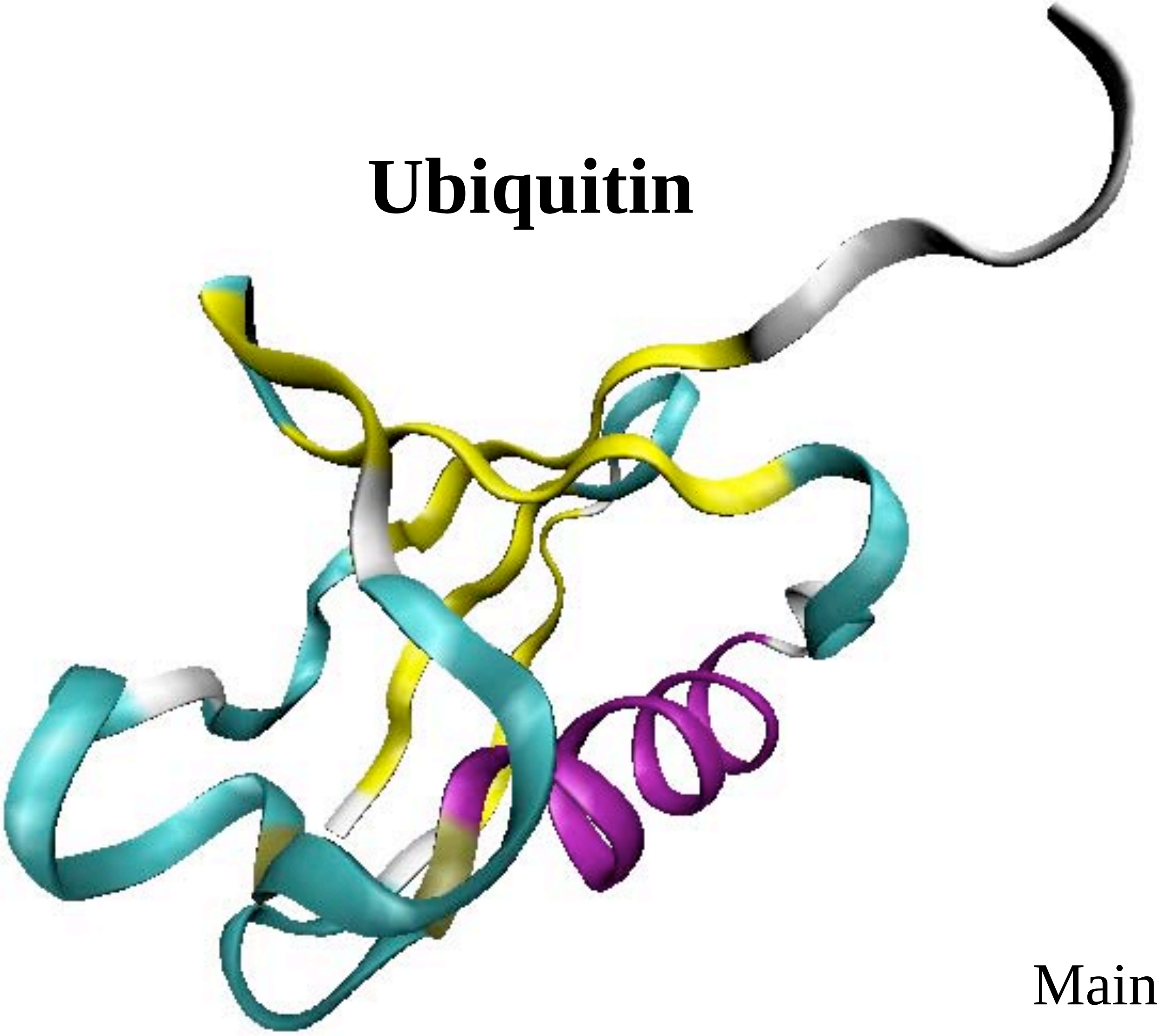
# Molecular Dynamics of Proteins



## Ubiquitin

Main funding:

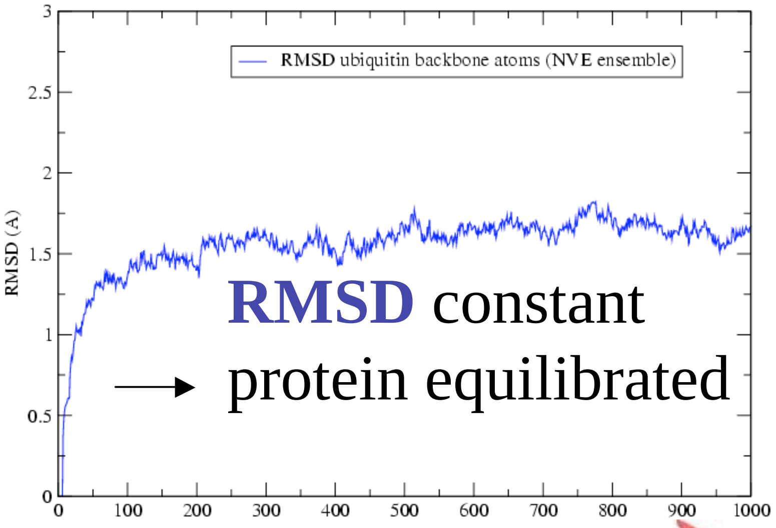
National Center for
Research Resources

# Equilibrium Properties of Proteins

## Ubiquitin

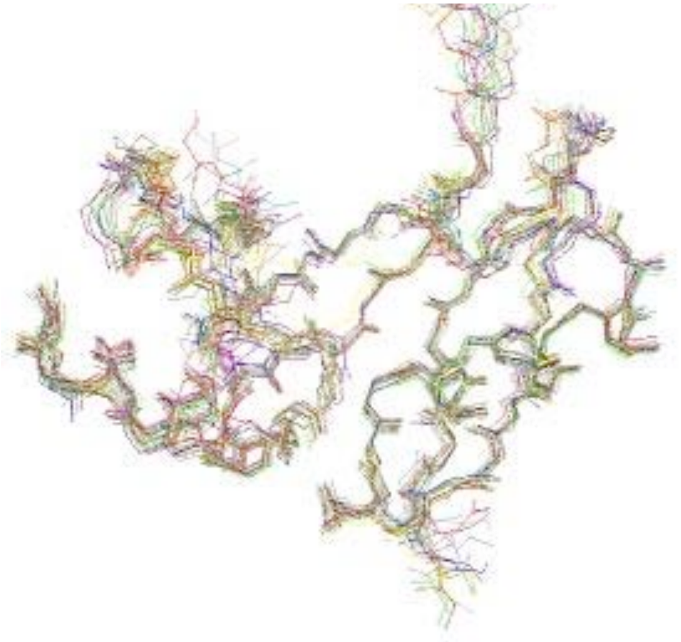
Root Mean Squared Deviation: measure for equilibration and protein flexibility

