Application and testing of chitosan protective layers for screen printed carbon electrodes (SPCE)

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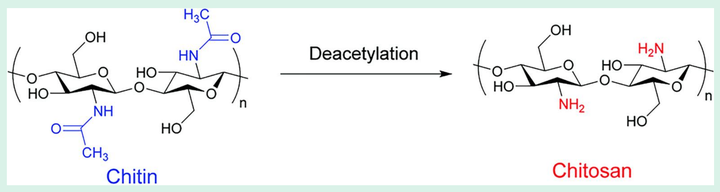
Introduction

Bioelectrodes play a critical role in electrochemical sensing, with applications in areas such as environmental monitoring (Tsai et al (2021)), glucose detection (Mross et al (2015)), and more. Enzyme-based bioelectrodes are key components of biosensors, leveraging the specificity of enzymes (biorecognition elements) to detect and quantify target substances in complex fluids, such as wastewater and biological fluids (George et al (2022)). However, enzymes are highly sensitive and prone to inactivation after prolonged exposure to high temperatures (Wang et al (2016)) and their performance is often hampered by interferences (Campuzano et al (2019)).Chitosan is a biopolymer derived from the partial deacetylation of chitin and is formally composed of glucosamine and N-acetylglucosamine units.



Aim of the study

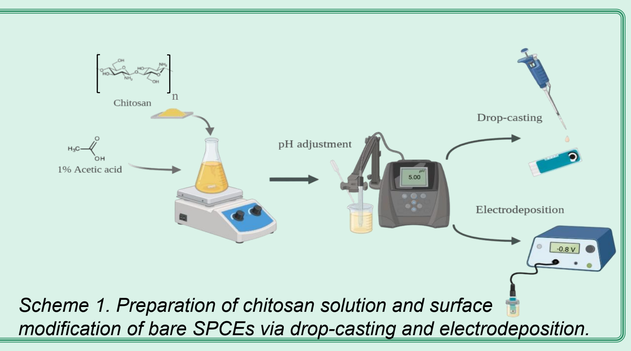
Thanks to its biocompatibility and pH-responsive behavior, chitosan is employed in electrochemical (bio)sensors, either as a matrix for immobilizing nanoparticles or biological recognition elements, or as a protective layer that provides antifouling properties, depending on the application (Bocchetta *et al* (2024); Kaushik *et al* (2009); Zhang *et al* (2014))..

In our study, commercial screen-printed carbon electrodes were modified with chitosan using two distinct deposition techniques: drop-casting and electrodeposition and evaluated for their ability to be deposited and subsequently be removed upon immersion in water (or any other simple (electro)-chemical procedure) leaving no residue on the electrode surface. The dissolution capability was evaluated using ATR-FTIR, Confocal Laser Scanning Microscopy (CLSM) and Atomic Force Microscopy (AFM). In addition, the selected materials will be modified to increase their water solubility under specific pH conditions

Materials and Methods

Screen-printed carbon electrodes (model C110, Metrohm) incorporating a three-electrode configuration (carbon working electrode, carbon counter electrode, and silver pseudo-reference electrode) were used as the electrochemical platform. A chitosan solution was prepared by dissolving finely powdered chitosan in 1% (v/v) acetic acid at an initial concentration of 10 mg/mL. The solution was subsequently diluted to 5 mg/mL by the dropwise addition of a dilute aqueous NaOH solution until the pH reached 5. For drop-casting, different volumes of the chitosan solution (5–20 µL) were applied onto the working electrode surface. Electrodeposition was carried out by applying a constant potential of -0.8 V for varying deposition times (3–10 minutes). The reversibility of these modifications was evaluated by immersing the modified electrodes in a 0.2 M HCl aqueous solution under stirring, followed by CV measurements after short periods of immersing times.

Electrochemical characterization of the electrodes before and after each modification was performed using cyclic voltammetry and electrochemical impedance spectroscopy. A 5 mM solution of potassium ferricyanide ($K_3[Fe(CN)_6]$) was used as the electroactive species, in 0.1 M KCl serving as the supporting electrolyte.



Scheme 1. Preparation of chitosan solution and surface modification of bare SPCEs via drop-casting and electrodeposition.

