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This book covers key areas of Chemistry and Biochemistry. The contributions by the authors include Titanium dioxide, cold spray, high velocity oxy-fuel spray, anodization, high velocity suspension flame spraying, pulsed laser deposition, zinc oxide nanoparticles doped with chlorine, crystallinity, anti-diabetic activity, , amylase inhibitors, electrode polarization, logarithmic derivatives and differences, matching Debye kernels, multivariate apart-together fitting, spectral resolution, symbolic differential operators, Synthesis, benzothiazole derivatives, heteroaromatic structures, heterocyclic molecules, Hexavalent chromium, hypoglycaemia, glycogenolysis, citric acid cycle, and electron transport chain. This book contains various materials suitable for students, researchers and academicians in the field of Chemistry and Biochemistry.

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<u>A Comparative Study on Amperometric Detection of Urea by Polyaniline and Poly (Oanisidine) Film under Galvanostatic Method</u>

Kiran Paithankar, Priyanka Choudhari, Ritesh Yadav , Suresh More, Dattatraya Galhe, Vikas Gade

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A Comparative Study on Amperometric Detection of Urea by Polyaniline and Poly (O-anisidine) Film under Galvanostatic Method

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ABSTRACT

The growth mechanism of polyaniline (PANI) and poly (O-anisidine) (POA) on an indium tin oxide (ITO) substrate with the supporting electrolyte HCIO4 has been explored. Polyaniline being ecological steady, many scientists covered thing leading polymer by intriguing on the redox properties related with the chain of nitrogen particles. The PANI and POA polymer film that was formed on the indium tin oxide substrate was created using the electrochemical polymerization process under galvanostatic conditions in an aqueous solution with the supporting electrolyte HCIO4 at a temperature of 27^oC. Analytical methods like UV-visible, FTIR, and FE-SEM studies were used to characterise the organised materials in order to compare the amperometric response of PANI and POA film on aurease enzyme in traditional sensor. The performance of developed sensor was evaluated and the obtained urea biosensor exhibited shorter response time (3 s), wider range 1 x 10⁻⁹ to 9 x 10⁻⁹ M and the detection limit found to be 1 x 10⁻⁹ M. About 80 % of enzyme activity is retained for about 40 days. A modified sensor performs better with polyaniline than one with poly (O-anisidine).

Keywords: Polyaniline; poly (o-anisidine); galvanostatic.

1. INTRODUCTION

There have been a lot of novel materials developed recently for a few creatively organised uses. One of the most promising new material classes being used in a few applications is the directed polymer [1]. Intriguing properties like electrochemical redox conduct and electro chromic reactant exercises are demonstrated by the unique type of leading electrodynamic natural polymer known

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as polyaniline [2]. Polyaniline being ecological steady, many scientists covered thing leading polymer by intri-guing on the redox properties related with the chain of nitrogen particles. Two types of aromatic polymers including polyaniline (PANI) and polypyrrole (PPy) are used to provide nitrogen atoms and control the morphology. PANI and PPy are both widely used conductive polymers. Their morphology can be controlled with different synthesis methods [3-5].

Polyaniline is one of the leading polymers, which can be utilized to improve the speed, affectability and selectivity of biosensors. Polyaniline have pulled in more enthusiasm as an appropriate network for ensnarement of proteins [6]. It is being utilized both as an immobilization lattice and as a physico-chemical transducers to changes over a compound flag to electrical flag.

In the present work, we report an execution of polyaniline (PANI) and poly(Oanisidine) (POA) film made by galvano- static electrochemical polymerization method strategy which is to be a decent adsorbent for analyte, for distinguishing of urea in research center examples utilizing amperometric procedure with expansion of urease chemical. This PANI and POA cathode by estimating amperometric reaction caused by immobilized urease response framework. The electrochemical cell was amassed in a customary one compartment three termi- nal framework plan, the working cathode was adjusted PANI and POA film comprise dropping a urease enayme permitted to homogenize for couple of hours [7]. The Ag/AgCl utilized as reference cathode and a graphite use as the counter terminal. The unrivaled execution of adjusted terminal (PANI/POA) is exhibited by the speciation and recognition of urea in pharma-ceutical species, pee test, ocean water tests [8]. The proposed amperometric technique has been approved by utilizing induc-tively coupled plasma-nuclear emanation spectrometry (ICP-AES) [9].