$$RMSD_\alpha = \sqrt{\frac{\sum_{j=1}^{N_t} \sum_{\alpha=1}^{N_\alpha} (\vec{r}_\alpha(t_j) - \langle \vec{r}_\alpha \rangle)^2}{N_\alpha}}$$

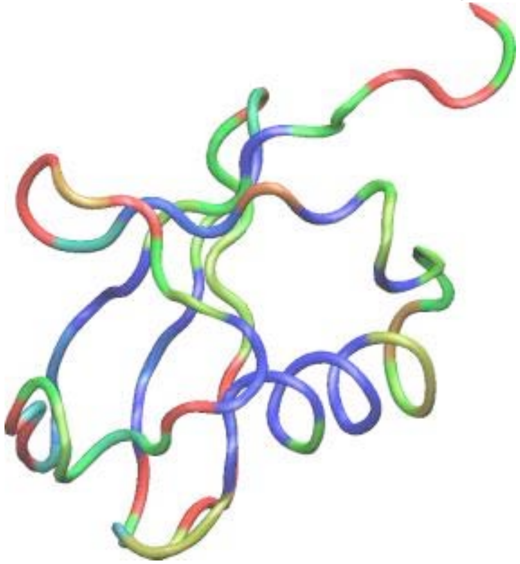


**RMSD** constant

→ protein equilibrated



**Protein sequence exhibits characteristic permanent flexibility!**



NMR structures

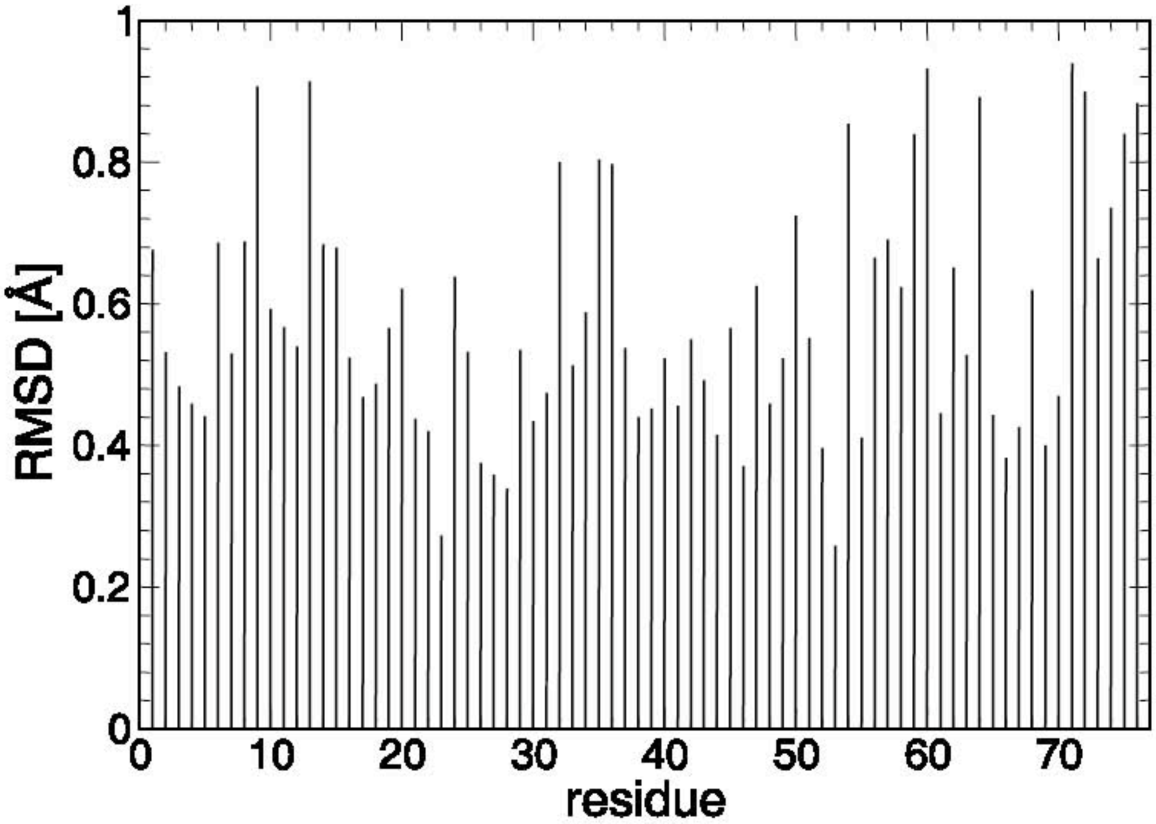
aligned together to see flexibility

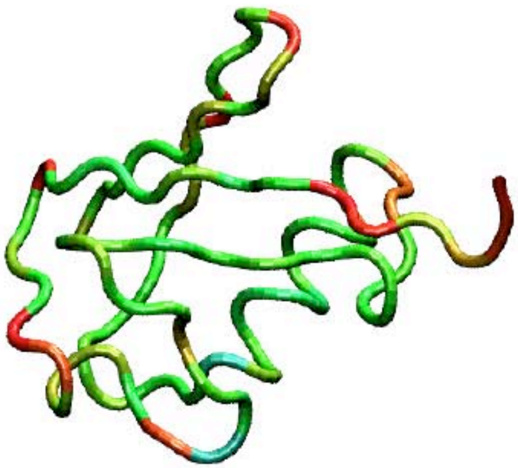
MD simulation

The color represents mobility of the protein through simulation (red = more flexible)

