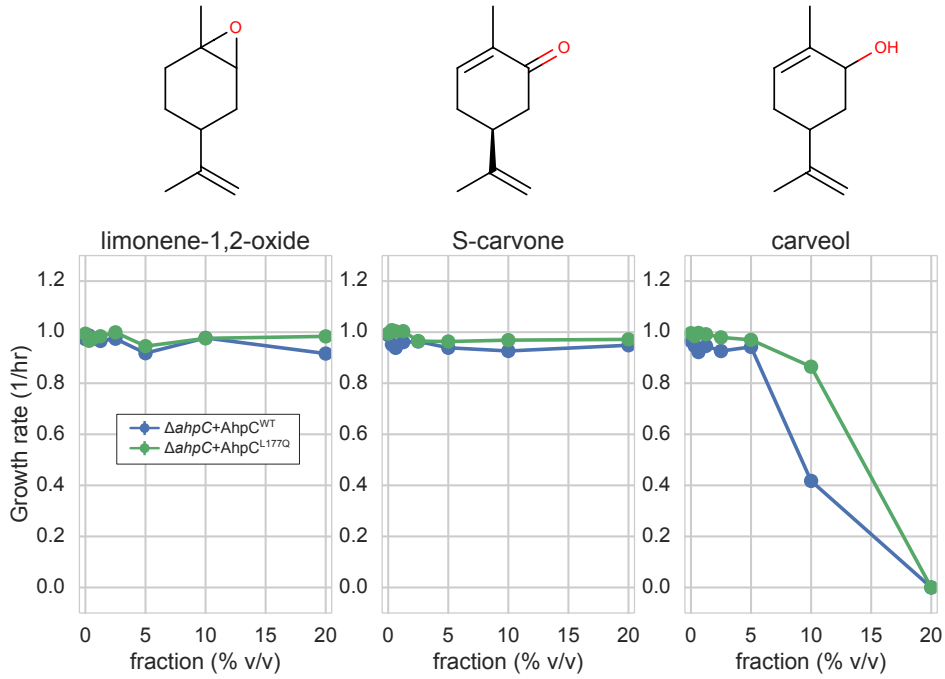
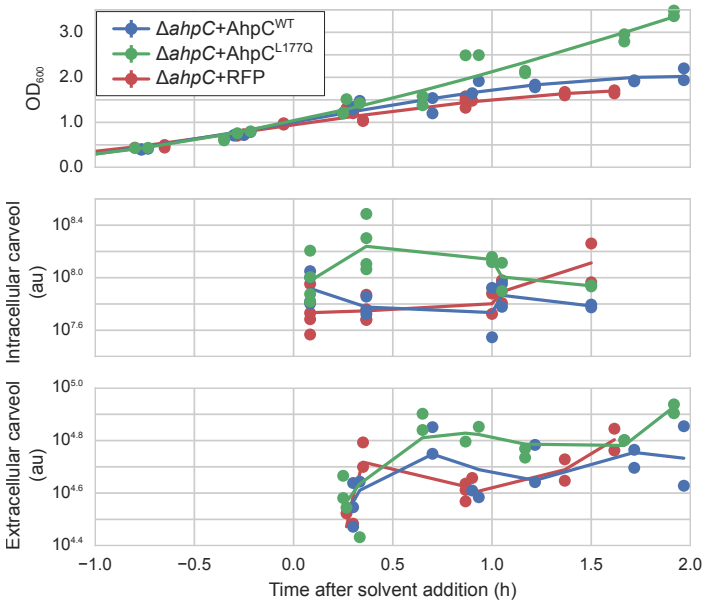
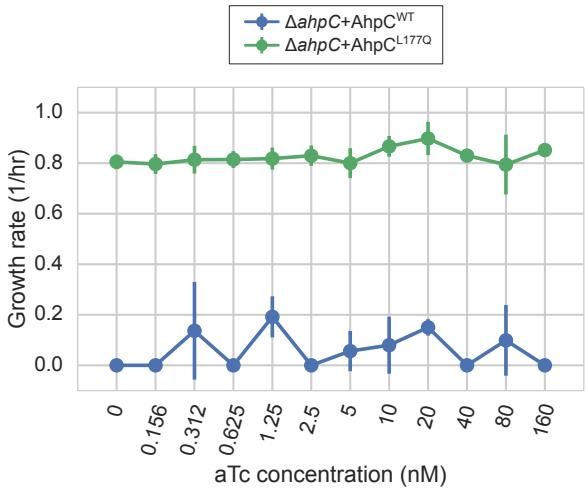


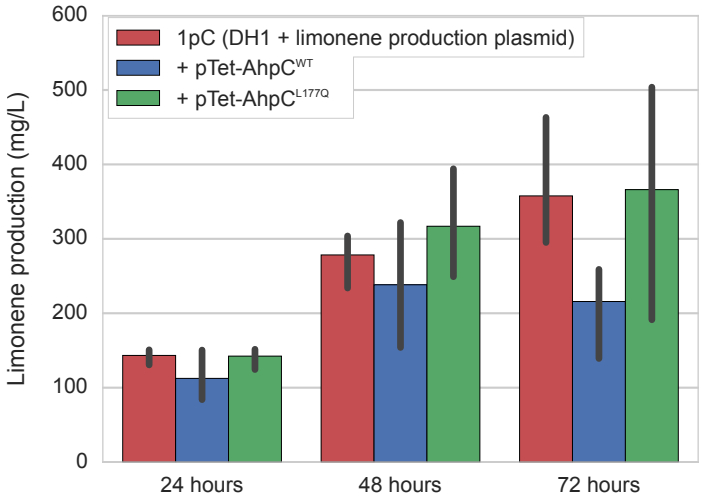
Supplementary Figure S1. Effect of other oxidized forms of limonene on growth. Non-oxidized, anaerobically stored limonene was mixed with limonene-1,2-oxide, S-carvone, or carveol, and this mixture was added at 1% v/v to the growth medium. Limonene-1,2-oxide and carveol were purchased as mixtures of isomers.



