

## Supplementary webappendix

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## APPENDIX MATERIAL

### **Artemisinin-resistant *Plasmodium falciparum* malaria in Pursat province, western Cambodia: a parasite clearance rate study**

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**Appendix 1: Summary of screening activities.**

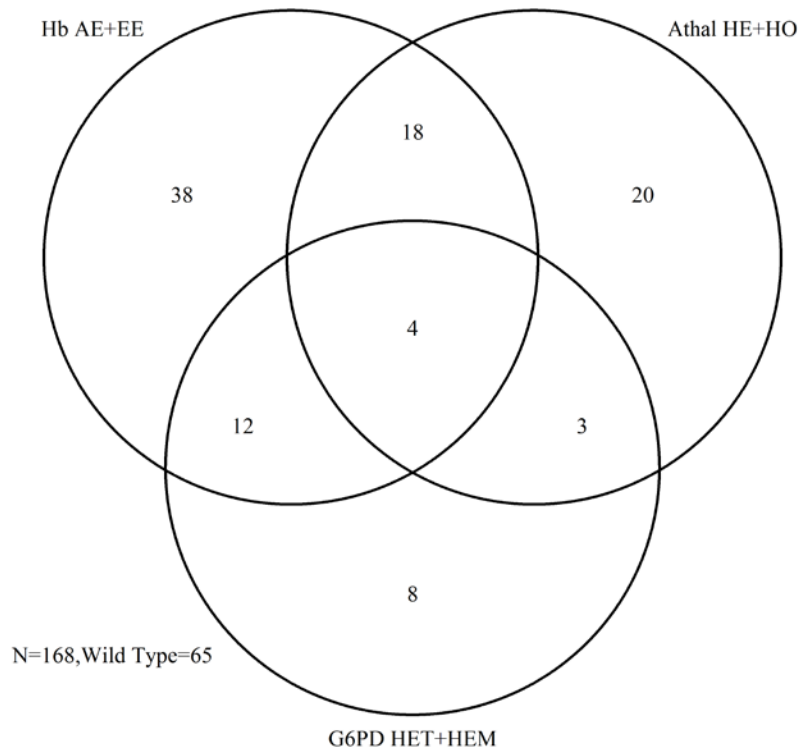
Year	2009			2010			2009 + 2010		
Screening period	June 19 – November 28			June 25 – December 26					
Screening site	SP <sup>a</sup>	SF <sup>b</sup>	All	SP	SF	All	SP	SF	All
<b>Total screened</b>	686	1066	1752	802	950	1752	1488	2016	3504
<i>Pf</i> +	136	294	430	173	109	282	309	403	712
<i>Pf</i> 4+ or 5+ <sup>c</sup>	88	56	144	118	54	172	206	110	316
<i>Pf</i> <10,000/µl	3	3	6	9	5	14	12	8	20
Severe malaria	22	3	25	24	6	30	46	9	55
Age <10 years	1	10	11	7	2	9	8	12	20
Pregnant	2	1	3	2	0	2	4	1	5
Refused to participate	2	1	3	6	3	9	8	4	12
Used antimalarial drug	13	1	14	5	4	9	18	5	23
Chronic illness	0	0	0	1	0	1	1	0	1
Enrolled	45	37	82	64	34	98	109	71	180
Removed from analysis <sup>d</sup>	3	0	3	7	2	9	10	2	12
Analyzed	42	37	79	57	32	89	99	69	168

<sup>a</sup> SP, Screening conducted at Sampov Meas Referral Hospital, Pursat town.

<sup>b</sup> SF, Screening performed on the main road from Pursat town to Veal Veng district.

<sup>c</sup> '4+' = 11-100 parasites, '5+' = >100 parasites per high-powered field.

<sup>d</sup> Patients were removed from analysis due to missing host genotype data.



**Appendix 2: Venn diagram showing the distributions of erythrocyte polymorphisms in 168 Cambodian patients with uncomplicated *P. falciparum* malaria enrolled in 2009 and 2010. HbAE=heterozygosity for HbA and HbE; HbEE=homozygosity for HbE; Athal HE=heterozygosity for  $\alpha$ -thalassaemia; Athal HO=homozygosity for  $\alpha$ -thalassaemia; G6PD HET= heterozygosity for G6PD deficiency ('Viangchan') allele; G6PD HEM= hemizyosity for G6PD deficiency ('Viangchan') allele.**

