PGI SOP

Specific activity of the glycolytic enzymes were measured in NAD(P)H/NAD(P)+ linked enzyme assays that were adapted from Teusink *et al*. [1] and measured at 340 nm in 96-well plates (Flat Bottom microplate, Greiner Bio-One, Kremsmünster, Austria) on a spectrophotometer (VarioSkan microplate reader, Thermo Electron Corporation, Waltham, Massachusetts, USA). The same buffer, (20 mM HEPES, 20 mM MgCl, 10 mM KCl and 20 mM NaCl), was used for all assays, with a pH set to 7.17, matching the cytosolic pH of *P. falciparum* D10 [2]. All of the linking enzymes were used at a non-limiting, final concentration of 5 U/mL. All reagents and enzymes were obtained from Sigma-Aldrich, St. Louis, Missouri, USA.

Phosphoglucoisomerase (PGI) activity was measured via the conversion of G6P (0 - 35 mM) to F6P, which was linked to the oxidation of NADH (0.8 mM) via αGlyPDH, ALD, TPI, PFK in the presence of ATP (2 mM). The reverse isomerisation of F6P (0 - 5 mM) to G6P was linked to reduction of NADP+ (0.8 mM) via G6PDH.

[1]  Teusink B, Passarge J, Reijenga C, Esgalhado E, van der Weijden C, et al. (2000) Can yeast glycolysis be understood in terms of *in vitro* kinetics of the constituent enzymes? testing biochemistry. Eur J Biochem 267: 5313-5329.

[2]  Wünsch S, Sanchez C, Gekle M, Grosse-Wortmann L, Wiesner J, et al. (1998) Differential stimulation of the Na+/H+ exchanger determines chloroquine uptake in *Plasmodium falciparum*. J Cell Biol 140: 335-345.