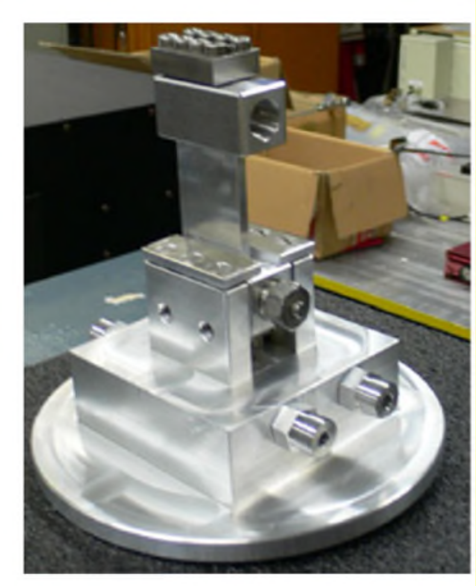
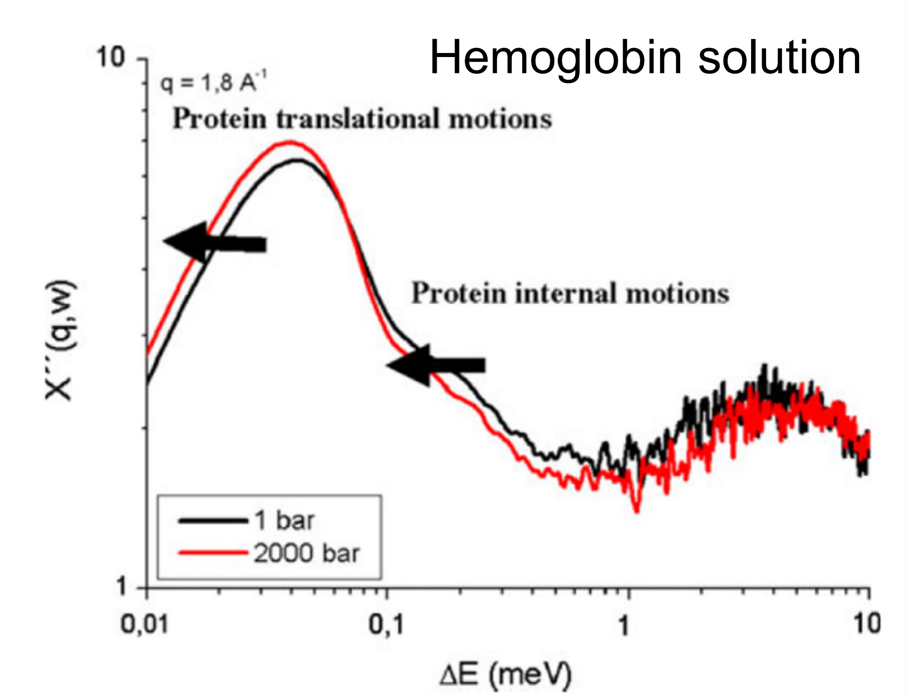
in memory of my former collaborator and friend Dr. Marie Sousai Appavou

† September 13, 2023



2 kbar high pressure cell for TOFTOF



High resolution problems with biological samples

Lit: www.bioneutron.de/publications.html

1) Availability: mg 100-200 mg required, QENS
2) biomolecule solutions: large water background, 80%
3) diffusion of water and biomolecules limit resolution

What to do?

a) Study motions via elastic scans versus T or resolution
b) D_2O- hydrated samples: no diffusion effects, wide temperature range: ‚Elastic resolution spectroscopy' (2001)

Elastic intensity Scattering function * resolution

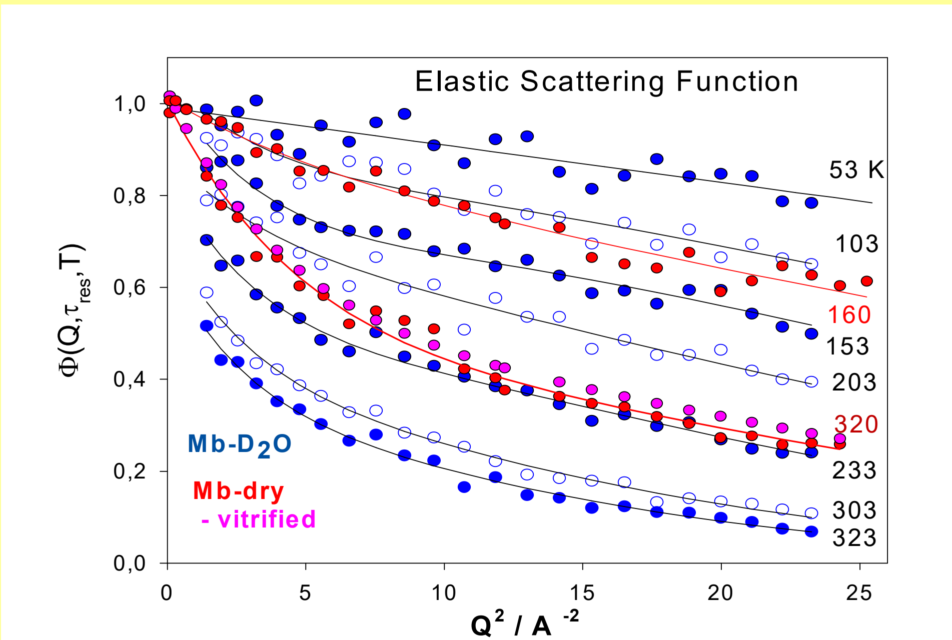
$$S_R(Q, \omega = 0, \Delta\omega) = \frac{1}{\pi} \int_0^{\infty} dt \cdot I_s(Q, t/\tau_c) \cdot R(t/\tau_{res}).$$

$$S_R(Q, \tau_{res}, \tau_c) = \tau_{res} \cdot F_R\left(Q, \frac{\tau_{res}}{\tau_c}\right).$$

Scaling function

Elastic Scattering Function (1):
vary temperature at fixed resolution

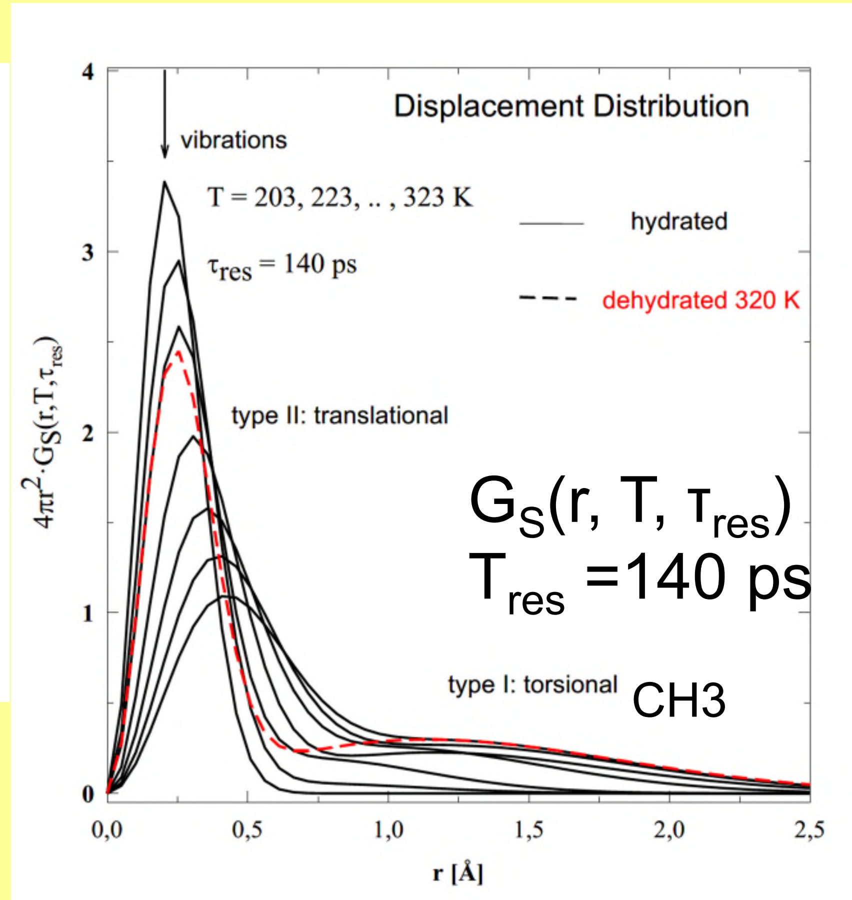
hydrated / dehydrated myoglobin
Backscattering IN13



