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**UNIVERSITY  
OF BERN**

# *u<sup>b</sup>* X-ray microtomography

## 485018-HS2025-0: Advanced Course II Ultraprecision Engineering

**David Haberthür**

Institute of Anatomy, September 16, 2025

# *u<sup>b</sup>* 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

*u*<sup>b</sup>

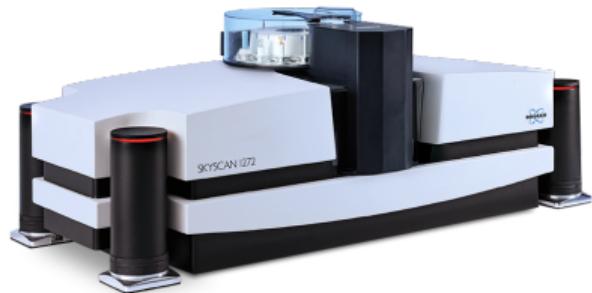
# Grüessech from the µCT group



[David.Haberthuer@unibe.ch](mailto:David.Haberthuer@unibe.ch) [Ruslan.Hlushchuk@unibe.ch](mailto:Ruslan.Hlushchuk@unibe.ch) [Oleksiy.Khoma@unibe.ch](mailto:Oleksiy.Khoma@unibe.ch)

# $\mu$ 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](http://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

# $\mu$ CT<sup>b</sup>

- 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

*u<sup>b</sup>*

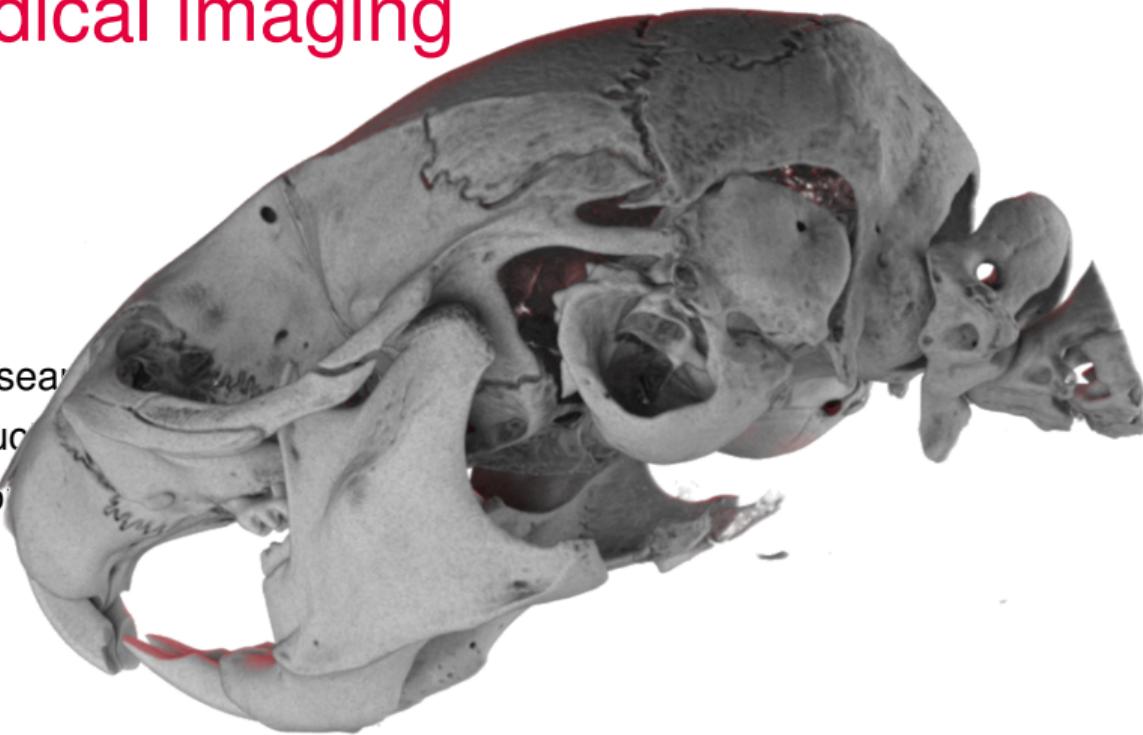
# Biomedical imaging

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



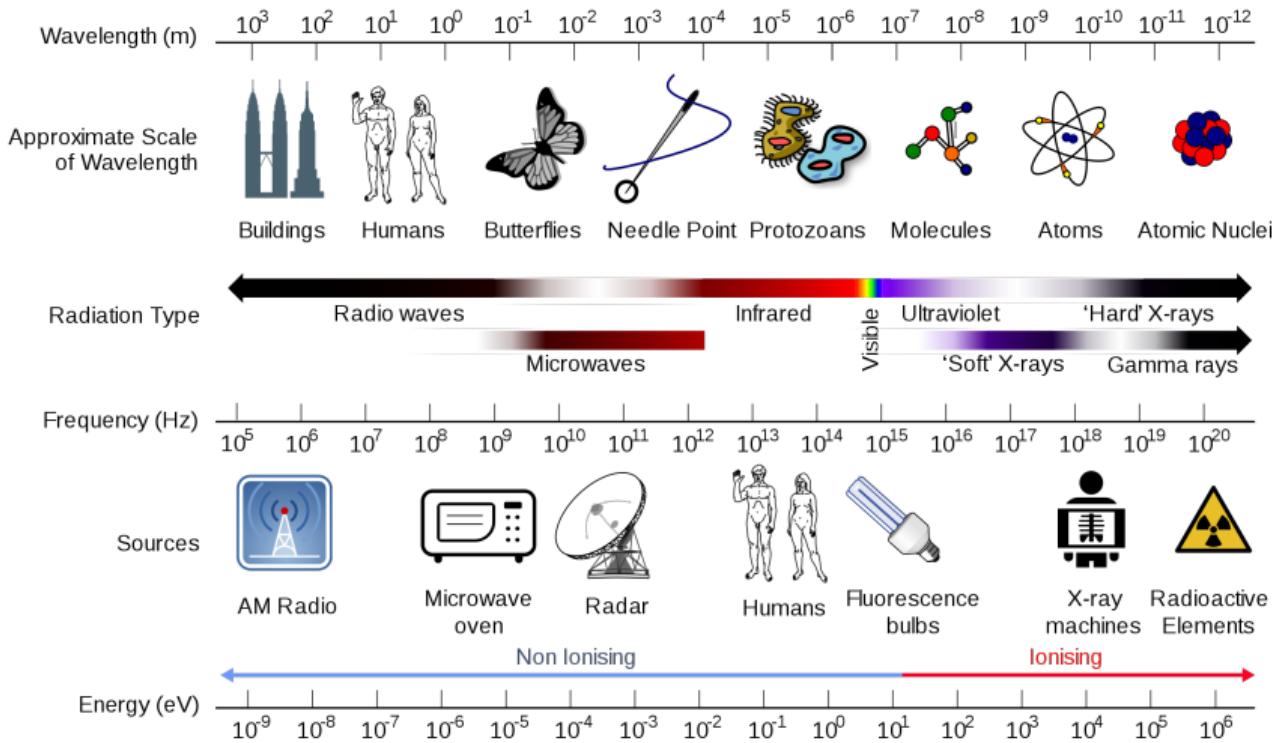
w.wiki/7g4 CC BY-NC-SA

# Biomedical imaging

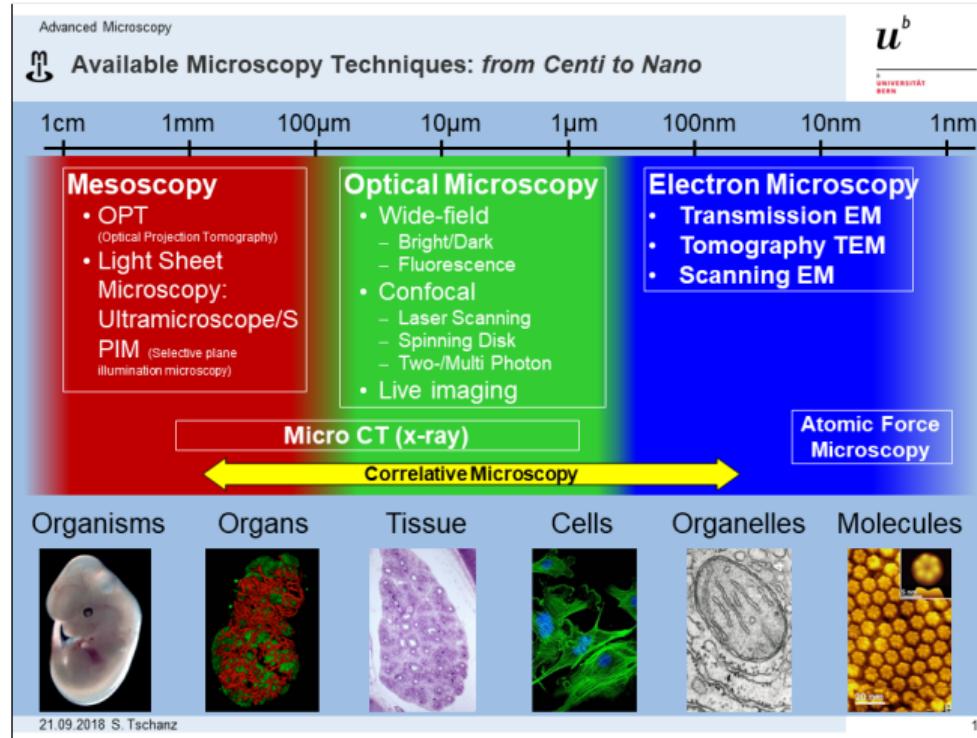


- Medical research
- Non-destructive
- (Small) Biology

# $u^b$ Wavelengths & Scales



# $u^b$ Wavelengths & Scales



Stefan Tschanz, with permission

# 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](https://youtu.be/2CWpZKuy-NE)

*u<sup>b</sup>*

# CT History

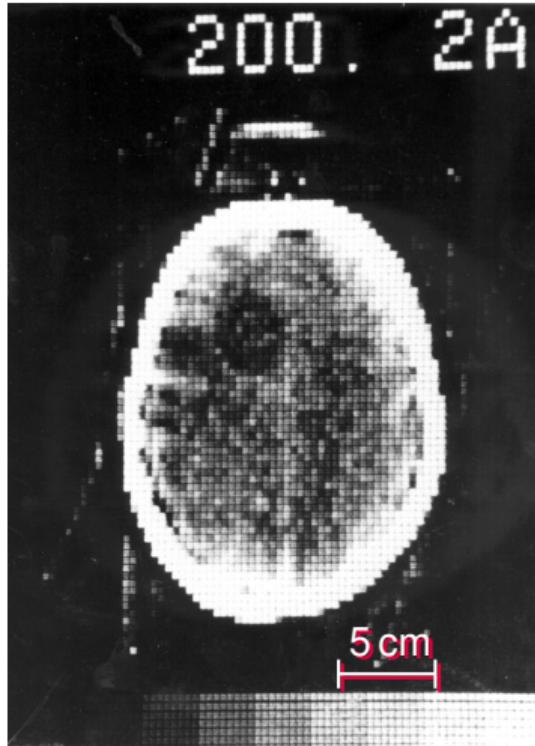
- 1895: Wilhelm Conrad Röntgen discovers X-rays



w.wiki/BHAN ©

# CT History

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



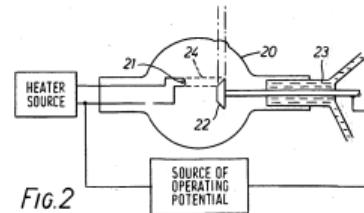
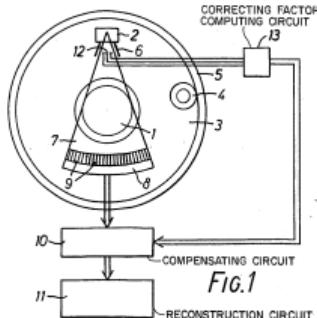
From [14], Figure 5

*u<sup>b</sup>*

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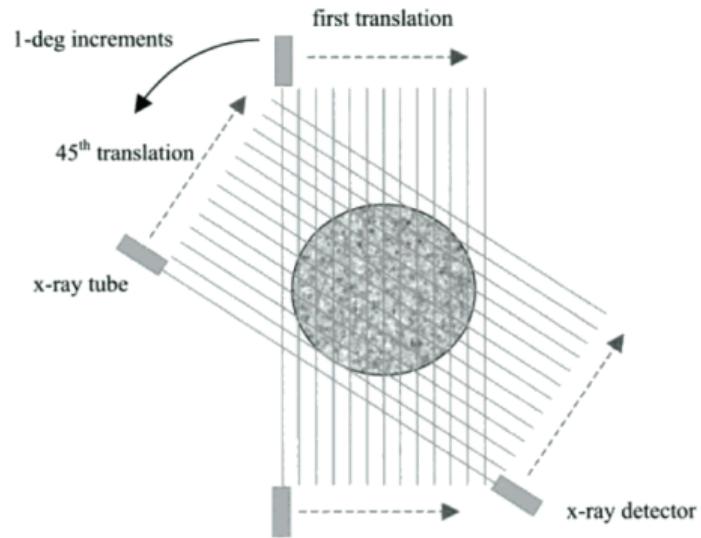
U.S. Patent Feb. 24, 1976 Sheet 1 of 2 3,940,625



