Dynamics of Hydration Water by QENS

Emiliano Fratini

fratini@csgi.unifi.it

Why Neutrons?

The typical wavelengths of the neutron in a beam range from ~10 Å to ~1 Å.

- comparable to interatomic and intermolecular distances

slower neutrons → longer wavelengths → longer length scales

The typical energies of a neutron beam range from ~1 meV up to ~100 meV.

- comparable to the time scale of many motions in materials

=> inelastic scattering from vibrations, diffusion, reorientations, and relaxational processes can be observed

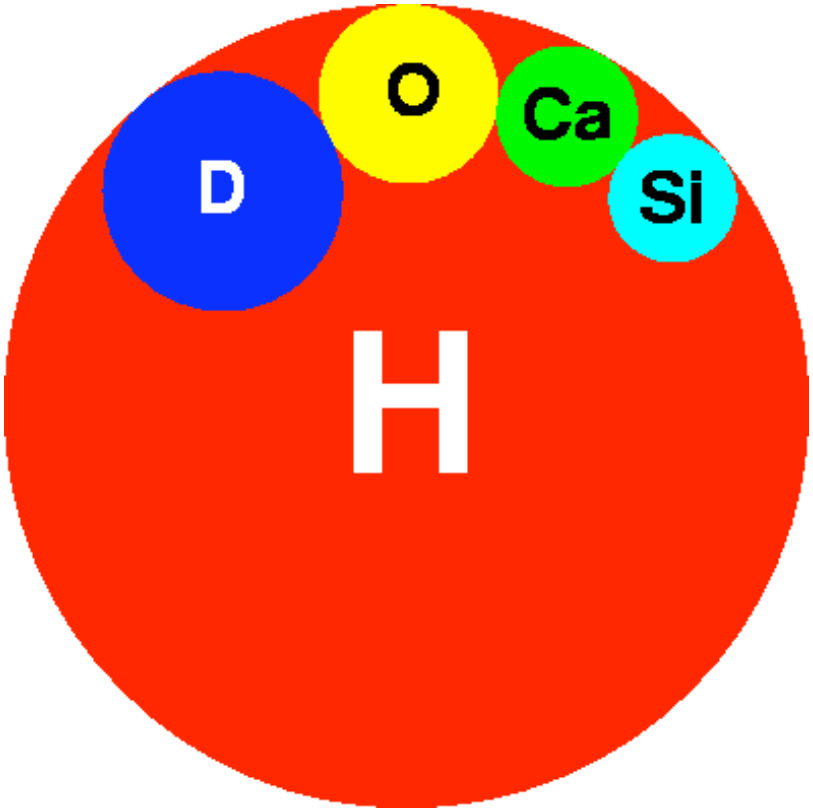
- light E ~ eV's λ ~ 1000 Å's (Q ~ 0)
- x-rays E ~ keV's λ ~ Å's

slower neutrons → lower energies → longer time scales

1 meV @ 8 cm^{-1} @ 240 GHz @ 12 K @ 0.1 kJ/mol ~ 1 ps

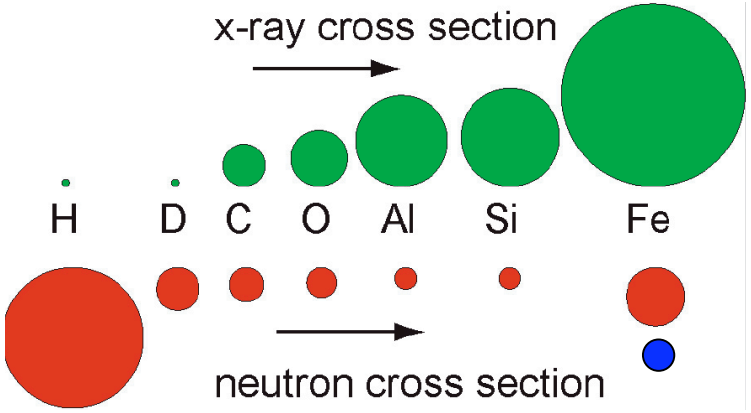
Scattering cross-sections

Scattering power varies "randomly" from isotope to isotope.



Cross section (s) - Area related to the probability that a neutron will interact with a nucleus in a particular way (*e.g.* scattering or absorption)

For a single nucleus s ~ 10^{-24} cm^2



D $\sigma_{coh} = 5.59\ barn$ $\sigma_{inc} = 2.504\ barn$

H $\sigma_{coh} = 1.76\ barn$ $\sigma_{inc} = 80.26\ barn$

Nuclear Interaction

Scattering power varies "randomly" from isotope to isotope.

The scattering also depends on nuclear spin state of the atom.

- If the scattered neutron waves from the different nuclei have definite relative phases, they do interfere

Coherent Scattering

- If the scattered neutron waves from the different nuclei have RANDOM relative phases, they don't interfere

Incoherent Scattering

Dynamic Structure Factor

$$S(Q,\omega) = S_{inc}(Q,\omega) + S_{coh}(Q,\omega)$$

$S_{coh}(Q,\omega)$ is the time and space Fourier transform of the ***PAIR*** correlation function (Collective Particle Dynamics)

$$S_{coh}(Q,\omega) = FT\left\{\left\langle \exp\left[-iQ\left(\underline{r}_i(t) - \underline{r}_j(0)\right)\right]\right\rangle\right\}$$

$S_{inc}(Q, \omega)$ is the time and space Fourier transform of the ***SELF*** correlation function (Single Particle Dynamics)

$$S_{inc}(Q,\omega) = FT\left\{\left\langle \exp\left[-iQ\left(\underline{r}_i(t) - \underline{r}_i(0)\right)\right]\right\rangle\right\}$$

Dynamic Structure Factor

$$g(r,t) = \frac{1}{N}\sum_{k,j}^{N}\langle\delta\{r + r_k(0) - r_j(t)\}\rangle.$$

Space Fourier Trasform

$$I(Q,t) = \frac{1}{2\pi}\int_{-\infty}^{+\infty} g(r,t)\exp\{-\mathrm{i}Q\cdot r\}\,\mathrm{d}r = \frac{1}{N}\sum_{k,j}\langle \mathrm{e}^{\mathrm{i}Q\cdot r_k(t)}\mathrm{e}^{-\mathrm{i}Q\cdot r_j(0)}\rangle$$

Time Fourier Trasform

$$S(Q,\omega) = \frac{1}{2\pi}\int_{-\infty}^{+\infty} I(Q,t)\exp\{-\mathrm{i}\omega t\}\,\mathrm{d}t$$

Incoherent Scattering

- Inelastic/Quasielastic Scattering
  - Single Particle Dynamics
    - Translations
    - Rotations
    - Vibrations

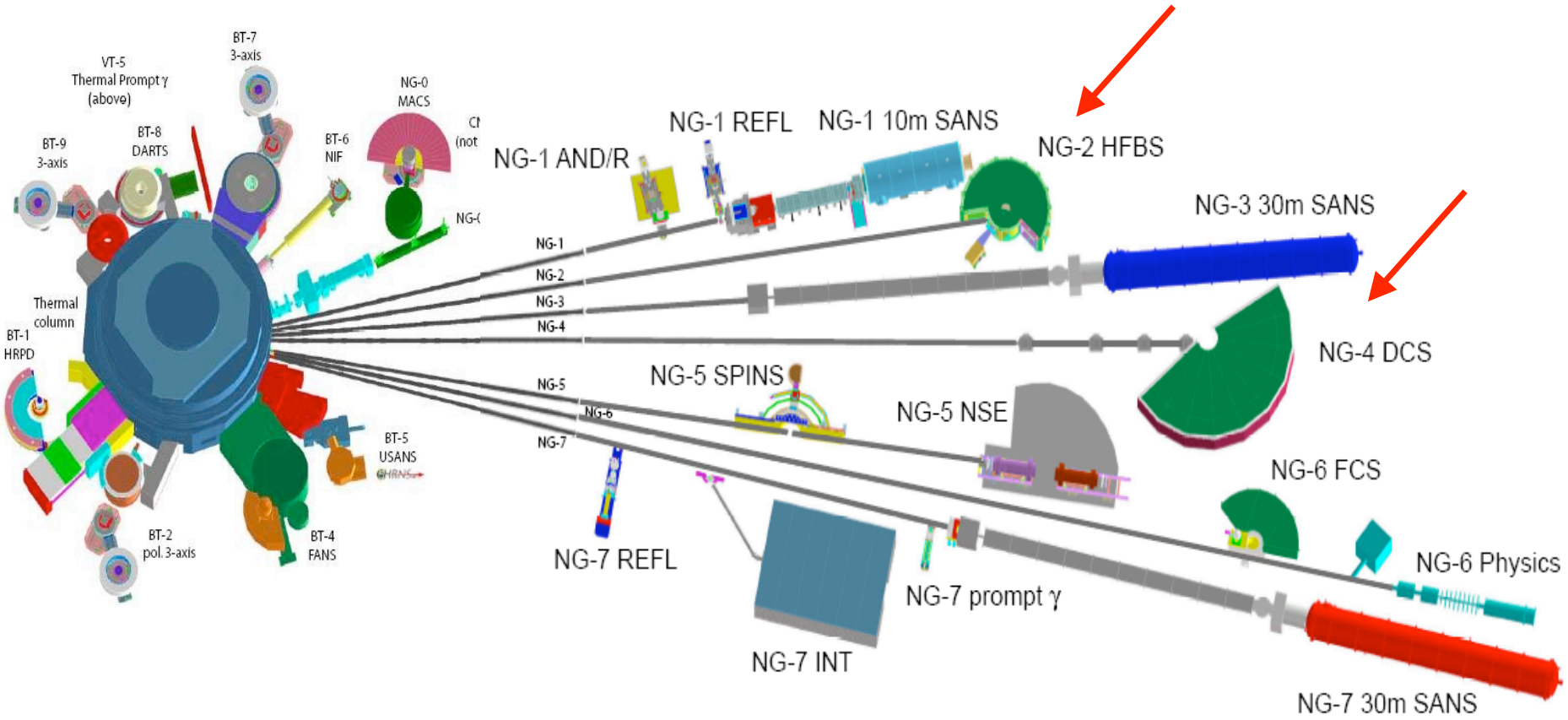
Protein Dynamics

• A protein is a nano-machine whose molecular structure was selected by evolution to perform specific biological functions.

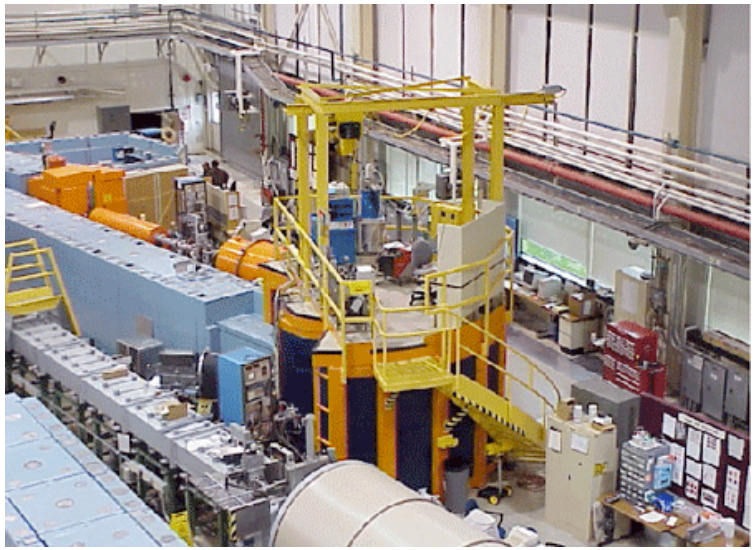
• Proteins perform these biological functions thanks to their flexibility.

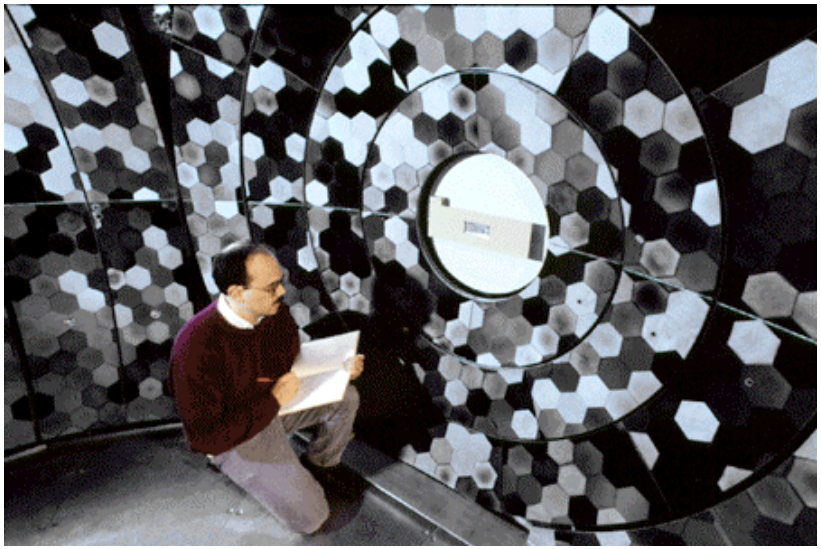
• Protein Dynamics extends from fs motions of side chains to multi-ms fluctuations on the length scale of clusters of interacting proteins and from the Angstroms scale up to tens of nm.

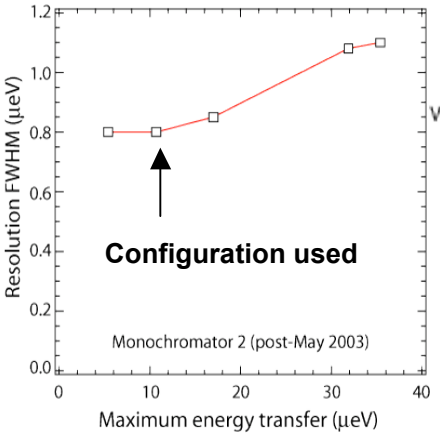
NIST Center for Neutron Research
picoseconds/nanoseconds dynamics

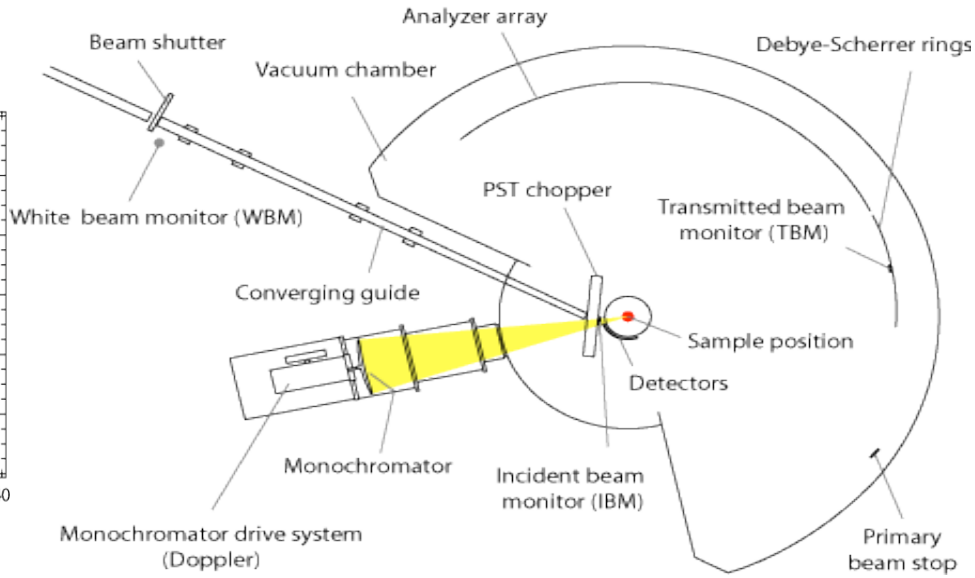


High-Flux Backscattering Spectrometer









Energy resolution: 0.8 μeV

Dynamic range: 11 μeV

Time range covered

60 ps to 20 ns

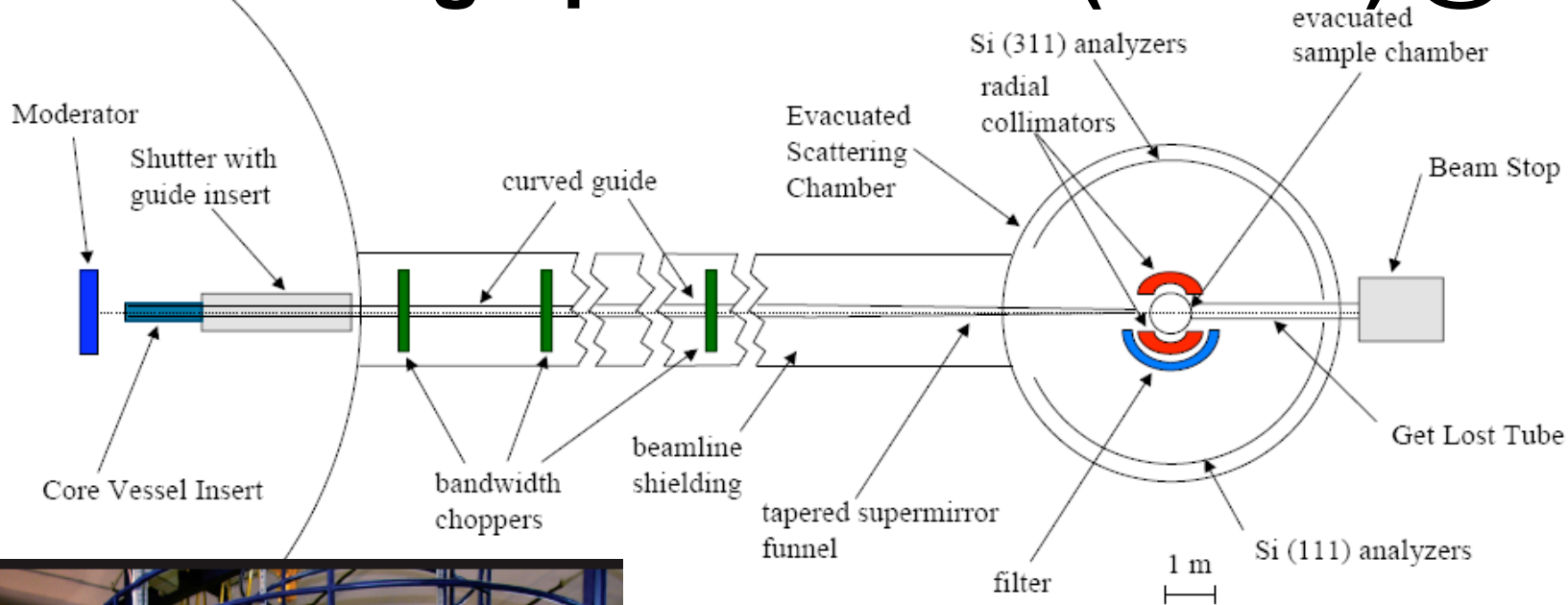
Incident energy:

2.08 meV (λ_0=6.271 Å)

Q range: 0.25 Å^{-1} ~ 1.75 Å^{-1}

Typical run time: 6 ~ 8 h

Backscattering Spectrometer (BASIS) @ SNS



Si 111	
Elastic energy	2.08 meV
Bandwidth	±258 μeV
Resolution (elastic)	3 μeV
Q range (elastic)	0.17 Å^{-1} < Q < 2.0 Å^{-1}
Solid angle	2.0 sr 4.0 sr (upgrade)

Si 311 (upgrade)	
Elastic energy	7.64 meV
Bandwidth	±1700 μeV
Resolution (elastic)	10 μeV
Q range (elastic)	0.35 Å^{-1} < Q < 3.8 Å^{-1}
Solid angle	4.0 sr

Sample Preparation: Example

- Protein has to be D-exchanged by dissolving ***1 g of protein in 10 g of D_2O***. After allowing hydrogen exchange to occur overnight at room temperature, the sample was freeze-dried into a powder. (then check by neutron prompt gamma-ray!!!)

- After lyophilization, hydration was achieved by of the deuterium-exchanged protein with saturated solutions of LiCl, NaCl, and K_2SO_4 in D_2O which resulted in hydration levels of h: 0.05, 0.18, and 0.30. Samples with h= 0.45, 0.50, and 0.80 were prepared by adding D_2O to the 0.30 h sample and equilibrating the powders for at least 12 hours.

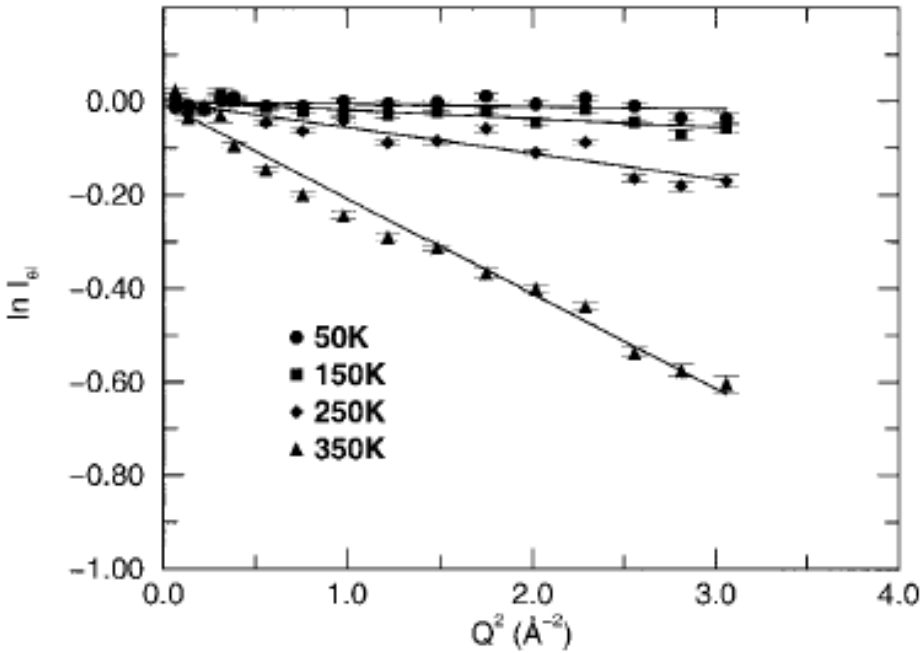
0.3 h = 0.3 g of D_2O/1 g of dry protein = 1 monolayer

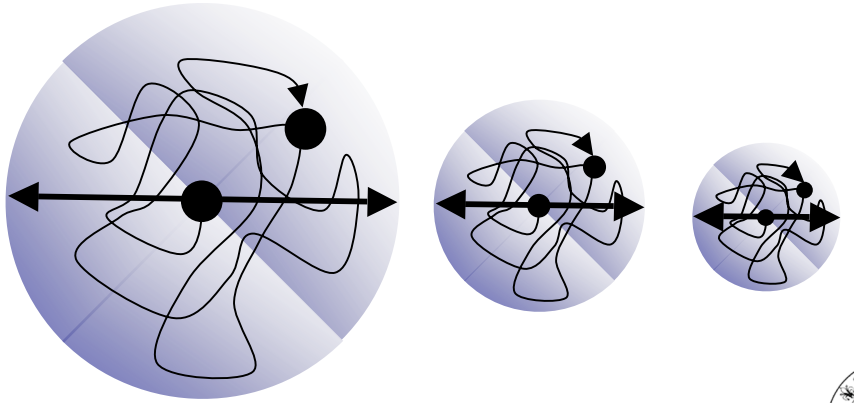
Elastic scans: EINS

In the Gaussian approximation the MSD $\langle x^2 \rangle$ values are calculated from the angular dependence of the incoherent scattered elastic intensity (within the instrumental resolution).

$$I(Q,0\pm\Delta\omega) = (\text{constant})[\exp(-\langle \bar{x}^2 \rangle Q^2)]$$

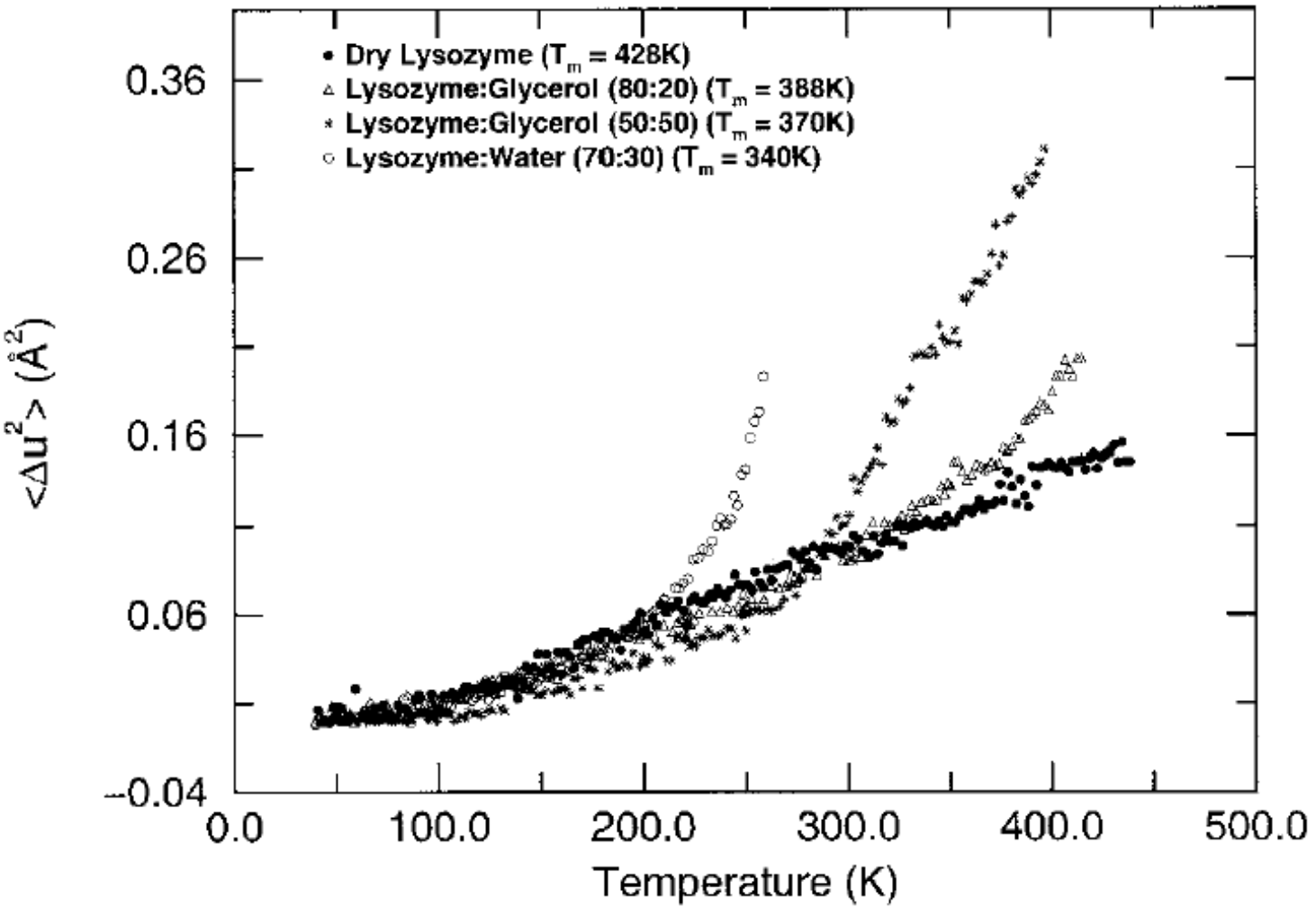
A.M. Tsai, et al., Biophysical J., 79, 2728 (2000).