Doster, Settles BBA 2005

Two components: torsional, CH3
local diffusion

displacement distribution function



$G_S(r, T, \tau_{res})$
$T_{res} = 140$ ps

CH3

r /A

Elastic Scattering Function (2): vary resolution at fixed T

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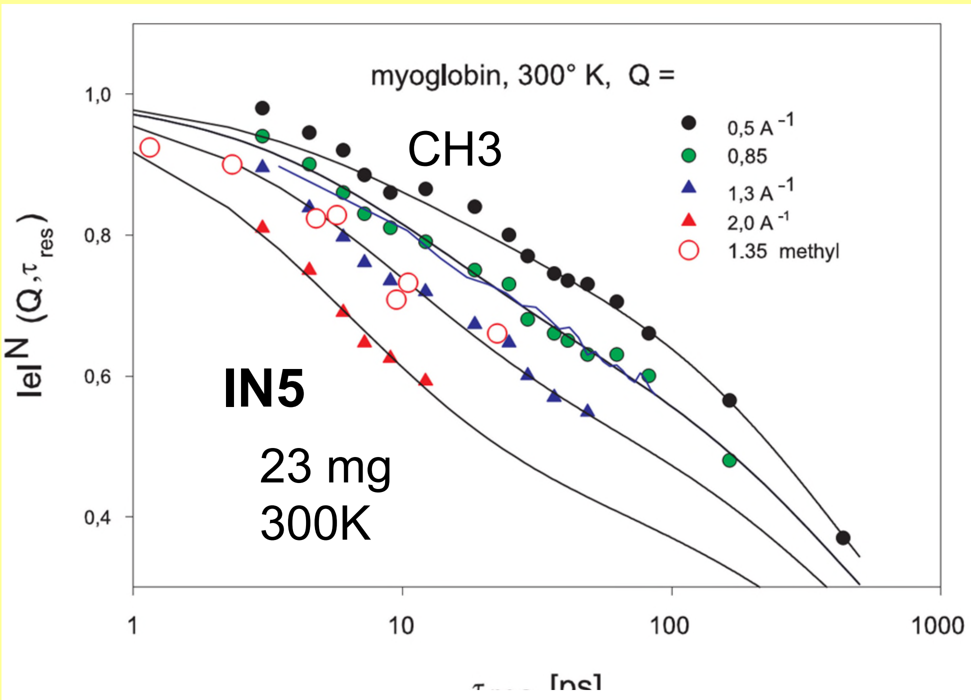
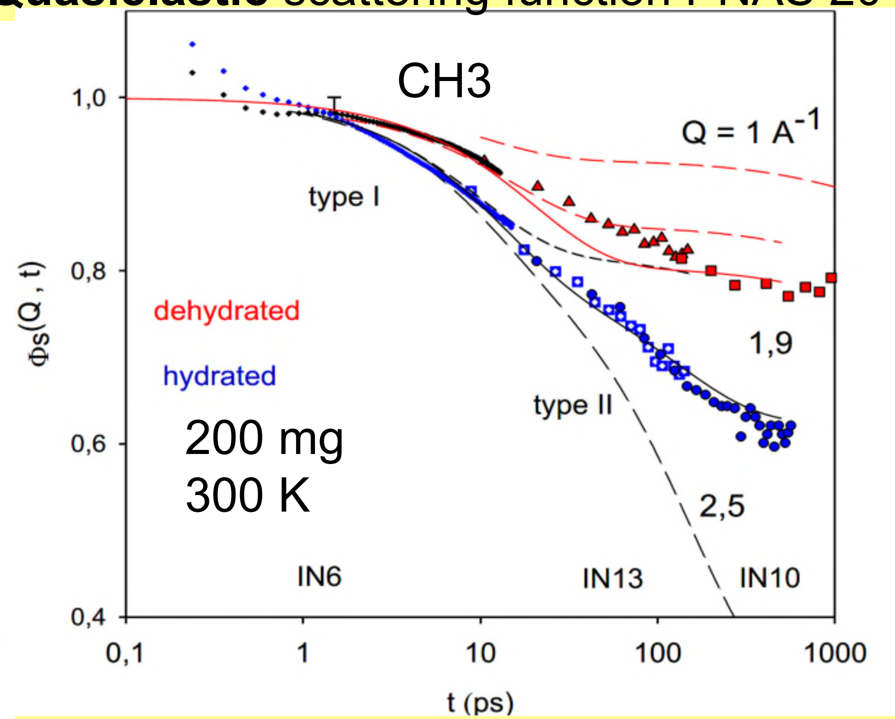
Elastic scattering function (2001)



