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Disposable electrochemical immunosensor for cortisol determination in human saliva

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Abstract

Cortisol is a steroidal hormone and an important stress marker. Free serum cortisol concentration has been identified to correlate well with free salivary cortisol. In this present work an electrochemical immunosensor was developed to determine cortisol concentration within the physiological concentration range found in human saliva. The immunosensor is based on a direct competitive enzyme linked immunoassay using a home-made cortisol-alkaline phosphatase (AP) conjugate synthesized in our laboratory with disposable graphite screen-printed electrodes (SPEs). 1-nalptyl phosphate (1-NP) was used as an enzymatic substrate and a square wave voltammetry (SWV) for electrochemical detection. To study method suitability for use with saliva samples, calibration curves were performed both in buffer and saliva. In buffer standard samples showed a limit of detection (LOD) of 0.6 ng/ml and working range (WR) of 0.2–44.6 ng/ml with good reproducibility (RSD 10%). Saliva matrix effect was removed effectively with Salivette Cortisol collection device (polyethylene) and a calibration curve showed similar characteristics as in buffer with LOD 1.7 ng/ml and WR 0.5–55.1 ng/ml (RSD 8%) demonstrating the possibility to determine human salivary cortisol within the desired human physiological range. Spiked saliva samples were analyzed with the developed immunosensor presenting excellent 92–114 % recovery.

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