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TOWARDS DEVELOPMENT OF A MASSTAG PCR ASSAY FOR THE SYNDROMIC DETECTION OF PATHOGENS THAT CAN CAUSE ENCEPHALITIS

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Abstract
MassTag PCR is a novel technology for the rapid, sensitive and simultaneous detection of multiple gene sequences. This technique, developed at the Center for Infection and Immunity (CII) at Columbia University, utilises a library of unique tags, each unique its molecular weight. MassTags are conjugated to oligonucleotide primers using a UV-cleavable linker that enables separation of primer and tag. Primers are labelled with a unique molecular weight tag and are used to amplify target nucleic acids in a multiplex RT-PCR. After removing unincorporated primers, tags are released by UV irradiation and analysed by mass spectrometry. Thus, amplification of the gene target produces a unique dual signal in mass spectrometry analysis that allows its identification. MassTag PCR offers an inexpensive and sensitive diagnostic platform suitable for high-throughput testing, and that can be adapted to suit diagnostic needs (e.g., syndrome-, vector-based). To date, MassTag PCR panels have been developed for the detection of respiratory pathogens and viruses that cause haemorrhagic fever. A third MassTag PCR assay is being developed to identify microbial agents that cause neurological disease in a North American/European diagnostic setting. In collaboration with the CII and PathWest Laboratory Medicine, we have begun modifying and developing this assay to address pathogens relevant to the Australasian region. Initial research has involved extensive bioinformatics analyses for the design of primer sets that are specific only for the cognate target pathogen and that will not cross-hybridise with existing primers comprising the panels. This has been followed by *in vitro* evaluation of multiplex panels using untagged primers and agarose gel analysis. Preliminary research experiences and progress towards development of this assay will be addressed in the accompanying presentation.

Personal Profile
David Williams is a Senior Research Fellow at the AB-CRC group located at Curtin University of Technology, Perth, Western Australia. His research interests include the molecular characterization and epidemiology of arboviruses of the Australasian region and novel microbial genome detection technologies for emerging pathogen identification.