

Biophysical Journal, Volume 114

Supplemental Information

Control of Transmembrane Helix Dynamics by Interfacial Tryptophan Residues

Matthew J. McKay, Ashley N. Martfeld, Anna A. De Angelis, Stanley J. Opella, Denise V. Greathouse, and Roger E. Koeppe II

Control of Transmembrane Helix Dynamics by Interfacial Tryptophan Residues

Matthew J McKay,¹ Ashley N Martfeld,¹ Anna A De Angelis,² Stanley J Opella,² Denise V Greathouse,¹ and Roger E Koeppe II¹

¹Department of Chemistry and Biochemistry, University of Arkansas, Fayetteville, Arkansas 72701 USA; and

²Department of Chemistry and Biochemistry, University of California, San Diego; La Jolla, California 92093 USA

Supplemental Figures

FIGURE S1. Reversed phase HPLC elution profile to confirm purification of GW^{4,20}ALP23.

FIGURE S2. MALDI-TOF mass spectrum confirming the synthesis and purification of GW^{4,20}ALP23.

FIGURE S3. Deuterium (²H) NMR spectra for labeled GW^{4,20}ALP23 in oriented DMPC bilayers at $\beta=90^\circ$ measured at 42 °C and 50 °C.

FIGURE S4. Deuterium (²H) NMR spectra for labeled GW^{4,20}ALP23 in oriented DLPC, DMPC, and DOPC bilayers at $\beta=0^\circ$ measured at 50 °C.

FIGURE S5. Illustration of PISA wheels that fit and do not fit the data for GW^{4,20}ALP23 in bicelles.

FIGURE S6. Deuterium (²H) NMR spectra for labeled GW^{4,20}ALP23 in oriented DMPC/DHoPC bicelles measured at 42 °C.

FIGURE S7. Rmsd contour plots for of GW^{5,19}ALP23 and GW^{4,20}ALP23 in DMPC/DHoPC bicelles using ¹⁵N + ²H data combined, or ²H data alone.

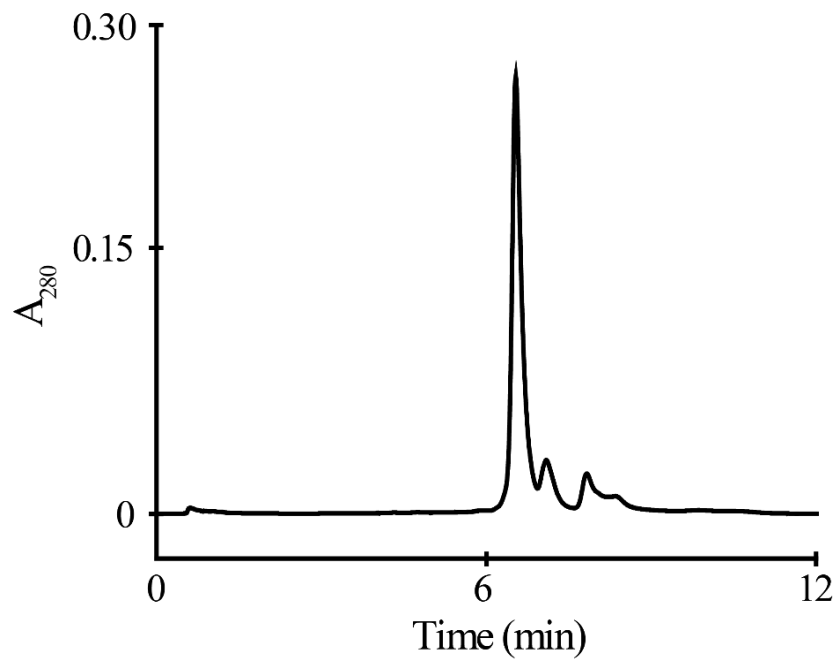


FIGURE S1

Reversed phase HPLC elution profile to confirm purification of GW^{4,20}ALP23.

