We will live on this planet at least for the next 10 000 years.

This statement leads to two GNEUS projects

We have time.

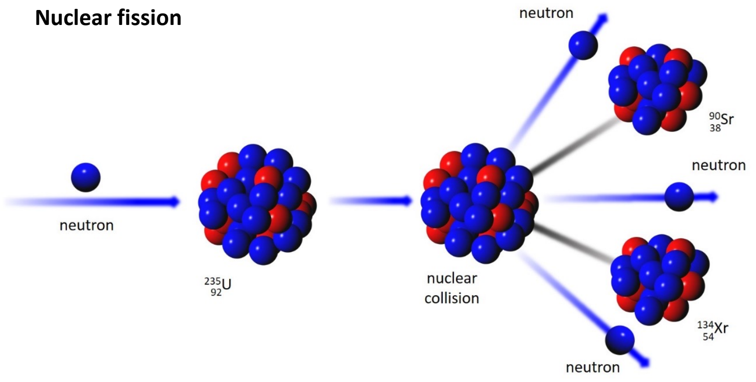
- There is no need to panic.
- We will have time to investigate the machinery of live (and other interesting phenomenon) in great depths.

GNEUS Project A: We will build a dedicated instrument for macromolecular crystallography on a HiCANS neutron source.

- Because this is the most cost efficient way to produce neutrons without adding too much entropy caused by the daughter nuclei of fission or the radioactivity of spallation.

How to get neutrons

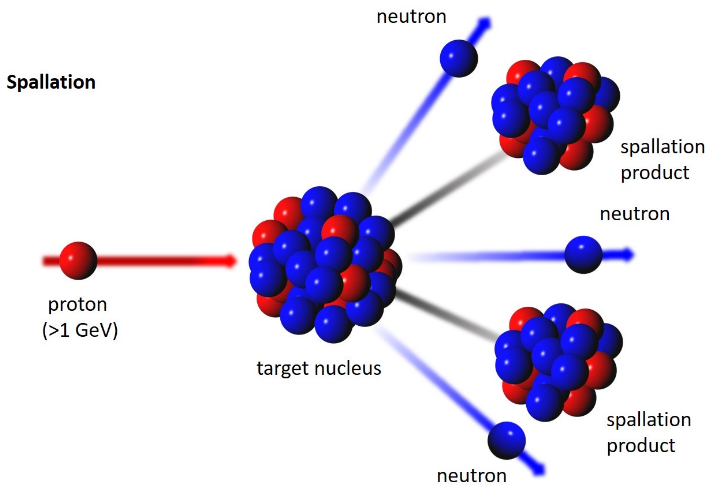
Nuclear fission



Reactor based neutron source

(ILL, FRM II, NIST, JINR, ANSTO a.m.m.)

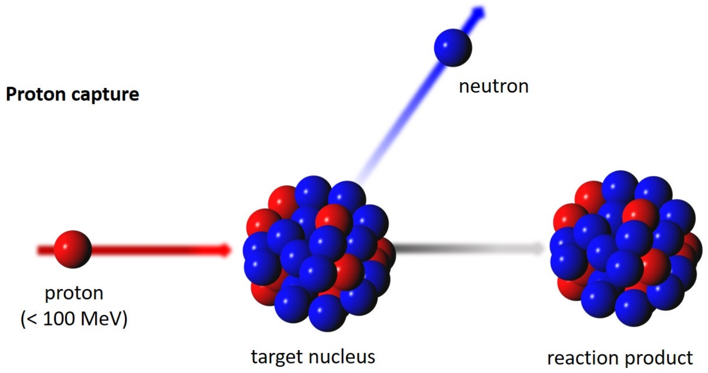
Spallation



Spallation based neutron source

(ESS, ISIS, SINQ, SNS, CSNS, J-PARC, KEK)

Nuclear processes



Accelerator based neutron source

(LENS, RANS, HUNS, NUANS, IREN a.o.)

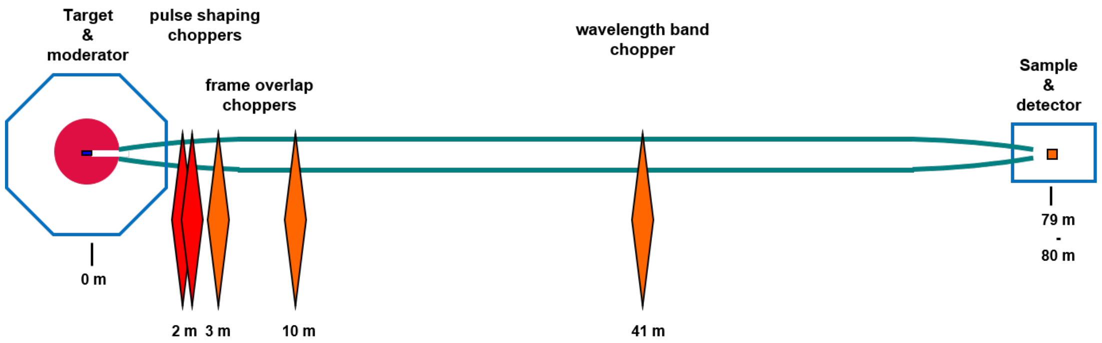
A 20 m instrument at the 96 Hz target station or a 80 m instrument at the 24 Hz target station?

