Fig. S5. Fluorescence spectra of 11.3.3-L before (black) and after (blue) acid denaturation. Purified protein was unfolded in 100 mM citrate (pH 1.9) containing 2 mM DTT at room temperature for 1 hr. The denatured protein solution was diluted 100-fold into 20 mM Tris (pH 8.0), 100 mM NaCl, and 1 mM DTT. Incubation of GFP at pH 1.9 is known to cause some irreversible denaturation (2); however, the kinetics of refolding are independent of the extent of irreversible denaturation and the spectrum of the refolded protein is consistent with refolding to an authentic native structure.