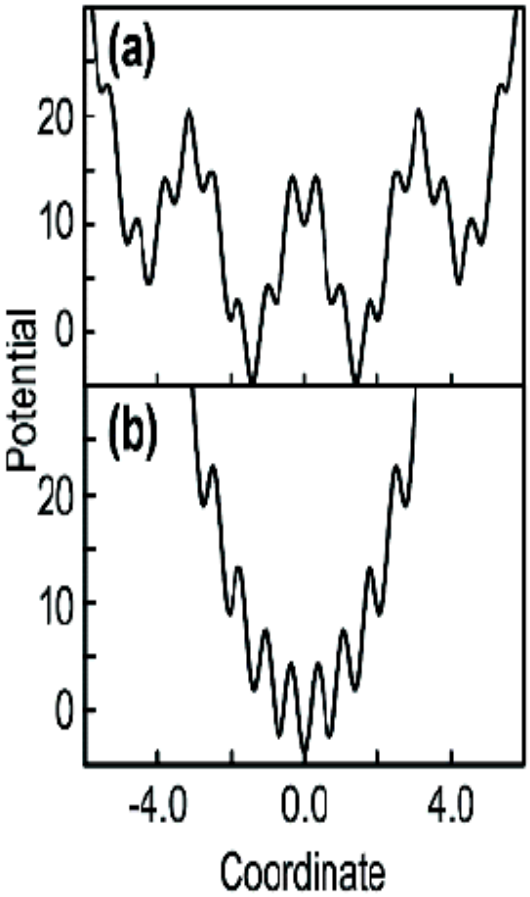
u^2

Effect of glycerol



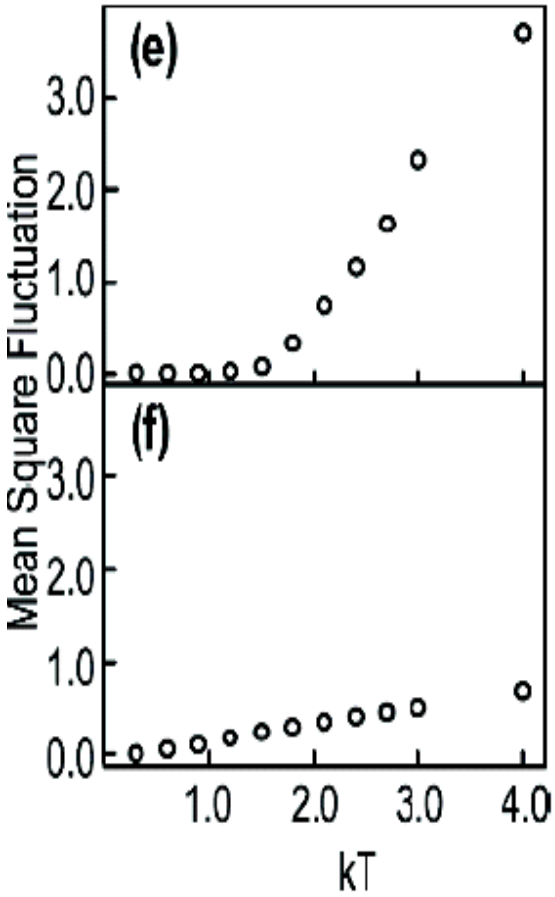
A.M. Tsai, et al., Biophysical J., 79, 2728 (2000).

Conformational Substates (k_BT)



Hydrated Protein

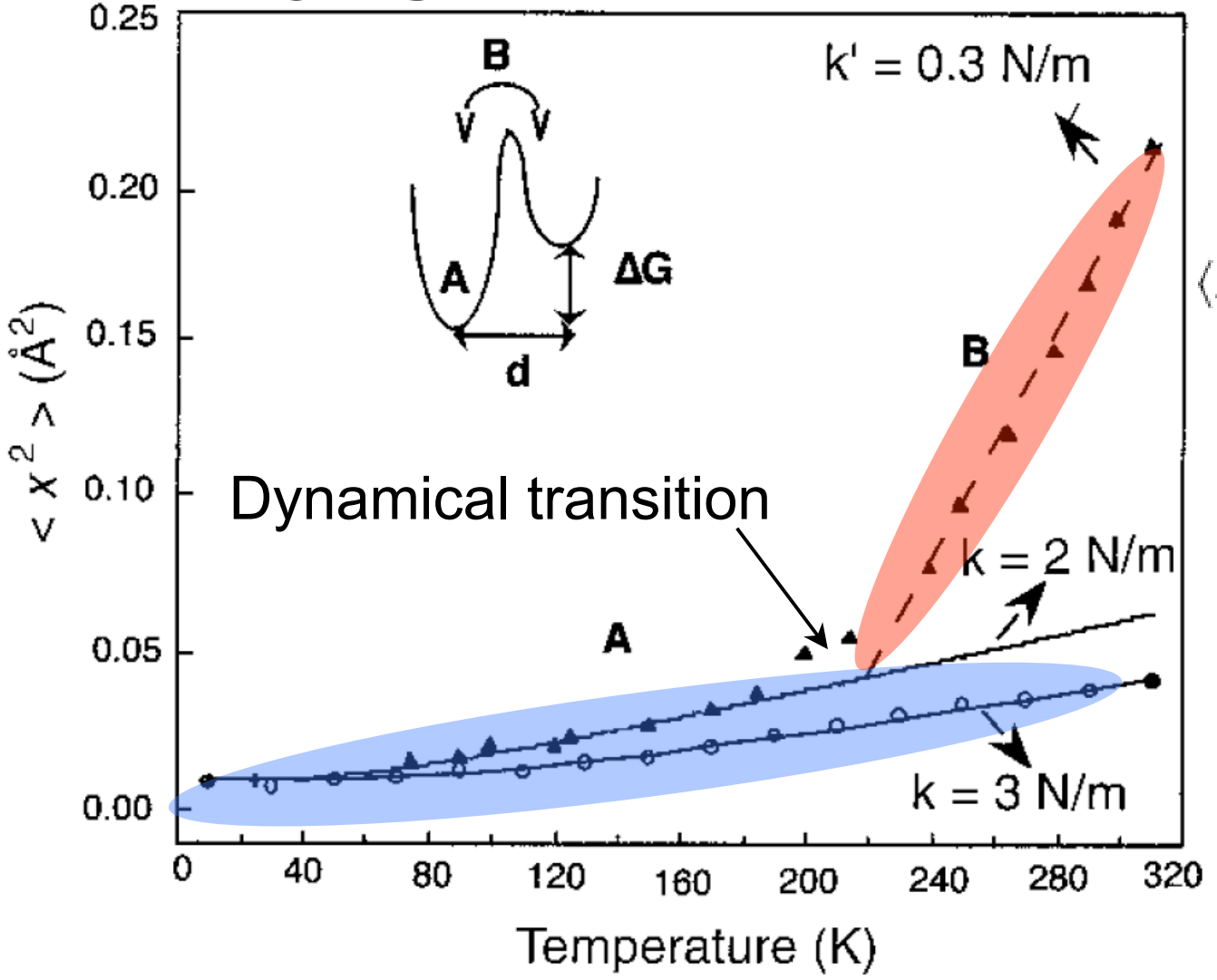
Dry Protein



Model one-dimensional (a) multiply hierarchical and (b) singly hierarchical energy surfaces, and results of Brownian dynamics simulation using these surfaces.Temperature dependences of mean-square fluctuations for (a) and (b) are shown in (e) and (f), respectively.
Yasumasa Joti, Akio Kitao, and Nobuhiro Go *JACS 127, 8705 (2005).*

Myoglobin

G. Zaccai, Science, 288 1604–7 (2000)



Mean environmental constant force:

$$\langle k \rangle = 0.00138/(d\langle 3x^2 \rangle/dT)$$

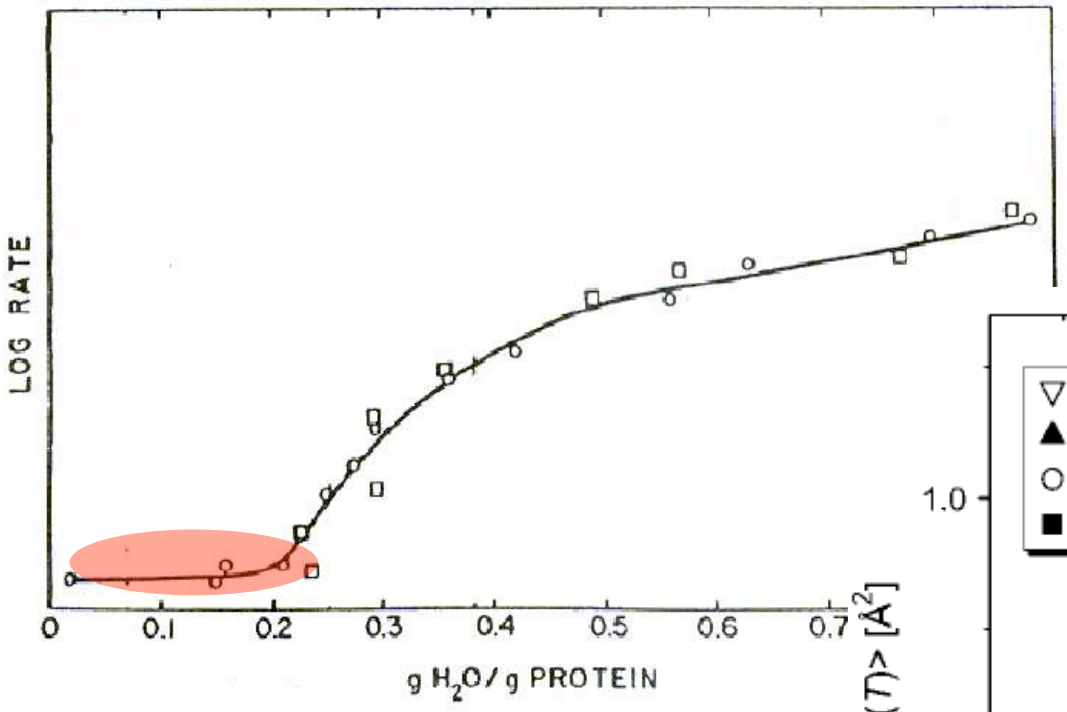
(N/m if *x* is in Å and T in K)

Trehalose: stiffens the protein, inhibits the transition and has a protective effect.

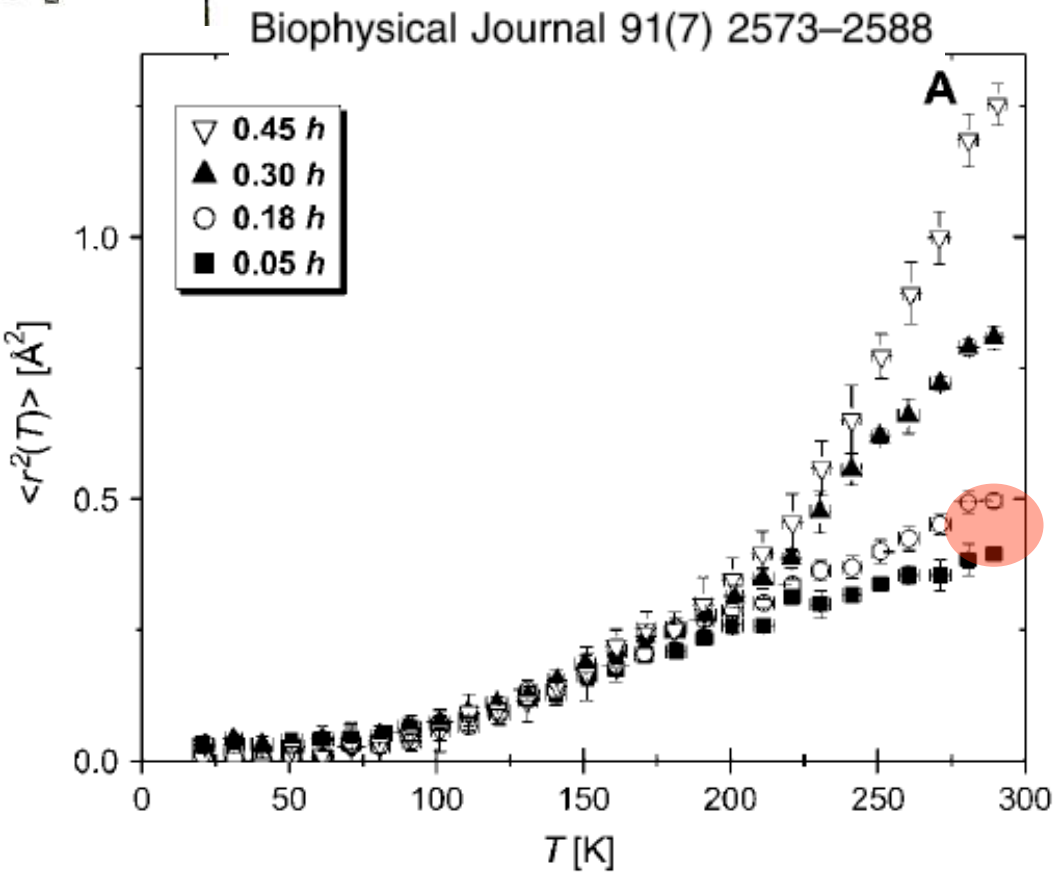
▲ In D_2O [W. Doster, S. Cusack, W. Petry, Nature 337, 754 (1989).]

O In Trehalose [L. Cordone, et al., Biophys. J. 76, 1043 (1999).]

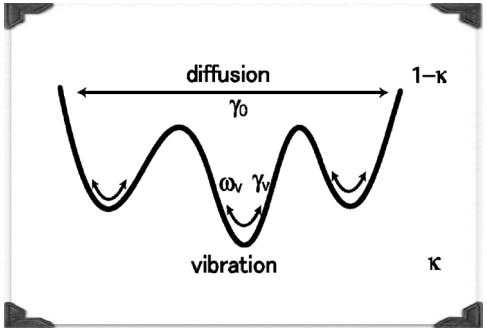
Protein Dynamics vs h

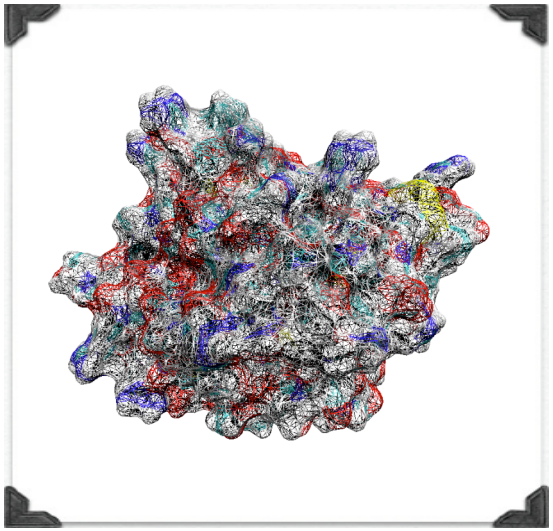


Rupley J.A., P.Yang and G.Tollin. "Water in Polymers" 1980 S.P.Rowland, Washington D.C.



Protein Dynamics





Biological activity

proper mobility of backbone and side-chains

Glass Transition

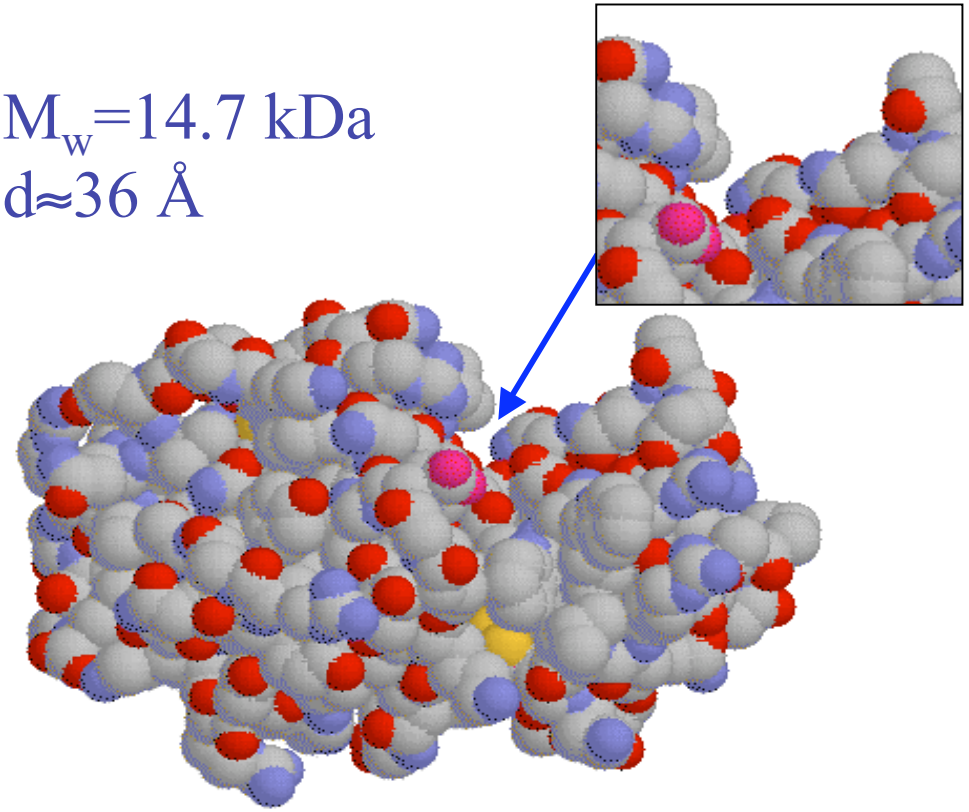
harmonic motion around equilibrium positions