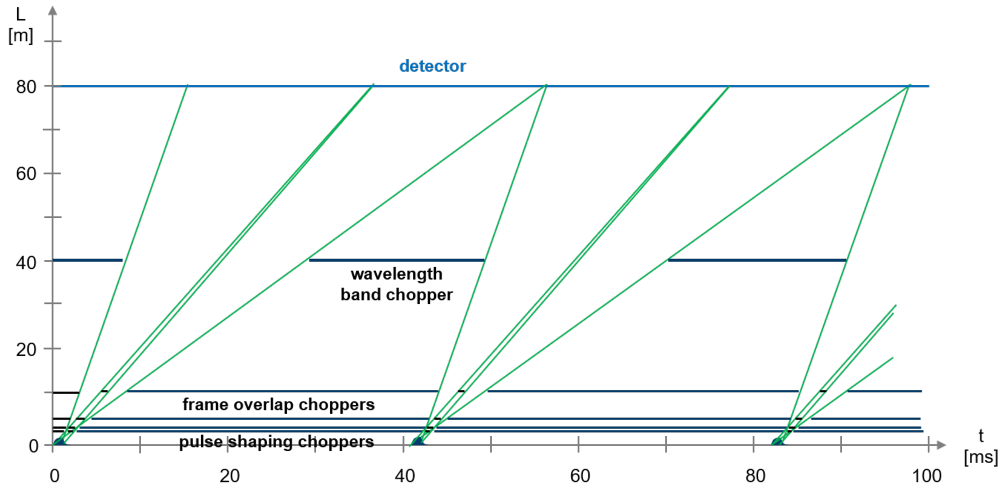
Instrument Name	Source	Flux	neutron pulse	instrument length	rep rate	div FWHM	total deltad/d at given scattering angle		
		n/s/cm^2	µs	m	Hz	° degree	5°	45°	85°
MANDI	SNS	4,50E+07	17,4	30	60	0,8	15,96%	1,40%	0,17%
EWALD	SNS		43,3	60	15	0,38	7,58%	0,68%	0,15%
iBIX	J-PARC	7,00E+07	500	40	25	0,2	4,69%	2,50%	2,47%
NMX	ESS		2860	156	14	0,1	4,14%	3,63%	3,63%
NMD	HBS 96Hz 20 m		254	20,4	96	0,7	14,18%	2,75%	2,47%
NMD	HBS 24Hz 80m		666,7	80	24	0,7	14,06%	2,05%	1,65%

Design considerations:

