Development of Novel Techniques for Monitoring Anti-oxidant Thiols

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Abstract

The clinical exploitation of physiological biomarkers could yield considerable improvements in diagnosis and treatment providing their measurement can be conducted speedily and preferably at the point of care. The project has sought to investigate the development of new methods that could allow such measurements to be made. The various biomarkers that could be exploited as the basis of a general "index" of physiological wellbeing have been identified and their potential clinical merit critically appraised. Anti-oxidant sulphur compounds (cysteine, glutathione, sulphite) were selected as potential targets on the basis of their physiological role in protecting the body from damage by free radicals. The variation in their concentration within biofluids is widely acknowledged as a useful diagnostic gauge as to the degree of oxidative stress that an individual may be experiencing. The main problem, from a clinical perspective, is the lack of a suitable procedure for monitoring their concentration speedily at, or by, the patient.

A brief assessment of the various electroanalytical options (encompassing voltammetric, amperometric and potentiometric methodologies) available for the detection of the sulphur anti-oxidants has been conducted. A potentiometric detection strategy was found to offer numerous advantages and a novel indicator family based on quinone interaction adopted and forms the foundation of the work presented herein. The reaction mechanism has been elucidated and the analytical applicability of the system

investigated using a variety of techniques – covering both chromatographic, spectroscopic and electrochemical methodologies.

The system has been characterised in terms of selectivity, sensitivity and its efficacy for the quantification of thiol containing pharmaceuticals and various biofluids (urine and plasma) has been demonstrated. The simplicity of the detection methodology is shown to markedly contrast alternative thiol detection strategies. The transfer of the technology to a mass production format through the adoption of screen print electrode formats has been achieved. A series of clinical trials were performed and the efficacy of the approach and the underlying technology format demonstrated. The results have been corroborated using standard techniques and the routes through which the system can be adopted within mainstream biomedical environments highlighted.

An alternative sensor system based on a composite polymer laminate approach was also investigated as a route through which prototype sensors could be speedily prepared and which would be more accessible to general chemistry laboratories. A new approach to the detection of sulphite – based on the quinone system – was used as the principal detection system to allow the system to be evaluated and proof of principle demonstrated. The fabrication methodology adopted has been found to provide a highly versatile option for the construction of polymer film electrodes.

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Chapter 1 Introduction to Decentralised Testing

Chapter 2 Experimental Details and Methodologies

Chapter 3 Electroanalytical Detection of Oxidative Stress Biomarkers: A Preliminary Assessment

Chapter 4 Preliminary Investigation of Quinone – Thiol Biomarker Interactions