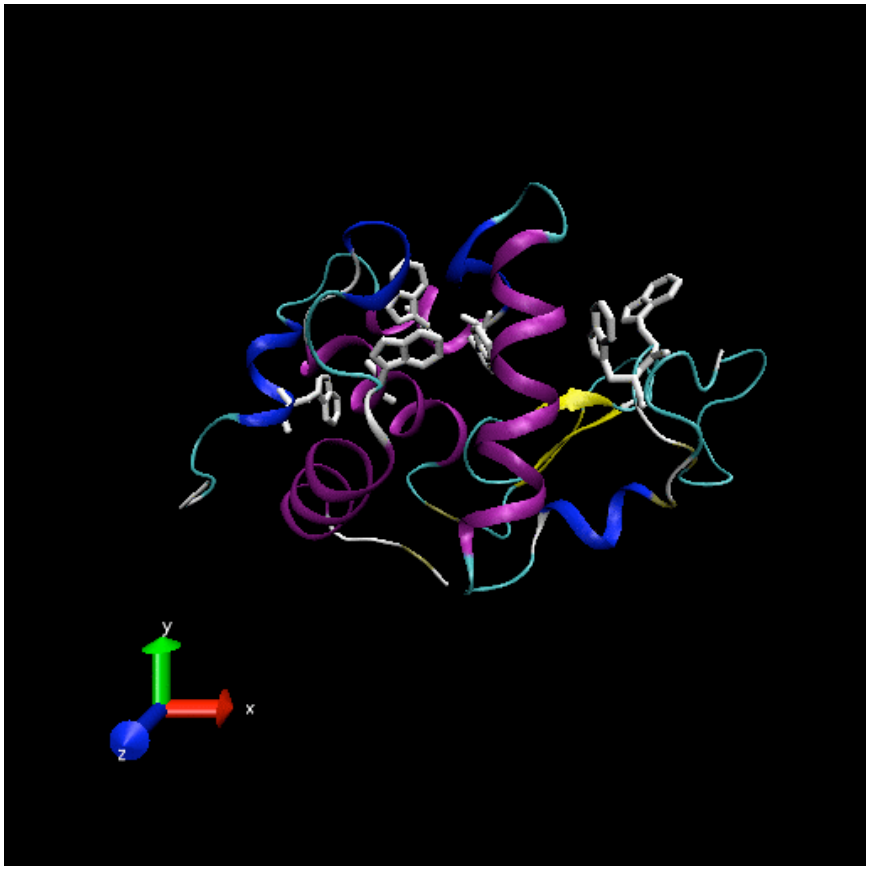
Denaturation

loss of tertiary and secondary structure

Hen-egg white Lysozyme

M_w=14.7 kDa
d≈36 Å

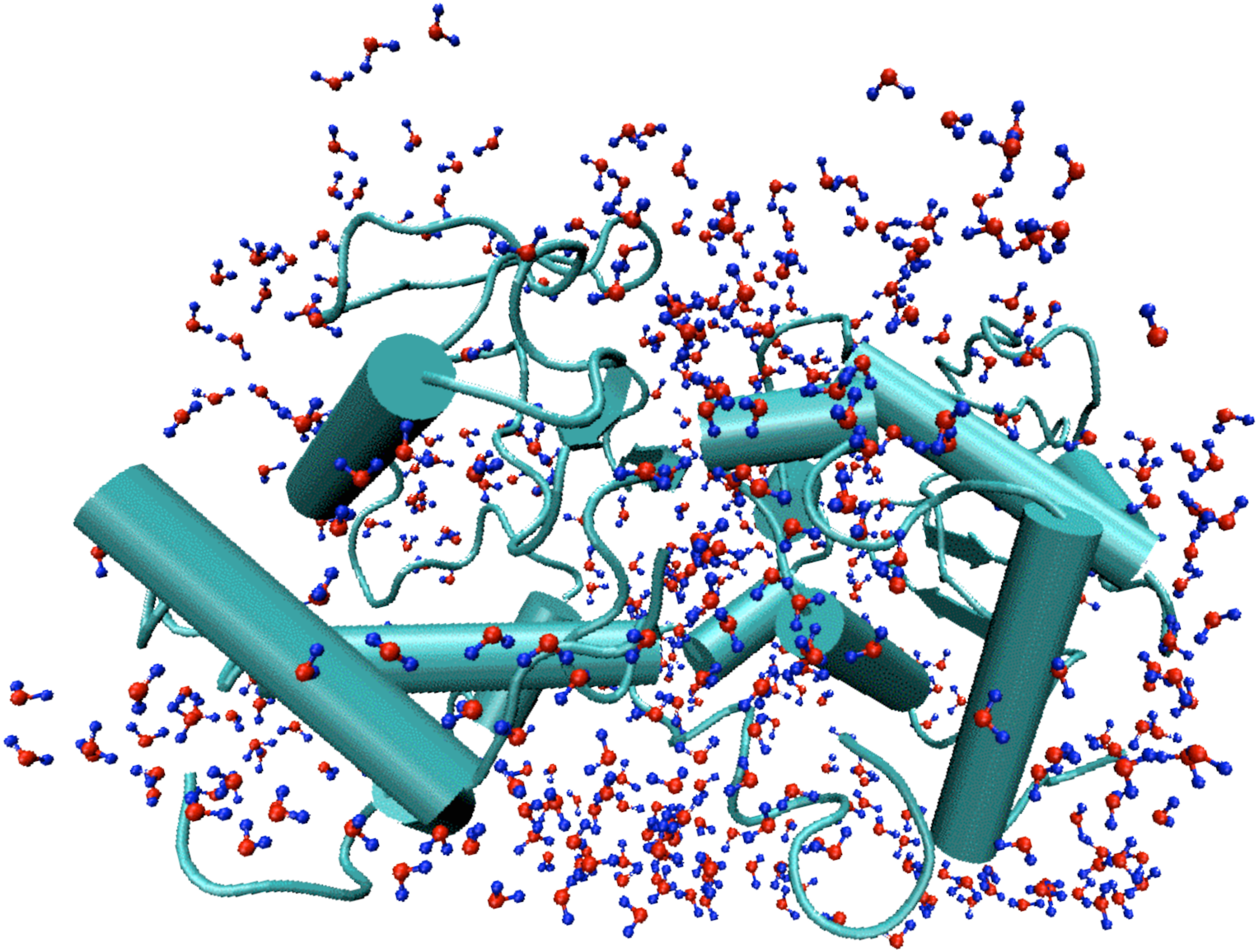




Lysozyme is an **enzyme** consisting of 129 aminoacid residues which folds into a compact globular structure having an ellipsoidal shape with dimensions a x b x b = 2.25 x 1.5 x1 5 nm^3. Molecular weight of Lysozyme is approximately 14.4 kDa. Lysozyme has an enzymatic cleft where the α-domain meets the β-domain. It is found in the mucosal secretions and the white of avian eggs. ***It hydrolyzes the polysaccharides found in many bacterial cell walls.*** This catalytic activity, which is a non-specific organism defence, is only effective against ***Gram-positive*** bacterial cells. In viruses (or bacteriophages), lysozyme is used as an agent to break into the host bacterial cell and lately to release new viruses.

Model of Hydrated Lysozyme Powder

Water: 484 TIP4P-EW molecules (h = 0.3); Protein: 2 lysozyme, OPLS-AA force field

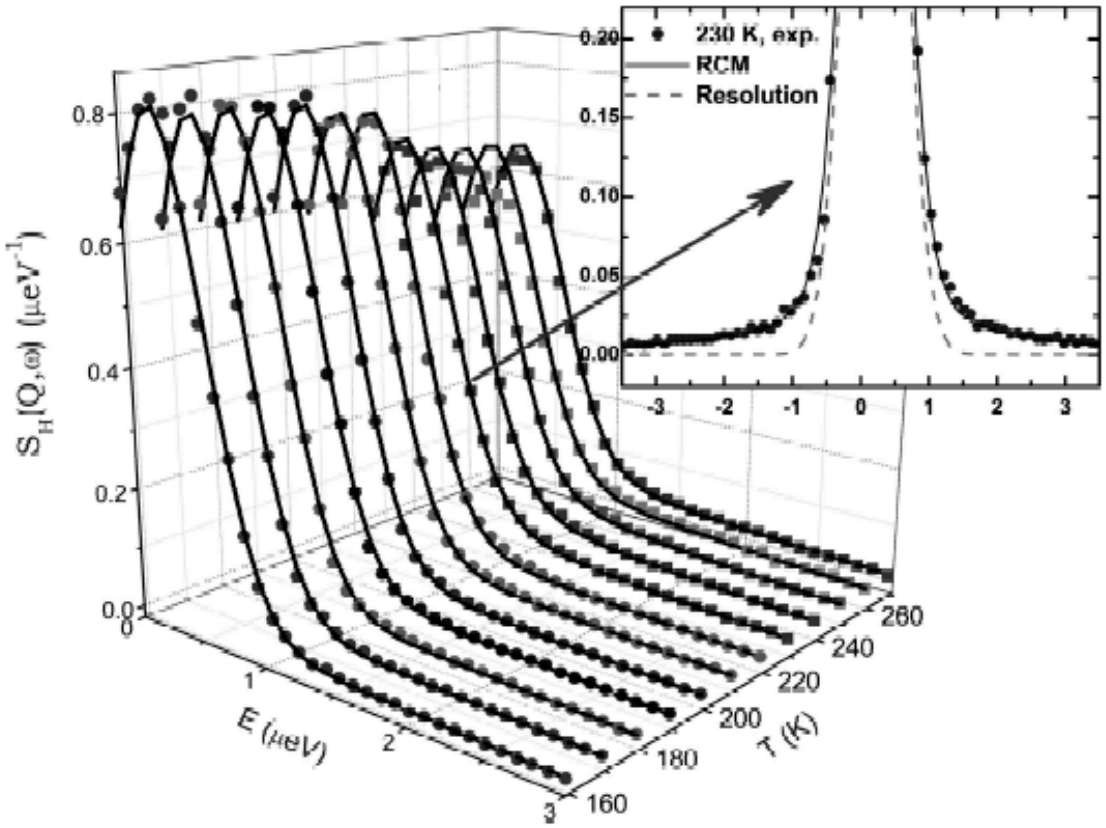


M. Lagi et al. *J. Phys. Chem. B, Vol. 112, (2008), 1571-1575*

Hydration Water in Lysozyme

Lysozyme/H_2O spectra - Lysozyme/D_2O spectra =

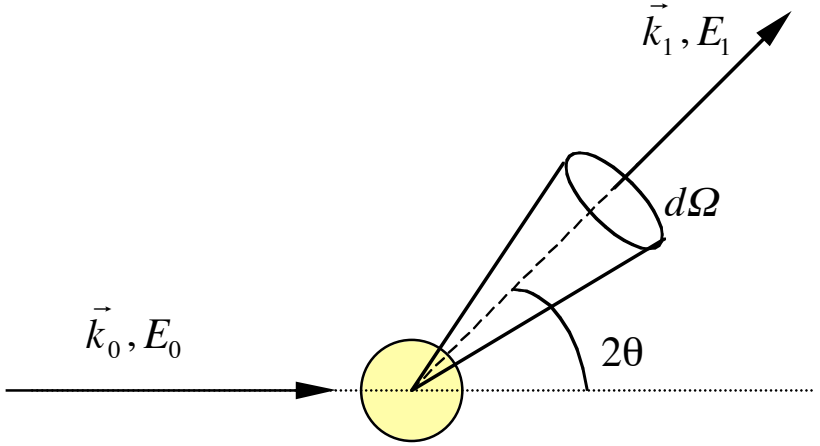
Only Scattering from the Hydration Water

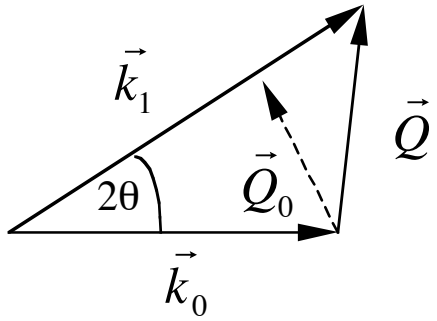


S.-H. Chen, et al., PNAS, 103, 9012 (2006).

QENS: Background

QENS = quasi-elastic neutron scattering





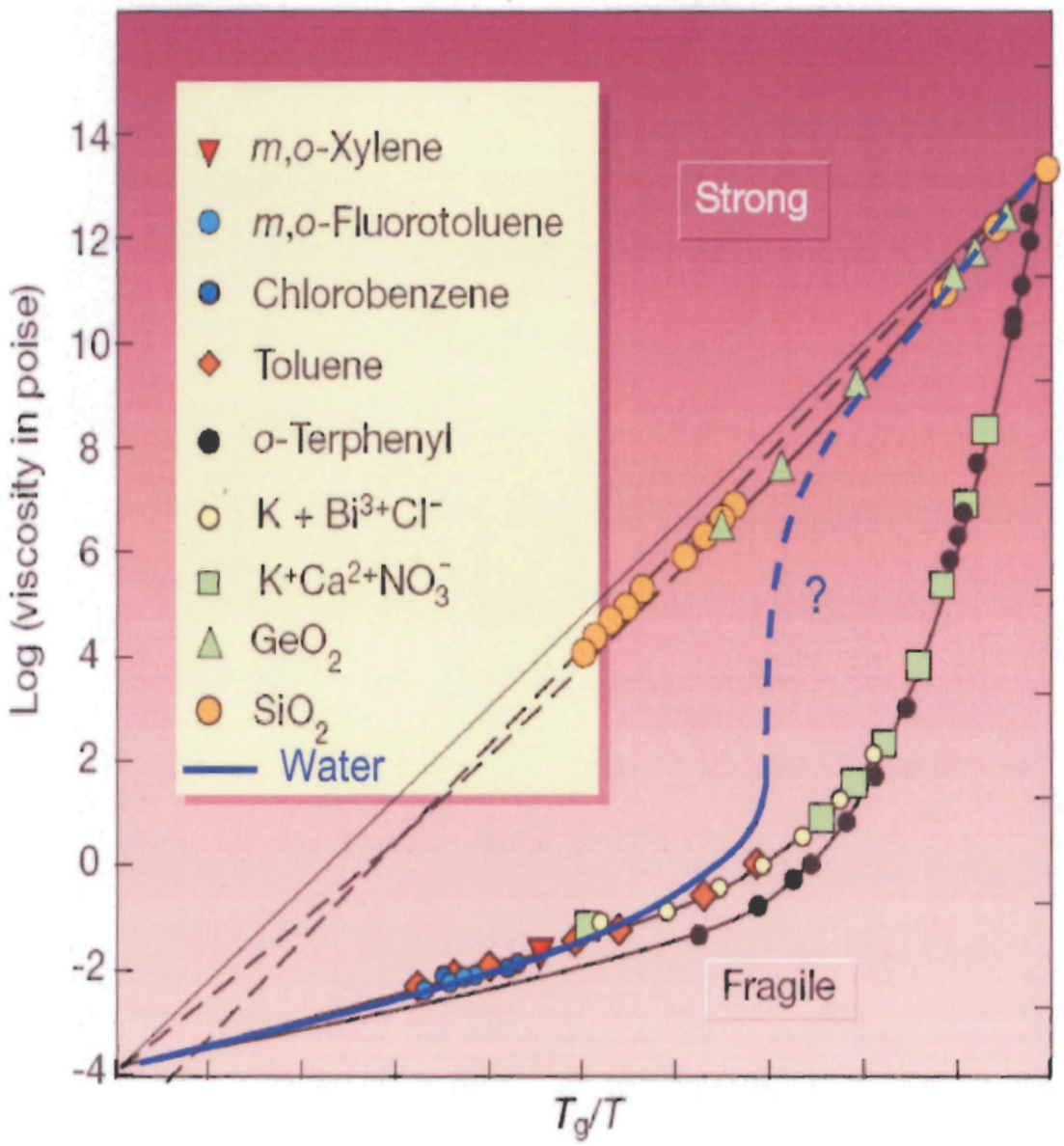
$$\vec{Q} = \vec{k}_1 - \vec{k}_0$$

$$\frac{d^2\sigma}{d\Omega d\omega} = S_{inc}\left(\vec{Q},\omega\right) \otimes R(\omega)$$

Measure the number of scattered neutrons as a function of Q and ω

$$S_{inc}\left(\vec{Q},\omega\right) = S_{inc}^{trans}\left(\vec{Q},\omega\right) \otimes S_{inc}^{rot}\left(\vec{Q},\omega\right) \otimes S_{inc}^{vib}\left(\vec{Q},\omega\right) \quad Q < 1.2 \text{ Å}^{-1}$$

Viscosity η or Equivalently the Structural Relaxation Time τ



P.G. Debenedetti, F.H. Stillinger, *Nature* **410**, 259 (2001)

$$\eta = G_\infty \tau \quad \text{(Maxwell's Relation)}$$

Fragile $\quad \tau = \tau_0 \exp\left[\frac{DT_0}{T - T_0}\right]$

Strong $\quad \tau = \tau_0 \exp\left[\frac{E_A/k_B}{T}\right]$

Fragile-to-Strong (F-S) transition is defined as a temperature T_L where:

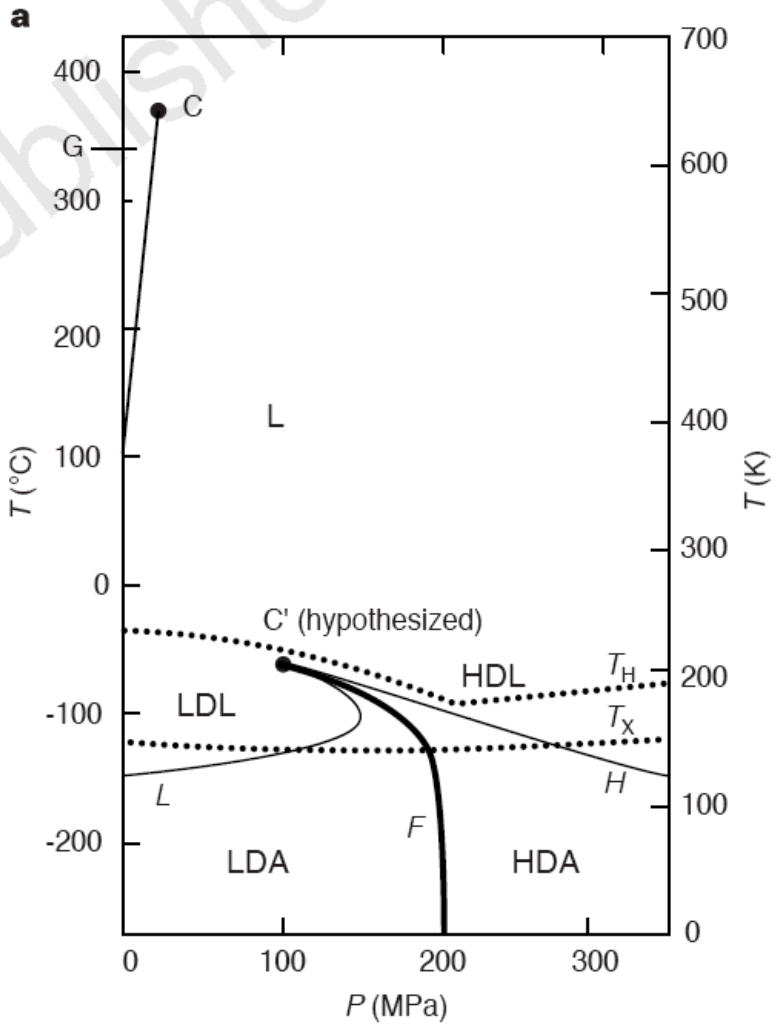
$$\exp\left[\frac{DT_0}{T_L - T_0}\right] = \exp\left[\frac{E_A/k_B}{T_L}\right]$$

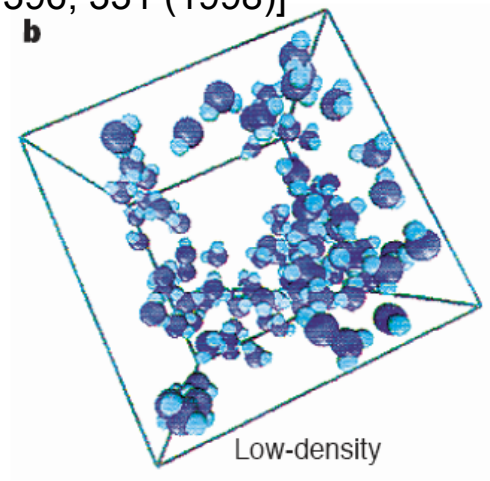
or: $\frac{1}{T_L} = \frac{1}{T_0} - \frac{Dk_B}{E_A}$

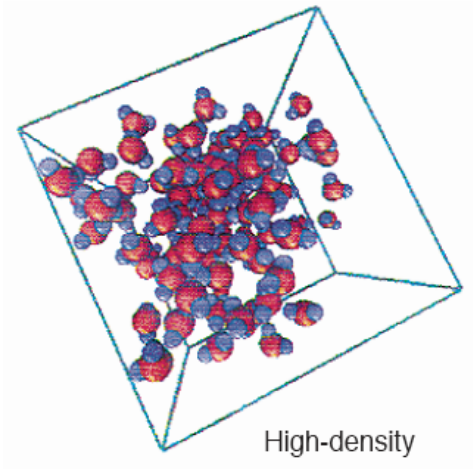
L Liu, SH Chen, *et al*, *Phys. Rev. Lett.* **95**, 117802 (2005).

The Two Critical Points Scenario in Water

[O. Mishima, and H. E. Stanley, Nature 396, 331 (1998)]

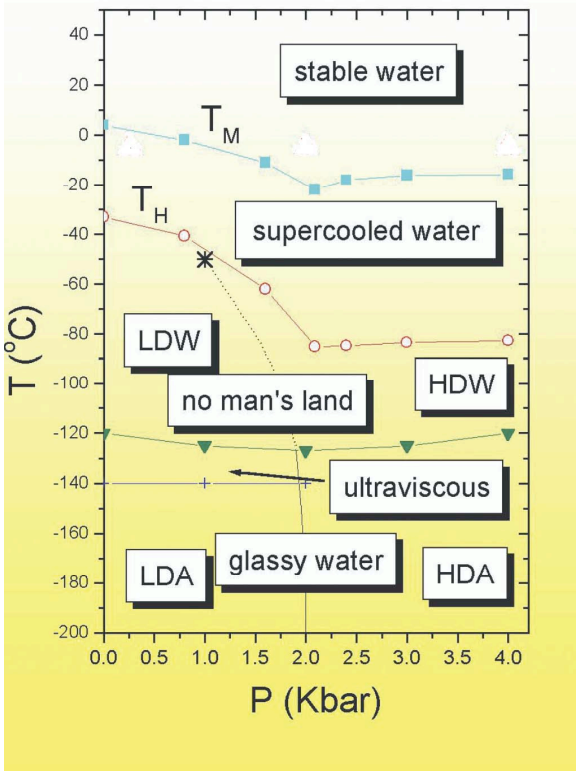




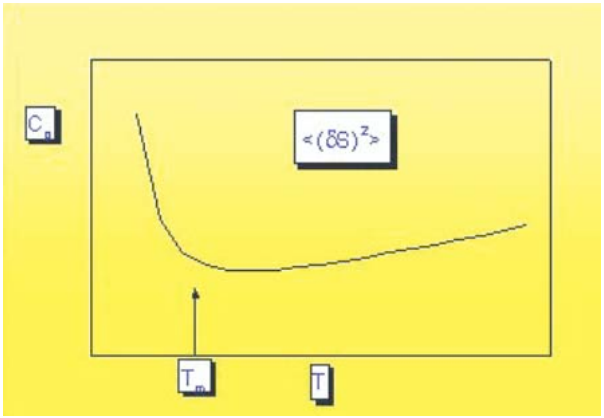


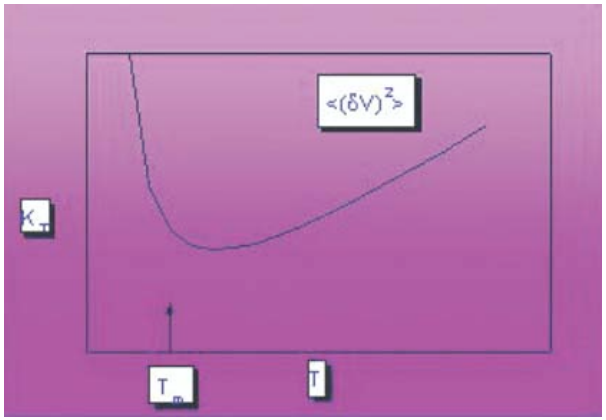
Water Phase Diagram

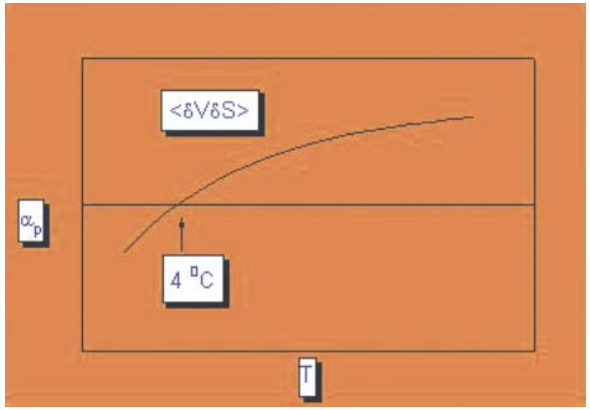
Phase diagram T vs P of water



H_2O thermodynamic anomalies





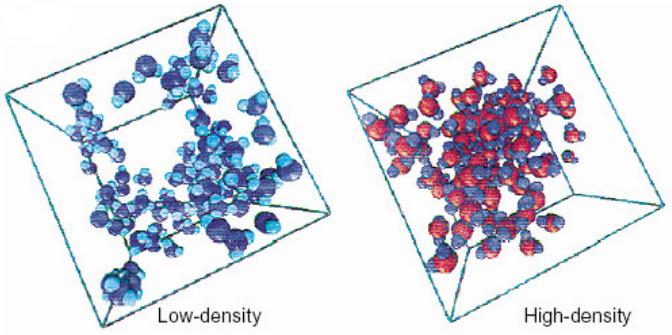


By courtesy of M.A. Ricci

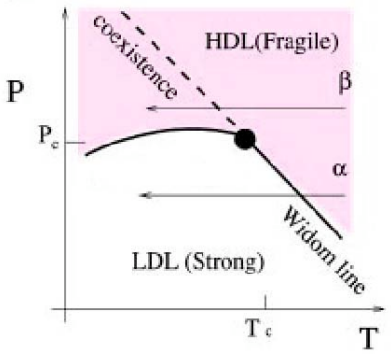
*: Hypothesized second critical point (p_c ~ 1 Kbar, T_c ~ 223 K)

