Supplemental Material Henard et al., Ribulose-1,5-bisphosphate carboxylase/oxygenase (RubisCO) is essential for growth of the methanotroph Methylococcus capsulatus.

Supplemental Figures

Figure S1. a) The percent of Methylovivimicrobium alcaliphilum 20ZR (Ma) and Escherichia coli (Ec) biomass derived from 13CO2 determined by isotopic elemental analysis after 72 h cultivation with 20% unlabeled CH4 and 8% 13CO2 (Ma) or 8% 13CO2 only (Ec) in the gas phase of serum vials. b) Growth of M. alcaliphilum 20ZR in a sealed serum vial or a continuous gas reactor (CGR) with 20% CH4 in air only (white bar) or supplemented with 2% CO2 (gray bar). c) Culture density (OD600) of two independent Methylococcus capsulatus Bath strains obtained from Mary Lidstrom’s laboratory (ML) or ATCC (strain 33009) and M. capsulatus Texas from ATCC (strain 19069) after 72 h of growth in a CGR supplied with 20% CH4 in air (CH4 only) or 20% CH4 and 2% CO2 in air (CH4/CO2) at 1 volume gas mixture/volume medium/minute. d) M. capsulatus growth in a CGR supplied with 20% CH4 in air at 0.1 volume gas mixture/volume medium/minute. The data represent the mean ± standard deviation of 4-6 biological replicates from two independent experiments.
Figure S2. Amino acid mass isotopomer distributions after $^{13}$CO$_2$ isotopic labeling. Amino acids and their respective metabolite precursors are color-coded: glycine and serine from 3-phosphoglycerate (3PG, light blue); histidine from ribose-5-phosphate (R5P, orange); phenylalanine and tyrosine from phosphoenolpyruvate (PEP, green) and erythrose-4-phosphate (E4P, green); alanine from pyruvate (PYR, gray); isoleucine, threonine, and aspartate from oxaloacetate (OAA, yellow); and glutamate and proline from alpha ketoglutarate (AKG, pink). The data represent the mean mass isotopomer distribution vector of three biological replicates.
Supplemental Tables

Table S1. *M. capsulatus* relative transcripts per million (TPM) during cultivation with continuous 20%CH₄/2% CO₂ in air.
See accompanying Excel spreadsheet.

Table S2. Isotopomer analysis.
See accompanying Excel spreadsheet.