



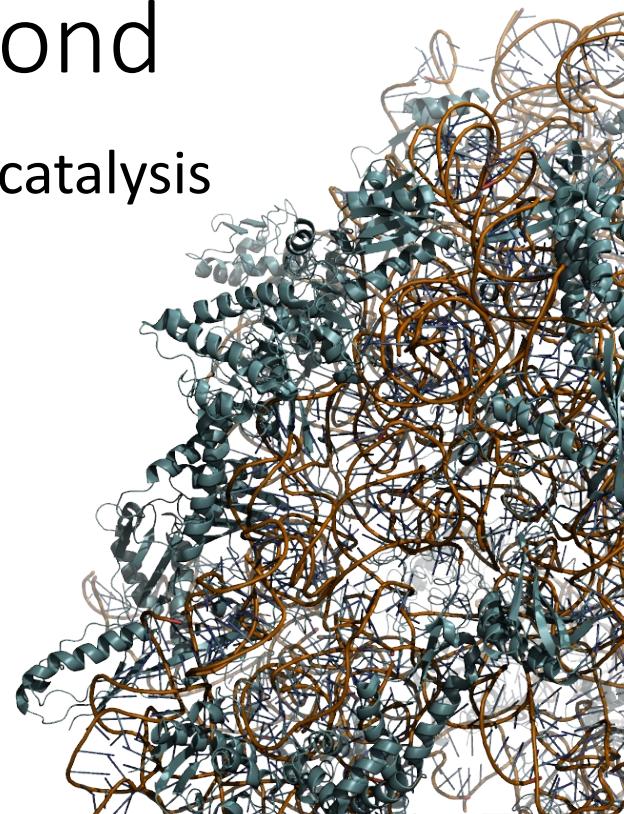
# Empirical Valence Bond

Molecular modeling of enzymatic catalysis

Biocat C8

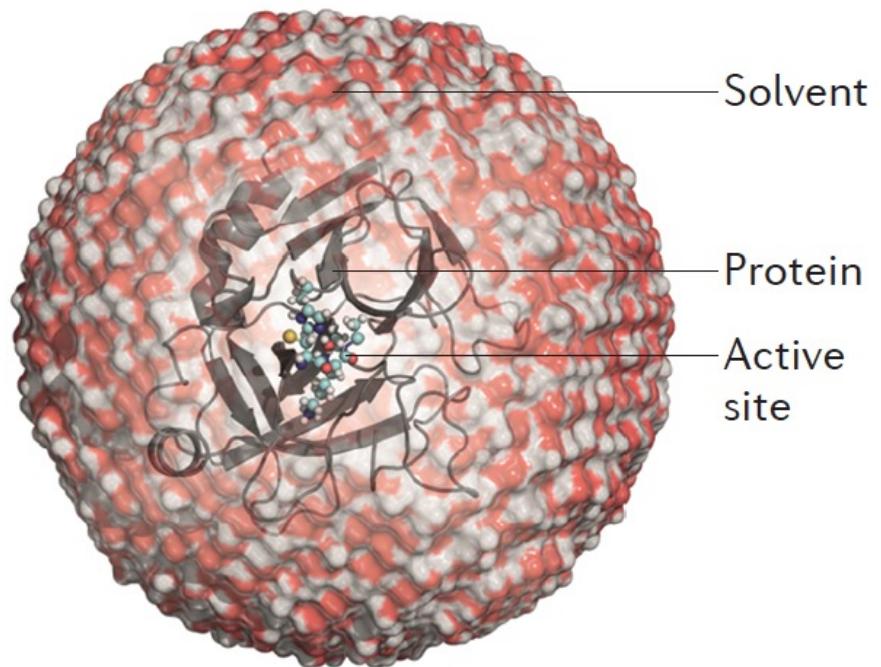
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# Outline

- What is EVB?
- Theory
- Examples / Qgui



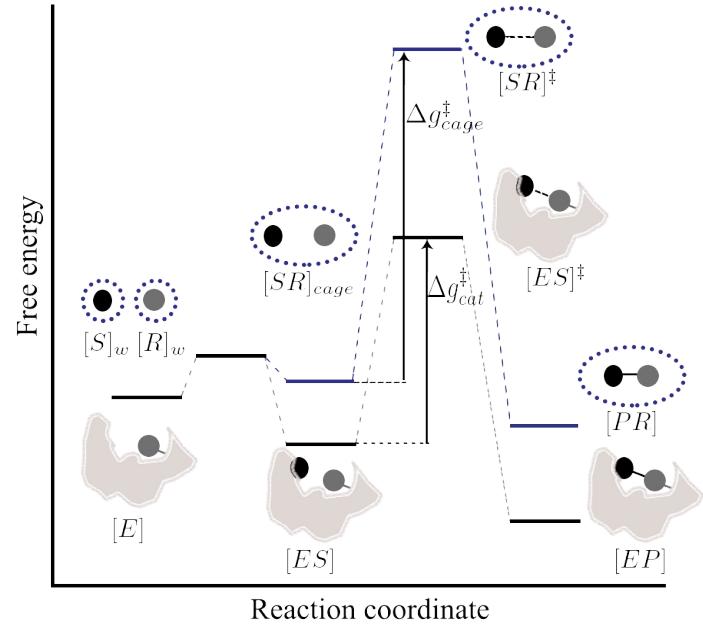
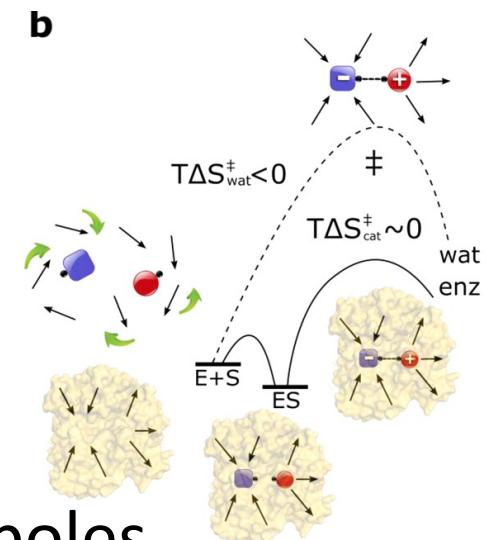
# 2 important findings with EVB

# Origin of enzyme catalysis

- Electrostatic stabilisation of  $[ES]^{\ddagger}$
- Polar solvents need to reorient their dipoles

