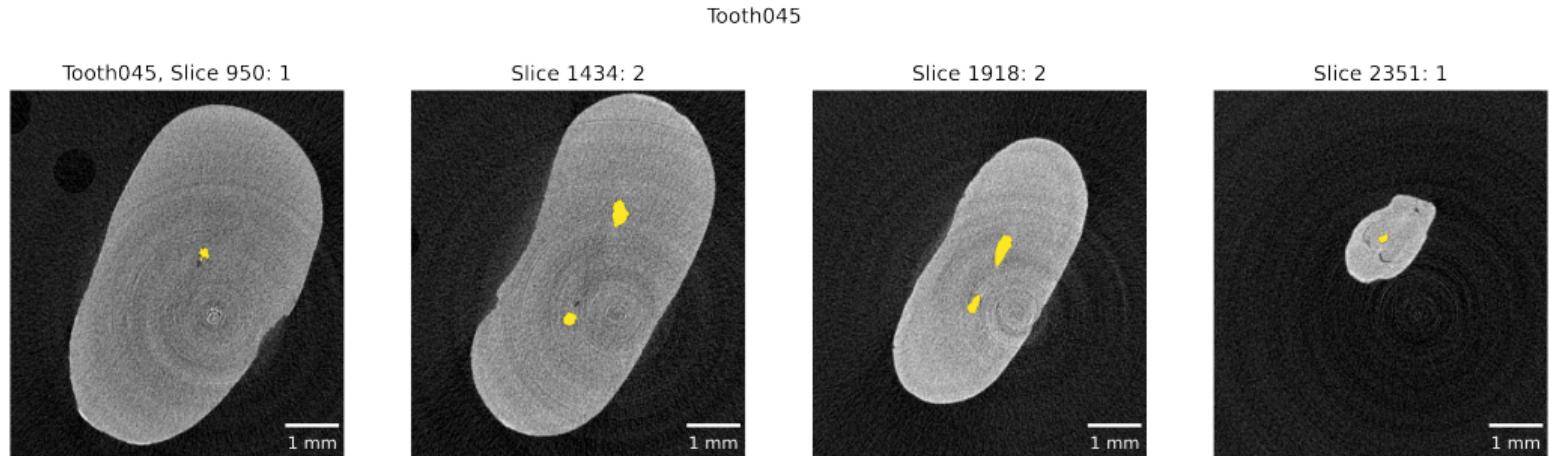
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Detection of enamel-dentin border



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Detection of enamel-dentin border



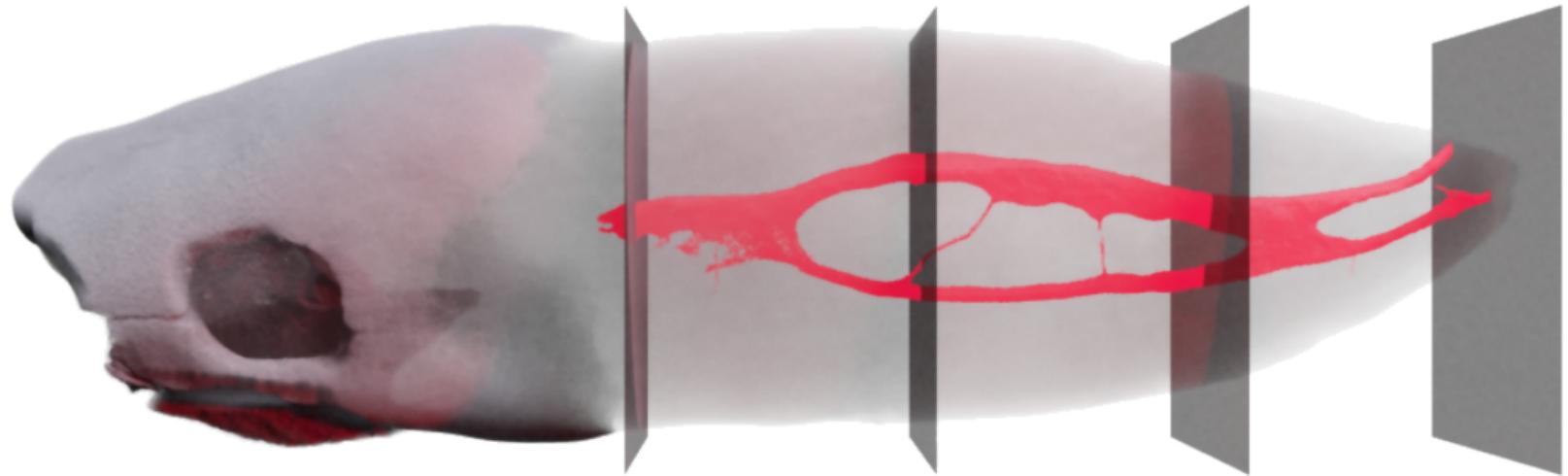
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Classification of root canal configurations

