Title: Pharmacogenetic Assessment of Tafenoquine efficacy in Patients with *Plasmodium vivax* Malaria

SUPPLEMENTARY MATERIAL

Study Details

DETECTIVE was a multi-centre, double dummy, double blind, parallel group, randomized, active control study conducted in two parts. Part 1 was a Phase 2b dose-ranging study with ITT patients randomized to 6 treatment arms: arms 1 to 4 contained different doses of tafenoquine (50mg, 100mg, 300mg, and 600mg) dosed on day 1 or 2, arm 5 contained primaquine (15mg) dosed over 14 days (days 2-15) and arm 6 contained chloroquine alone [4]. Based on Part 1 efficacy and safety, the tafenoquine 300mg dose was studied in the pivotal Part 2 Phase 3 study [5]. The aim of DETECTIVE Part 2 was to investigate the safety and efficacy of the selected tafenoquine dose in the treatment and radical cure of *P. vivax* malaria. Part 2 included 522 mITT patients and contained 3 treatment arms randomized 2:1:1 to i) 300mg tafenoquine dosed on day 1 or 2, ii) 15mg primaquine dosed daily on days 2-15, and iii) chloroquine alone, respectively. GATHER was a multi-centre, double dummy, double blind, parallel group, randomized, active control phase 3 study to assess the incidence of hemolysis, safety and efficacy of tafenoquine [6]. 251 mITT patients were randomized 2:1 to i) 300mg tafenoquine on day 1 or 2 or ii) 15mg primaquine on days 2-15.

Outcomes

Three efficacy outcomes were assessed: 1) percentage of patients who were recurrence-free at 6 months, 2) percentage of patients who were recurrence-free at 4 months and 3) time to recurrence up to 6 months post-dosing. Freedom from recurrence was defined as initial clearance of the *P. vivax* infection, no recurrence of *P. vivax* parasitemia, no receipt of other antimalarial treatments, and a negative blood smear at the 4 or 6-month assessment. Patients were excluded from analysis of the

recurrence-free efficacy outcomes if they were censored prior to final assessment due to non-clearance of the original infection, use of a concomitant medication with anti-malarial properties, or no final assessment.

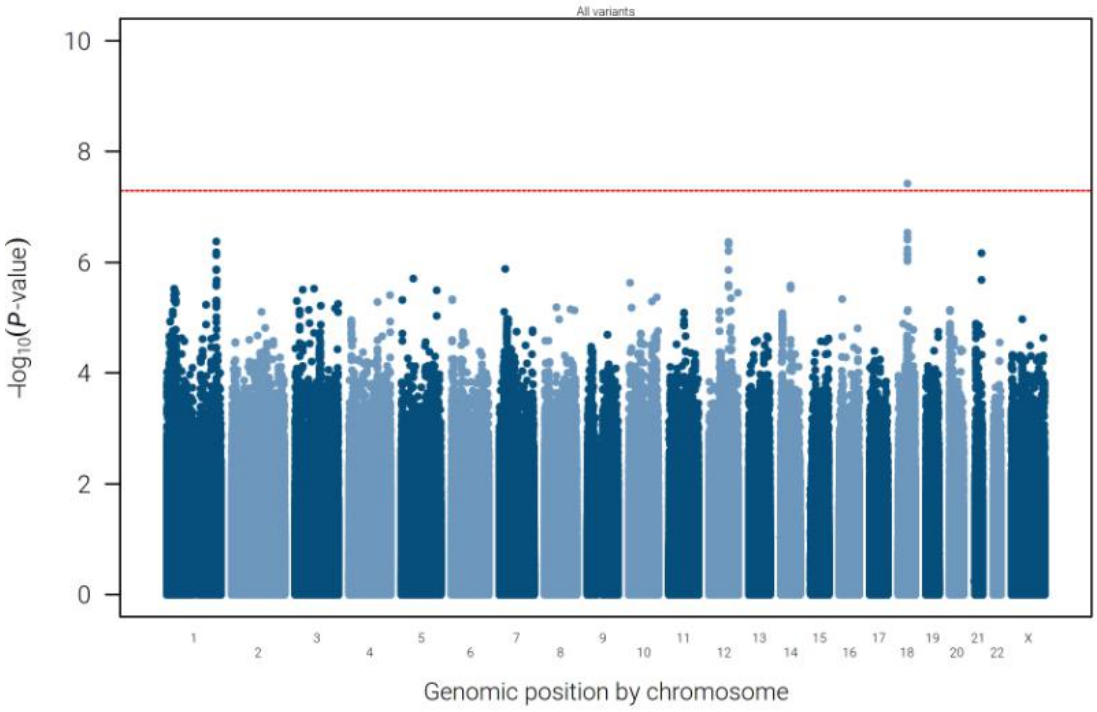
Genotyping, quality controls and imputation

