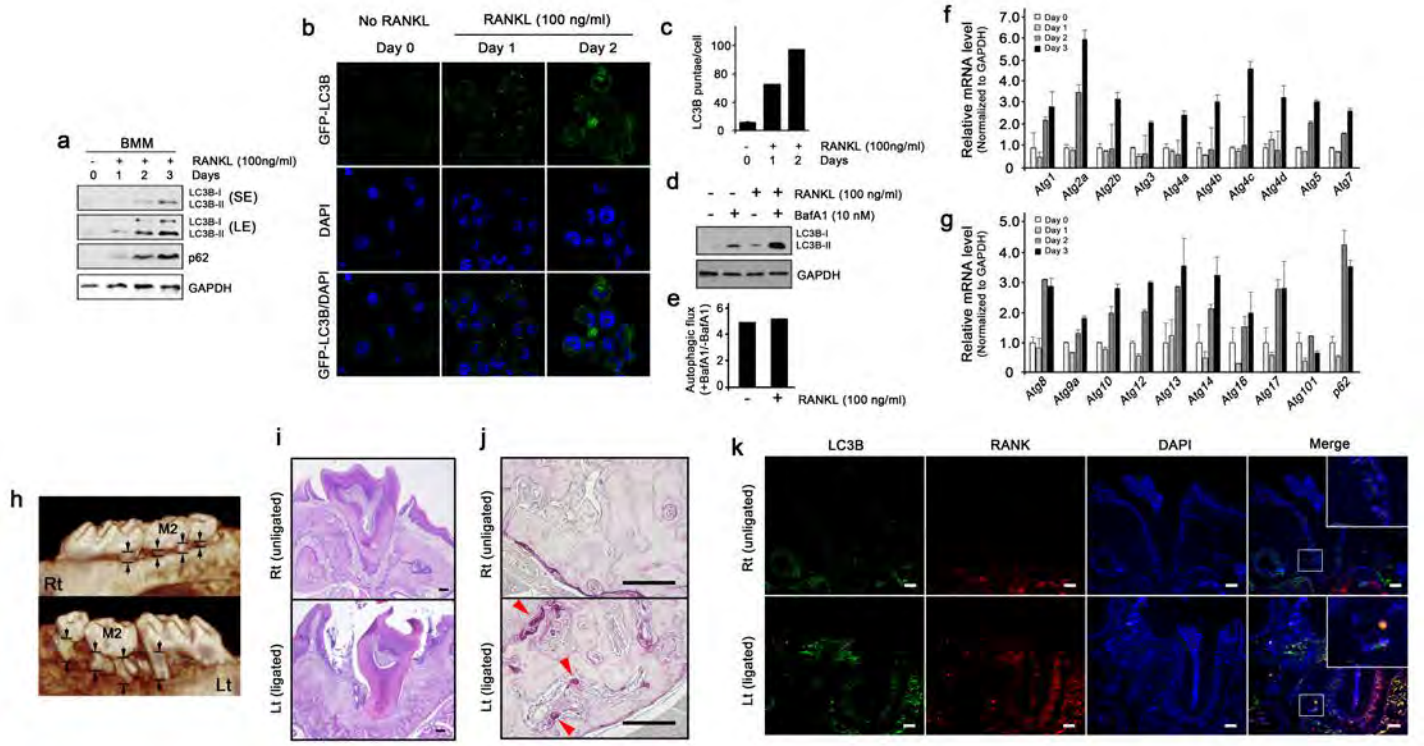
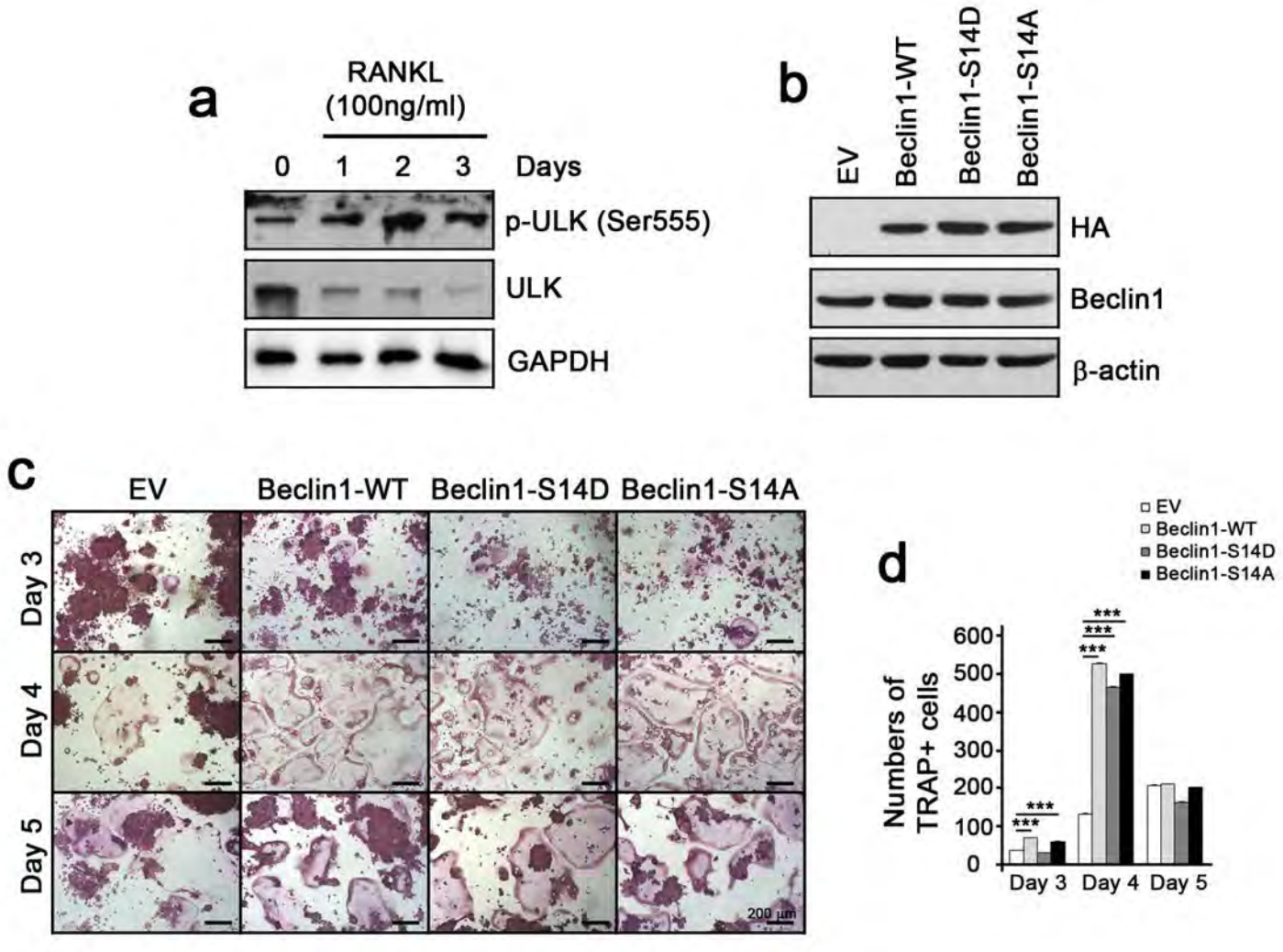
Supplementary Figure 1



