Electronic Supplementary Material (ESI) for Analyst.

Supplemental Information

A Fluorometric Assay for High-throughput Phosphite Quantitation in Biological and Environmental Matrices

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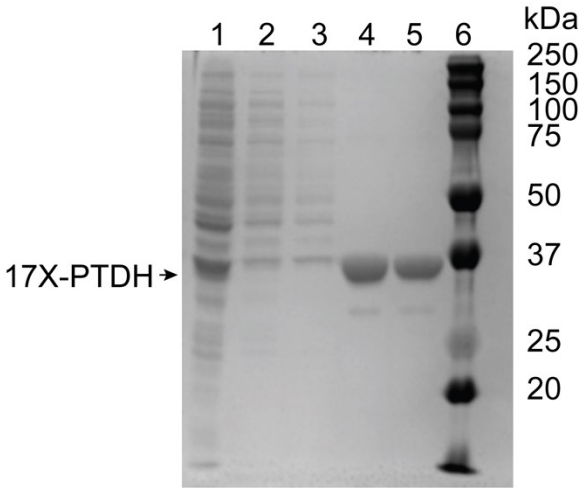

Figure S1. SDS-PAGE gel describing the purification of 17X-PTDH. Lanes contain the *E. coli* BL21(DE3) lysate with overexpressed 17X-PTDH (1), Ni-NTA column flow-through (2, 3), eluted final protein fraction (4,5), and molecular weight ladder (6). The 17X-PTDH is 36 kDa.

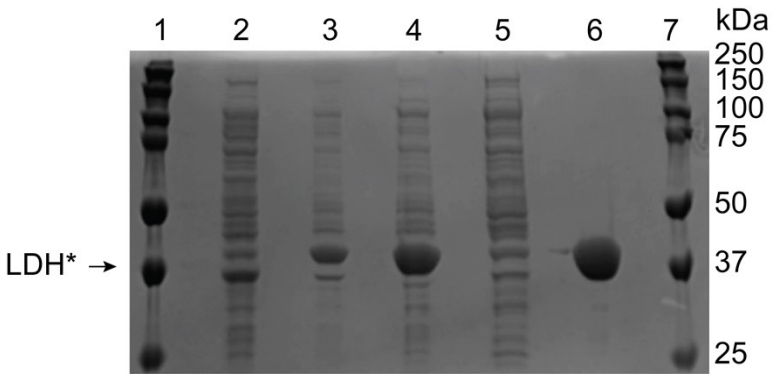

Figure S2. SDS-PAGE gel describing the purification of LDH*. Lanes contain the molecular weight ladder (1), pre-induction *E. coli* BL21(DE3) lysate (2), post-induction *E. coli* BL21(DE3) lysate with overexpressed LDH* (3), cell lysate (4), Ni-NTA column flow-through (5), final elution protein fraction (6), and the molecular weight ladder (7). The LDH* is 37 kDa.

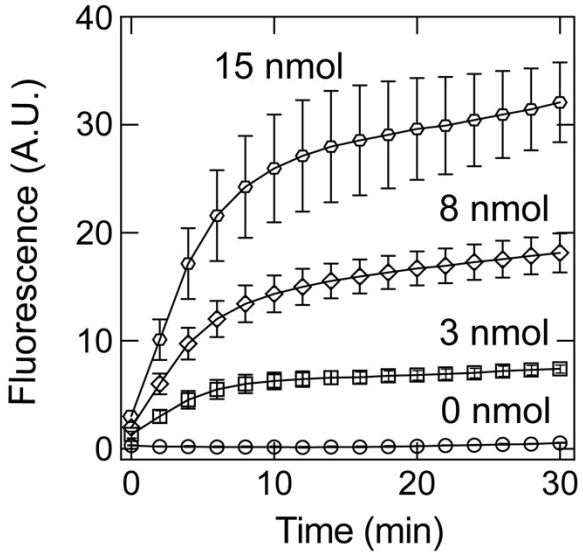

Figure S3. Phosphite assay kinetic response for 200 nM 17X-PTDH and 100 mM PMS. Assay reactions containing 0 (circles), 3 (squares), 8 (diamonds), and 15 (hexagons) nmol Phi are shown.