Supplementary Figure S2. Kinetics of carveol accumulation upon limonene addition to growing cultures. 1% v/v of partially oxidized limonene was added at t=0. Four independent biological replicates (from two different days) are shown together. Lines are spline fits.



Supplementary Figure S3: Growth rate as a function of inducer concentration. 2% v/v of partially oxidized limonene with an intermediate level of toxicity was used.



Supplementary Figure S4. Production of limonene by engineered *E. coli*. Strain 1pC from Alonso-Guttierez et.al (1) was transformed with no plasmid (red), or plasmids expressing $AhpC^{WT}$ (blue) or $AhpC^{L177Q}$ (green). Following protocols identical to previous work (1), cells were grown in EZRich media with 10 g/L glucose. Limonene was captured by a dodecane overlay and quantified by GC-MS. See Supplementary Methods below for additional details.

	Name	Description
Plasmid	pVC5006	pTet-*AhpC*^{L177Q}
Plasmid	pVC5005	pTet-*AhpC*^{WT}
Plasmid	pBbS2k-RFP	pTet-*RFP*
Plasmid	JPUB_004931	pBbA5c-MTSA-T1f-MBI(f)-T1002-Ptrc-trGPPS(co)-LS (limonene biosynthesis pathway)
Strain	JBx_038308	FM003. Spontaneous mutant evolved from KEIO *ΔdmsD*
Strain	JBx_038290	BW25113+pVC5006
Strain	JBx_038289	BW25113+pVC5005
Strain	JBx_038288	BW25113+pBbS2k-RFP
Strain	JBx_038293	*ΔahpC*+pVC5006
Strain	JBx_038292	*ΔahpC*+pVC5005
Strain	JBx_038291	*ΔahpC*+pBbS2k-RFP
Strain	JBx_038299	*ΔacrB*+pVC5006
Strain	JBx_038298	*ΔacrB*+pVC5005
Strain	JBx_038297	*ΔacrB*+pBbS2k-RFP
Strain	JBx_038296	*ΔacrA*+pVC5006
Strain	JBx_038295	*ΔacrA*+pVC5005
Strain	JBx_038294	*ΔacrA*+pBbS2k-RFP

Supplementary Table S1. Plasmids and strains used in this work. pTet: tetracycline inducible promoter (2). BW25113 and all single-gene knockout strains are from the KEIO collection (3) with the kanamycin resistance cassette removed as described in the reference.

Supplementary Methods

Production of limonene was done using the plasmid and protocol used for strain 1pC as previously described (1). Briefly, *E. coli* DH1 was transformed with the plasmid pBbA5c-MTSA-T1f-MBI(f)-T1002-Ptrc-trGPPS(co)-LS (encoding enzymes responsible for the conversion of acetyl-CoA derived from glucose into limonene) and optionally, a second low-copy plasmid containing either the wild type *ahpC* gene or $ahpC^{L177Q}$.

E. coli strains harboring the limonene production plasmid were grown in tubes containing 8 mL of EZ-Rich defined medium (Teknova), supplemented with 10g/L glucose and appropriate antibiotics (chloramphenicol and kanamycin at 30 and 12.5 μg/mL, respectively). Production cultures were induced at an OD_{600} of 0.8-1.2 with a final concentration of 25μM isopropyl β-d-1-thiogalactopyranoside (IPTG) and 50nM anhydrous tetracycline (aTc), and allowed to continue growth at 30C. A dodecane layer (10% v/v) was added to the culture upon induction to trap limonene, and 10 μL of the organic phase was sampled and analyzed using gas-chromatography mass-spectrometry (GC/MS) at 24, 48, and 72 hours after induction.

References

1. **Alonso-Gutierrez J, Chan R, Batth TS, Adams PD, Keasling JD, Petzold CJ, Lee TS**. 2013. Metabolic engineering of *Escherichia coli* for limonene and perillyl alcohol production. Metabolic Engineering **19**:33–41.
2. **Lee TS, Krupa RA, Zhang F, Hajimorad M, Holtz WJ, Prasad N, Lee SK, Keasling JD**. 2011. BglBrick vectors and datasheets: A synthetic biology platform for gene expression. Journal of Biological Engineering **5**:12.
3. **Baba T, Ara T, Hasegawa M, Takai Y, Okumura Y, Baba M, Datsenko KA, Tomita M, Wanner BL, Mori H**. 2006. Construction of *Escherichia coli* K-12 in-frame, single-gene knockout mutants: the Keio collection. Mol Syst Biol **2**:2006.0008.