1. Flux
2. Flux
3. Resolution in reciprocal space
4. Round or Flat top uniform beam profile

The 80 m Single Crystal Diffractometer

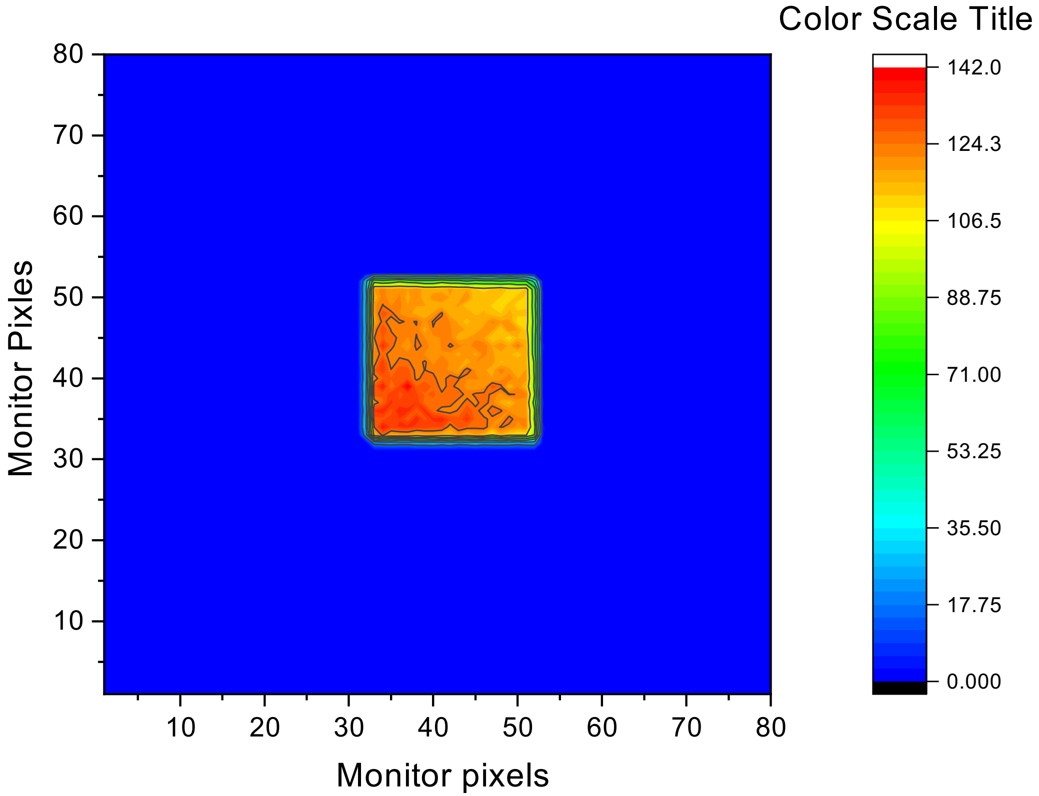




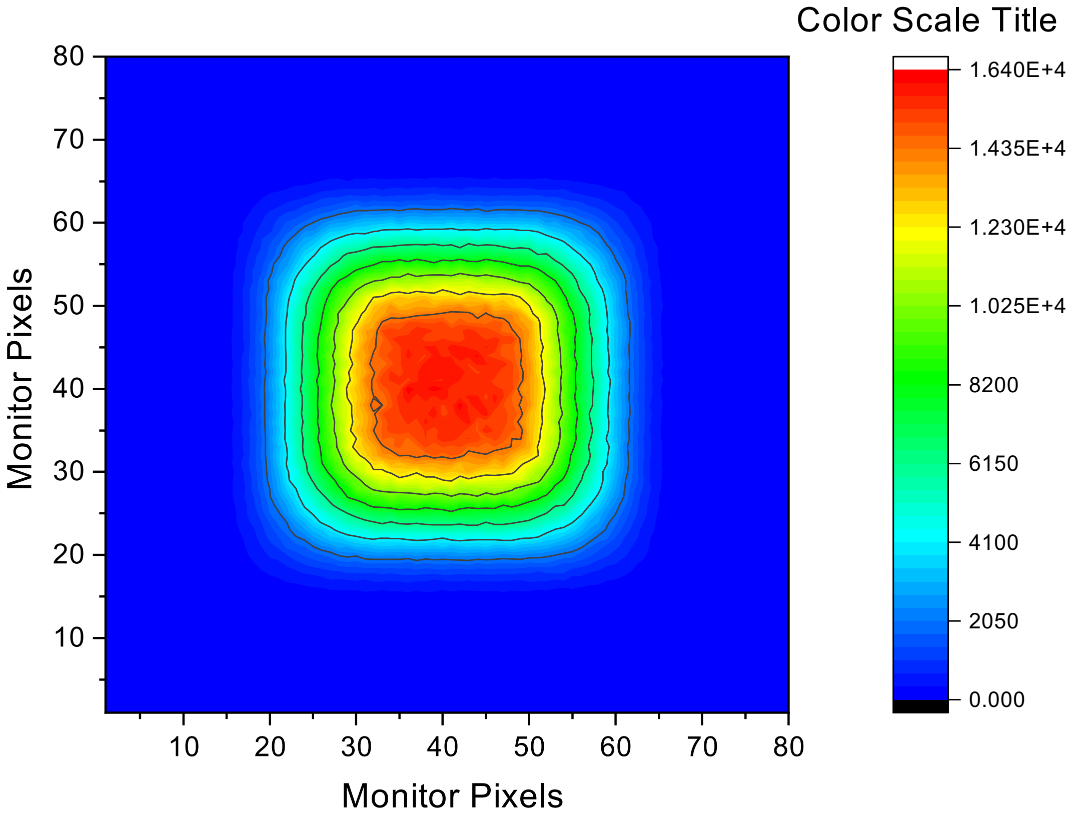
- **Applications**
  - All kinds of powder samples
- **Concept and Requirements**
  - TOF diffractometer using pulse shaping and wavelength frame multiplication
  - Variable up to very high resolution
- **Choices**
  - Thermal moderator
  - Low frequency (24 Hz, 667 µs)
  - 80 m length (source to detector)
  - Detector range: 7° - 175°
- **Characteristics**
  - Bandwidth: 1.65 Å, standard: 0.75 – 2.4 Å
  - d-range : 0.32 – 16.7 Å
  - High-Resolution option (100 µs pulse)
    - 0.17 – 0.59% (θ>90°), 0.04% for 175°
    - Estimated flux at sample: $1 \cdot 10^6$ n/(cm²s)
  - High-Intensity option (667µs pulse)
    - 0.47 – 1.4% (θ>90°)
    - Estimated flux at sample: $5 \cdot 10^8$ n/(cm²s)

Comparison between the two instruments:

The 20 m instrument:



