

Development of chitosan-based electrochemical sensors for copper detection

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Abstract

We report on interdigitated electrodes functionalized with layer-by-layer (LbL) films of carboxymethylcellulose (CMC) and chitosan (CHI) for copper ion detection. Variations in CMC/CHI bilayers (1, 3, and 5) and the degree of deacetylation (DD \approx 95%, 75%, 55%) of high molecular weight chitosans (approx. 10^6 g/mol) were explored for their impact on electrode sensitivity using anodic stripping voltammetry. Sensors produced with 95% deacetylated chitosan displayed nearly double the current intensity highlighting amino group influence on sensitivity. Rougher films showed enhanced copper detection. Optimal performance was achieved with films containing three bilayers, demonstrating good reproducibility and repeatability (standard deviation 2%). The copper ion concentration range was linear from 0.5 to 2.5 ppm, with a detection limit of 0.05 ppm. The enhancement mechanism of CMC/CHI coatings is mainly attributed to their physical-chemical characteristics and the available amino groups in CHI. The nitrogen atoms in CHI's amino groups assist in the copper ion complexation, enhancing sensor effectiveness. CHI's DD provides novel control over LbL film properties, boosting the sensor's analytical capability for highly sensitive metal ion determination.

Keywords: chitosan; copper; electrochemical sensor; anodic stripping voltammetry.

1. Introduction

Heavy metals are often linked to contamination and toxicity, making their monitoring essential for environmental and public health safety, especially in water contamination cases [1]. Typically, metal ion detection in water is performed in labs with methods like Atomic Absorption Spectroscopy (AAS), which require costly equipment and expert training. This highlights the need for simpler, faster, and more affordable *in situ* alternatives [2]. One such alternative is the use of electrochemical sensors that have gained significant prominence in several research areas due to advancements in electroanalytical techniques. They are low-cost, easy to reproduce and handle, highly sensitive, and portable, making them versatile for various environmental applications [3].

A promising alternative is using chitosan (CHI) for sensors, due to its chelating capacity from unshared nitrogen electrons. Chitosan, a

polysaccharide derived from the deacetylation of chitin, contains amino and hydroxyl groups in its structure [4]. The composition of chitosan varies based on its average degree of deacetylation (DD), which measures the quantity of amino groups within the polymer chain. Consequently, the DD affects the chelating capacity of the polymer, as the interaction between chitosan and metal ions primarily involves the amino groups available [5].

Chitosan can be used for metal ion detection as films, where its structure and thickness can be precisely controlled [6]. These films can be obtained by the Layer-by-Layer (LbL) method, which involves a thin and alternating polyanion and polycation layer formation via adsorption [7]. For LbL films with chitosan, carboxymethylcellulose (CMC) can be used as the polyanion. CMC, an anionic polyelectrolyte derived from cellulose, has hydroxyl and carboxymethyl groups in its chemical structure [8]. As reported in the literature [9], the LbL technique with natural polymers effectively detects copper ions.

Natural polymers like chitosan and CMC can be used as electrode modifiers in film form, since these films have charged sites on their surface that can interact with the species to be detected, enabling the development of highly sensitive sensors [10].

In this study, gold electrodes were modified via adsorption with chitosan-based films containing different degrees of deacetylation (DD) to investigate their interaction with copper ions through electrochemical measurements. The results highlight the significant potential of these polymers for metal ion detection devices.

2. Methodology

CHI, PEI, and CMC solutions were prepared at 1.0 g/L in ultrapure water, with CHI dissolved in 0.1 M acetic acid and PEI adjusted to pH 4.0 with 0.1 M NaCl. Chitosans of varying DD were prepared at 0.1 g/L in ultrapure water with HCl. The pH of CMC and CHI solutions was set to 3.0 using 1.0 M HCl or 1.0 M NaOH. All solutions were stirred for 24 hours.

Interdigitated gold electrodes were prepared on a glass substrate following established methods [11]. The electrodes were washed sequentially in isopropanol, ethanol, and Milli-Q water, then exposed to O₂ plasma for 11 minutes. Subsequently, a positively charged monolayer of PEI was deposited by a 15-minute immersion, followed by three rinses with Milli-Q water at pH 4.0 for 2, 1, and 1 minute, consecutively.

Nanometric CMC/CHI films were deposited on electrodes using the LbL method by alternating 10-minute immersions in CMC and CHI solutions, with three rinses in Milli-Q water (2 min, 1 min, 1 min) between each. Films with 1, 3, and 5 bilayers were created to find the optimal sensor configuration, labeled as (CMC/CHI)_x, where x refers to the number of bilayers. For chitosans with different degrees of deacetylation (DD), 3 bilayer films were developed and labeled as (CMC/CHI₉₅)₃, (CMC/CHI₇₅)₃, and (CMC/CHI₅₅)₃ for DDs of 95%, 75%, and 55%, respectively.