Quasielastic scattering function PNAS 2019



$$t_{res} = 2\hbar / \Delta E_G .$$

t /ps

$$\Delta E_G = (273400/\lambda^3 S) \quad \text{meV Å}^3 \text{ rpm}$$

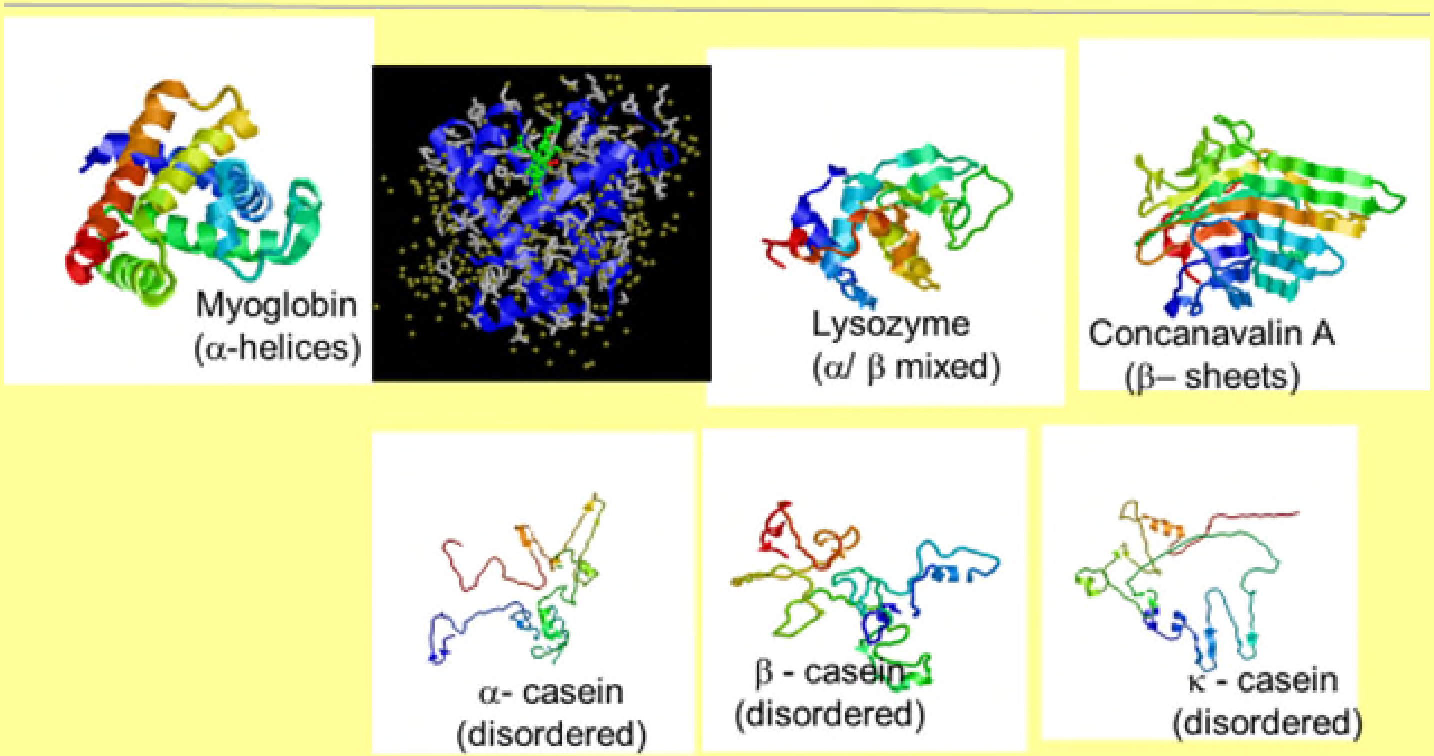
Two components:Torsion
local diffusion

Dynamical transition at room temperature

Protein Solutions

Are protein motions structure dependent?

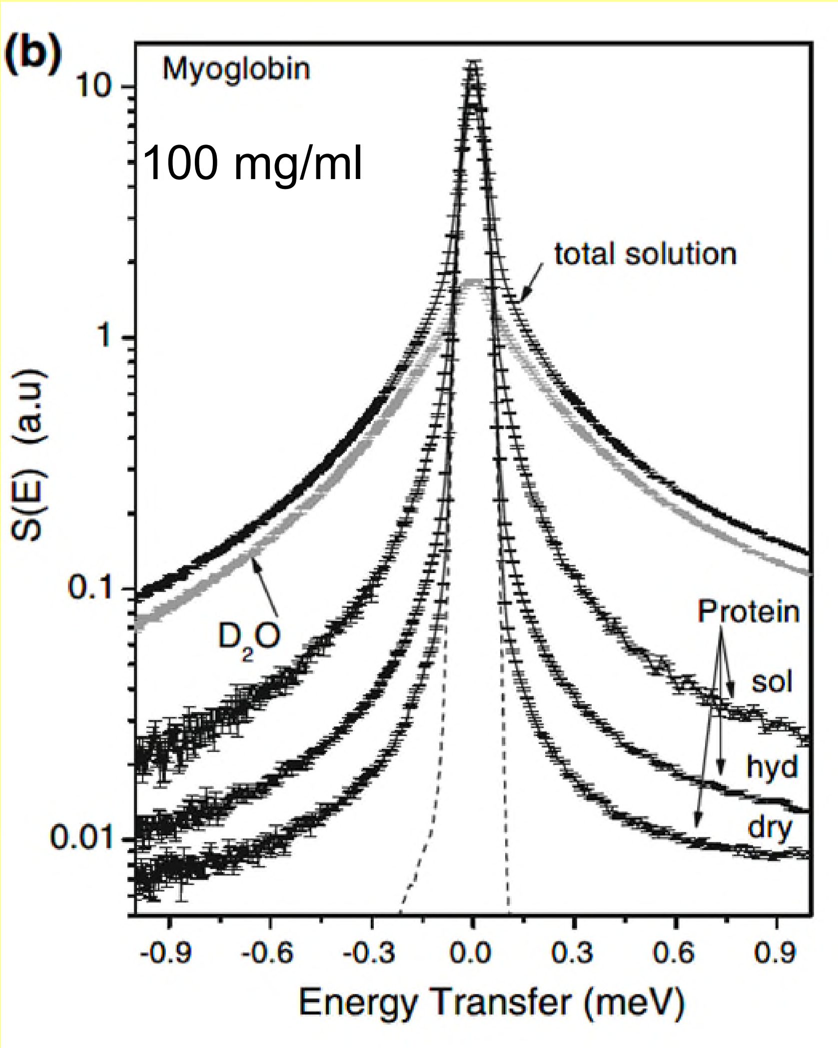
Proteins with different secondary structure