From [US3940625A], p. 2

# CT History

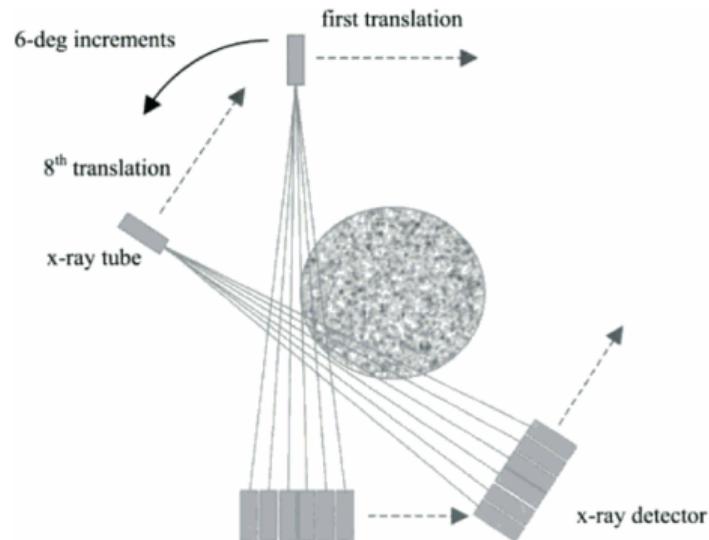
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- CT scanner generations
  - First generation



From [15], Figure 1.12

# CT History

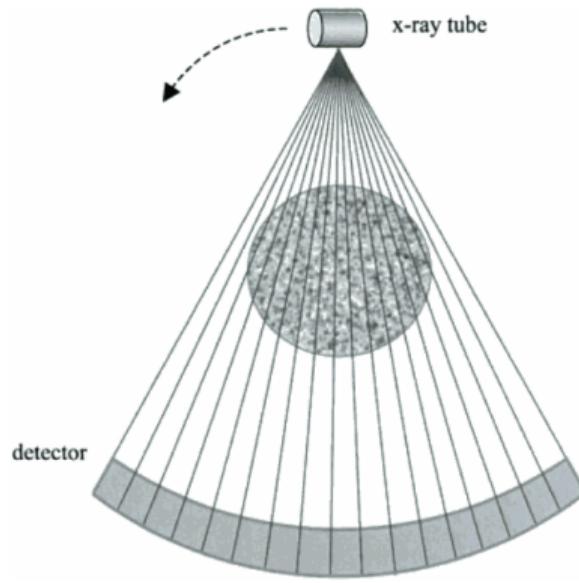
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  - First generation
  - Second generation



From [15], Figure 1.13

# CT History

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



From [15], Figure 1.14

# $\mu$ CT History I

- X-ray computed tomography began to replace analog focal plane tomography in the early 1970s [**Lin2019**]
- Non-medical use in the late 1970s, for detection of internal defects in fabricated parts and equipment
- Lee Feldkamp [16] 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 [**Feldkamp1983**]

# $\mu$ CT History II

- $\mu$ CT was first reported in the 1980s, for scanning gemstones
- Early 1990s: Manufacturers like SkyScan and Scanco Medical made  $\mu$ 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  $\mu$ 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.” ([19])
  - Photoelectric absorption ( $\tau$ ) 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 [20, i. e. Beer-Lamberts law]:  $I(t) = I_0 e^{-\alpha z}$

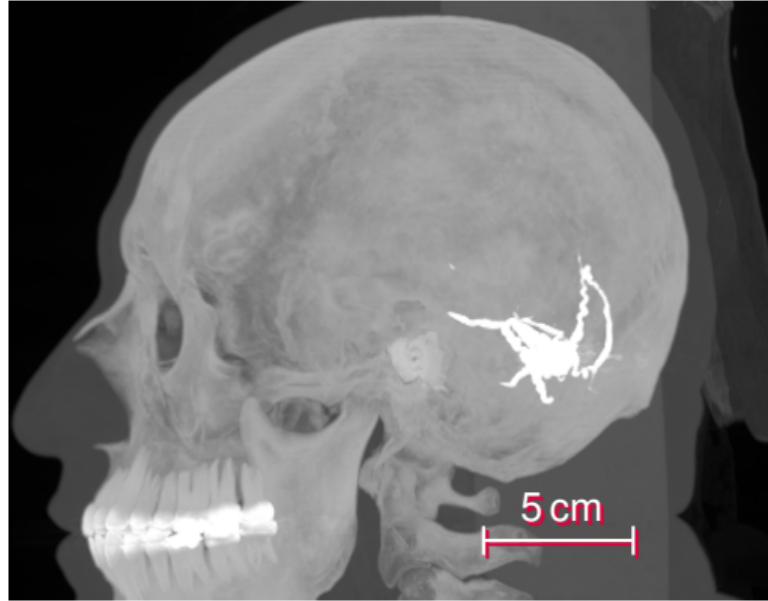
# Composition of biological tissues

Tissue: content by mass percentage

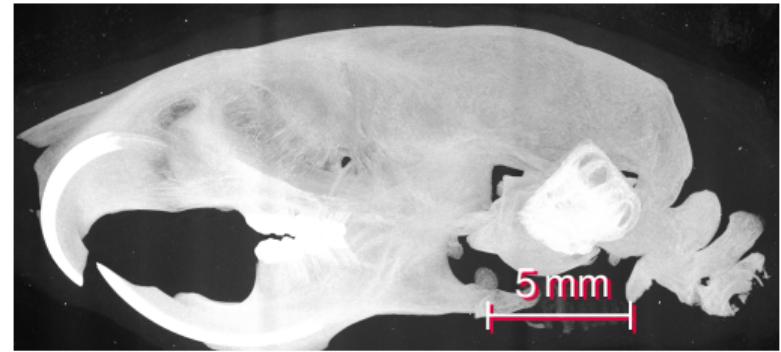
Element Atomic number	H 1	C 6	N 7	O 8	Na 11	P 15	S 16	Cl 17	K 19	Ca 20
Fat	11.4	59.8	0.7	27.8	0.1		0.1	0.1		
Water	11.2			88.8						
Blood	10.2	11	3.3	74.5	0.1	0.1	0.2	0.3	0.2	
Liver	10.2	13.9	3	71.6	0.3	0.2	0.3	0.2	0.3	
Brain	10.7	14.5	2.2	71.2	0.2	0.4	0.2	0.3	0.3	
Bone	3.4	15.5	4.2	43.5	0.1	10.3	0.3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

*u*<sup>b</sup>

# Why $\mu$ CT?

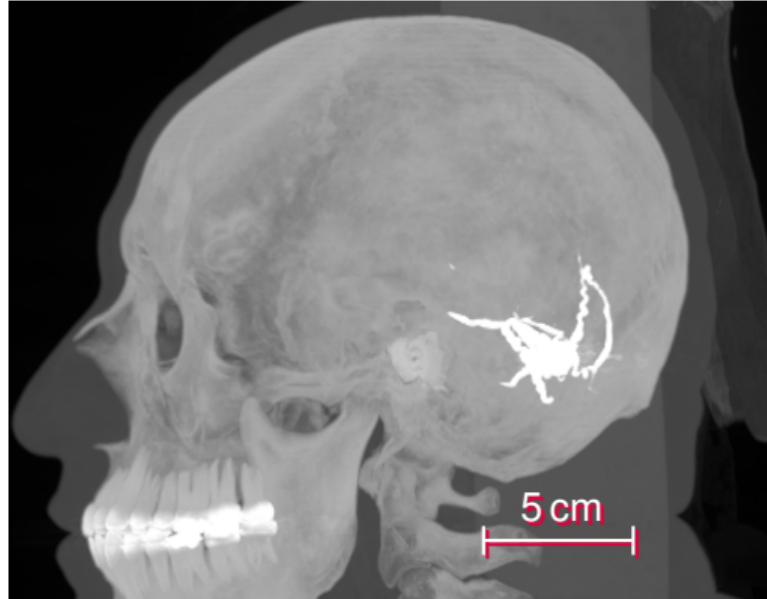


From [21], Subject C3L-02465



$u^b$

# Why $\mu$ CT?

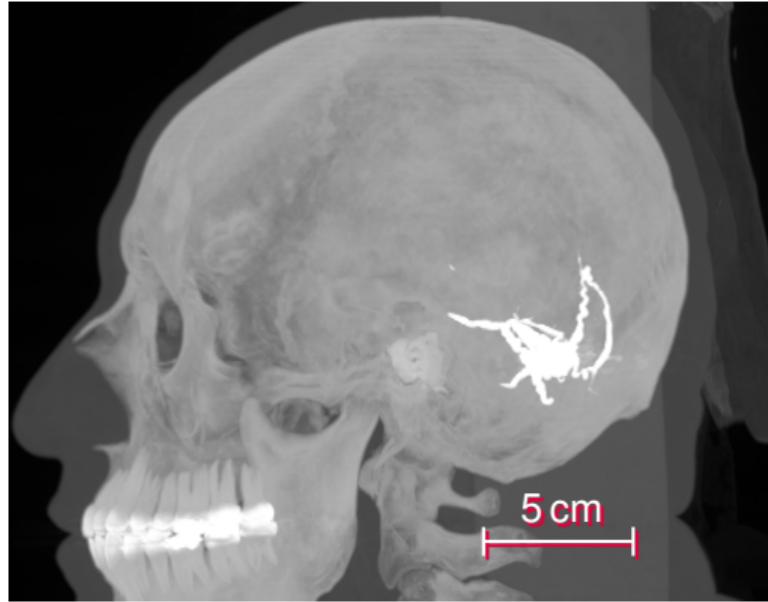


From [21], Subject C3L-02465

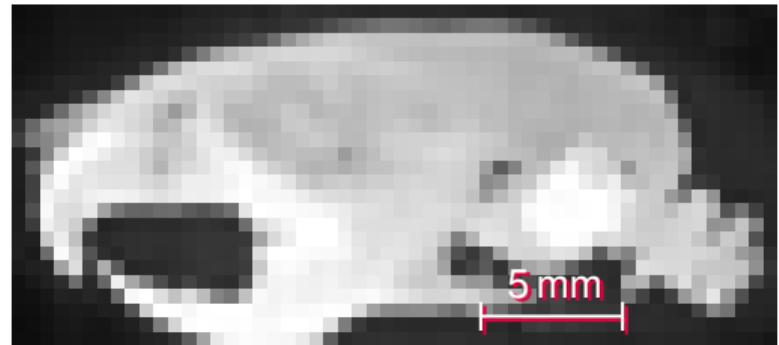


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# Why $\mu$ CT?

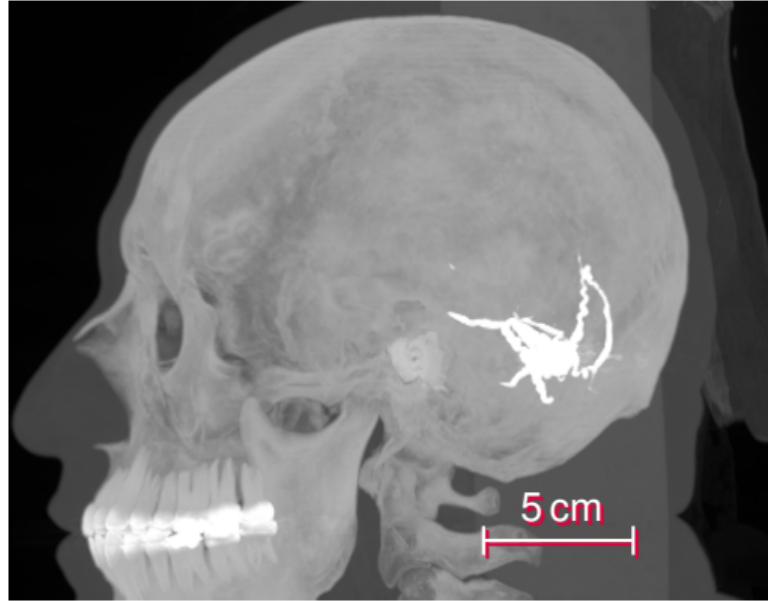


From [21], Subject C3L-02465

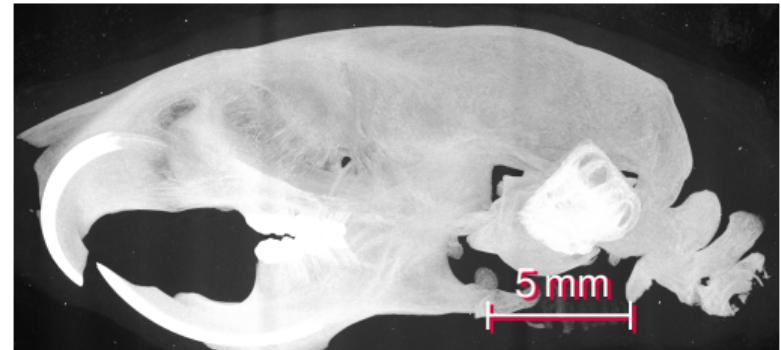


*u*<sup>b</sup>

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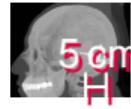


