Laser Induced Graphene Sensor with MIP for the Detection of Reactive Black 5 Dye

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Introduction

Water is an essential resource for biological survival, ecological stability, and agricultural sustainability. Beyond its physiological necessity, water serves as a universal solvent across industrial and domestic sectors. However, global water security is severely threatened by human activity, rapid urbanization, and unmanaged industrialization [1]. According to the World Health Organization (WHO), only 25% of the global population currently has access to safely managed drinking water [1]. Industrial effluents frequently discharge hazardous substances into aquatic ecosystems, causing widespread contamination of groundwater, surface water, and subsurface soils [2]. The global water crisis is defined by two major challenges: acute scarcity of potable water and increasing exposure of hydrological and food systems to hazardous synthetic chemicals [3]. Driven by population growth and industrial expansion, global water demand is projected to increase by 55% between 2025 and 2050, leaving over half of the world's population in water-stressed regions [4].

A major contributor to global water contamination is industrial effluent, which contains diverse hazardous pollutants including synthetic dyes, pathogens, heavy metals, pesticides, phenols, polycyclic aromatic hydrocarbons (PAHs), and polychlorinated biphenyls (PCBs) [5]. Among these, synthetic dyes represent one of the most critical environmental threats. The textile manufacturing sector—a major economic engine valued at over US $1 trillion globally and contributing ~2% to global GDP—is the largest source of dye-laden wastewater [6]. Over 10,000 commercial synthetic dyes are manufactured worldwide, with major production and consumption concentrated in industrial hubs such as China [7].

Dyes are structurally diverse chemical compounds designed to impart persistent color to textiles, plastics, leather, and food products . Although natural dyes were historically dominant, synthetic variants are now overwhelmingly preferred due to their superior color fastness, shade diversity, and cost-effectiveness [8,9]. Based on chemical structure and application performance, synthetic dyes are categorized into classes such as azo, triarylmethane, anthraquinone, cationic, acid, vat, and reactive dyes [9]. Most synthetic dyes are toxic, bioaccumulative, and persistent, posing severe ecotoxicological risks and potential carcinogenic effects in human populations [9,10].

Among reactive dyes, Reactive Black 5 (RB5) is widely utilized in textile processing due to its excellent fixation stability and low manufacturing cost. However, RB5 features a complex bis-azo chemical structure that exhibits extreme resistance to natural photolytic, chemical, and microbial degradation under ambient conditions [11,12]. Consequently, un-treated RB5 discharged into natural bodies of water persists indefinitely, bioaccumulating in aquatic food chains and disrupting photosynthetic biological processes.

Monitoring and quantifying emerging pollutants like RB5 requires robust analytical methodologies. Conventional techniques such as UV-Vis spectrophotometry and High-Performance Liquid Chromatography (HPLC) [13] offer precision but are hampered by high operational costs, complex sample preparation, bulky instrumentation, and unsuitable profiles for real-time on-site detection. In contrast, electrochemical analysis provides an exceptional alternative characterized by high sensitivity, rapid response, low instrumentation cost, portability, and minimal sample volume requirements [14]. To enhance selectivity and sensitivity, nanomaterials and advanced matrices—such as carbon nanotubes, nanofibers, MXenes, and molecularly imprinted polymers (MIPs)—are increasingly integrated into electrochemical sensor platforms [15].

Molecularly Imprinted Polymers (MIPs) function as artificial antibody-like receptors. Synthetic polymers are synthesized in the presence of a target template molecule, creating rigid, custom-tailored binding cavities that match the shape, size, and functional chemical groups of the target analyte [16]. Molecular imprinted polymer was synthesized following the protocol [16] with some adaptations. MIP-based electrochemical sensors offer exceptional chemical and mechanical stability, superior target selectivity, low limits of detection (LOD), reusability, and straightforward preparation.