# Thermal Motion of Ubiquitin from MD

RMSD values per residue



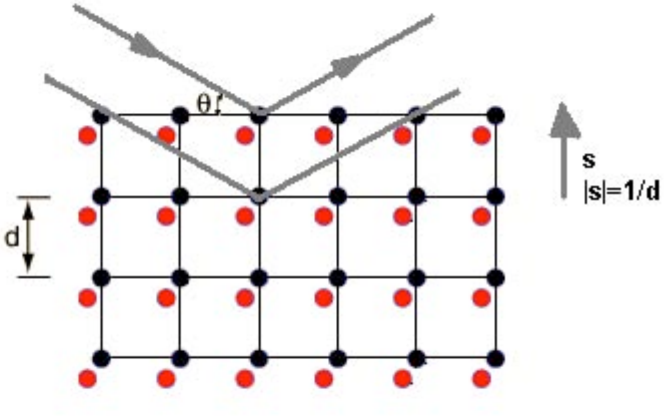


# Thermal Motion of Ubiquitin from MD

## *Temperature Dependence of Crystal Diffraction (Debye-Waller factor)*

Bragg's law

$$2\,d\,\sin\theta \;=\; \lambda$$



structure factor

$$f_j \exp[-i\vec{s}\cdot\vec{r}_j]$$

The diffraction signal is the sum of the structure factors of all atoms in the crystal.

But the atom carries out thermal vibrations around equilibrium position $\vec{x}_j$

$$\vec{r}_j(t) \;=\; \vec{x}_j \;+\; \vec{u}_j(t)$$

Accordingly:

$$\langle f_j \exp[-i\vec{s}\cdot\vec{r}_j]\rangle \;=\; f_j \exp[-i\vec{s}\cdot\vec{x}_j]\,\langle \exp[-i\vec{s}\cdot\vec{u}_j]\rangle$$

# Thermal Motion of Ubiquitin from MD

## *Temperature Dependence of Crystal Diffraction (Debye-Waller factor)*

One can expand:

$$\langle \exp[-i\vec{s}\cdot\vec{u}_j]\rangle = 1 - i\underbrace{\langle \vec{s}\cdot\vec{u}_j\rangle}_{=0} - \tfrac{1}{2}\langle(\vec{s}\cdot\vec{u}_j)^2\rangle \quad + \ldots$$

Spatial average: $\langle(\vec{s}\cdot\vec{u}_j)^2\rangle = \frac{1}{3}s^2\langle u_j^2\rangle$

One can carry out the expansion further and show

$$\langle \exp[-i\vec{s}\cdot\vec{u}_j]\rangle = \exp\left[-\tfrac{1}{6}s^2 \ \langle u_j^2\rangle\right]$$

Using for the thermal amplitude of the harmonic oscillator

$$\tfrac{1}{2}m\omega^2u_j^2 = \tfrac{3}{2}k_BT$$

one obtains

$$\langle f_j \exp[-i\vec{s}\cdot\vec{r}_j]\rangle = f_j \overbrace{\exp[-s^2k_BT/2m\omega^2]}^{\text{Debye-Waller factor}}\exp[-i\vec{s}\cdot\vec{x}_j]$$

# Equilibrium Properties of Proteins

## Energies: kinetic and potential

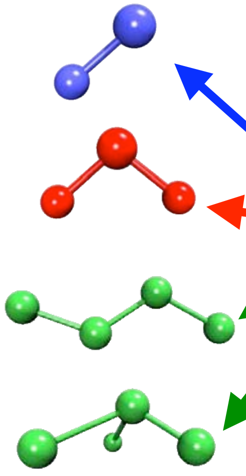
?

$$\leftarrow \langle \sum_j \frac{1}{2} m_j v_j^2 \rangle$$

Kinetic energy (quadratic)

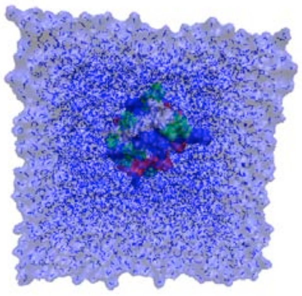
**temperature**

$\longrightarrow$



**from ubiquitin.psf**

```
26190   !NATOM:     atoms
17867    !NBOND:    bonds
10572    !NTHETA:   angles
 3293      !NPHI:      dihedrals
  204     !NIMPHI:    impropers
```