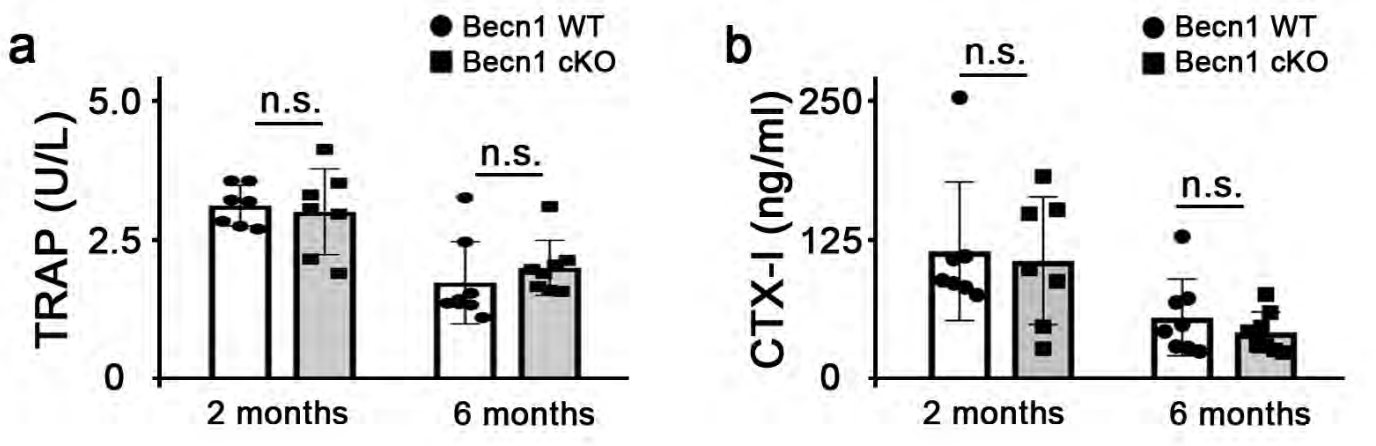
Supplementary Figure 1. Autophagy is induced during osteoclast differentiation. (**a**) Western blotting against LC3B, p62, and GAPDH in BMMs treated with RANKL (100 ng/ml) and M-CSF (10^4 U/ml) in a time-dependent manner. (**b**) Confocal microscopy for the fluorescent punctae in GFP-LC3B-overexpressing RAW 264.7 cells treated with RANKL (100 ng/ml). (**c**) Quantification of the fluorescent punctae per cell (n=50). (**d**) Western blotting against LC3B in BMMs treated with RANKL/M-CSF and bafilomycin A1 (BafA1, 10 nM) for 1 day. (**e**) Quantification of autophagic flux using ImageJ from 3-independent Western blotting. (**f,g**) Real-time qPCR for autophagy-related genes in BMMs treated with RANKL/M-CSF in a time-dependent manner (n=3). (**h**) Lateral μCT views from the buccal sides spanning from the first to the third maxillary molars. (**i**) H&E staining of the secondary maxillary molars from the unligated side (left panel) and the ligated side (right panel) (**j**) TRAP staining of the unligated (upper panel) and ligated (lower panel) tooth areas obtained from the mouse maxilla in the ligature-induced periodontitis model. Arrow indicates the TRAP+ multinucleated osteoclasts. Bar = 100 μm. (**k**) Immunofluorescent staining for LC3B and RANK at the unligated (upper panel) and ligated (lower panel) tooth areas obtained from the mouse maxilla. Bar = 200 μm.