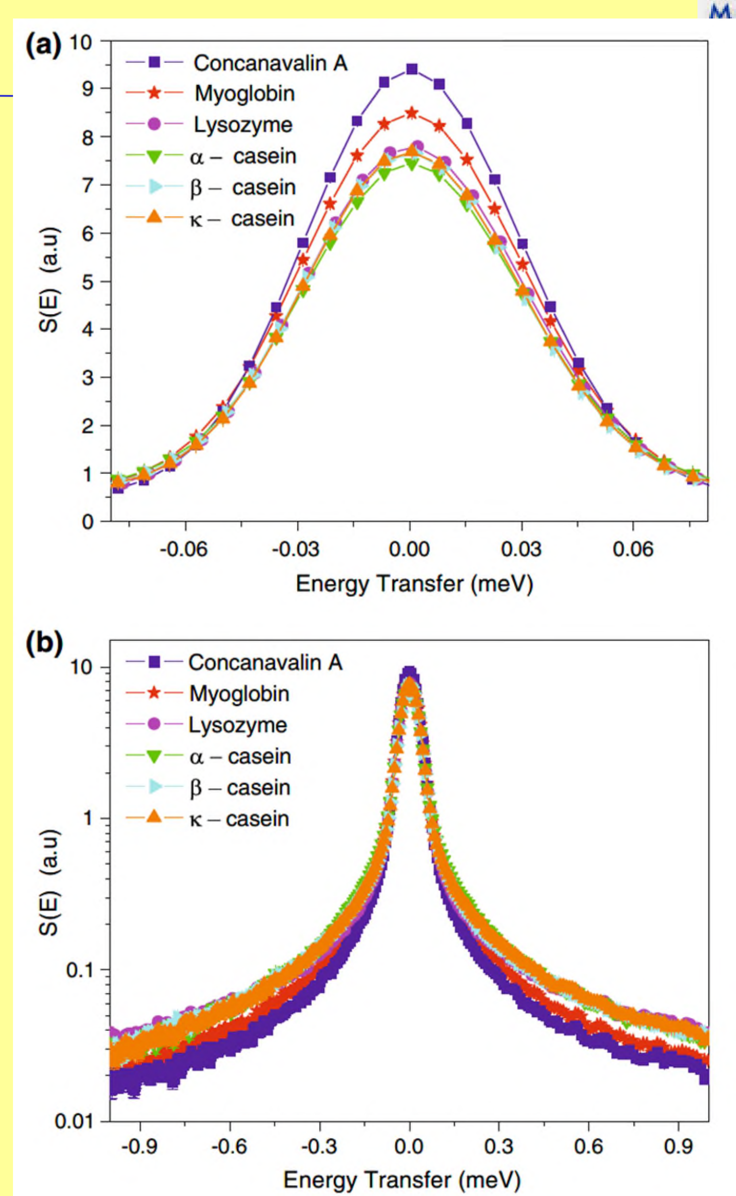


TOFTOF spectra of protein solutions

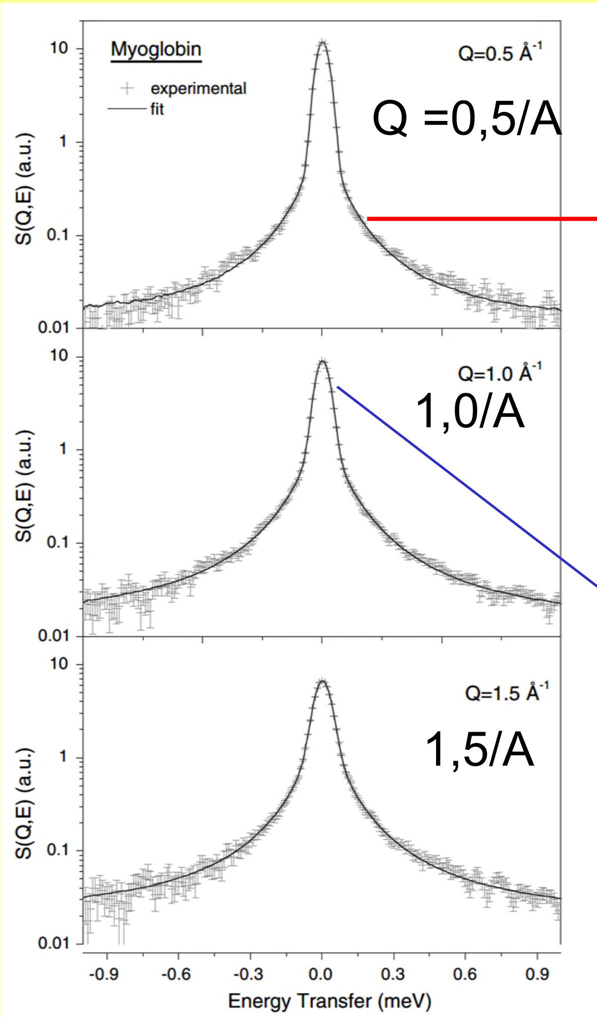
Anna Gaspar et al. EBJ 2008

log scale



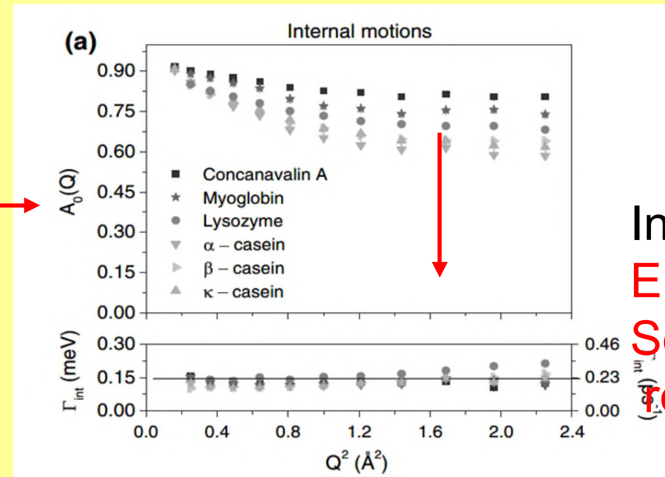


2-component analysis:
(1) internal motion and (2) global diffusion



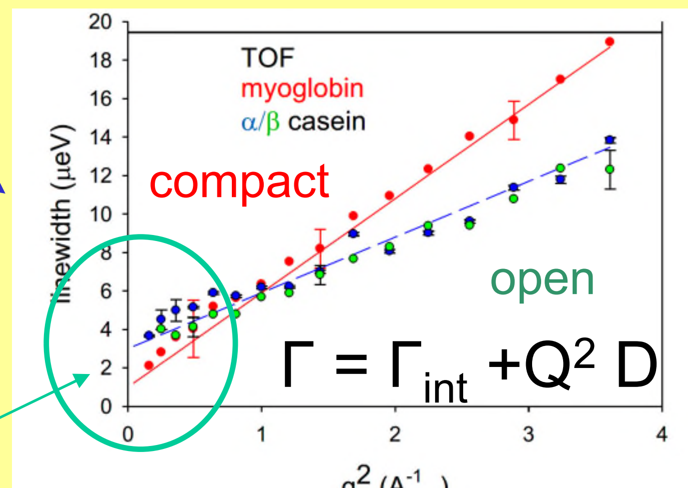
internal

Global
Diffusion



EISF(Q)

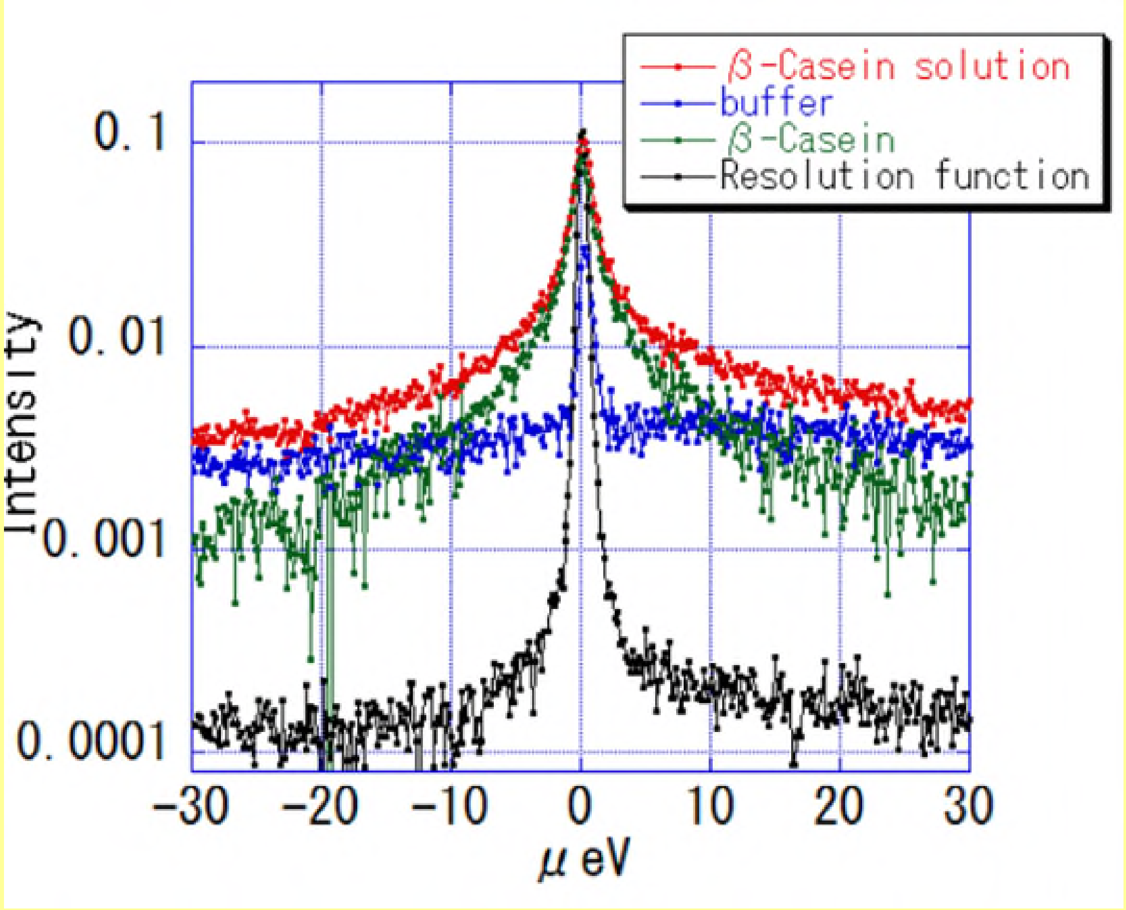
Internal dynamics
EISF depends on
Secondary structure
relaxation times: 5 ps



Offset?