Roots	RCC	#	%	
Single (N=98)	1-1-1/1	73	74.5	
	1-1-1/2	14	14.3	
	1-1-1/3	1	1.0	
	1-1-1/4	2	2.1	
	1-1-2/1	1	1.0	
	1-2-1/1	4	4.1	
	1-2-1/2	1	1.0	
	1-2-2/2	1	1.0	
	2-3-1/1	1	1.0	
Double (N=3)	Buccal	1-1-1/1	2	66.6
		1-2-1/1	1	33.3
	Lingual	1-1-1/1	2	66.6
		1-1-1/2	1	33.3

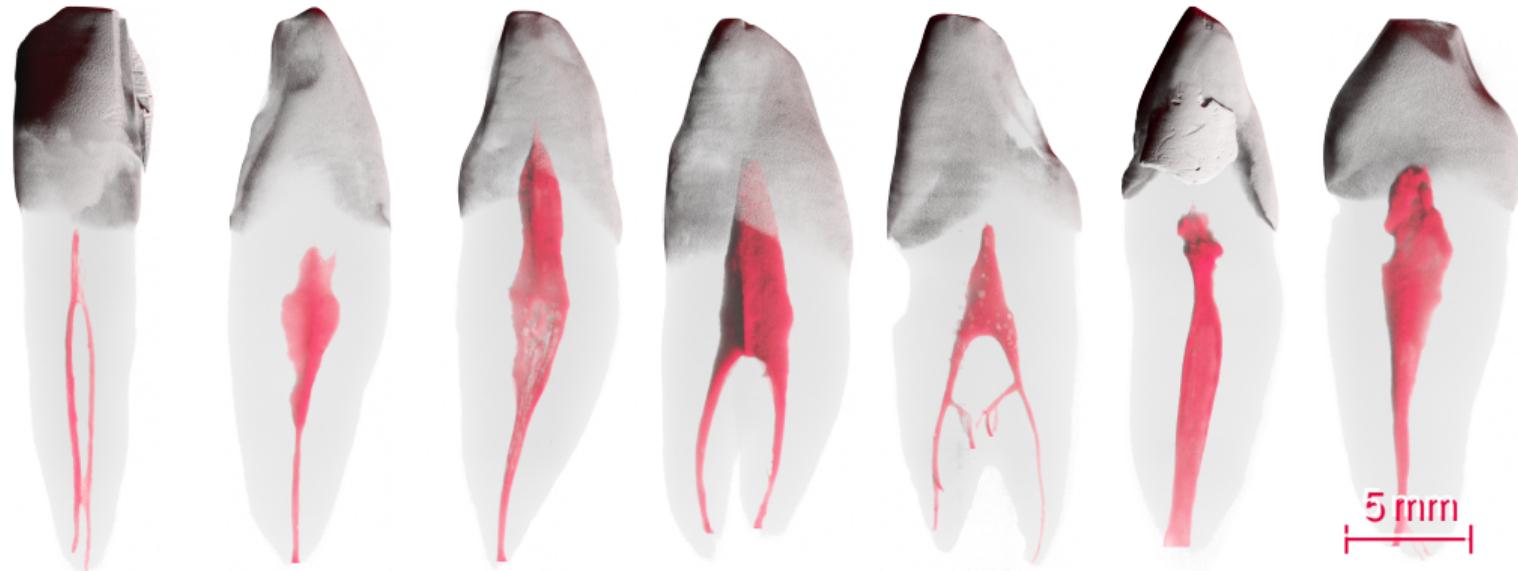
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Extraction of root canal space