Supplementary Figure 2



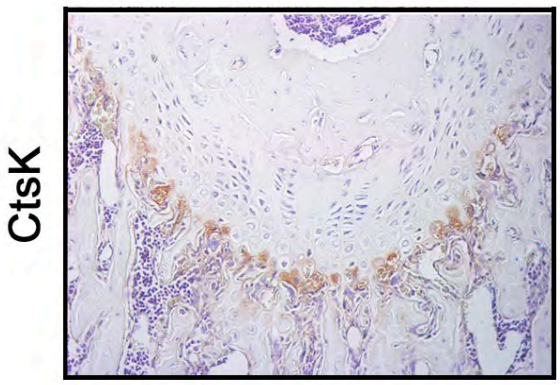
Supplementary Figure 2. Phosphorylation of Beclin1 at S14 is dispensable in RANKL-induced osteoclast differentiation. (**a**) Western blotting for p-ULK (Ser555), ULK and GAPDH in RANKL-treated RAW 264.7 cells. (**b**) Western blotting against HA, Beclin1, and β-actin in RAW 264.7 cells infected with retroviruses harboring empty vector (EV) or wildtype Beclin1 (Beclin1-WT), constitutively phosphorylated mutant Beclin1 at S14 (Beclin1-S14D), and phosphorylation incompetent mutant Beclin1 at S14 (Beclin1-S14A). (**c**) TRAP staining following RANKL treatment on RAW 264.7 cells for 3, 4 and 5 days. Bar = 200 μm. (**d**) Quantification of TRAP+ cells.

Supplementary Figure 3



Supplementary Figure 3. The serum amounts of TRAP and CTX-I in the Becnl cKO mice. (**a**) TRAP ELISA was performed from serum obtained from Becn1 WT and cKO mice. (**b**) CTX-I ELISA was performed from serum obtained from Becn1 WT and cKO mice.

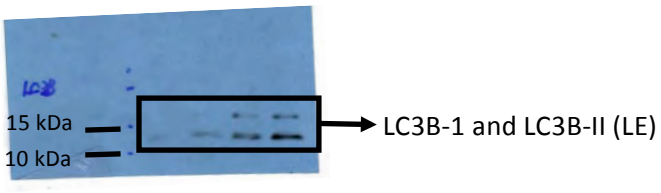
Supplementary Figure 4

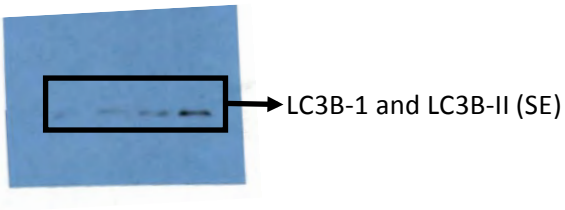


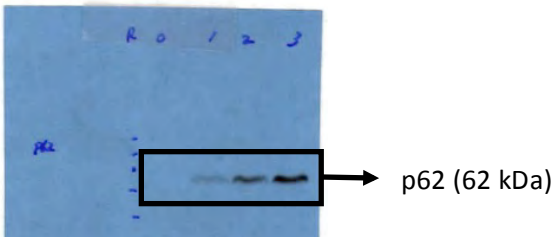
Supplementary Figure 4. CtsK is expressed at the hypertrophic zone of the cartilage.
Immunohistochemical staining against CtsK.

Supplementary Figure 5: Uncut raw Western data

Figure 1a







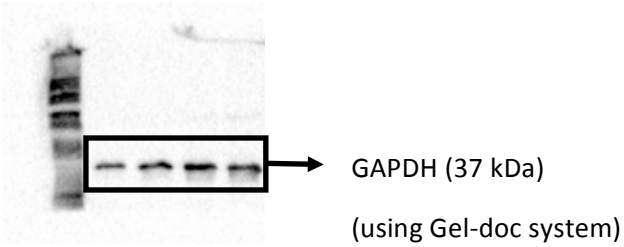
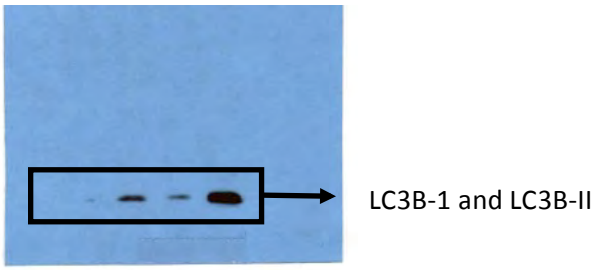