From [21], Subject C3L-02465

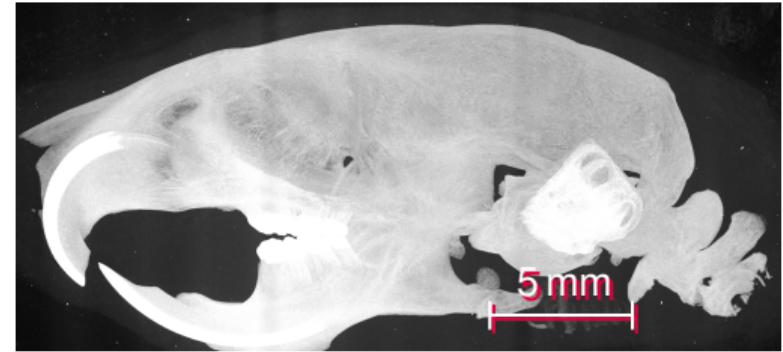


$u^b$

# Why $\mu$ CT?



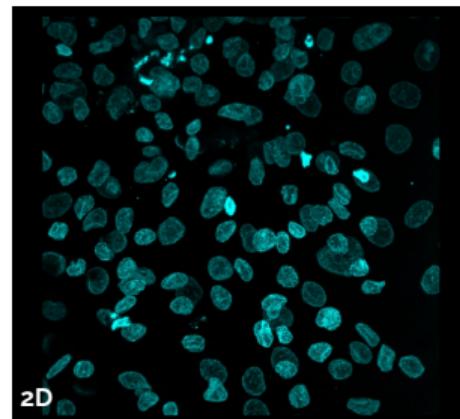
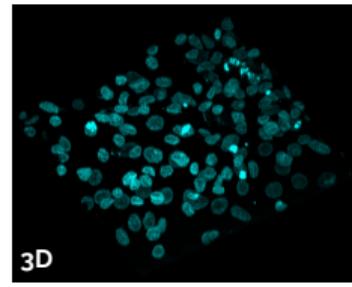
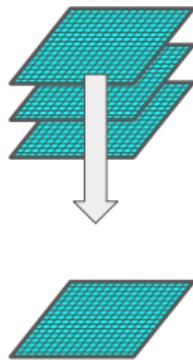
From [21], Subject C3L-02465



# 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  $\mu\text{m}$  (*in vivo*)
  - Voxel size around 0.5  $\mu\text{m}$  (*ex vivo*)
- Synchrotron CT
  - Voxel size down to 160 nm



flic.kr/p/D4rbom

# Machinery

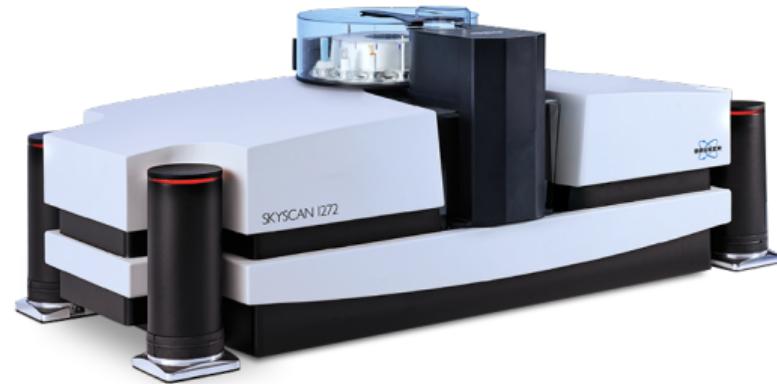
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[flic.kr/p/fpTrGu](https://flic.kr/p/fpTrGu)

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[bruker.com/skyscan1272](http://bruker.com/skyscan1272)

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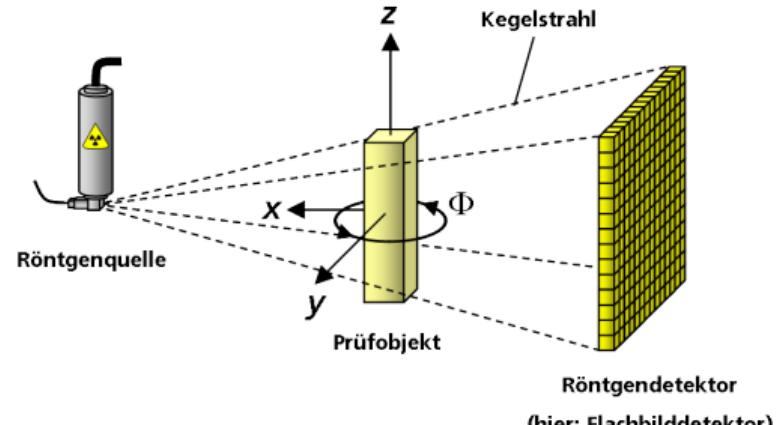
flic.kr/p/7Xhk2Y