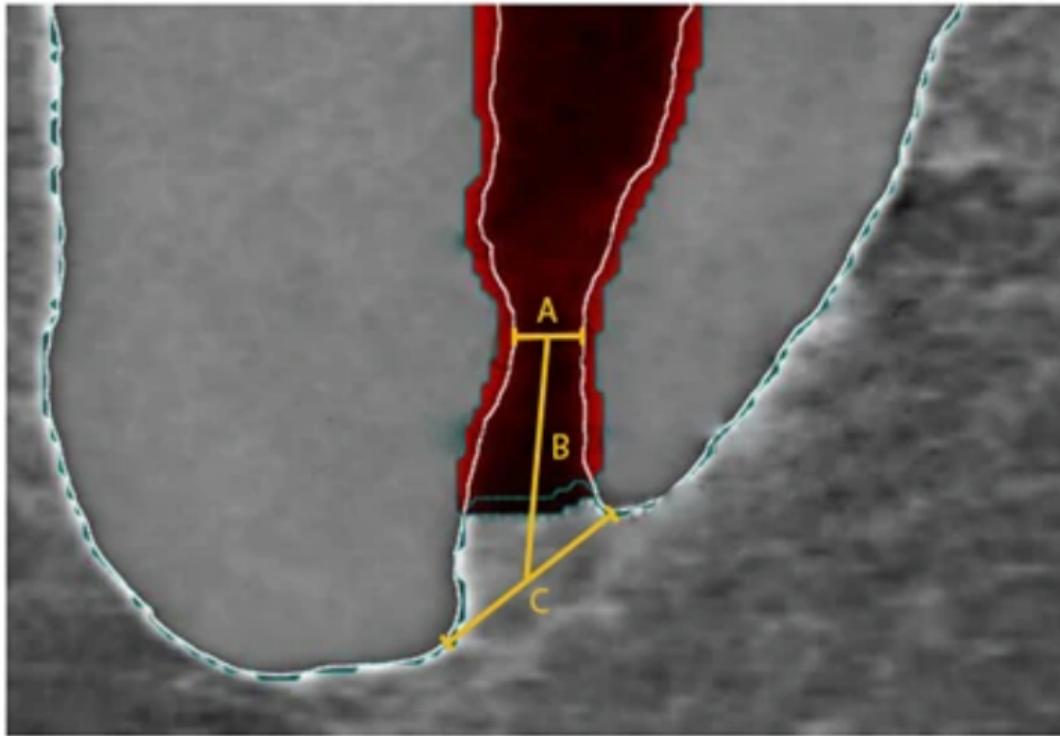
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Results of root canal space extraction



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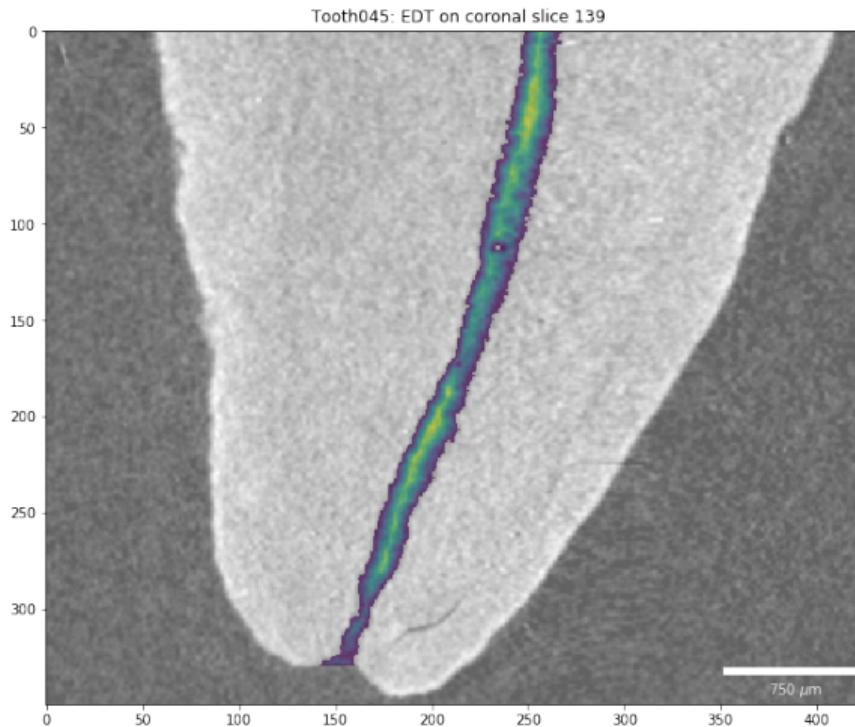
Physiological foramen geometry