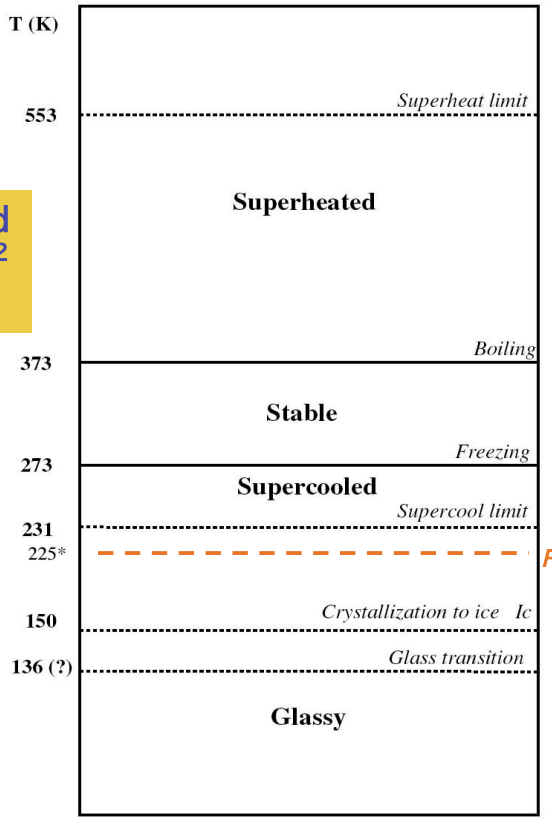
FSC in water

Dynamic crossover from a *fragile* - high-density (HDW) to a *strong* - low-density (LDW) liquid around 225 K* at ambient pressure



Hypothesized second liquid-liquid critical point[2] (1 Kbar, 223 K)





'Strong' ➡ Arrhenius behavior of transport properties and relaxation processes (es. SiO_2, GeO_2, As_2S_3)

'Fragile' ➡ 'Non-Arrhenius' behavior (es. Ionic liquids, $ZnCl_2$, $CH_3Py^+Cl^-$, $K^+Ca^{2+}NO3^-$ or aromatic hydrocarbons (toluene, clorobenzene, o-terphenyl)

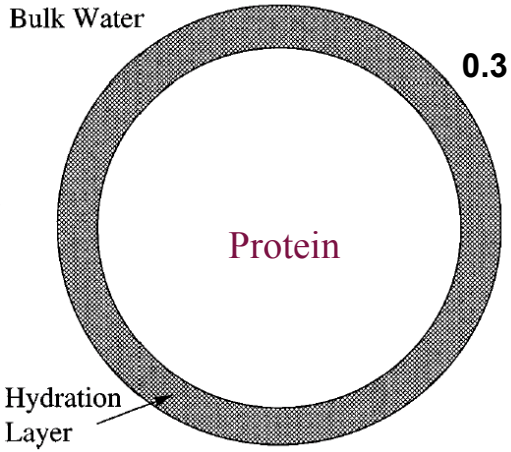
Glass-like dynamical transition at T ~ 200 K in hydrated proteins

Confining system: lysozyme surface

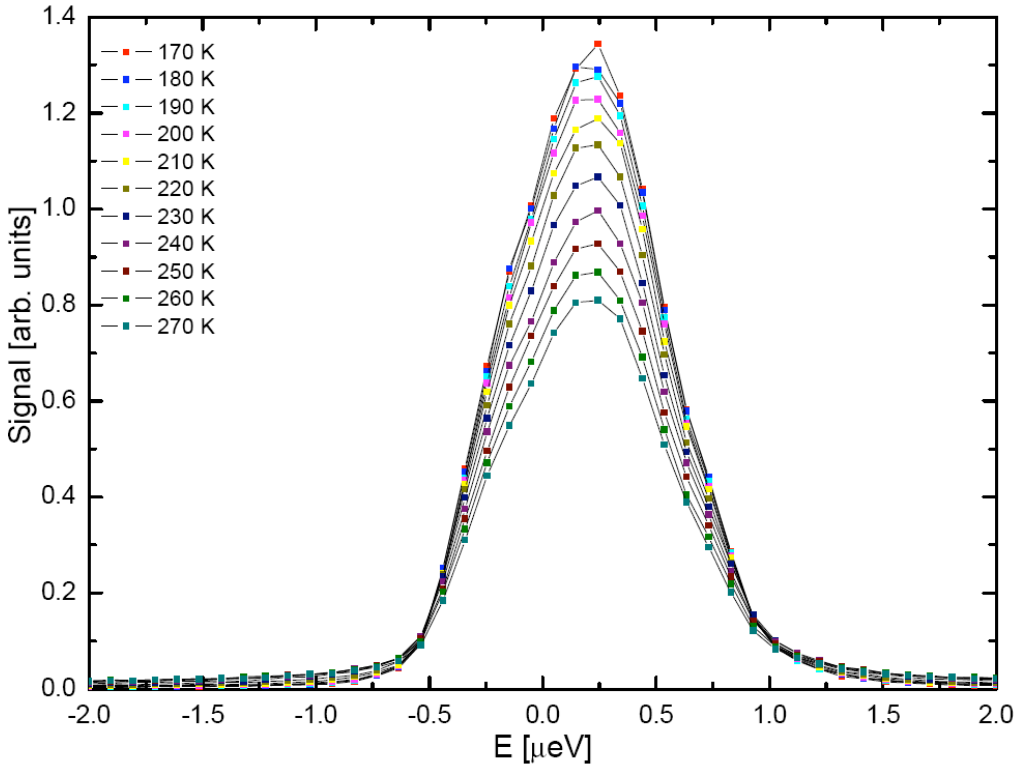
The glass transition in proteins is triggered by FST in hydration water
Water as a *plasticizer* of proteins

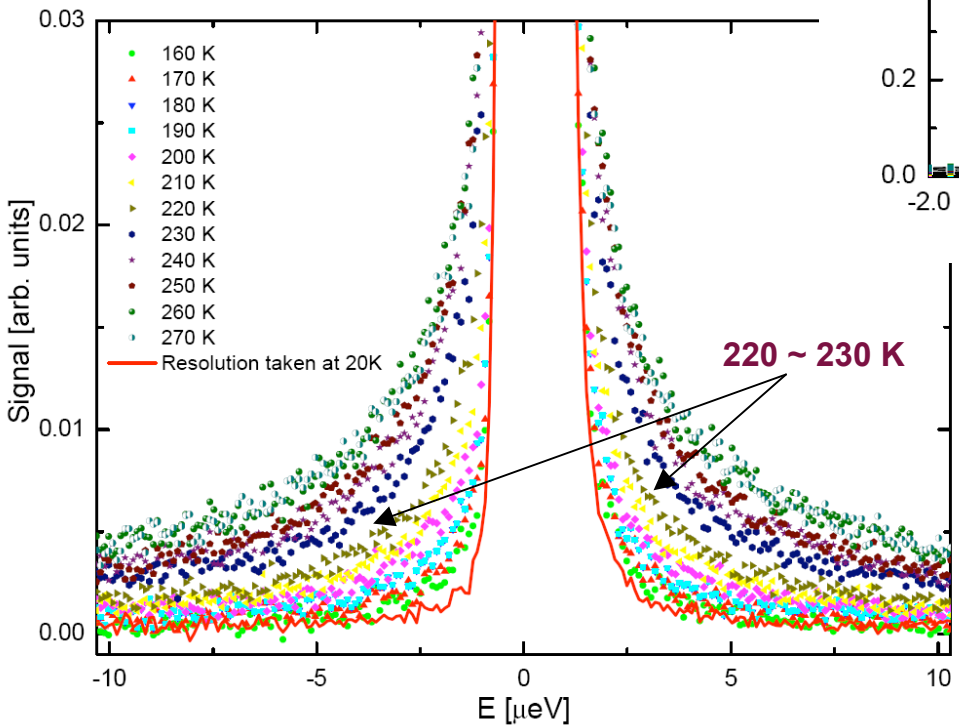
[1]Angell, C. A. *Science*, **1995**, 267, 1924
[2]Stanley,E. et al. *PNAS* **2005**, 102, 16558

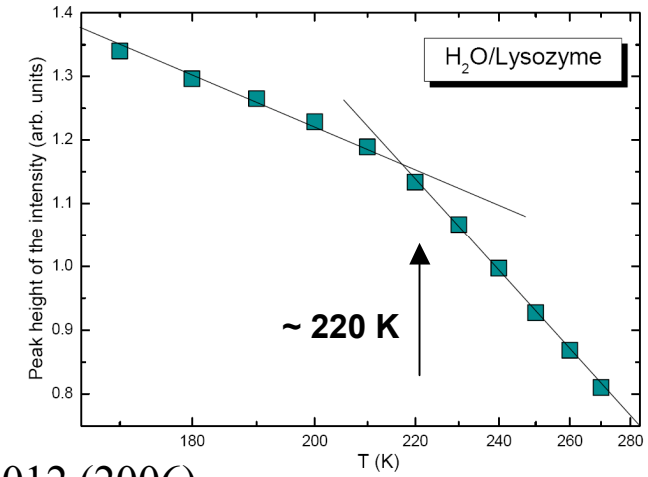
Dynamics of Protein Hydration Water. QENS



0.3 g water/ 1 g of protein







S.-H. Chen, et al., PNAS, 103, 9012 (2006).

Model for ISF and SF

$$F_s(Q,t) = p + (1-p)F_v(Q,t)\ \exp\left[-(t/\tau)^{\beta}\right]$$

(Intermediate Scattering Function)

$$\tau_T(Q) \cong \tau_0(aQ)^{-\gamma}$$

$$\langle\tau_T\rangle = \tau_0\Gamma(1/\beta)/\beta$$

Elastic part

QuasiElastic part:

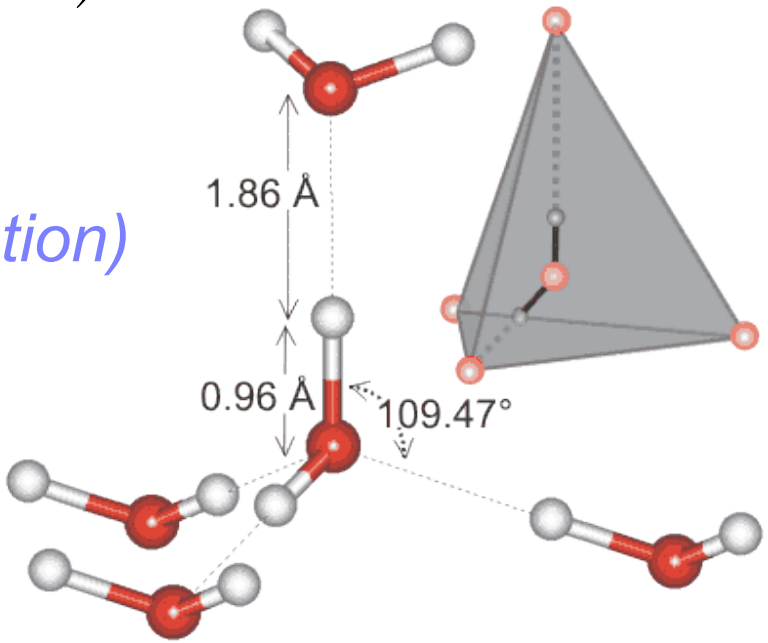
(in cage oscillation + cage relaxation)

Fitting parameters:

p = immobile water

β = exponent (α-relaxation)

τ = relaxation time



$$S_{inc}(Q,\omega) = \frac{1}{2\pi}\int_{-\infty}^{\infty} F_S(Q,t)e^{-i\omega t}dt$$

(Dynamic Structure Factor)

Two-step Relaxation of ISF in Supercooled Water

Bulk Supercooled Water

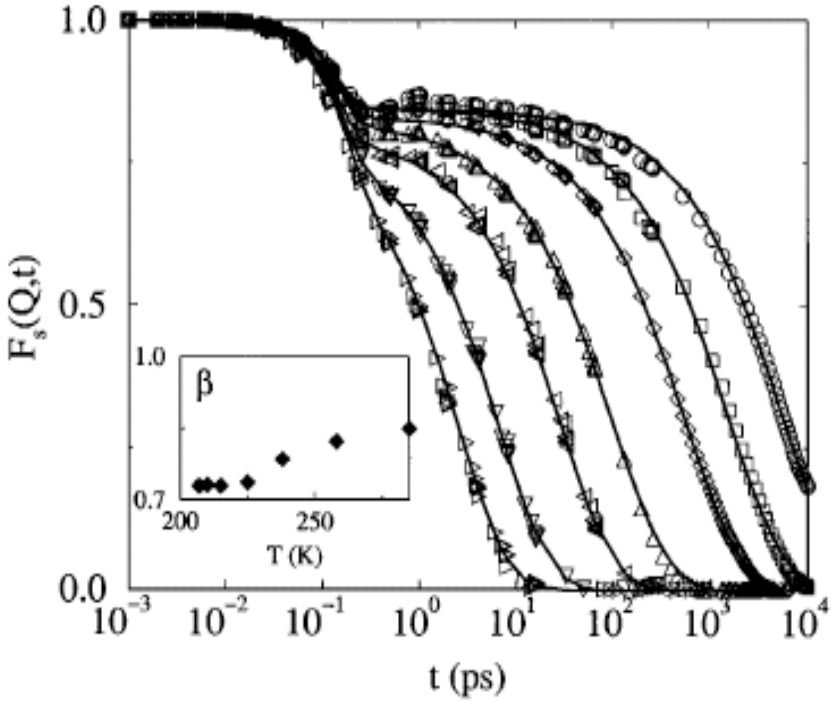


FIG. 2. $F_s(Q_{max}, t)$ vs time (symbols as in Fig. 1). Solid lines are calculated according to Eq.(1). The inset shows the T dependence of the exponent β associated with the slow relaxation.

$$F_T(Q,t) = F_T^S(Q,t)\exp[\ -\ (t/\tau(Q))^{\beta}]$$

P. Gallo, F. Sciortino, P. Tartaglis, S. H. Chen, "Slow dynamics of water molecules in supercooled states", *PRL* **76**, 2730 (1996)

Water in Vycor Glass

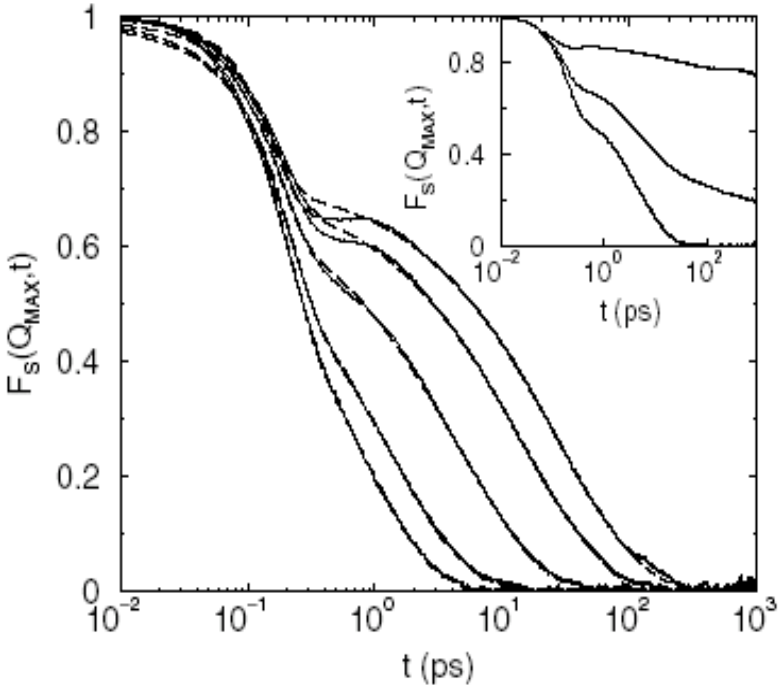
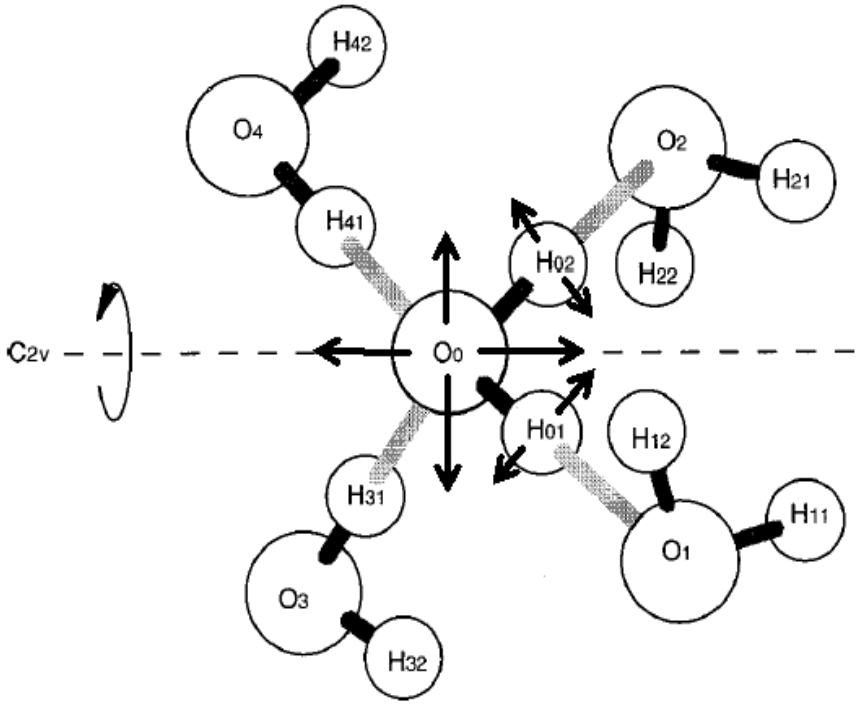


FIG. 2. Intermediate scattering function (ISF) for free water in the xy direction at the peak of the structure factor (Q_{MAX} = 2.25 Å^{-1}) for the five investigated temperatures. Curves on the top correspond to lower temperatures. Full lines are the MD data and long-dashed lines are the fit by Eq. (1). In the inset the full layer analysis is shown for T = 240 K. The central curve is the total ISF, the upper curve is the bound water contribution, and the lower curve is the free water contribution to the total ISF.

P. Gallo, M. Rovere, E. Spohr, "Supercooled confined water and the mode coupling crossover temperature", *PRL* **85**, 4317 (2000)

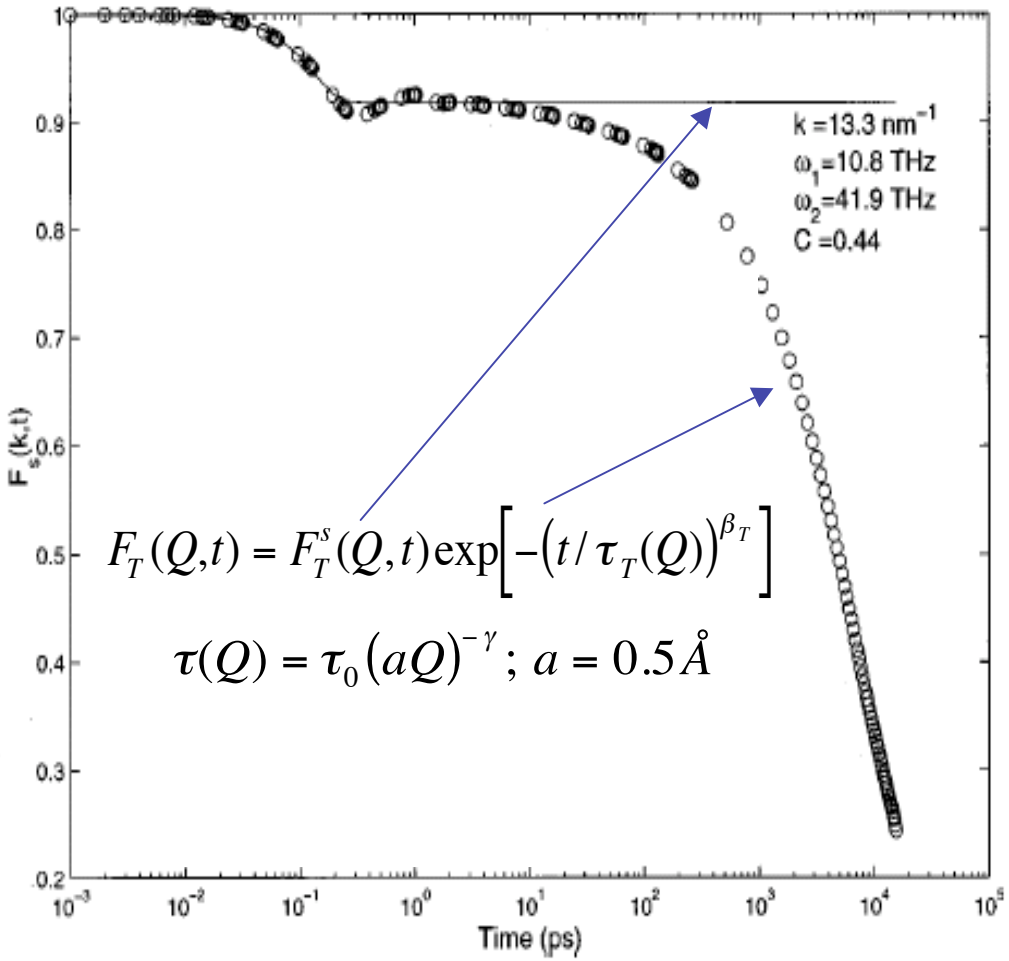
Relaxing-Cage Model

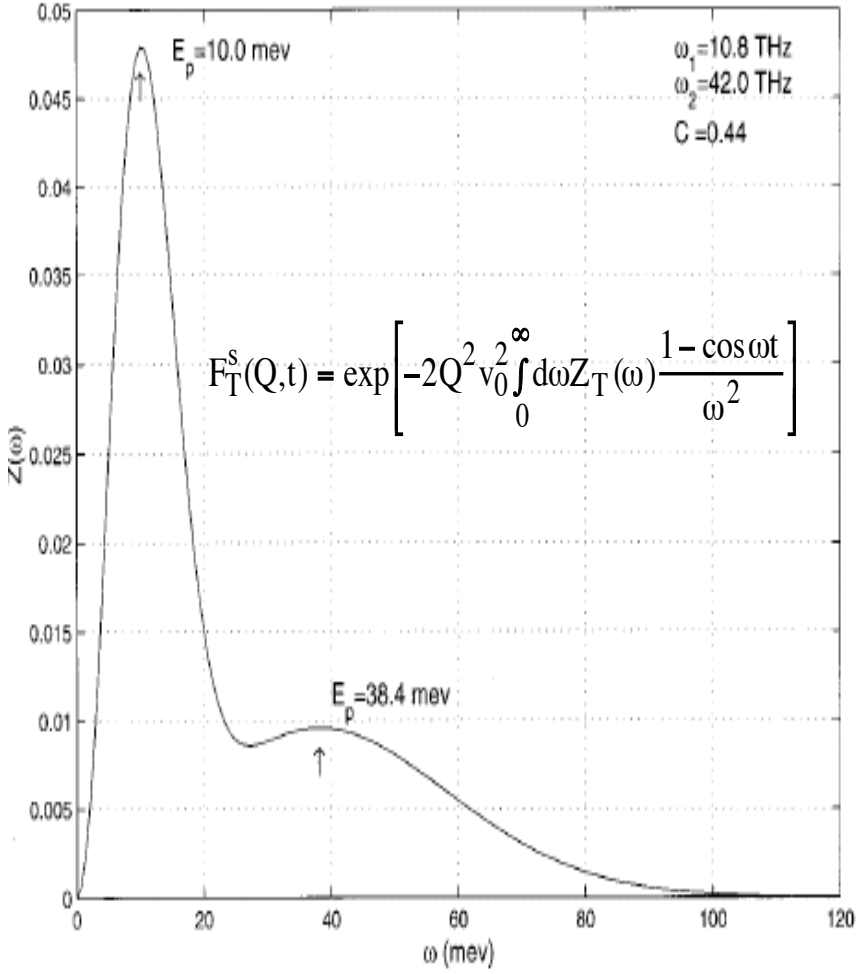
On lowering the temperature below the freezing point, there is a tendency to form around a given water molecule a hydrogen-bonded, tetrahedrally coordinated first neighbor shell (cage). At short times, less than 0.05 ps, the water molecule performs harmonic vibrations and librations inside the cage. At long times, longer than 1.0 ps, the cage eventually relaxes and the trapped particle can migrate through the rearrangement of a large number of particles surrounding it. Thus, there is a strong coupling between the single particle motion and the density fluctuations of the fluid.



