

S3 Table. Genotypic and phenotypic characteristics of sequenced auxotrophic and prototrophic genotypes. The genomic features are also illustrated in Fig. 5. *Strain identity* code: X-Y-Z where X refers to a particular population (1-8), Y refers to the regime, i.e. AA = amino acid, or NA = no amino acid, and Z refers to the phenotype, i.e. either AT = auxotrophic or PT = prototrophic.

Strain identity	Genotype	Amino acid auxotrophies
2-AA-AT	<i>ptsP</i> (M604R), <i>yoaA</i> (D121Y), <i>insF1-mdtB</i> (Δ 13,852 bp)	Arg, Asn, Asp, Cys, Gly, His, Phe, Ser, Thr, Val
3-AA-AT	<i>hemF</i> (G127V), <i>dhaM</i> (V196A), <i>rph</i> (-82 bp)	Ala, Arg, Asn, Asp, Cys, Gln, Glu, Gly, His, Ile, Leu, Lys, Met, Phe, Pro, Ser, Thr, Trp, Tyr, Val
4-AA-AT	<i>rph</i> (-82 bp), <i>yfjM</i> / <i>yfjL</i> [(CATAGTGC) $6 \rightarrow 7$], <i>cra</i> / <i>ilvH</i> (C \rightarrow T)	Arg, Asn, Asp, Cys, Gln, Gly, His, Phe, Ser & Thr
5-AA-AT	<i>rph</i> / <i>pyr</i> (-1 bp), <i>sspA</i> (Q24*), <i>gltD</i> N354D, <i>crr</i> / <i>ptsI</i> (A \rightarrow C)	Arg, Asn, Asp, Cys, Gln, Gly, His, Phe, Ser, Thr, Val
6-AA-AT	<i>gltD</i> (N354D), <i>stpA</i> (R49S), <i>rcnR</i> / <i>thiM</i> (A \rightarrow G), <i>wcaN</i> (-1 bp), <i>uspC</i> / <i>flhD</i> (+4 bp), <i>ykcC-proB</i> (Δ 13,518 bp);	Pro
8-AA-AT	<i>yhdW</i> (C \rightarrow A), <i>gltD</i> (N354D), <i>rcnR</i> / <i>thiM</i> (A \rightarrow G), <i>wzxC</i> / <i>wcaK</i> (+85 bp),	Gly
7-NA-AT	<i>yqiB</i> (Q117K), <i>rpoB</i> (T135P)	Lys, Trp
8-NA-AT	<i>yqiB</i> (Q117K), <i>rpoB</i> (T135P), <i>metG</i> (R600P), <i>dxr/frr</i> (T \rightarrow A), <i>ynaE/pinR</i> (A \rightarrow G)	Lys, Trp
3-AA-PT	<i>rph</i> / <i>pyr</i> (-1 bp), <i>cra</i> (L191F)	NA
4-AA-PT	<i>rph</i> / <i>pyr</i> (-1 bp), <i>cra</i> (L191F)	NA
5-AA-PT	<i>rpsK</i> (I116N), <i>gltD</i> (N354D), <i>crr/ptsI</i> (A \rightarrow C), <i>rcnR/thiM</i> (A \rightarrow G)	NA
6-AA-PT	<i>rph</i> (-82 bp), <i>fecR</i> (A12G), <i>hflC</i> (G268D), <i>gltD</i> (N354D), <i>rcnR/thiM</i> (A \rightarrow G)	NA
7-NA-PT	<i>wzxC/wcaK</i> (+85 bp)	NA
8-NA-PT	<i>rph</i> / <i>pyr</i> (-1 bp), <i>wzxC/wcaK</i> (+85 bp), <i>mppA/pgrR</i> (IS5 +4 bp)	NA