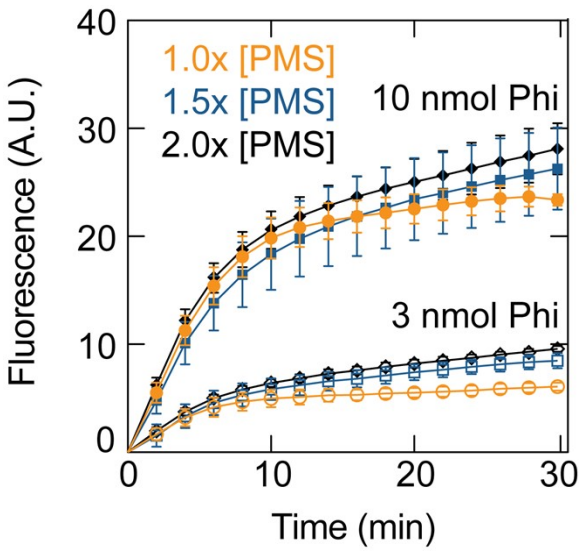

Figure S4. Phosphite assay kinetics profile for 2 μg 17X-PTDH and 100 (orange circles), 150 (blue squares), or 200 mM (black diamonds) PMS. Assay reactions containing 3 nmol (open shapes) and 10 nmol (filled shapes) Phi are shown.

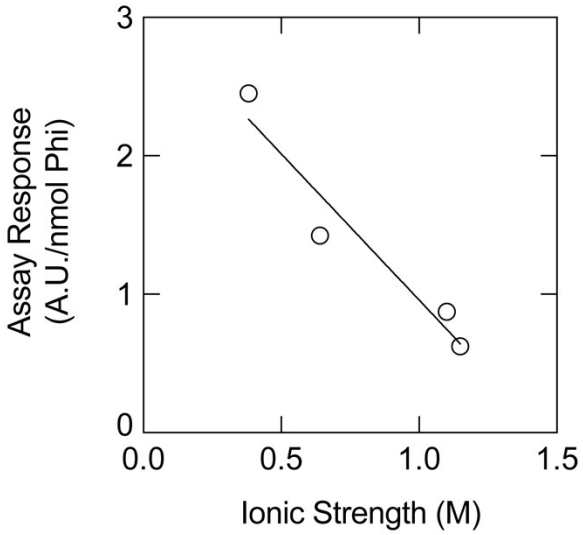

Figure S5. Dependence of ionic strength of sample on Phi assay response. Assay response is defined as the slope of the standard addition curve in fluorescence (A.U.) versus nmol Phi added.

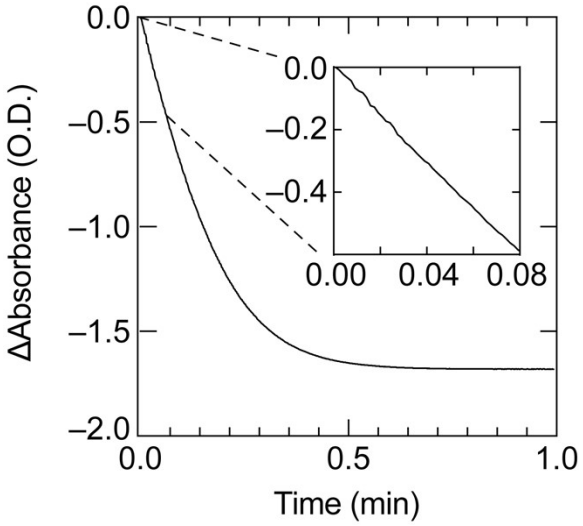

Figure S6. Pyruvate-dependent NADH oxidation activity of purified LDH* in STET buffer as determined by the change in absorbance at 340 nm. The k_{cat} was determined from the pseudo-first order regime within the first 5 seconds to be 1,330 ± 30 s^{-1} at room temperature.

