E07-10-1092 Debnath

SUPPLEMENTAL FIGURES:

Supplemental Figure 1: Reduced Punctate GFP-LC3 in ATG-Depleted Cells During HBSS Starvation or EGF Withdrawal. MCF-10A cells expressing GFP-LC3 were transfected with control, ATG5, 6 or 7 siRNA and HBSS starved for 3h or EGF withdrawn for 24h. The number of GFP-LC3 dots per cell was enumerated using the "count nuclei" plug-in in Metamorph (v6.0); results are the mean+/-SEM enumerated from 40-80 cells for each condition.

Supplemental Figure 2: ECM signals regulate detachment-induced autophagy. (A) MCF-10A cells were grown attached for 48h or detached for 24h or 48h as indicated. In rBM "+" lanes, laminin-rich reconstituted basement membrane (2% Matrigel (v/v) final concentration) was added at 24h. (B) Left: MCF-10A cells were incubated for 24h with rat IgG isotype control or anti-β1 integrin subunit function-blocking antibody (A2B2), both at 10 μg/ml. Right: MCF-10A cells were incubated for 24h with rat IgG isotype control or anti-E-cadherin function-blocking antibody (DECMA), both at 50 μg/ml. All lysates were subject to immunoblotting with α -LC3 and α - tubulin antibodies.

Supplemental Figure 3: Bim is induced during *anoikis* in ATG-depleted cells. Cells transfected with the indicated siRNAs were grown attached (A) for 48h, or suspended for the indicated times. Cells were lysed, and subject to immunoblotting with α -Bim and α -tubulin antibodies. Note that the same blot was probed in experiments shown in Figure 4D; hence, the same tubulin loading control is shown for these two figures.





