**Supplementary File 1. Predicted and measured apparent transfer free energies of mGnRHR TM6.**

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| --- | --- | --- | --- |
| **Insert TMD** | **Kapp** ‡  **(Int. G1 / Int. G2)** | **ΔGapp** ‡  **(kcal/ mol)** | **ΔGapp, pred** †  **(kcal/ mol)** |
| WT TMD6 | 1.2 ± 0.2 | -0.1 ± 0.1 | 0.26 |
| V276T TMD6 | 0.7 ± 0.1 | 0.2 ± 0.1 | 0.805 |

‡Values represent the average of three experimental replicates, and errors represent the standard deviation.

†Values predicted from the TM6 sequence using the ΔG Predictor.