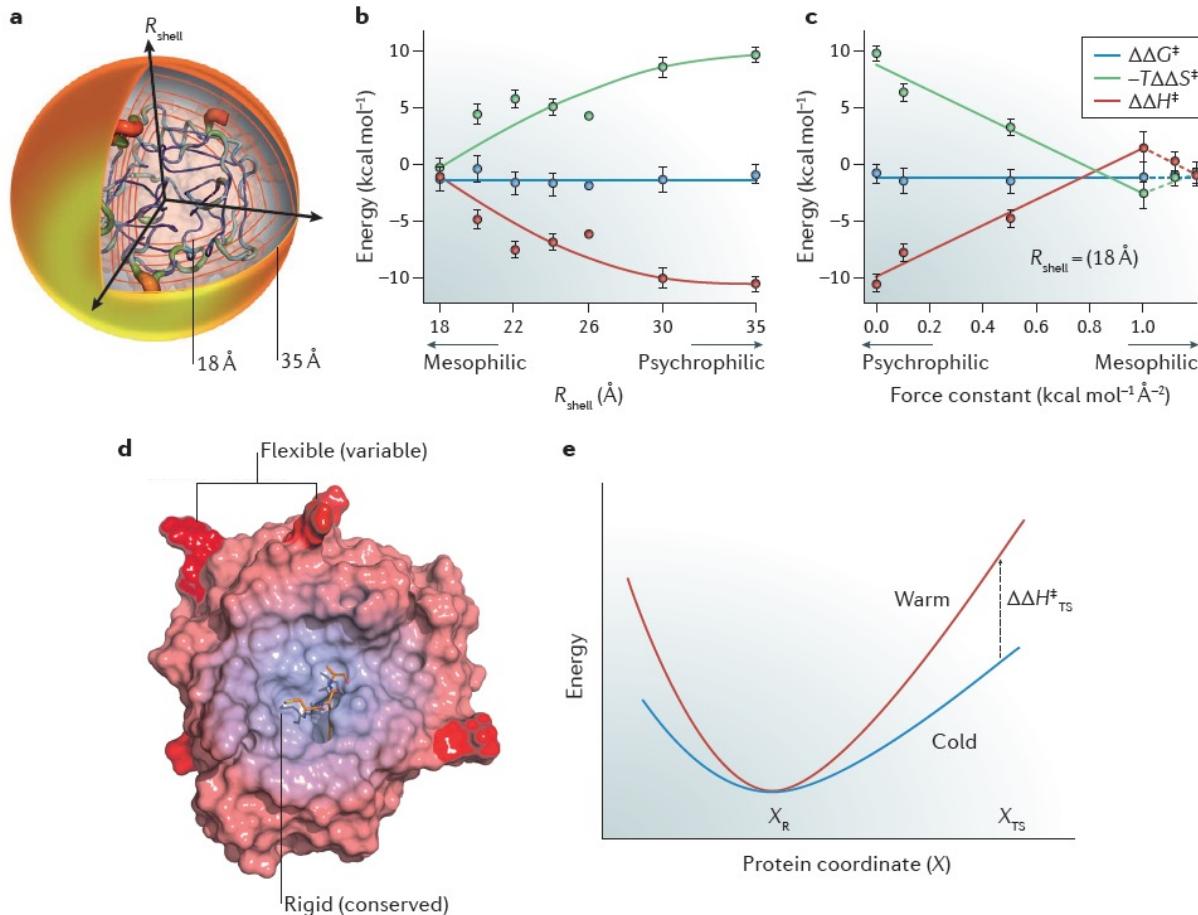
$$\Delta G_{\text{sol}} \cong \langle \Delta U_{q\mu} \rangle + \langle \Delta U_{\mu\mu} \rangle \cong \frac{1}{2} \langle \Delta U_{q\mu} \rangle$$

- $\Delta U_{\mu\mu}$  repulsion paid in folding
- → Reduced reorganization E
- → Enzymes are preorganized
  - $[ES]^{\ddagger}$



\* Warshel A (1978) "Energetics of Enzyme Catalysis" Proc Nat Acad USA 75(11):5250-5254

# Origin of enzyme cold-adaptation



\* Isaksen, GV., Åqvist, J., Brandsdal, BO. (2014) "Protein surface softness is the origin of cold adaptation of trypsin" *PLOS Comp Biol* 10(8)

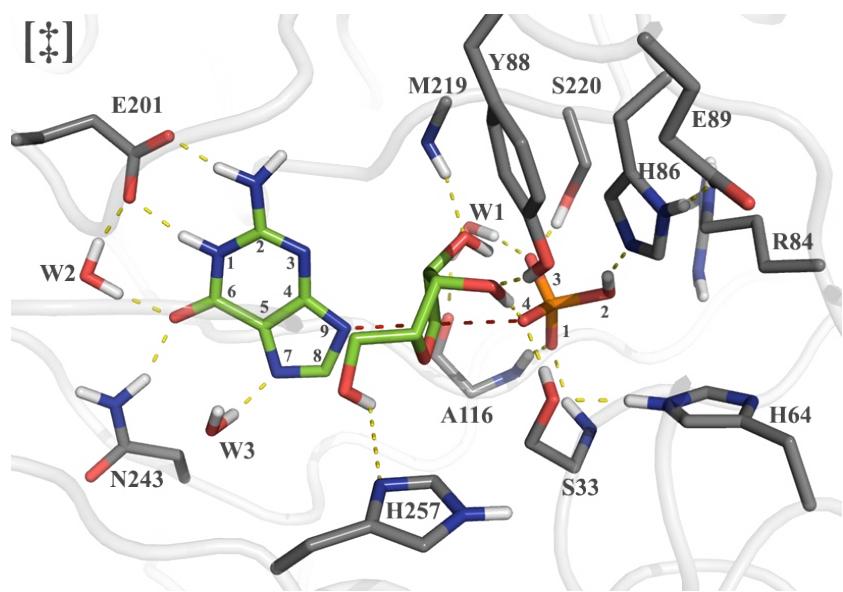
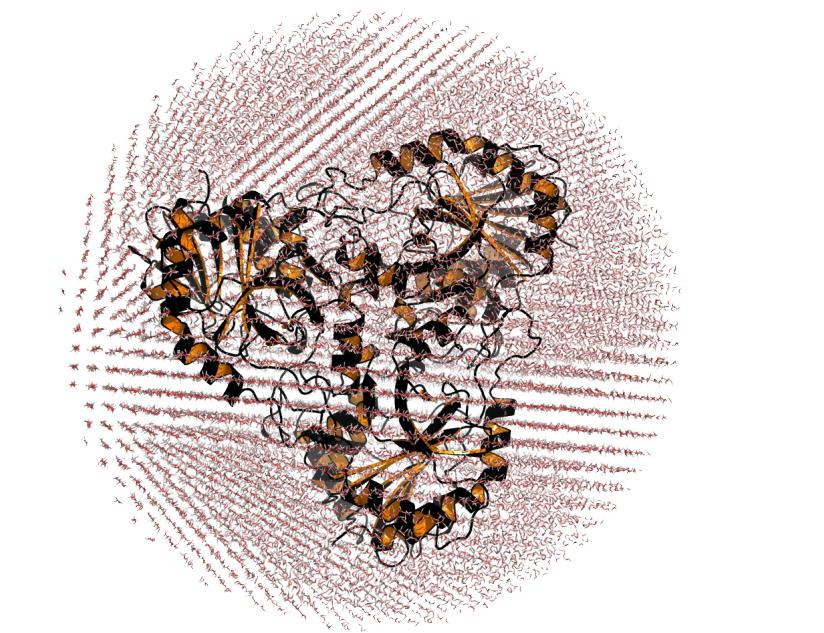
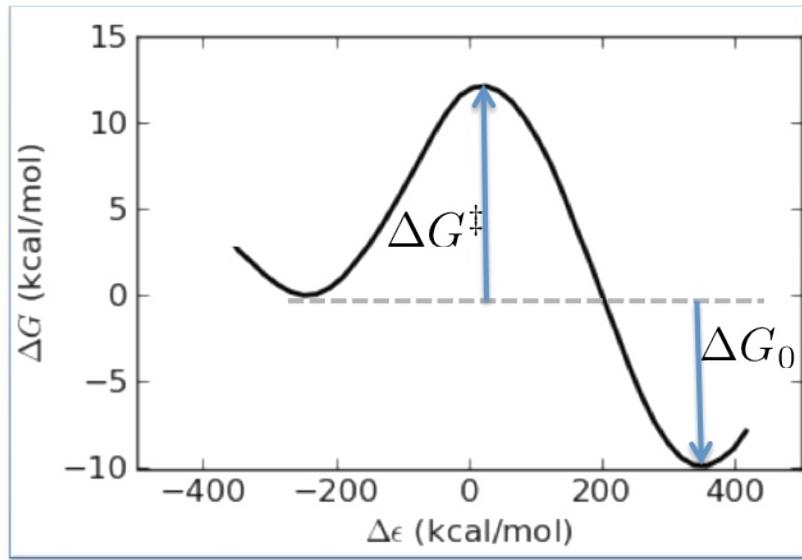
\* Isaksen, GV, Brandsdal, BO, Åqvist, J (2016) "Enzyme surface rigidity tunes the temperature dependence of catalytic rates" *Proc Nat Acad USA* 113(28):7822-7827