Copper solutions were prepared by dissolving pentahydrated copper sulfate in ultrapure water at concentrations of 0.5, 1.0, and 2.0 ppm, with the addition of 0.1 M NaNO₃ and pH maintained to approximately 6.5. Film roughness was assessed using AFM, and amino group availability was evaluated by staining with Rose Bengal dye following the method described in the literature [12]. Copper determination was performed using a

three-electrode cell with anodic stripping voltammetry, involving metal pre-concentration and determination through cyclic voltammetry. A potentiostat controlled the potential sweep from -0.2 V to 0.6 V. Uncoated electrodes were tested to validate the modifications, and repeatability was tested with consecutive measurements at 2 ppm copper concentration. An analytical curve was generated for concentrations from 0.5 to 2.5 ppm.

3. Results

The topography and Root-mean-squared (RMS) values are shown in Fig. 1, revealing that film roughness increases up to 3 bilayers, but decreases from 5 bilayers onward. This result is explained by the island-like formation of polymer films, as reported in the literature [13]. As more layers are incorporated, more polymer molecules are deposited, and the spaces between the islands are filled, leading to a smoother topography, as shown in the 5-bilayer AFM images (Fig. 1D).

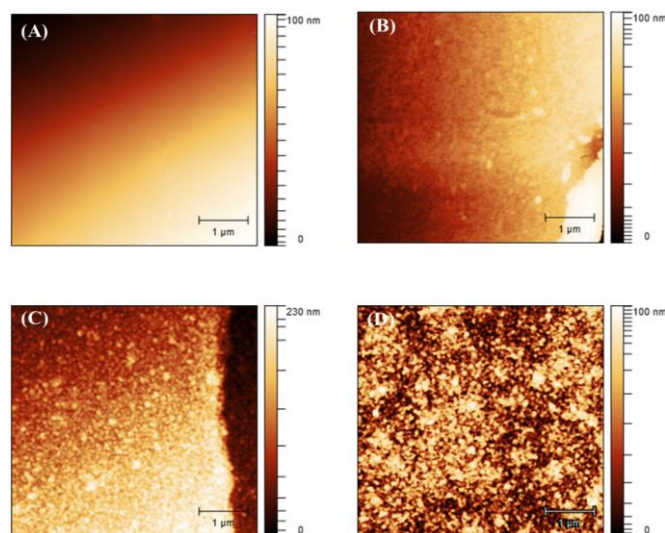


Fig. 1. AFM topography images for uncoated electrodes (A), (CMC/CHI)₁ (B), (CMC/CHI)₃ (C), and (CMC/CHI)₅ (D). RMS values are respectively 9, 16, 28, and 11 nm.

The absorbance spectra [Fig. 2] indicate that increasing the number of bilayers enhances Rose Bengal absorbance, reflecting a higher availability of chitosan amino groups. For chitosans with varying DD, higher deacetylation degrees also resulted in greater absorbance, with (CMC/CHI₉₅)₃ films showing the highest peak, followed by

(CMC/CHI75)₃ and (CMC/CHI55)₃. However, a much larger increase in (CMC/CHI95)₃ peak was noted, indicating a significantly higher availability of amino groups.

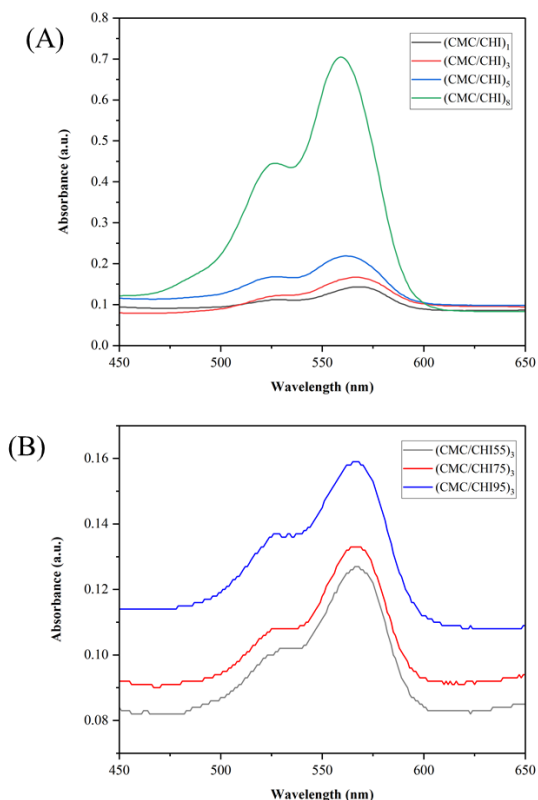


Fig. 2. Rose bengal absorbance spectra for CMC/CHI films with (A) different numbers of bilayers and (B) different average chitosan degrees of deacetylation.

