

Determination of the K_D for GS-Me and TCHQ dehalogenase in the presence of saturating TriCHQ. Aliquots of a solution containing GS-Me (50 mM), TCHQ dehalogenase (100 μ M), TriCHQ (170 μ M) and ascorbate (0.1%) in 200 mM potassium phosphate, pH 8.0, were added to a cuvette containing the same solution lacking only GS-Me. Absorbance spectra were recorded and the change in absorbance at 360 nm was plotted versus concentration of GS-Me (Supplementary Figure 1). The data were fit to a quadratic binding equation (Equation 1) and the dissociation constant of GS-Me was determined to be 55 μ M \pm 10 μ M.

$$A = A_0 + \Delta A \left(\frac{(K_D + E_0 + S_0) - \sqrt{(K_D + E_0 + S_0)^2 - 4E_0S_0}}{2E_0} \right) \quad \text{Eq. 1}$$

Figure 1.

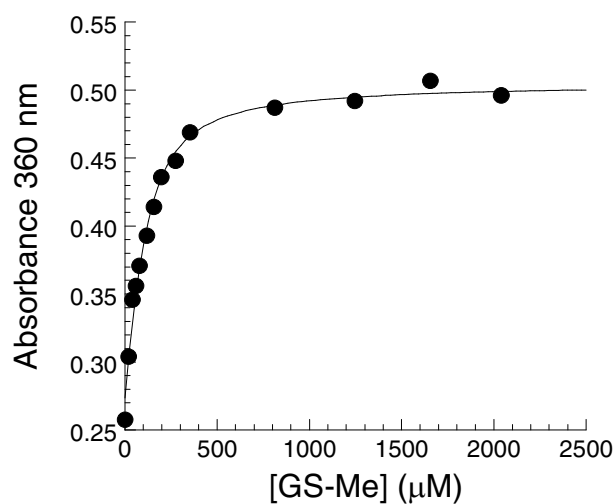


Figure 1. Absorbance at 360 nm of TCHQ dehalogenase (100 μM) in 200 mM potassium phosphate, pH 8.0, containing 0.1% ascorbate, 170 μM TriCHQ, and variable concentrations of GS-Me (0 to 800 μM). The data were fit with equation 1 to give a K_D , $_{\text{GS-Me}} = 55 \mu\text{M} \pm 10 \mu\text{M}$.