S. H. Chen, C. Liao, F. Sciortino, P. Gallo, P. Tartaglia, *Phys. Rev. E* **59**, 6708 (1999)

Model for the Translational Dynamics

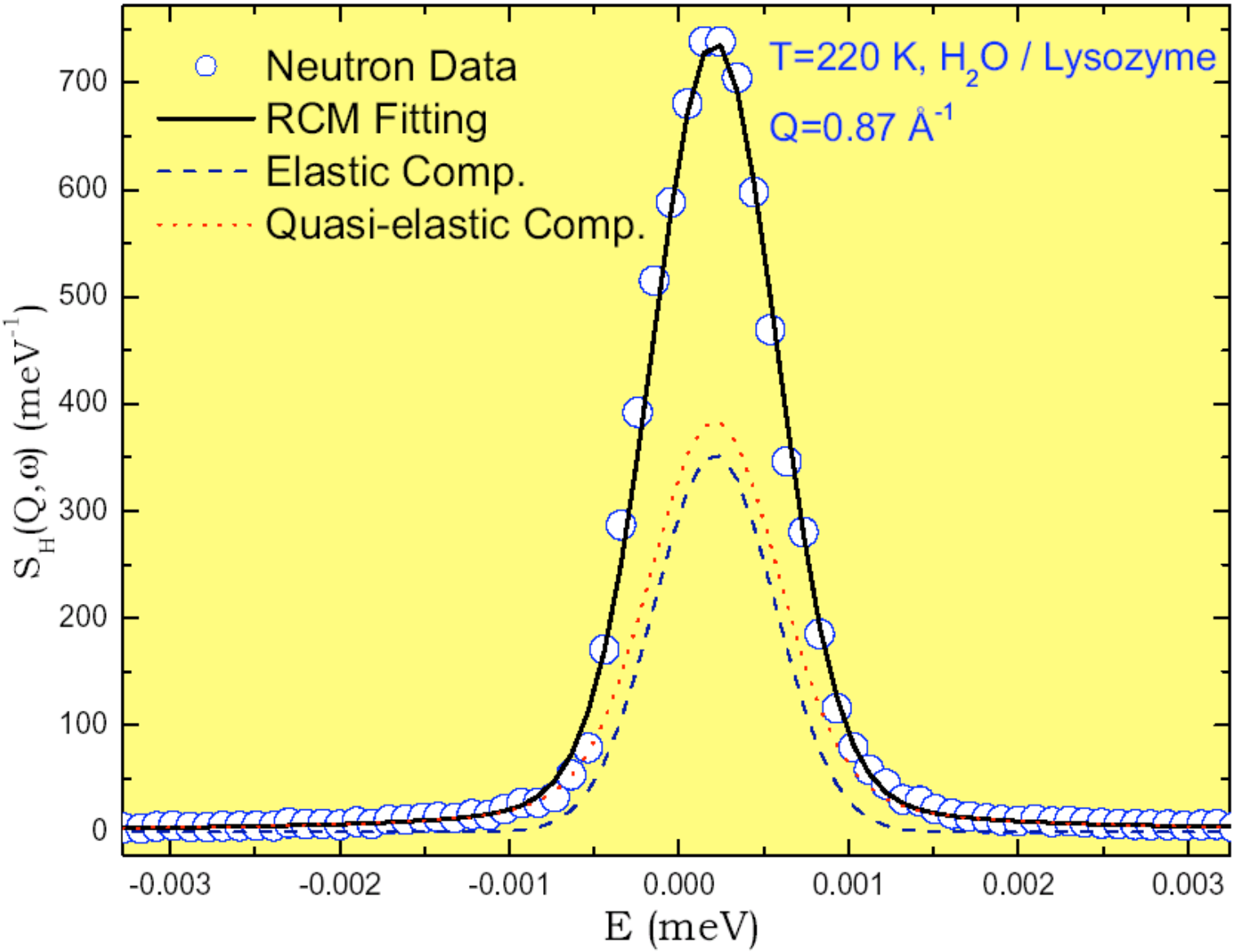




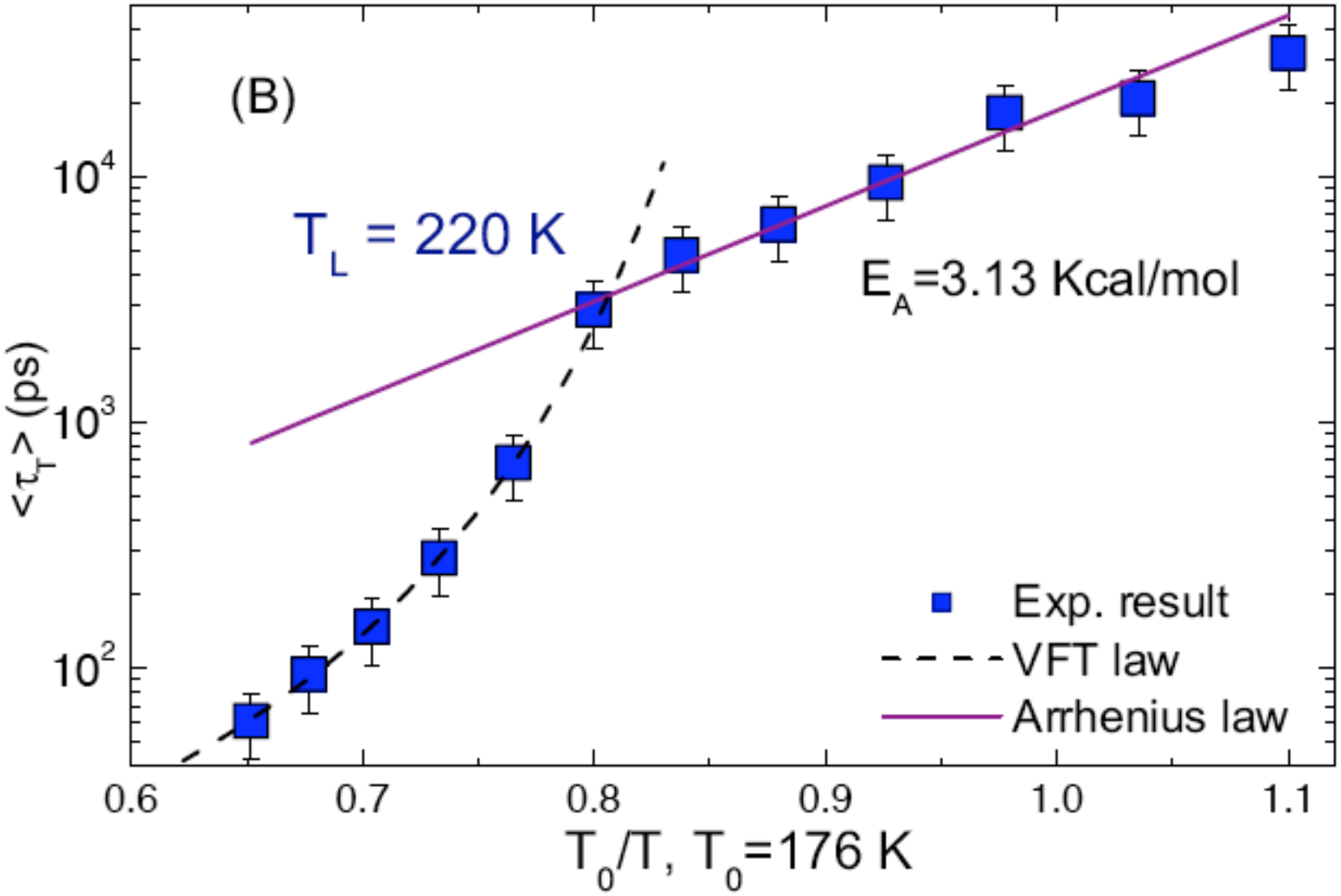
S. H. Chen, C. Liao, F. Sciortino, P. Gallo, P. Tartaglia, "Model for single-particle dynamics in supercooled water", *Phys. Rev. E* **59**, 6708 (1999)

RCM Analysis of QENS spectra

S.-H. Chen, et al. PNAS **103**, 9012-9016 (2006).

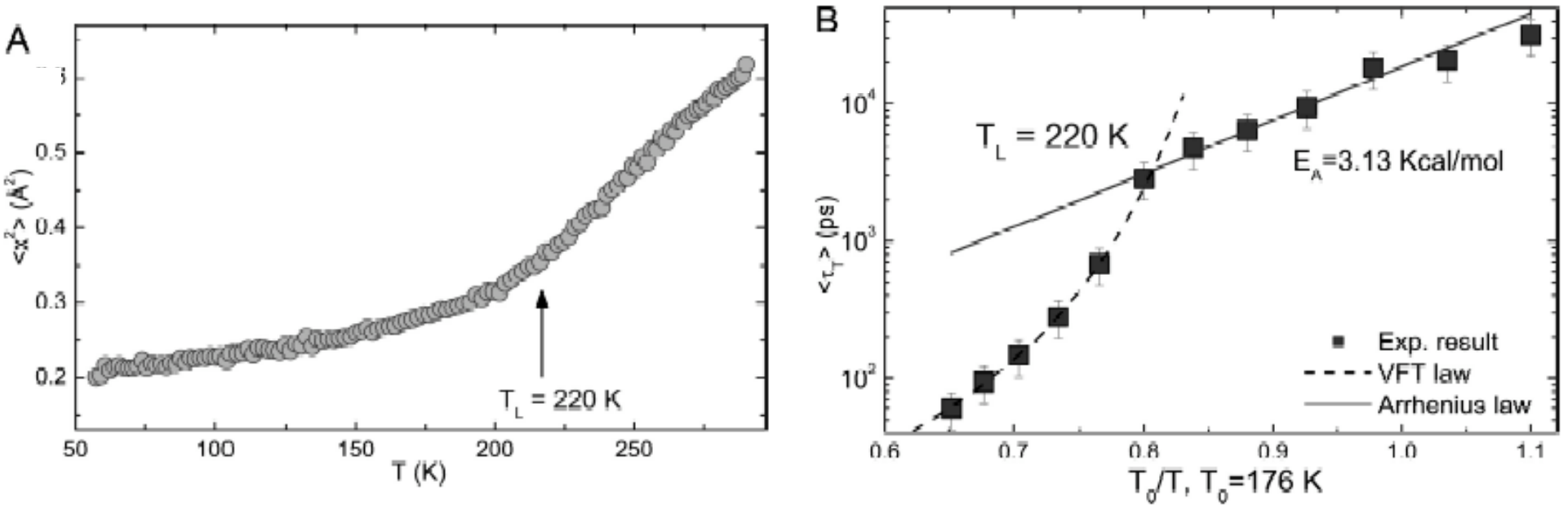


Evidence for dynamic crossover at 220 K in Protein Hydration Water



S.-H. Chen, L. Liu, E. Fratini, P. Baglioni, A. Faraone and E. Mamontov, "Observation of Fragile-to-Strong Dynamic Crossover in Protein Hydration Water," PNAS **103**, 9012-9016 (2006).

Is Lysozyme Dynamics Controlled by the Hydration Water?



The increase in the Dynamics of Lysozyme takes place at the same temperature as the fragile-to-strong transition of the hydration water.

S.-H. Chen, et al. PNAS **103**, 9012-9016 (2006).

Coincidence of Dynamical Transitions in a Soluble Protein and Its Hydration Water: Direct Measurements by Neutron Scattering and MD Simulations

Kathleen Wood,[†,⊥] Andreas Frölich,[†,¶] Alessandro Paciaroni,[‡] Martine Moulin,[§] Michael Härtlein,[§] Giuseppe Zaccai,[§] Douglas J. Tobias,[*,∥] and Martin Weik[*,†]

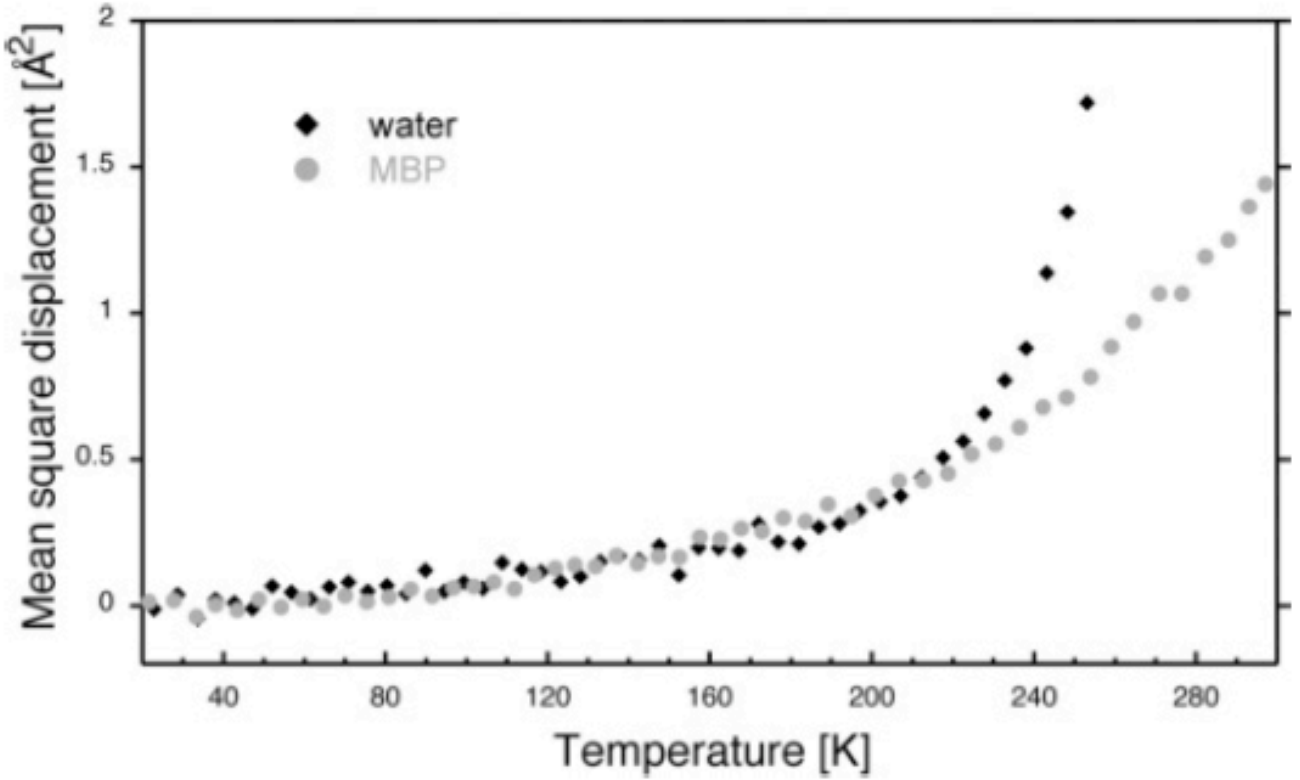


Figure 1. Mean square displacements of ns − ps motions in maltose binding protein (H−MBP−D_2O sample; gray circles) and in its hydration water (D−MBP−H_2O sample; black diamonds). Dynamical transitions (changes in slope of temperature-dependent mean square displacements) in the protein and in its hydration water take place at similar temperatures (~220 K).

J. AM. CHEM. SOC. 2008, *130*, 4586−4587

In general

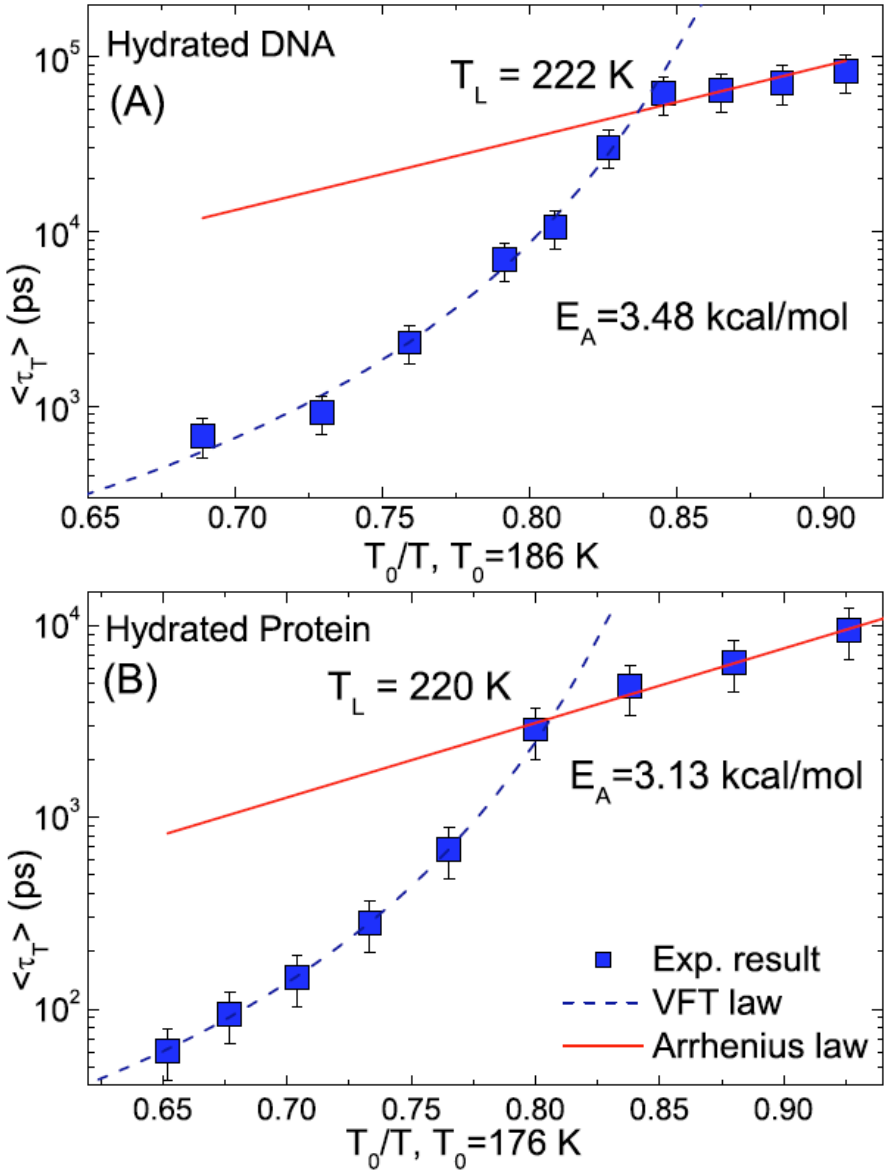
- At low T globular proteins exist in a glassy state having no conformational flexibility and show hardly any biological activities.
- For hydrated proteins above about 200 K, the flexibility is restored, able to sample more conformational sub-states, thus becoming biologically active.

❑ This “dynamical transition” is universal to all biopolymers. Believe to be triggered by their strong coupling with their hydration water, which shows a similar “dynamical transition” at approximately the same temperature.

❑ We show experimentally that this sudden switch in dynamical behavior of hydration water on Lysozyme, B-DNA and RNA occurs precisely at 220 K and can be described as a Fragile-to-Strong dynamic crossover (FSC).

❑ At FSC, the structure of hydration water makes a transition from predominantly high density form (HDL), a more fluid state, to predominantly low density form (LDL), a less fluid state, derived from the existence of a second critical point at an elevated pressure.

Comparison of FSC in DNA and Protein hydration water



Comparison of the temperature dependence of the average translational relaxation times of hydration water:

(A) in hydrated DNA

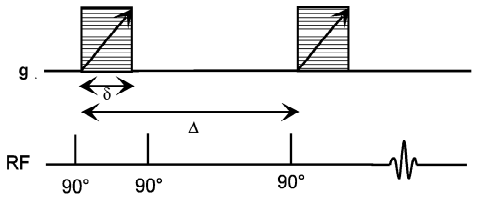
(B) in hydrated Protein.

They both show a cusp-like dynamic crossover phenomenon at temperatures around 220 K. Dash line and solid line are a VFT law and an Arrhenius law fits respectively.

S.-H. Chen et al., "Experimental Evidence of Fragile-to-Strong Dynamic Crossover in DNA Hydration Water," JCP **125**, 171103 (2006).

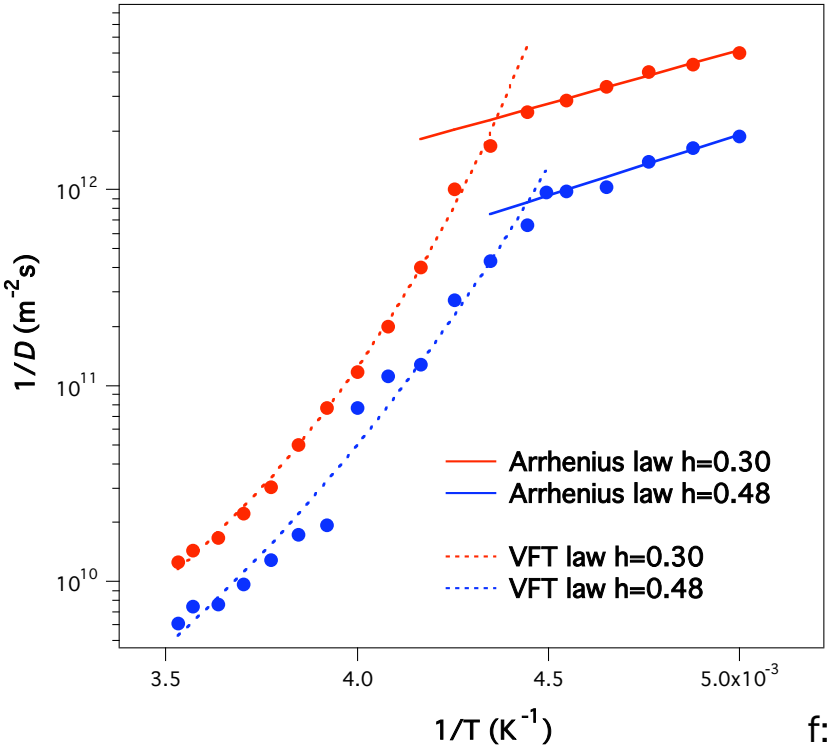
Other evidences of the FSC: NMR

^{1}H-PGSE NMR[1]



$$I = I_0 \exp\left[-D\gamma^2 g\delta^2\Delta\right]$$

δ = 0÷10 ms
Δ= 1 ms÷1 s
g = 1÷20 T/m



T > T_L: Non-Arrhenius behavior
(Vogel-Fulcher-Tamman law)

$$1/D = d\exp\left[\frac{fT_0}{T-T_0}\right]$$

T < T_L: Arrhenius behavior

$$1/D = d\exp\left[\frac{E_A}{RT}\right]$$

f: fragility parameter
T_0: ideal glass transition temperature

D: average self-diffusion coefficient of ^{1}H

h (g H_2O/g lys)	*f*	T_0 (K)	E_A (kcal/mol)	τ_{FST}(K)
0.30	3.47	179.3	2.53	229
0.48	7.66	139.7	2.86	225

EVIDENCE OF FSC at ~ 227 K

[1]Mallamace, F. *et al. J. Chem. Phys.*, **2007**, 127, 045104
[2]Chen, S.-H. *et al. Procl. Natl. Acad. Sci.* **2006**, *103*, 9012

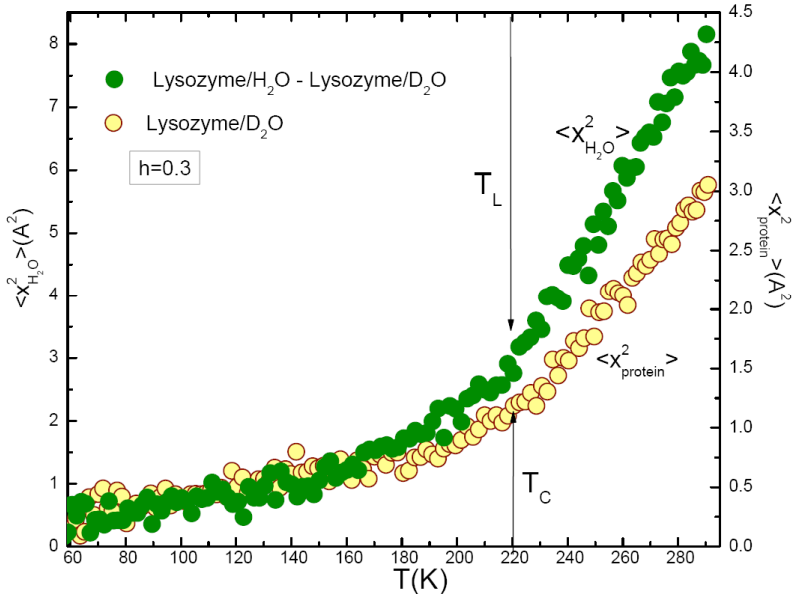
Other evidences of the FSC:

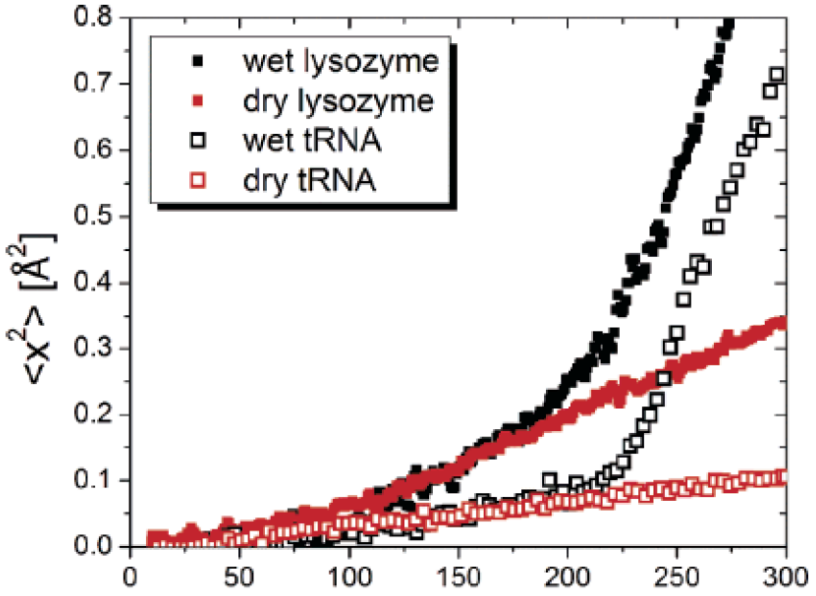
In DSC a well defined “freezing” peak appears at 225K only when h<0.4 and the bulk water peak vanishes.