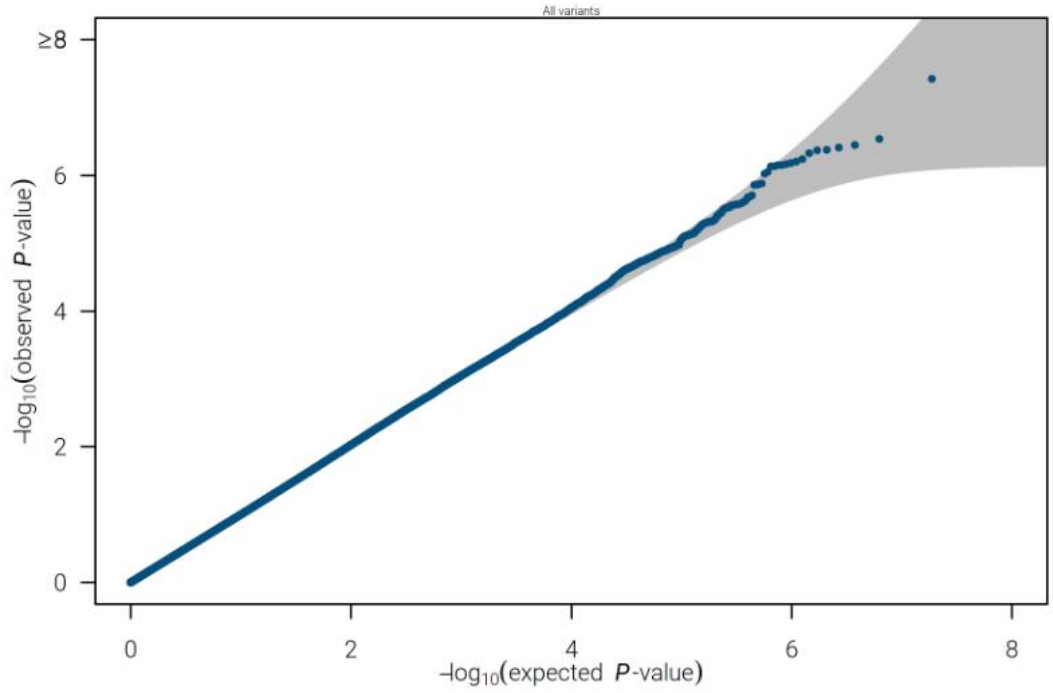
Venous blood was collected from each participant who provided written informed consent. Genomic DNA was extracted using the Gentra Puregene kit on the Autopure LS (Qiagen, Valencia, CA) by Quest Diagnostics (Valencia, CA, USA or Heston, UK). Genotyping was performed using the Affymetrix Axiom array (Santa Clara, CA) with GSK custom content v2 by the Bioprocessing Solutions Alliance under Brooks Automation Inc. (Piscataway, NJ). Standard quality control exclusions were applied. The array data was used to i) generate genetic ancestry principal components (PCs) to correct for confounding due to population stratification [1] and ii) impute variants, with a reference set of 1000 Genomes [2] haplotypes, across the genome including the HLA region using MaCH, minimac and HiBAG [3-5].

