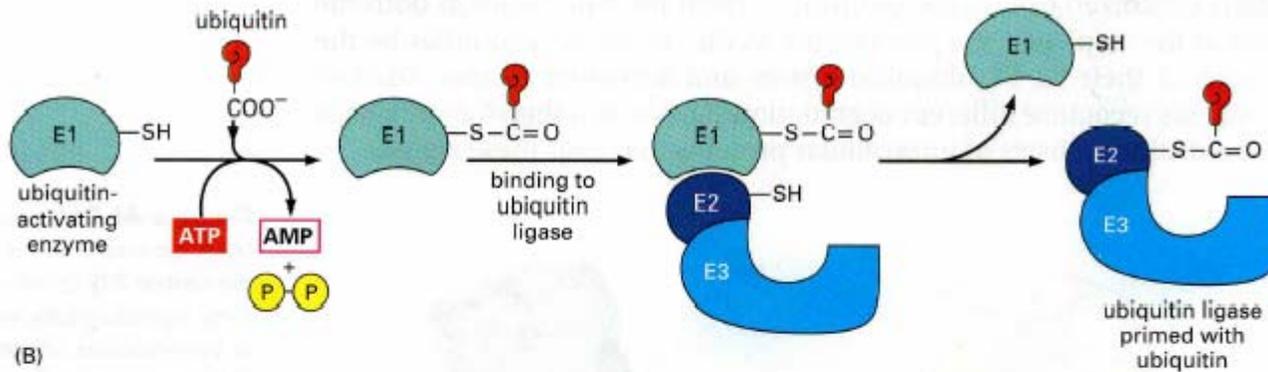


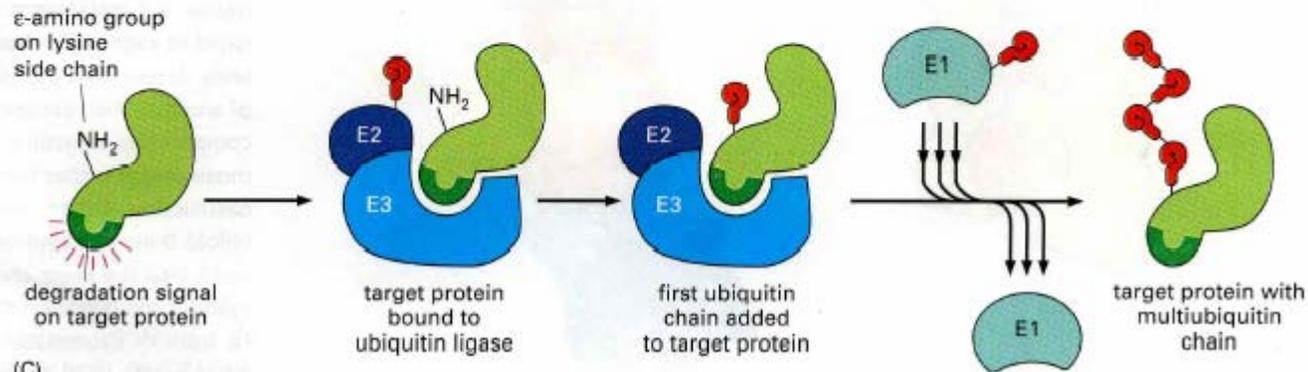
How Polyubiquitin Chains are Made and Unmade

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How a protein may become polyubiquitinated?



(B)



(C)

(Alberts, *The Cell*, p. 360)

Test of E1 as a Continuous Ub Source

E1.AMP-Ub*.S-Ub* (PULSE)