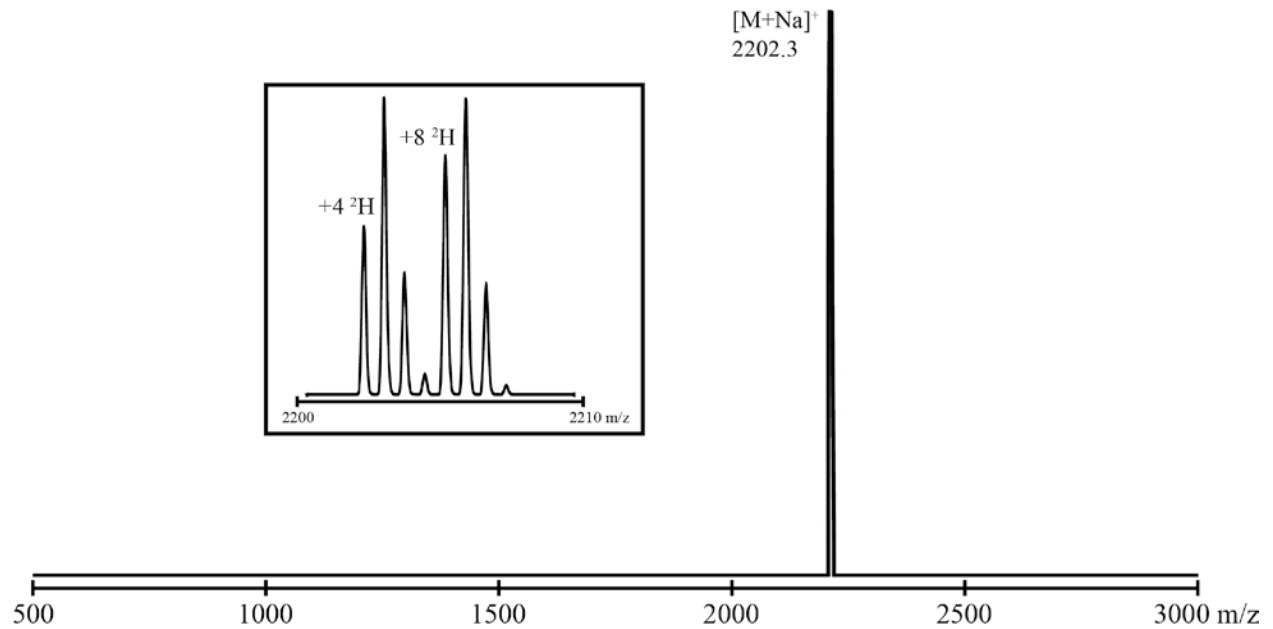


FIGURE S2.

MALDI-TOF mass spectrum confirming the synthesis and purification of GW^{4.20}ALP23. The expected monoisotopic mass of the undeuterated peptide with Na⁺ is 1198 daltons. The observed mass is 2202.3 with Na⁺ and 4 deuterons, or 2206.3 with Na⁺ and 8 deuterons. Adjacent peaks differ in mass by \pm one atom of naturally abundant ^{13}C (1.1% abundance).

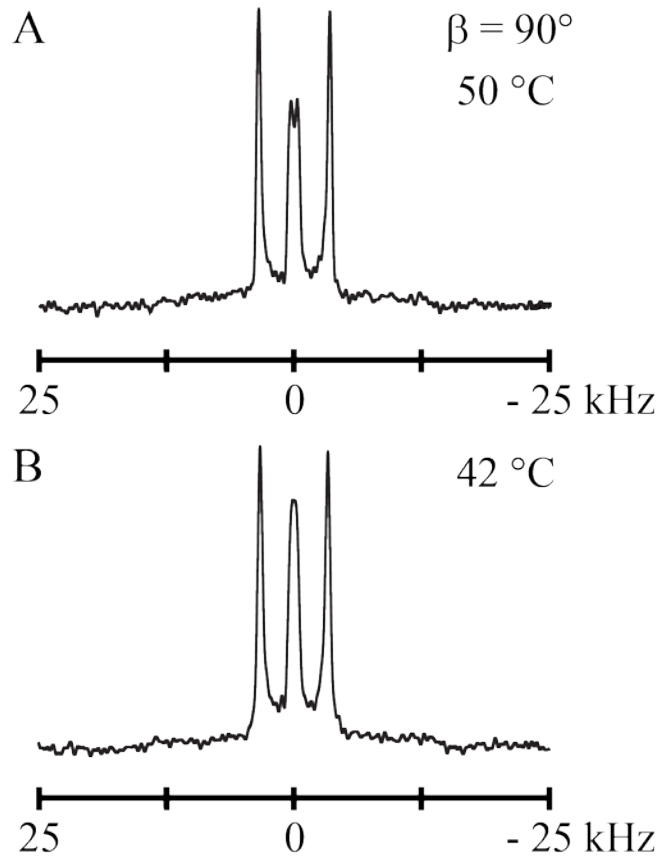


FIGURE S3.

Deuterium (^2H) NMR spectra for labeled $\text{GW}^{4,20}$ ALP23 in oriented DMPC bilayers at $\beta=90^\circ$ measured at (A) 50 °C and (B) 42 °C. ^2H labeled alanine positions are 3 and 21 and were labeled with 100% and 50% abundances respectively.