REFERENCES FOR SUPPLEMENTAL MATERIALS

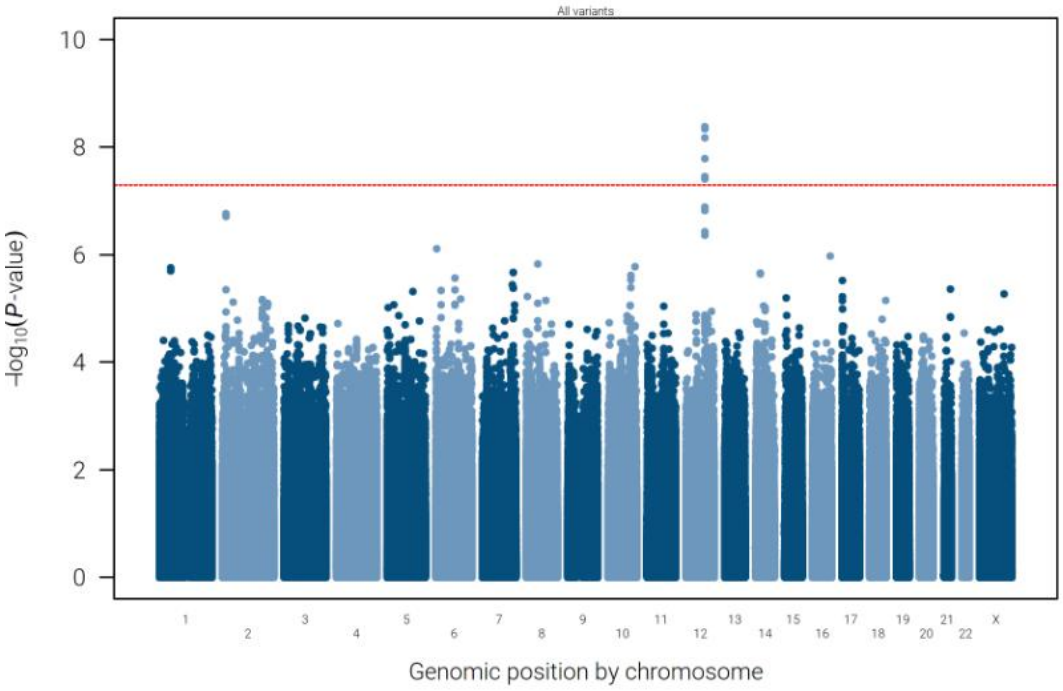
1. Price AL, Patterson NJ, Plenge RM, Weinblatt ME, Shadick NA, Reich D. (2006) Principal components analysis corrects for stratification in genome-wide association studies. *Nature Genetics* **38**(8):904-909.
2. The 1000 Genomes Project Consortium. A global reference for human genetic variation. Nature. 2015; **526**:68-74.
3. Li Y, Willer CJ, Sanna S, Abecasis GR. Genotype imputation. Annual Review Genomics and Human Genetics. 2009; **10:**387-406.
4. Howie B, Fuchsberger C, Stephens M, Marchini J, Abecasis GR. Fast and accurate genotype imputation in genome-wide association studies through pre-phasing. Nature Genetics. 2012; **44**:955-959.

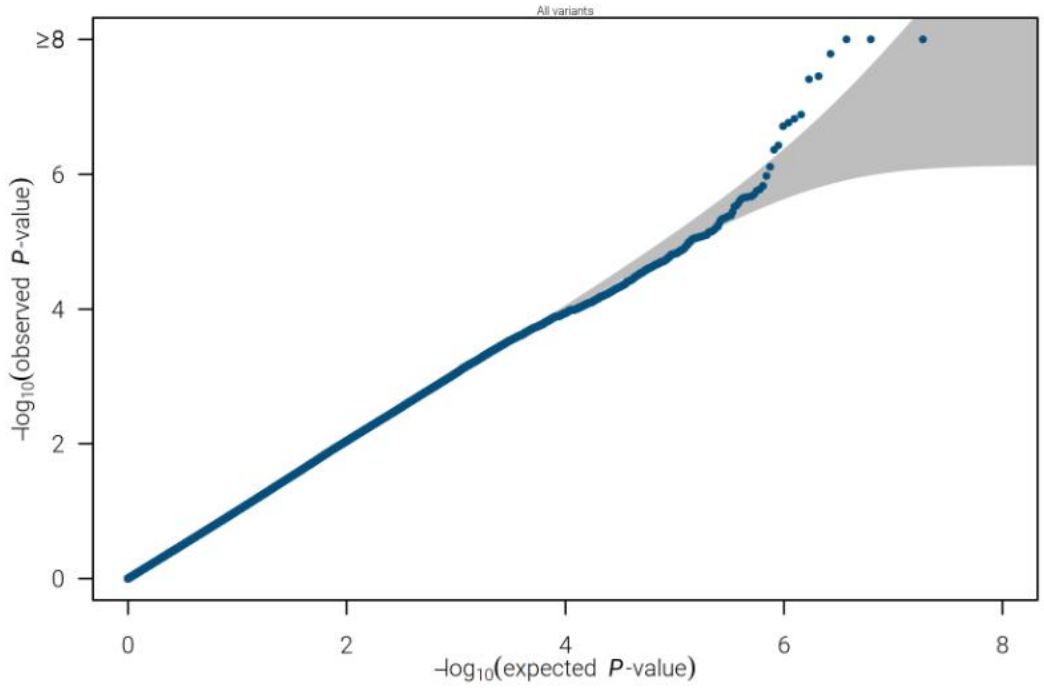
5. Zheng X, Shen J, Cox C, Wakefield J, Ehm M, Nelson M, Weir BS. HIBAG -- HLA genotype imputation with attribute bagging. Pharmacogenomics Journal. 2014; **14**:192-200.