Despite significant progress in dye sensor development, no electrochemical sensor modified with a Molecularly Imprinted Polymer (MIP) has yet been reported for the selective detection of Reactive Black 5 dye. This study addresses this gap by developing a low-cost, high-selectivity MIP-modified electrochemical sensor paired with Non-Imprinted Polymer (NIP) control studies. The

proposed platform offers a promising route for environmental monitoring and rapid screening of RB5 in industrial wastewater effluents.

Materials and methods

A three-electrode architecture was used to fabricate the electrochemical chip which consisted of a working electrode (WE), counter electrode (CE) and reference electrode (RE). The layout of the electrode was designed using Xtool S1 software. Direct laser writing (LDW) process was employed on a polyimide film (Kapton), which was fixed to a support plate and processed utilizing a diode laser system (Xtool S1) (Figure 1). The pyrolysis of the polymer was carried out under controlled conditions to preserve the conductivity and geometry of the electrode. The process was carried out under controlled parameters: a temperature of 2500 °C, a scan speed of 100 mm/s, a laser power 1.8 W, and an engraving density of 200 lines per centimeter. The precise manufacturing of sensor architecture was enabled by following this protocol from Gonçalves (2026) [14].

The modified Laser Induced-Graphene (LIG) electrodes were prepared via a drop-casting protocol. First, a homogeneous dispersion was prepared by suspending 1.0 mg of Molecularly Imprinted Polymer (MIP) obtained by bulk methodology (figure 2) in 1.0 mL of isopropyl alcohol, followed by ultrasonication for 3 minutes to prevent particle agglomeration. To functionalize the sensing surface, 20 µL of the prepared MIP suspension was drop-casted onto the working electrode surface of the SPE. The modified electrodes were allowed to dry at room temperature for 4 hours to form a stable, adherent polymer film. To optimize electrical contact between the electrode pads and the potentiostat edge connector, a conductive silver ink was applied to the contact areas. Finally, a hydrophobic hot-glue barrier was deposited around the electroactive sensing zone to precisely define the cell area and contain the droplet volume during subsequent electrochemical measurements.

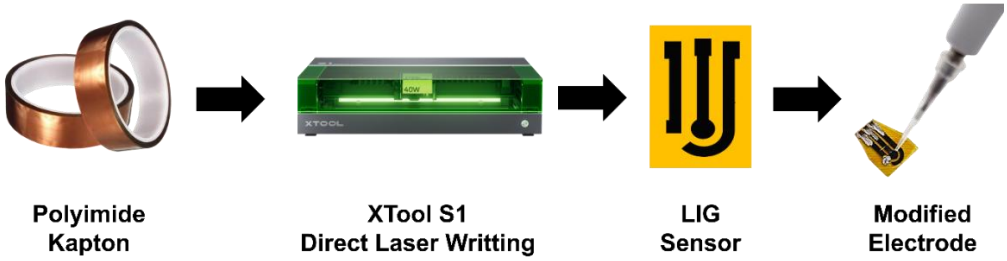

***Figure 1**. Sensor fabrication and modification.*

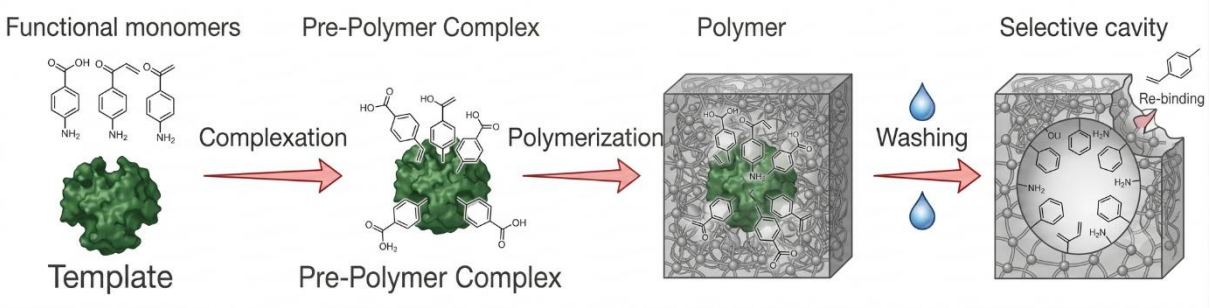

Figure 2. Synthesis of molecular imprinted polymer

Results and discussions

Morphological Characterization (SEM Analysis)

