Free energy landscapes as a function of WE iterations 5 ΔG (kcal/mol) 3 2 0.8 0.0 0.2 0.4 1.0 3 3.5 5.0 5.5 6.0 0.8 1.0 1.6 1.8 2.0 SI_noCHOL **FLC CEI** 240 - 250 290 - 300 340 - 350 390 - 400 440 - 450 490 - 500 Configurational distribution as a function of WE iterations 500 400 WE iteration ΔG (kcal/mol) 300 200 100 0.0 0.2 1.0 3.0 4.0 5.5 6.0 **0.8** 1.0 1.8 4.5 SI_noCHOL **FLC** CEI Flux profile between states 0.010 0.008 **Plux** (T-1) 0.006 0.002 0.000 10 20 30 40 50 10 20 Iteration intervals Iteration intervals Iteration intervals ← Flux from separated to mixed ← Flux from mixed to separated State population profile 1.0 0.8 0.6