From [Wolf2017], Fig. 1

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Physiological foramen geometry

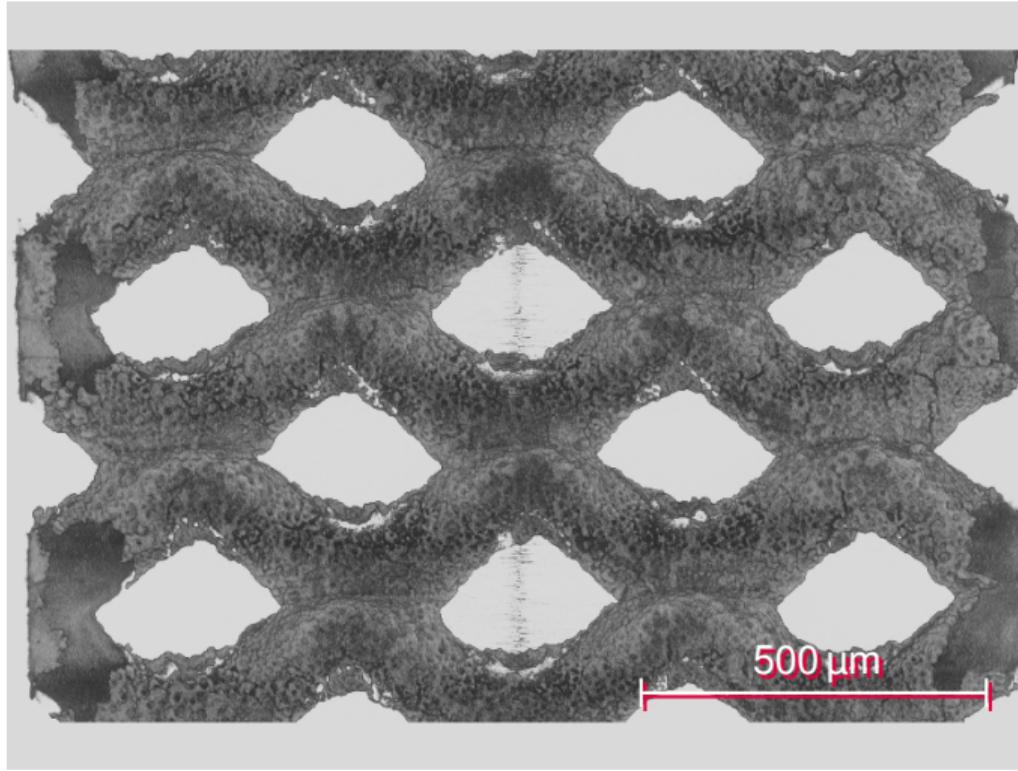


Conclusion ZMK

- Efficient use of time, e. g. more teeth does not mean more (human) work
- Reproducible analysis with *free and open-source* software, usable by *anyone*
- Objective analysis, e. g. no operator bias