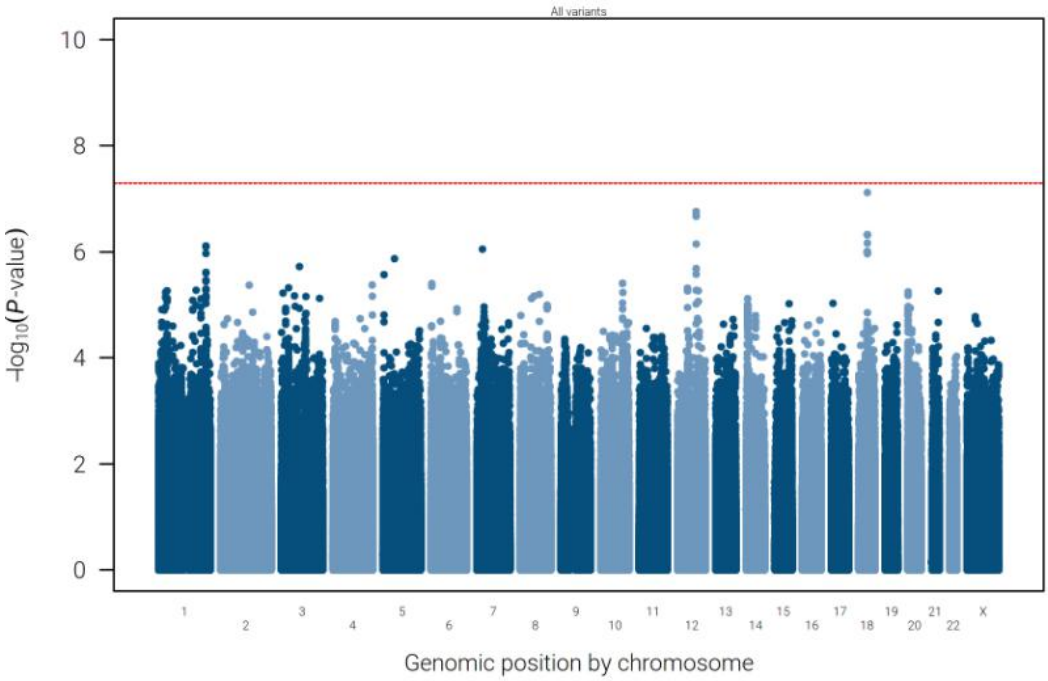


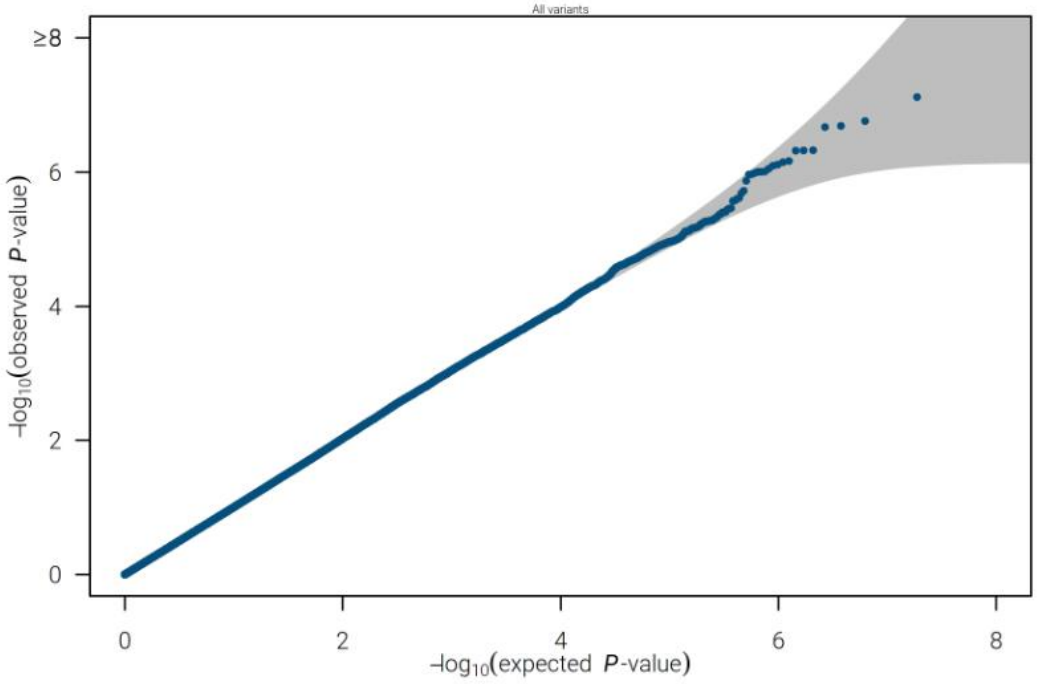
Supplementary Figure 1a and b. Manhattan and QQ plots for recurrence free efficacy at 6 months analysis in the PGx TQ group.