\* Åqvist, J, Isaksen, GV, Brandsdal, BO (2017) "Computation of enzyme cold adaptation" *Nat Rev Chem* 51(1):1-14

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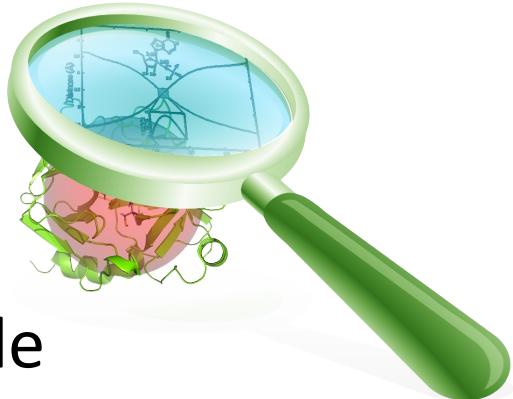
# What is EVB?

- Enzyme & solution reactions
- Large and complex systems



\* Warshel & Weiss 1980, 1981

# What is EVB?



- Relate to experimentally measurable thermodynamic properties
- Dissecting observed energies into individual contributions

$$k_{\text{cat}} = \kappa \frac{k_B T}{h} \exp \left( \frac{-\Delta G^\ddagger}{k_b T} \right)$$

$$\Delta G^\ddagger = \Delta H^\ddagger - T\Delta S^\ddagger$$

$$\Delta H^\ddagger = \Delta U^\ddagger + p\Delta V^\ddagger$$

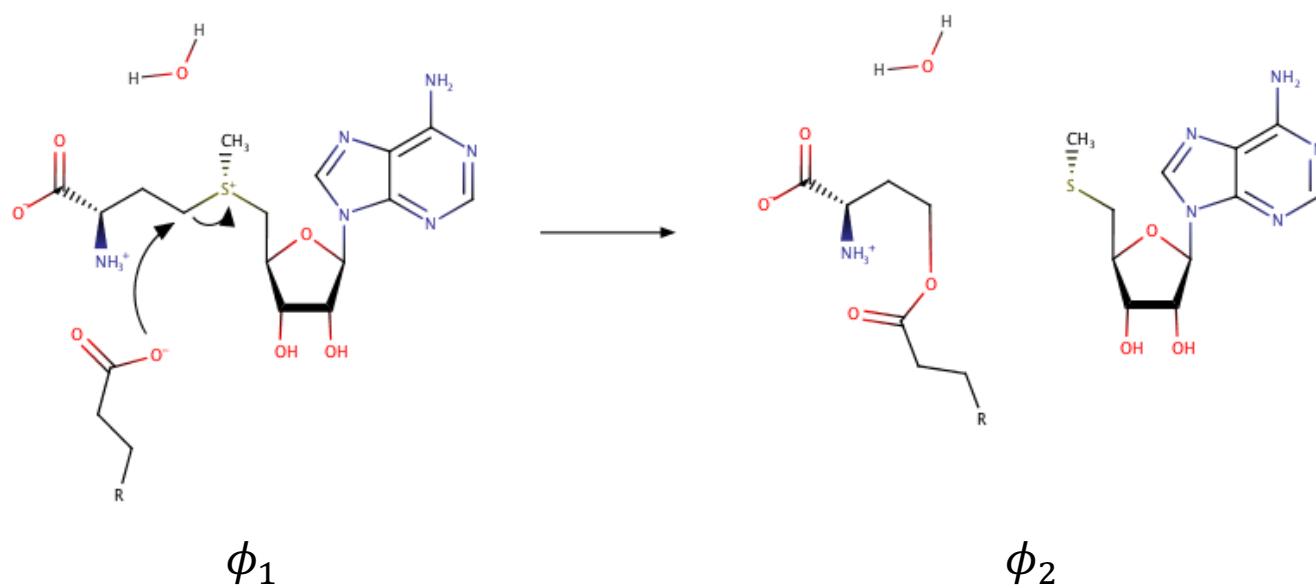
$$\Delta U^\ddagger = \Delta U_{rr}^\ddagger + \Delta U_{rs}^\ddagger + \Delta U_{ss}^\ddagger$$

$$\Delta U_{ss}^\ddagger = \Delta U_{pp}^\ddagger + \Delta U_{pw}^\ddagger + \Delta U_{ww}^\ddagger$$