Figure 1d



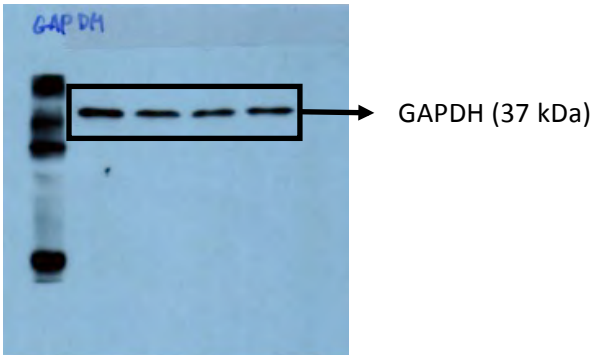


Figure 2b

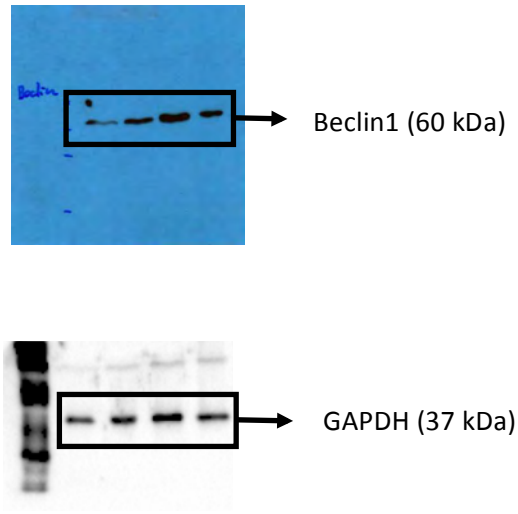


Figure 2c

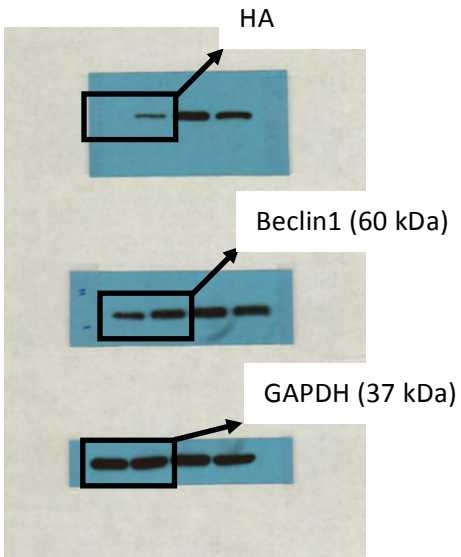
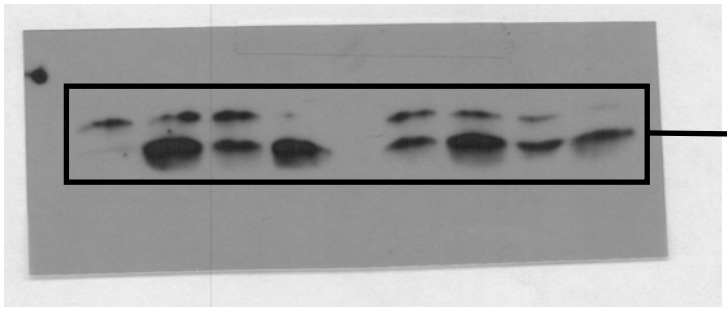