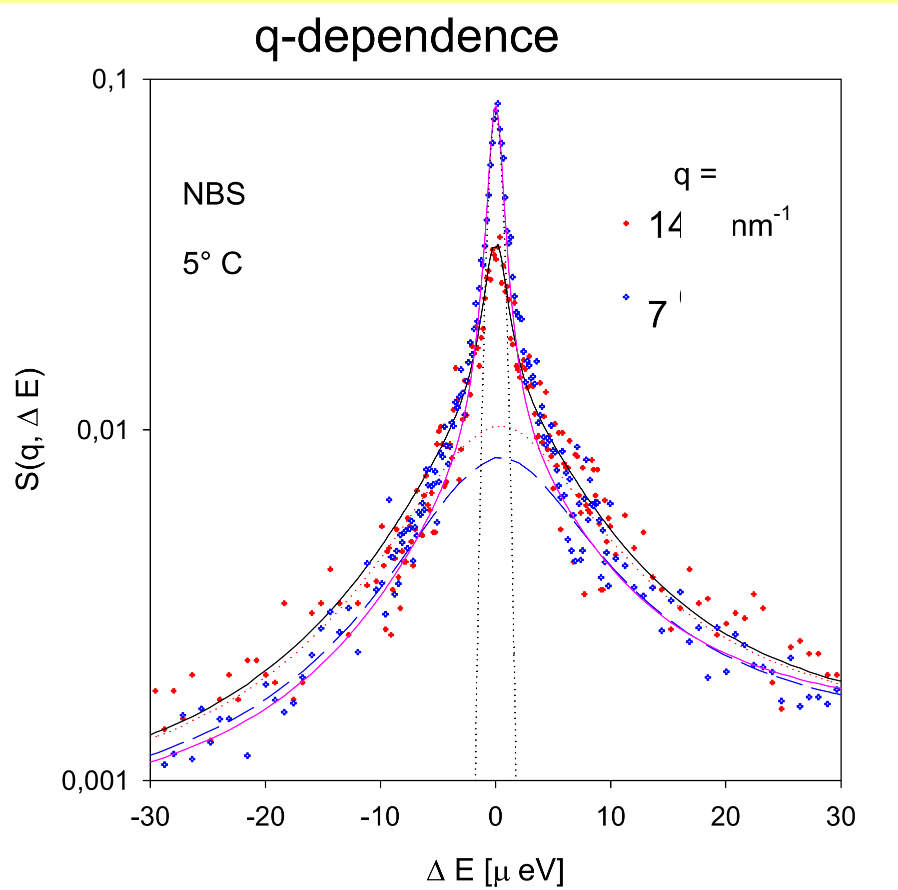
Global diffusion
Offset Q-> 0
casein:
New process?
Resolution?

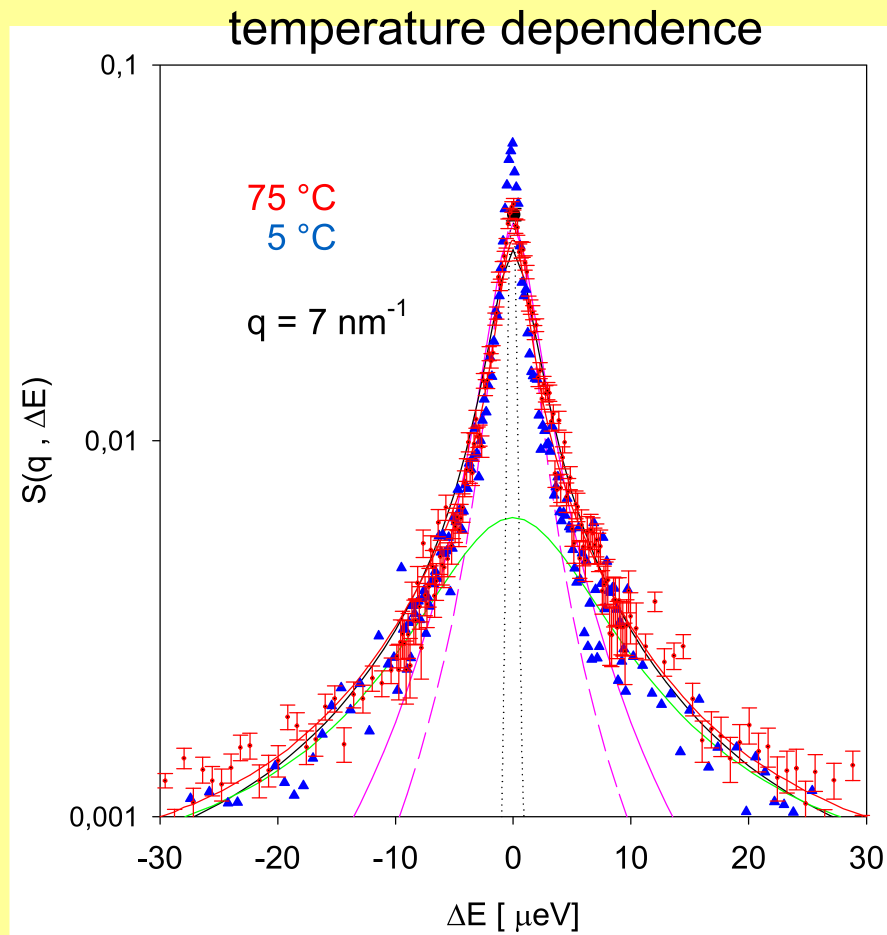
Backscattering Spectroscopy: SPHERES
β-casein solutions Nakagawa et al. Biophys.J 2022



$$S_{\beta\text{-casein}}(q, E) = S_{solution}(q,E) - \alpha\, S_{buffer}(q, E)$$