$u^b$

# What is happening?

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

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



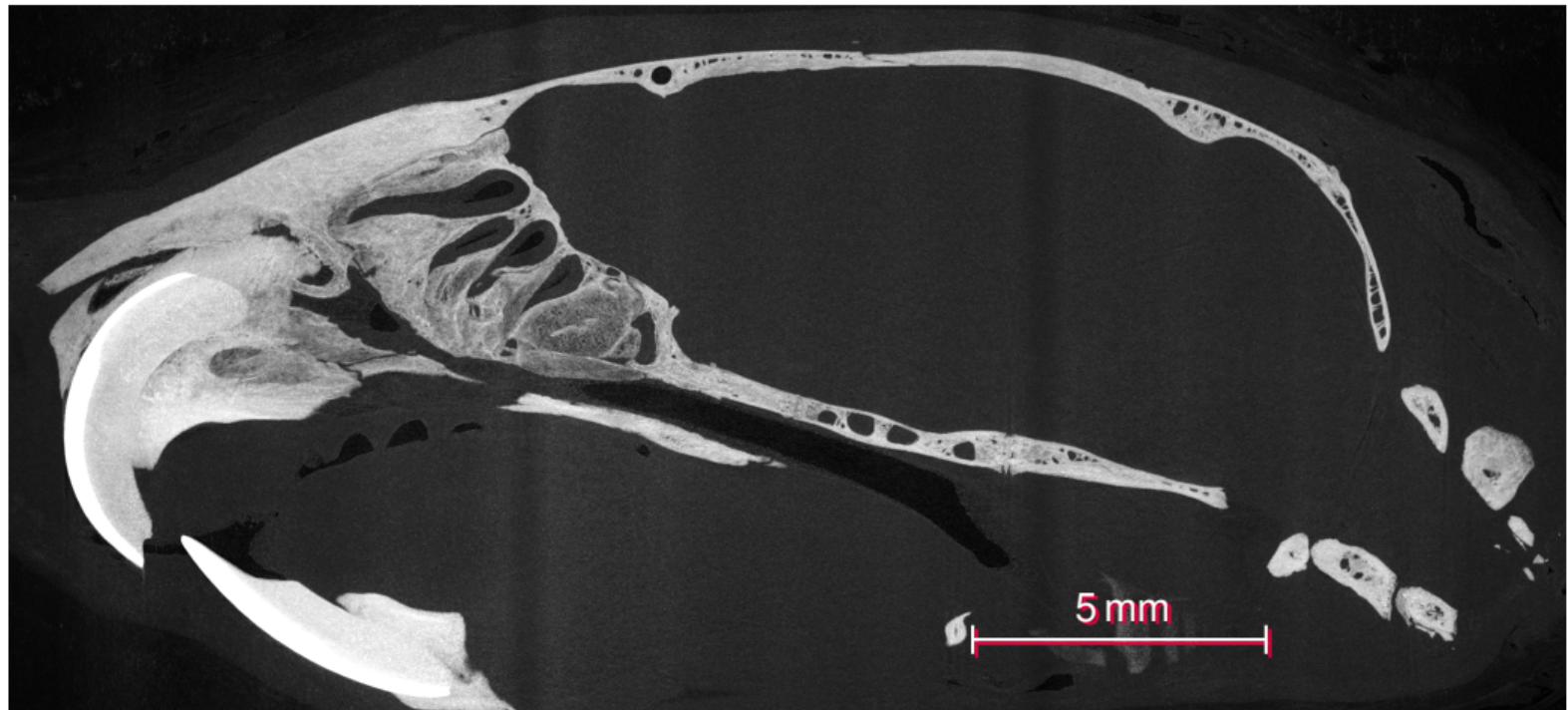
w.wiki/7g3 @①②

*u<sup>b</sup>*

# Machinery

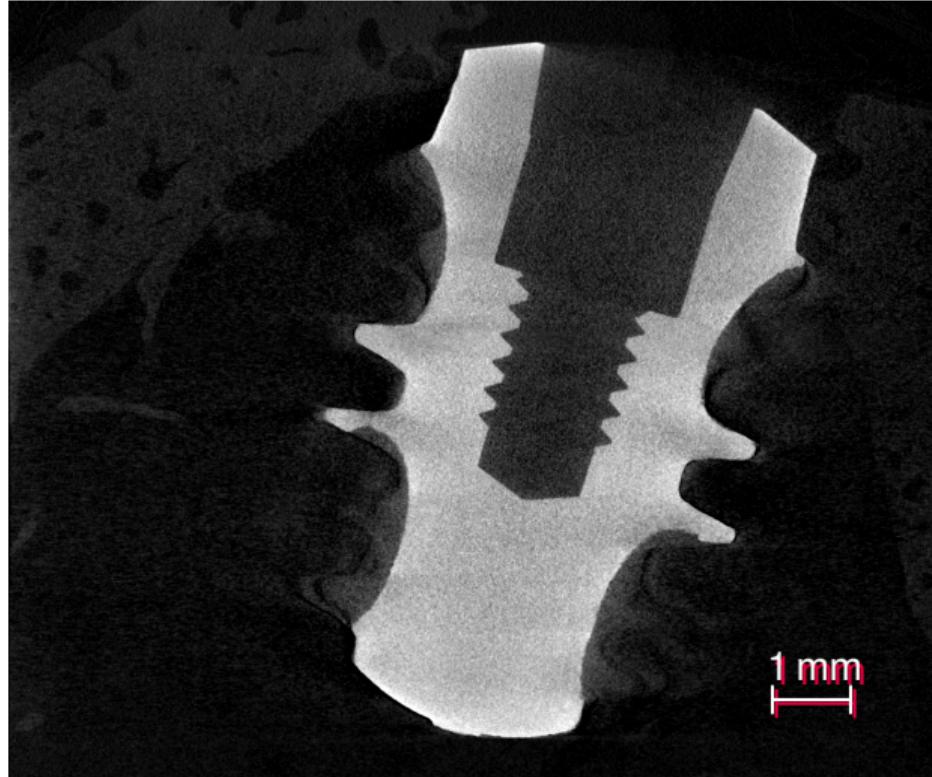
*u*<sup>b</sup>

# Examples



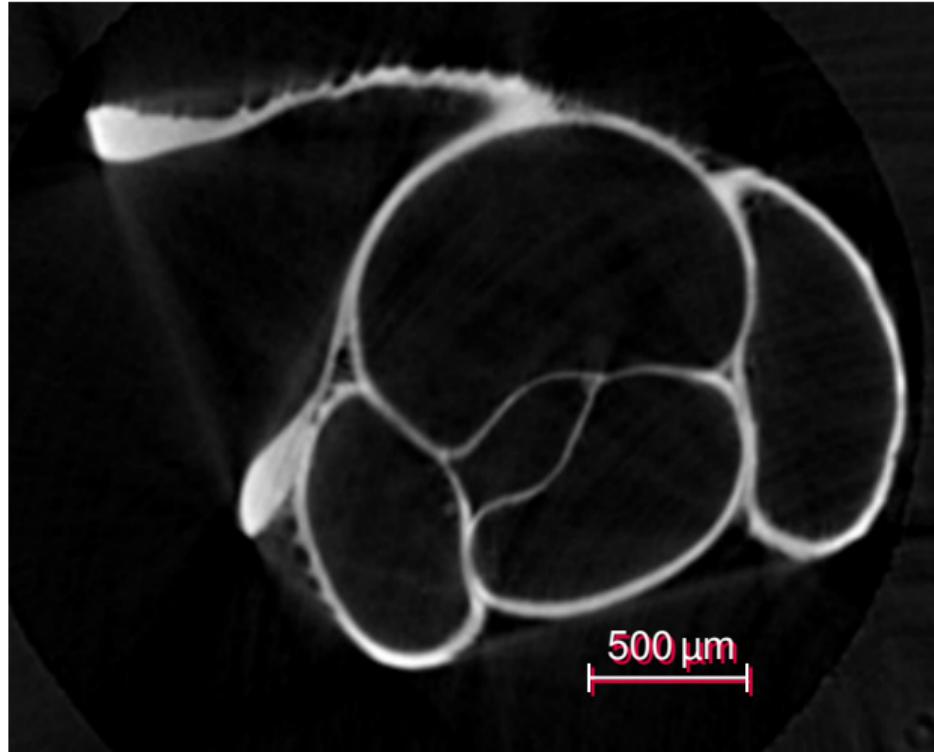
$u^b$

# Examples



$u^b$

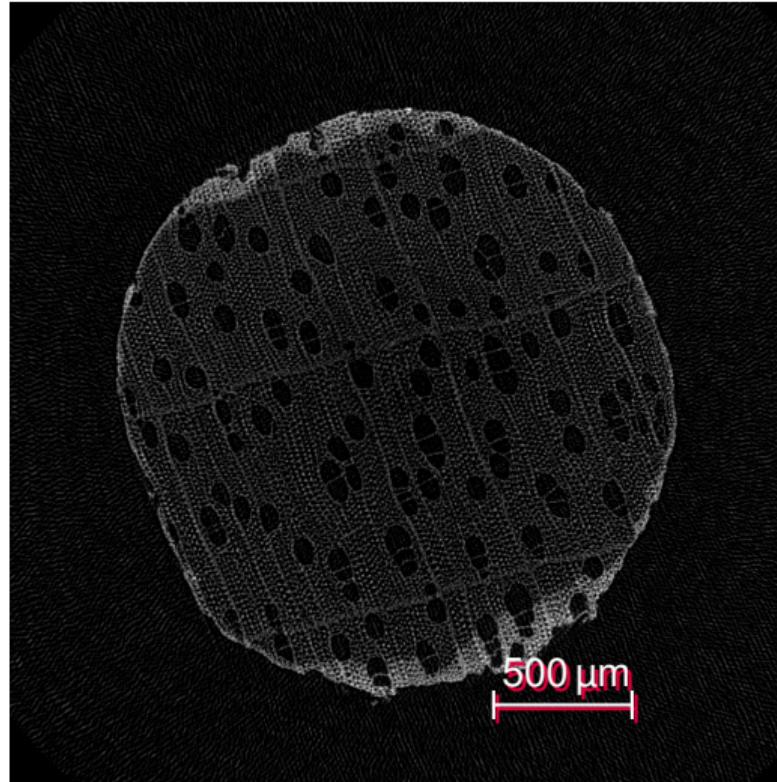
# Examples



From [8], *Diancta phoenix*

$u^b$

# Examples



*u*<sup>b</sup>

# Examples



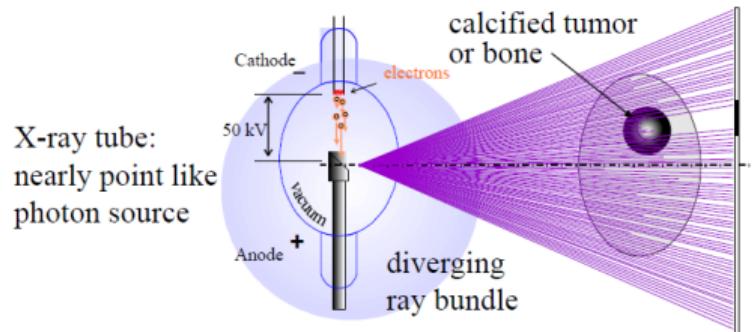
*u<sup>b</sup>*

# Preparation

- Study design
- Sample preparation

# Projections

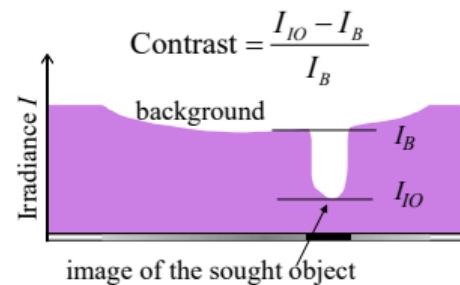
## X-ray generation and contrast



X-ray tube:  
nearly point like  
photon source

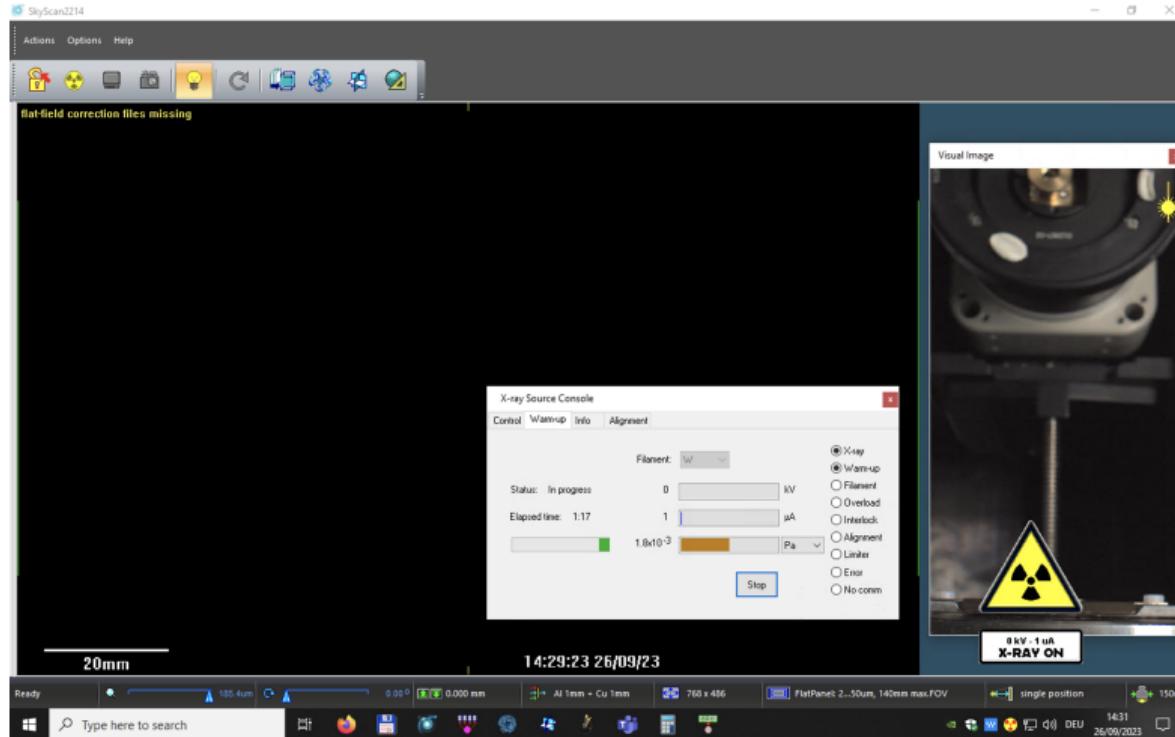
Contrast is given by  
absorption of intensity I

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



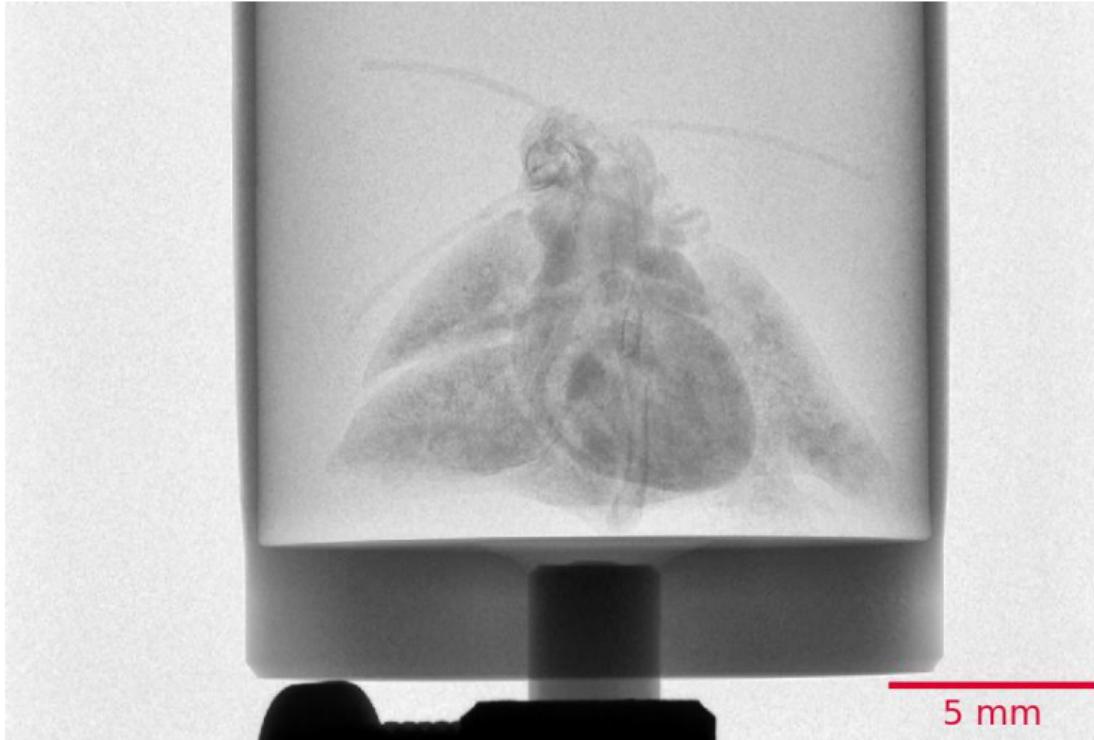
*Contrast, Magnification and Resolution—Laws of Physics for Microscopists (1, 2022) by Martin Frenz, Slide 21*

