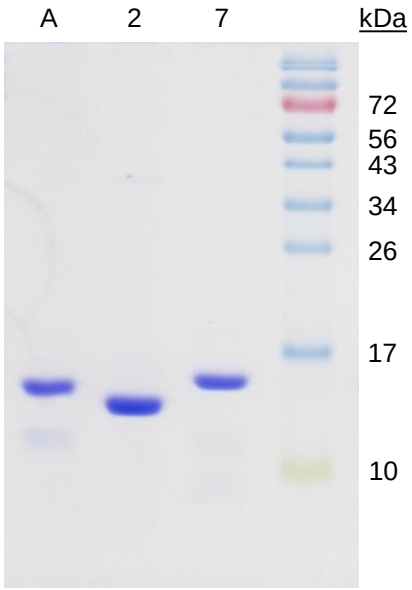
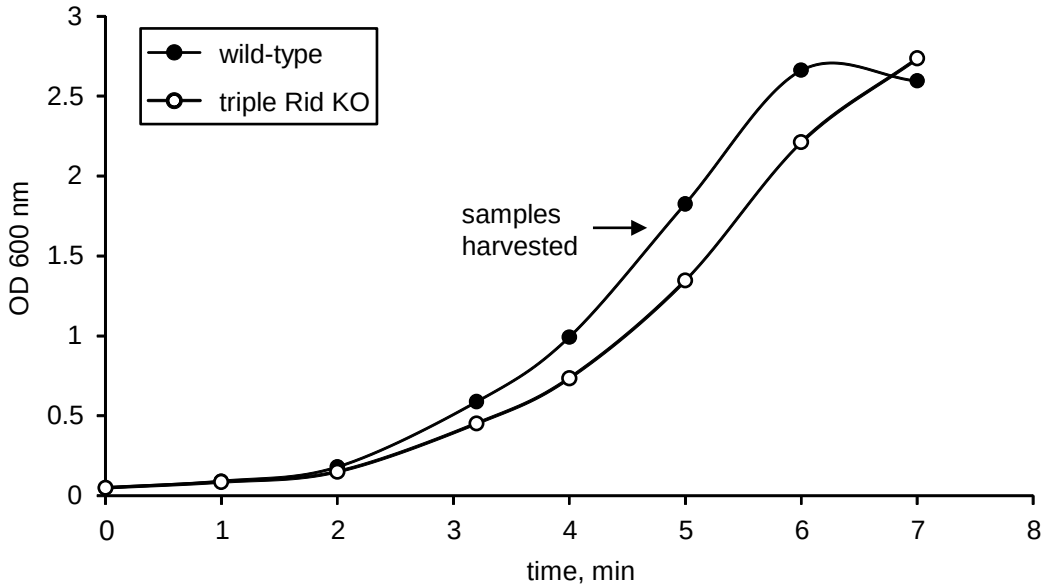


Supplemental Figure 1. Recombinant proteins used in this study were purified to near homogeneity. 5 µg of Ni-affinity purified *Salmonella enterica* RidA (A), Rid2 (2), and Rid7 (7) were analyzed by SDS-PAGE (15% gel) with Coomassie staining. Each protein preparation was judged to be ≥90% pure.

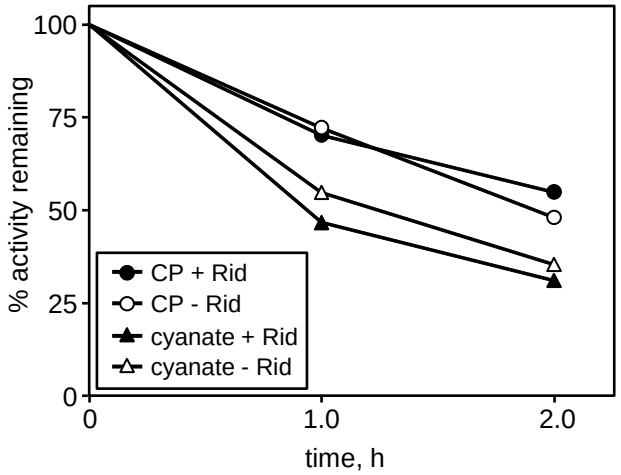


Supplemental Figure 2. Growth of wild-type *S. enterica* and triple Rid KO (*ridA Rid2 Rid7*; DM14100) cells in M9 medium (0.2% glucose). Fresh medium was innoculated with overnight cultures to an OD (600 nm) of 0.05 and cultures were grown at 37°C. Data shown are representative of at least three independent experiments done on separate days. For metabolomics, samples were harvested at OD 600 nm = 1.7±0.1.

A

Rate of cyanate breakdown in presence of Rid proteins	
sample	µmol NH_3 formed
control	<1.0
RidA	<1.0
Rid2	<1.0
Rid7	<1.0

B



Supplemental Figure 3. *Salmonella enterica* Rid proteins do not increase the rate of breakdown of cyanate or carbamoyl phosphate. **(A)** Assays (50 µL) contained 50 mM KPi, pH 8.0, 100 µM sodium cyanate, with or without 100 µM *S. enterica* RidA, Rid2, or Rid7 and were incubated for 30 min at 22°C. Afterwards, 50 µL of GDH master mix containing 50 mM Kpi, pH 8.0, 7 mM 2-oxoglutarate, 200 µM NADPH, and 6 U L-glutamic dehydrogenase from bovine liver (Sigma-Aldrich) was added and the optical density at 340 nm was monitored for 5 min at 22°C. The amount of NADPH oxidized was calculated (ε340 nm = 6200•M^{-1}•cm^{-1}). **(B)** Preincubation reactions (25 µL) contained 50 mM KPi, pH 7.5, 1 mM isoleucine, 10 µM *Arabidopsis thaliana* threonine dehydratase [9], with (closed symbols) or without (open symbols) 100 µM each of *S. enterica* RidA, Rid2, and Rid7, and 25 mM of either carbamoyl phosphate (CP, circles) or cyanate (triangles) and were incubated at 37°C. At the indicted times, 5 µL of preincubation assay was added to 95 µL of assay mixture containing 50 mM KPi, pH 8.0, 5 mM threonine, and the absorbance at 248 nm was recorded at 22°C. Data is reported as the percent of the rate of 2-oxobutanoate formed at t=0.

Table S1

Relative rate of semicarbazone formation for various amino acid substrates incubated with *Crotalus adamanteus* L-amino acid oxidase.

Amino acid	**Relative rate** [a]
Leu	100
Met	79.1
Phe	46.1
Lys	6.5
Ser	0.4
Glu	0.3
Gln	21.3

[a]rate is expressed as a percent of the rate of semicarbazone formation with Leu as the substrate