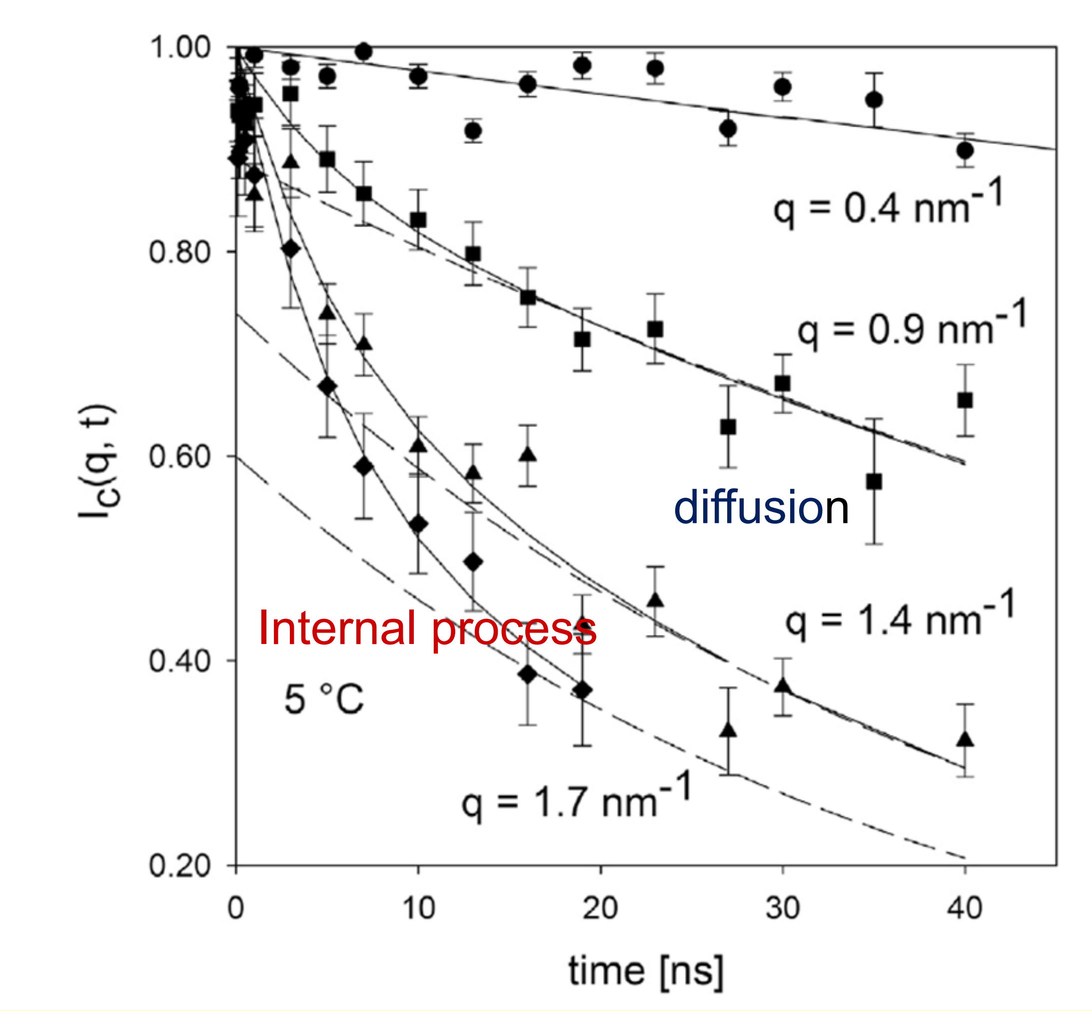
Backscattering SPHERES
β-Casein in Solution



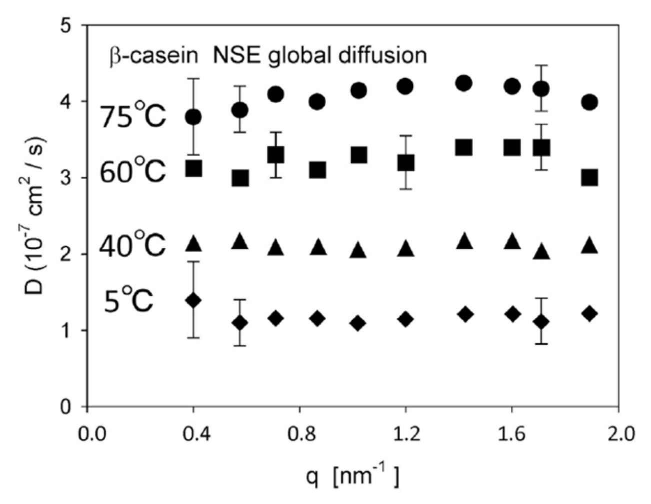


$$S_{inc}(q,\omega) = A_0(q)\cdot S_{res}(q,\omega) \otimes \left[\frac{1}{\pi}\frac{q^2 D}{\omega^2 + \left(q^2 D\right)^2}\right] \otimes \left[\sum_i (1 - A_i)\delta(\omega) + \frac{A_i}{\pi}\frac{\tau_i}{1+\omega^2\tau_i^2}\right] + B(q,\omega)$$

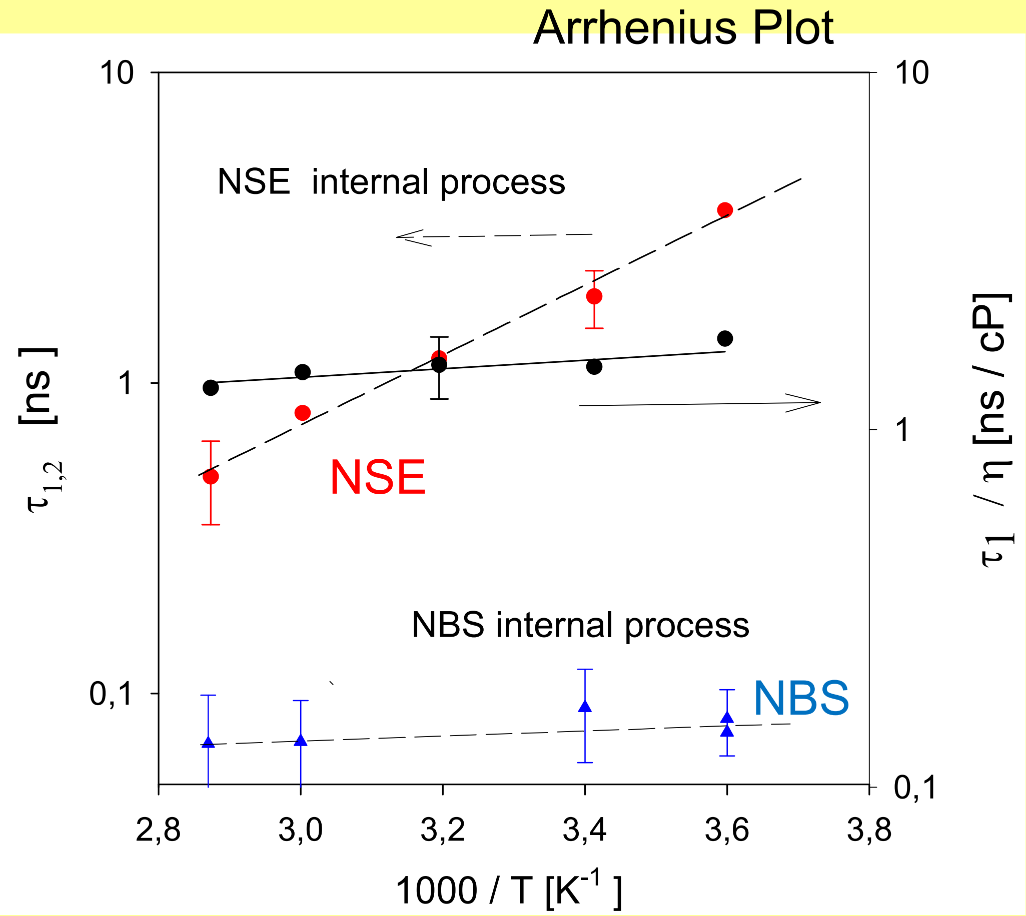
NSE study of βcasein solution: diffusion + internal process (3)