## $u^b$ Projection acquisition



*u*<sup>b</sup>

# Projections

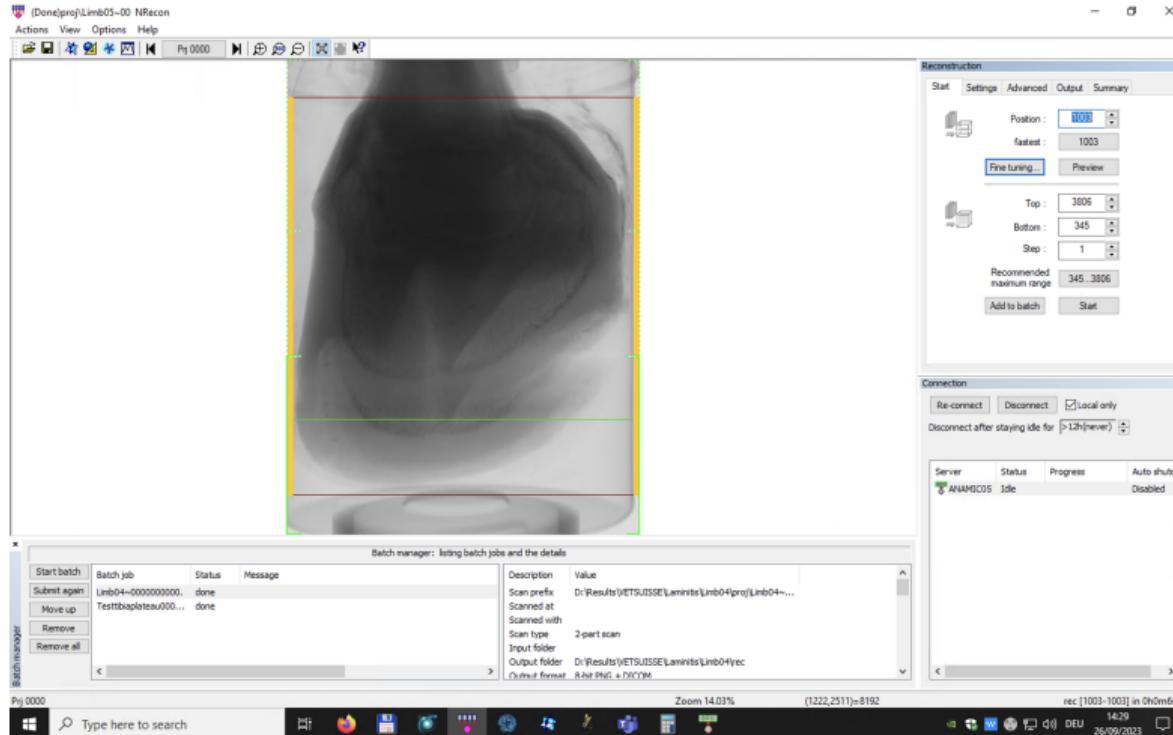


# Projections

- 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

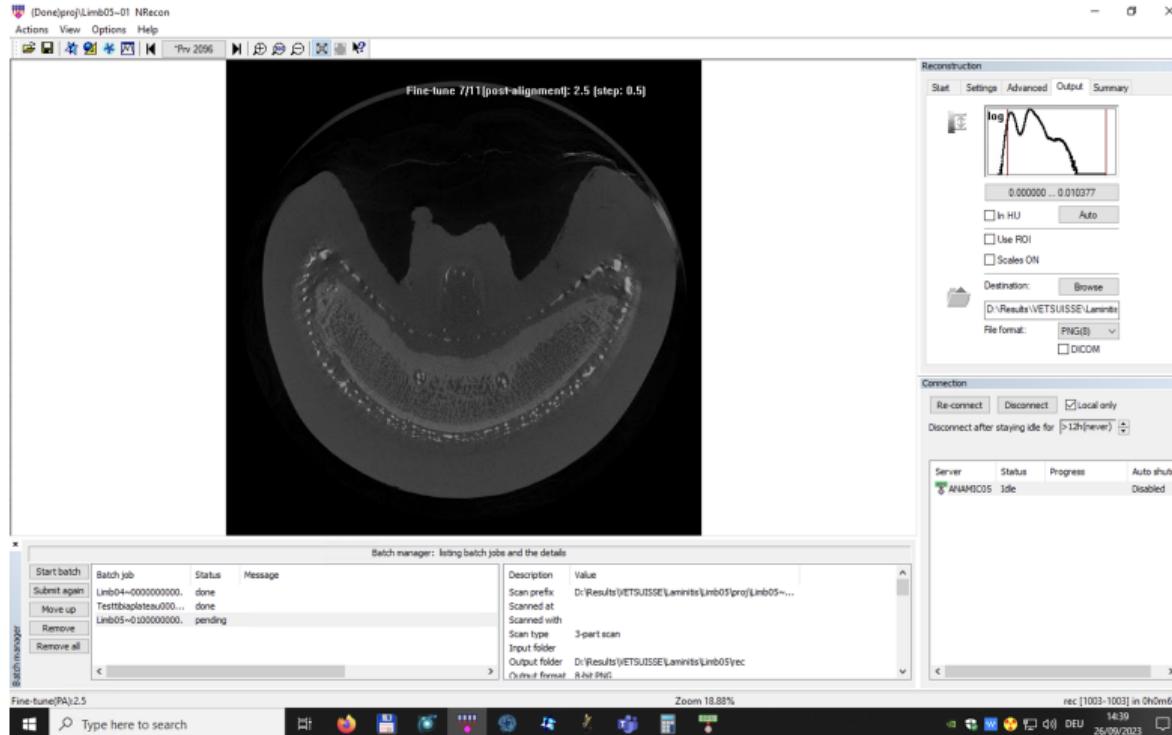
*u*<sup>b</sup>

# Reconstructions



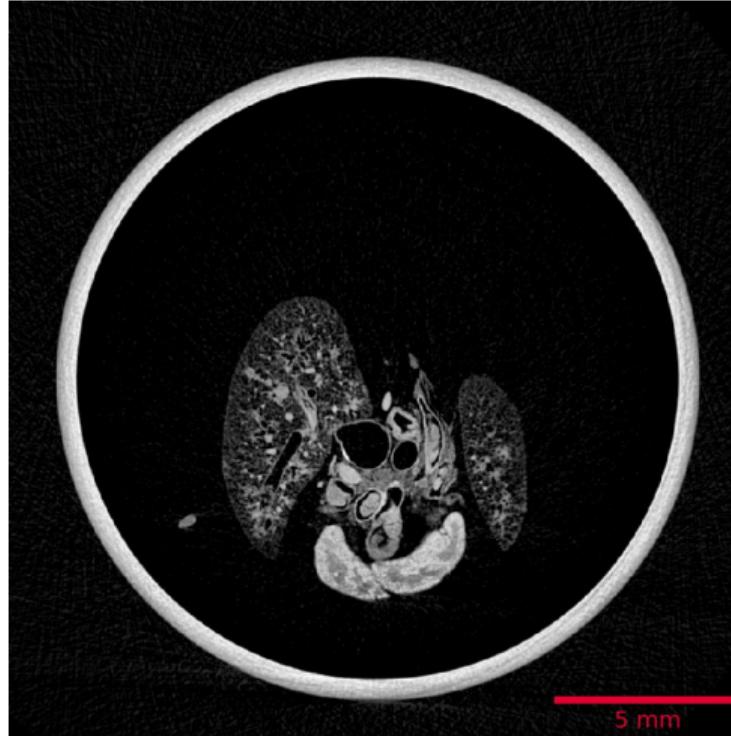
*u*<sup>b</sup>

# Reconstructions



*u*<sup>b</sup>

# 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 [16]
- Corrections (beam hardening, etc.)
- Writing to stack

$u^b$

# Visualization



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

*u*<sup>b</sup>

# What to use?

- ImageJ/Fiji [22]
- Also see *Fundamentals of Digital Image Processing* by Guillaume Witz
- Reproducible research
  -  in Jupyter [23]
  - **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

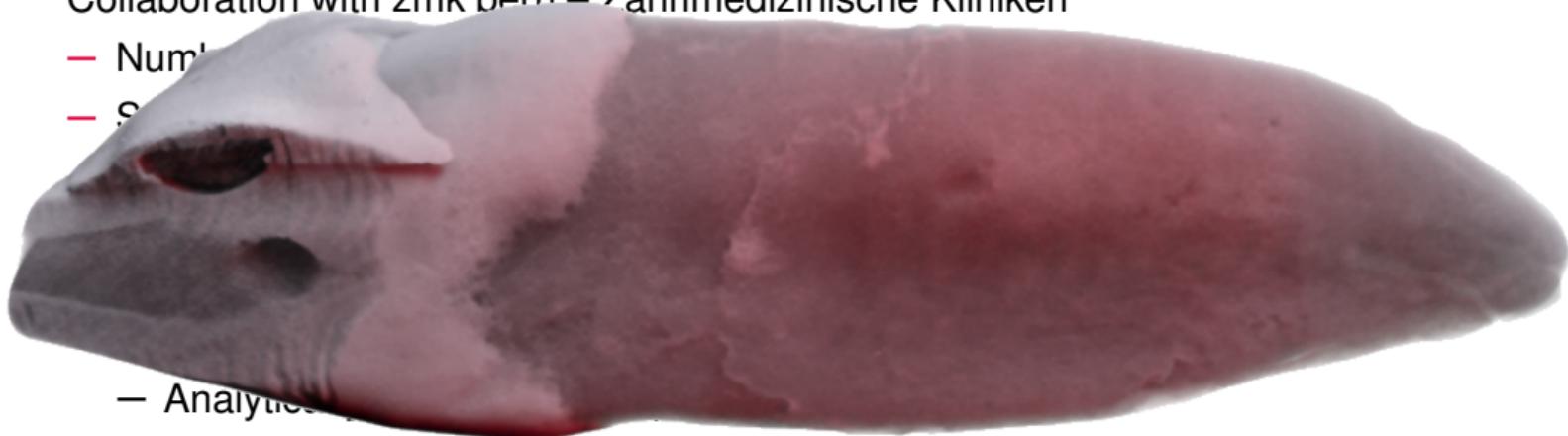
*u<sup>b</sup>*

# Internal morphology of human teeth

Collaboration with zmk bern – Zahnmedizinische Kliniken

- Numbering
- Sectioning

- Analytics

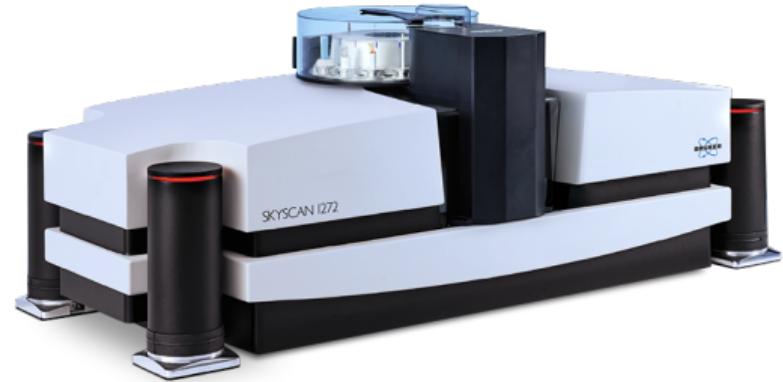


# How?

- 104 extracted human permanent mandibular canines
- $\mu$ CT imaging
- Root canal configuration, according to Briseño-Marroquín et al. [25]
- *Reproducible* analysis [26], e. g. you can click a button to double-check or recalculate the results yourself!



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[bruker.com/skyscan1272](http://bruker.com/skyscan1272)