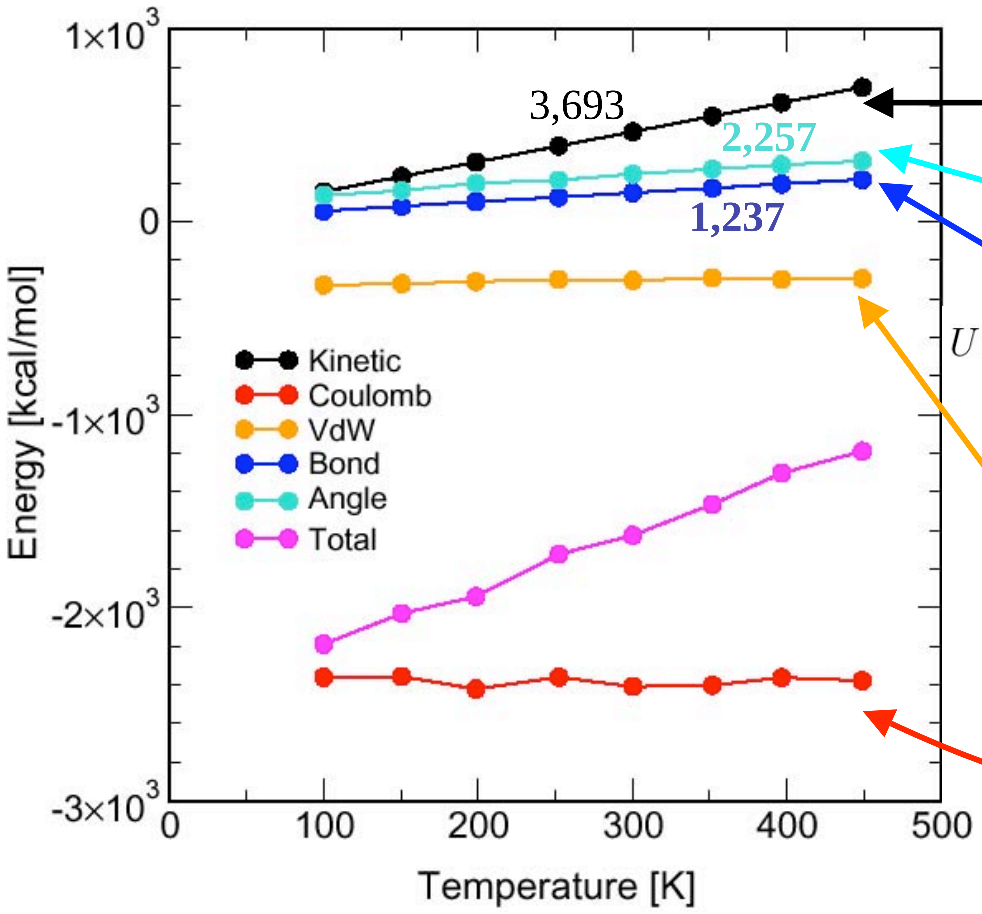


$$U(\vec{R}) = \underbrace{\sum_{bonds} k_i^{bond}(r_i - r_0)^2}_{U_{bond}} + \underbrace{\sum_{angles} k_i^{angle}(\theta_i - \theta_0)^2}_{U_{angle}} + \underbrace{\sum_{dihedrals} k_i^{dihe}[1 + \cos(n_i\phi_i + \delta_i)]}_{U_{dihedral}} + \underbrace{\sum_i \sum_{j \neq i} 4\epsilon_{ij}\left[\left(\frac{\sigma_{ij}}{r_{ij}}\right)^{12} - \left(\frac{\sigma_{ij}}{r_{ij}}\right)^{6}\right] + \sum_i \sum_{j \neq i} \frac{q_i q_j}{\epsilon r_{ij}}}_{U_{nonbond}}$$

Potentialc energy (not all quadratic)

# Equilibrium Properties of Proteins

## Energies: kinetic and potential



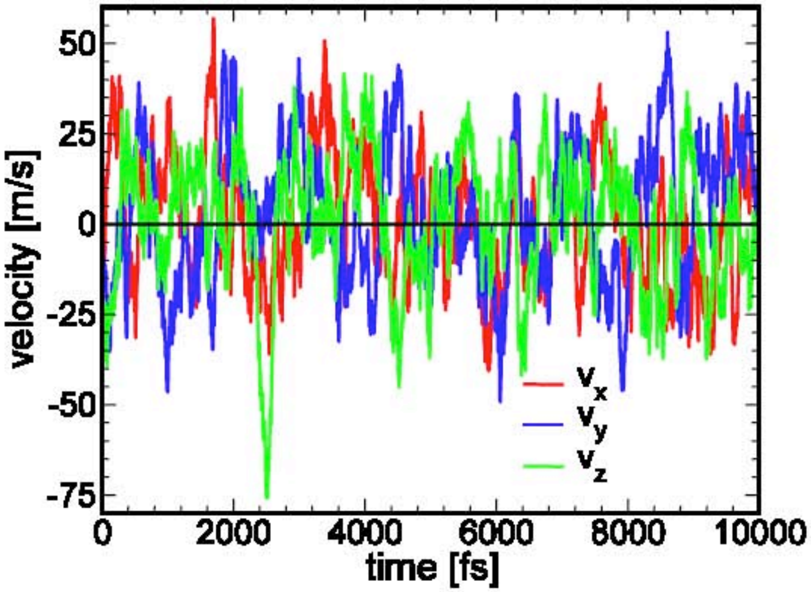
$$\langle \sum_j \frac{1}{2} m_j v_j^2 \rangle = \frac{3}{2} N\, k_B\, T$$

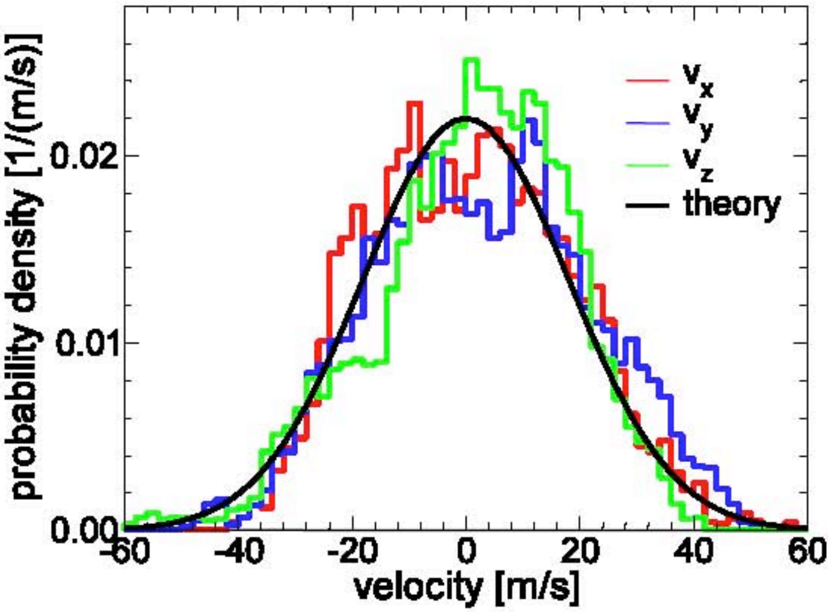
Kinetic energy (quadratic)

$$U(\vec{R}) = \underbrace{\sum_{bonds} k_i^{bond}(r_i - r_0)^2}_{U_{bond}} + \underbrace{\sum_{angles} k_i^{angle}(\theta_i - \theta_0)^2}_{U_{angle}} + \underbrace{\sum_{dihedrals} k_i^{dihe}[1 + \cos(n_i \phi_i + \delta_i)]}_{U_{dihedral}} + \underbrace{\sum_i \sum_{j \neq i} 4\epsilon_{ij}\left[\left(\frac{\sigma_{ij}}{r_{ij}}\right)^{12} - \left(\frac{\sigma_{ij}}{r_{ij}}\right)^{6}\right] + \sum_i \sum_{j \neq i} \frac{q_i q_j}{\epsilon r_{ij}}}_{U_{nonbond}}$$