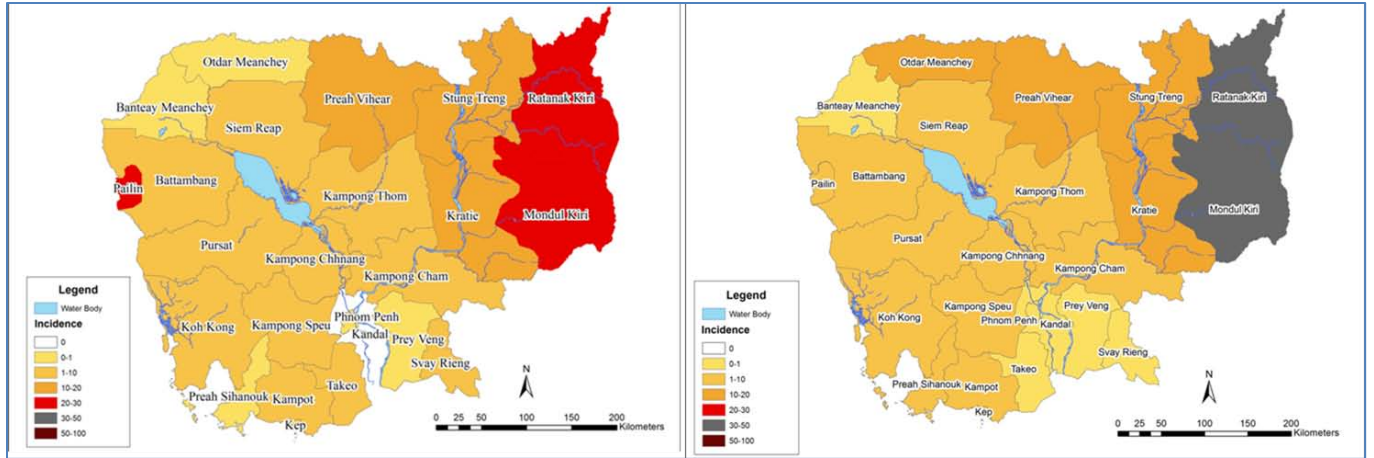
**Appendix 3: Results of regression analysis of parasite clearance rate (half-life) estimates.**

	Estimate	Std. Error	t value	Two-sided p-value	Lower 95% Confidence Limit	Upper 95% Confidence Limit
<b>(Intercept)</b>	5.112	2.274	2.247	0.027	0.599	9.624
<b>Sex Male</b>	0.958	0.512	1.873	0.064	-0.057	1.973
<b>Age 21+ years</b>	-0.211	0.36	-0.588	0.558	-0.925	0.502
<b>Village VV or KV</b>	-0.029	0.37	-0.079	0.937	-0.763	0.705
<b>Hemoglobin E</b>	0.551	0.309	1.781	0.078	-0.063	1.164
<b><math>\alpha</math>-thalassemia</b>	-0.334	0.375	-0.89	0.376	-1.079	0.411
<b>G6PD deficiency</b>	0.123	0.25	0.491	0.624	-0.374	0.62
<b>Ln(initial parasite density)</b>	-0.037	0.208	-0.177	0.86	-0.449	0.376
<b>Year 2010</b>	0.680	0.368	1.847	0.068	-0.051	1.41
<b>Parasite Group 1</b>	0.790	0.365	2.164	0.033	0.066	1.515

Parameter estimates and related statistics from the linear model with parasite clearance  $T_{1/2}$  as the response. The intercept is the estimated  $T_{1/2}$  in hours for the reference group (Female, Age < 21 years, Village not VV or KV, Hemoglobin AA, wildtype  $\alpha$ -globin genotype, no G6PD<sup>VC</sup> genotype, Ln(initial parasite density) equal to 0, Year 2009, and Parasite Group 2). Estimates are shifts in  $T_{1/2}$  in hours for changing each variable one at a time: the binary variables are changed compared to the reference group and the Ln(initial parasite density) is changed by increasing it by one unit.

**Appendix 4: *Ex vivo* IC<sub>50</sub> values for antimalarial drugs, stratified by parasite clone group.**

	Geometric mean (range) IC <sub>50</sub> (nM)		p (Mann-Whitney)
	Group 1	Group 2	
<b>Chloroquine</b>	341.90 (27.64-730.00)	236.30 (68.90-617.00)	0.1166
<b>Quinine</b>	257.90 (95.60-668.00)	362.20 (185.60-796.60)	0.1827
<b>Mefloquine</b>	16.46 (7.60-54.26)	36.59 (18.72-71.30)	<b>0.0007</b>
<b>Dihydroartemisinin</b>	3.14 (1.81-6.09)	3.78 (1.31-9.76)	0.3013



Appendix 5: Incidence of *P. falciparum* malaria per 1000 people in 2009 and 2010, by Cambodian province.