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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=A1 1mm
Study Date and Time=02 Jul 2020
08h:23m:34s
Scan duration=0h:39m:51s
```

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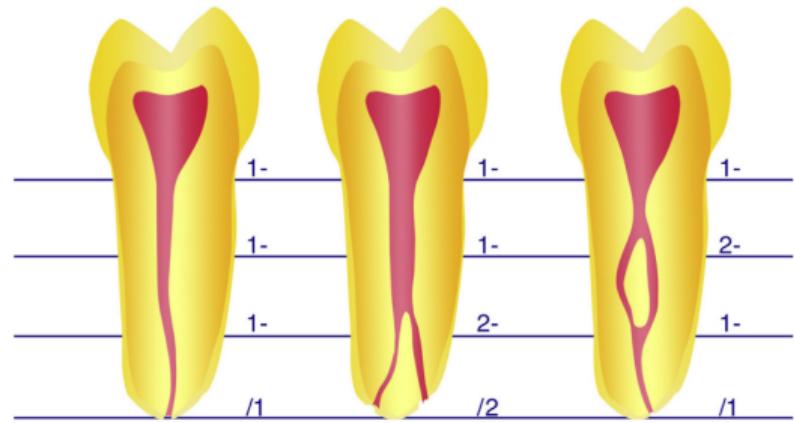
*Sample changer* on the SkyScan 1272

In total:

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

# How?

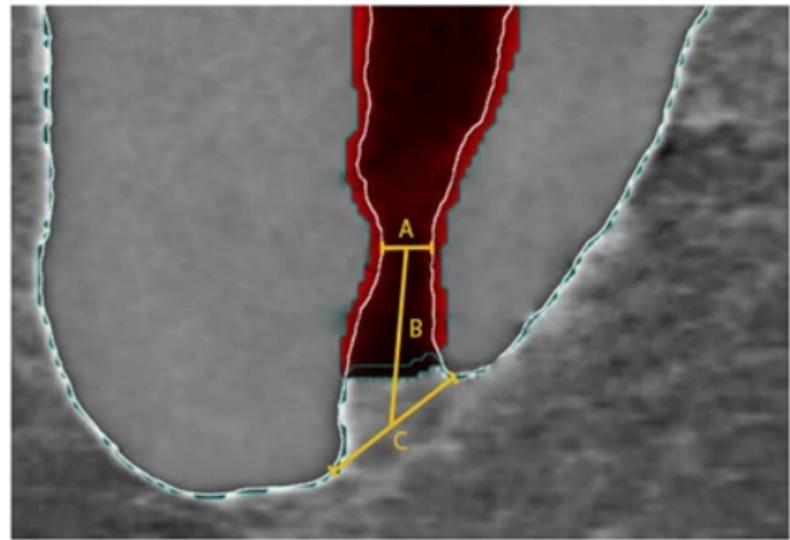
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From [25], Fig. 2

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From [27], Fig. 1

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[gph.is/2nqkple](http://gph.is/2nqkple)

$u^b$

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The screenshot shows a GitHub repository interface. At the top, it displays 'master' branch, '1 branch', '1 tag', 'Go to file', 'Add file', and a green 'Code' button. Below this is a list of commits from user 'habi' for an 'Update actions file'. The commits are:

File	Description	Time Ago
.github/workflows	Update actions file	20 days ago
.gitignore	Only 'mode' changes	2 months ago
DownloadFromOSF.ipynb	Clean run of download script	22 days ago
README.md	Typo in Binder badge & link to full repo on Binder	22 days ago
Tooth.Border.jpg	Only 'mode' changes	2 months ago
Tooth.Characterization.ipynb	Only 'mode' changes	2 months ago
ToothAnalysis.ipynb	Only select a subset if we actually have data:wq	22 days ago
ToothAxisSize.ipynb	Clean run of notebook	22 days ago
ToothDisplay.ipynb	Display Tooth045 for manuscript	22 days ago
requirements.txt	We also need this	2 months ago
treebeard.yaml	Add treebeard configuration	20 days ago