Potentialc energy (not all quadratic)

# Maxwell Distribution of Atomic Velocities





$$p(v_\sigma) \;=\; \sqrt{\frac{m}{2\pi k_B T}}\;\exp\left[-\frac{m v_\sigma^2}{2 k_B T}\right]$$

$$\sigma \;=\; x,\; y,\; z$$

# Mean Kinetic Energy

## *Exercise in Statistics*

$$\left\langle \tfrac{1}{2} m v^2 \right\rangle \;=\; \int_{-\infty}^{\infty} dv \left(\tfrac{1}{2} m v^2\right)\; p(v)$$

$$=\; \sqrt{\frac{m}{2\pi k_B T}} \int_{-\infty}^{\infty} dv \left(\tfrac{1}{2} m v^2\right)\; \exp\left[-\frac{m v^2}{2 k_B T}\right]$$

$$=\; k_B T\; \sqrt{\frac{1}{\pi}} \int_{-\infty}^{\infty} \sqrt{\frac{m}{2 k_B T}}\; dv \left(\frac{m v^2}{2 k_B T}\right)\; \exp\left[-\frac{m v^2}{2 k_B T}\right]$$

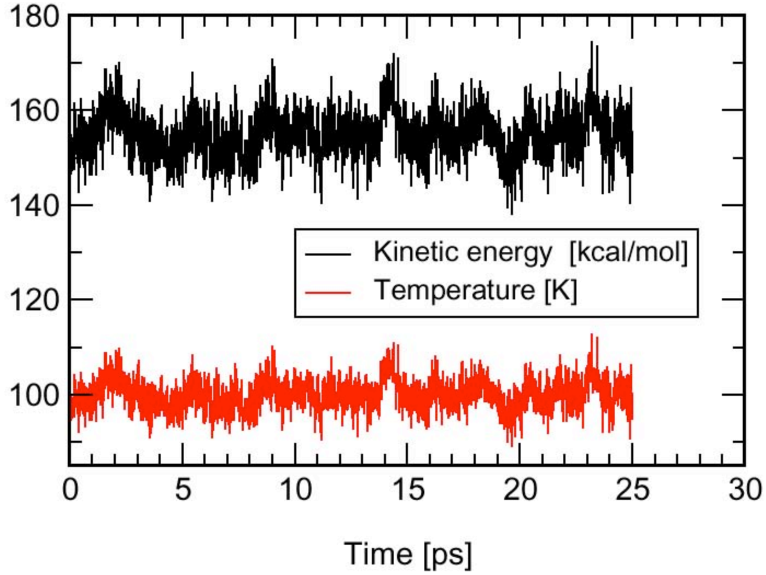
$$=\; k_B T\; \sqrt{\frac{1}{\pi}} \int_{-\infty}^{\infty} dy\, y^2 \exp - y^2]$$

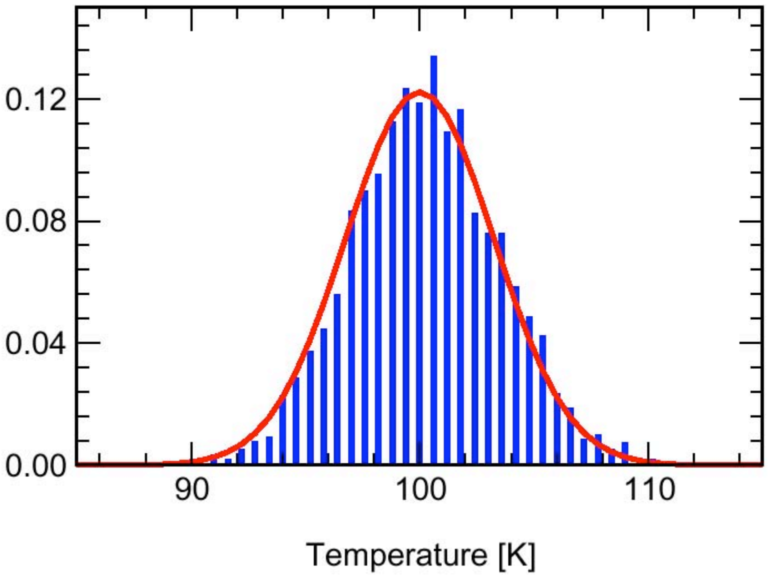
Use formula below: $\left\langle \tfrac{1}{2} m v^2 \right\rangle \;=\; \tfrac{1}{2} k_B T$

$$\int_0^{\infty} dy\, y^m\; \exp[-y^2] \;=\; \tfrac{1}{2}\, \Gamma\left(\tfrac{m+1}{2}\right)$$

$$\Gamma(x+1) \;=\; x\, \Gamma(x)\;, \quad \Gamma\left(\tfrac{1}{2}\right) \;=\; \sqrt{\tfrac{1}{2}}$$

