DXP Assay

2011年12月12日 15:28

Ziru Li

Measure concentration of DXP with a substrate-limiting reaction

Last modified 2009.12.02

Dilute 1mg of DXP in 500uL DI H2O and make 50uL aliquots. Mix 5uL of DXP stock solution with 95uL of DI H2O to make 1:19 DXP solution

40uL 1.0M triethonalamine-HCl buffer (pH7.5)
3uL 1.0M DTT
4uL 1.0M MgCl2
5uL DXP solution
5uL 10mM NADPH
>2ug DXR
DIH2O to 1000uL

Mix all components except DXR and incubate at room temperature for 5 minutes. Set the spectrophotometer to amplification = 20, chart recorder speed = 20cm/hr, filter = 0.5s, signals A and B = 0.940 (between 0.8 and 1.0). Add DXR to initiate the reaction, after obtaining a steady baseline.