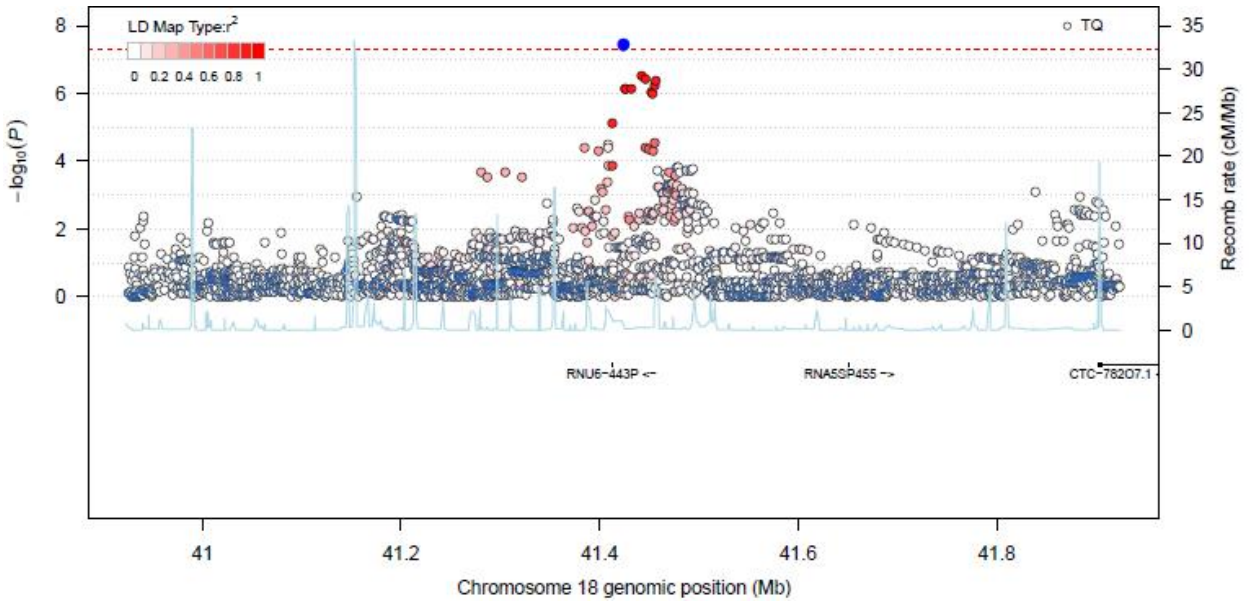


Supplementary Figure 1c and d. Manhattan and QQ plots for recurrence free efficacy at 4 months analysis in the PGx TQ group.