# Analysis of $E_{kin}$, T (free dynamics)





## Definition of Temperature

$$\langle \sum_j \frac{1}{2} m_j v_j^2 \rangle \;=\; \frac{3}{2} N\, k_B\, T$$

$$T \;=\; \frac{2}{3N\, k_B}\, \langle \sum_j \frac{1}{2} m_j v_j^2 \rangle$$

**The atomic velocities of a protein establish a thermometer, but is it accurate?**

# Maxwell Kinetic EnergyDistribution

## *Second Exercise in Statistics*

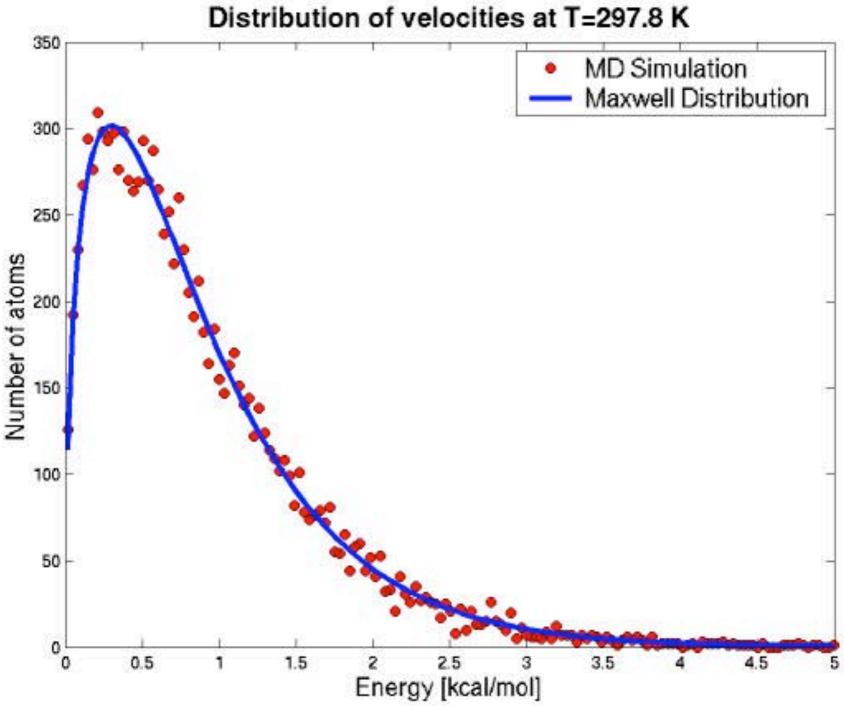
One-dimensional kinetic energy: $\epsilon_k \;=\; \frac{1}{2}mv_\sigma^2$

$$\tilde{p}(\epsilon_k) \;=\; p(v_\sigma)\frac{dv_\sigma}{d\epsilon_k} \;\longrightarrow\; \tilde{p}(\epsilon_k) \;=\; \sqrt{1/\pi k_B T}\sqrt{1/\epsilon_k}\;\exp[-\epsilon_k/k_BT]$$

(factor 2 from restriction of integration to positive values)

For the total kinetic energy
(in three dimensions)
holds then

$$\tilde{p}(\epsilon_k) \;=\; \frac{4}{\sqrt{\pi}}\,(k_BT)^{-\frac{3}{2}}\sqrt{\epsilon_k}\;\exp[-\epsilon_k/k_BT]$$



# Temperature Fluctuations

**The atomic velocity thermometer is inaccurate due to the finite size of a protein!**

Maxwell distribution

$$dP(v_n) \;=\; c\,\exp(-m\,v_n^2/2k_BT)\,dv_n \tag{7}$$

Individual kinetic energy $\epsilon_n \;=\; mv_n^2/2$

$$dP(\epsilon_n) \;=\; (\pi T_0\epsilon_n)^{-1/2}\exp(-\epsilon_n/k_BT_0)\,d\epsilon_n \tag{8}$$

One can derive

$$\langle\epsilon_n\rangle \;=\; T_0/2 \tag{9}$$

$$\langle\epsilon_n^2\rangle \;=\; 3\,T_0^2/4 \tag{10}$$

$$\langle\epsilon_n^2\rangle - \langle\epsilon_n\rangle^2 \;=\; T_0^2/2 \tag{11}$$

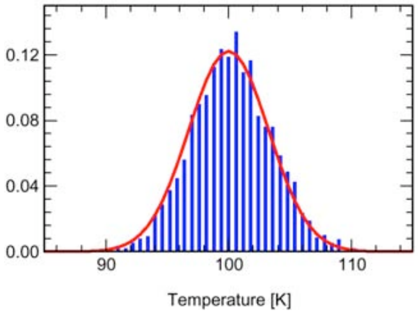
The distribution of the total kinetic energy $E_{kin} \;=\; \sum_j \frac{1}{2}m_jv_j^2$, according to the central limit theorem, is approximately Gaussian