The surface morphologies of the synthesized molecularly imprinted polymer (MIP) and non-imprinted polymer (NIP) were characterized using Scanning Electron Microscopy (SEM) to evaluate structural differences arising from the imprinting process as shown in figure 2. SEM micro-graphs revealed distinct morphological features between the two polymer matrices. MIP surface exhibited a highly porous, irregular morphology characterized by well-defined micro-cavities distributed homogeneously throughout the polymer framework. These specific cavities correspond to the spatial and chemical imprints generated during polymerization using Reactive Black 5 (RB5) as the template molecule. Subsequent extraction of the template created accessible, shape-selective recognition sites tailored for targeted rebinding.

NIP surface synthesized under identical conditions in the absence of the RB5 template, the NIP demonstrated a comparatively smooth, compact, and non-porous surface structure lacking specific recognition cavities. This pronounced morphological contrast confirms the successful creation of imprinted binding sites within the MIP network, providing the high specific surface area and selectivity required for the targeted electrochemical detection of Reactive Black 5.

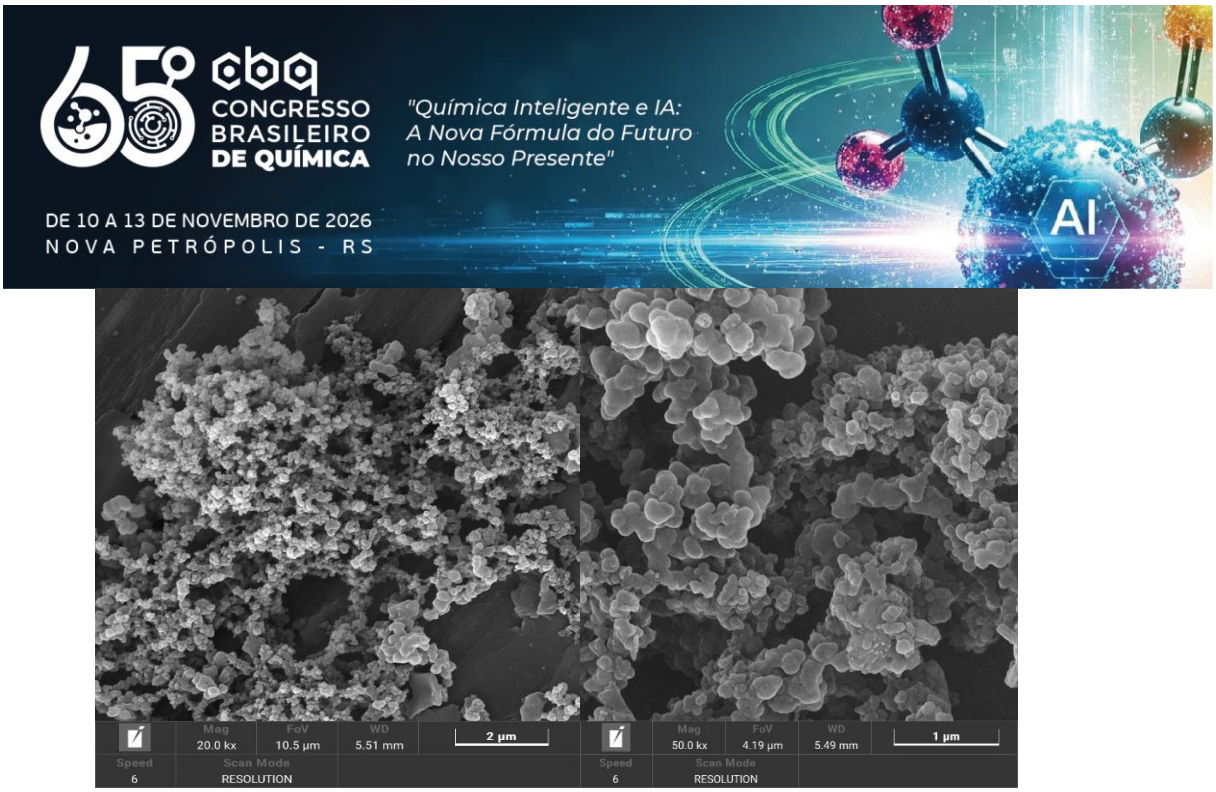

***Figure 3**. SEM images a) Molecular imprinted polymers. b) Non-imprinted polymers*

Electrochemical behavior of Reactive Black 5 dye

The electrochemical behavior of Reactive Black 5 (RB5) was investigated using cyclic voltammetry (CV) as shown in figure 3. At an analyte concentration of 3.53×10^{-5} mol L^{-1}, RB5 exhibited a well-defined anodic oxidation peak. On the Molecularly Imprinted Polymer (MIP)-modified sensor, a prominent peak was observed at 0.47V, corresponding to the electrochemical oxidation of the target dye molecules bound within the imprinted cavities. A comparative