Computational Discovery of Drug Resistance Mechanism(s) of the Malaria Parasite to Tetracyclines and Chloroquines

BY

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INTRODUCTION

- Research has shown that it is impossible to discover the resistance mechanism of Plasmodium falciparum (P.f) at the genomic or transitional level, but on a proteomic level.
- Over the last 40years, biochemical investigations has discovered a consistent image of cellular metabolism popularly called biological interaction network, and this has allowed us to be able to envisage the static state of the cell.
- Also, the advent of DNA microarrays has allowed us to explore a major subset or all genes of an organism under variety of conditions.

DESCRIPTION OF GENE EXPRESSION DATA USED

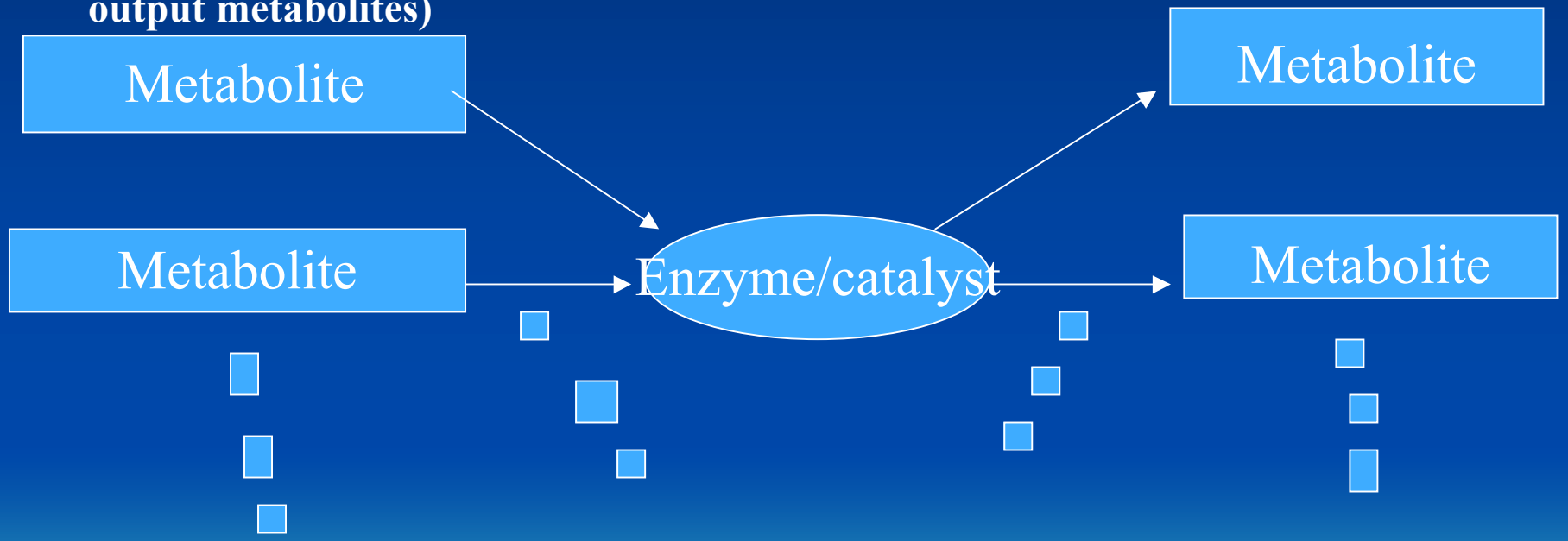
- The chloroquine induced data of P.f is a SAGE (Serial analysis of gene expression) tags from the work of Gunasekera et al. , 2001. We mapped the tags to the genes they represented using BLAST.
- And the microarray time point's data for P.f while treated with tetracycline was made available from the work of Dahl et al., 2006.

MODEL FOR ANALYSING GENE EXPRESSION DATA ON METABOLIC NETWORKS

1. CONSTRUCTION OF METABOLIC NETWORK FROM PLASMOCYC (BIOCYC)
2. NETWORK CLUSTERING USING CONSECUTIVE ONES
3. MAPPING GENE EXPRESSION DATA ONTO REACTIONS AND FEATURES EXTRACTION
4. ANALYSIS OF STIMULATED OR REPRESSED PATHWAYS

1. CONSTRUCTION OF METABOLIC NETWORK FROM PLASMOCYC (BIOCYC)

List of metabolites (nodes or input) and their product of reaction (edge or output metabolites)