Low temperature (273-183K) FT-NIR spectra (first overtone of OH stretching mode 6000-7200 cm^{-1}) do not show any hexagonal ice formation in samples with h<0.4. The band maximum frequency shifts showing a discontinuity at 225K.

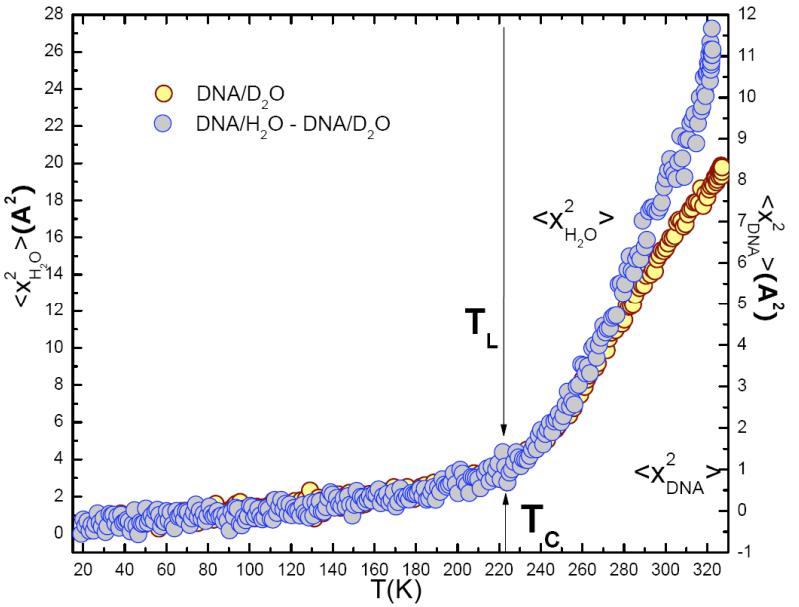
Appearance of a well-defined boson peak.

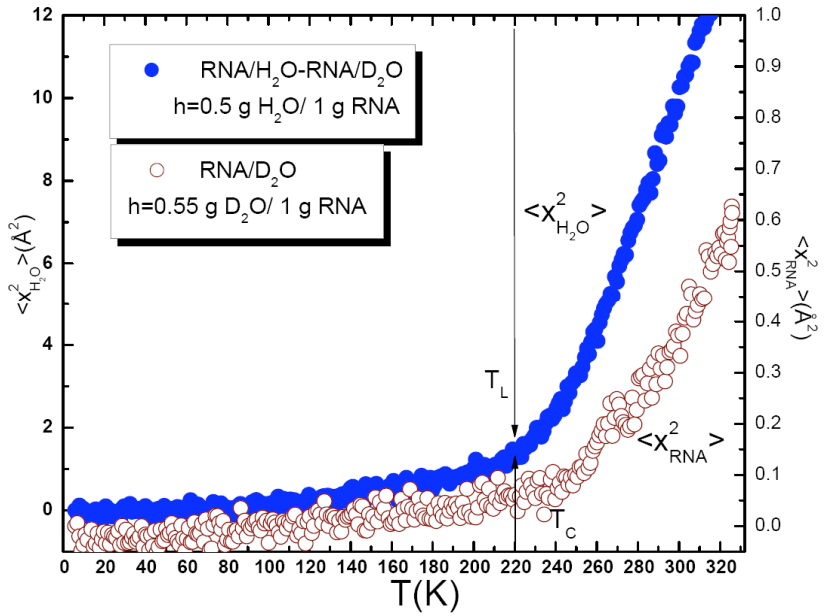
Other Biopolimers:





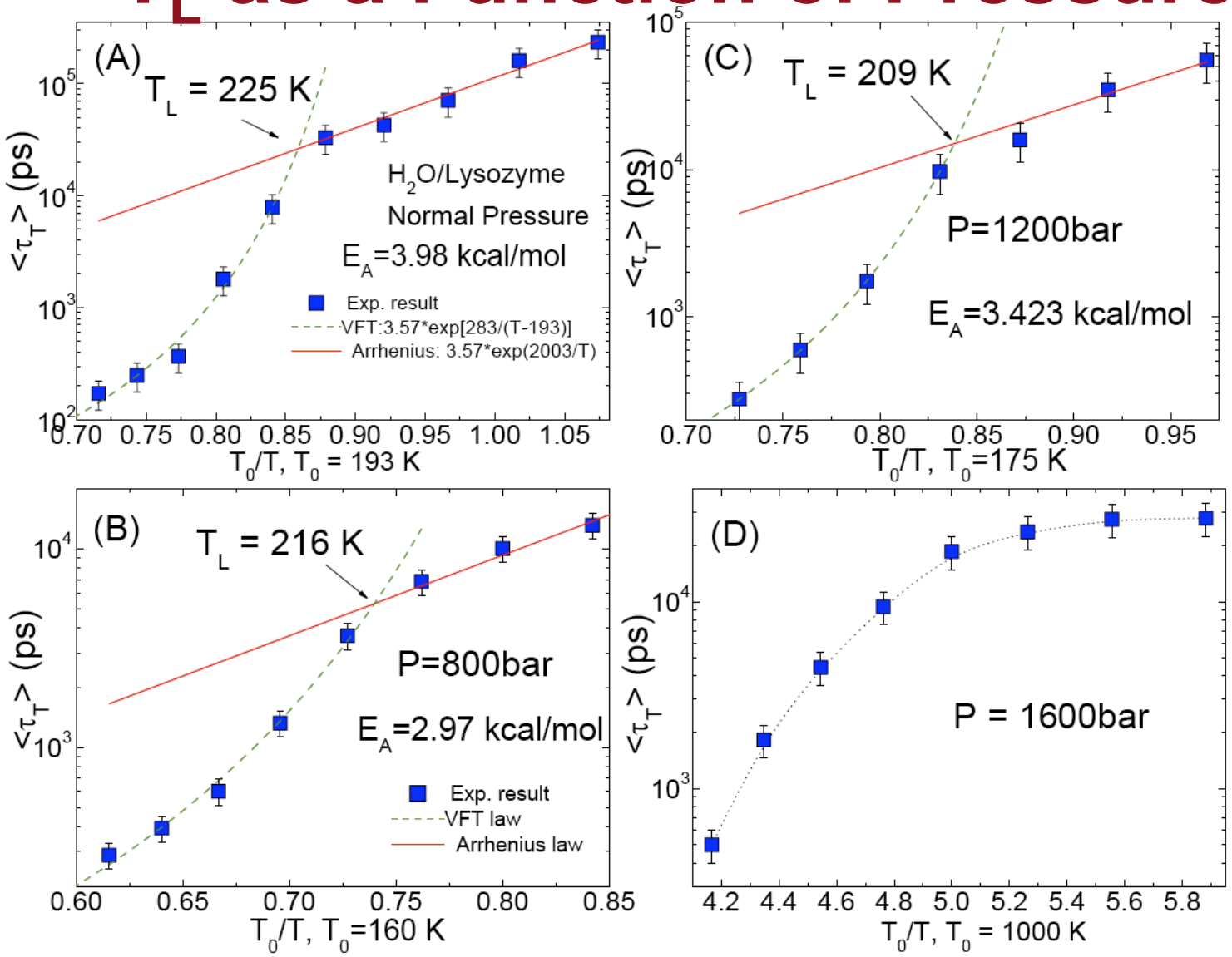
G. Caliskan *et al.*, ***JACS*** 128, 32-33 (2006).





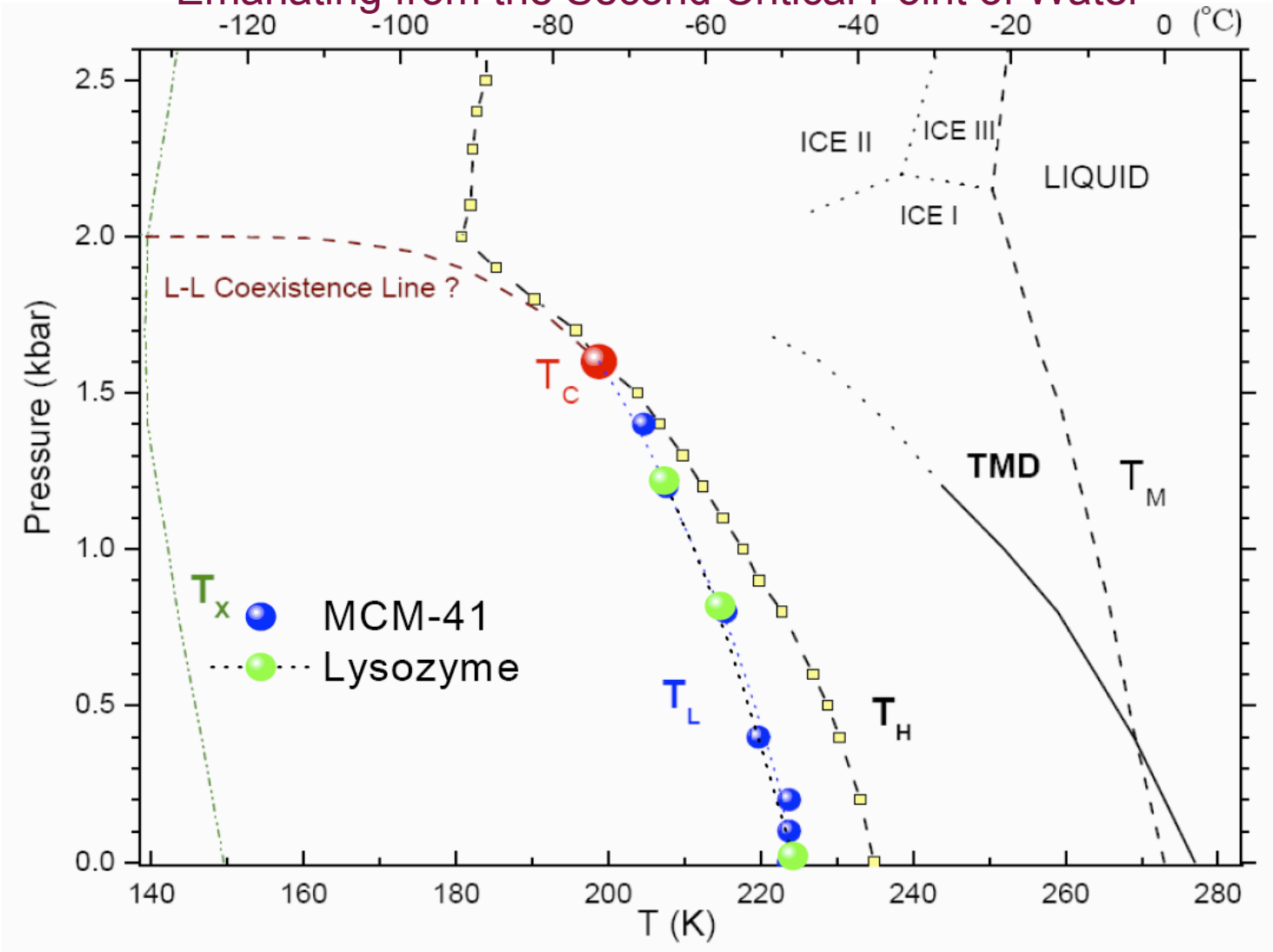
Chen SH, Liu L, Chu X, Zhang Y, Fratini E, Baglioni P,et al. JCP **125**, 171103, 2006

T_L as a Function of Pressure



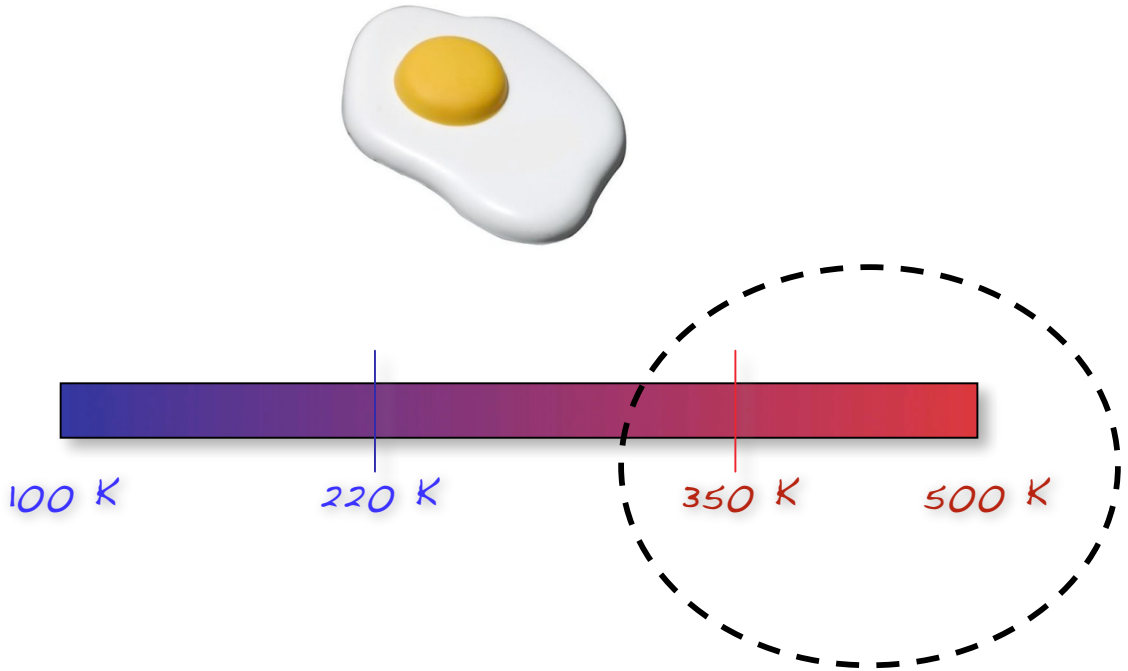
X.-Q. Chu, A. Faraone, C. Kim, E. Fratini, P. Baglioni, J. Leao, and S.-H. Chen "*Observation of Pressure Dependence of the Dynamic Crossover Temperature in Protein Hydration Water*" submitted to PRL.

Tracking of T_L in Hydration Water of Lysozyme Along the Widom Line Emanating from the Second Critical Point of Water

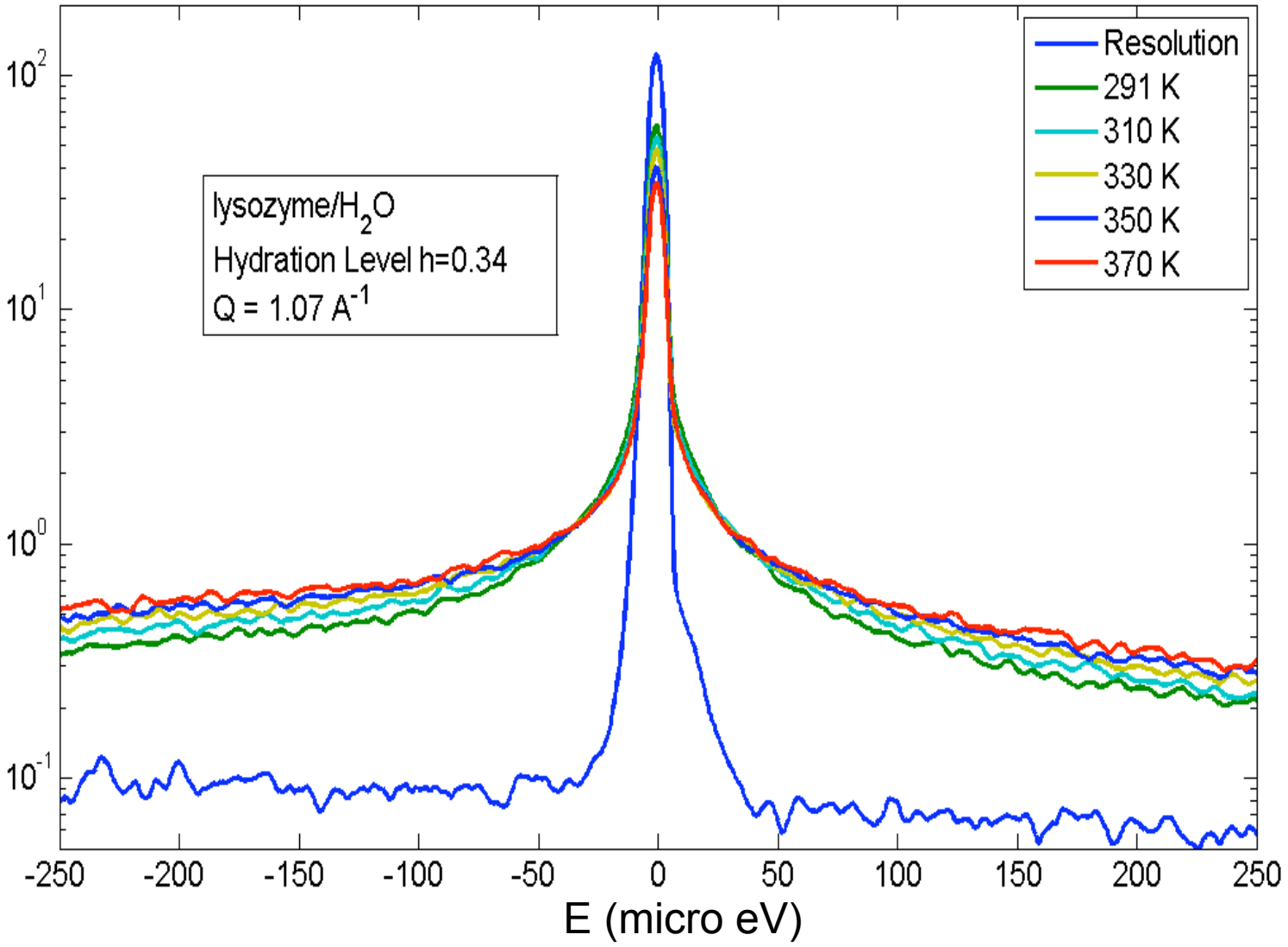


X.-Q. Chu, A. Faraone, C. Kim, **E. Fratini**, P. Baglioni, J. Leao, and S.-H. Chen "*Observation of Pressure Dependence of the Dynamic Crossover Temperature in Protein Hydration Water*" submitted to PRL.