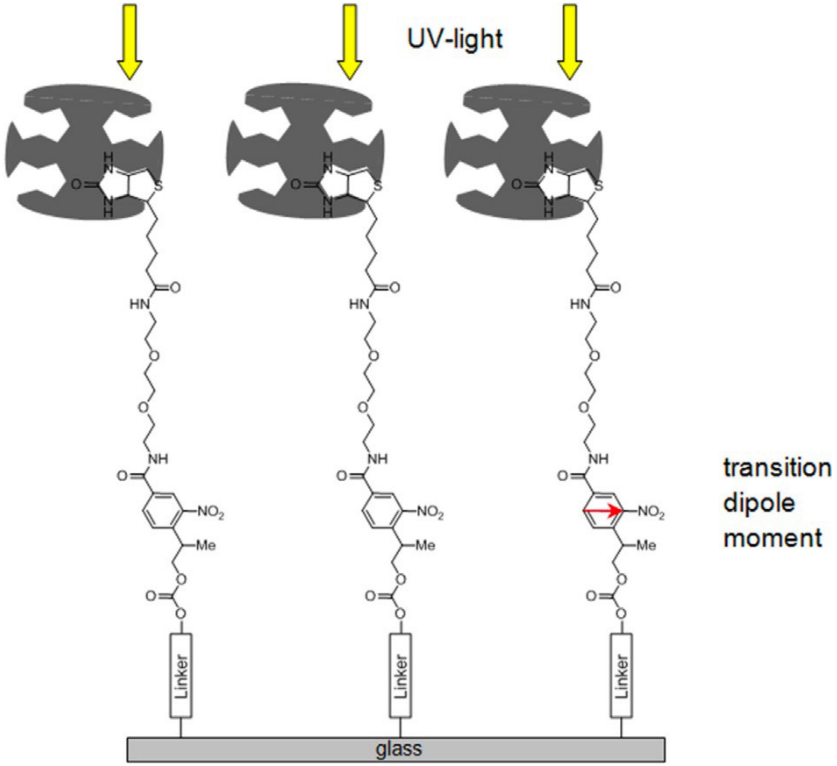
The 80 m instrument:



The 80 m instrument offers twice the flux of the 20 m instrument
in a wavelength band of 2-4 Å.
Disadvantage of the 80 m instrument: A lot of neutrons have to be absorbed near the sample position.

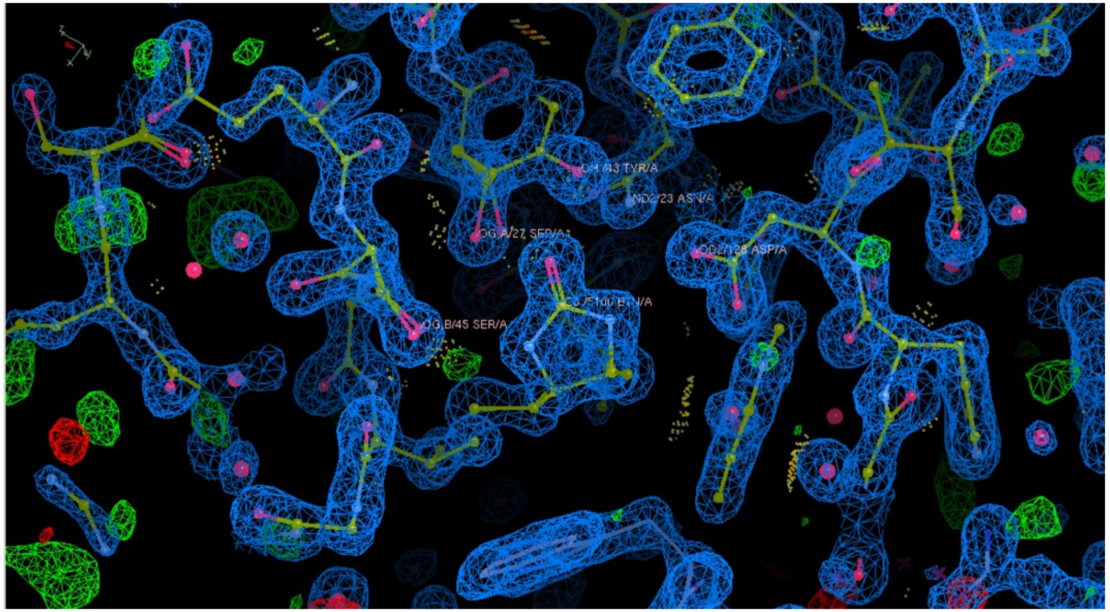
Hands on experience for the GNEUS postdoc with a real sample: The Streptavidin story

- Streptavidin and its ligand biotin are used as linkers on functionalized surfaces