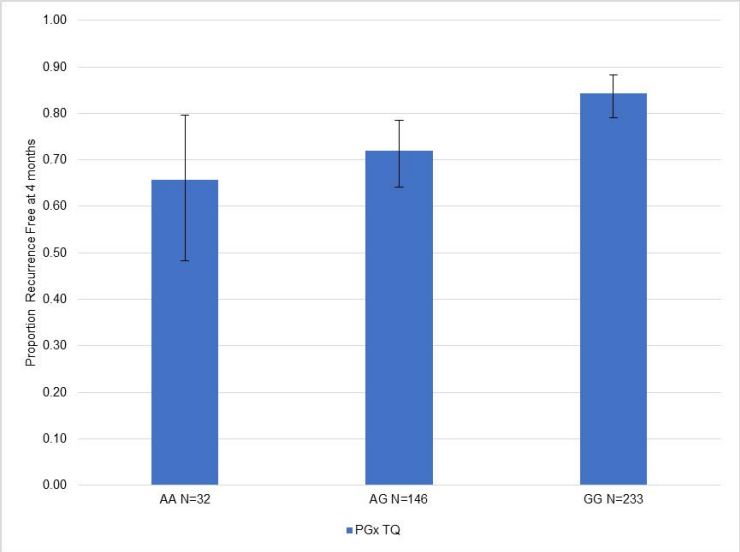


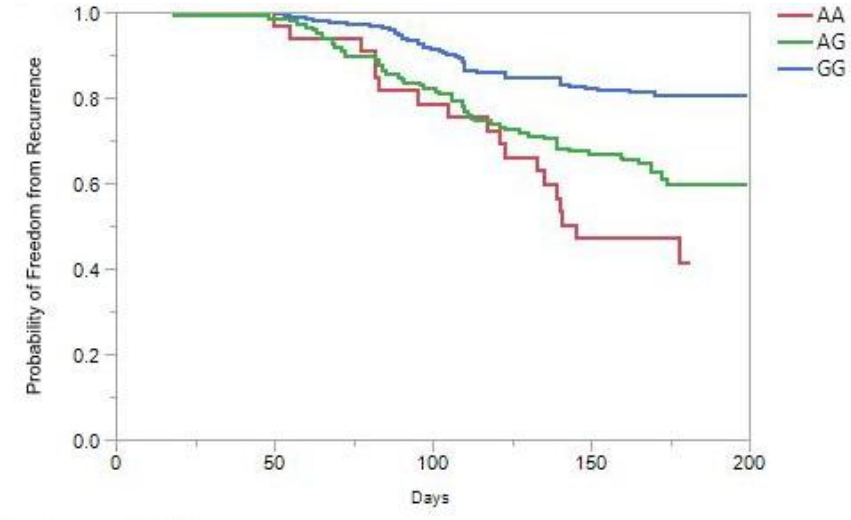


Supplementary Figure 1e and f. Manhattan and QQ plots for time to recurrence up to 6 months post dosing analysis in the PGx TQ group.