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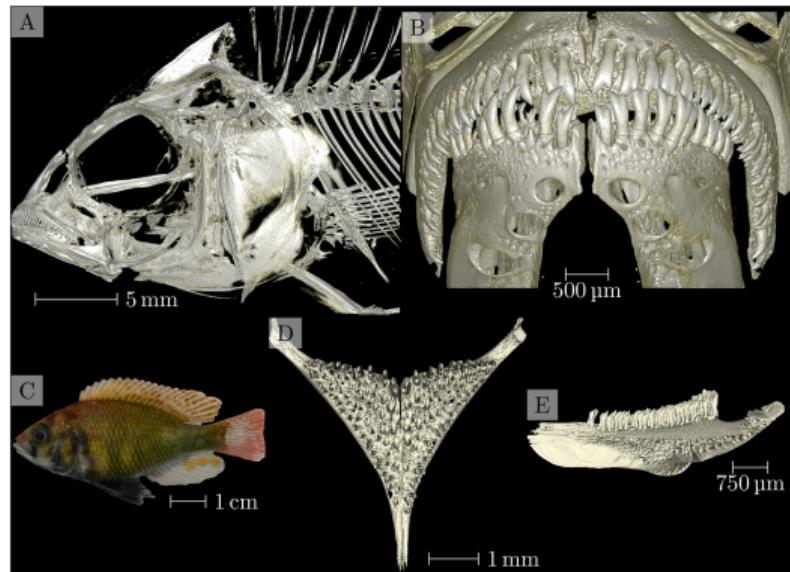
Metal foam



Data wrangling by example: Cichlids

Collaboration with the Institute of Ecology and Evolution [11]

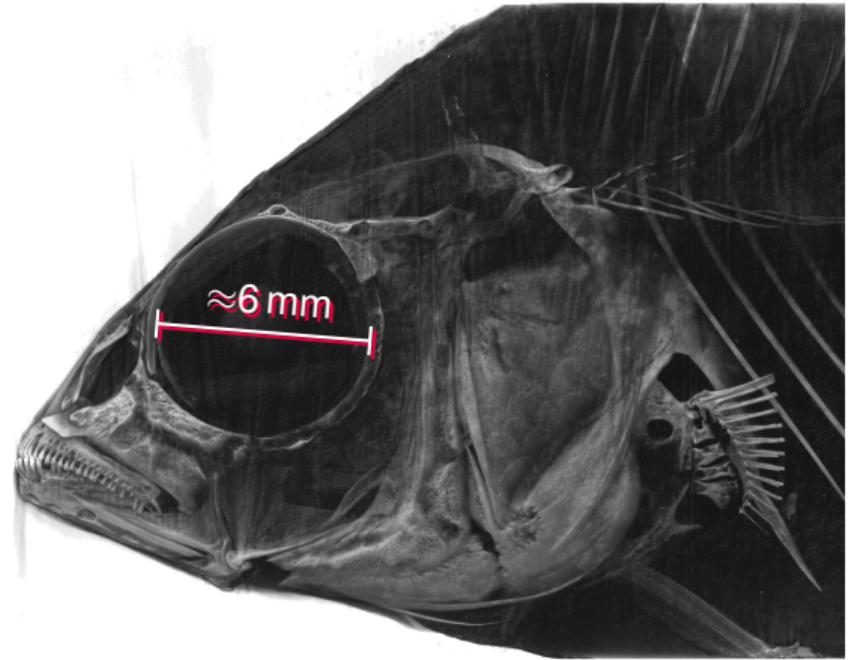
- 133 Cichlids from Lake Victoria, East Africa
 - Functional anatomy of the skulls and jaws
 - 6–18 cm in size
- 375 scans in total
 - Voxelsizes from 3.5–50 μm
 - 46 days of scanning time
 - 9.8 TB of raw data
 - 1.5 TB/+1 000 000 reconstructions