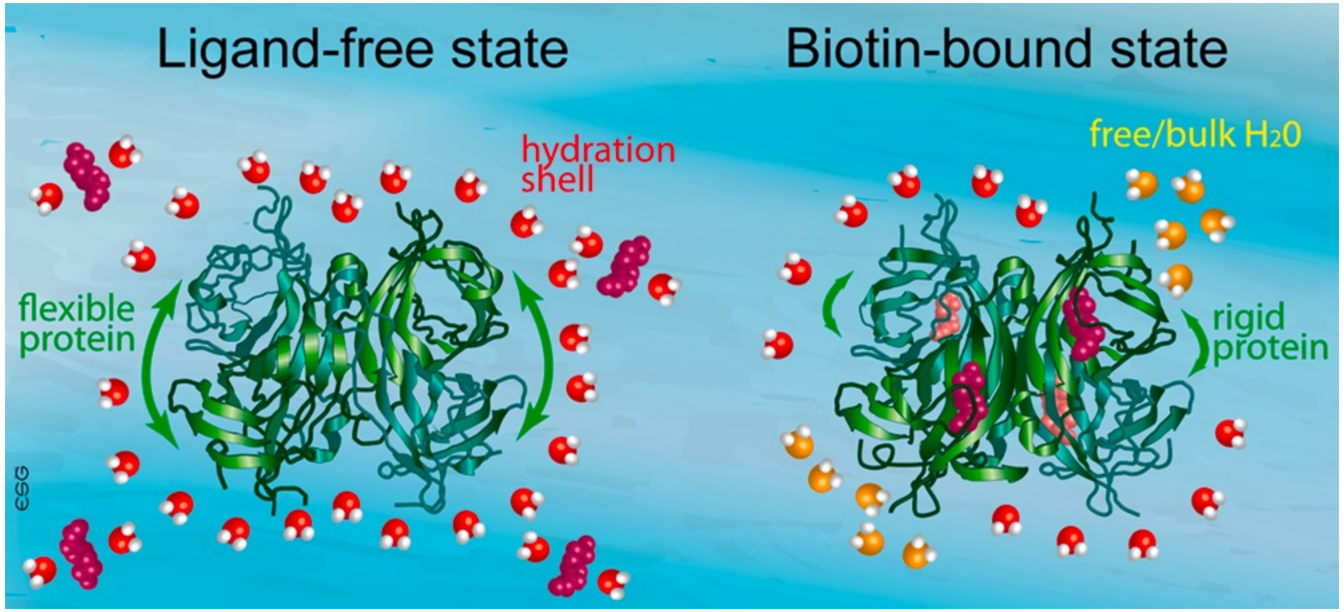


But a neutron structure at room temperature is still missing…

- … and can be compared to Mona Sarter et al.



Made with Coot from 1mk5.pdb



J. Phys. Chem. B 2020, 124, 324–335

Scientific Questions to be adressed:

1. Does the use of polarized neutrons make sense at such an instrument?
2. How can one mitigate the problems associated with the non-uniform beamprofile of the 20 m instrument?
3. How can one assess to which unit cell size one can go using Monte-Carlo Simulations?
4. What is the performance of the 80 m instrument when it is optimised?
5. How can one account for the neutrons to be absorbed near the sample position? How much background do they produce?
6. Do we go out of line of sight from the moderator or do we use a time zero chopper?
7. Does pulse shaping make sense?
8. Does it make sense to go to a smaller bandwidths (1 A instead of 2 A) but afford a shorter instrument?

Secondment: Mirrortron, Budapest, Hungary

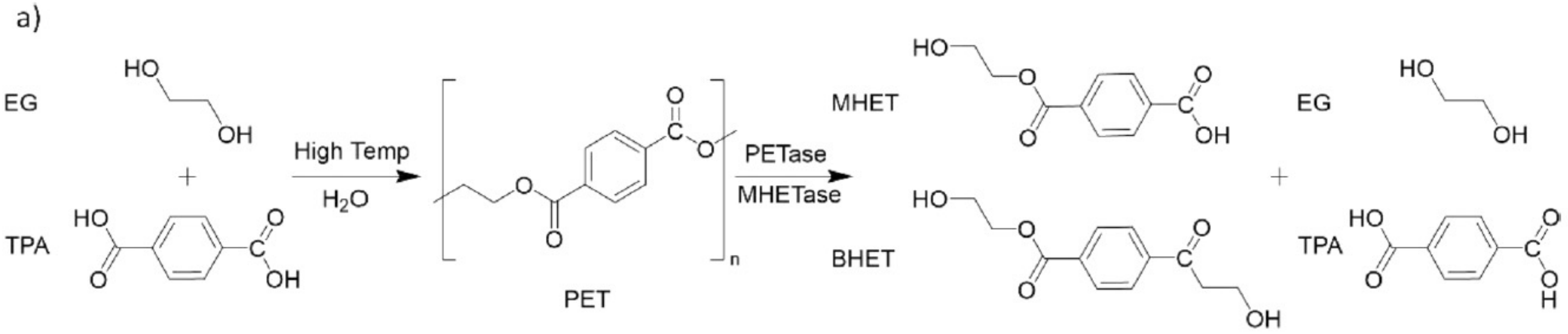
There is no alternative to 100% recycling.

- If we do not recycle 1 % it will pile up over the years.
- We have to tackle even the plastic waste which we have produced by now.
- We have to find ways to produce environmentally friendly plastic materials.

A part of the solution: GNEUS project B: Mechanistic insights into the enzymatic activity of PETases