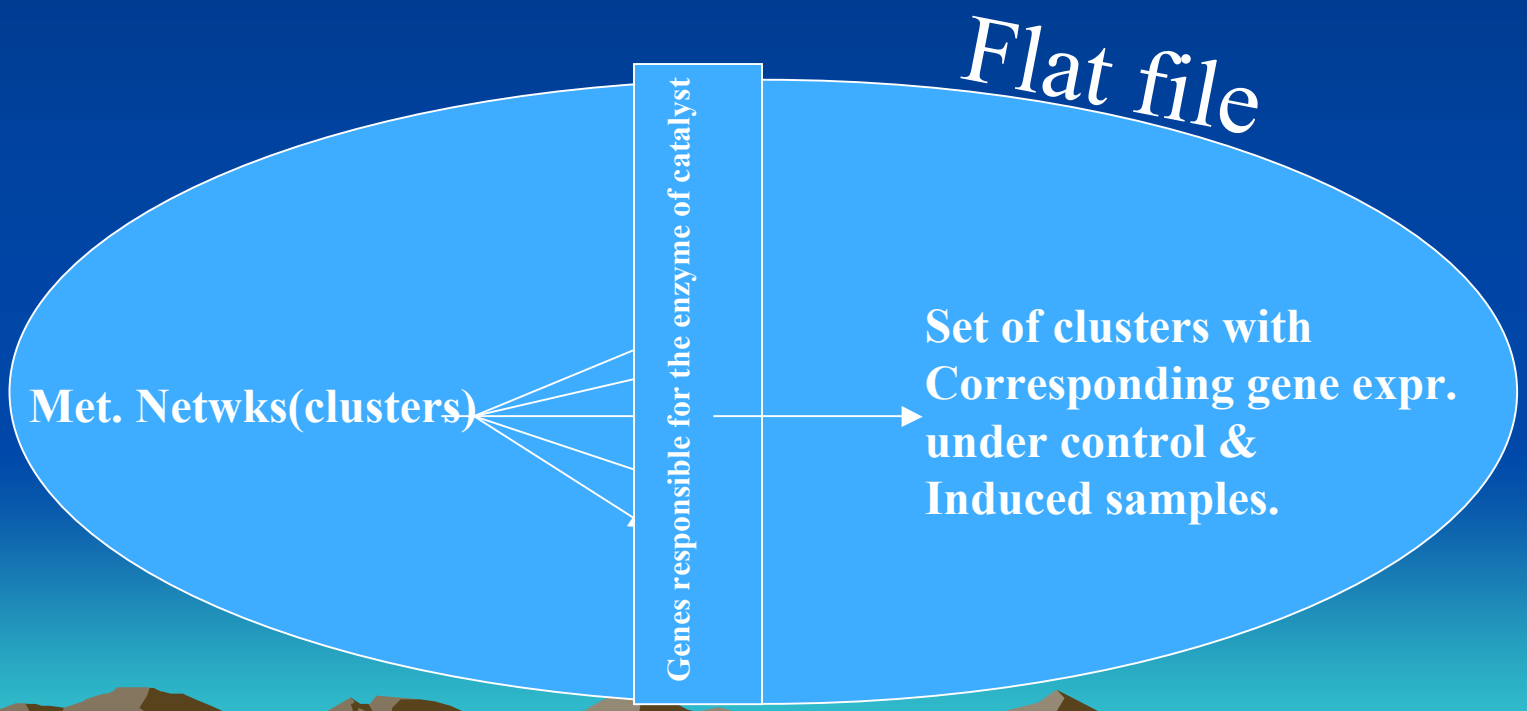


2. NETWORK CLUSTERING USING CONSECUTIVE ONES

The Heuristic based algorithm used here solved the simultaneous consecutive ones prob.