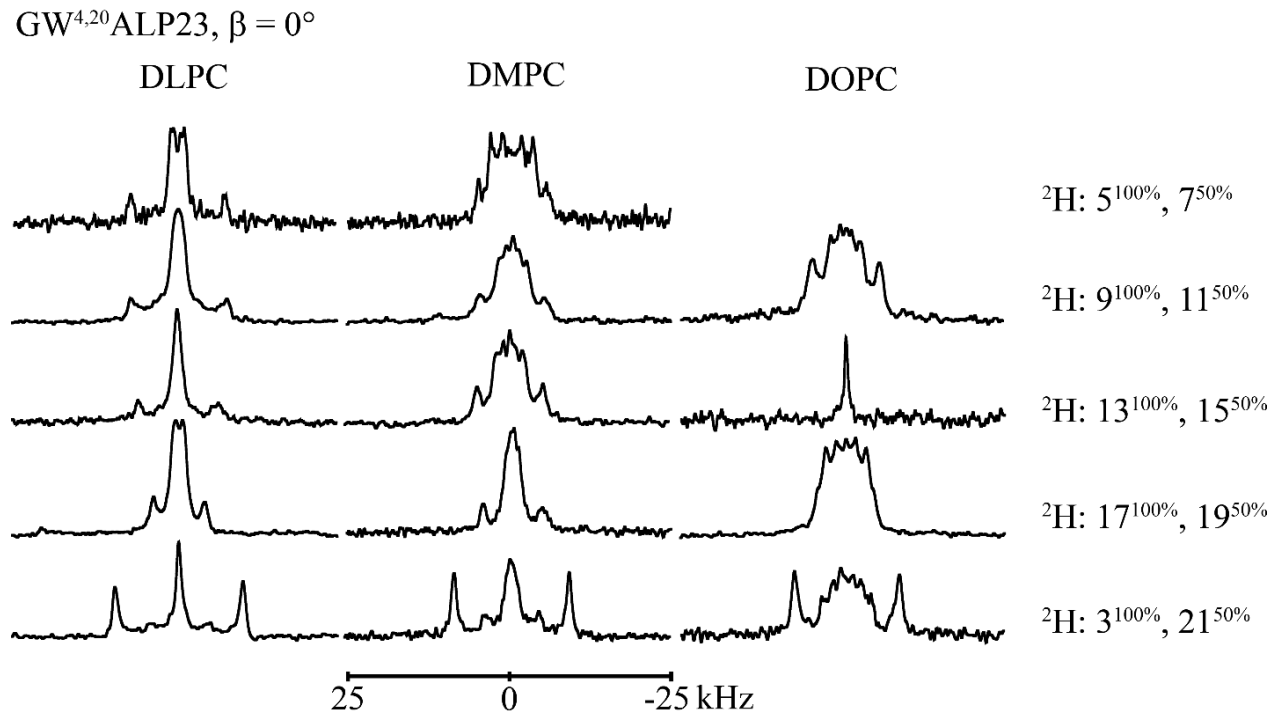


FIGURE S4.

Deuterium (^2H) NMR spectra for labeled GW^{4,20}ALP23 in oriented DLPC, DMPC, and DOPC bilayers at $\beta=0^\circ$ measured at 50 °C. ^2H labeled alanine positions are depicted on the right.

