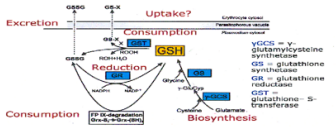
A Systems Biology approach to model the Glutathione pathway in *Plasmodium falciparum*.

[1]Brian B. Tarimo, [2]Sylke Müller, [2]Xu Gu, and [3]David R. Gilbert

[1]Ifakara Health Institute (Tanzania), [2]University of Glasgow (UK), and [3]Brunel University (UK)

Background

- Malaria morbidity and mortality is a severe burden, no where is this more apparent than in sub-Sahara Africa (SSA).
- There is great efforts to control Malaria in SSA but this faces great challenges notably to drug resistance, especially to classical and affordable antimalarials, such as Chloroquine (CQ).
- Glutathione (GSH) has its metabolism greatly associated with the mode of action of CQ.
- This mathematical model is a series of biochemical reactions of this pathway in terms of its biosynthesis, reduction, consumption, and efflux over a specified period of time.

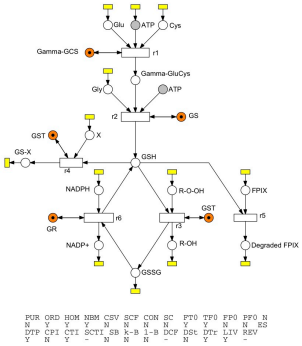


Objectives

- Broad:
  - ➢ To construct a mathematical model that elucidates the metabolism of the GSH pathway.
- Specific:
  - ➢ To collect kinetic data of metabolites involved in GSH metabolism from published literature.
  - ➢ To construct and validate of a Petri net (PN) model of the GSH pathway using the software Snoopy and Charlie respectively.
  - ➢ To construct an ordinary differential equation (ODE) based model using MATLAB and its annotation using the software CellDesigner.
  - ➢ To subject the ODE based model to analysis by performing inhibition studies.

Methods

PN model construction & Validation



PUR ORD HOM NBM CSV SCF CON SC FT0 TP0 FP0 FP0_N
N Y Y Y N N N N Y Y N N_ES
DTP CPI CTI SCTI SB k-B 1-B DCF DSt DTr LIV REV
Y N Y - N N N - N Y Y -

ODE based model with MATLAB

- Biosynthesis:

$$V_{\gamma\text{-}GCS} = \frac{V_{max}[Glu][Cys][ATP]}{(K_m + [Glu])(K_m + [Cys])(Km + [ATP])}$$

$$V_{GS} = \frac{V_{max}[\gamma\text{-}GluCys][Cly][ATP]}{(K_m + [GluCys])(K_m + [Gly])(Km + [ATP])}$$

- Reduction:

$$V_{GR} = \frac{V_{max}[GSSG][NADPH]}{(K_m + [NADPH^+) + (Km + [GSSG])}$$

$$V_{NADP+} = K[NADP^+]$$

- Consumption:

$$V_{GST} = \frac{V_{max}[X][GSH]}{(K_m + [X])(K_m + [GSH])}$$

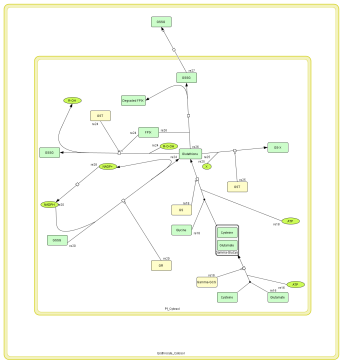
$$V_{GST} = \frac{V_{max}[GSH][R\text{-}O\text{-}OH]}{(K_m + [GSH])(K_m + [R\text{-}O\text{-}OH])}$$

$$V_{FPIX} = k[FPIX][GSH]$$

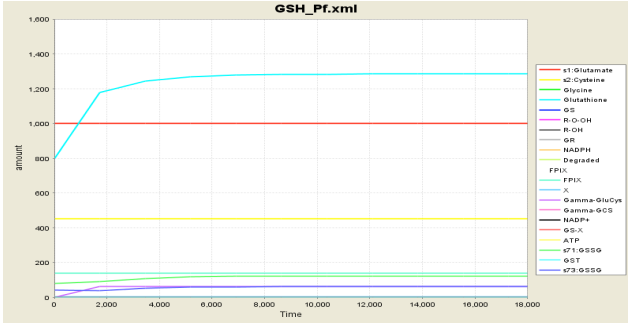
- Excretion/Efflux:

$$V_{GSSG\ Efflux} = \frac{V_f[GSSG_{Pf}]/K_{mGSSGPf} \times (1 - (GSSG_{RBC}/GSSG_{Pf})/K_{eq}}{1 + GSSG_{RBC}/K_{mGSSGRBC} + GSSG_{Pf}/K_{mGSSGPf}}$$

Annotation of the ODE based model using CellDesigner



Results



- Inhibition studies (50 µM of Methylene Blue (MB)):
  - ➢ Concentration of GSH increased by 4-folds (1290 to 350µM).
  - ➢ Concentration of oxidized Glutathione (GSSG) in Pf increased by 200 folds (122 to 28,234µM).
  - ➢ Concentration of GSSG (in RBC) increased by over 100 folds (61 to 7671µM).

Discussion & Conclusion

- 50 µM of MB affects the GSH/GSSG ratio to such levels that are deleterious to *Plasmodium falciparum*.
- The reduction reaction of the GSH pathway/metabolism has the potential for the development of a drug target.
- MB has effective antimalarial activity and hence could be developed into an antimalarial drug.
- More research has to be conducted on this, and it should focus on how MB can be carefully administered so that it only affects *Plasmodium falciparum* and not its human host.

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