2. EXPERIMENTAL

Urea (99 %), urease, aniline (99.5 %) and poly (O-anisi- dine) (98 %), perchoric acid (70 %), ethanol (99.9 %), The working electrodes was indium tin oxide (ITO) was purchased from Sigma Aldrich, graphite fine powder extra pure (particle size 240×10^{-6} m) obtained from Lobachemie Pvt. Ltd. India, Paraffin liquid heavy or mineral oil (viscosity at 37 °C is 64 cS) purchased from High purity lab, Mumbai, India. UV-visible spectra were recorded in air at room temperature in the wavelength range of 200-800 nm using a Jena specord 210 spectrophotometer. FT-IR spectra were recorded on a Ocean optics HPX-2000 (Fiber coupled) spectrometer in the range of 4000-500 cm⁻¹. FE-SEM carried by JEOL JSM-7500F. Ampero- metric response characteristics were studied with a 4^{1/2} Digit True RMS Multimeter (MODEL 1085).

2.1 Electrochemical Synthesis of Polymer Film (PANI and POA)

The polymers was double distilled before use. Perchloric acid was used by supporting electrolyte. The thin films were synthesized by electrochemical

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polymerization of aniline and O-anisidine on ITO substrate under potentiostatic conditions at room temperature using in electrochemical polymerization system. The electrolyte was prepared using deionized water at room temperature. The experiment was performed in a system by containing 0.2 M of polymer (polyaniline & poly O-anisi- dine) and 0.4 M HCIO4. The potential 1.4 V is constant for the whole experiment. The pH for aniline is 2 and for O-anisidine is 1 kept constant. The time for this films preparation is 20 min are kept constant. The synthetic films (PANI and POA) usedfor detection of urea by amperometric method.

3. RESULTS AND DISCUSSION

3.1 SEM

Scanning electron microscopy of the synthesized polymers (polyaniline & poly(Oanisidine) films with opti- mized process parameters for $HCIO_4$ are shown in Fig. 1. Scan- ning electron micrographs were recorded using JEOL JSM- 6360 A analytical SEM. It shows very good uniformity and porosity. It can be seen that the surface morphology is more porous and uniform like structure [10-12].

3.2 UV-visible Analysis

The optical absorption spectrum of synthesized PANI and POA films with optimized parameters for $HCIO_4$ is shown in Fig. 2. It was recorded in the wavelength range of 200-800 nm using UV-visible spectrophotometer Jasco spectrometer (V-670). All the spectra were recorded in the wavelength range of 200-800 nm and the peak is appearing at 667 nm for PANI and the peak appeared at 674 nm for POA, which shows a very good resemblance with earlier work [12].

3.3 FTIR Analysis

The FTIR spectroscopy was utilized for auxiliary portrayal of incorporated polvaniline and POA films Fig. 3a shows the sub-atomic structure of PANI incorporated examples in the 4000-400 cm⁻¹ region by FTIR spectra. The bands 1463 and 1597 cm⁻¹ compares to C=C extending vibrations of benzenoid and quinonoid rings individually [9]. The crest at 1259 cm⁻¹ is the trademark band of fragrant C-N extending vibration and a powerless crest at 3462 cm⁻¹ is allocated to extending method of N-H [11]. The assimilation band shows up at 1101 cm⁻¹ is observed as vibration band of nitrogen quinine (N=Q=N). Likewise, the band at 860 cm⁻¹ can be attributed to C-H out of plane twisting vibrations for the ring which obviously underpins the arrangement of PANI. The polymer demonstrates the ingestion groups at 2968 cm⁻¹ are because of lopsided C-H extending and symmetric C-H extending vibra- tions [11]. These groups relates to the attributes groups of aniline. Fig. 3b demonstrates the atomic structure of POA integrated examples in the 4000-400 cm⁻¹ region by FTIR spectra. The bands 1689 cm⁻¹ compares due to presence of carbonyl group [1] and extending vibrations of guinonoid ring. The band at 1438 cm⁻¹ relates to C-O and the extending vibrations of benze- noid ring. The assimilation band shows up at 929 cm⁻¹ compares to O-C=O

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in the plane disfigurement. The bands at 626 and 1259 cm⁻¹ is due to C-C-O in plane twisting. The band region 3631-3446 cm⁻¹ relates to N-H, while the band at 1489 cm⁻¹ is due to CH3 gathering. The band at 1489 cm⁻¹ might be because of CH3 gathering or because of aromaticity [7]. The C-H extending vibrations relate to 2945 cm⁻¹ bonds.

3.4 XRD Analysis

The pattern shows sharp and well define peaks which indicates crystalline nature of both polymers (PANI and POA) (Fig. 4). The plains of benzenoid and quinonoid rings of polymers are responsible for crystalline nature. The inter planer distance and crystalline size are estimated by Bragg SLaw and Debye Scherrer equation:

 $\mathsf{D} = (0.8 \times \lambda) / \beta \cos \theta$

where, β is (FWHM) full width half maxima.