evaluation among the unmodified laser-induced graphene (LIG) sensor, the Non-Imprinted Polymer (NIP)-modified control, and the MIP-modified sensor demonstrated a distinct trend in current response. Unmodified LIG exhibited a baseline capacitive response with minimal signal enhancement. NIP-LIG sensor displayed a slight current increase due to non-specific surface adsorption. MIP-LIG sensor produced the highest peak current density, confirming that the high concentration of pre-formed recognition cavities significantly enhances mass transport and selective pre-concentration of RB5 at the electrode interface. The comparison is shown in figure 3.

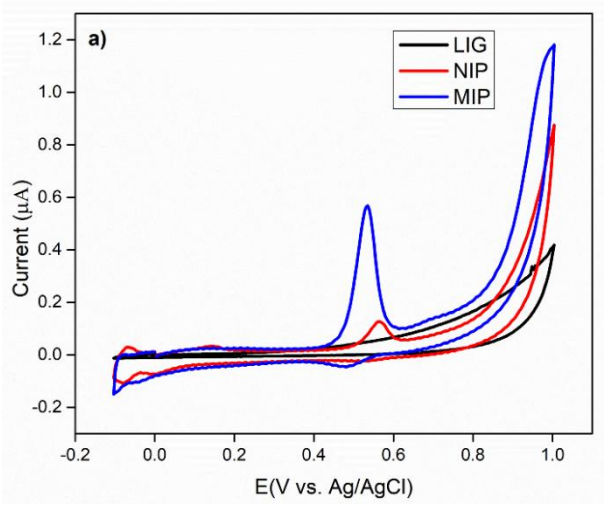

Figure 4. *Cyclic voltammogram comparing bare LIG, LIG/NIP and LIG/MIP.*

Conclusions

In this work, a novel, cost-effective electrochemical sensing platform was successfully developed by combining laser-induced graphene (LIG) substrate technology with a Reactive Black 5 (RB5)-templated molecularly imprinted polymer (MIP). The MIP/LIG modified sensor demonstrated superior electrochemical performance, high sensitivity, and excellent selectivity toward RB5 across a broad linear concentration range, accompanied by strong operational stability and signal reproducibility. Practical utility was further validated through the successful detection and quantification of RB5 in complex real environmental wastewater matrices.

By eliminating the need for tedious sample preparation, expensive infrastructure, and time-consuming workflows typical of conventional chromatographic and spectroscopic methods, this LIG–MIP platform offers a rapid, reliable, and scalable alternative. Overall, these findings highlight the strong potential of the developed sensor for real-time environmental monitoring, industrial effluent control, and on-site water quality management.

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