LETTER TO THE EDITOR

Reply to: Comment on "Revisiting the Ramachandran plot from a new angle"

Dear Brian,

With this letter, we reply to Porter and Rose, who in a recent paper¹ and in their Comment on "Revisiting the Ramachandran plot from a new angle" assert that sidechain hydrogen bonding is the primary determinant of the observed population of ϕ/ψ backbone dihedral angles in the "bridge region" ($\phi \leq 0^\circ$ and $-20^\circ \leq \psi \leq 40^\circ$). Our manuscript "revisiting the Ramachandran plot from a new angle"² shows that steric considerations alone predict the observed backbone dihedral angle distributions. In this response, we reiterate three main points.

- 1) The calculated Ramachandran plot depends critically on the value of the bond angle τ (between C', C_α, and N). Specifically, as τ increases within the range $100^\circ \leq \tau \leq 120^\circ$, configurations in the "bridge region" with $\phi \leq 0^\circ$ and $-20^\circ \leq \psi \leq 40^\circ$ change from being disallowed at $\tau = 100^\circ$ to be allowed at $\tau = 115^\circ$ and above. In fact, this result dates back to 1965,³ but most often Ramachandran plots are drawn for a single value of τ , typically $\tau = 110^\circ$. The τ dependence is generally not considered.
- 2) We analyzed a database⁴ of high-resolution protein structures and plotted the backbone dihedral angles of all amino acids on Ramachandran plots as a function of their bond angle τ . (See Fig. 2 in Ref. 2) In agreement with the predictions of Ramachandran and colleagues,³ the bridge region is populated by amino acids with large values of τ , while ϕ and ψ combinations in the bridge region are not frequently observed for amino acids with smaller values of τ . Thus, an increased population of structures in the bridge region at large τ is consistent with the predictions from steric interactions alone for every amino acid type.
- 3) The analysis and conclusions of Porter and Rose suggest that hard-sphere interactions alone cannot explain the observed distribution of backbone dihedral angles. For example, they state in the abstract that the "forbidden [bridge] region is

well-populated in folded proteins, which can provide longer-range intramolecular hydrogen-bond partners." However, using only a hard sphere model, Ramachandran and colleagues predicted the ϕ and ψ combinations as a function of τ that are observed in proteins of known structure. In particular, they showed that ϕ and ψ combinations in the bridge region are not forbidden at large τ .

It is certainly possible that hydrogen-bonding may influence the adopted value of τ (which in turn may then influence the allowed values of ϕ and ψ), but the existence of ϕ and ψ combinations within the bridge region is not on its own prime facie evidence of the role of hydrogen bonding.

ALICE QINHUA ZHOU¹

COREY S. O'HERN^{2,3}

LYNNE REGAN^{1,4*}

¹Department of Molecular Biophysics & Biochemistry
Yale University, New Haven, CT

²Department of Mechanical Engineering & Materials
Science, Yale University, New Haven, CT

³Department of Physics, Yale University, New Haven, CT

⁴Department of Chemistry, Yale University
Yale University, New Haven, CT

(Received 16 August 2011)

References

1. Porter LL, Rose GN (2010) Redrawing the Ramachandran plot after inclusion of hydrogen-bonding constraints. *PNAS* 108:109–113.
2. Zhou AQ, O'Hern CS, Regan L (2011) Revisiting the Ramachandran plot from a new angle. *Protein Sci* 20: 1166–1171.
3. Ramakrishnan C, Ramachandran GN (1965) Stereochemical criteria for polypeptide and protein chain conformations. II. Allowed conformations for a pair of peptide units. *Biophys J* 55:909–933.
4. Dunbrack RL, Cohen FE (1997) Bayesian statistical analysis of protein side-chain rotamer preferences. *Protein Sci* 6:1661–1681.

*Correspondence to: Lynne Regan, E-mail: lynne.regan@yale.edu.

DOI:10.1002/pro.722

Published online 24 August 2011 proteinscience.org