Supplementary Material

ATG5 Promotes Death Signaling in Response to the Cyclic Depsipeptides Coibamide A and Apratoxin A

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Calculation of Cell Doubling Time in Cultured MEFs

MEFs were seeded in a 6-well plate (Corning) with 24,000 cells/well. Cell doubling time was calculated when cells were in the log phase of growth: $DT = T \ln 2/\ln(Xe/Xb)$ Where:

T = incubation time in any units Xb = cell number at the start of incubation time Xe = cell number at the end of incubation time

Results

MEFs ATG +/+

Mean \pm SEM 24.1 \pm 1.8; *N*=4

MEFs ATG -/-

Mean \pm SEM 19.7 \pm 2.4; *N*=4

Figure S1: page 2

Comparison of cellular proliferation and doubling time in untreated wild-type, ATG5-null MEFs.

Figure S2: page 3

Analysis of ATG5 expression in wild-type, ATG5-null and ATG5-null mouse embryonic fibroblasts re-expressing GFP-ATG5.

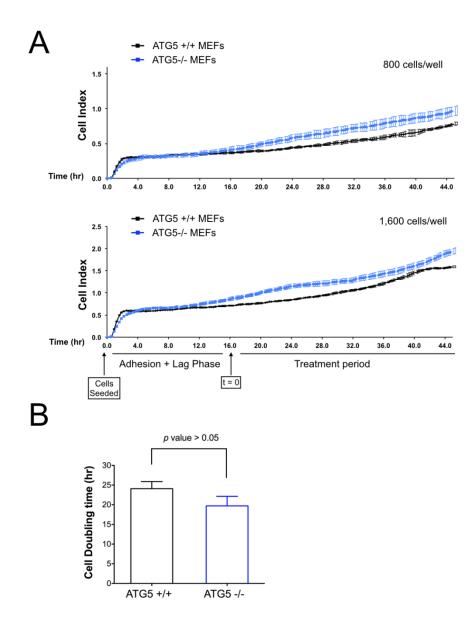


Figure S1. Comparison of cellular proliferation and doubling time in untreated wild-type, ATG5-null MEFs. (A) Continuous assessment of cellular proliferation using an xCELLigence real-time cellular analysis (RTCA) DP instrument (ACEA Biosciences, San diego, CA). Wildtype and ATG5-/-MEFs were seeded in E-plate with density of 800 or 1600 cells/well in triplicate in complete medium. Cells were allowed to equilibrate at room temperature for 30 min after seeded. E-plates were inserted into the xCELLigence instrument and impedance measurements were taken at 5 min interval for 8 h, and then at 15 min intervals for the remainder of the study. (B) Doubling time (h) of cells used in the study

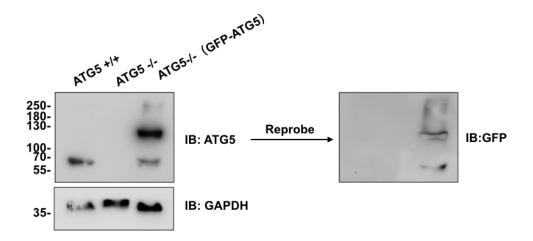


Figure S2. Analysis of ATG5 expression in wild-type, ATG5-null and ATG5-null mouse embryonic fibroblasts re-expressing GFP-ATG5. Immunoblot analysis of ATG5 expression (detected in the context of the covalent ATG5-ATG12 complex) in wild-type and ATG5-/- cells expressing GFP-ATG5. Whole cell lysates were probed with primary antibodies as indicated: anti-ATG5, anti-GFP, and anti-GAPDH.