

Supporting Information

Bicarbonate is essential for protein tyrosine phosphatase 1B (PTP1B) oxidation and cellular signaling through EGF-triggered phosphorylation cascades

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- Running title: PTP1B oxidation and signaling by EGF requires bicarbonate

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Materials included:

Figure S1

Figure S2

Figure S3

Figure S4

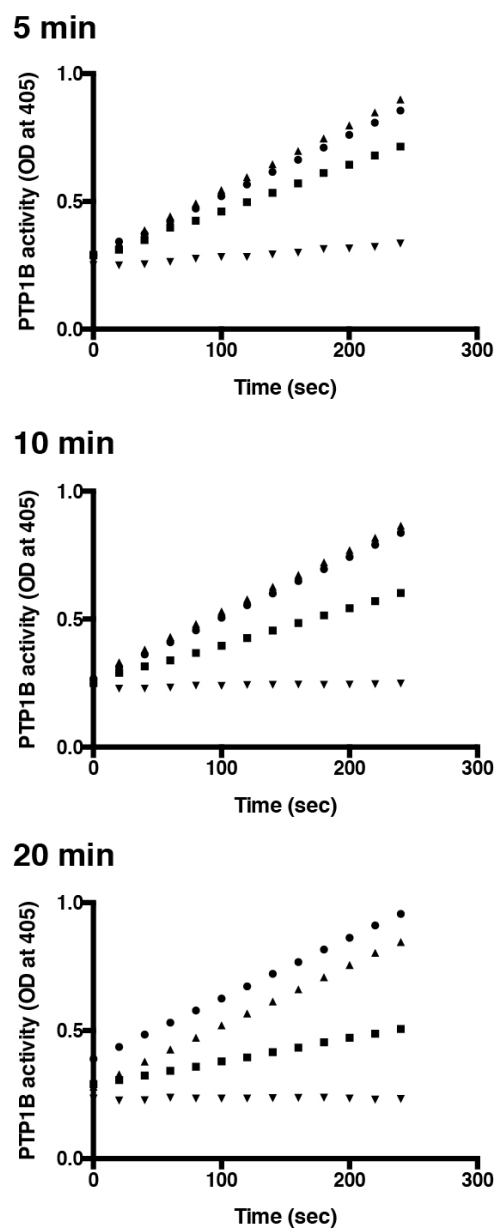


Figure S1. Bicarbonate potentiates H_2O_2 -dependent inactivation of PTP1B. PTP1B was treated with buffer control (●), 25 mM bicarbonate (▲), or 50 μ M H_2O_2 (■) or 50 μ M H_2O_2 and 25 mM bicarbonate (▼) as described in figure 1A and B. The activity was measured at indicated time points by following the rate of absorbance increase at 410 nm.

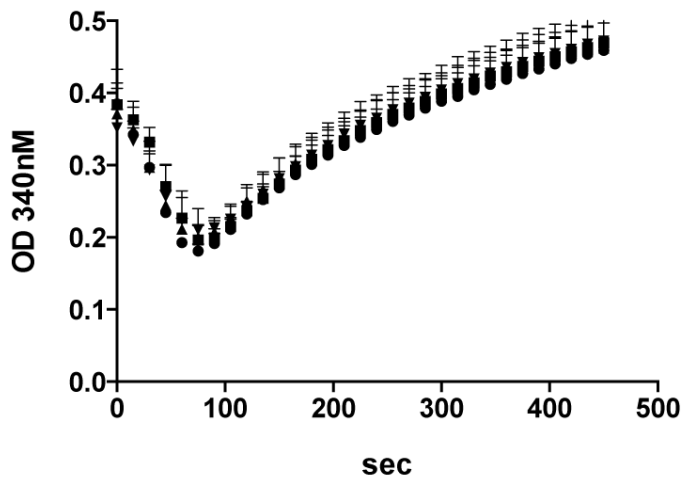
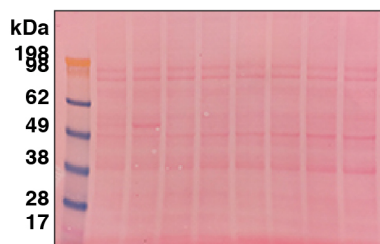


Figure S2: The Trx system is not affected by treatment with bicarbonate alone or in combination with H₂O₂. The effects of 25 mM bicarbonate, alone (▲), or in combination with 100 μM H₂O₂ (▼) or 100 μM H₂O₂ alone (■), compared to buffer control (●), on the Trx system activity as determined following the NADPH consumption at 340 nm using 0.16 mM insulin as a substrate. The initial decrease of absorbance reflects NADPH (300 μM) consumption during insulin (0.16 mM) disulfide reduction by Trx1 (20 μM) and its recycling by TrxR1 (1 μM), while the subsequent increase in absorbance is due to turbidity increase as reduced insulin precipitates.

A) Ponceau staining as loading control for Fig 3A



B) Ponceau staining as loading control for Fig 3B

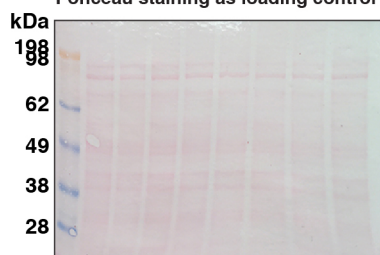
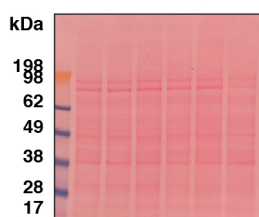


Figure S3. Loading control for figure 3A and 3C. The membranes were stained with Ponceau for total protein loading.

A) Ponceau staining as loading control for Fig 4A



B) Ponceau staining as loading control for Fig 4B

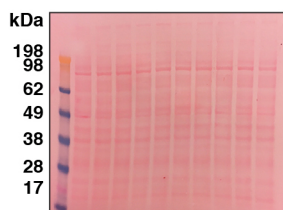


Figure S4. Loading control for figure 4A and 4B. The membranes were stained with Ponceau for total protein loading.