Malaria Diagnosis Using ATR-FTIR Spectroscopy

Bayden R. Wood[1], Phil Heraud[1], David Perez-Guita[1], Aazam Khoshmanesh[1], Patcharee Jearanaikoon[2], Matthew W. A. Dixon[3], Leann Tilley[3] and Don McNaughton[1]

[1]Centre for Biospectroscopy, School of Chemistry, Monash University, 3800, Victoria, Australia
[2]Department of Clinical Chemistry, Faculty of Associated Medical Sciences, Khon Kaen University, Khon Kaen 40002, Thailand
[3]Department of Biochemistry and Molecular Biology and Bio21 Molecular, Science and Biotechnology Institute, The University of Melbourne, Melbourne, Victoria 3010 Australia

Malaria is one of the most deadly diseases resulting in over 600,000 fatalities per annum.(*1*) Accurate and early diagnosis followed by the immediate treatment of the infection is essential in reducing mortality(*2*). New technologies to diagnose malaria must be cost effective and have high sensitivity to enable the detection of premature parasitic forms in peripheral blood. During the course of its life the malaria parasite transgresses through several developmental stages including a sexual and an asexual reproductive pathway. The detection of the rings and gametocytes at low parasitemia in peripheral blood is critical for early diagnosis and treatment. Here we show that Attenuated Total Reflection-Fourier Transform Infrared (ATR-FTIR) spectroscopy in combination with a partial least squares regression modeling has the required sensitivity and ease of sample preparation to become a laboratory standard for malaria detection and most importantly quantification. The absolute quantification limit was found to be 0.001% (50 parasites/uL of blood) for cultured ring stage and gametocyte parasites in a suspension of normal erythrocytes. The absolute detection limit was found to be 0.00001% for laboratory cultured parasites.(*3*) The method is simple, quick and only requires the whole blood to be spun down and the plasma and white cells removed. The red blood cells are then fixed in methanol and a 5 μL aliquot of packed cells is placed on the diamond window of the ATR-FTIR spectrometer, rapidly dried and a spectrum recorded in approximately 20 seconds. In December 2014 we commenced a pilot trial in North East Thailand investigating the potential of the technology to diagnose malaria in a field/clinical setting. Samples were collected from two independent clinics and analysed with two independent ATR-FTIR spectrometers. The results were combined and a Partial Least Squares Discriminant Analysis (PLS-DA) model developed and tested on a totally independent test set. Although the sample number was small 28 negatives and 81 positives (including patients infected with *P. falciparum*, *P. vivax* and a mixture of both) the replicate number was high as three aliquots for each sample were analysed. The percentage of correctly diagnosed spectra based on comparison with antibody capture rapid diagnostic test (RDT) was 97 %. The high sensitivity, low cost, ease of use, portability and robustness of the ATR-FTIR technique could see it become a standard diagnostic tool in both the clinic and remote field locations.

References

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