Below the commits is a preview of the 'README.md' file. It contains:

README.md

DOI: 10.5281/zenodo.3999402 treebeard.yaml failing

Launch binder

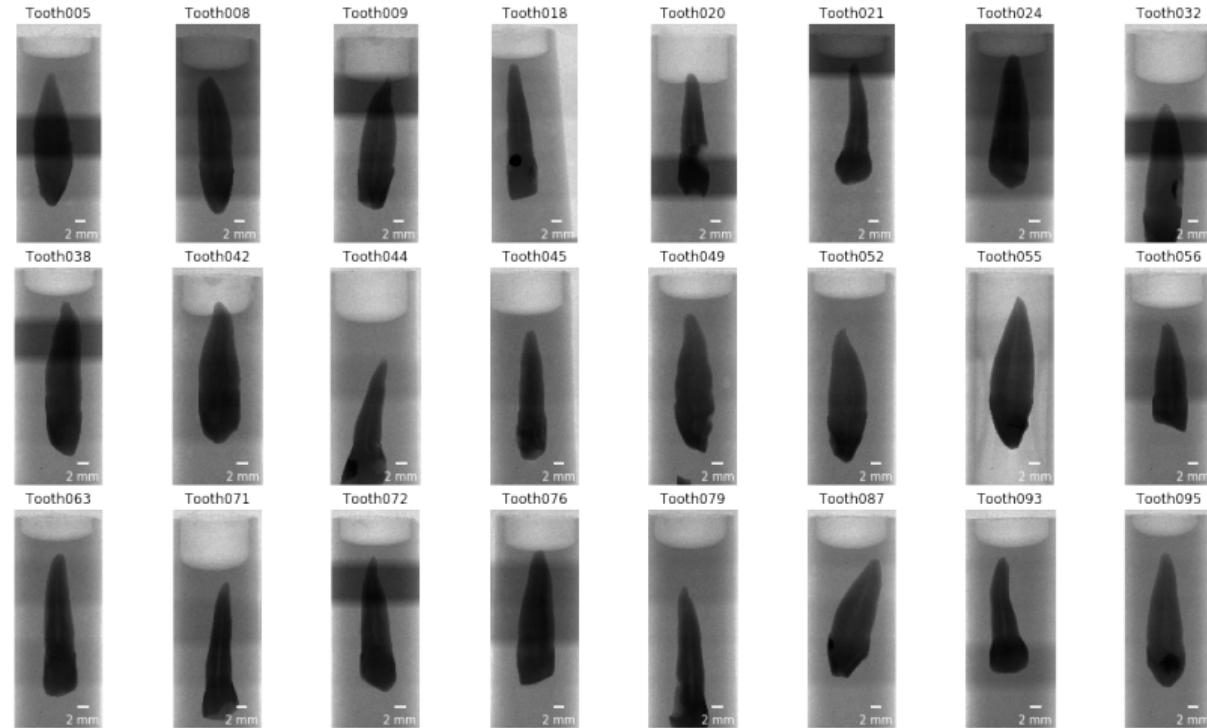
A big tooth cohort

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](#).

$\mu$ b

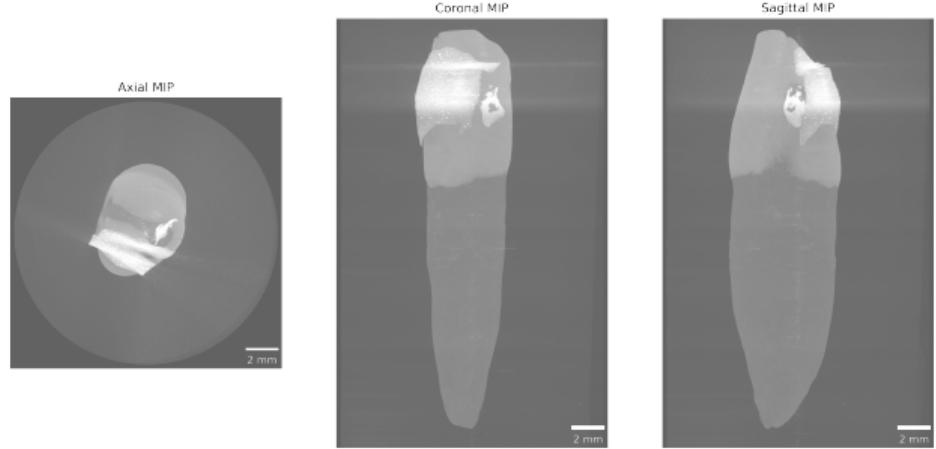
# $\mu$ CT imaging



*u*<sup>b</sup>

# Dataset cropping

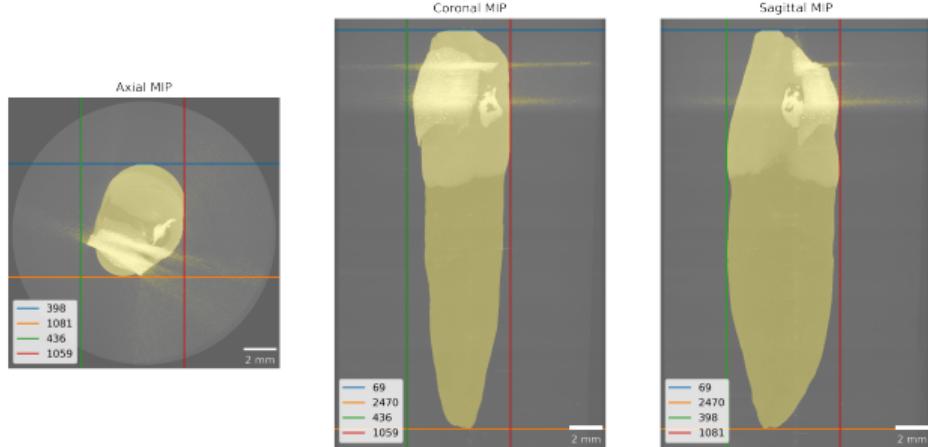
- Full datasets: 326 GB
- Cropped datasets: 115 GB



*u*<sup>b</sup>

# Dataset cropping

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



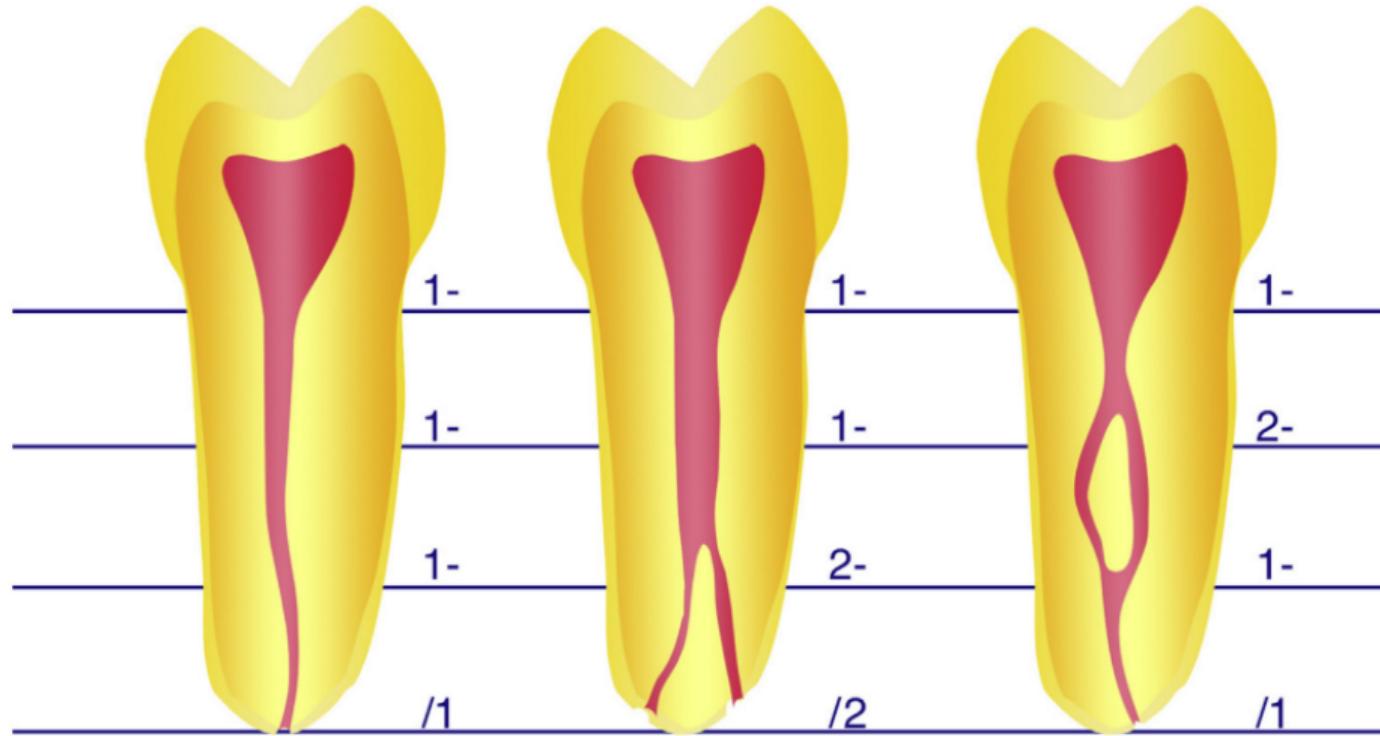
*u*<sup>b</sup>

# Tooth morphology



*u<sup>b</sup>*

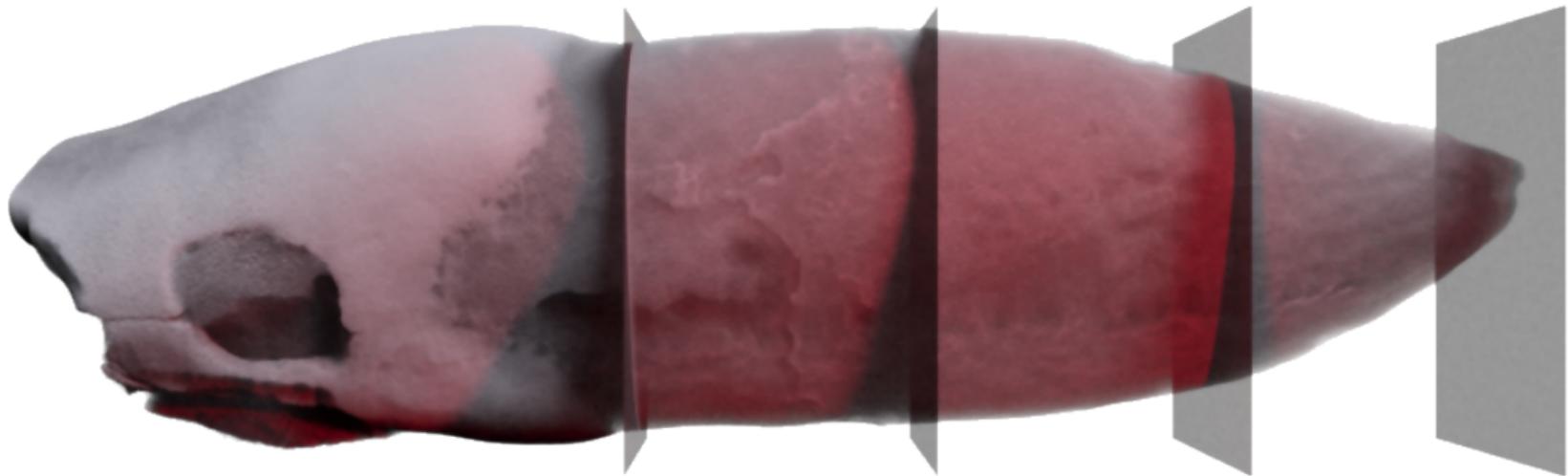
# Tooth morphology



From [25], Fig. 2

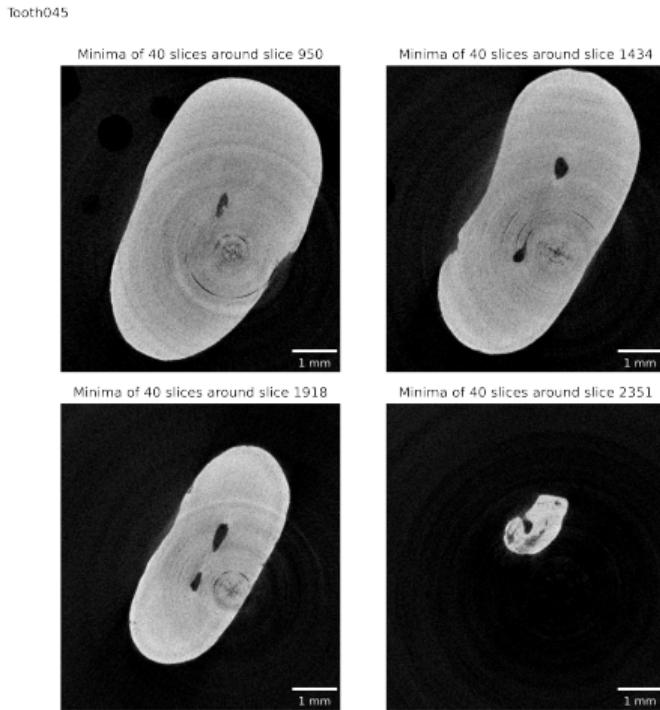
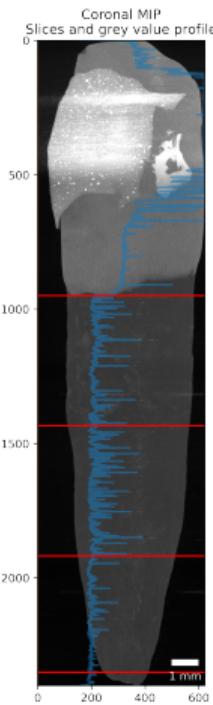
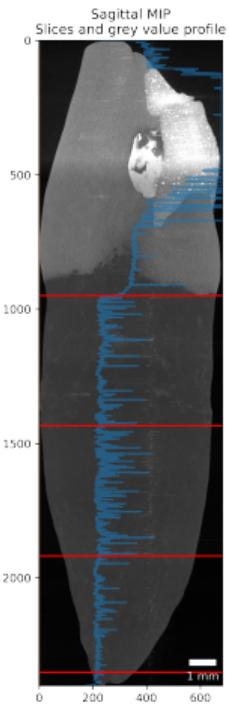
*u<sup>b</sup>*

# Tooth morphology



*u<sup>b</sup>*

# Detection of enamel-dentin border



*u*<sup>b</sup>

# Detection of enamel-dentin border

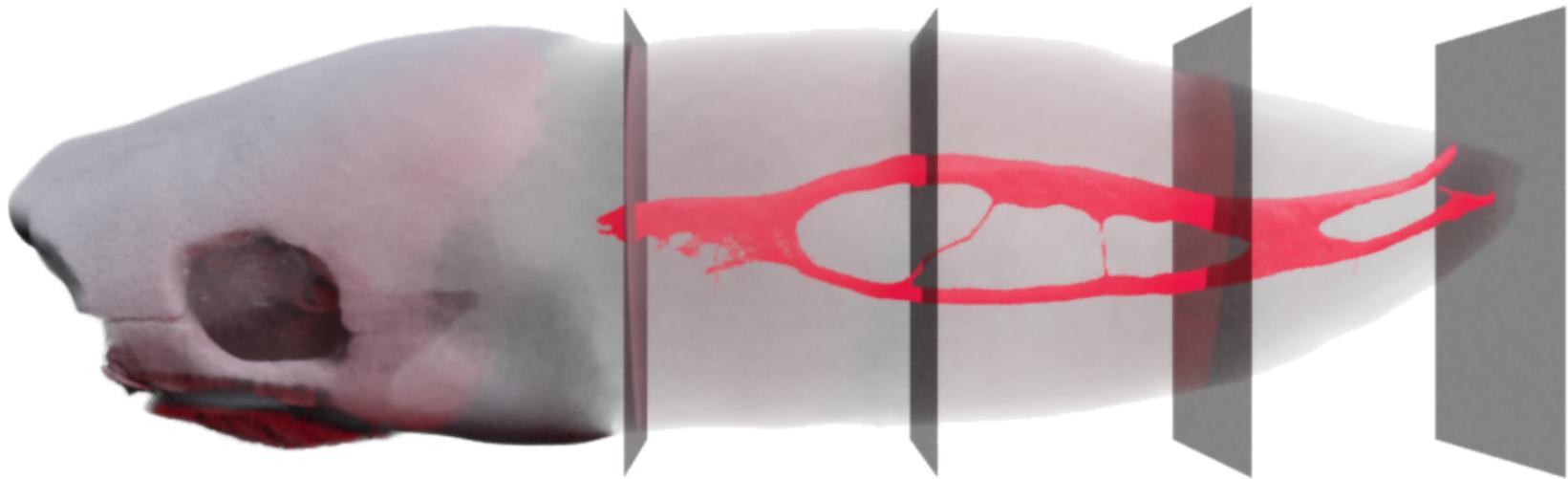


# Classification of root canal configurations

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
		1-2-1/1	1
	Lingual	1-1-1/1	2
		1-1-1/2	1

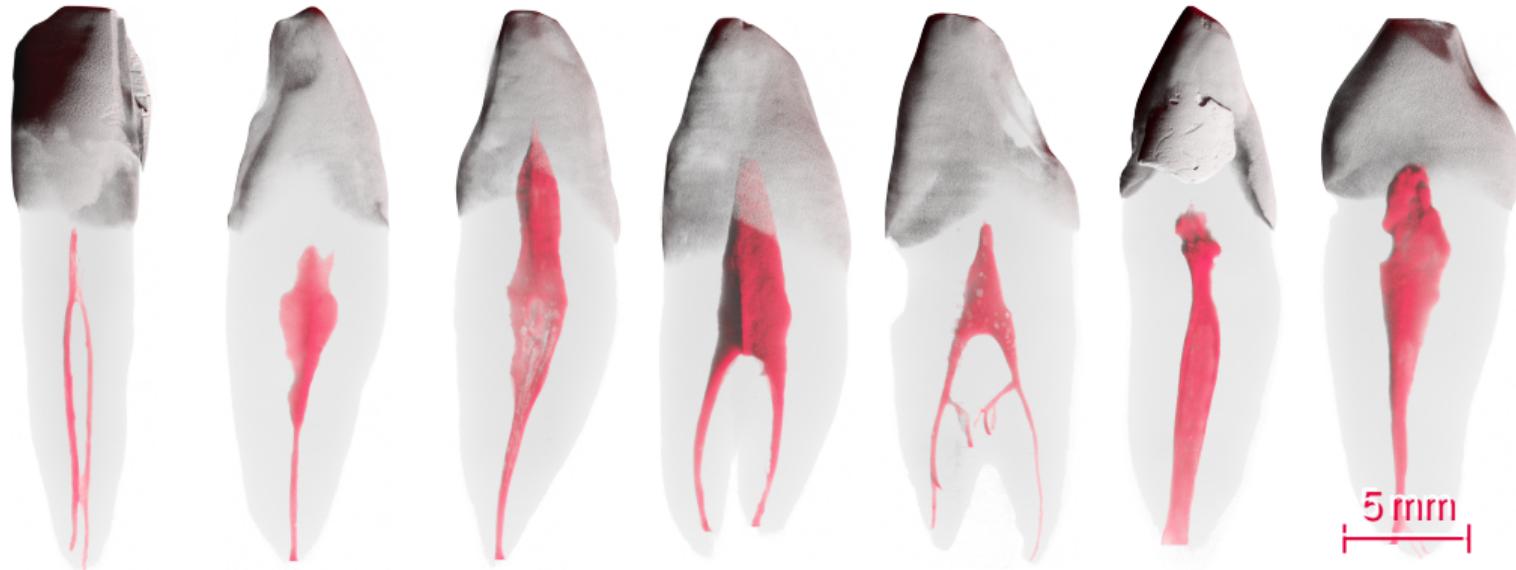
*u<sup>b</sup>*

# Extraction of root canal space



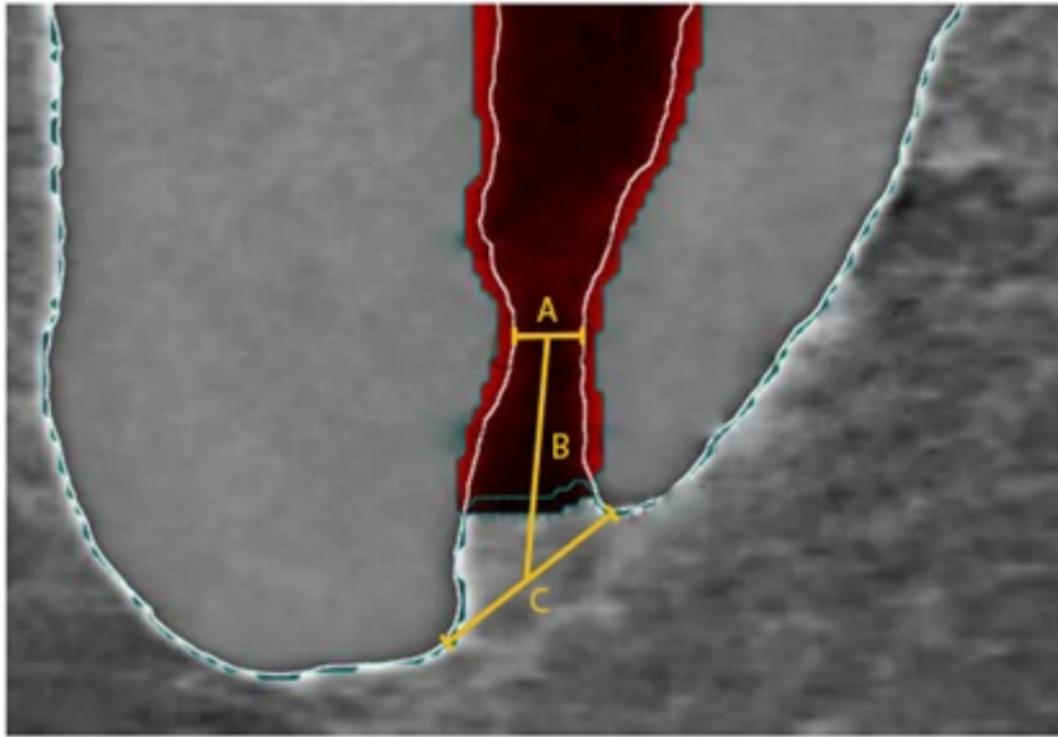
*u<sup>b</sup>*

# Results of root canal space extraction



*u*<sup>b</sup>

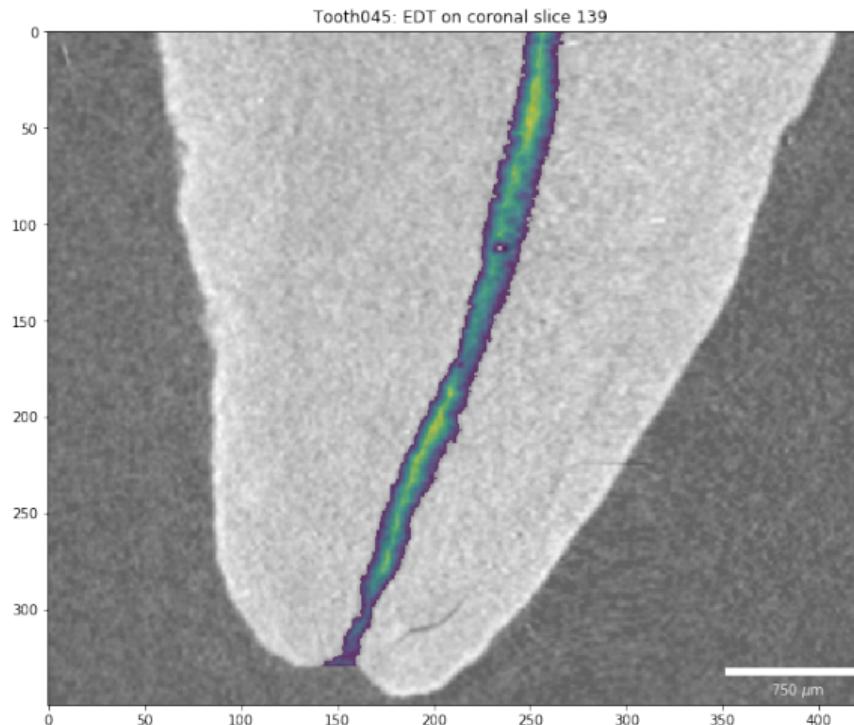
# Physiological foramen geometry



From [27], Fig. 1

*u*<sup>b</sup>

# Physiological foramen geometry

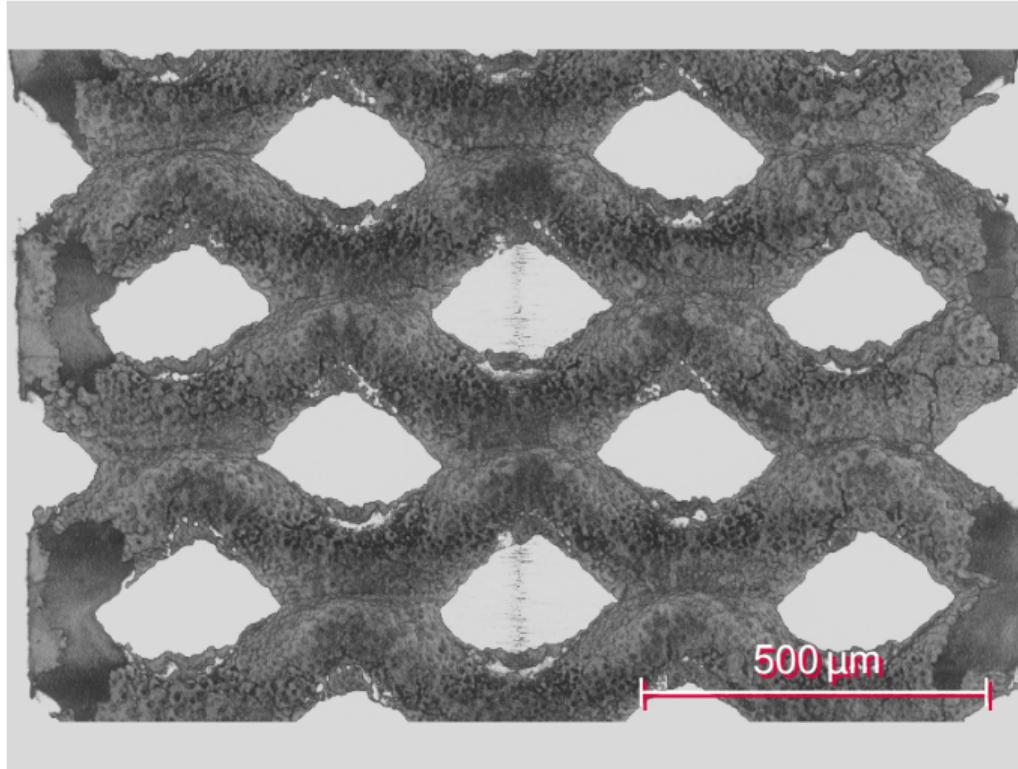


# Conclusion ZMK

- 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

$u^b$

# Metal foam



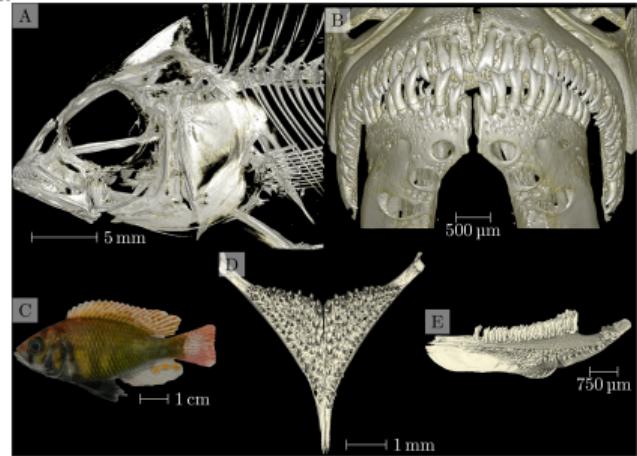