DOI:gsst8t, Fig. 1

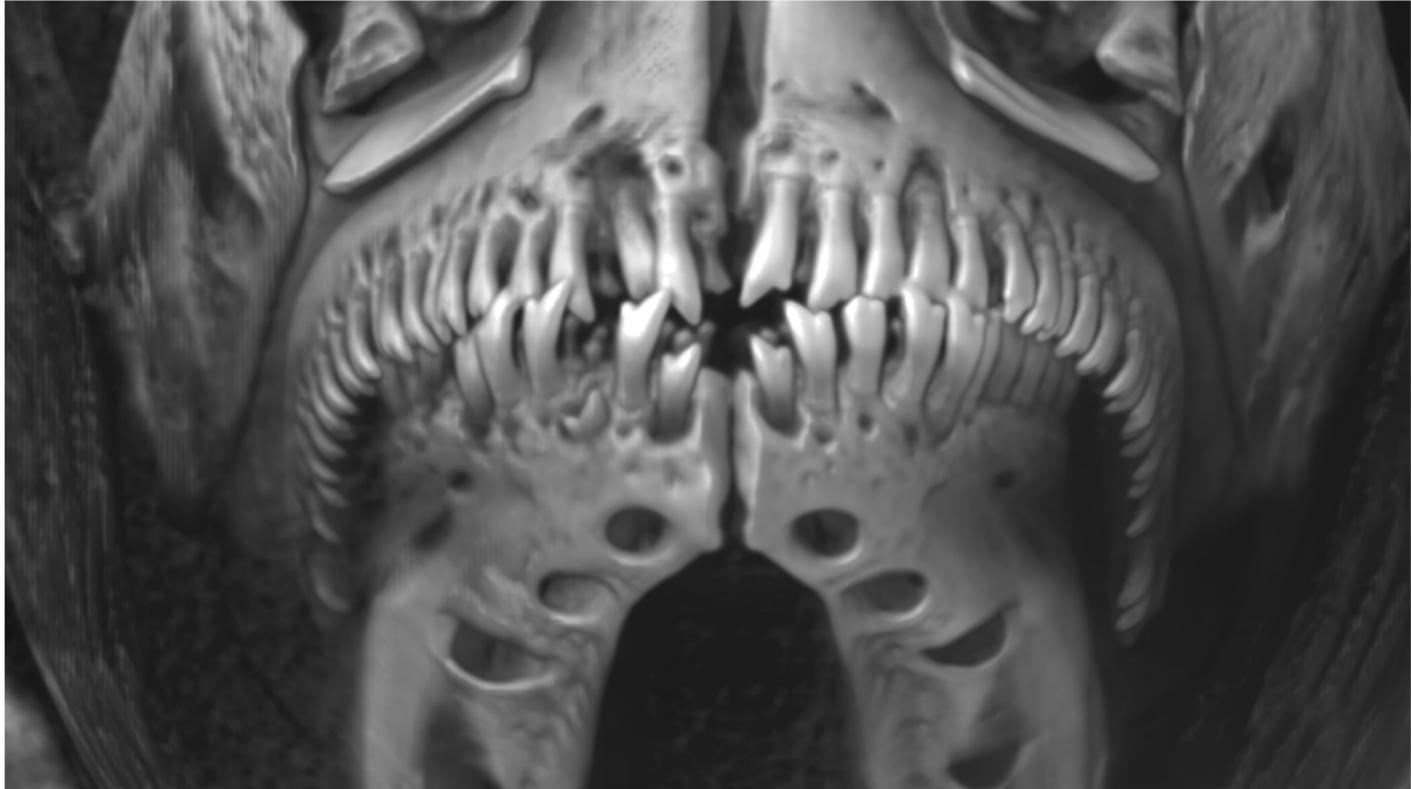
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Visualization of cichlid head



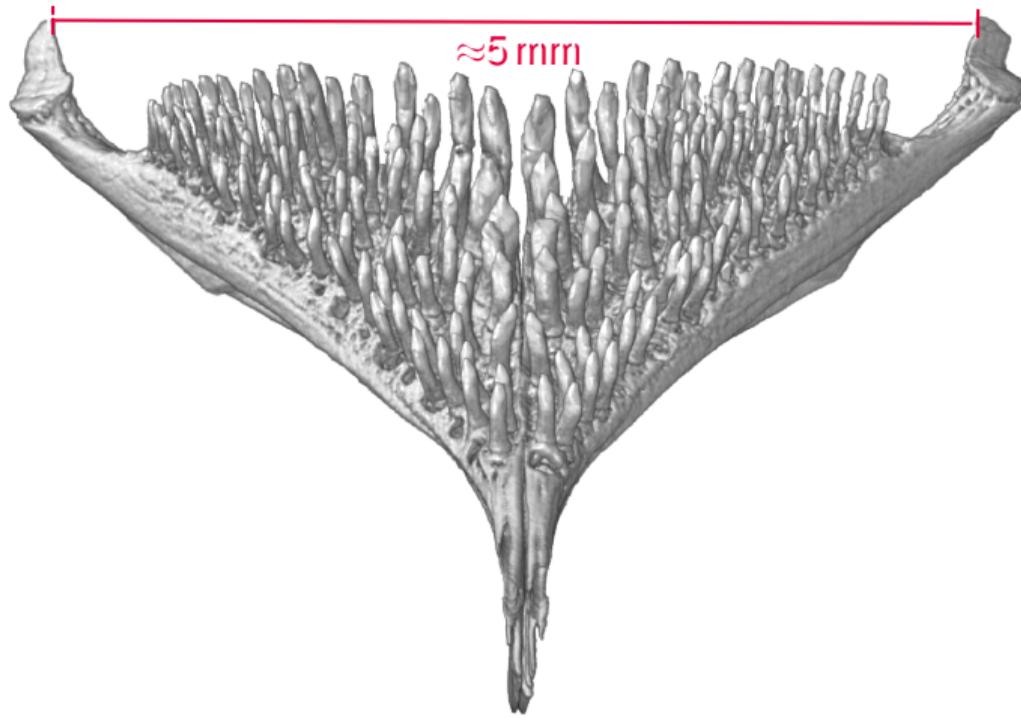
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Visualization of cichlid head



