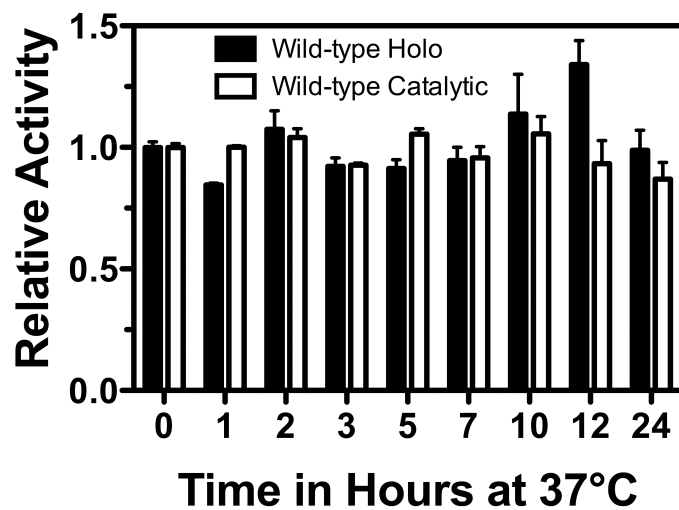


Supplemental Figure 1. Detection of two forms of human GCLC. Embryonic fibroblasts obtained from GCLC null mice were transiently transfected with wild-type or mutant human GCLC. hGCLC expression was monitored via western analysis of whole cell lysates. The primary antibodies used are anti-hGCLC (mouse polyclonal antibody, Covance; 1:4000) (**Panel A**) and anti-flag (mouse monoclonal antibody, Sigma; 1:1000) (**Panel B**). Goat anti-mouse Alexa Fluor 680 (Invitrogen), and goat anti-rabbit IRDye 800 (Rockland Immunochemicals) were used as secondary antibodies at 1:5000 dilution. Secondary antibodies were visualized using the Odyssey Infrared Imaging System (LI-COR Biosciences). As shown in **Panel A**, the polyclonal hGCLC antibody detected two forms of hGCLC, whereas the flag-specific antibody only detected only the upper band (**Panel B**). An overlay of the two detection methods is shown in **Panel C**. This suggests that the lower band may be a truncation of the intact protein, since the flag epitope is located at the C-terminus of the protein.



Supplemental Figure 2. Wild-type GCLC protein stability. Wild-type GCLC was incubated at 37 °C and relative enzymatic activity was monitored as a function of time in the absence (white bars) or presence (black bars) of hGCLM. During the 24 hour time course, wild-type enzyme did not show a significant loss of activity.