r: reacting fragment  
s: surroundings  
p: protein  
w: water

# Morse potentials

- Standard MM FFs use harmonic bond potentials



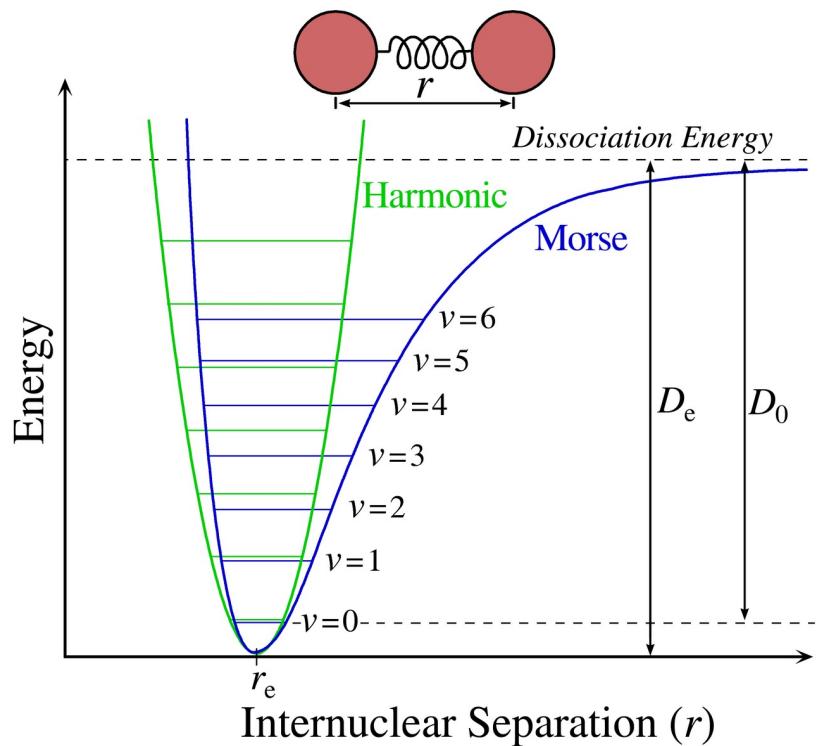
# Morse potentials

- Form/break bonds with MM methods

$$U(r) = \frac{1}{2} k_b (r - r_e)^2$$



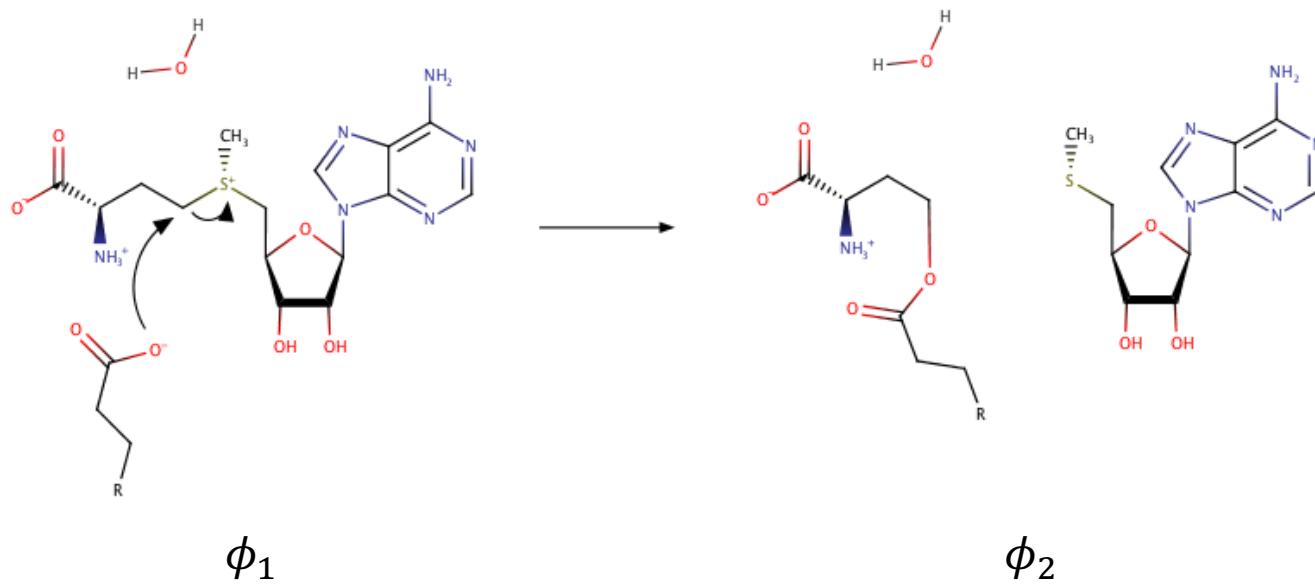
$$U(r) = D_e \left(1 - e^{-a(r-r_e)}\right)^2$$



# Free Energy Perturbation

- Free energy difference between two states:

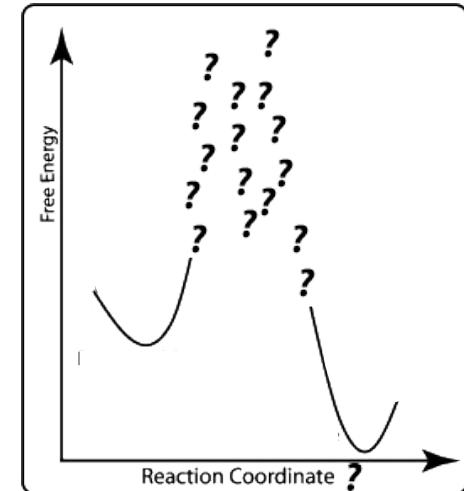
- $\Delta G = -\beta^{-1} \ln \langle \exp(-\beta\Delta\epsilon) \rangle_{\phi_1}$  (NPT)
  - $\beta = 1/k_B T$



# Free Energy Perturbation

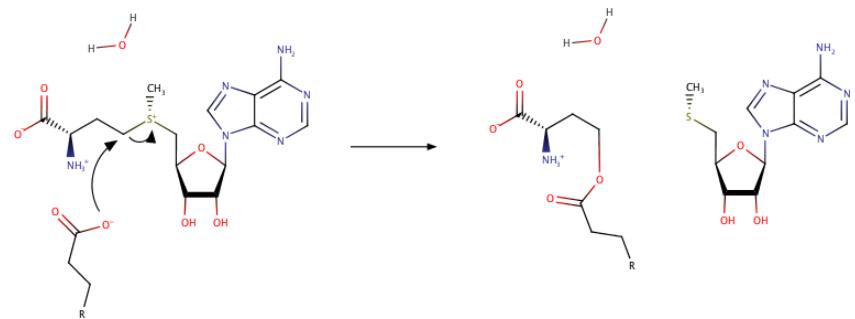
- Free energy difference between two states:

- $\Delta G = -\beta^{-1} \ln \langle \exp(-\beta\Delta\epsilon) \rangle_{\phi_1}$ 
  - $\beta = 1/k_B T$



- The potentials ( $\epsilon_i, \epsilon_{i+1}$ ) must significantly overlap
- Intermediate mapping potentials ( $\epsilon_m$ ):

- $\epsilon_m = (1 - \lambda_m)\epsilon_1 + \lambda_m\epsilon_2$ 
  - $\lambda_m \in [0,1]$