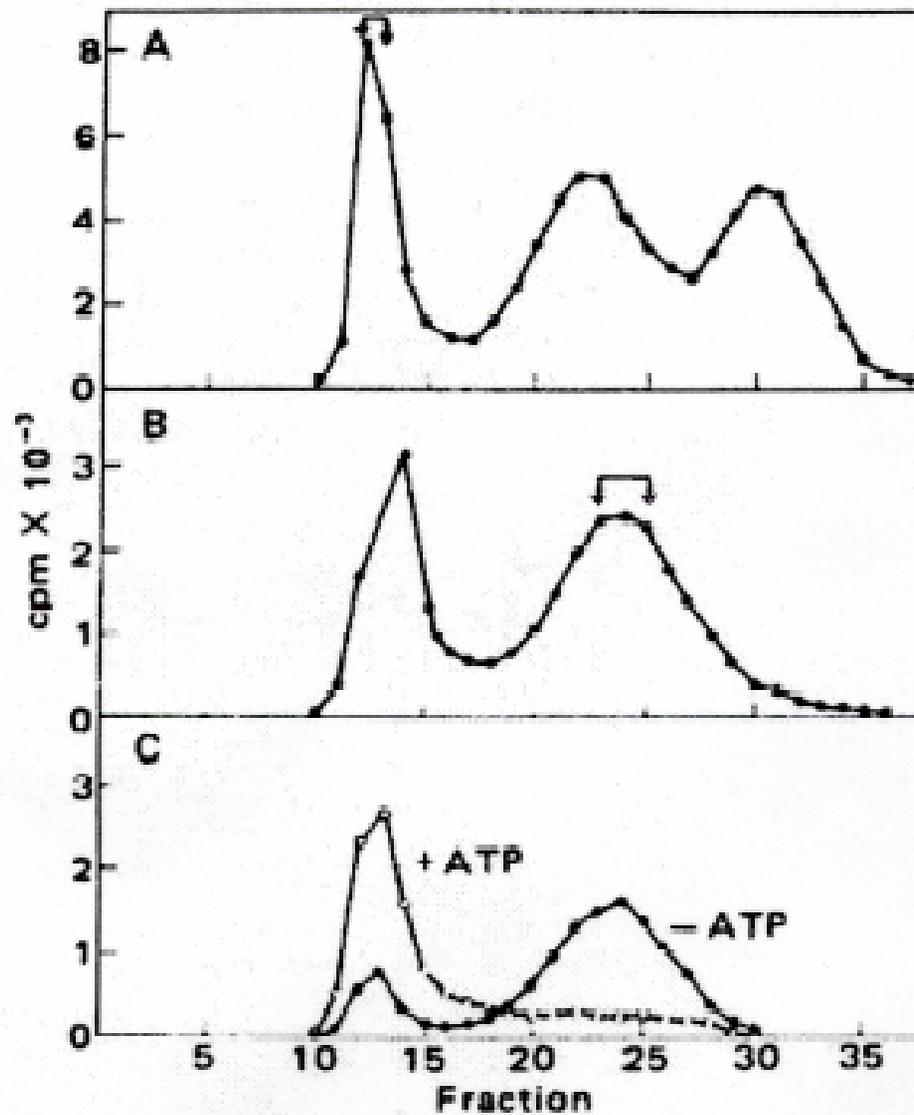


E2.E3.Protein + ATP+Ub (CHASE)



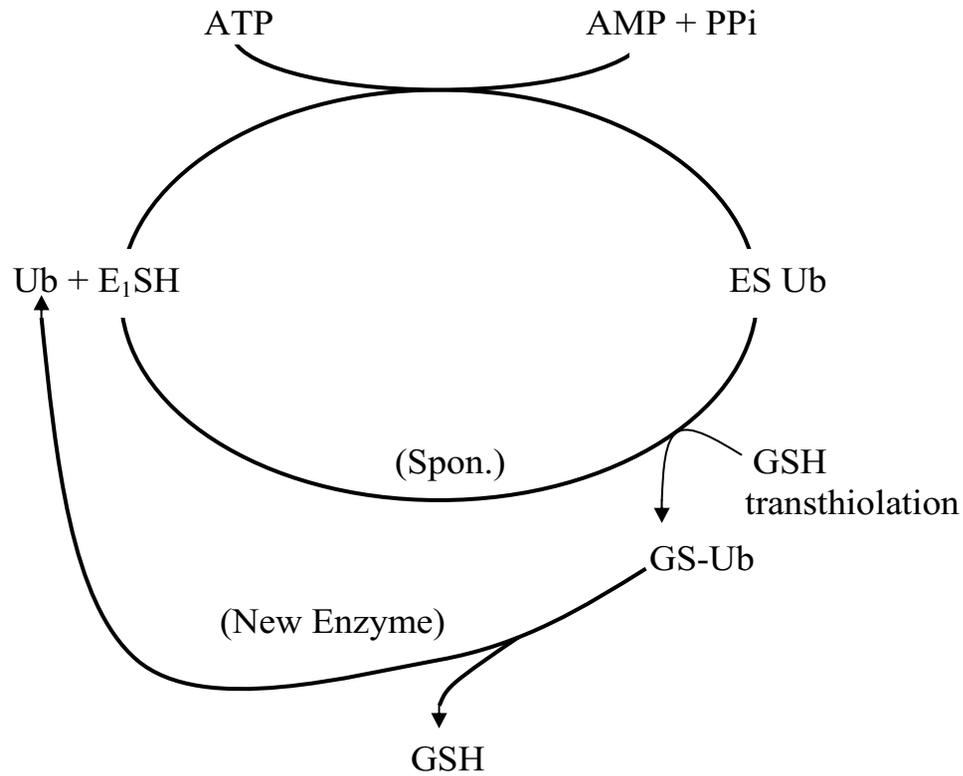
Protein-Ub-Ub*-Ub* if E1AMP-Ub → ES-Ub is fast

