

Integration of multi-omics data reveals evolutionary traces of *Escherichia coli*

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We have got 7 independent glycerol evolved strains from wild type *E. coli* K-12 MG1655. The results from parallel, replicate adaptive evolution experiments of *E. coli* grown on glycerol minimal media showed that growth phenotypes at the endpoint of evolution are convergent and reproducible; however, endpoints of evolution have different underlying gene expression states. We have fully resequenced 5 independent glycerol evolved strains. Surprisingly, one of them had only two mutations of whole genome in the endpoint. We added these two mutations into the wild type strain by site-directed mutagenesis. The double knock-in mutant had similar specific growth rate of evolved strain. The two mutations thus seem to fully be responsible for the growth rate change during adaptive evolution. Moreover, intracellular metabolome analysis showed that guanosine triphosphate level of evolved strain and double mutant was almost half of wild type strain at early and mid exponential phase.