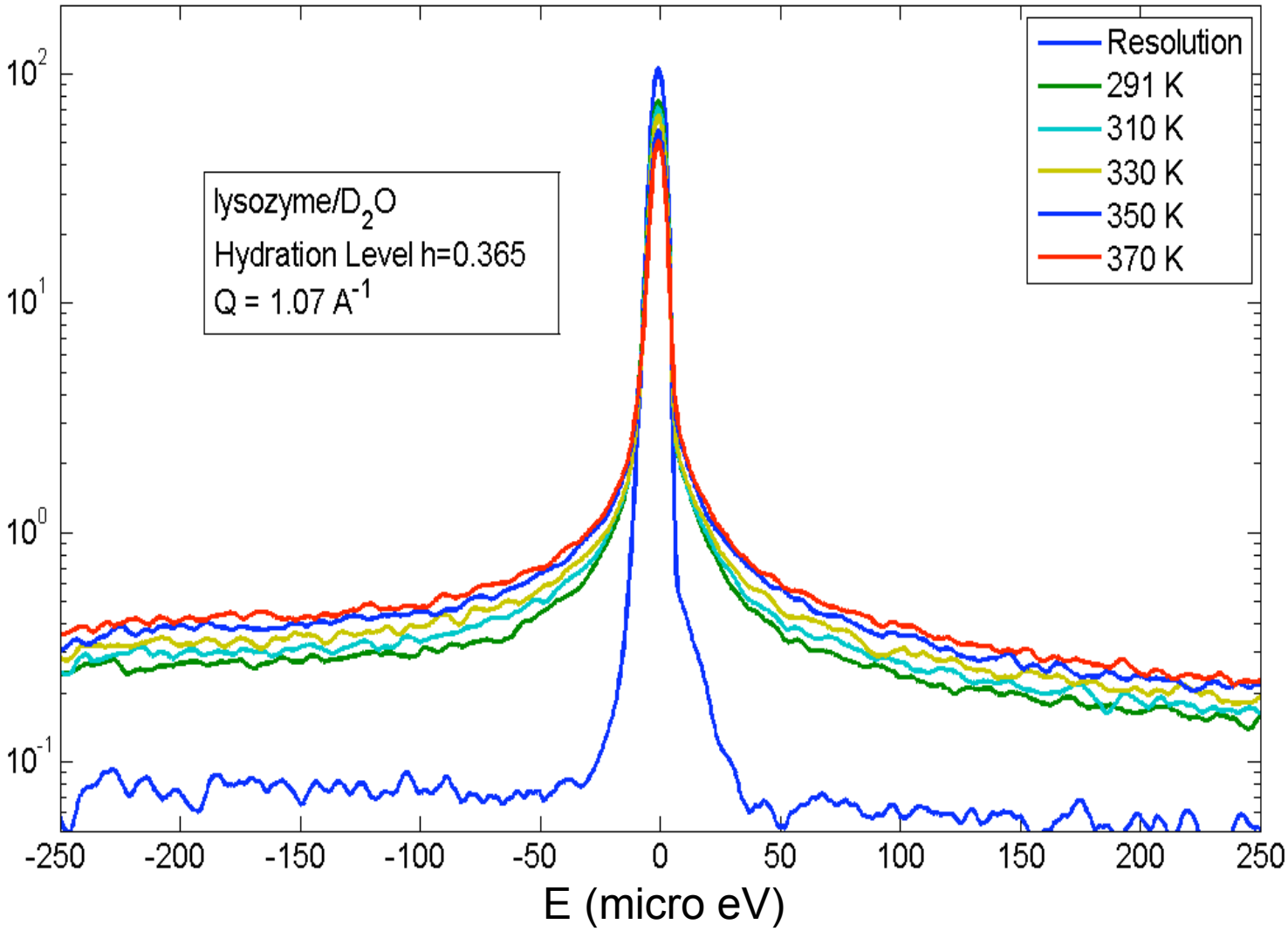
High T Region



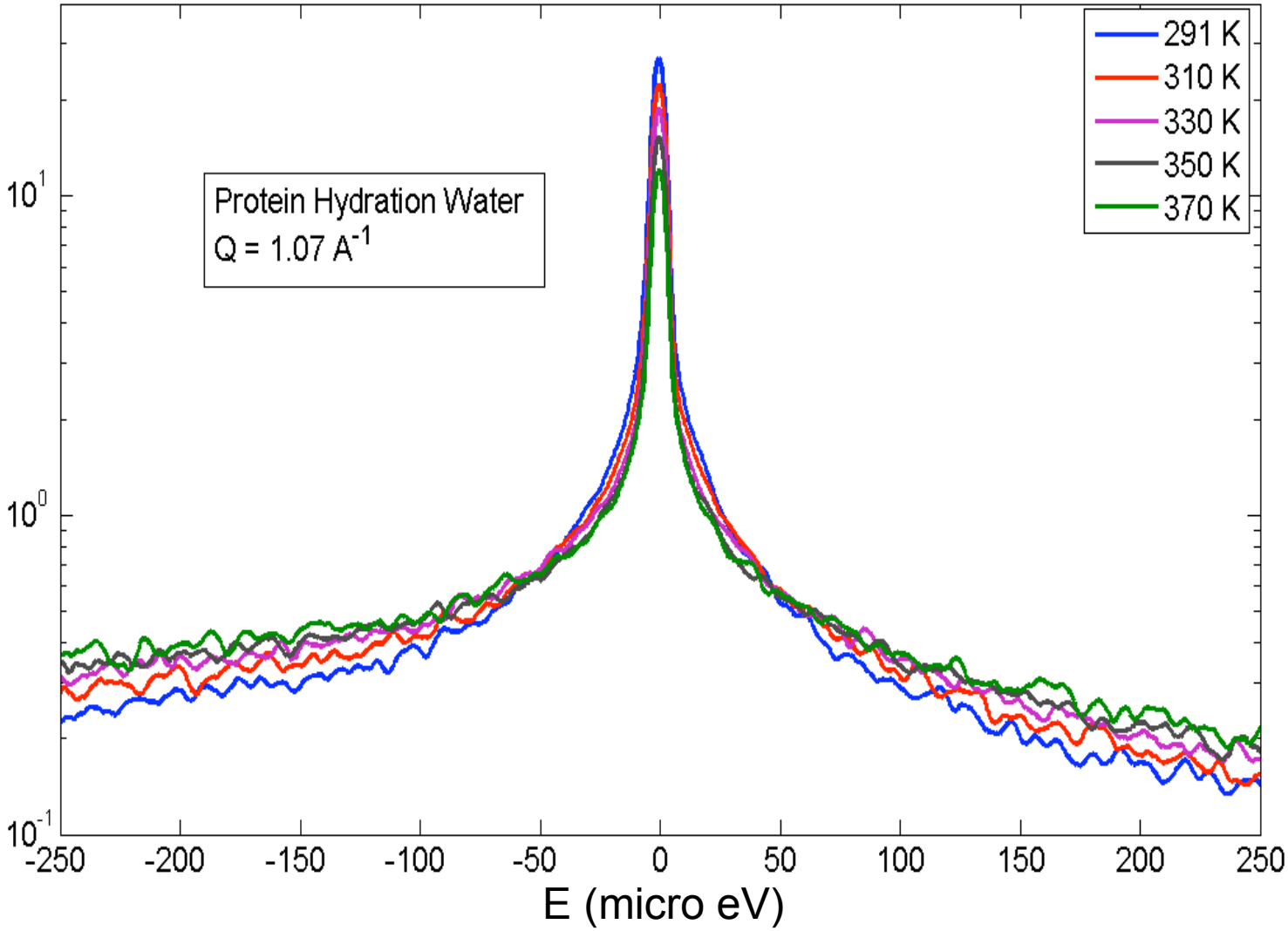
QENS Spectra of H_2O Hydrated Lysozyme



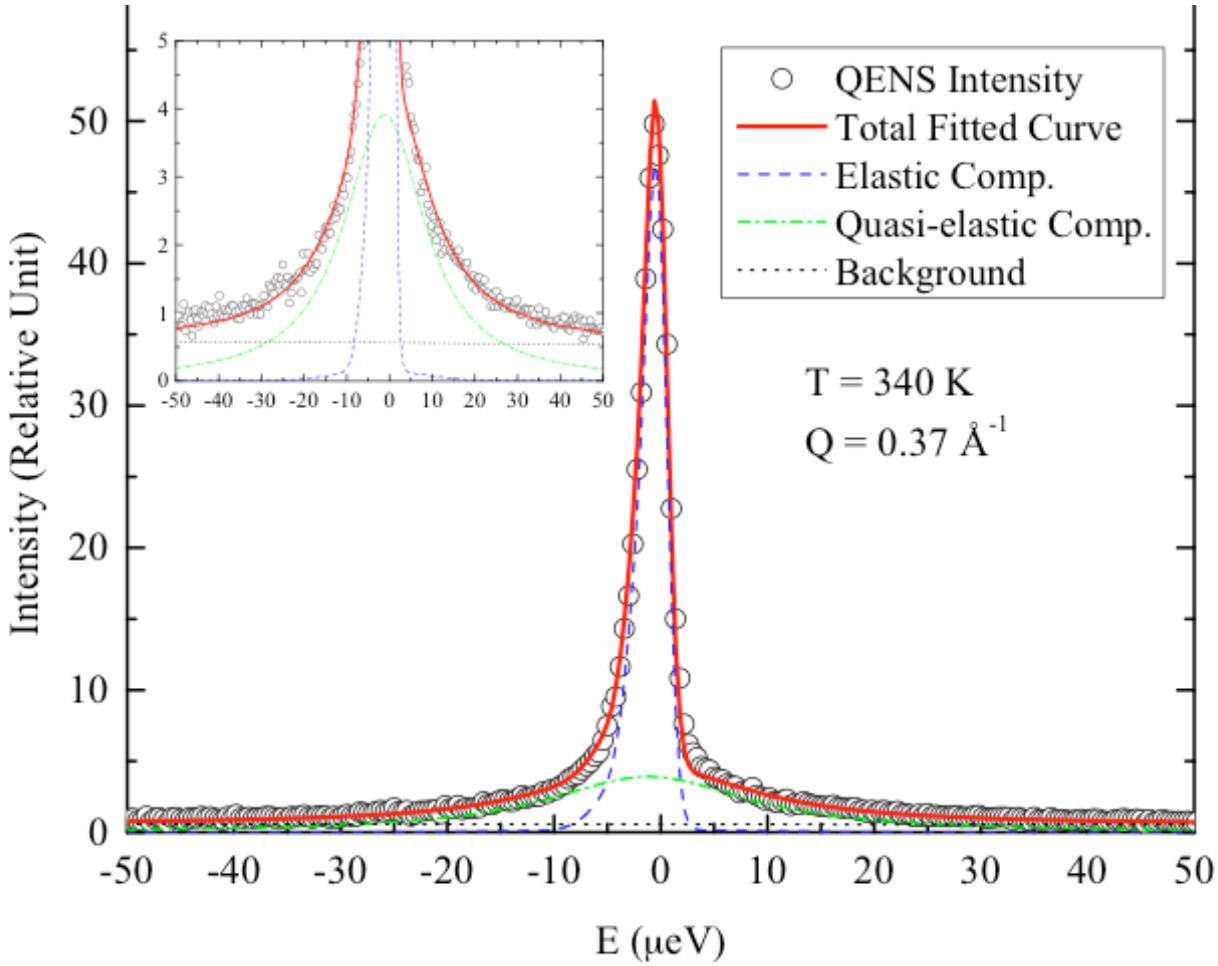
QENS Spectra of D_2O Hydrated Lysozyme



QENS Difference Spectra of H_2O and D_2O Hydrated Lysozyme

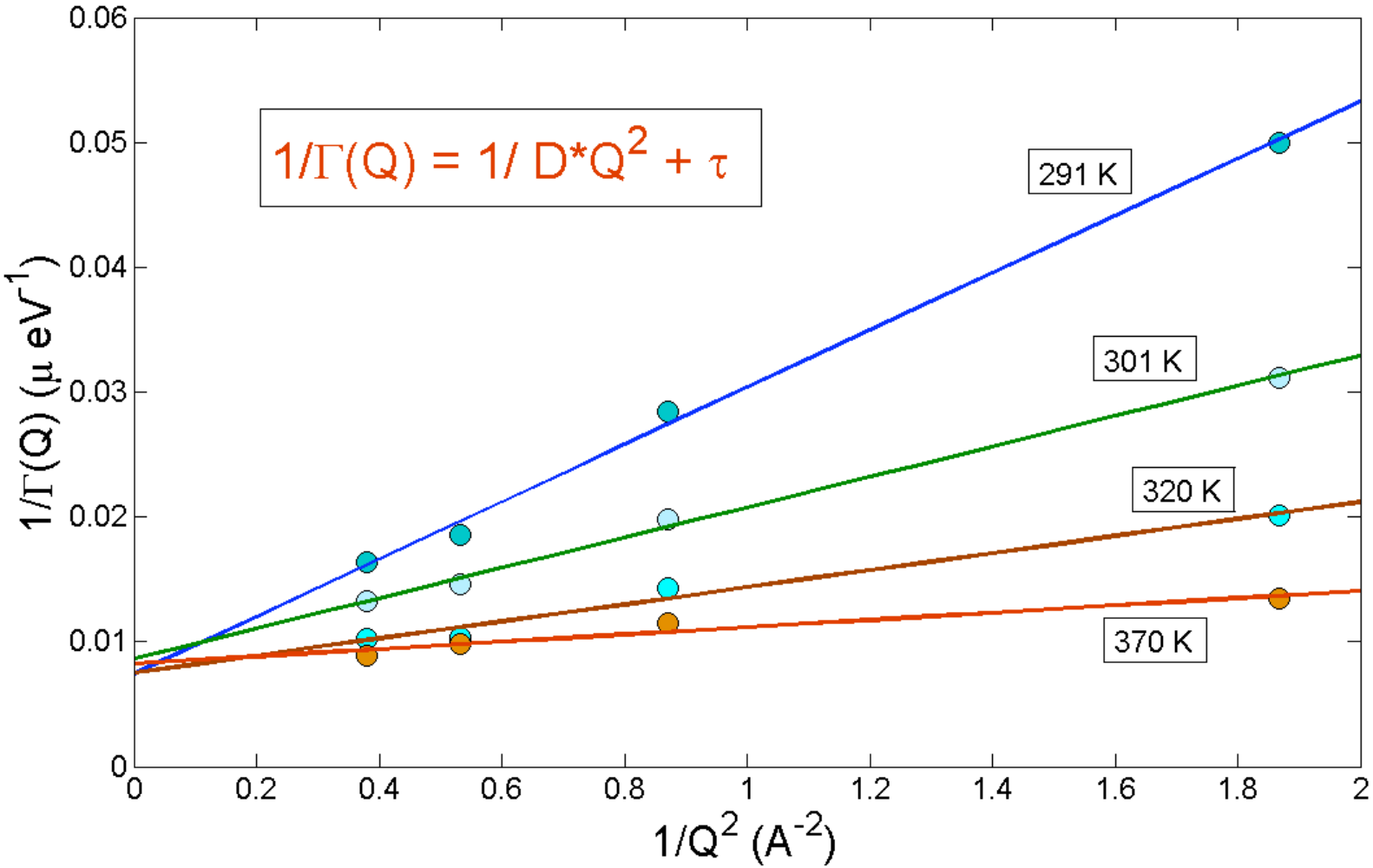


Standard Lorentzian Fit of the Spectra



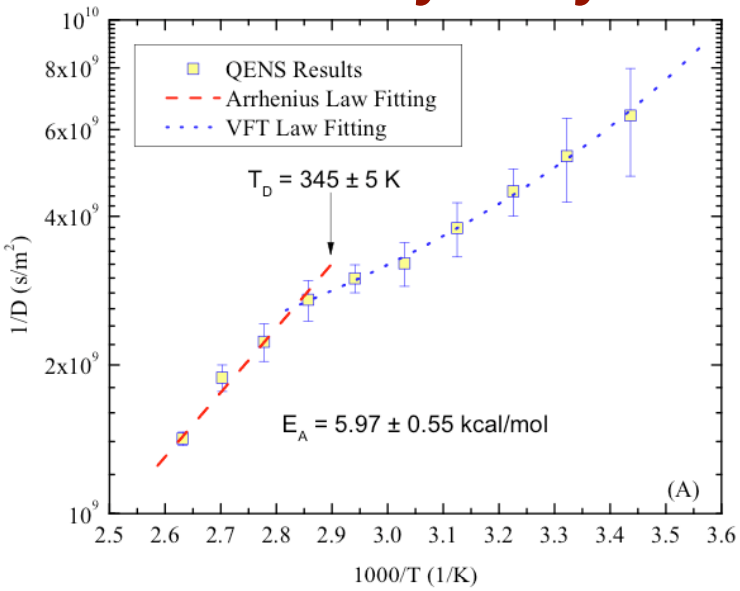
Y. Zhang, M. Lagi, D. Liu, F. Mallamace, E. Fratini, P. Baglioni, E. Mamontov, S.-H. Chen "Observation of high-temperature dynamic crossover in protein hydration water and its relation to reversible denaturation of lysozyme" submitted to JCP.

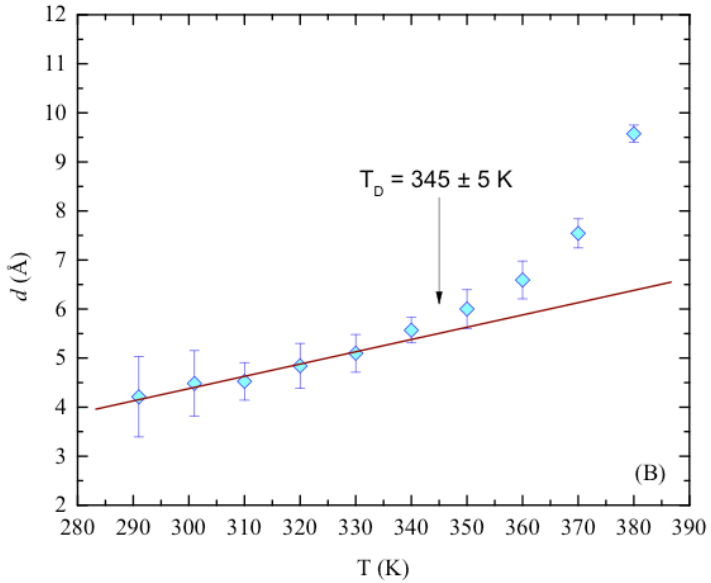
Analysis of Linewidth Based on Jump Diffusion Model

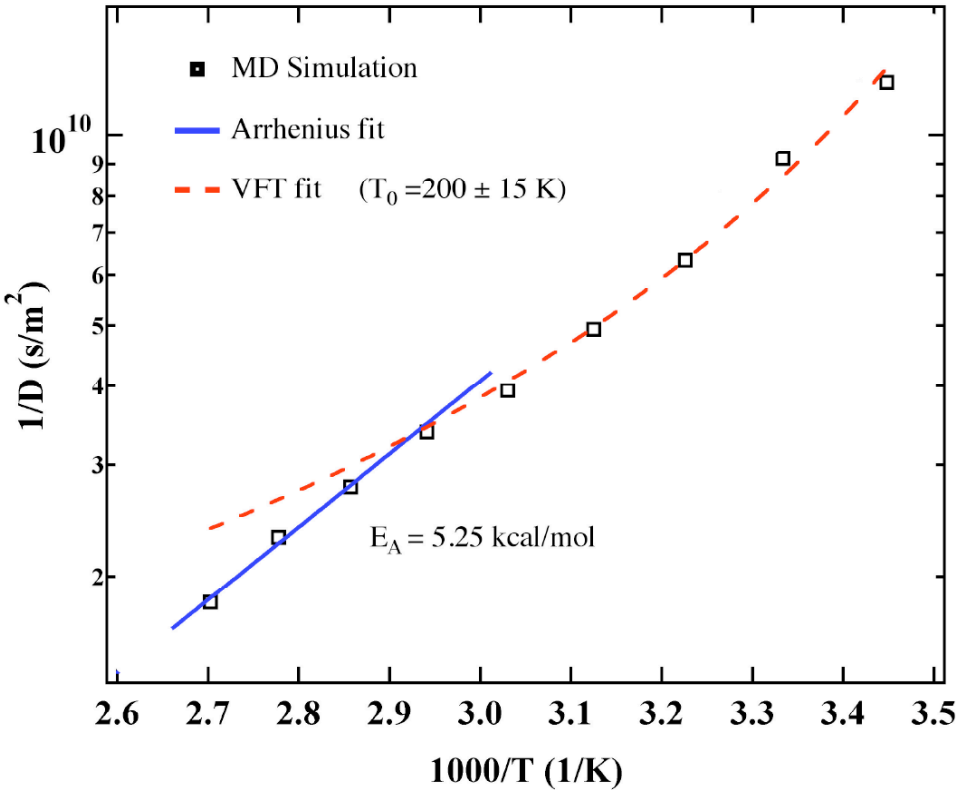


Y. Zhang, M. Lagi, D. Liu, F. Mallamace, E. Fratini, P. Baglioni, E. Mamontov, S.-H. Chen "Observation of high-temperature dynamic crossover in protein hydration water and its relation to reversible denaturation of lysozyme" submitted to JCP.

High-T Dynamic Crossover Phenomenon in Lysozyme Hydration Water

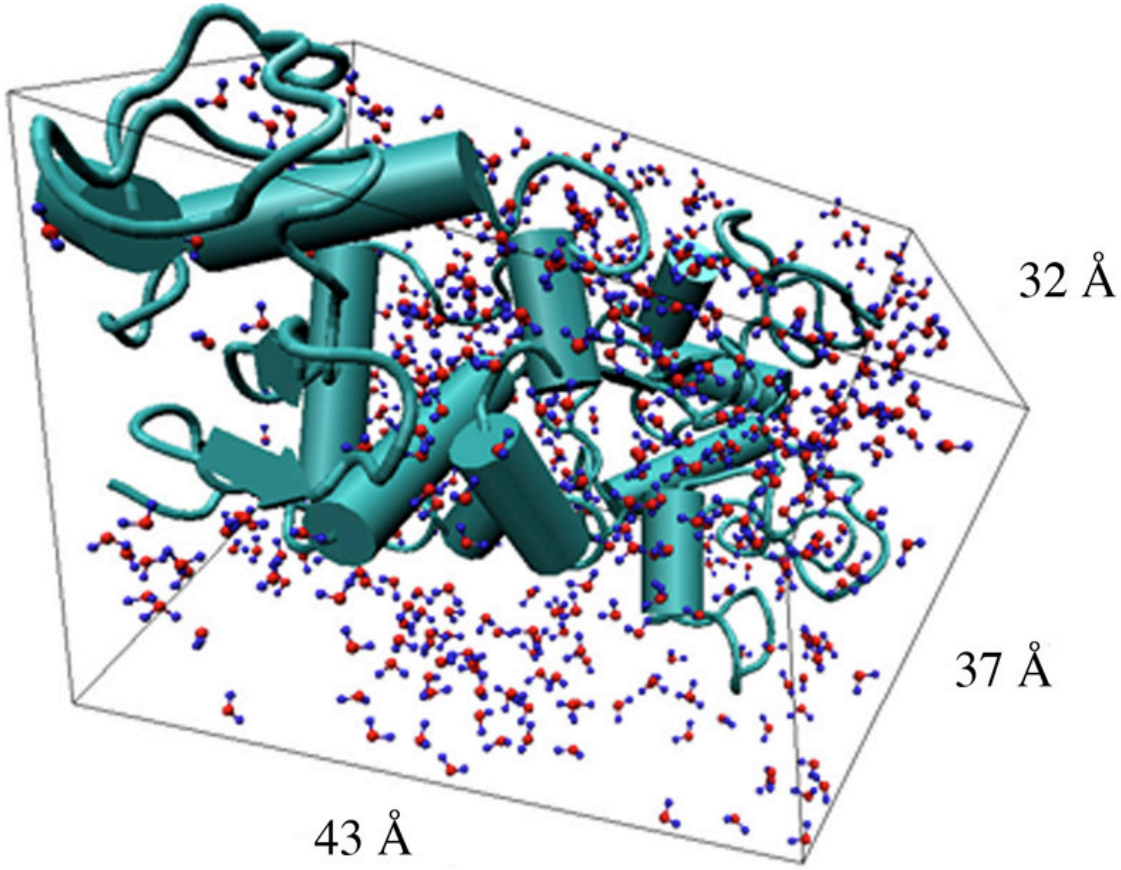






Y. Zhang, M. Lagi, D. Liu, F. Mallamace, E. Fratini, P. Baglioni, E. Mamontov, S.-H. Chen submitted to JCP.

MD Experimental Details



2 Lysozyme molecules

protein was modeled with OPLS-AA force field

484 water molecules

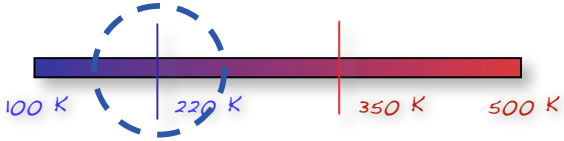
water was modeled with TIP4P-Ew force field

h = 0.3

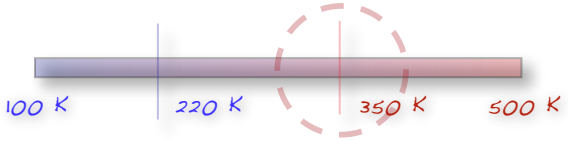
hydration level, grams protein/grams water

NPT ensemble

20 trajectories of 50 ns, from 180 to 380 K
8 CPU = 8 trajectories in about 2 weeks