Met. Netwks(clustering) ⟶ Sets of Nodes of highly connected sub-graphs

3. MAPPING GENE EXPRESSION DATA ONTO REACTIONS AND FEATURES EXTRACTION

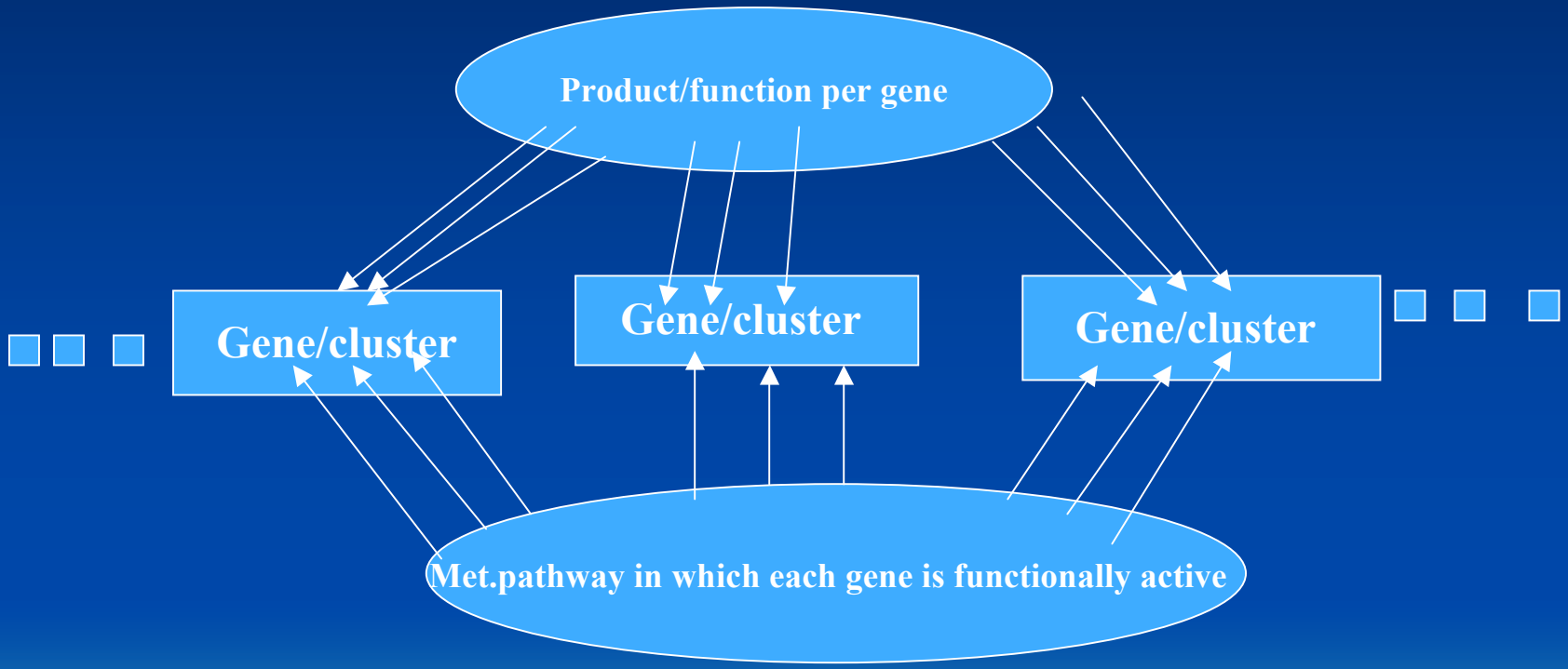


MAPPING GENE EXPRESSION DATA ONTO REACTIONS AND FEATURES EXTRACTION CONTD.

- We calculated a p-value for every possible expression pattern of every groups of genes within a cluster that may show essential differences between samples of different conditions.
- Here, we have microarray data under normal and drug induced conditions at various time points.
- The idea is that if those p-values differ a lot, the group of genes in that clusters should be differentially expressed.
- And there should be a sharp difference as regard the p-values of the stimulated or repressed pathways (clusters) when compare to the control samples.

- Under a condition, the clusters are ranked according to their p-values.
- Note that each cluster is labelled with a serial number that indicates its serial position among the clusters of the output of the clustering algorithm.
- This is also been used to identify the differential expression of genes within clusters under different time points and conditions.

4. ANALYSIS OF STIMULATED OR REPRESSED PATHWAYS



- We expect our patterns extraction tool to capture the distinct differential expression of these genes between the drug induced and the control samples.
- We look out for cases that do not show this format, such cases have been found to give us hints on what collection of genes differentially co-expressed to possibly de-activate the effectiveness of the drug on the targeted pathway.

RESULTS AND DISCUSSION
Dahl et al. results on tetracyclines

- Dahl et al. are the first to analys the tetracycline treated microarray data, and they demonstrated that tetracyclines specifically block expression of the apicoplast genome.
- This results in the distribution of non-functional apicoplasts into the daughter merozoites.
- And the loss of apicoplast function in the progeny of treated parasites leads to a slow but potent antimalarial effect.

RESULTS AND DISCUSSION CONTD.

OUR FINDINGS

- We have an output of 12 clusters but 2 clusters (clusters 21 and 15) exhibit different expression patterns btw control and tetracyclines induced samples.
- Our results depicted in Cluster 21(figs 1,2) confirmed Dahl et al. findings.

RESULT AND DISCUSSION CONTD.

