Bioelectrochemical response of thiol compounds in quinone mediated systems.

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Anti-oxidants are known to play a crucial role in the prevention of free radical induced oxidative damage and there are many incidences where being able to monitor their concentration in the body would be highly beneficial [1, 2]. The sulphydryl thiols, cysteine and glutathione, are biochemically the amino acid building blocks of proteins, as well as antioxidants capable of reducing the damage induced in oxidative stress conditions. Such conditions are enhanced by diseases such as diabetes, where the thiol content in blood can vary and be an indicator of a specific condition [3]. For these reasons it is important to assess the efficacy of utilising such biomarkers in a point of care monitoring context.

The suitability of electrochemical techniques to this type of analysis is clear. It should be possible to obtain a quick response to the analyte concentration, that should minimise problems of sample degradation that areassocuated with traditional, laboratory based techniques. The methods are also easily miniaturised, which improves their suitability to mass production, and subsequent availability to patients in self assessment[4].

In this presentation, the electrochemistry of glutathione has been investigated with respect to a benzoquinone mediated system and it potential for use within biomedical contexts appraised. The three possible mechanisms for the mediation are given in **Figure 1**. Route 1 illustrates benzoquinone (**I**) oxidising the RSH to RSSR and in the process being converted to hydroquinone (**II**). The alternative route is highlighted in 2 where the reaction of RSH with benzoquinone to yield the quinone conjugate of SR (**III**). Sequentially, route 3 shows the further conversion by the chemical oxidation of the reduced form of the thiol conjugate (**III**) by another molecule of benzoquinone, yielding the hydroquinone and the oxidised thiol conjugate (**IV**). It is our contention that the analytical signal is derived from the electrochemical oxidation of hydroquinone, and not via the sole electro-oxidation of the reduced thiol conjugate (**III**).

Elucidation of the mechanism is clearly important as this approach does possess considerable analytical merit. The viability of the benzoquinone mediated system is shown in **Figure 2** where the direct oxidation of glutathione in pH 7 buffer is examined. Direct oxidation at a bare carbon electrode shows a poor response. However, the response of the same electrode/analyte system in the catalysed system is significantly enhanced. Both series of experiments show the introduction of 15µmol l^{-1} aliquots of glutathione. The technological requirements for the development of a working sensor are not too dissimilar to those utilised in blood glucose monitoring and data, highlighting the applicability of the technique to the analysis of thiols within common physiological fluids, will be presented.

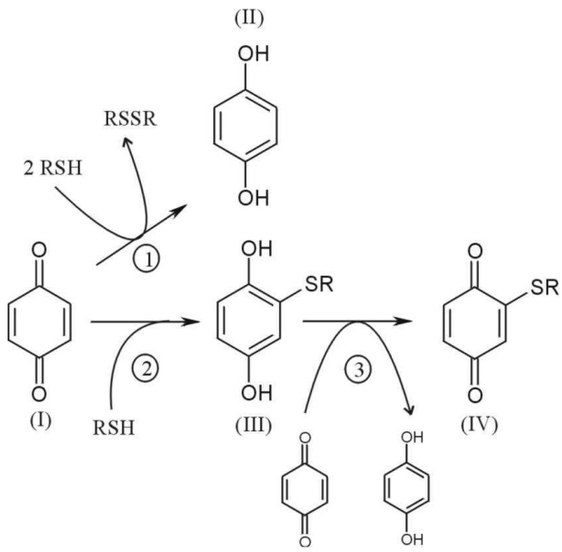

Figure 1. Possible reaction mechanisms for mediation.

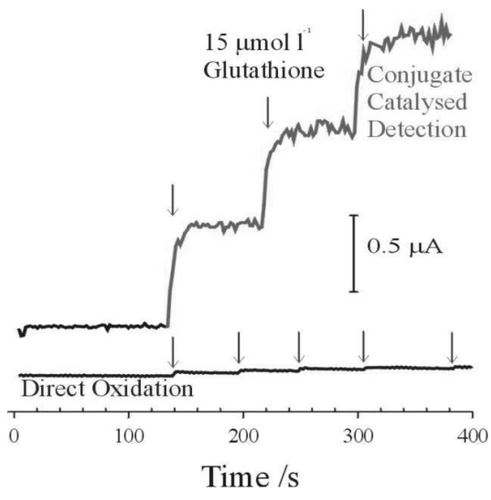

Figure 2. Amperometric response of glutathione at bare carbon electrode in the presence and absence of benzoquinone.

References:

1. West, I.C., *Radicals and oxidative stress in diabetes.* Diabetic Medicine, 2000. **17**: p. 171.
2. Lang, C.A., et al., *Blood glutathione decreases in chronic diseases.* Journal of Laboratory and Clinical Medicine, 2000. **135**: p. 402.
3. Gutteridge, M.C. and B. Halliwell, *Antioxidants in nutrition, health and disease.* 1996: p. 96-97.
4. Lawrence, N.S., et al., *Advances in the Voltammetric Analysis of Small Biologically Relevant Compounds.* Analytical Biochemistry, 2002. **303**: p. 1-16.