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**Dual-aptasensor array: Optimization of the aptamers immobilization.**

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In the present work it is intended to establish the optimal experimental conditions that allow enhancing the performance of an electrochemical dual-aptasensor array for the detection of osteopontin, which has been reported as a cancer biomarker. The aptamer concentration, time and temperature of immobilization onto dual-screen printed gold electrodes (Dual-SPGE), as well as the aptamer-protein interaction time were evaluated using a 2<sup>k</sup> factorial experimental design. The preliminary results allowed identifying the optimal experimental conditions, namely: (i) aptamer concentration of 0.5 µM for both DNA and RNA aptamers; (ii) incubation temperature of 4°C; (iii) time for aptamer immobilization onto the Dual-SPGE at 4°C of 20 min; and, (iv) aptamer-protein interaction time of 30 min. The evaluation of the best experimental conditions was carried out by cyclic and square wave voltammetry using a ferro/ferricyanide solution ( $[\text{Fe}(\text{CN})_6]^{3-/4-}$ ) as a redox probe. Aptamers were immobilized onto the Dual-SPGE via streptavidin-biotin interaction. With the preliminary optimal experimental conditions herein obtained, we will further assess the sensitivity of the Dual-SPGE using synthetic plasma.

**Keywords:** Aptasensor; Aptamers immobilization.

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