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$\phi_1$

$\phi_2$

# Free Energy Perturbation

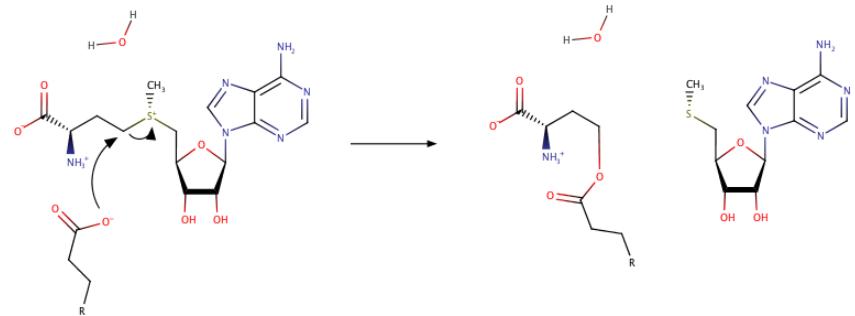
- Free energy difference between two states:

$$\bullet \Delta G = -\beta^{-1} \ln \langle \exp(-\beta \Delta \epsilon) \rangle_{\phi_1}$$



$$\bullet \Delta G_m = \beta^{-1} \sum_{m=0}^{n-1} \ln \langle \exp[-\beta(\epsilon_{m+1} - \epsilon_m)] \rangle_m$$

- $\beta = 1/k_B T$
- $\epsilon_m = (1 - \lambda_m)\epsilon_1 + \lambda_m\epsilon_2$ 
  - $\lambda_m \in [0,1]$



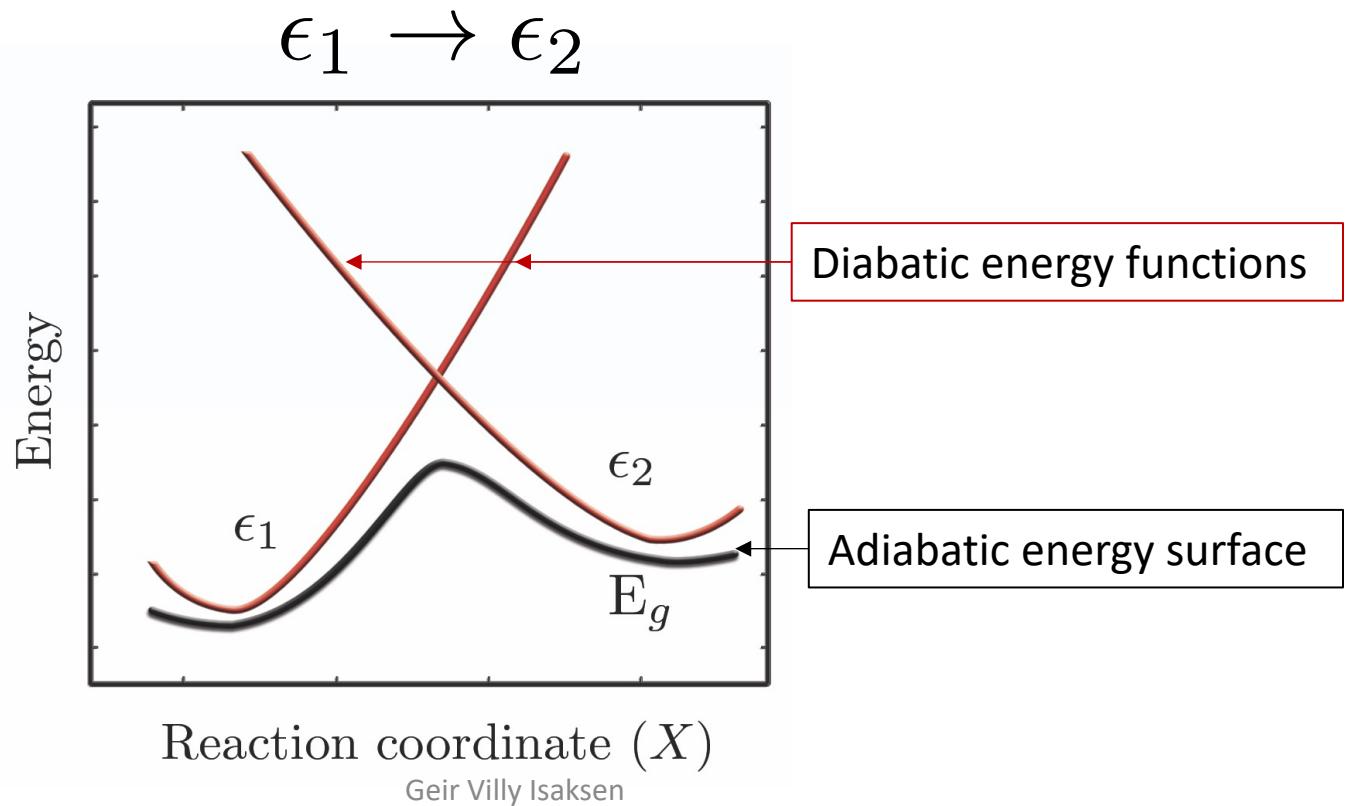
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$\phi_1$

$\phi_2$

# How does EVB work?

- Rudy Marcus electron transfer theory of metals
  - Applied to chemical catalysis in solution



# The EVB Hamiltonian

$$\widehat{H}_{EVB} = \begin{pmatrix} H_{ii} & \cdots & H_{ij} \\ \vdots & \ddots & \vdots \\ H_{ji} & \cdots & H_{jj} \end{pmatrix}$$

- Each state in a chemical reaction is described as VB states ( $\phi_1, \phi_2, \dots, \phi_n$ )
- Diagonal elements (diabatic energy functions):

$$H_{ii} = \epsilon_i = U_{bnd}^i + U_{ang}^i + U_{tor}^i + U_{nb,rr}^i + U_{nb,rs}^i + \alpha^i + U_{ss}$$

- $\alpha$  = intrinsic gas-phase energy at infinite separation
- Off-diagonal elements (adiabatic mixing)

$$H_{ij} = \sum_{k,l} A_{ij}^{k,l} \exp[-\mu_{i,j}^{k,l} r_{k,l}]$$

$$H_{ij} = \sum_{k,l} A_{ij}^{k,l}$$

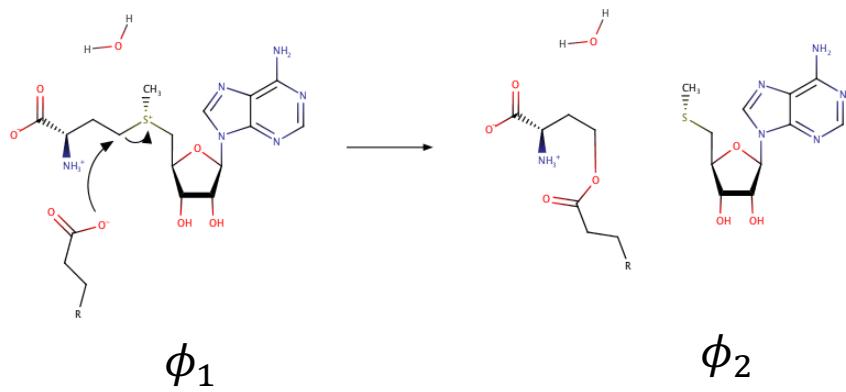
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$$H_{ij} = \sum_{k,l} A_{ij}^{k,l}$$