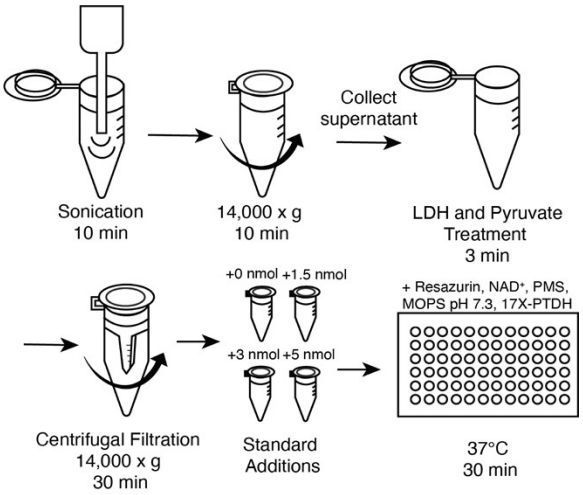

Figure S7. Schematic representation of sample preparation for the Phi assay.

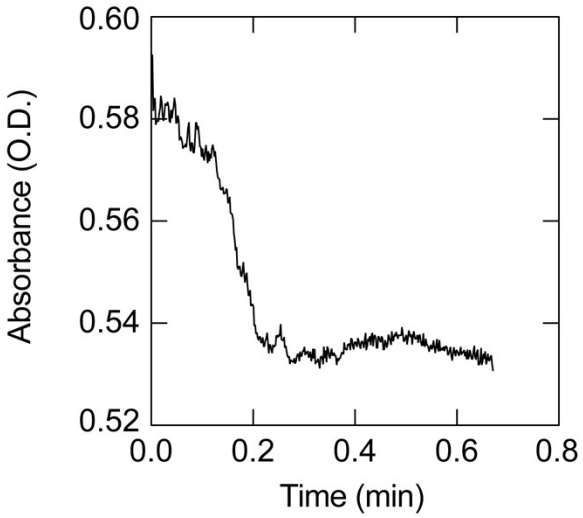

Figure S8. The effect of LDH* and pyruvate treatment on *E. coli* cell lysate based on the absorbance of NAD(P)H at 340 nm.

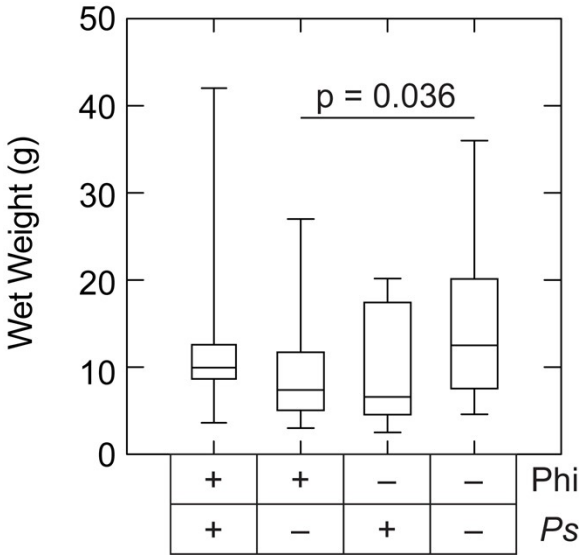

Figure S9. Box-and-whisker plot of radish wet weight following growth in soil containing Phi alone (n = 27), *P. stutzeri* (*Ps,* n = 7), both (n = 34), or neither (n = 23). Samples containing just Ps or both *Ps* and Phi exhibited $p > 0.05$ with respect to the null.