PETase have been optimized by machine learning

- An optimization process which took only a few weeks…

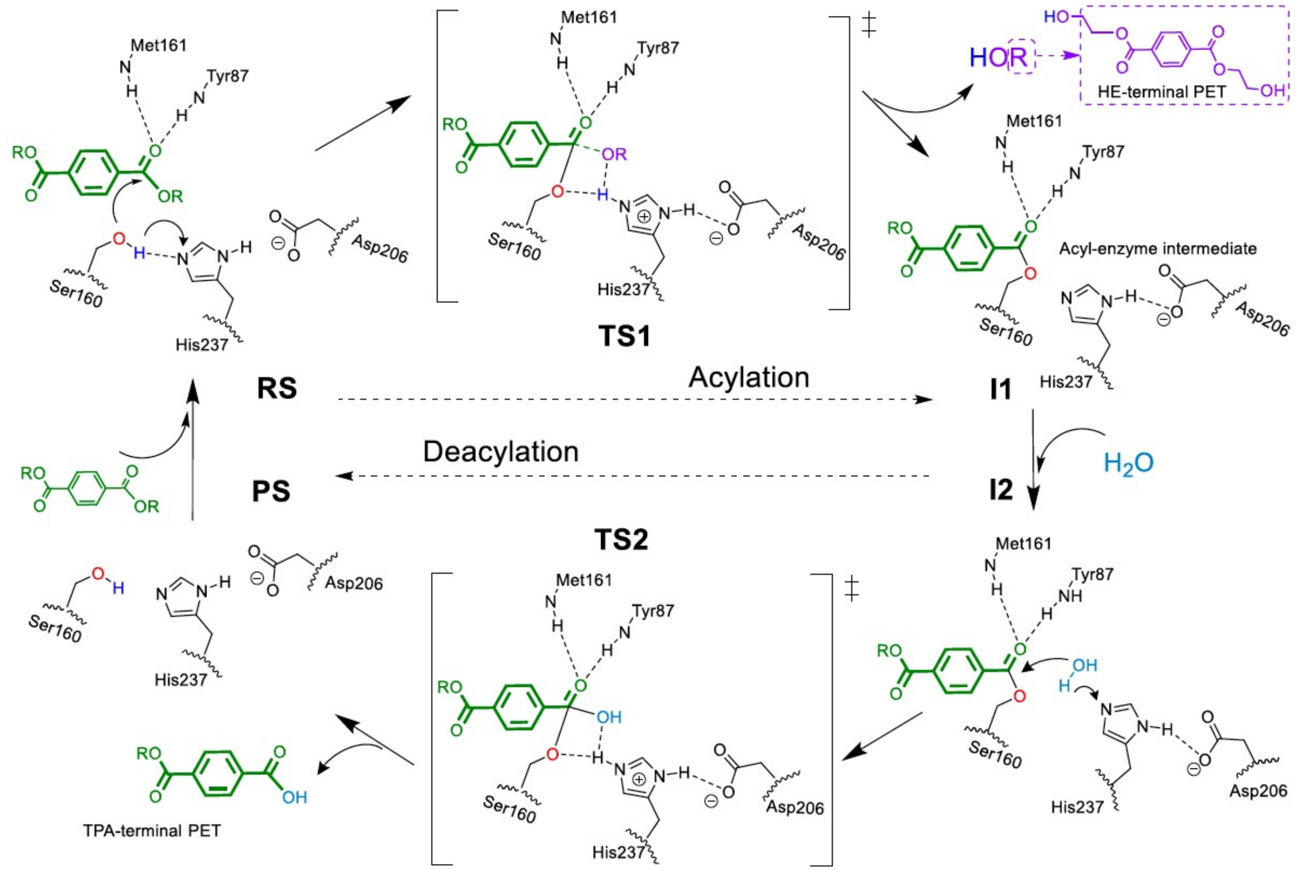


J. Am. Chem. Soc. 2023, 145, 19243–19255

Hydrogen atoms important for the catalytic process

- But they are not observed directly…

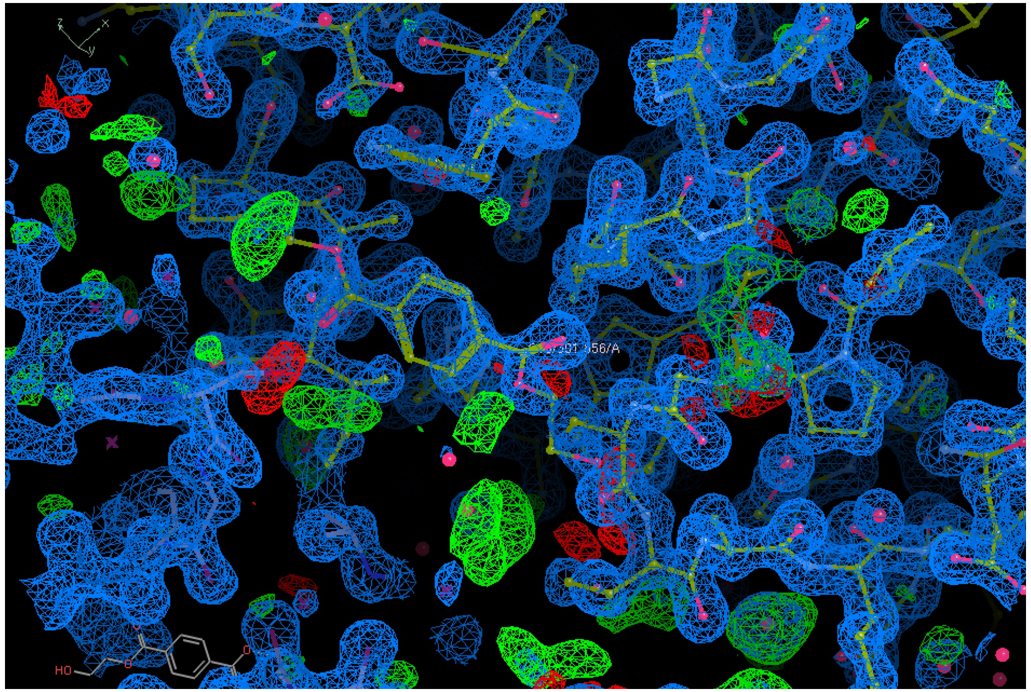
Scheme 1. Schematic Representation of the PETase's Reaction Mechanism