Etienne Berner NanoElectroCatalysis Group

*u*<sup>b</sup>

# Data wrangling by example: Cichlids

Collaboration with team of *Aquatic Ecology & Evolution*, from the Institute of Ecology and Evolution<sup>a</sup>

- 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

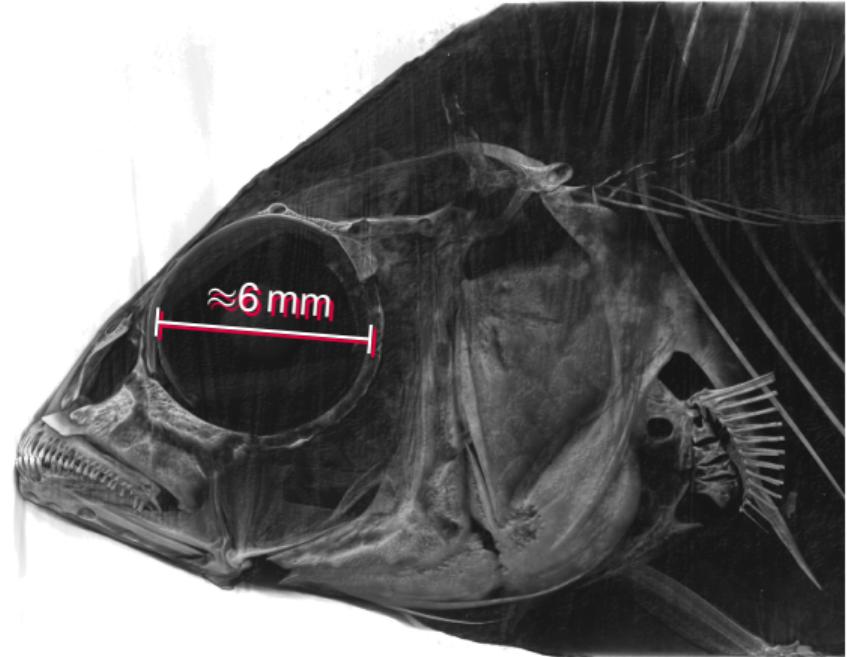


DOI:gsst8t, Fig. 1

<sup>a</sup>11.

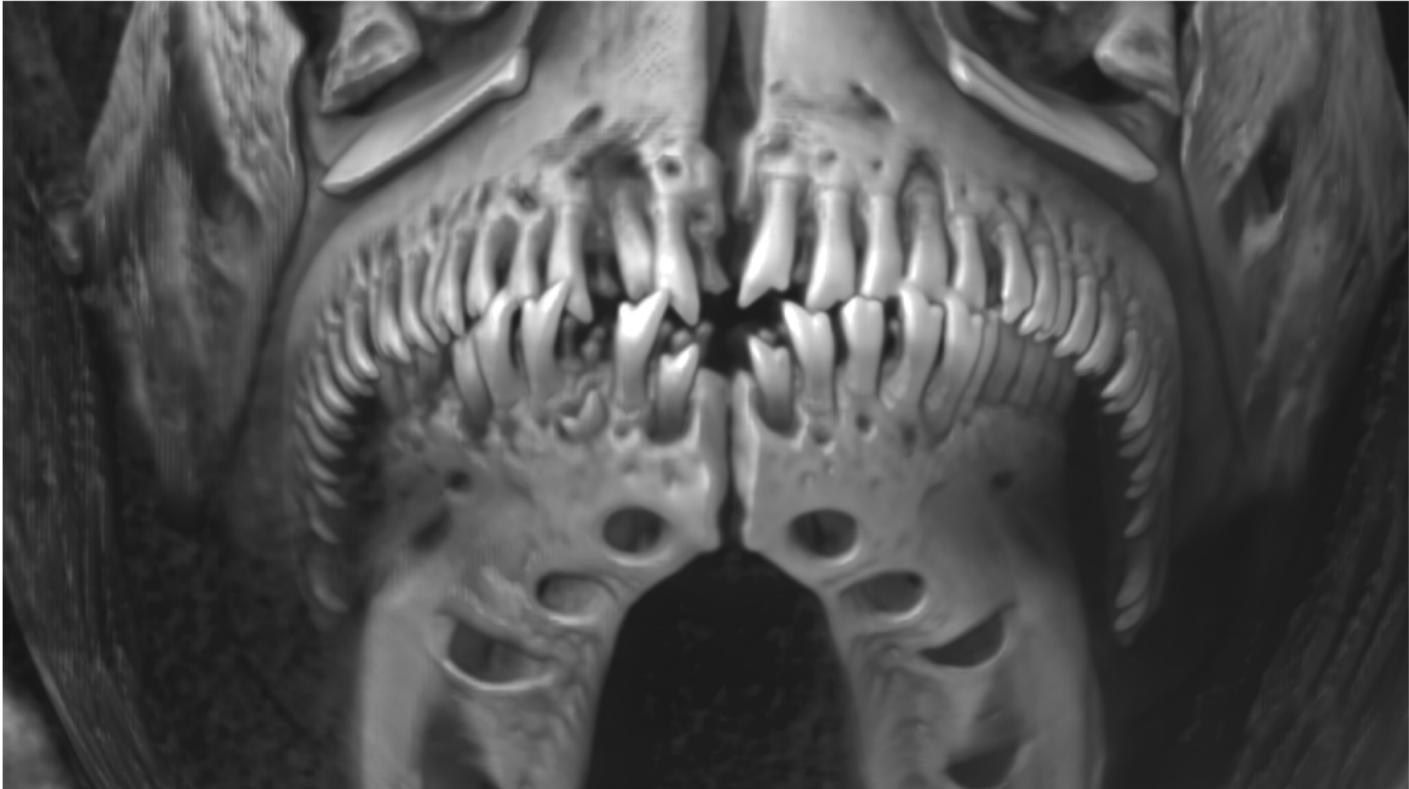
*u<sup>b</sup>*

# Visualization of cichlid head



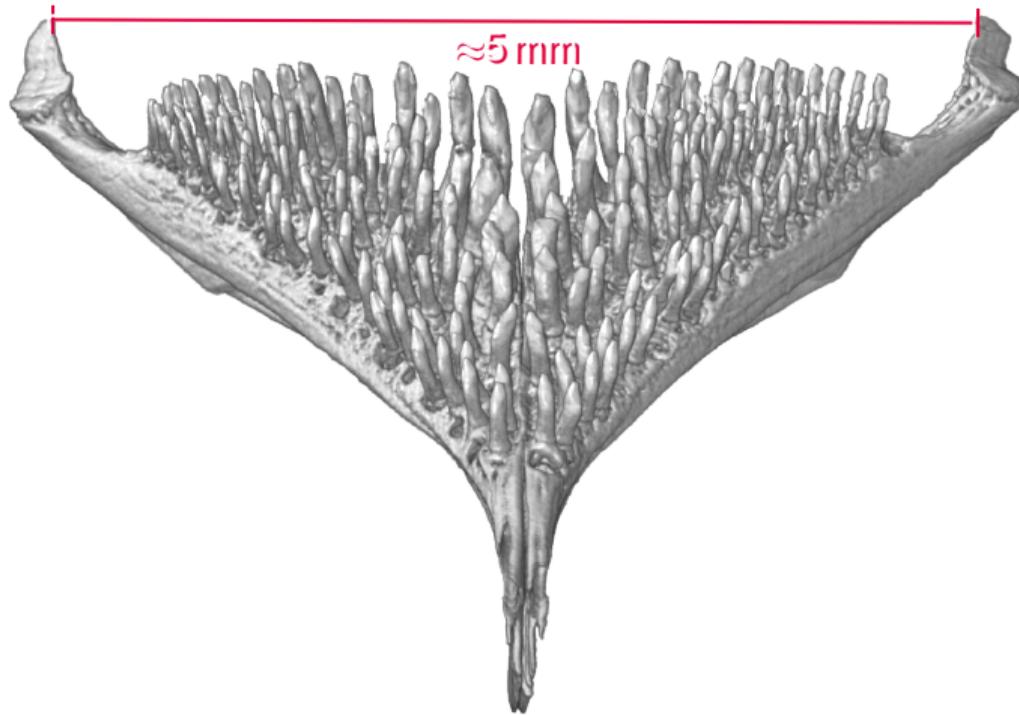
*u*<sup>b</sup>

# Visualization of cichlid head



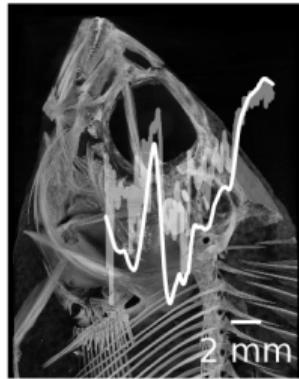
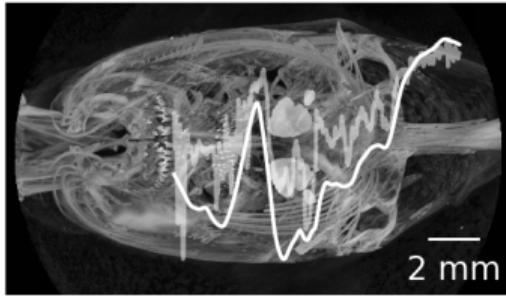
*u*<sup>b</sup>

# Visualization of segmented pharyngeal jaw



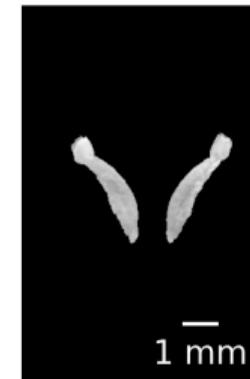
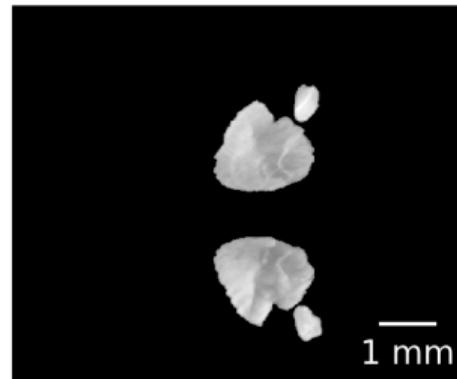
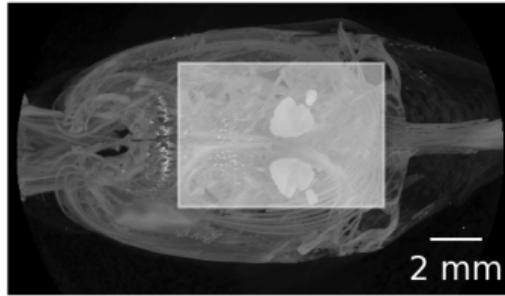
*u*<sup>b</sup>

# Data wrangling by example: Cichlids



*u*<sup>b</sup>

# Data wrangling by example: Cichlids



*u*<sup>b</sup>

# Thanks!

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

# Colophon

- This BEAMER presentation was crafted in  $\text{\LaTeX}$  with the (slightly adapted) template from *Corporate Design und Vorlagen* of the University of Bern.
  - Complete source code: [git.io/fjpP7](https://git.io/fjpP7)
  - The  $\text{\LaTeX}$  code is automatically compiled with a GitHub action to a (handout) PDF which you can access here: [git.io/JeQxO](https://git.io/JeQxO)
- Did you spot an error?
  - File an issue: [git.io/fjpPb](https://git.io/fjpPb)
  - Submit a pull request: [git.io/fjpPN](https://git.io/fjpPN)
  - Send me an email: [david.haberthuer@unibe.ch](mailto:david.haberthuer@unibe.ch)

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