

Molecular Biology Laboratory  
BP Koirala Institute of Health Science  
RGQ Machine Nr RGQ\_01  
qPCR Experiment No: Q199  
Date : 26/07/2018  
PCR Name Malaria 5 qPCR  
SOP No. MLB\_BP\_0062\_1.1

1. To perform the Malaria multiplex PCR of clinical sample  
2. Required Material:

SN	Chemical /Reagents	Brand	Catalogue Number	Lot Number
1	PCR grade water	Himedia	ML024	0000117943
2	Primer/Probe Mix	AIT biotech	300230	E229-10
3	Enzyme/Reaction Mix	AIT biotech	300230	E229-10
4	Positive Control	AIT biotech	300230	E229-10
Number of reaction tubes		24		

SN	RXN components	Stock Concentration	Final concentration	Volumes/ tube (µl)	Final volume (µl)
1	Primer/Probe Mix	NA	NA	2	48
2	Enzyme/Reaction Mix	NA	NA	6	144
15	Water Nuclease free	NA	NA	12	288
16	DNA Template			5	
Total				25	480

Aliquot 20µl of master mix in each 23 Tubes: 21 Samples + 1 Positive Controls + 1 No template control (NTC)

3. Name of Rotor Gene Q: Rotatory ramp, Convexional Heating

4. Programme Condition:

Temp (°C)	Time	Cycles	Optics
95	2	min	1
95	5	sec	45
60	20	sec	On

5. qPCR Analysis:

Location of the Folder on Computer: D:\Molecular Lab\Google\_drive\qPCR\  
Name of the RGQ file: Q199\_Malaria\_qPCR\_V1 2018-07-26

Tube No	Sample code	ct VALUE					PCR RESULT	Malaria Species
		FAM (P.falciparum)	HEX (P.malariae)	ROX (P.vivax)	Cy5 (P.ovale)	Quasar 705 (P.knowlesi)		
1	All Positive Control	28.01	29.09	24.69	28.85	24.71	POSITIVE	Pos control
2	No template Control						NEGATIVE	
3	MP135						NEGATIVE	
4	MP136						NEGATIVE	
5	MP137						NEGATIVE	
6	MP138						NEGATIVE	
7	MP139						NEGATIVE	
8	MP140						NEGATIVE	
9	MP141						NEGATIVE	
10	MP142			37.81			POSITIVE	P.vivax
11	MP143						NEGATIVE	
12	MP144						NEGATIVE	
13	MP145			20.71			POSITIVE	P.vivax
14	MP146			15.24			POSITIVE	P.vivax
15	MP147			32			POSITIVE	P.vivax
16	MP148			30.53			POSITIVE	P.vivax
17	MP149						NEGATIVE	
18	MP150			32.56			POSITIVE	P.vivax
19	MP151			34.91			POSITIVE	P.vivax
20	MP152						NEGATIVE	
21	MPC1						NEGATIVE	
22	MPC2						NEGATIVE	
23	MPC3						NEGATIVE	

