



**Figure S1.** E2 conjugating enzymes showing no activity in combination of TRIM32 full-length,  $\Delta$ RING or  $\Delta$ B-Box. Recombinant TRIM32 full-length (FL),  $\Delta$ RING ( $\Delta$ R) or  $\Delta$ B-Box ( $\Delta$ BB) was used as E3 ligase in *in vitro* ubiquitination reactions with a panel of E2 conjugating enzymes. In each panel the first lane is the reaction with no E3 (-). Control reactions without the addition of E2 enzymes are shown (No E2). Proteins were resolved by SDS-PAGE and membranes incubated with anti-Ubiquitin (top panels) or anti-TRIM32 (bottom panels). Asterisks indicate SDS-resistant high molecular weight species likely representing TRIM32 oligomers (see text).