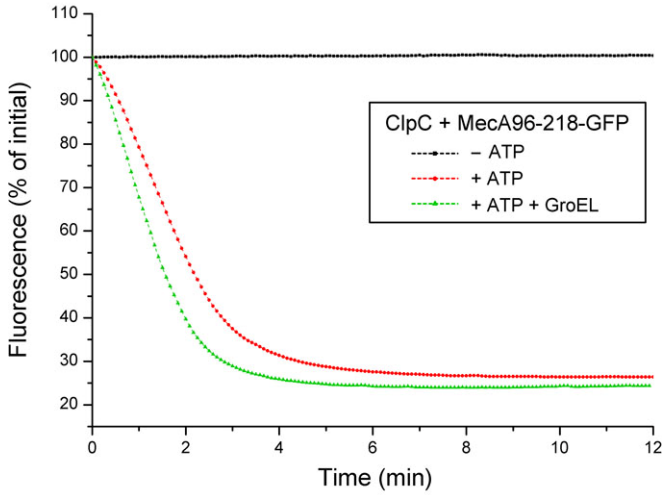
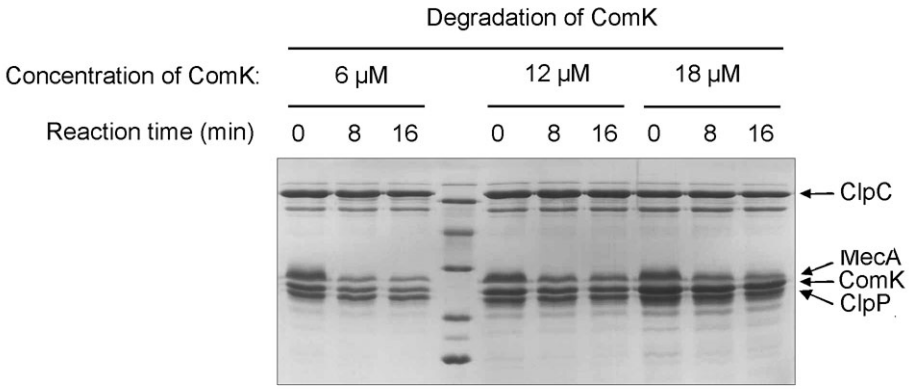
Supplementary Fig. 1



Supplementary Fig. 1. Refolding of the substrate protein only partially accounted for the observation that fluorescence intensity remained relatively steady after 7 minutes. Addition of the GroEL trap, which sequesters unfolded substrate protein, only reduced the fraction of folded substrate protein by 2% after 7 minutes. This observation suggests that binding equilibrium may be the primary reason for the "steady-state" of unfolding reaction.

Supplementary Fig. 2



Supplementary Fig. 2. MecA may not be reusable within the assembled ClpCP protease. A fixed concentration of MecA (3 μM) was incubated with ComK of three different concentrations (6, 12, and 18 μM) in the presence of ClpC, ClpP, and ATP. The extent of ComK degradation was examined by SDS-PAGE and coomassie staining. These results show that, while similar amounts of MecA was consumed in each of these three sets of reactions, higher concentrations of ComK failed to increase the extent of ComK degradation. This analysis suggests that MecA may not be reusable within the assembled ClpCP protease.