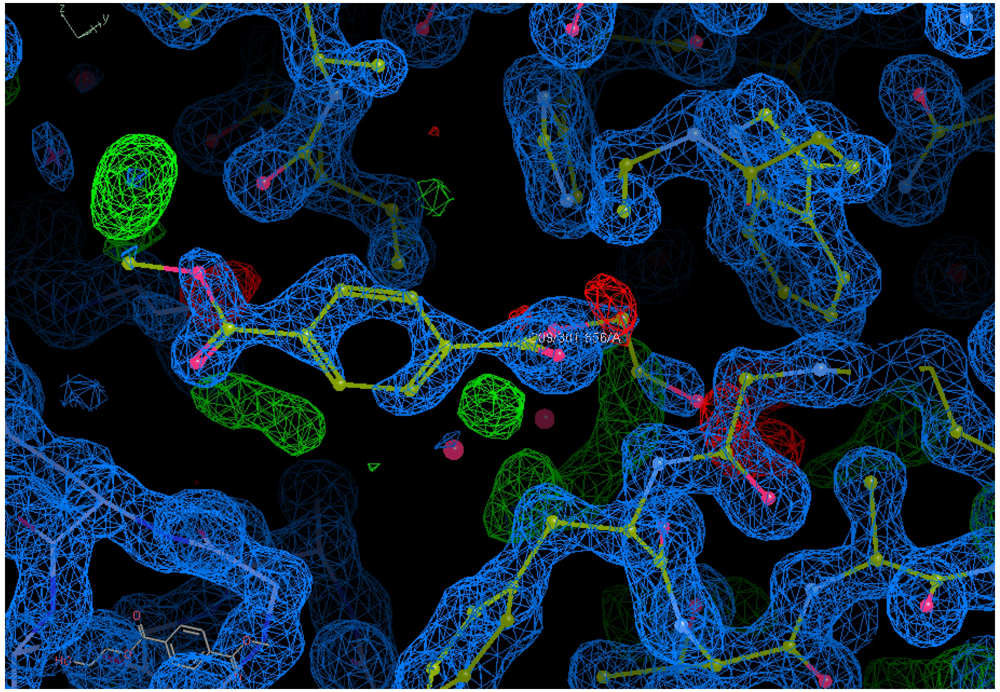


J. Am. Chem. Soc. *2023, 145, 19243–19255*

X-ray structure with substrate bound, 1.3 A resolution

- A lot of unexplained difference density…





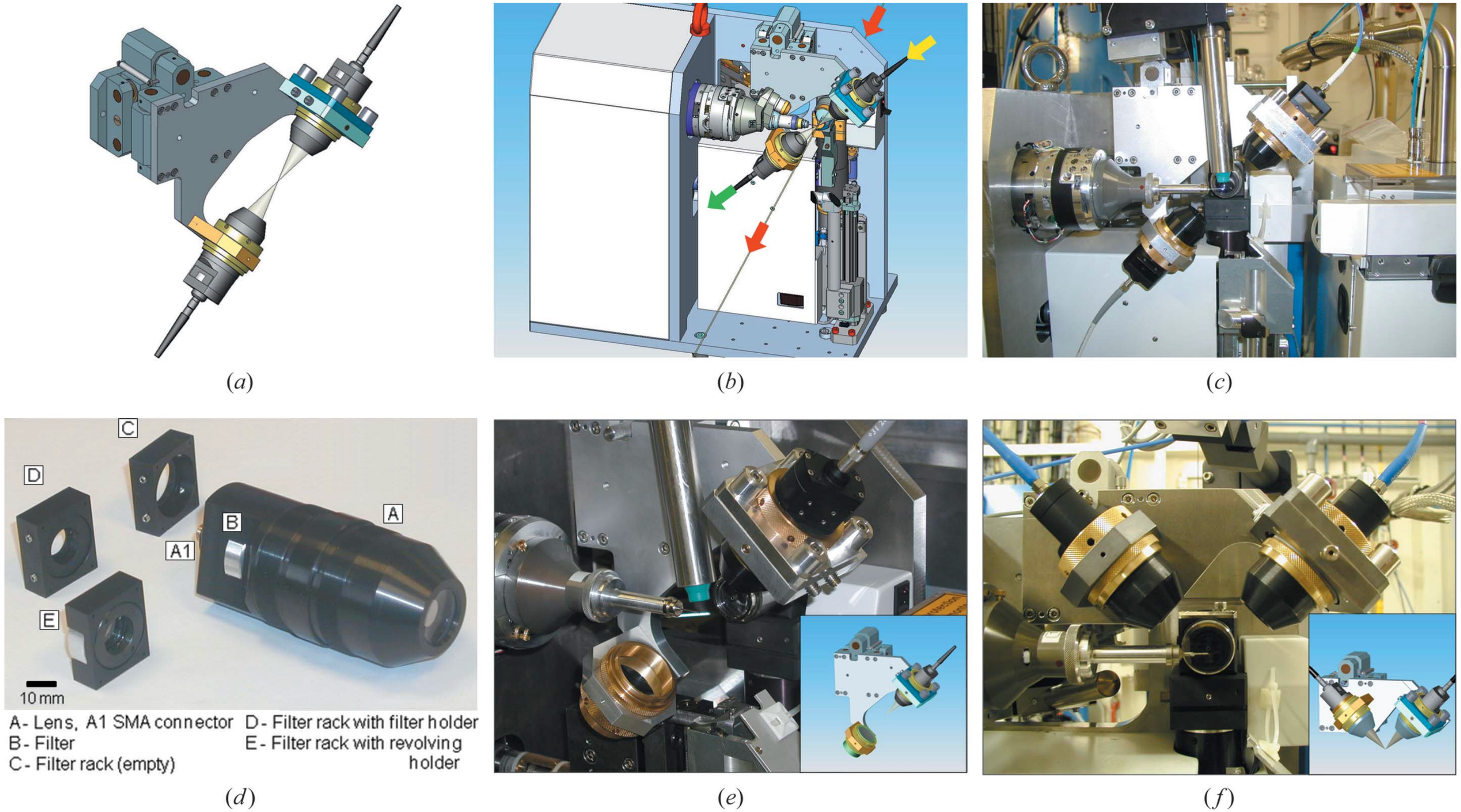
Pictures made with COOT from 5XH3.pdb

Ideas, questions…

- Can we use machine learning also to improve the crystallization conditions in order to get crystal volumes necessary for neutron diffraction?
- Is the optimization process of machine learning efficient? Does it produce new mechanisms?