CMC/CHI sensors showed a strong correlation between amino groups, roughness, and copper detection. The 3-bilayer sensor proved optimal, performing best at 2 ppm. Fig. 3 reveals that the (CMC/CHI95)₃ sensor had the highest copper oxidation peak, likely due to its increased amino groups, which enhance protonation and copper adsorption, a phenomenon widely reported in the literature regarding the intrinsic interaction between copper and chitosan molecules in various materials [14]. The (CMC/CHI75)₃ and (CMC/CHI55)₃ electrodes exhibited similar voltammetric responses, with only slight differences in current peaks, reflecting their comparable amino group availability, as indicated by the Rose Bengal absorbance in Fig. 2B.

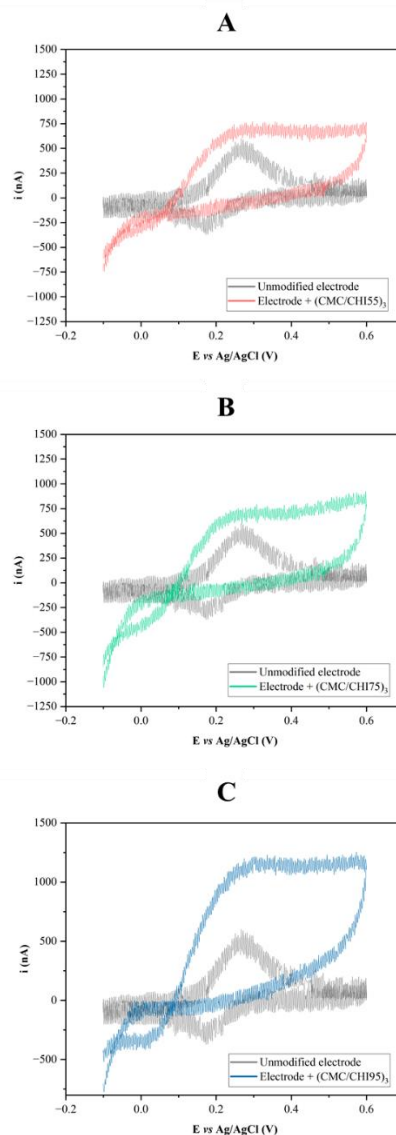


Fig. 3. Voltammograms for copper determination in a 2.0 ppm solution for uncoated electrodes and electrodes modified by (CMC/CHI55)₃ (A), (CMC/CHI75)₃ (B), and (CMC/CHI95)₃ films (C).

The repeatability study [Fig. 4] demonstrate the sensor's stability, allowing for repeated copper detection with minimal variation. The relative standard deviation was about 2%, confirming good repeatability. The analytical curve showed linearity, represented by $i \text{ (nA)} = 69.4 + 220.4[\text{Cu}] \text{ (ppm)}$, with $R^2 = 0.9942$. A detection limit of 0.05 ppm was obtained, demonstrating the sensor's ability to detect copper in low-concentration solutions.

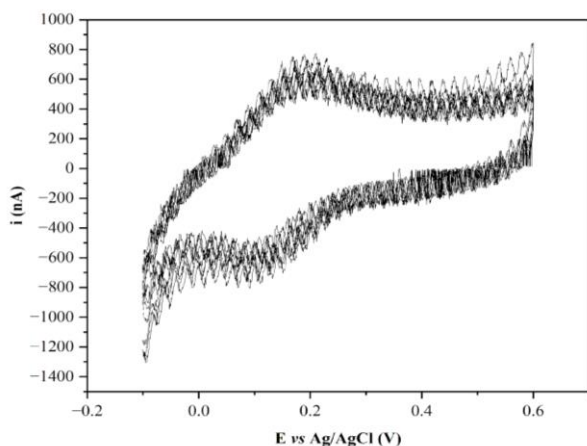


Fig. 4. Voltammograms after 9 consecutive measurements at 2.0 ppm.

4. Conclusion

This study validated the hypothesis that chitosan effectively detects copper ions in water, enhancing sensor sensitivity, particularly with controlled chitosans. Film roughness played a key role in detection, with 3-bilayer films identified as optimal. The sensor demonstrated good reusability, with a detection limit of 0.05 ppm. The performance analysis underscored the strong correlation between amino group availability and copper detection, positioning controlled chitosans as promising materials for improving sensor sensitivity in Cu^{2+} detection.

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