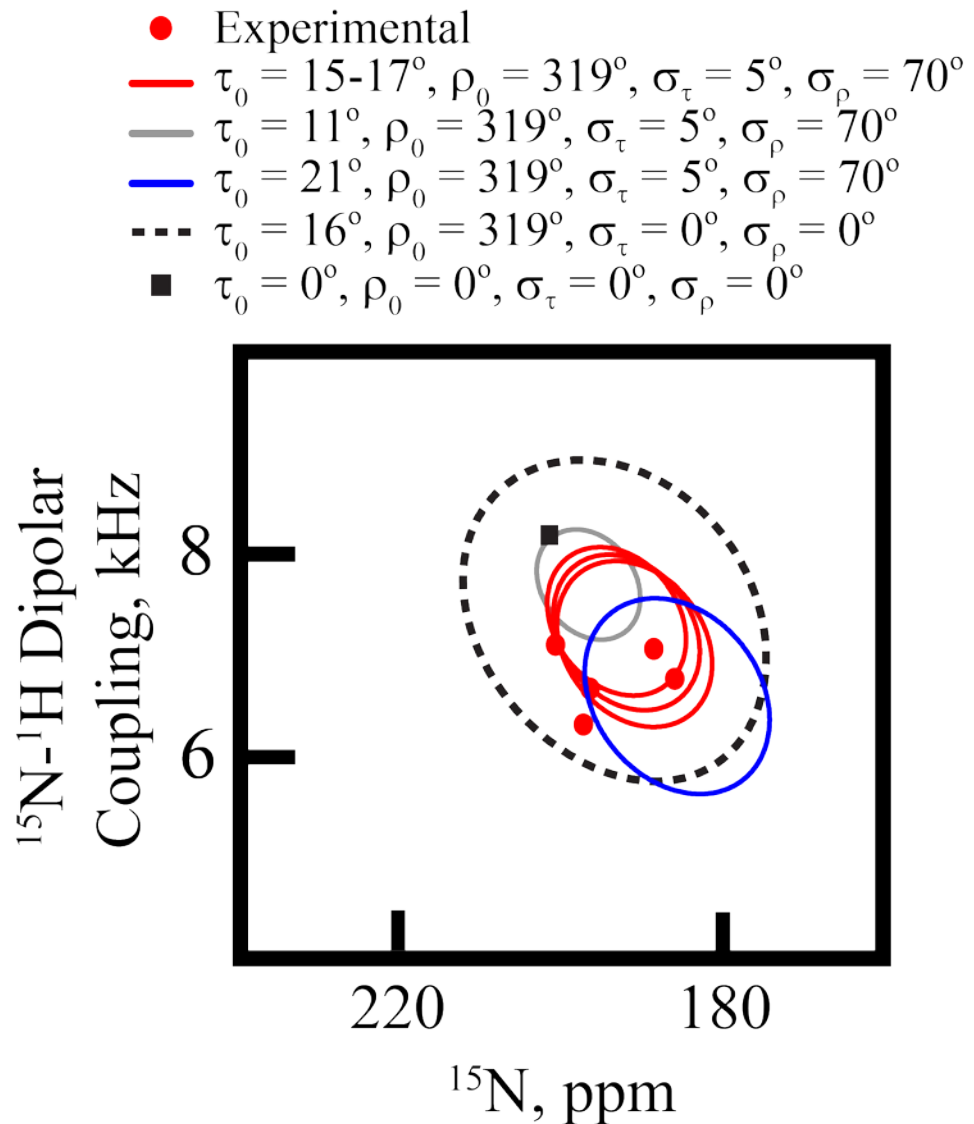


FIGURE S5.

Illustration of PISA wheels (for sample orientation $\beta = 0^\circ$) that fit and do not fit the data for GW^{4,20}ALP23, labeled with ^{15}N in residues 13-17, in oriented bicelles of DMPC/DHoPC (ether). The red PISA wheels are approximate fits to the red data points, with helix tilt values of 15° , 16° and 17° ; and moderate motion represented by σ_τ of 5° and σ_ρ of 70° . Larger or smaller values of the tilt angle (blue and gray wheels) do not fit the data. An ellipse with a correct tilt angle but no motional averaging (- - -) also does not fit. For a helix with zero tilt, the data points would collapse to the black square. Note that the red wheel sizes are much smaller and the motional averaging much more extensive than observed with GW^{5,19}ALP23 (see figure 6 of the main article).