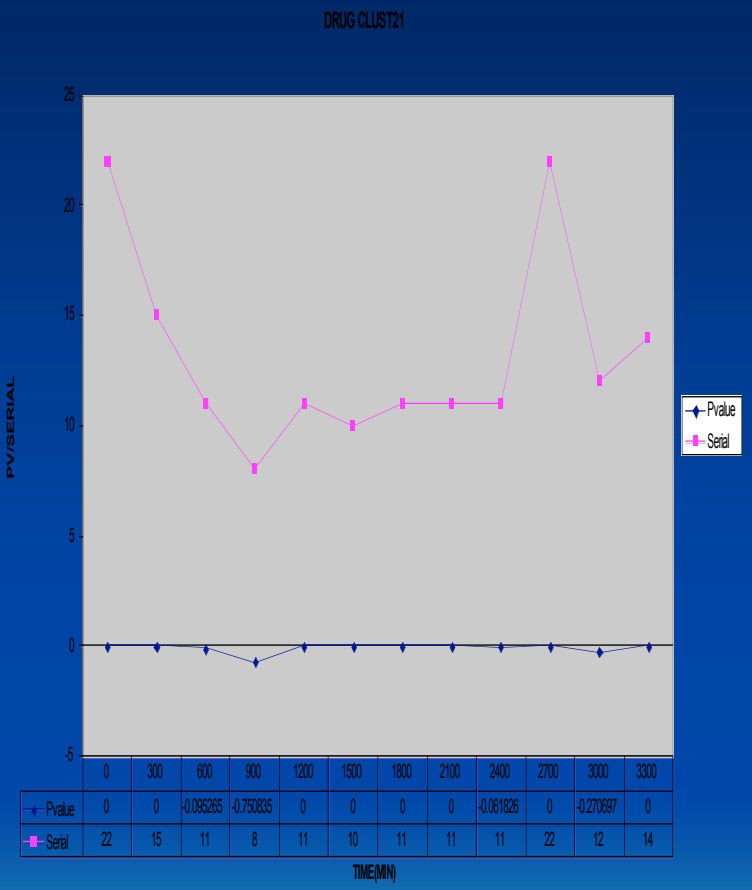


Fig 1. Differential expressions of the genes in cluster 21 under the tetracycline-induced sample

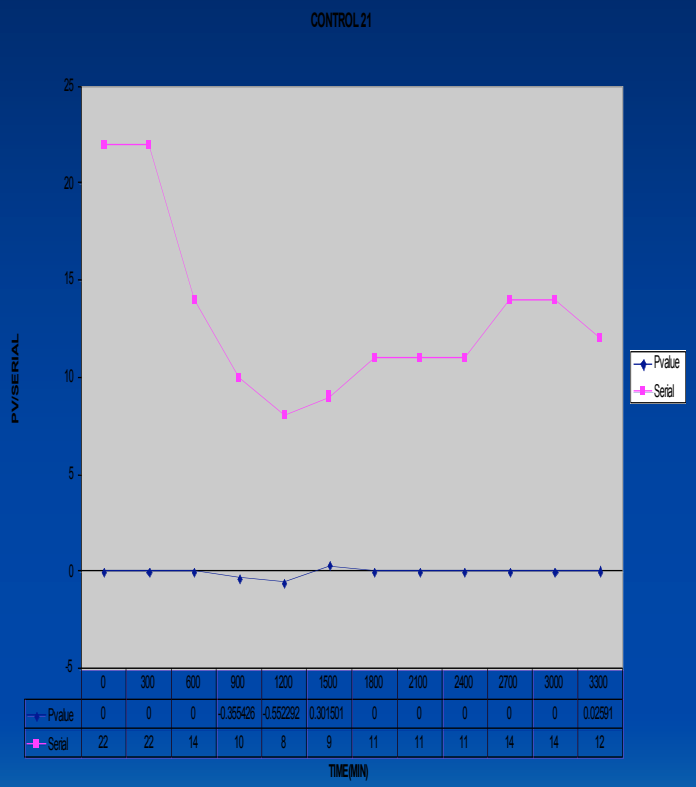


Fig 2. Differential expressions of the genes in cluster 21 under the control sample

RESULT AND DISCUSSION CONTD.

- Also, our result suggests that excess glucose is been generated to resist the effect of tetracycline, because some P.f genes in cluster 15 (figs 3,4). are active in glycolysis pathway and are more excited beyond their expressions under the control sample.
- Thus, the slow effect of tetracyclines is hypothesize in this work to be due to this excess production of glucose.

RESULT AND DISCUSSION CONTD.