or Protein-Ub-Ub-Ub* if E3.Protein dissociates faster

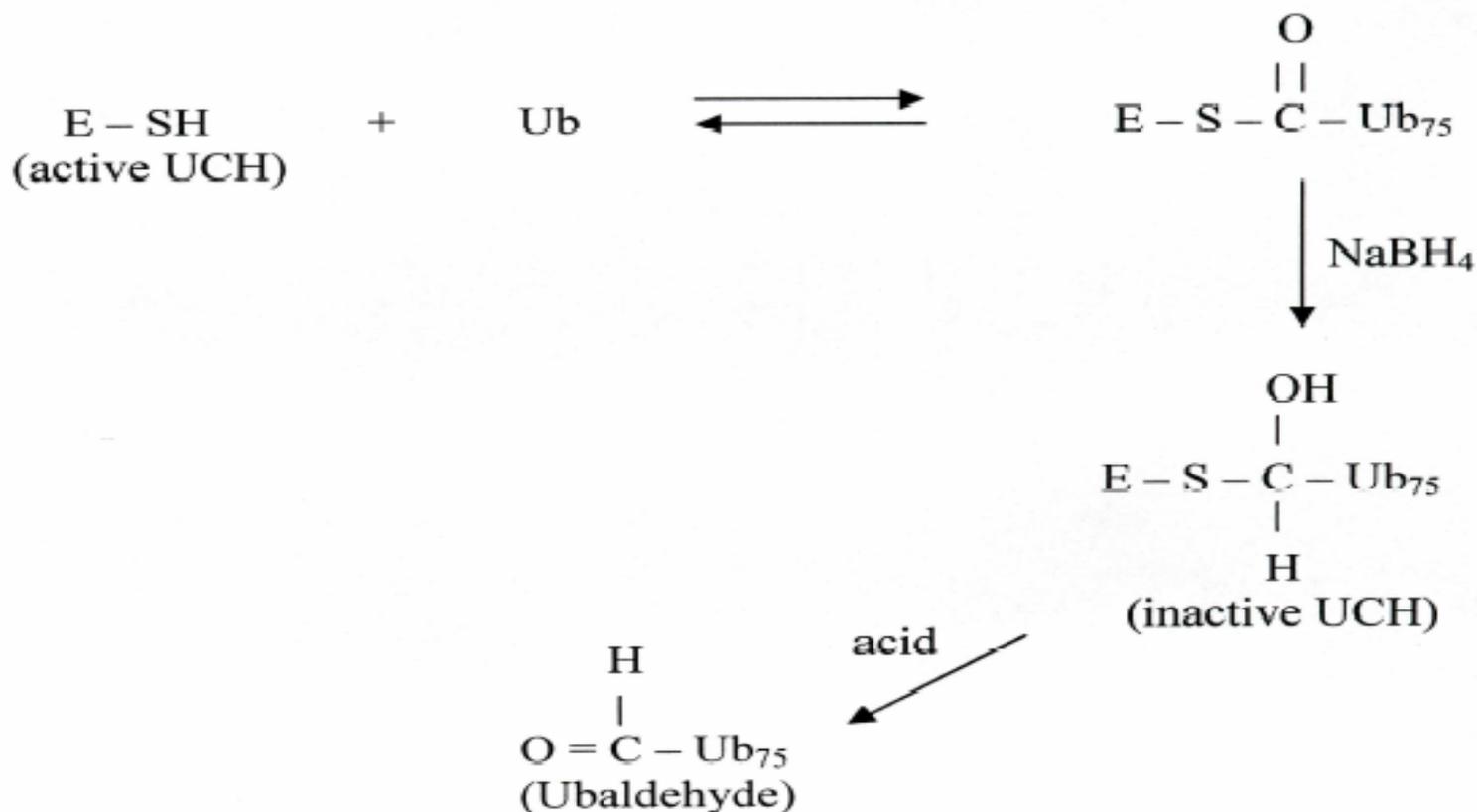


High mw conjugates formed with labeled Ub in reticulocyte extracts on G75 (A ∇) are shown to breakdown to Ub if ATP is withdrawn as in B as shown by their regeneration when ATP is added to (B ∇) (Hershko, et al, PNAS, 1980)

Ubiquitin Carboxy-terminal Hydrolase



Inactivation of UCH by NaBH₄



-
1. Free Ubal + NaBH₄ → Ub₇₅-ethanolamine
 2. Ubal binds UCH 1000x tighter than Ub
(Pickart and Rose, 1986)

Q: Why is Ubal a good inhibitor?