2-state EVB



$$\widehat{H} = \begin{pmatrix} \epsilon_1 & A_{1,2} \\ A_{2,1} & \epsilon_2 \end{pmatrix} = \begin{pmatrix} \epsilon_1 & A_{1,2} \\ A_{1,2} & \epsilon_2 \end{pmatrix}$$

# The EVB Hamiltonian

$$\widehat{H}_{EVB} = \begin{pmatrix} H_{ii} & \cdots & H_{ij} \\ \vdots & \ddots & \vdots \\ H_{ji} & \cdots & H_{jj} \end{pmatrix}$$

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$$\widehat{H} = \begin{pmatrix} \epsilon_1 & A_{1,2} \\ A_{1,2} & \epsilon_2 \end{pmatrix}$$

- The adiabatic ground-state energy:

$$|\widehat{\mathbf{H}} - \mathbf{I}E_g| = 0$$

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$$\left\| \begin{bmatrix} \epsilon_1 & A_{1,2} \\ A_{1,2} & \epsilon_2 \end{bmatrix} - \begin{bmatrix} E_g & 0 \\ 0 & E_g \end{bmatrix} \right\| = 0$$

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$$\left[ \begin{bmatrix} \epsilon_1 & A_{1,2} \\ A_{1,2} & \epsilon_2 \end{bmatrix} - \begin{bmatrix} E_g & 0 \\ 0 & E_g \end{bmatrix} \right] = 0$$



$$E_g^2 + E_g(-\epsilon_1 - \epsilon_2) + \epsilon_1\epsilon_2 - A_{1,2}^2 = 0$$



$$E_g = \frac{(\epsilon_1 + \epsilon_2) - \sqrt{(\epsilon_1 - \epsilon_2)^2 + 4A_{1,2}}}{2}$$

# The EVB Hamiltonian

$$\hat{H} = \begin{pmatrix} \epsilon_1 & A_{1,2} \\ A_{1,2} & \epsilon_2 \end{pmatrix}$$

- Or, solving with the eigenvector coefficients:

$$\begin{aligned}\hat{\mathbf{H}}\mathbf{C} &= E_g \mathbf{C} \\ \downarrow \\ \mathbf{C}^* \hat{\mathbf{H}} \mathbf{C} &= E_g \\ \downarrow \\ [c_1 \quad c_2] \begin{bmatrix} \epsilon_1 & A_{1,2} \\ A_{1,2} & \epsilon_2 \end{bmatrix} \begin{bmatrix} c_1 \\ c_2 \end{bmatrix} &= E_g \\ \downarrow \\ c_1^2 \epsilon_1 + 2c_1 c_2 A_{1,2} + c_2^2 \epsilon_2 &= E_g\end{aligned}$$

The lowest eigenvalue will always have  $c_1 c_2 < 0$ :

$$E_g = c_1^2 \epsilon_1 + c_2^2 \epsilon_2 - 2|c_1 c_2| A_{1,2}$$

# The EVB Hamiltonian

$$\widehat{H} = \begin{pmatrix} \epsilon_1 & A_{1,2} \\ A_{1,2} & \epsilon_2 \end{pmatrix}$$

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$$E_g = c_1^2 \epsilon_1 + c_2^2 \epsilon_2 - 2|c_1 c_2| A_{1,2}$$

$$\Delta U^\ddagger = \Delta(c_1^2 U_1)^\ddagger + \Delta(c_2^2 U_2)^\ddagger + \Delta(c_2^2 \alpha_2)^\ddagger - \Delta(2|c_1 c_2| H_{12})^\ddagger$$

$$c^2 U = \sum_i w_i c(\lambda_i)^2 U(\lambda_i); \lambda_i \in [\Delta\epsilon(rs), \Delta\epsilon(ts)]$$

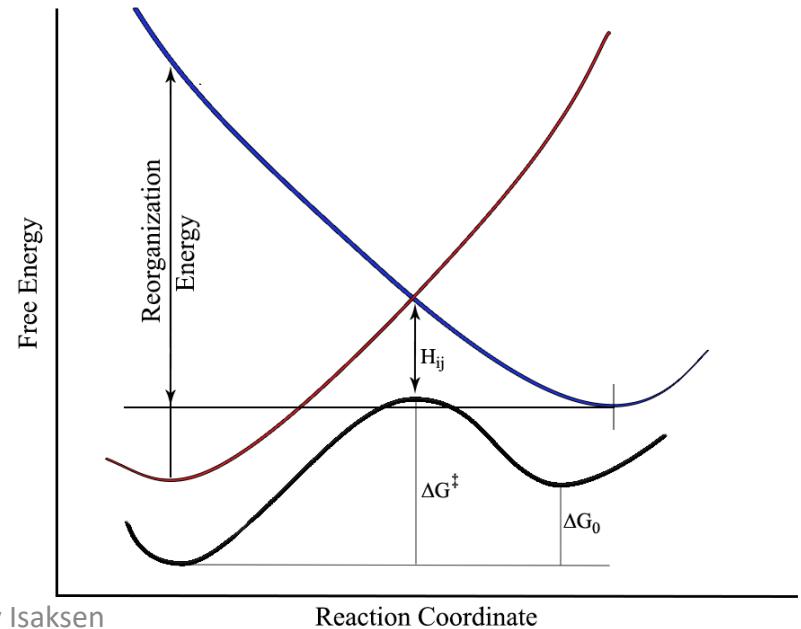
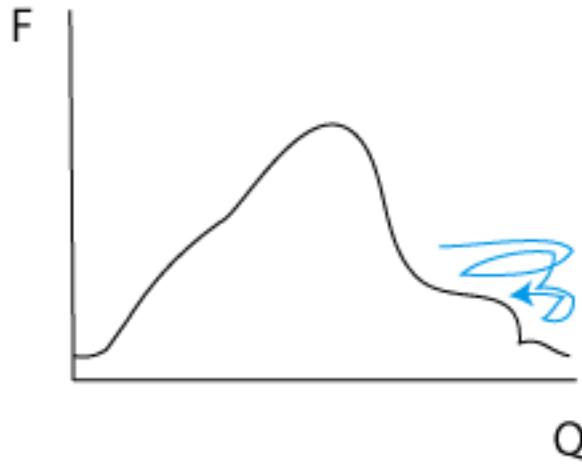
$$\sum_i w_i = 1$$

# The free energy functional

- FEP-Umbrella sampling (FEP/US) method

$$\Delta G(\Delta\epsilon_n) = \sum_{m \supset \Delta\epsilon_n} w_m \left( \Delta G_m - \beta^{-1} \ln \left\langle \exp \left[ \beta (E_g(\Delta\epsilon_n) - \epsilon_m(\Delta\epsilon_n)) \right] \right\rangle_m \right) / \sum_{m \supset \Delta\epsilon_n} w_m$$