Diffusion coefficient is q-independent



β-casein solution: NSE+NBS: two internal processes



Chain motion
Viscosity effect

local
residue diffusion

Fourier transforms of averaged TOF and NBS spectra put together

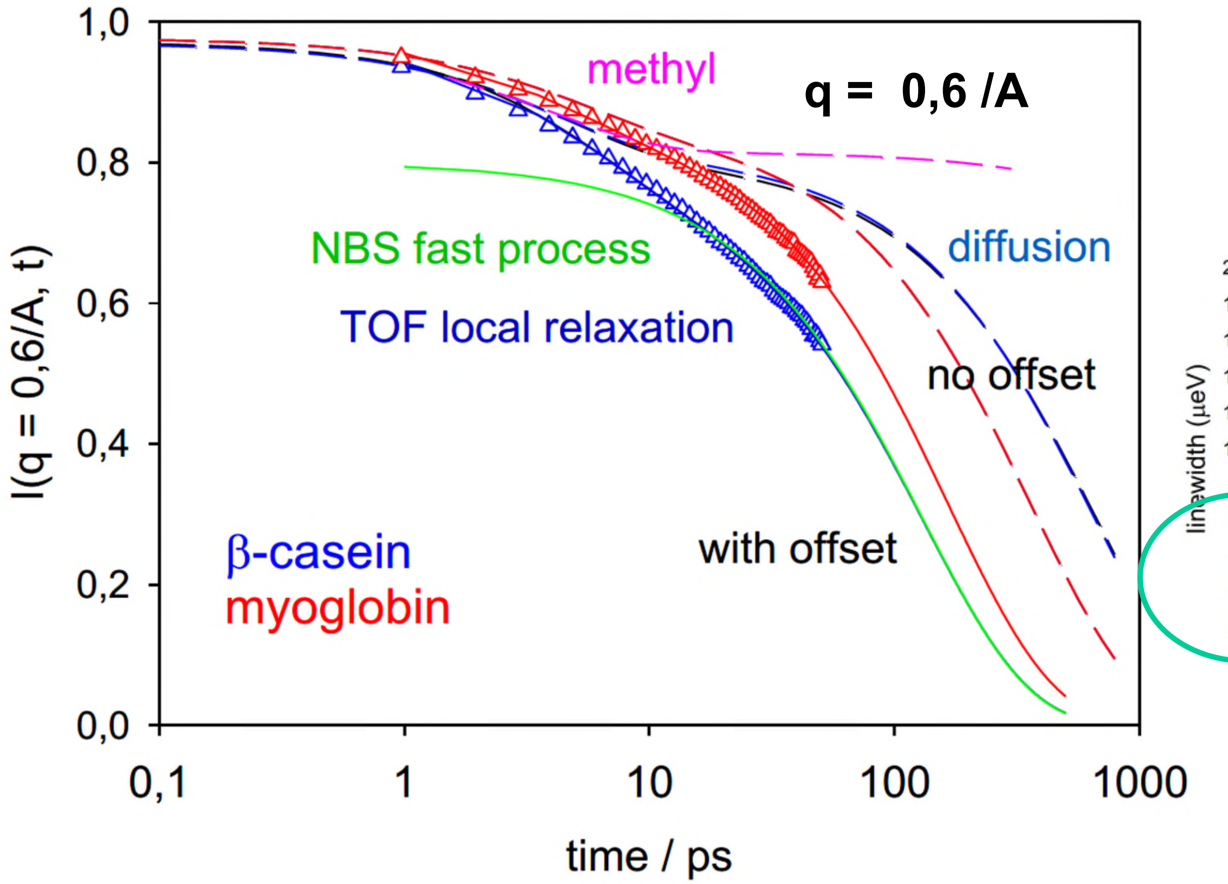
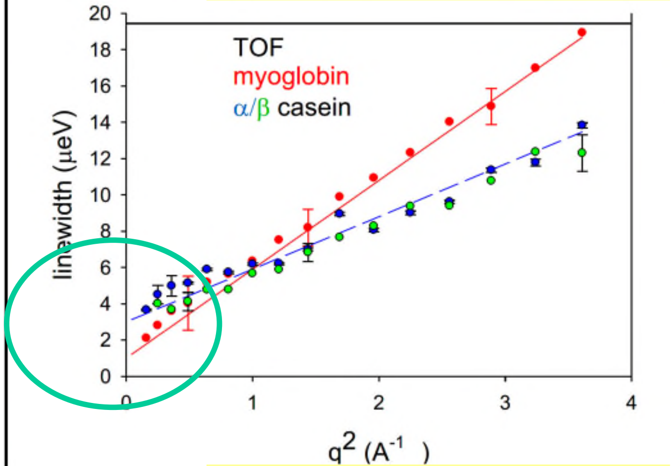


Fig. 16: incoherent intermediate scattering functions of myoglobin and β-casein (TOF and NBS)

Offset?



TOF offset agrees with process of backscattering experiment

Parameters of two component analysis

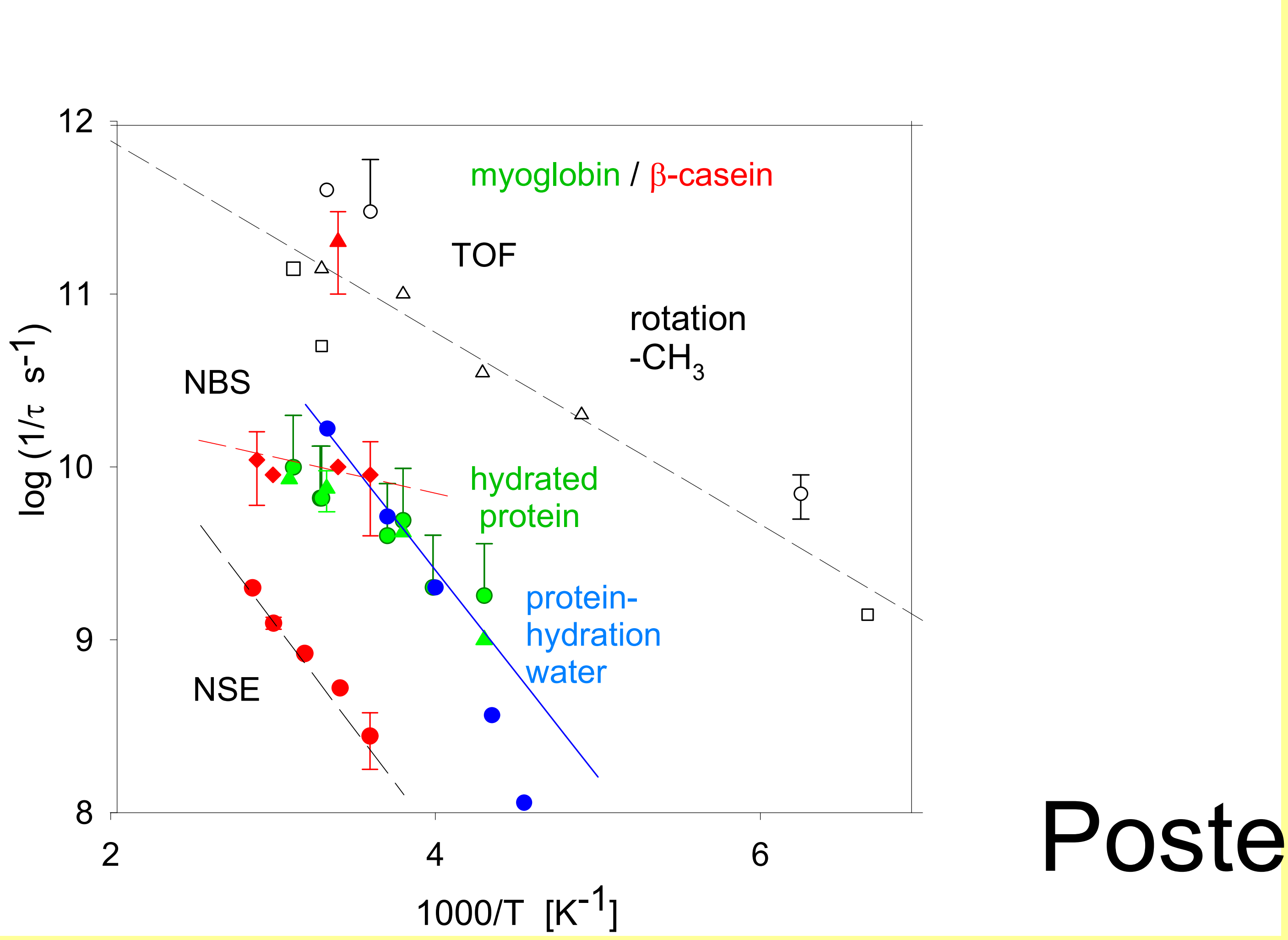
		q	A_{met}	τ_{met} ps	$\tau_{diff}(q = 0{,}6\ A^{-1})$ ps
β-casein	TOF	0,6 (1/A)	0,19	5,2 (± 0,2)	130 (± 10)
	NBS	fast process			130
myoglobin	TOF	0,6	0,11	5,2	165 (± 20)
	NBS	fast process			150