Figure 2k



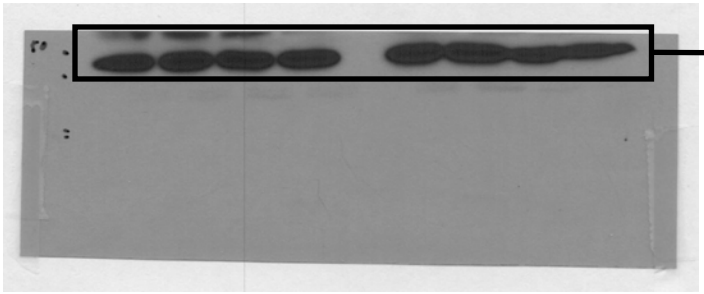


Figure 3a

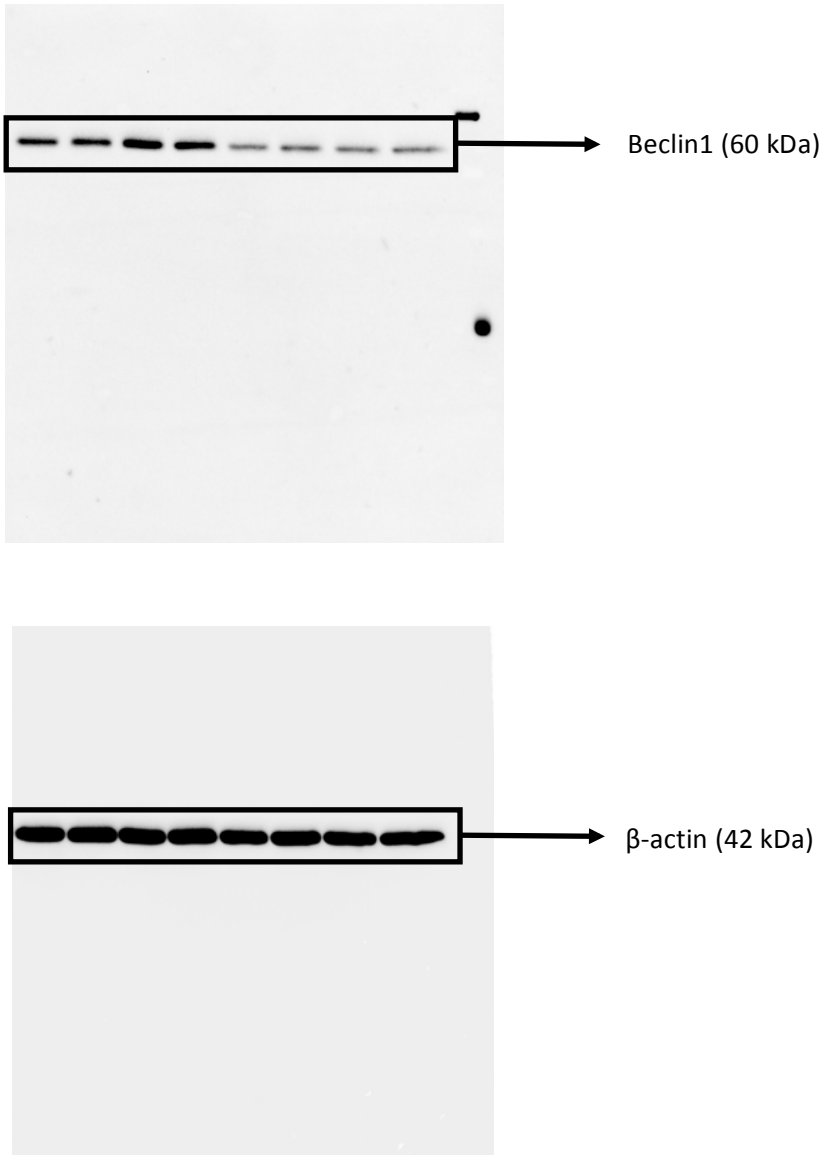
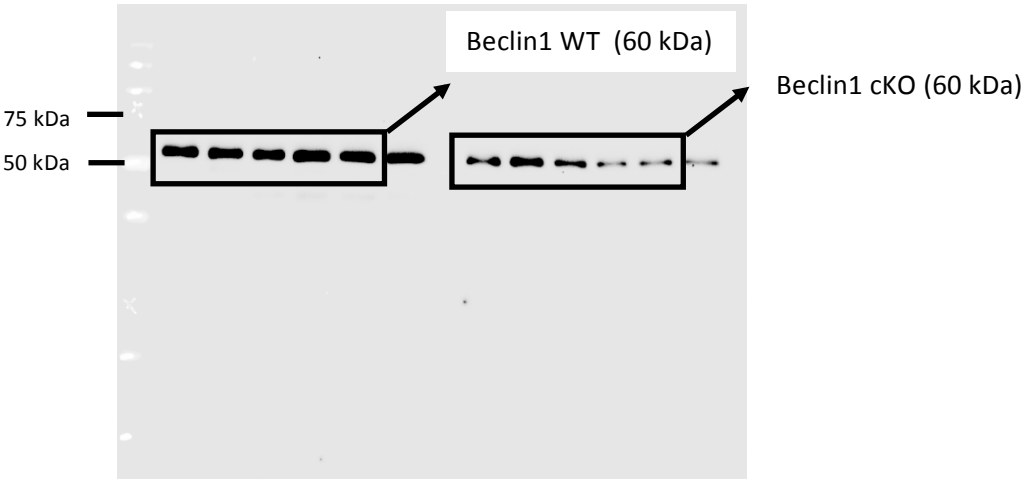


