Statistical and thermodynamic analysis of the binding of trans-activation response–binding proteins to HIV-1 TAR RNA

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In a recent article (1), the authors examine the binding of lab-evolved trans-activation response (TAR)–binding proteins (TBPs) to HIV-1 TAR RNA. Here, we show our analysis of the thermodynamic data of the binding that identifies three quantitative features of the binding, which may provide further insight into the interactions.

(1) The binding of TBPs to HIV-1 TAR RNA exhibits enthalpy–entropy compensation (2, 3), which suggests that the bindings of the six TBPs follow a common mechanism in the binding (Fig. 1*A*).
(2) Two other variants of TBPs do not fit the linear regression (Fig. 1*A*), according to a regression diagnostic test using studentized deleted residuals (4). This suggests that the rest structure of TBPs other than the TAR-binding β2-β3 loop, which is absent in the two outliers, contributes to the binding.
(3) The ΔG° values of the six TBPs at 310.15 K calculated using Equation 1 are normally distributed, according to the Shapiro–Wilk normality test (W = 0.933, p = 0.607):

$$\Delta G^\circ = \Delta H^\circ - T\Delta S^\circ \quad (1)$$

where ΔH° and ΔS° are experimentally measured values reported in the original paper (1). Based on the statistical parameters, probability density of the ΔG° can be generated using Equation 2:

$$p(\Delta G^\circ) = \frac{1}{\sigma\sqrt{2\pi}} \times \exp\left(-\frac{1}{2} \times \left(\frac{\Delta G^\circ - \Delta G^\circ_{\text{mean}}}{\sigma}\right)^2\right) \quad (2)$$

where σ = 1.9407 kJ/mol and $\Delta G^\circ_{\text{mean}}$ = −41.3 kJ/mol. The distribution allows one to relate the probability to find a TBP variant to its binding affinity. For example, statistically expected maximum ΔG° of the highest-affinity variant of TBPs (Fig. 1*B*) is obtained by solving Equation 3 for x to be −46.8 kJ/mol:

$$1 - \int_x^\infty p(\Delta G^\circ) d\Delta G^\circ = \frac{1}{400} \quad (3)$$

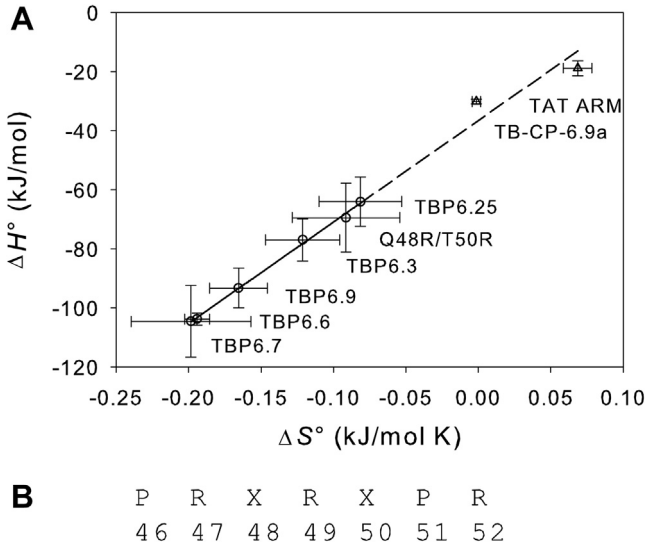

Figure 1. Statistical and thermodynamic analysis of the binding of TBPs to HIV-1 TAR RNA. *A*, enthalpy–entropy compensation in the binding. The solid line is the best fit to the data without two outliers (Tat ARM and TB-CP-6.9a), ΔH° = 343.3 × ΔS° − 36.6 (R^2 = 0.9972), and the dotted line is its extrapolation. *B*, the β2-β3 loop sequence of the lab-evolved TBPs. X indicates two positions of mutation, leading to 400 possible variations of the sequence. All statistical analyses were conducted using SigmaPlot (version 11; Systat Software). TAR, trans-activation response; TBP, TAR-binding protein.

Conflict of interest—The authors declare that they have no conflicts of interest with the contents of this article.

Abbreviations—The abbreviations used are: TAR, trans-activation response; TBP, TAR-binding protein.

References

1. Chavali, S. S., Mali, S. M., Jenkins, J. L., Fasan, R., and Wedekind, J. E. (October 13, 2020) Co-crystal structures of HIV TAR RNA bound to lab-evolved proteins show key roles for arginine relevant to the design of cyclic peptide TAR inhibitors. *J. Biol. Chem.* https://doi.org/10.1074/jbc.RA120.015444
2. Fox, J. M., Zhao, M., Fink, M. J., Kang, K., and Whitesides, G. M. (2018) The molecular origin of enthalpy/entropy compensation in biomolecular recognition. *Annu. Rev. Biophys.* **47**, 223–250
3. Kang, J., and Auerbach, J. D. (2009) Thermodynamic characterization of dissociation rate variations of human leukocyte antigen and peptide complexes. *Mol. Immunol.* **46**, 2873–2875
4. Pardoe, I. (2012) *Applied Regression Modeling*, 2nd Ed., Wiley, Hoboken, NJ: 190

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