Supplementary Figure 1. HECT cysteine residue of KIAA10-CD is required for activity (autoradiograph). Steady-state chain synthesis assays (Ub74 substrate) were done with no E3 (lanes 1, 2), KIAA10-CD (lanes 3, 4), or a version of KIAA10-CD in which the HECT cysteine was mutated to alanine (lanes 5, 6). The low level of Ub2 seen in lane 6 is largely attributable to a weak background reaction of UbcH5A (see lane 2).