Titration of RNase with 2'-CMP

High protein concentration

**Cell:** RNase 64 uM  
**Syringe:** 2'-CMP 2.19 mM  
2 uL injections

The area underneath each injection peak (top panel) is equal to the total heat released for that injection. When this is plotted against the molar ratio of ligand added to macromolecule in the cell, a complete binding isotherm for the interaction is obtained (bottom panel).

The fitted isotherm provides the following binding parameters:

Data: RNaseEwa11_NDH  
Model: OneSites  
Chi^2 = 9521.08

N = 0.9617 ± 0.001882 (Stoichiometry; mass ratio)  
K = 7.566 ± 0.1958 E5 (Binding constant)  
delta H = -1.692 E4 ± 48.84 (Enthalpy of binding reaction)  
delta S = -28.90 (Entropy of binding reaction)
Low protein concentration

<table>
<thead>
<tr>
<th></th>
<th>Low protein concentration</th>
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<tbody>
<tr>
<td></td>
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<tr>
<td>RNase</td>
<td>11.4 uM</td>
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<tr>
<td>2'-CMP</td>
<td>0.544 mM</td>
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<td></td>
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<tr>
<td>RNase</td>
<td>40 uM</td>
</tr>
<tr>
<td>2'-CMP</td>
<td>2.72 mM</td>
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</table>

![Graphs showing time in minutes and microcalories per second](https://www.med.yale.edu/wmkeck/biophysics/RNAse_ITC.html)

**Data:** RNAseEwa112602_NDH  
**Model:** OneSites  
**Chi\(^2\) = 22106.6**  
**N = 0.8972 ± 0.00832**  
**K = 7.902 ± 0.369 E5**  
**delta H = -1.783 E4 ± 226.8**  
**delta S = -31.83**

**Data:** RNAseEwa112502_NDH  
**Model:** OneSites  
**Chi\(^2\) = 8499.21**  
**N = 1.006 ± 0.00352**  
**K = 6.806 ± 0.2378 E5**  
**delta H = -1.655 E4 ± 92.44**  
**delta S = -27.91**