Internal fast process TOF

internal slow TOF/NBS

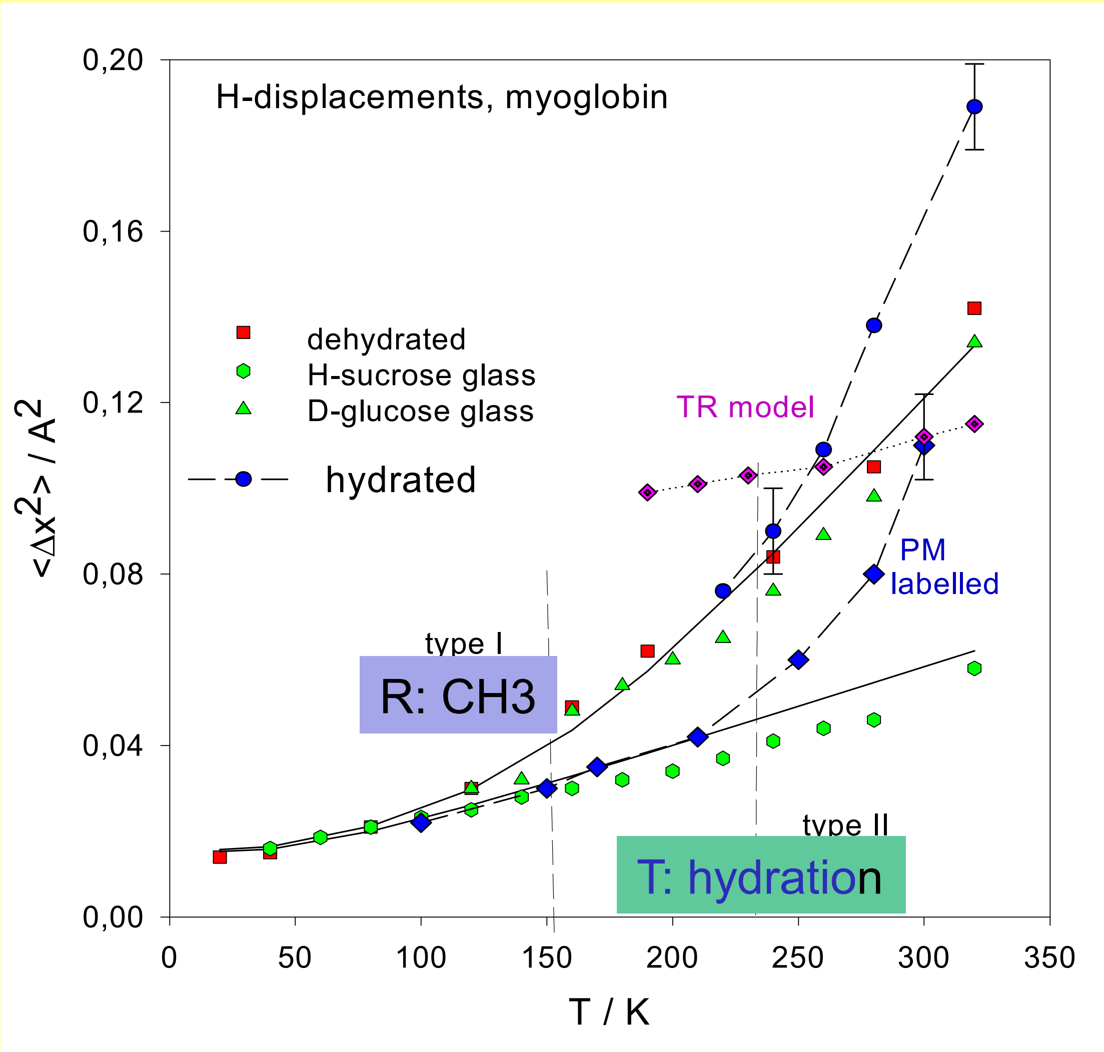
Arrhenius Plot of Protein Rates hydrated and solvated

Chain Diffusion, Residue Diffusion , Rotational Transitions



Poster

Mean Square Displacements
Two transitions R, T: resolution effect, CH3 rotation also in the glass



D-hydrated / CH3

dehydrated

D-Glass

- TR Model: no transition

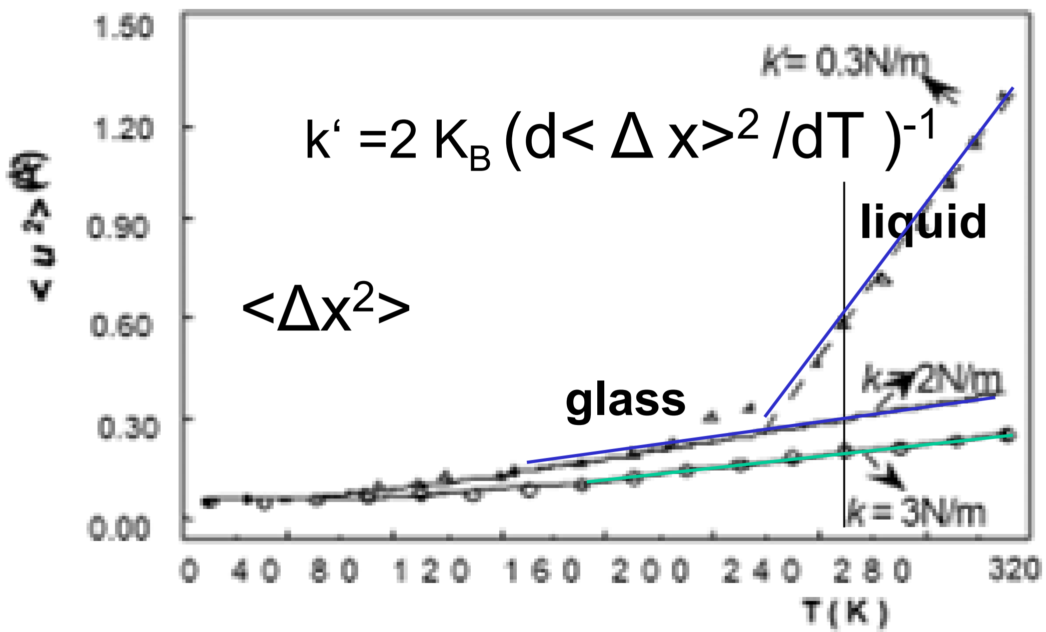
D-hydrated, CD3

H- Glass

Dynamic Softening of Protein Force Constants from fixed resolution elastic scans

J. Zaccai, Science 2000 to QENS 2022
Walter Hälg prize 2013

- **Hydrogen mean square displacements ,**



Transition of Protein Resilence in hydrated but not in glassy matrix

Myoglobin in D_2O-hydrated powder
k' = 2 N/m → 0,3 N/m
240 K

Myoglobin **in D_2O exchanged trehalose glass**
k' = 3 N/m

IN 13, 8μ eV,
100 ps

Using polarization analysis to separate the coherent and incoherent scattering from protein samples

Ana M. Gaspar [a,b,*], Sebastian Busch [a,b], Marie-Sousai Appavou [a,c], Wolfgang Haeussler [b,d], Robert Georgii [b,d], Yixi Su [c], Wolfgang Doster [a]

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BBA 1804 (2010) 76