Área: **ELE**

Thiolated chitosan: a sustainable alternative for metal surface functionalization for biosensor-based dengue diagnosis

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Highlights

- Development of a new biosensing platform based on chitosan modified with thiol groups.
- haracterization of thiolated chitosan using electrochemical techniques and surface plasmon resonance.

Abstract

Chitosan, a natural polysaccharide, stands out for several advantages, such as low cost, biocompatibility, ability to form stable films, and ease of chemical modification, which allows the incorporation of different functional groups depending on the desired application. Naturally, chitosan is released into the environment by crustaceans during exoskeleton shedding; however, human activities can also contribute to this increase and accumulation of material in water, affecting marine life. Therefore, the search for new applications for chitosan has become essential to reduce its environmental impact. In this context, chitosan presents itself as a sustainable substitute for thiol alkanes in metal surface functionalization for sensor development. In this study, some terminal amino groups of chitosan were modified with thiol groups, enabling the development of a chitosan-thiol-based biosensing platform. This modification was carried out through a coupling reaction via EDC:NHS of the amino groups with 3-mercaptopropionic acid. The success of the proposed modification was confirmed via surface plasmon resonance (SPR), where it was shown that thiolated chitosan exhibited greater affinity for gold compared to native chitosan. This behavior can be attributed to the formation of covalent bonds between the thiol groups and gold, promoting more efficient surface functionalization. The characterization of the gold substrate functionalization was performed using different electrochemical techniques, including Cyclic Voltammetry (CV), Electrochemical Impedance Spectroscopy (EIS), and Square Wave Voltammetry (SWV). In these analyses, the effect of surface charge with the incorporation of the new material was investigated, using different electrochemical mediators, in addition to studying the apparent pKa of the surface and the desorption reduction to determine surface coverage. After optimizing the substrate functionalization conditions, studies were conducted on the biosensor construction steps, using SPR and the previously mentioned electrochemical techniques. During the immobilization of the recognition unit – the Dengue virus envelope protein – the influence of pH was assessed, with more efficient anchoring observed under slightly acidic conditions (pH = 5.8). Finally, the proposed platform allowed the differentiation of positive and negative human serum samples for Dengue antibodies via SPR. In brief, thiolated chitosan appears to be highly promising for the construction of biosensors, as its three-dimensional structure provides an expanded surface area, with more available sites for biorreceptor anchoring, thereby enhancing the efficiency of the immobilization process. It is important to highlight the need for new Dengue diagnostic platforms, as the disease's spread is increasingly influenced by climate change, which promotes the proliferation of its vector, *Aedes aegypti*.

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