

Supplemental Figure 5

S5 Fig. Reactive oxygen and pH stresses do not regulate the *CTR3* promoter. *CTR3* promoter activity in 1 μ M CuSO₄ (low *CTR3* promoter activity, black bars) or 10 nM CuSO₄ (high *CTR3* promoter activity, red bars) at a range of H₂O₂ concentrations (**A**) or pH (**B**). (**A**) *H. capsulatum* yeasts were incubated in 3M medium (containing 1 μ M CuSO₄ or 10 nM CuSO₄) with H₂O₂ (0 μ M to 250 μ M). (**B**) *H. capsulatum* yeasts were incubated in 3M medium (containing 1 μ M CuSO₄ or 10 nM CuSO₄) with H₂O₂ (0 μ M to 250 μ M). (**B**) *H. capsulatum* yeasts were incubated in 3M medium (containing 1 μ M CuSO₄ or 10 nM CuSO₄) buffered to different pH with MES (4.5 to 6.0) or HEPES (6.5 to 7.0). The *CTR3* promoter activity was assessed by fluorescence of wild-type yeasts with the *CTR3* promoter-*gfp* fusion (P*cTR3*) after normalization to yeasts with the *TEF1* promoter-gfp fusion (P*tEF1*) grown in identical conditions. After 72 hours incubation at 37°C, culture turbidity (optical density at 595nm) and GFP fluorescence (485 nm excitation, 528 emission) were measured. *TEF1* or *CTR3* promoter activity (GFP fluorescence) was normalized to the yeast density (OD₅₉₅) and the *CTR3* promoter activity then compared to that of the constitutively expressed *TEF1* promoter. Data represent the average relative *CTR3* promoter activity ± standard deviation among biological replicates (n = 3).