3.5 Current Response

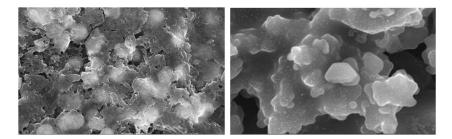
Fig. 5 demonstrates the present reaction for different convergence of urea. Fig. 5a demonstrates ampero-metric reaction for PANI and Fig. 5b indicates reaction for POA. At point when the capability of compound anode was set at 0.5 V is as appeared in Fig. 5. It was noted that reaction current of chemical terminal effectively reaches to consistent state. The connection between reaction current and urea fixation in 0.1 M phosphate support pH 7 is appeared. It was discovered that present increments with expanding urea focus in the scope of 0.1×10^{-9} to 1.1×10^{-9} M amperometric reaction of PANI (an) is bigger than POA (b) cathode. In the present case, accepting that the catalyst is consistently conveyed all through the terminal, the response happens generally on the surface of anode in the minor fixation, at the point when urea is not part in response. Notwithstanding, the response on the surface of cathode and dissemination happened at the same time at higher focuses defers the reaction time.

Fig. 6 demonstrates the relentless state potential reliance alignment bend for every individual urea fixation. In Fig. 6a, the reaction of PANI to urea was observed to be wide direct scope of 2×10^{-9} to 8×10^{-9} M and for Fig. 6b POA it turn out to be short 3×10^{-9} to 6×10^{-9} M. This linearity go is in well similarity with that got in the amperometric reaction of sensoris legitimate in extent to urea focus.

3.6 Storage Stability

Long term stability is one of the most important features required for the satisfactory application of a biosensor. In order to evaluate the storage stability, both sensors were tested for 40 days of storage in 0.1 M phosphate buffer pH 7 at 20°C. There is a slight decrease in sensitivity of PANI sensor of about 10 % from the initial value (Fig. 7), revealed a good preservation of bioactivity of sensor in comparison to POA [9].

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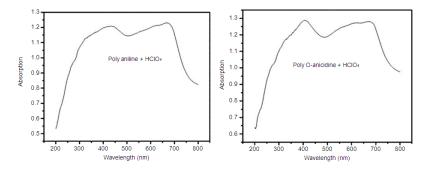


Fig. 2. UV-visible study of (a) PANI (b) POA

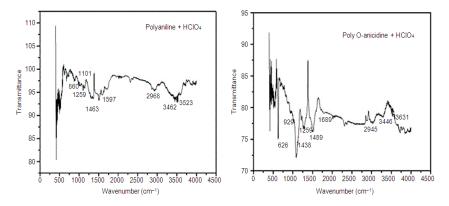
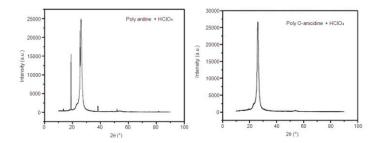
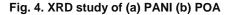


Fig. 3. FTIR study of (a) PANI (b) POA

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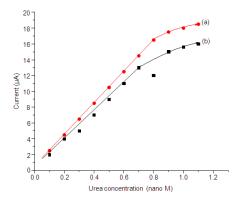


Fig. 5. (a-b) Current–concentration curve a) PANI (b) POA at 0.7 V and pH 7 in 0.1 M PBS for different urea solution of 0.1 × 10-9 to 1.1 × 10-9 M

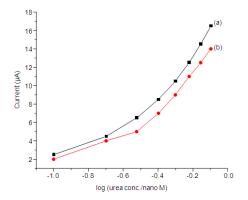


Fig. 6. (a-b) Steady-state potential dependence calibration curve of biosensor (a) PANI (b) POA

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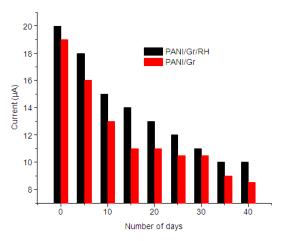


Fig. 7. Stability of the a) PANI (b) POA electrode on storage in 0.1 M PBS (pH 7) for 40 days

4. CONCLUSION

Polyaniline (PANI) and poly(O-anisidine) (POA) films have been developed and utilized to detect the presence of urea. An identification point of confinement of 0.1×10^{-9} M for urea was accomplished with the utilization of PANI. This strategy gives advantages such favourable circumstances as high affectability, low location restrict, simple taking care of opposition against surface fouling and minimal effort. There- fore, this strategy is suggested for the examinations of phosphate, antimony, glucose, creatinine in clinical and in addition quality control research facilities.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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Special Award: He received RULA AWARD.

Any other Remarkable Point(s): He developed BIOSENSOR.

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