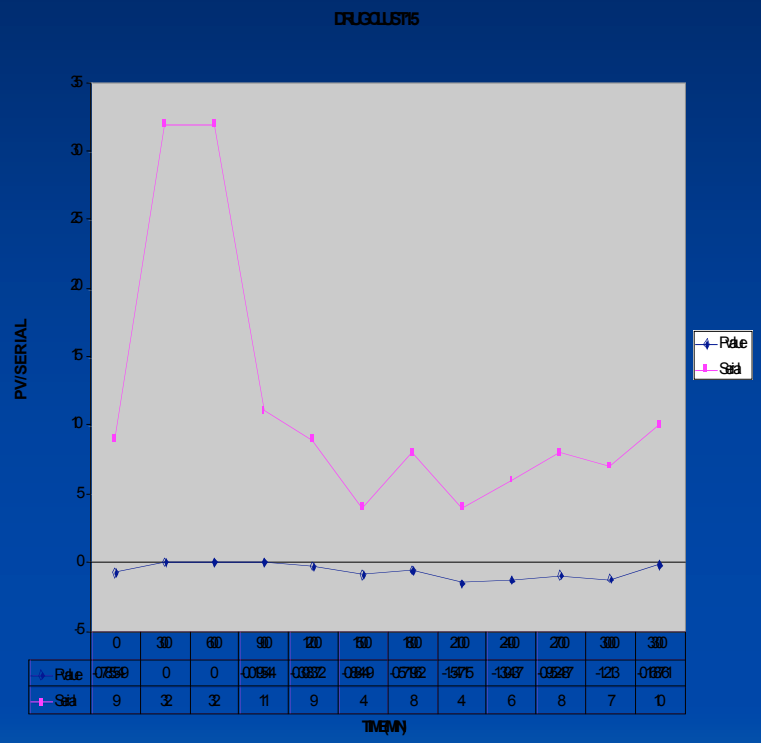


Fig 3. Differential expressions of the genes in cluster 15 under the tetracycline-induced sample

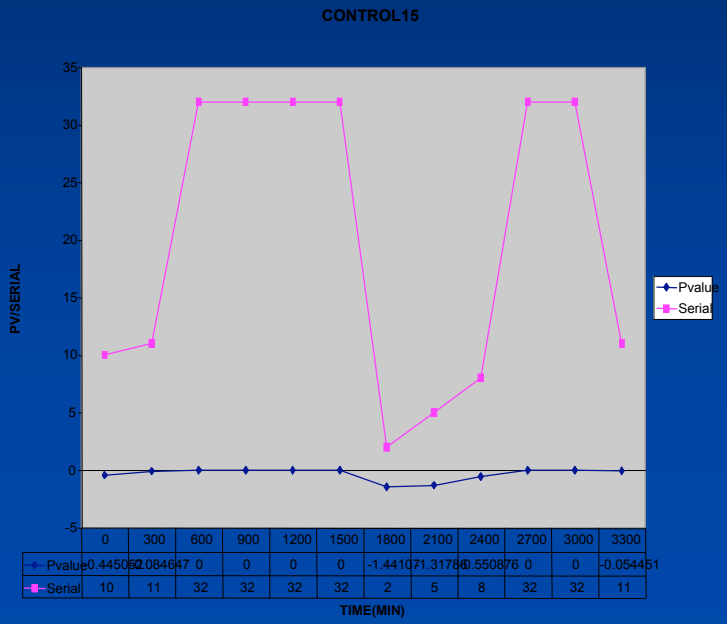


Fig 4. Differential expressions of the genes in cluster 15 under the control sample

RESULTS AND DISCUSSION CONTD.
P.f treated with chloroquine

- We did not find any significant differential expression between any clusters in the Chloroquine induced and control samples.
- Same conclusion was also reached by Le Roch et al.

CONCLUSION

- We have been able to use the biochemical network of P.f to deduce its drugs resistance mechanism(s) using the two microarray data obtained when P.f is treated with tetracycline and chloroquine.
- Our work is the first that developed and applied computational means toward the elucidation of these mechanisms in P.f.
- Our results also suggest to medical practitioner how existing drugs, whose effectiveness has been weakly, can be combined effectively.

NEXT LOOK

- The future work that is been planed as a follow-up to this project is the generation of microarray expression data of Pf under the influence of chloroquine.
- We also observed that quite a number (19 out of 22) of the enzymes encoded by the genes active in the pathway (Ferriprotoporphyrin & Heamoglobin digestion) targeted by chloroquine have not been identified.
- We believe any study that will be able to elucidate the resistant of P.f to this important drug, will require this information for significant discovery to be made in this direction.

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MANY THANKS FOR LISTENING

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