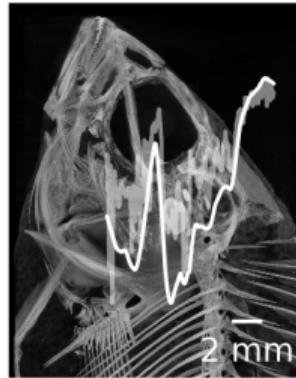
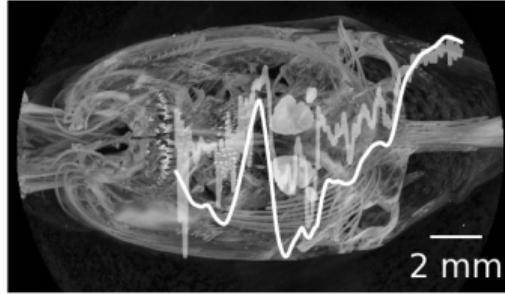
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Visualization of segmented pharyngeal jaw



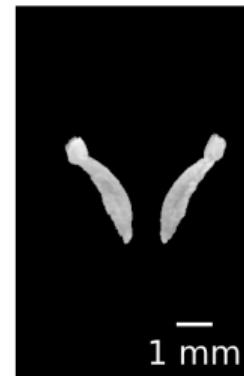
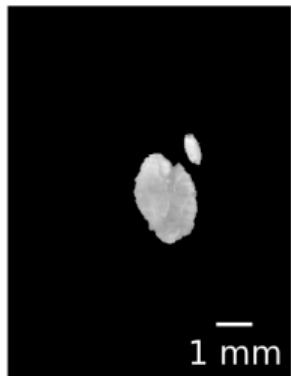
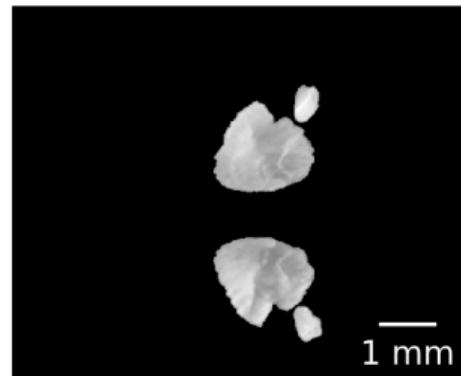
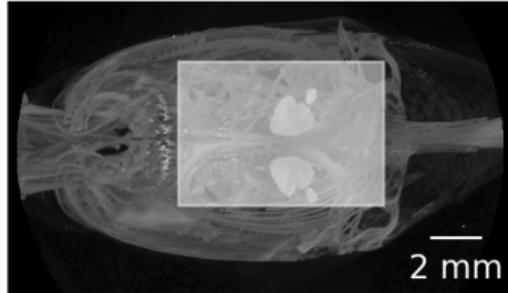
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Data wrangling by example: Cichlids



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Data wrangling by example: Cichlids



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Thanks!

- Thanks for listening to me!
- What questions do you have for me?

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Colophon

- This BEAMER presentation was crafted in \LaTeX with the (slightly adapted) template from *Corporate Design und Vorlagen* of the University of Bern.
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