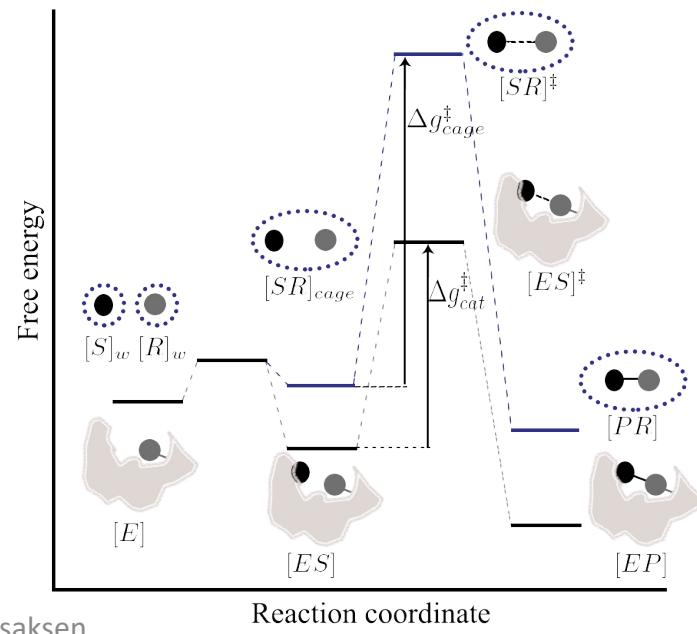
$$\left( \Delta G_m = \beta^{-1} \sum_{m=0}^{n-1} \ln \langle \exp[-\beta(\epsilon_{m+1} - \epsilon_m)] \rangle_m \right)$$



# Calibrating $\hat{H}_{\text{EVB}}$

$$\hat{H} = \begin{pmatrix} \epsilon_1 & A_{1,2} \\ A_{1,2} & \epsilon_2 \end{pmatrix}$$

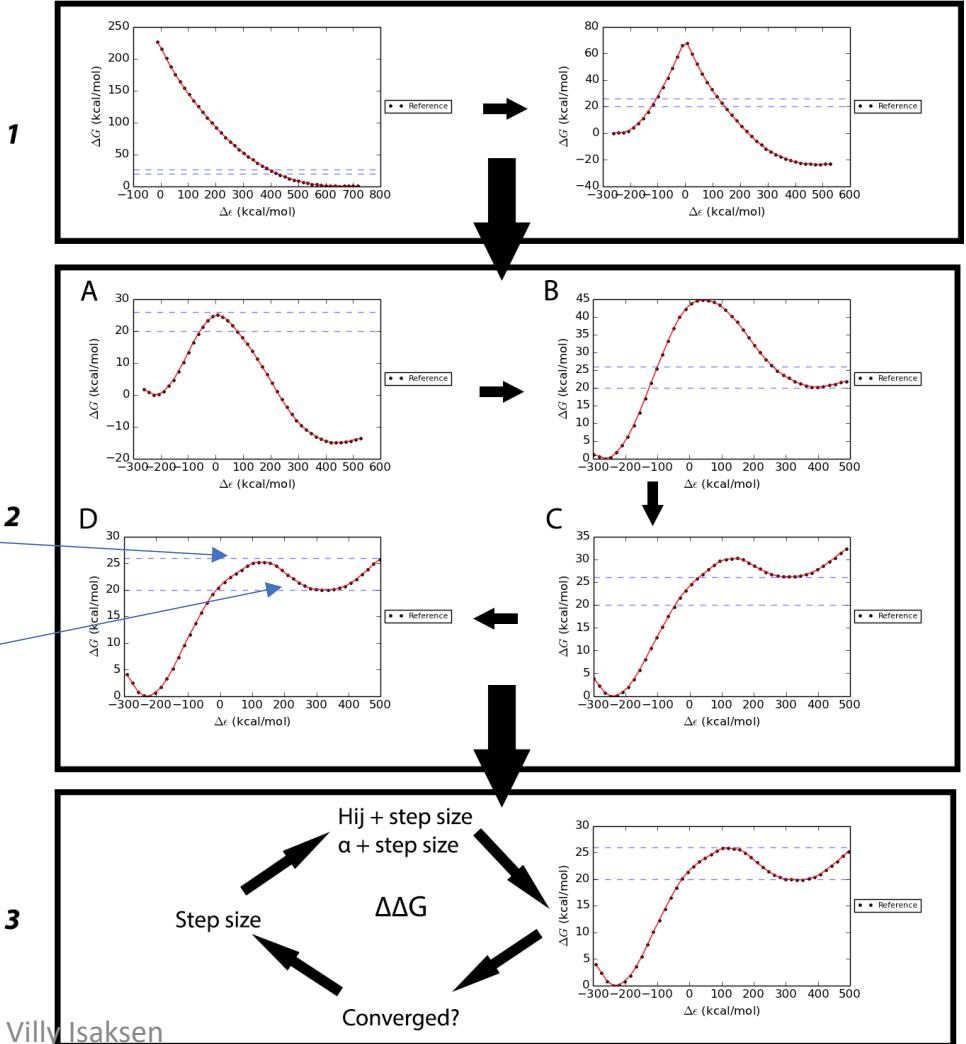
- Calibration of the empirical parameters ( $\alpha$  and  $A_{1,2}$ )
  - Key feature of EVB
- Reference reaction → Enzyme
  - Same parameters



# Calibrating $\hat{H}_{\text{EVB}}$

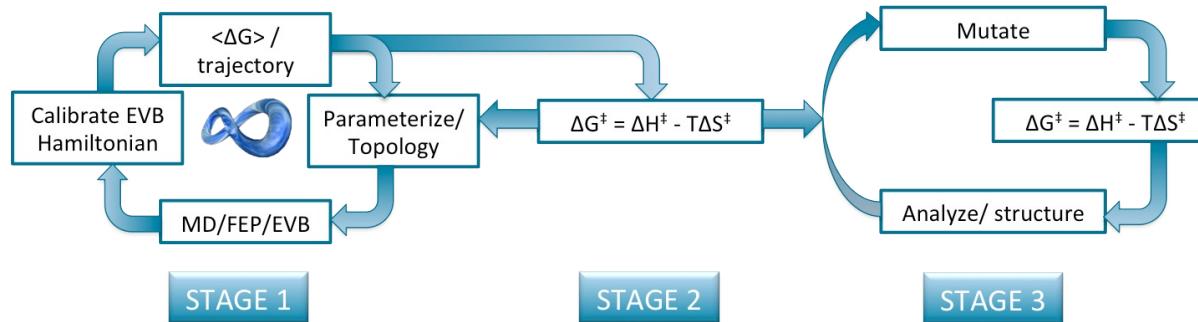
$$\hat{H} = \begin{pmatrix} \epsilon_1 & A_{1,2} \\ A_{1,2} & \epsilon_2 \end{pmatrix}$$