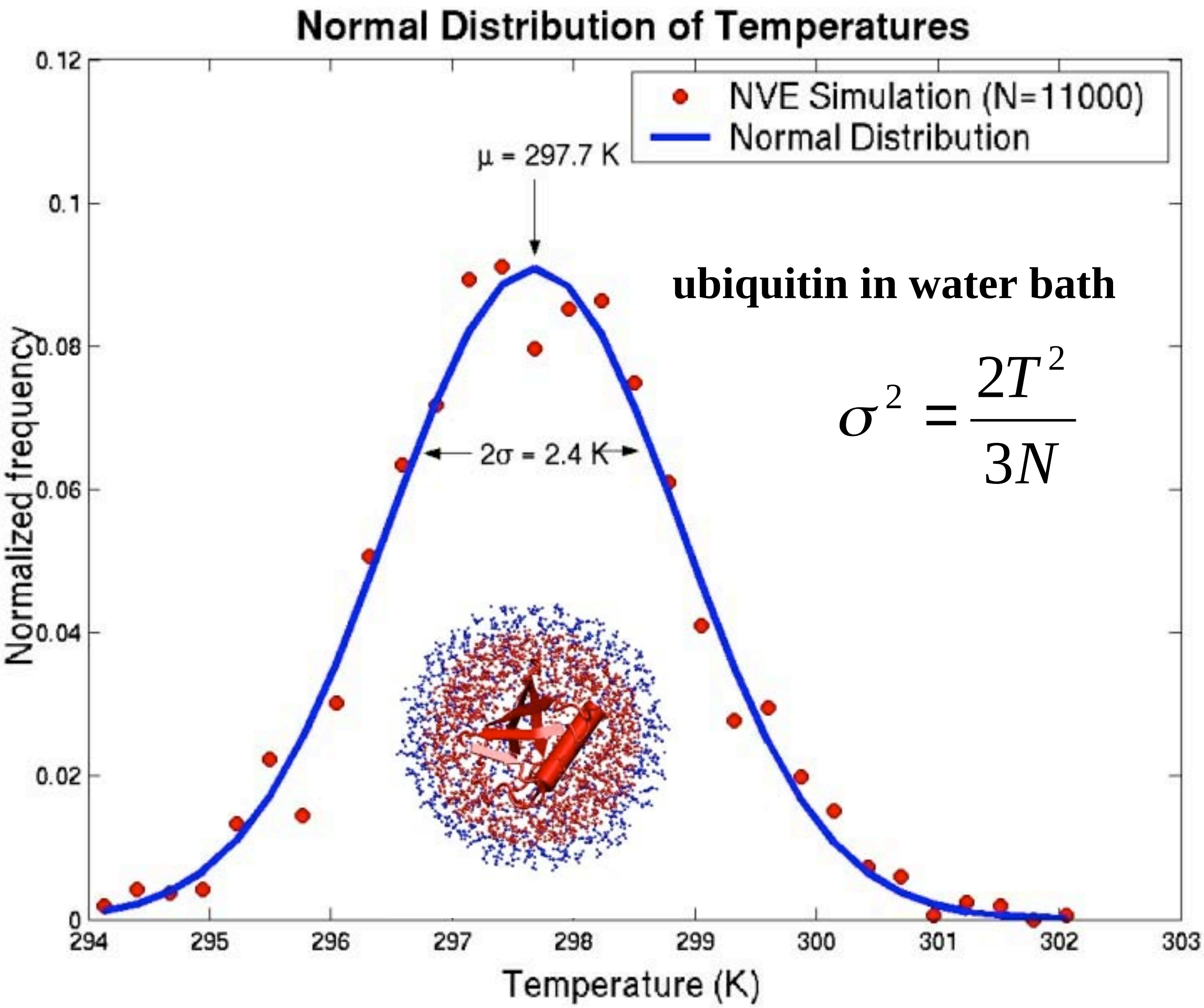
$$P(E_{kin}) \;=\; c\;\exp\left(\frac{-(E_{kin} - \langle E_{kin}\rangle)^2}{2\left(\frac{3Nk_B^2T_0^2}{2}\right)}\right) \tag{12}$$

The distribution function for the temperature ($T \;=\; 2E_{kin}/3k_B$) fluctuations $\Delta T \;=\; T - T_0$ is then

$$P(\Delta T) \;=\; c\,\exp[-(\Delta T)^2/2\sigma^2]\,, \qquad \sigma^2 \;=\; 2\,T^2/3N \tag{13}$$

For $T_0 \;=\; 100$K and $N \;=\; 557$, this gives $\sigma \;=\; 3.6$.