Figure 5b



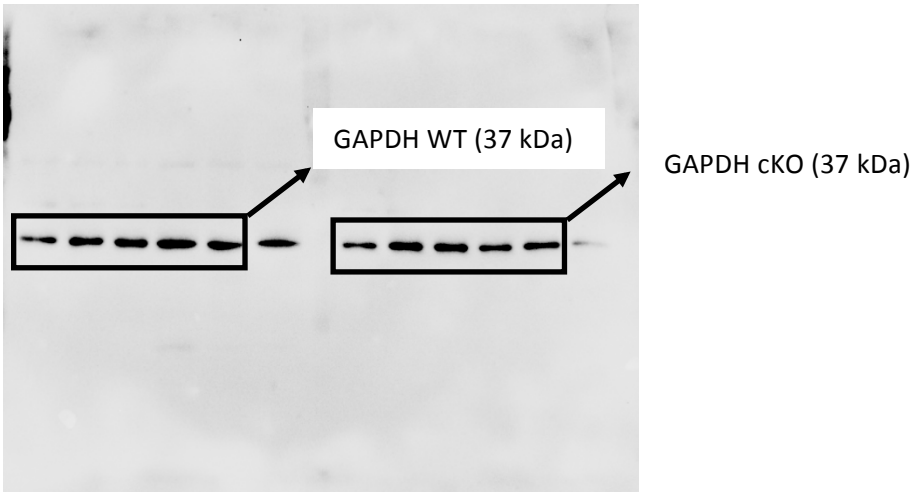


Figure 5i

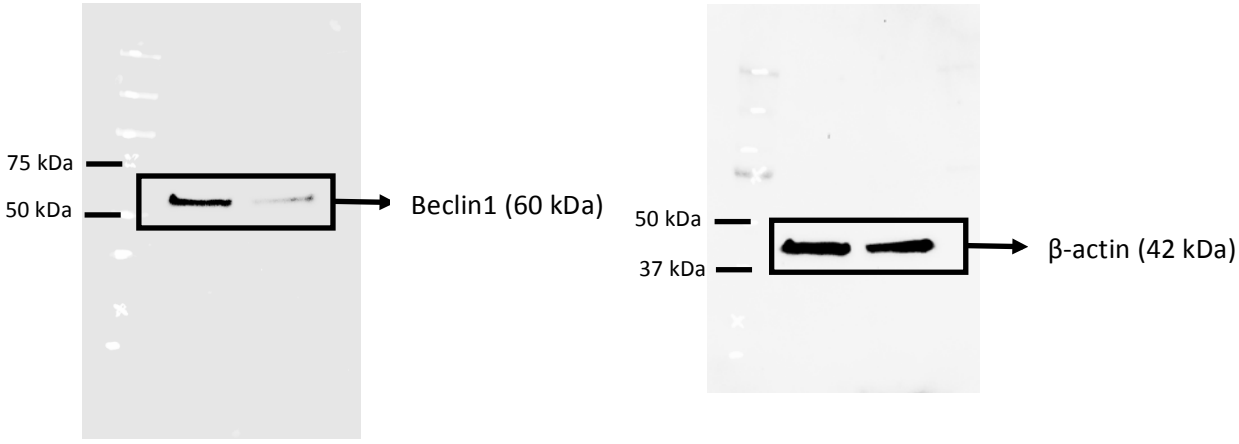


Figure 5j

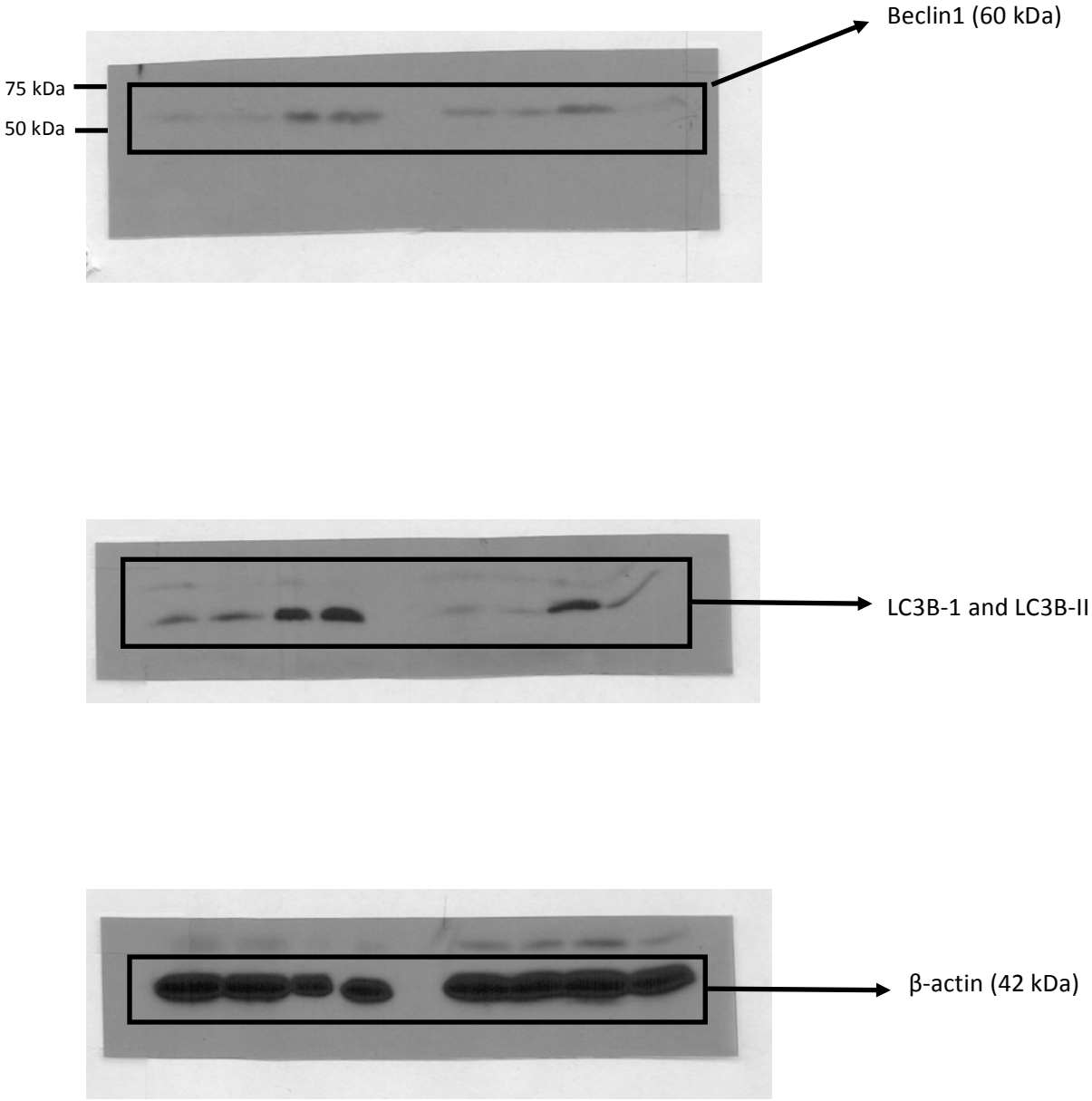
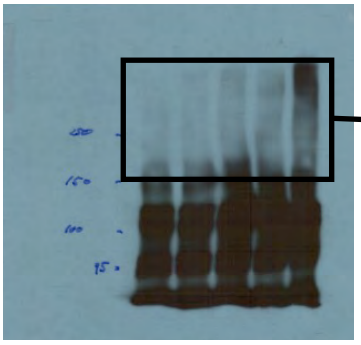
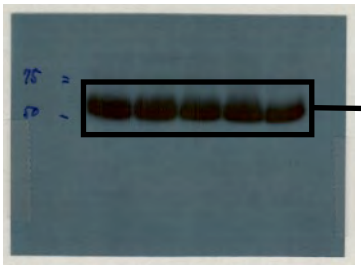


