

Supporting Information

Supporting experimental procedures

Yeast growth conditions

To prepare SCA medium, 0.5% Bacto™ Casamino Acid (Difco) was added to YNB medium.

Supporting Figure S1. Other nutrients except glucose and ethanol have no major impact on yeast growth in this study's condition. (A) Wild-type cells were inoculated into SCA medium, which contain only casamino acids as carbon sources, at a starting OD₆₀₀ of 0.01, and cultured in an automatically recording incubator. The OD₆₀₀ of the culture was measured every 10 minutes. Each result represents the mean ± S.E. ($n = 6$). Plots of OD₆₀₀ (grey plots and line) which are derived from Fig. 1B are depicted for comparison. (B) Ammonium concentration of the SD_{0.2}CA medium before or after cultivation were measured. To prepare after cultivation medium, wild-type cells were inoculated into SD_{0.2}CA medium at a starting OD₆₀₀ of 0.01, cultured for 48 hours, and collected the supernatant by centrifugation. Each bar represents mean ± S.E. ($n = 3$). (C) Wild-type cells were inoculated into SD_{0.2}CA medium (SD_{0.2}CA (1x NH₄⁺)) or SD_{0.2}CA medium supplemented with 5 g/L ammonium sulfate (SD_{0.2}CA (2x NH₄⁺)) at a starting OD₆₀₀ of 0.01. After 48 h-cultivation, OD₆₀₀ was measured. Each bar represents mean ± S.E. ($n = 5$).

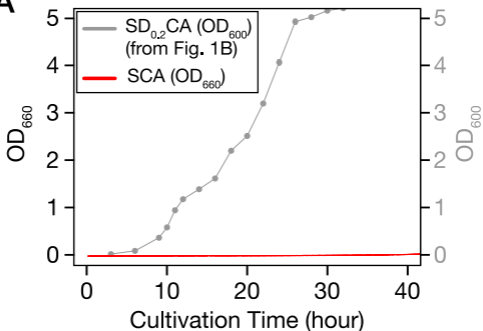
Supporting Figure S2. Vacuolar membrane with LD was invaginated into the vacuolar lumen. Wild-type cells were cultured for 22 hours (ethanol-utilizing) in SD_{0.2}CA medium. Images were acquired by transmission electron microscopy. Arrowheads indicate LDs associated with the vacuolar lumen. Bars: 1 μm.

Supporting Movie S1. Autophagic bodies were observed during ethanol-utilizing and ethanol-depleted phases. *atg15Δ* and *atg2Δatg15Δ* cells expressing GFP-Atg8 were inoculated into SD_{0.2}CA medium at a starting OD₆₀₀ of 0.01, and then cultured for 10 hours (glucose-utilizing), 22 hours (ethanol-utilizing), or 46 hours (ethanol-depleted). The cells were observed by fluorescence microscopy. Images were sequentially acquired (exposure time: 32 milliseconds). Movies were re-constructed using these images (0.5 × play speed). Bars: 5 μm.

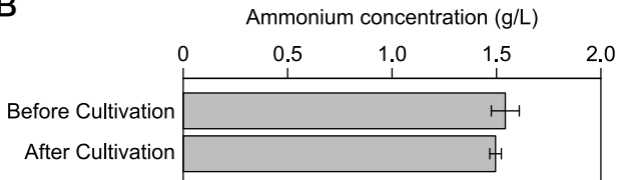
Supporting Movie S2 LDs are incorporated into the vacuolar lumen. Wild-type cells were cultured for 52 hours in SD_{0.2}CA medium. Cells were stained with BODIPY and observed under a fluorescence microscope. Images were sequentially acquired (exposure time: 32 milliseconds). Movies were re-constructed using these images (0.5 × play speed). Bar: 5 μm.

Figure S1

A



B



C

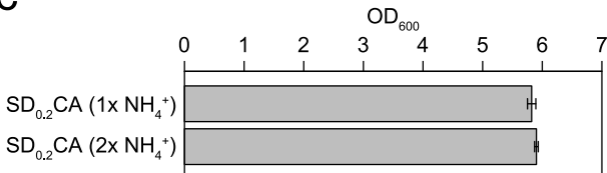


Figure S2

Ethanol-utilizing