Results

Modification via drop-casting

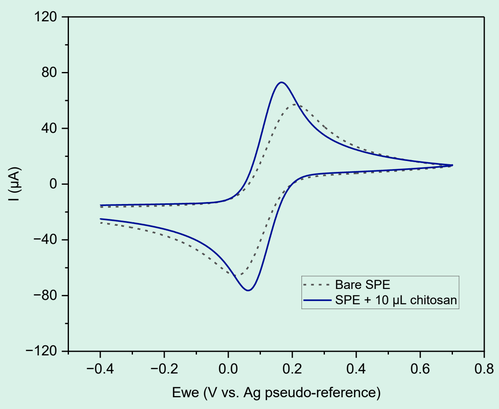


***Figure 1**. Cyclic voltammograms of bare SPCE and SPCE modified with 10 µL of chitosan solution by drop-casting.*

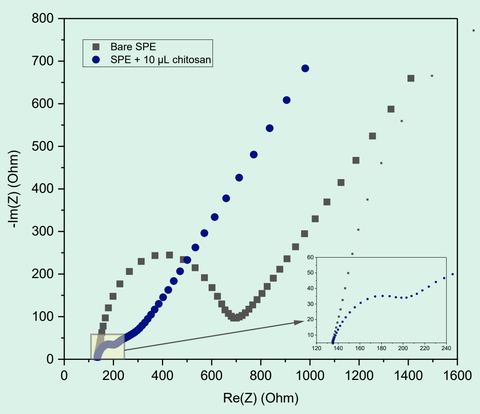


***Figure 2**. Nyquist plot of bare SPCE and SPCE modified with 10 µL of chitosan solution by drop-casting.*

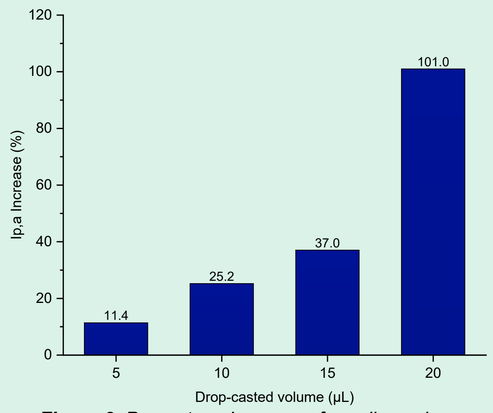


***Figure 3**. Percentage increase of anodic peak current compared to bare SPCEs after surface modification with chitosan by drop-casting using different solution volumes.*

Modification via electrodeposition

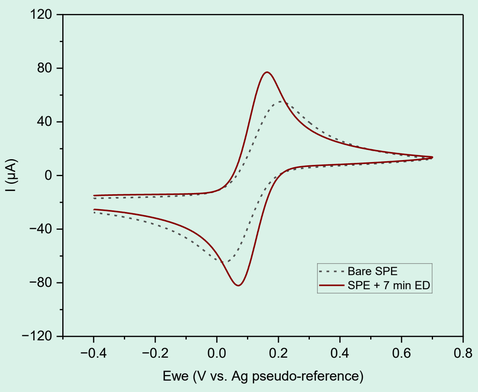


***Figure 4**. Cyclic voltammograms of bare SPCE and SPCE modified with chitosan by electrodeposition at -0.8 V for 7 min.*

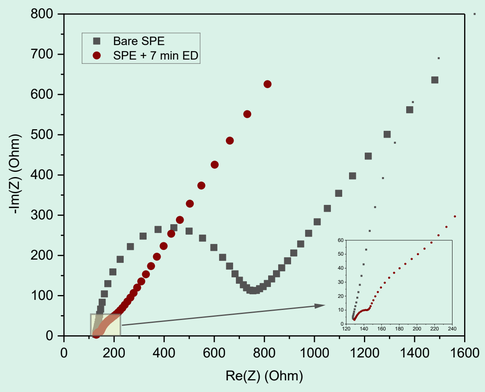


***Figure 5**. Nyquist plot of bare SPCE and SPCE modified with chitosan by electrodeposition at -0.8 V for 7 min.*

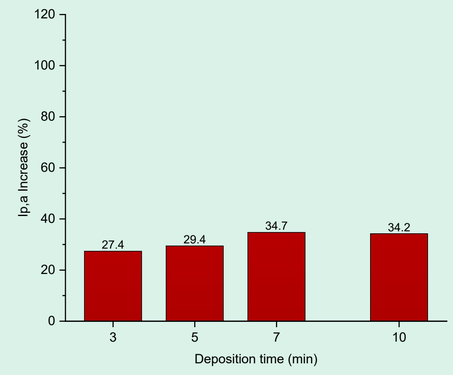


***Figure 6**. Percentage increase of anodic peak current compared to bare SPCEs after surface modification with chitosan by electrodeposition for different deposition times.*

Modification of the screen-printed carbon electrode surface with chitosan, using both drop-casting and electrodeposition methods, resulted in a notable enhancement of the electrochemical response. As shown in the cyclic voltammograms (**Figures 1 and 4**), the modified electrodes exhibited increased peak currents and a decrease in peak-to-peak separation compared to the bare SPCEs, indicating improved electron transfer kinetics. Additionally, electrochemical impedance spectroscopy revealed a significant reduction in the charge transfer resistance (Rct) for both modification approaches, as observed in the Nyquist plots (**Figures 2 and 5**). The percentage increase in anodic peak current (Ip,a) for different drop-casted volumes is presented in **Figure 3**, while the corresponding values for various electrodeposition durations are shown in **Figure 6**. The electrochemical response of the chitosan-modified electrodes decreased progressively and tended to approach that of the bare SPE as the immersion time in the dilute HCl solution increased, as shown in **Figure 7**. This reduction is likely attributed to the more effective removal of chitosan from the electrode surface over time.

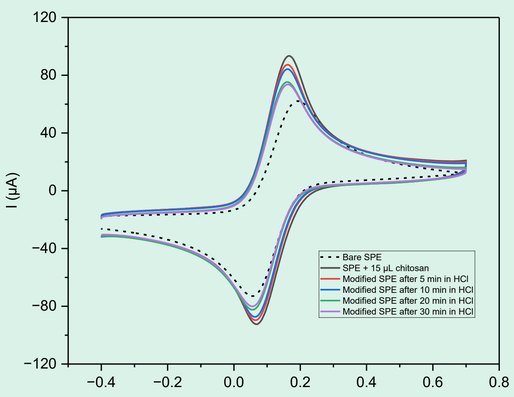


***Figure 7**. Cyclic voltammograms of chitosan-modified SPCE by drop-casting after different immersion times in dilute HCl solution.*

Conclusions

Screen-printed carbon electrodes were successfully modified with chitosan using drop-casting and electrodeposition methods. Cyclic voltammetry and electrochemical impedance spectroscopy revealed that the modified electrodes exhibited enhanced electron transfer kinetics compared to the unmodified ones. The improved electrochemical response of the modified electrodes highlights the potential use of chitosan as a functional layer for electrochemical biosensors. This response can be partially reversed by immersion in a dilute hydrochloric acid solution; however, this method is not recommended in the presence of biological elements.

References

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