Figure 6a

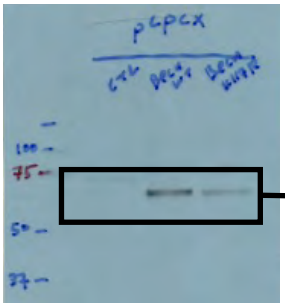


Blot: Ub

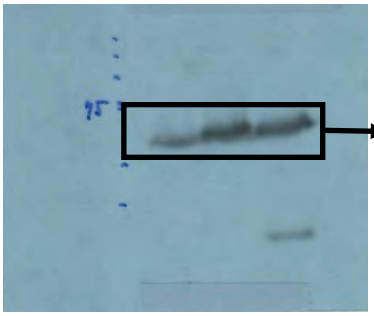


Blot: HA

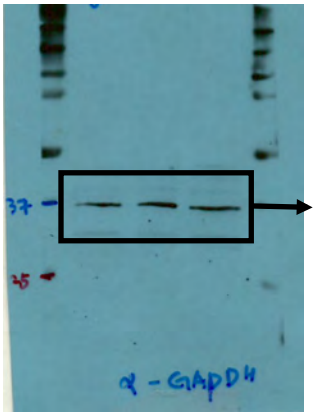
Figure 6c



HA



Beclin1 (60 kDa)



GAPDH (37 kDa)

