



Figure S3. Selective constraint for each amino acid pair estimated from WAG and from LG. The ML estimate, $-\hat{w}_{ab}^{\text{WAG-ML91+}}$ in (A) and $-\hat{w}_{ab}^{\text{LG-ML91+}}$ in (B), of selective constraint on substitutions of each amino acid pair in the ML-91+ models fitted to the 1-PAM matrices of WAG and LG is plotted against the mean energy increment due to an amino acid substitution, $(\Delta\hat{\varepsilon}_{ab}^c + \Delta\hat{\varepsilon}_{ab}^v)$ defined by Eqs. S1-4, S1-5, and S1-6. The estimates \hat{w}_{ab} for the least exchangeable class of multi-step amino acid pairs are not shown. Plus, circle, and cross marks show the values for one-, two-, and three-step amino acid pairs, respectively.