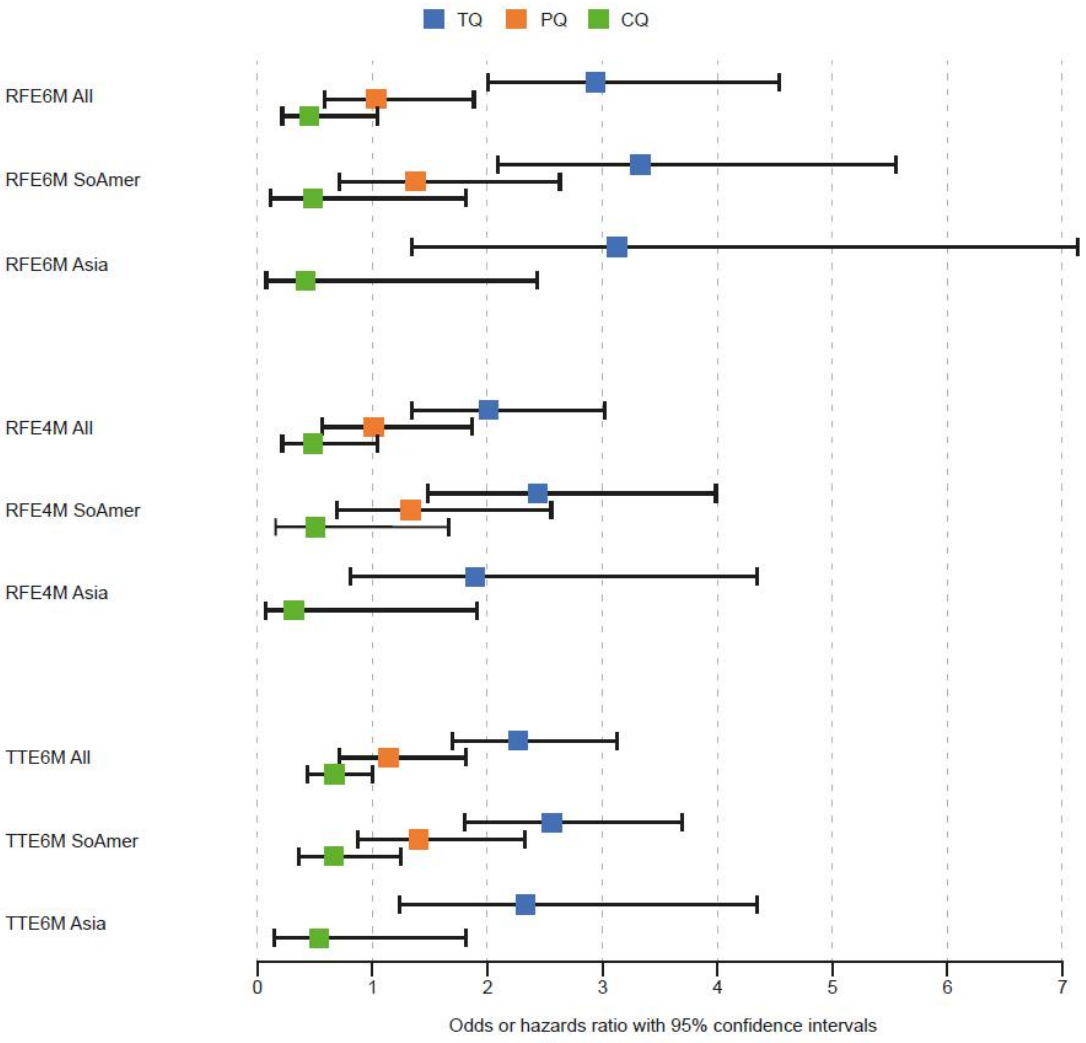


Supplementary Figure 2. Region plot for rs62103056 and recurrence free efficacy at 6 months in PGx TQ using hg19 map build.