Figure 6f

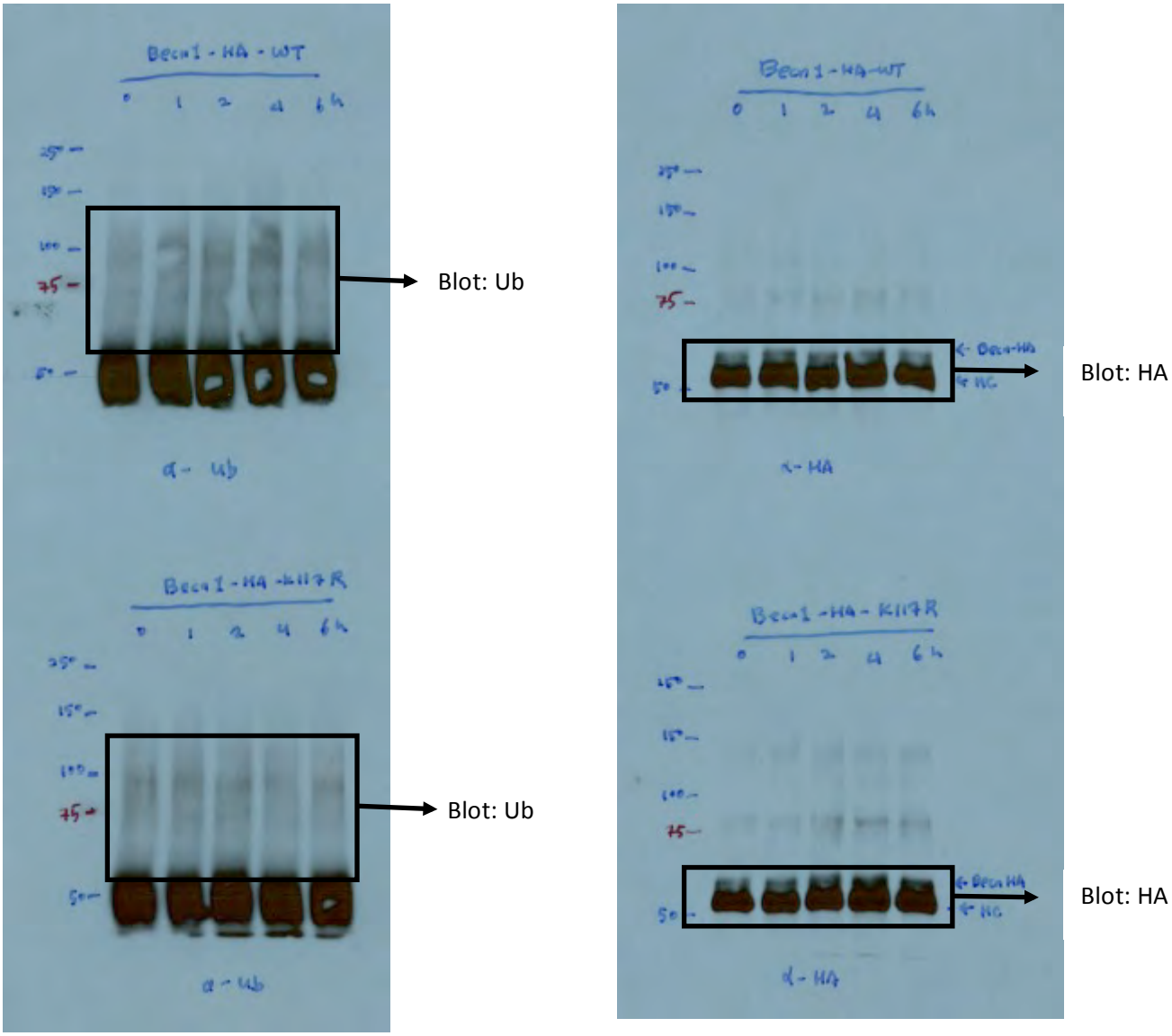
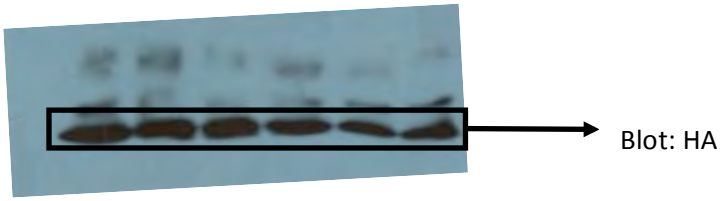

Becn1-HA-WT
0 1 2 4 6h
Blot: Ub
Becn1-HA-K117R
0 1 2 4 6h
Blot: Ub
Becn1-HA-WT
0 1 2 4 6h
Blot: HA
Becn1-HA-K117R
0 1 2 4 6h
Blot: HA

Figure 6g


Blot: Ub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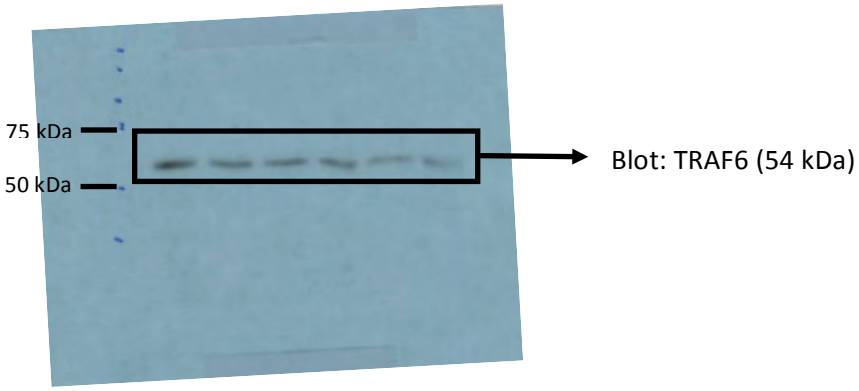
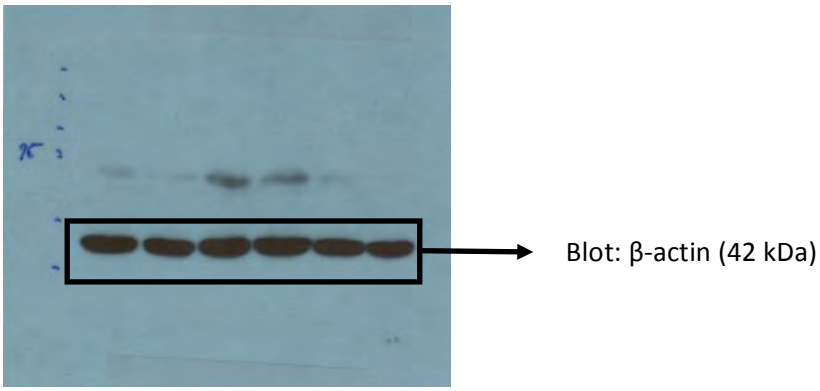

75 kDa
50 kDa
Blot: TRAF6 (54 kDa)


Blot: β-actin (42 kDa)