**ubiquitin in water bath**

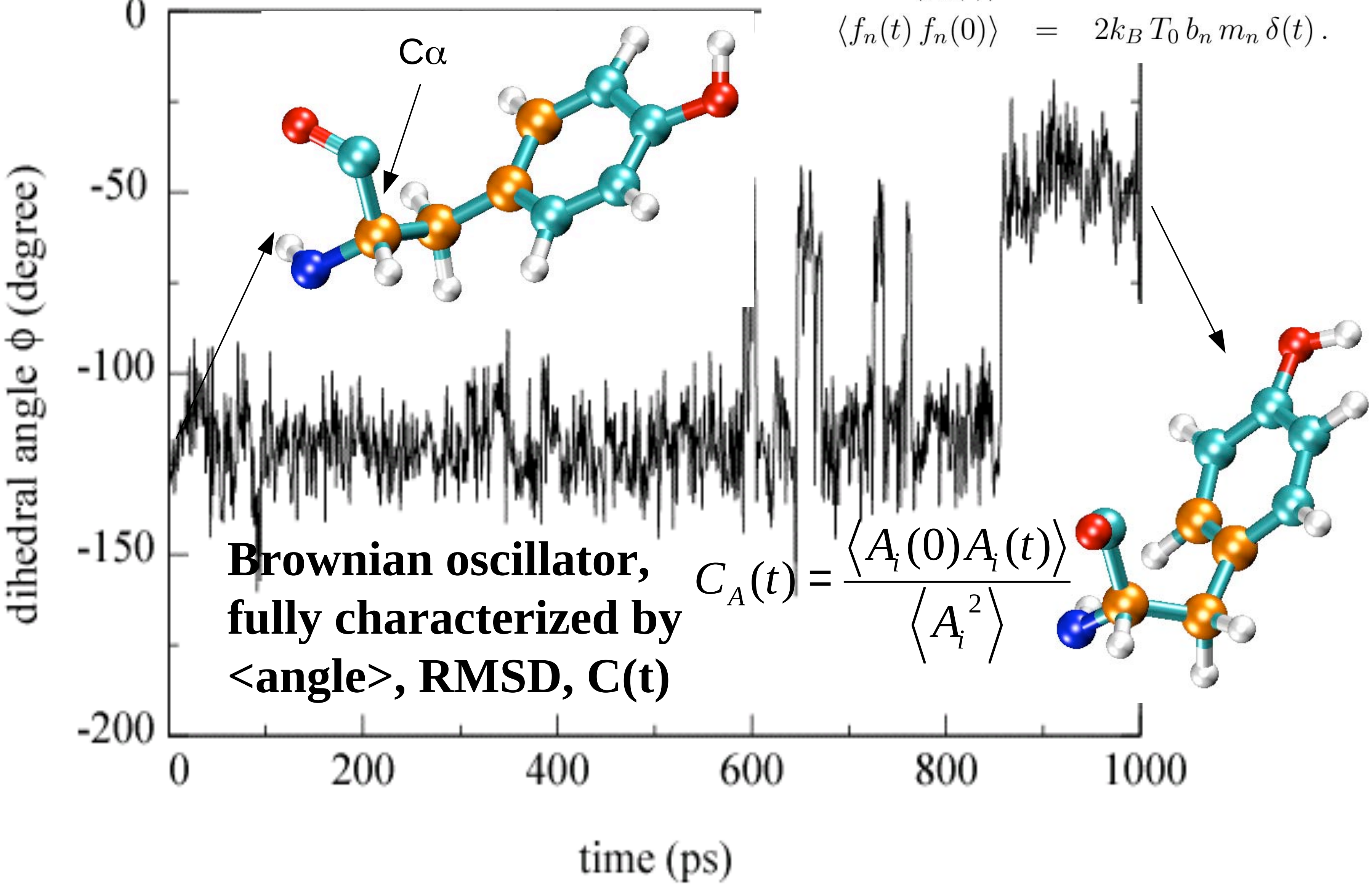
$$\sigma^2 = \frac{2T^2}{3N}$$

# Dihedral Angle

$$m_n \frac{d^2 x_n}{dt^2}(t) \;=\; \nabla_{x_n} V \;-\; m_n b_n \frac{dx_n}{dt}(t) \;+\; f_n(t)$$

Langevin dynamics in strong friction limit

$$\begin{aligned} \langle f_n(t) \rangle &= 0 \\ \langle f_n(t)\, f_n(0) \rangle &= 2 k_B\, T_0\, b_n\, m_n\, \delta(t)\,. \end{aligned}$$

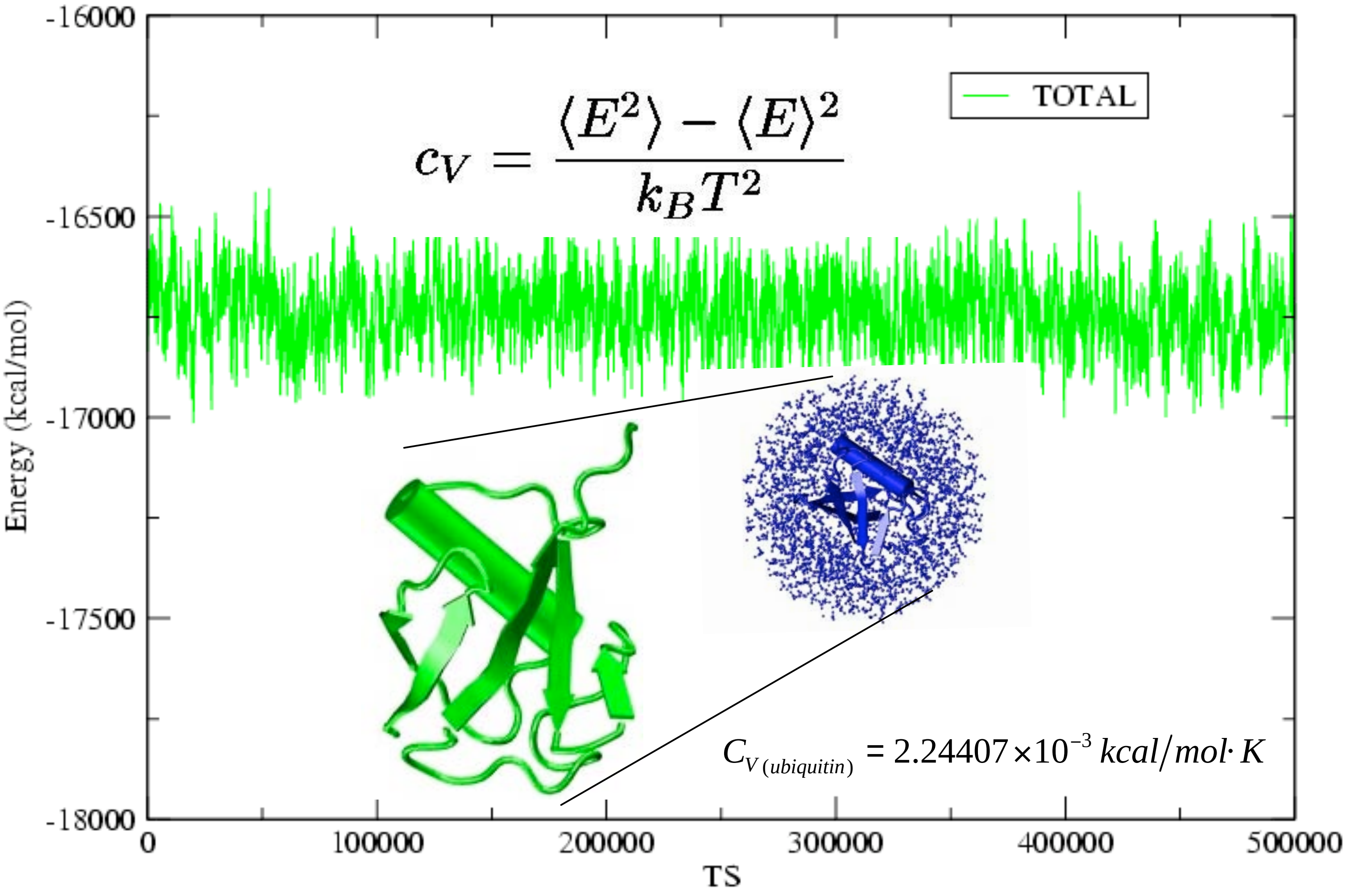


**Brownian oscillator, fully characterized by <angle>, RMSD, C(t)**

$$C_A(t) = \frac{\langle A_i(0) A_i(t) \rangle}{\langle A_i^2 \rangle}$$

# Specific Heat of a Protein

Total energy of ubiquitin (NVE ensemble)



$$c_V = \frac{\langle E^2 \rangle - \langle E \rangle^2}{k_B T^2}$$

$$C_{V\,(ubiquitin)} = 2.24407 \times 10^{-3}\, kcal/mol \cdot K$$