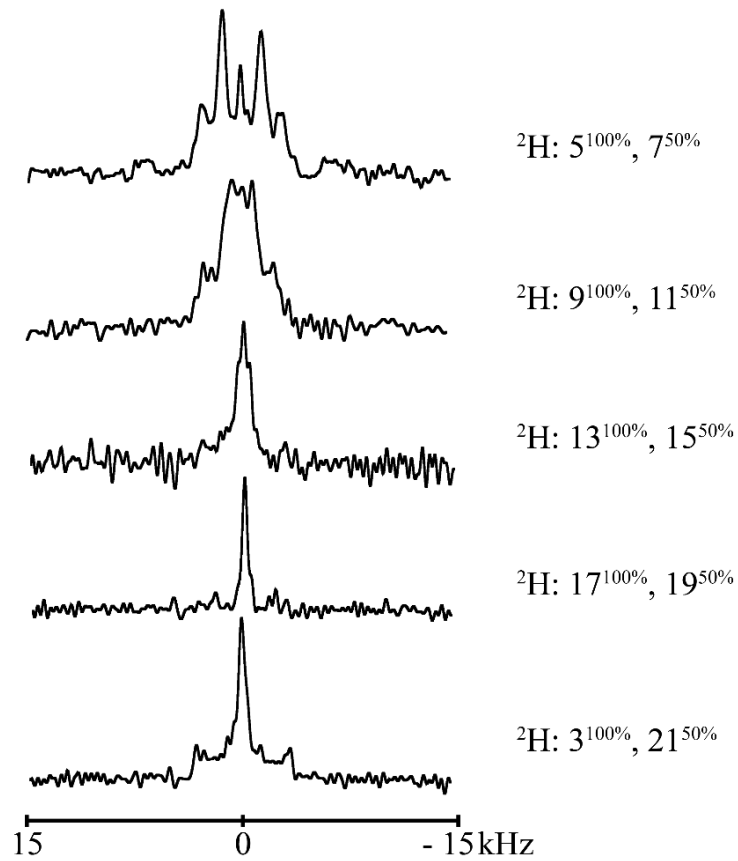
GW^{4,20}ALP23, DMPC/DH(o)PC Bicelles

FIGURE S6.

Deuterium (^2H) NMR spectra for labeled GW^{4,20}ALP23 in oriented DMPC/DH(o)PC bicelles measured at 42 °C. ^2H labeled alanine positions are depicted on the right.

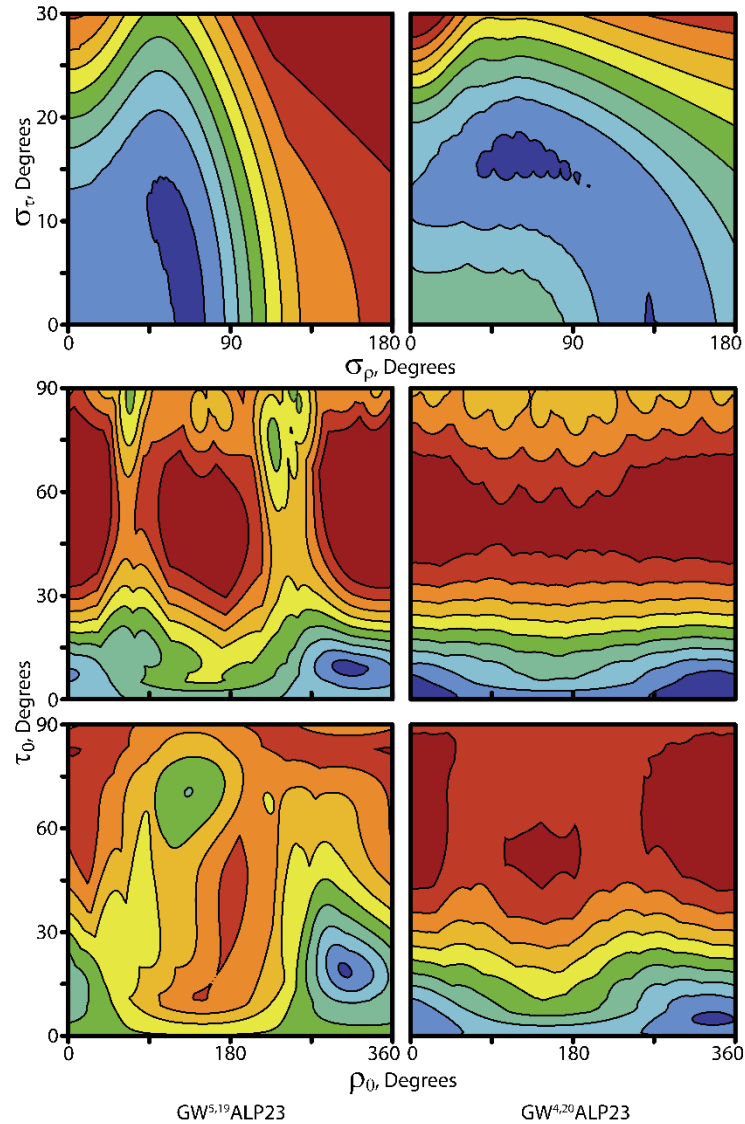


FIGURE S7.

Rmsd contour plots for $\text{GW}^{5,19}\text{ALP23}$ and $\text{GW}^{4,20}\text{ALP23}$ in DMPC/DHoPC bicelles using $^{15}\text{N} + ^2\text{H}$ data combined (Gaussian analysis) or ^2H data alone (GALA analysis). Contour levels are drawn using 10 contours starting at 0 kHz (blue) to the highest value (red). (Top) Gaussian distributions of the helix wobble σ_τ and rotational slippage σ_ρ (rmsd max left: 4.0 kHz; right: 3.9 kHz). (Middle) Average tilt and rotation from the GALA analysis (rmsd max left: 27.8 kHz; right: 25.6 kHz). (Bottom) Average tilt and rotation from the Gaussian analysis (rmsd max left: 9.0 kHz; right: 13.9 kHz).