MD Simulation



Molecular Dynamics Trajectories

Mean Square Deviation

$< \chi^2(t) > = < [r(t)-r(0)]^2 >$

$\lim_{t\to\infty} \langle X^2(t) \rangle = 2Dt$

Diffusion Constant

Intermediate Scattering Function

$ISF(Q,t) = < \exp(iQr(t)) \exp(-iQr(0)) >$

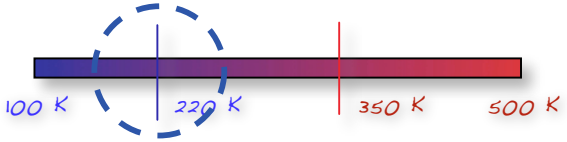
stretched exponential fit

Translational Relaxation Time

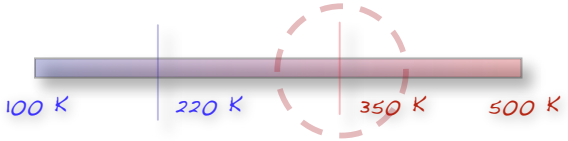
Stokes-Einstein Relation

$D \sim k_B T / \tau$

$D \tau / T \sim k_B$



MSD

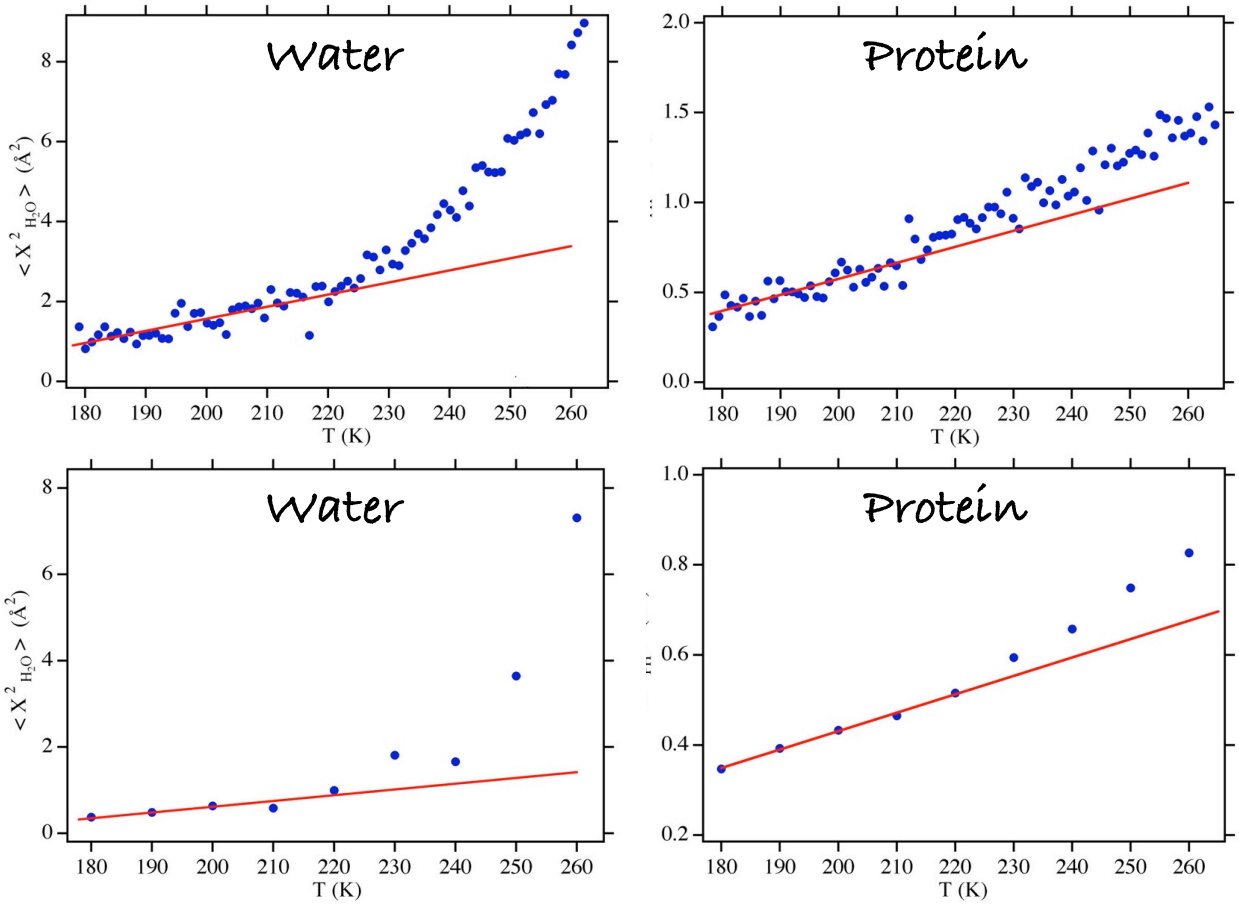


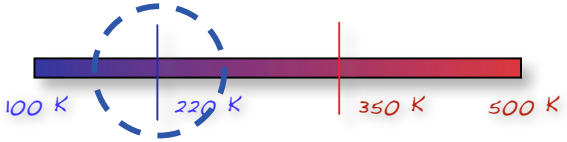
Mean Square Displacement

discontinuity in both water and protein mobility at 220 K

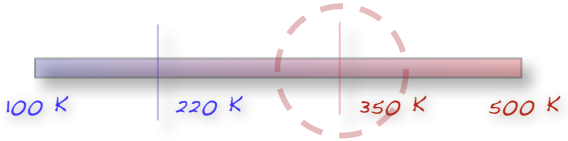
Neutron Scattering

Molecular Dynamics

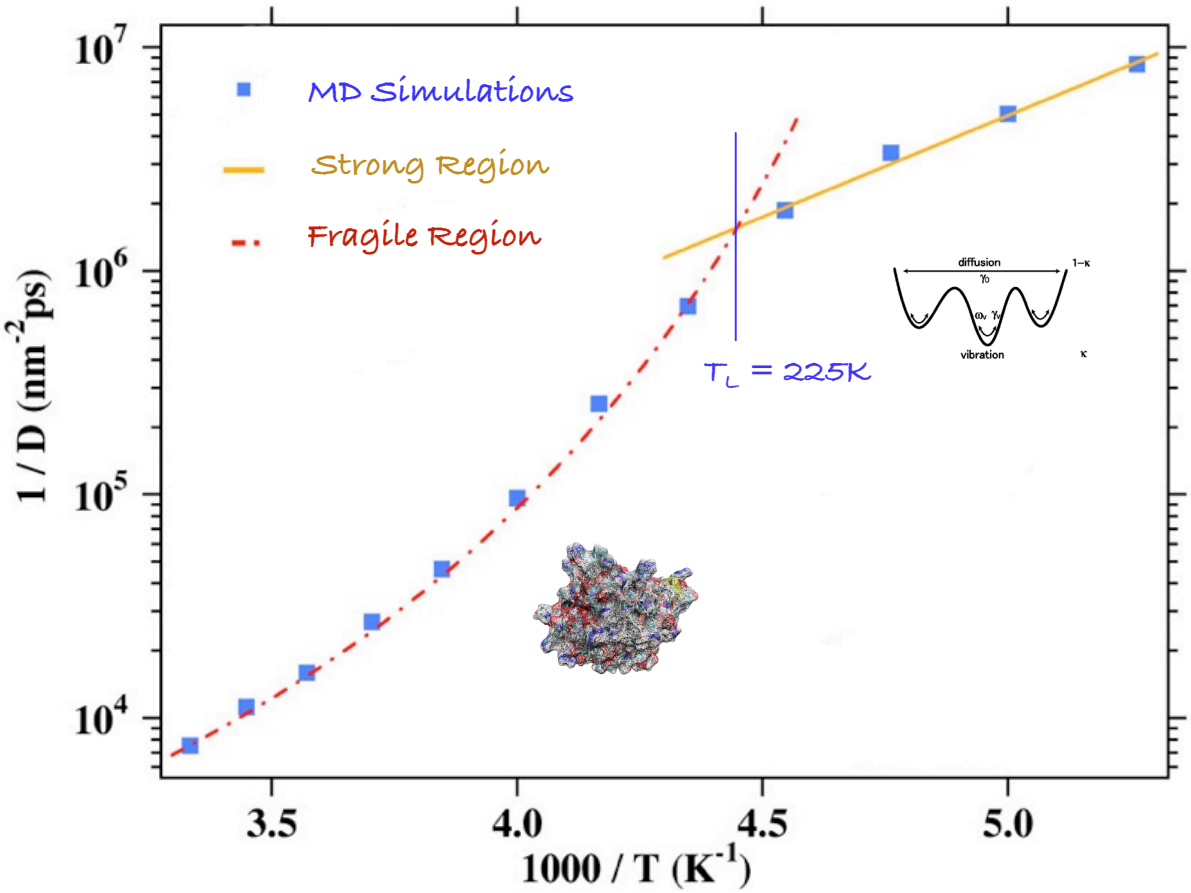




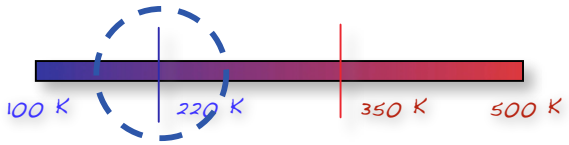
Water Diffusion Constant

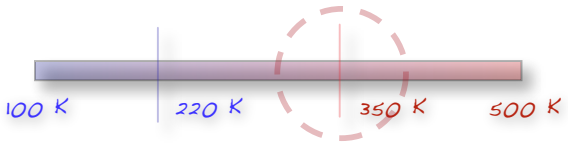


Fragile-to-Strong Crossover at T_L = 225 K

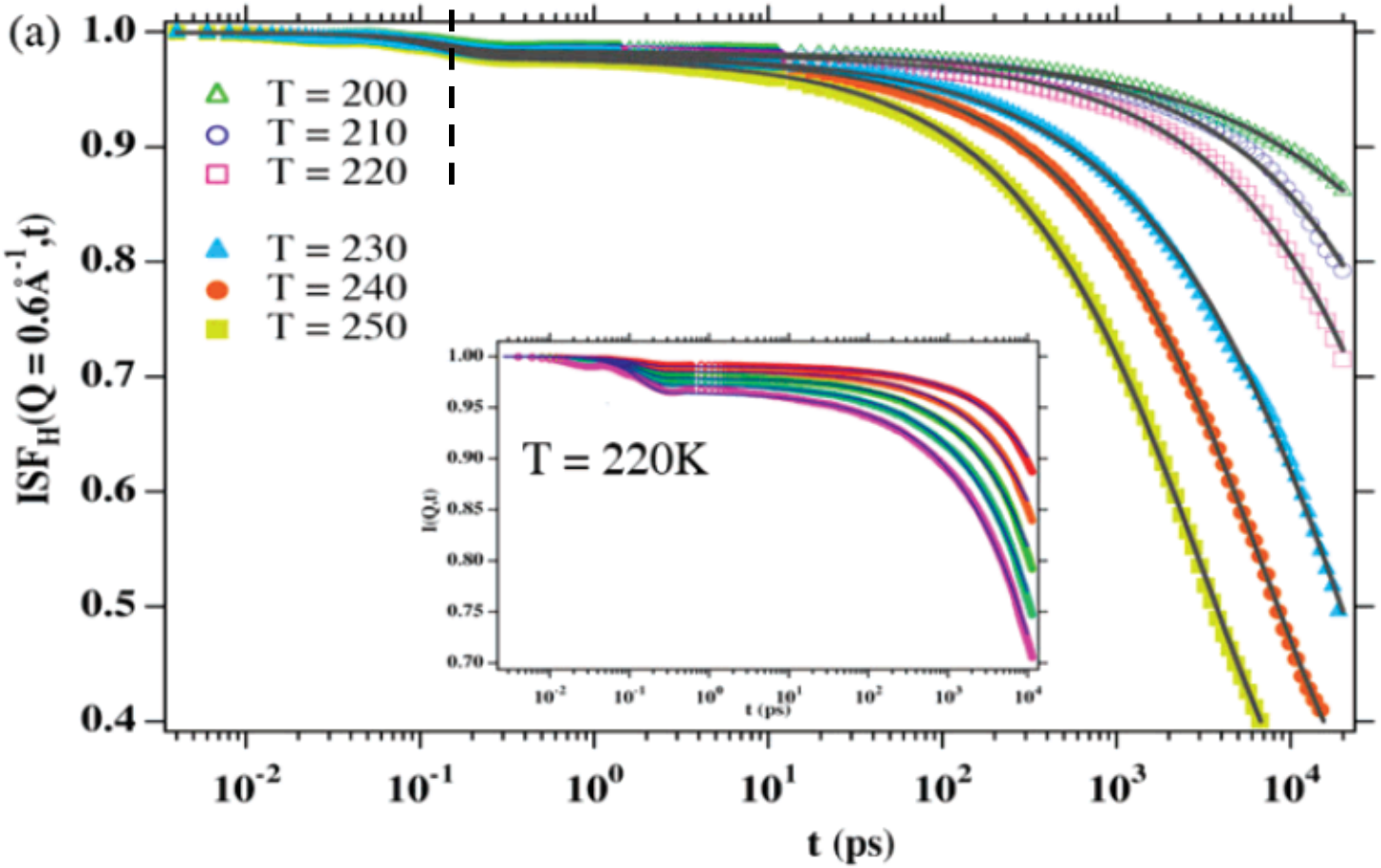


ISF from trajectories

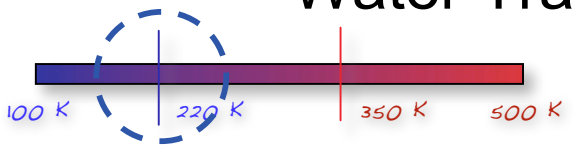




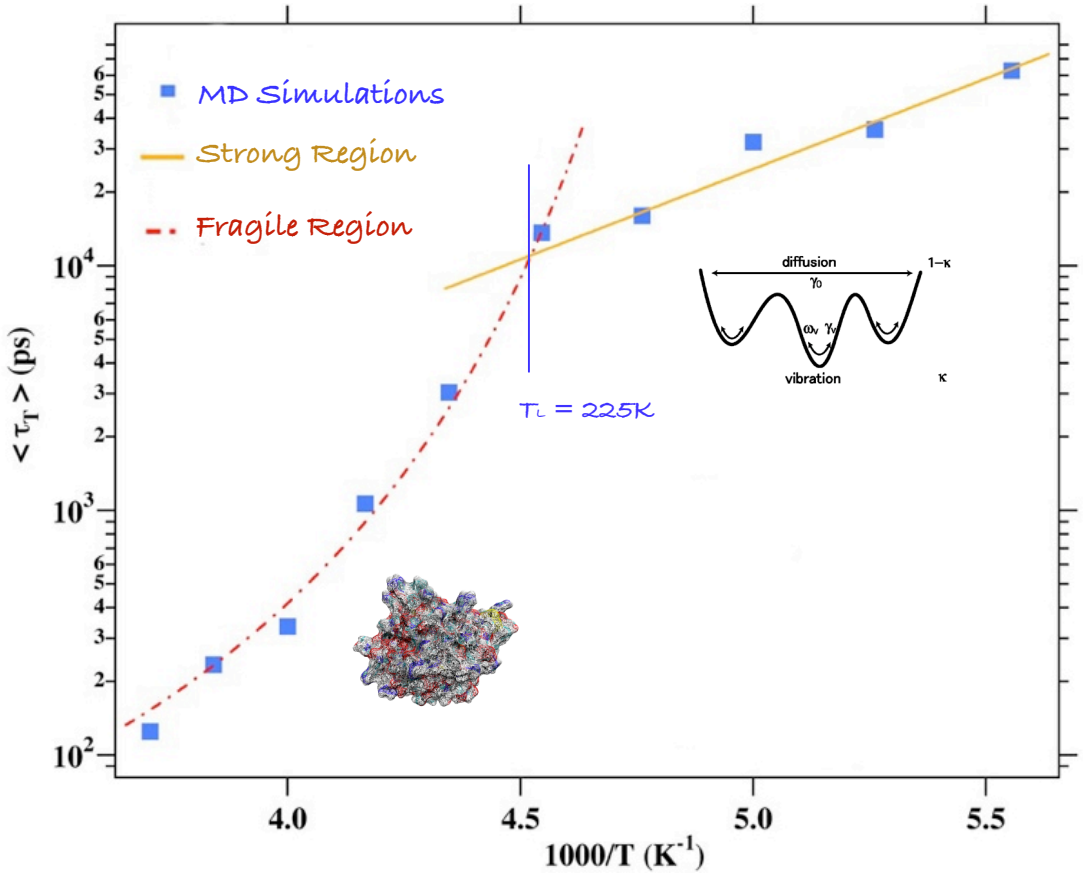
In cage motions | α-relaxation



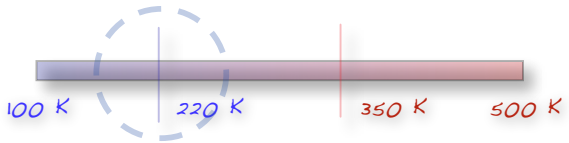
Water Translational Relaxation Time from ISF

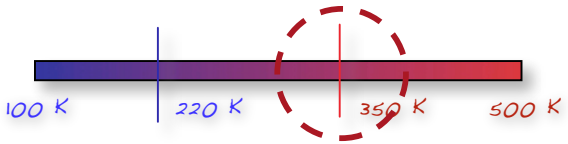


fragile-to-strong crossover at T_L = 223 K

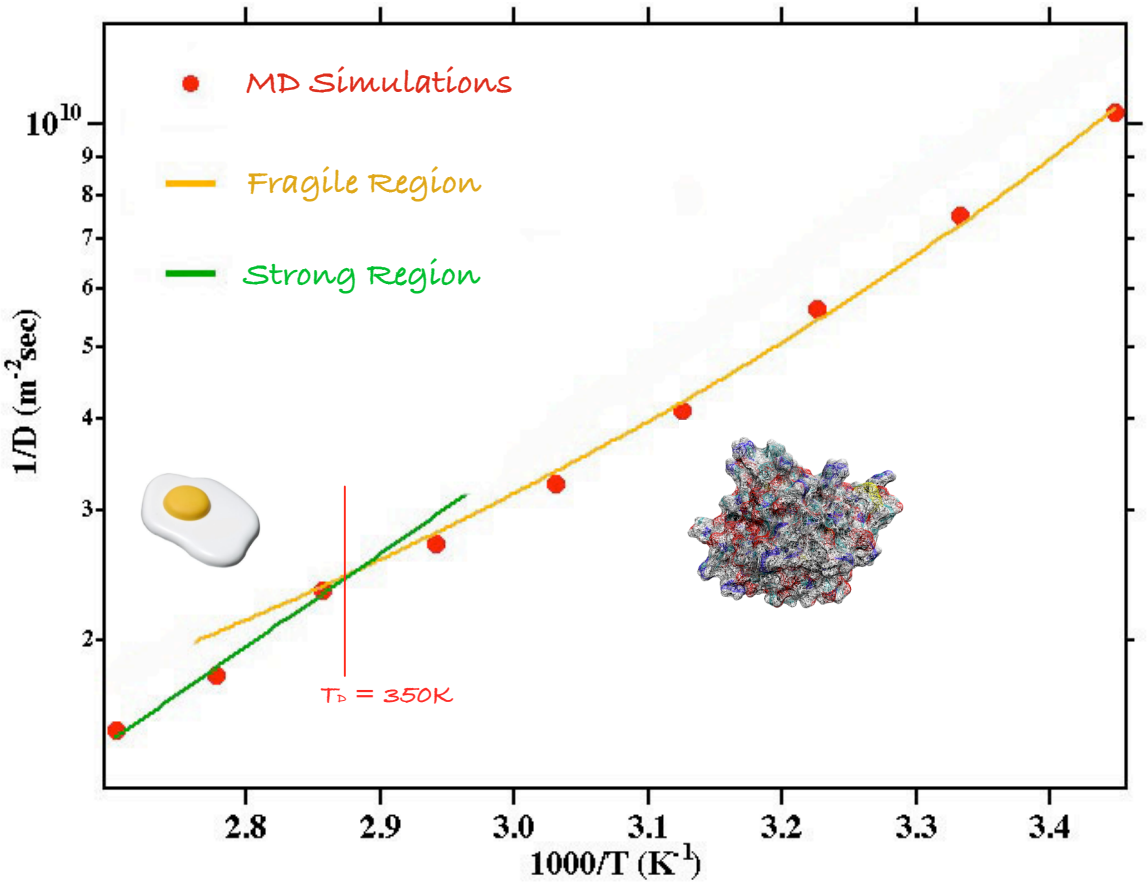


Water Diffusion Constant





Strong-to-Fragile Crossover at T_D = 350 K

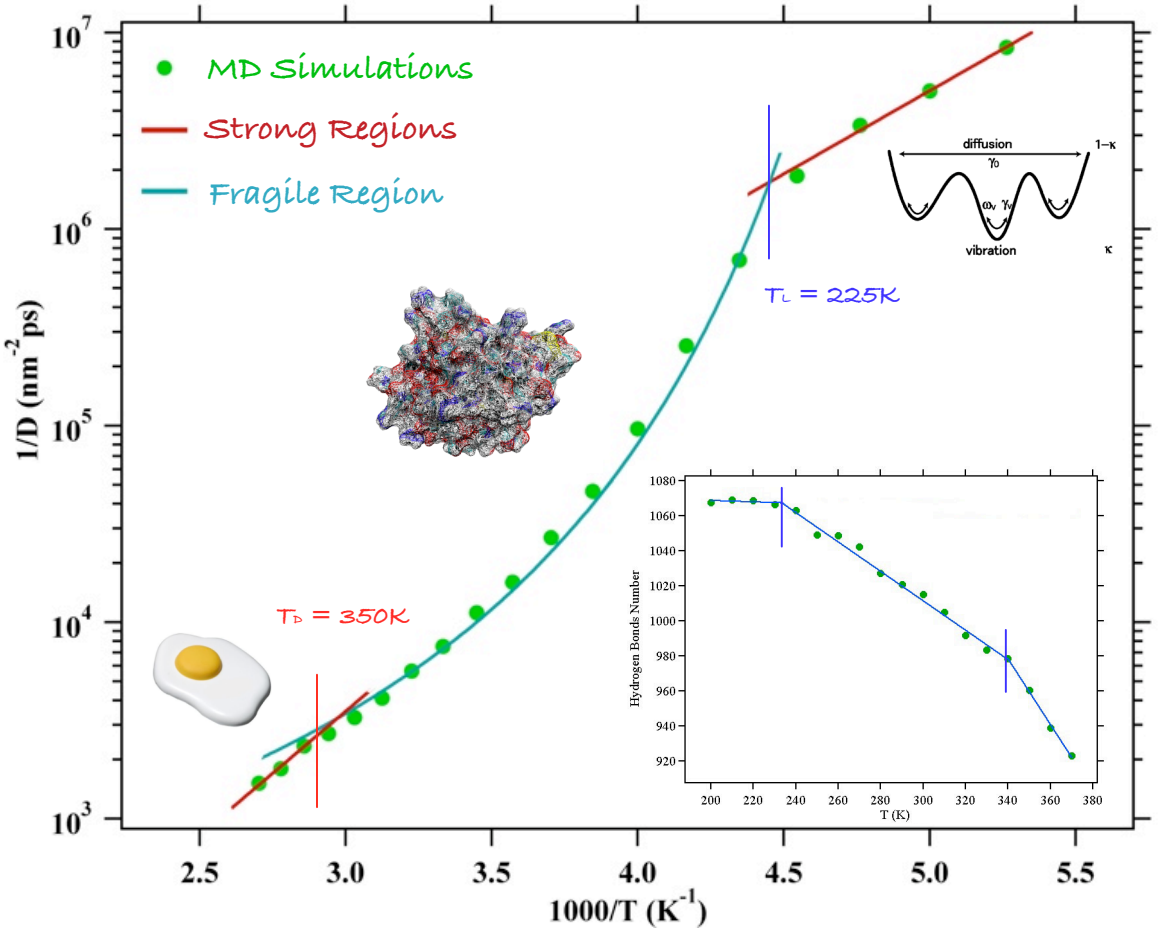


Conclusions

1. NMR studies of lysozyme hydration water show the existence of two dynamic crossovers in the protein hydration water.
2. Below the low-temperature crossover, at about 220 K, the hydration water displays a fragile-to-strong dynamic crossover, resulting in the loss of the protein conformational flexibility.
3. Above the high-temperature crossover, at about 346 K, where the protein unfolds, the dynamics of the hydration water appears to be dominated by the non-hydrogen-bonded fraction of water molecules.
4. Our recent experiment done in BASIS confirms the existence of the high temperature dynamic crossover in protein hydration water.

The Complete Picture

Strong-to-Fragile-to-Strong



Y. Zhang, M. Lagi, D. Liu, F. Mallamace, E. Fratini, P. Baglioni, E. Mamontov, S.-H. Chen "Observation of high-temperature dynamic crossover in protein hydration water and its relation to reversible denaturation of lysozyme" submitted to JCP.

Acknowledgments

- S.-H. Chen, MIT
- P. Baglioni, CSGI @ University of Florence
- M. Lagi, CSGI @ University of Florence & MIT
- C. Vannucchi, CSGI @ University of Florence
- A. Faraone, NCNR
- L. Liu, RPI
- E. Mamontov, ORNL
- F. Mallamace, University of Messina & MIT

Thank you for your attention!!!