

ITC experiments (Interaction of peptides with Gallium at pH 3.0)

Parameters:

- Buffered ITC solution contained 80 mM sodium acetate, 150 mM NaCl at pH 3.0
- Unbuffered ITC solution contained only 150 mM NaCl, 1 mM HCl
- Peptides had a concentration of 0.1 – 0.3 mM in the respective ITC solution
- Gallium was provided as $\text{Ga}(\text{NO}_3)_3$ with a concentration of 2 – 6 mM in the respective ITC solution (other metals with similar concentrations)

Peptide C3.8 [TMHHAAIAHPPH]

- Results indicate only weak interaction with Ga and no interaction with As
- 1:1 complex formation appears most likely
- Interaction doesn't depend on the presence of acetate buffer

| Ga Interaction (150 mM NaCl, pH 3.0) | |
|--------------------------------------|--|
| Binding affinity | $5.0 \cdot 10^2 \text{ M}^{-1}$ |
| ΔH (enthalpy) | $0.76 \text{ J} \cdot \text{mol}^{-1}$ |
| ΔS (entropy) | $0.15 \text{ J} \cdot \text{mol}^{-1}$ |

Peptide C3.130 [NDLQRHRLTAGP]

- Results indicate only weak interaction with Ga and no interaction with As
- 1:1 complex formation appears most likely
- The presence of acetate buffer seems to weaken complex formation, probably to competitive effect of acetate ions (?)

| Ga Interaction (150 mM NaCl, pH 3.0) | |
|--------------------------------------|--|
| Binding affinity | $9.0 \cdot 10^2 \text{ M}^{-1}$ |
| ΔH (enthalpy) | - $4.31 \text{ J} \cdot \text{mol}^{-1}$ |
| ΔS (entropy) | $0.072 \text{ J} \cdot \text{mol}^{-1}$ |

Peptide C3.15 [NYLPHQSSSPSR]

- Results indicate no interaction with As
- Complex formation with Ga seems to involve two reactions (or maybe two peptide molecules?), but 1:1 complex formation appears most likely from my data sets
- one exothermic reaction followed by one endothermic reaction (might depend on each other)
- Reactions take place independent from the presence of acetate
- Reaction enthalpy indicates stable complex formation
- But it's striking that the change in entropy is very high

| Ga Interaction (150 mM NaCl, pH 3.0) | | |
|--------------------------------------|--|---|
| Binding affinity | $4.6 \cdot 10^3 \text{ M}^{-1}$ | $2.0 \cdot 10^3 \text{ M}^{-1}$ |
| ΔH (enthalpy) | - $297.28 \text{ J} \cdot \text{mol}^{-1}$ | $58.62 \text{ J} \cdot \text{mol}^{-1}$ |
| ΔS (entropy) | - $1.17 \text{ J} \cdot \text{mol}^{-1}$ | $1.97 \text{ J} \cdot \text{mol}^{-1}$ |

Peptide C3.108 [SQALSTSRQDLR]

- Results indicate no interaction with As
- Complex formation with Ga seems to involve two reactions (or maybe two peptide molecules?)
- one exothermic reaction followed by one endothermic reaction (might depend on each other)
- Presence of acetate seems to weaken complex formation, probably to competitive effect of acetate ions (?)
- Heat uptake is higher than heat release

| Ga Interaction (150 mM NaCl, pH 3.0) | | |
|---|---|--|
| Binding affinity | $4.8 \cdot 10^3 \text{ M}^{-1}$ | $1.9 \cdot 10^3 \text{ M}^{-1}$ |
| ΔH (enthalpy) | $-146.55 \text{ J} \cdot \text{mol}^{-1}$ | $381.02 \text{ J} \cdot \text{mol}^{-1}$ |
| ΔS (entropy) | $-0.43 \text{ J} \cdot \text{mol}^{-1}$ | $1.35 \text{ J} \cdot \text{mol}^{-1}$ |

Peptide C3.129 [HTQHIQSDDHLA]

Peptide SDM-M3 [HTCHIQSCDHLA]

Peptide SDM-M4 [HTQCIQSDCHLA]

- Results indicate no interaction with As
- No Ga complexes are formed in the absence of acetate
- Ga complex formation occurs with the highest affinity for the most structured peptide (SDM-M3) and the highest heat release for the most flexible sequence (C3.129)

| Ga Interaction (80 mM sodium acetate, 150 mM NaCl, pH 3.0) | | | |
|--|--|---|---|
| | C3.129 | SDM-M3 | SDM-M4 |
| Binding affinity | $8.0 \cdot 10^2 \text{ M}^{-1}$ | $1.5 \cdot 10^4 \text{ M}^{-1}$ | $3.7 \cdot 10^3 \text{ M}^{-1}$ |
| ΔH (enthalpy) | $-35.89 \text{ J} \cdot \text{mol}^{-1}$ | $-4.86 \text{ J} \cdot \text{mol}^{-1}$ | $-5.86 \text{ J} \cdot \text{mol}^{-1}$ |
| ΔS (entropy) | $0.065 \text{ J} \cdot \text{mol}^{-1}$ | $0.034 \text{ J} \cdot \text{mol}^{-1}$ | $0.050 \text{ J} \cdot \text{mol}^{-1}$ |