Adjusting  $\alpha$  and  $A_{1,2}$  iteratively



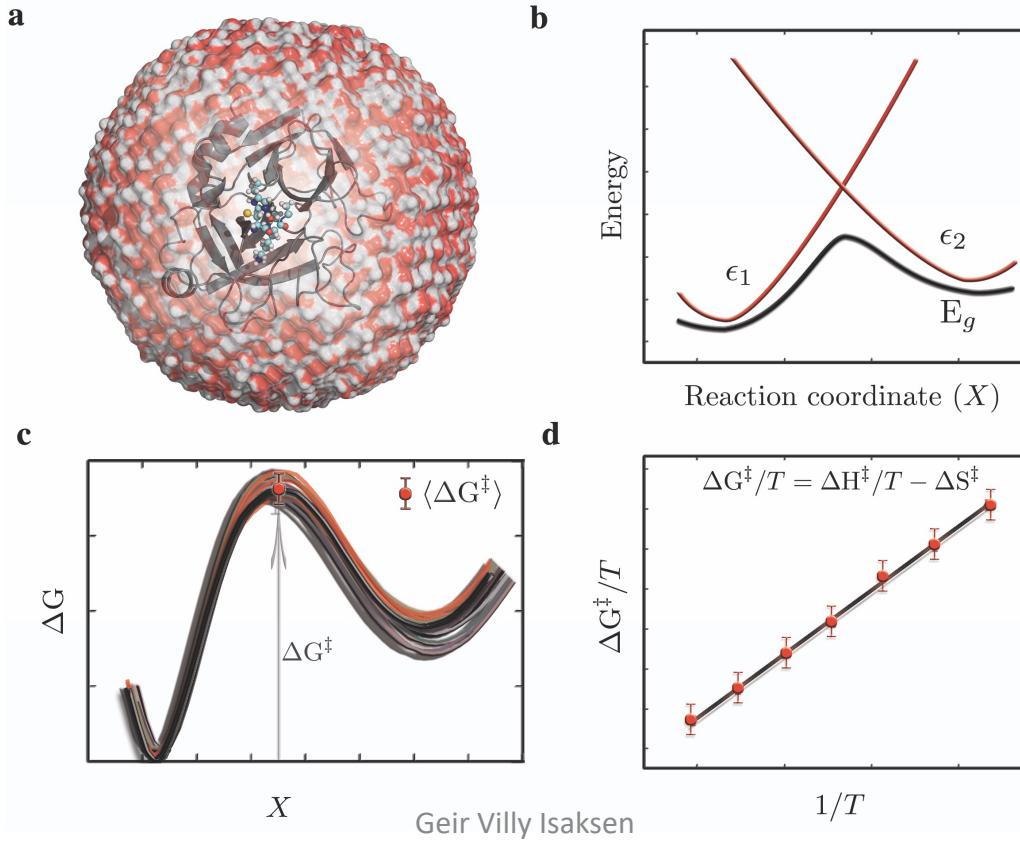
# Example workflow

- Calculating thermodynamic activation parameters

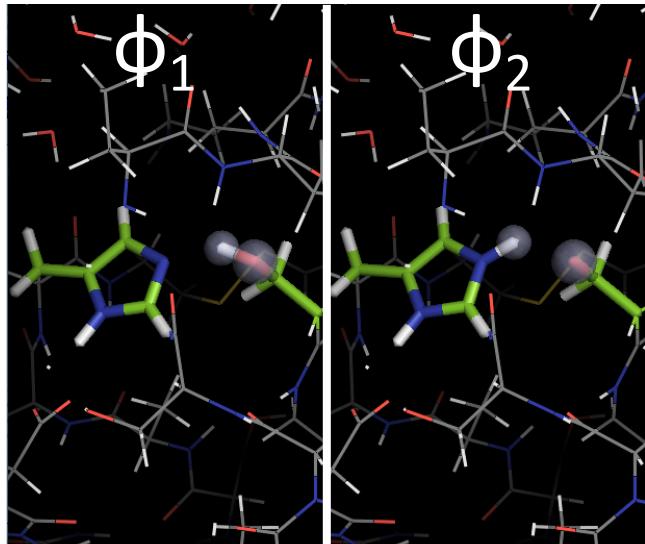


# Example workflow

- Calculating thermodynamic activation parameters



# The manual challenges



**17 Q-atoms:**

12 Atomtypes

34 Charges

17 Bond parameters

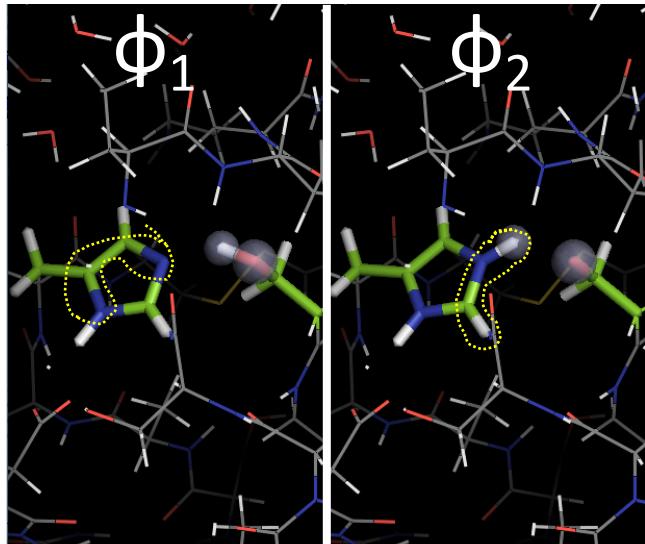
26 Angle parameters

105 (35\*3) Torsion parameters

8 Improper torsion parameters

→ **202 parameters**

# The manual challenges



**17 Q-atoms:**

12 Atomtypes

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17 Bond parameters

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105 (35\*3) Torsion parameters

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→ **202 parameters**

# Qgui

- <https://youtu.be/sQTYAZztY4Y>

# References

- Warshel A (1978) "Energetics of Enzyme Catalysis" *Proc Nat Acad USA* 75(11):5250-5254
- Warshel A. and Weiss R. M., (1980). "An empirical valence bond approach for comparing reactions in solutions and in enzymes", *Journal of the American Chemical Society*, **102**, 6218-6226.
- Warshel A. and Weiss R. M., (1981). "Empirical valence bond calculations of enzyme catalysis", *Annals of the New York Academy of Sciences*, **367**, 370-382

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$$H_{ij} = \sum_{k,l} A_{ij}^{k,l}$$

3-state EVB

$$\begin{pmatrix} \epsilon_1 & A_{1,2} & A_{1,3} \\ A_{2,1} & \epsilon_2 & A_{2,3} \\ A_{3,1} & A_{3,2} & \epsilon_3 \end{pmatrix} \longrightarrow \begin{pmatrix} \epsilon_1 & A_{1,2} & A_{1,3} \\ A_{1,2} & \epsilon_2 & A_{2,3} \\ A_{1,3} & A_{2,3} & \epsilon_3 \end{pmatrix}$$