- What is the largest contribution to the optimization by machine learning? Optimization of substrate binding, reducing the activation energy?
- How can neutron data sets be used as input for machine learning?
- Secondment: Felix Briza from Eitle Hoffmann Patentanwälte

Microspectroscopy as fall back solution…

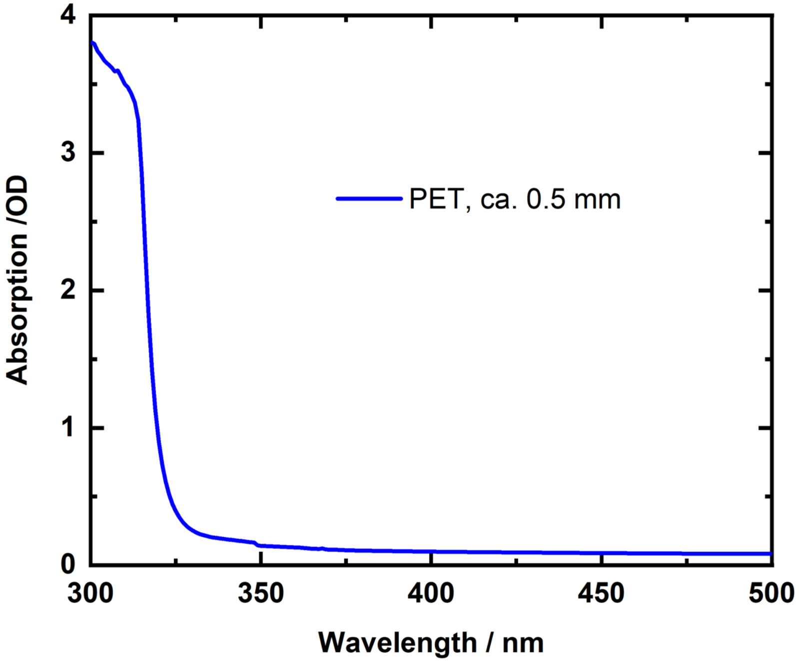
- Publication from Martin Weik:

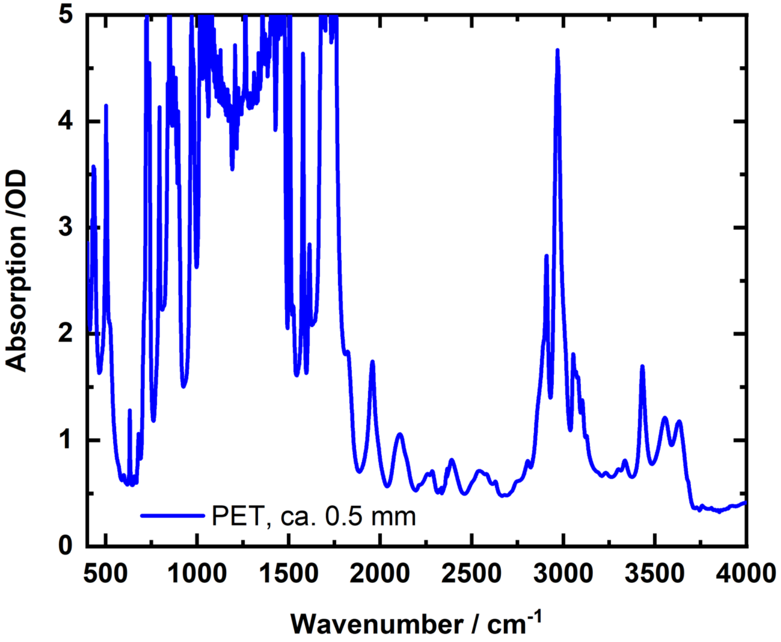


J. Synchrotron Rad. (2009). 16, 163–172

PET – Polyethylenterephthalat

- A very common type of plastic container





Thank you!