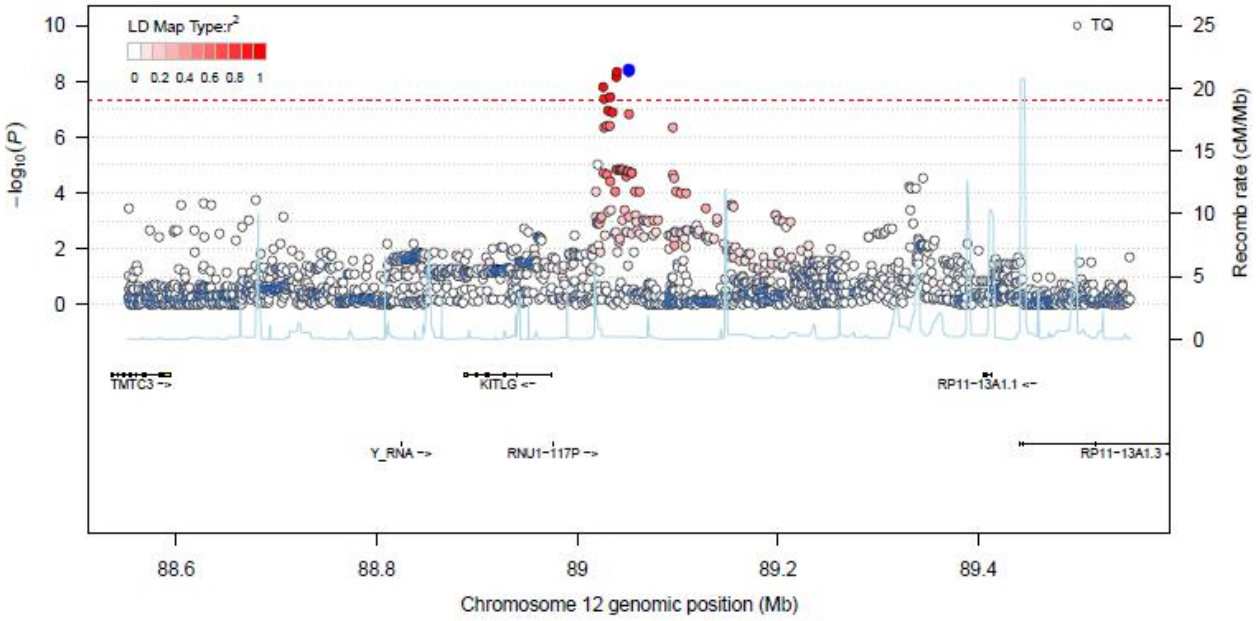




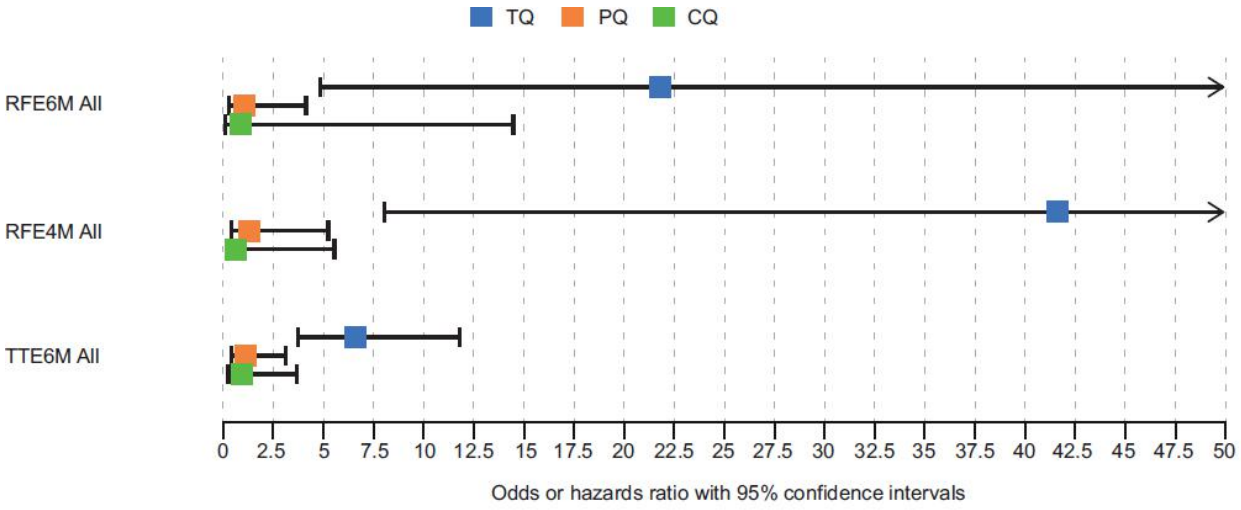
Supplementary Figure 3a and b. Results of rs62103056 in PGx TQ on a) recurrence free efficacy at 4-months with a per A allele odds ratio of 2.01 (95% CI 1.33, 3.02), p-value =0.0009 and b) Kaplan-Meier analysis of freedom from recurrence of *P. vivax* malaria up to 6 months post-dosing in PGx TQ with a per A allele hazards ratio of 2.30 (95% CI 1.71, 3.09), p-value = 7.63E-8; the number of participants with AA, AG and GG genotypes was 33, 153, and 252.



Supplementary Figure 4. Forest plot of rs62103056 results (odds or hazards ratio) across outcomes and PGx analysis groups. RFE6M=recurrence-free efficacy at 6 months, RFE4M=recurrence-free efficacy at 4 months, TTE6M=time to recurrence up to 6 months post-dosing, All = all patients without respect to geographic region, SoAmer = patients from South American region, Asia=patients from Asian region. Estimates within PQ Asia are unstable for all 3 endpoints and are not displayed.



Supplementary Figure 5. Region plot for rs11104986 and recurrence free efficacy at 4 months in PGx TQ using hg19 map build.



Supplementary Figure 6. Forest plot of rs11104986 results (odds or hazards ratio) across outcomes and PGx analysis groups. RFE6M=recurrence-free efficacy at 6 months, RFE4M=recurrence-free efficacy at 4 months, TTE6M=time to recurrence up to 6 months post-dosing, All = all patients without respect to geographic region. Due to the low minor allele frequency, estimates within the South American and Asian sub-groups are not shown.