

Cianciola and Carlin, <http://www.jcb.org/cgi/content/full/jcb.200903039/DC1>

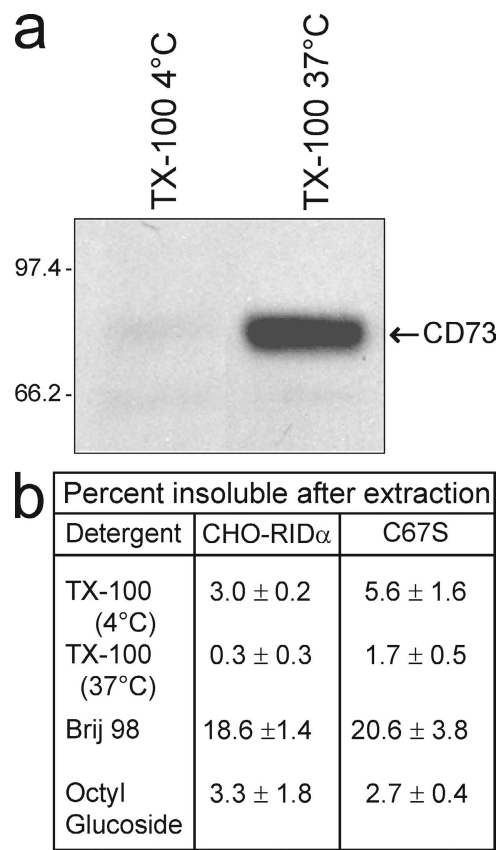


Figure S1. **Detergent extraction controls and quantification.** (a) Glycosylphosphatidylinositol-anchored protein CD73 is insoluble in Triton X-100 (TX-100) at low temperature. Cells were extracted with Triton X-100 at the indicated temperatures, and CD73 immunocomplexes from detergent-soluble and -insoluble fractions were immunoblotted with an antibody to the same protein. Molecular mass is indicated in kilodaltons. (b) Detergent extraction results for wild-type and RID- α (C67S) proteins from three independent experiments were scanned on a densitometer (GS-800; Bio-Rad Laboratories), and bands were quantified using ImageJ software (National Institutes of Health). Data are expressed as percent detergent insoluble relative to total RID- α and indicated as mean \pm SD.

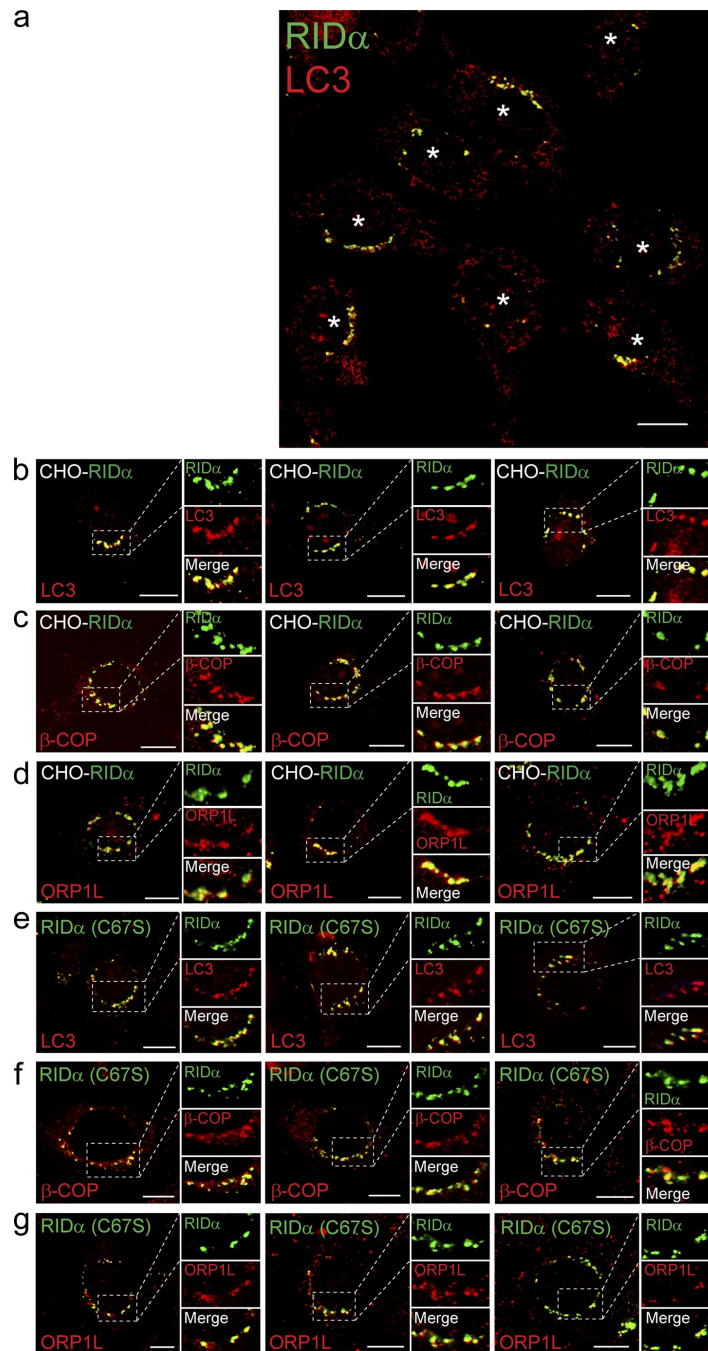


Figure S2. **RID- α induces formation of a hybrid organelle with characteristics of both endocytic and autophagic vesicles.** (a) Wide-field confocal image of CHO-RID- α cells stained for LC3 and RID- α , with asterisks indicating that all cells in the field have RID- α /LC3-positive vesicular compartments. (b–d) Multiple confocal images of CHO-RID- α cells stained for RID- α and LC3 (b), β -COP (c), or ORP1L (d). (e–g) Multiple confocal images of CHO-RID- α (C67S) cells stained for RID- α and LC3 (e), β -COP (f), or ORP1L (g). (b–g) Boxed areas show regions of the image that were magnified. Bars, 10 μ m.

Table S1. **Gene, probe, and primer details for real-time PCR**

Gene target	Primer	Oligonucleotide sequence
<i>GAPDH</i> (X52123)	Forward	5'-ATCTACTGGCGTCTTCACCAC-3'
Roche UPL probe #133	Reverse	5'-GGAGATGATGACCCCTCTTGG-3'
<i>HMGR</i> (X00494)	Forward	5'-CACGTCTCCTGTCGTGACC-3'
Roche UPL probe #50	Reverse	5'-CAGAGGCTCCCGTCTACAAC-3'
<i>CYP7B</i> (L04690)	Forward	5'-GGCAAACACTATTCCTGCAAC-3'
Roche UPL probe #50	Reverse	5'-CACTCAAATTGCTATCCACCTG-3'
<i>LDLR</i> (M94387)	Forward	5'-ACCATTTTGGAAGATGAGAAGC-3'
Roche UPL probe #133	Reverse	5'-CGTCCAAAATACTTTGCCTCA-3'

GAPDH, glyceraldehyde 3-phosphate dehydrogenase; UPL, universal probe library. Accession numbers (in parentheses) are from GenBank/EMBL/DDBJ.