

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

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The growth mechanism of polyaniline (PANI) and poly(O-anisidine) (POA) with supporting electrolyte HClO<sub>4</sub> on indium tin oxide (ITO) substrate has been investigated. The polymer film of PANI and POA deposited on indium tin oxide substrate have been synthesized by electrochemical polymerization method under galvanostatic condition in aqueous solution with supporting electrolyte HClO<sub>4</sub> at 27 °C. To compare the amperometric response of PANI and POA film on urease enzyme in conventional sensor, the organized materials were characterized by analytical techniques such as UV-visible, FTIR and FE-SEM analyses. The performance of developed sensor was evaluated and the obtained urea biosensor exhibited shorter response time (3 s), wider range  $1 \times 10^{-9}$  to  $9 \times 10^{-9}$  M and the detection limit found to be  $1 \times 10^{-9}$  M. About 80 % of enzyme activity is retained for about 40 days. Modified sensor gives better result by polyaniline than poly(O-anisidine) sensor.

**Keywords:** Polyaniline, Poly(O-anisidine), Galvanostatic.

### INTRODUCTION

In the prior days, there are large numbers of new materials coming up for a few innovative arranged applications. The directing polymer is a standout amongst the most encouraging new classes of materials being utilized for a few applications [1]. Polyaniline is a unique sort of leading electrodynamic natural polymer which demonstrates a many intriguing properties, for example, electrochemical redox conduct, electrochromic reactant exercises [2]. Polyaniline being ecological steady, many scientists covered thing leading polymer by intriguing on the redox properties related with the chain of nitrogen particles.

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 [3]. 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(O-anisidine) (POA) film made by galvanostatic 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 terminal framework plan, the working cathode was adjusted PANI and POA film comprise dropping a urease enzyme permitted to homogenize for couple of hours [4]. 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 pharmaceutical species, pee test, ocean water tests [5]. The proposed amperometric technique has been approved by utilizing inductively coupled plasma-nuclear emanation spectrometry (ICP-AES) [6].

### EXPERIMENTAL

Urea (99 %), urease, aniline (99.5 %) and poly(O-anisidine) (98 %), perchloric 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  $240 \times 10^{-6}$  m) obtained from Lobachemie Pvt. Ltd. India,

Paraffin liquid heavy or mineral oil (viscosity at  $37^\circ\text{C}$  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  $4000\text{-}500\text{ cm}^{-1}$ . FE-SEM carried by JEOL JSM-7500F. Amperometric response characteristics were studied with a  $4^{1/2}$  Digit True RMS Multimeter (MODEL 1085).

**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 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-anisidine) and 0.4 M  $\text{HClO}_4$ . 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) used for detection of urea by amperometric method.

## RESULTS AND DISCUSSION

**SEM:** Scanning electron microscopy of the synthesized polymers (polyaniline & poly(O-anisidine) films with optimized process parameters for  $\text{HClO}_4$  are shown in Fig. 1. Scanning 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 [7-9].

**UV-visible analysis:** The optical absorption spectrum of synthesized PANI and POA films with optimized parameters for  $\text{HClO}_4$  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 [9].

**FTIR analysis:** The FTIR spectroscopy was utilized for auxiliary portrayal of incorporated polyaniline and POA films Fig. 3a shows the sub-atomic structure of PANI incorporated examples in the  $4000\text{-}400\text{ cm}^{-1}$  region by FTIR spectra. The bands  $1463$  and  $1597\text{ cm}^{-1}$  compares to  $\text{C}=\text{C}$  extending vibrations of benzenoid and quinonoid rings individually [6]. The crest at  $1259\text{ cm}^{-1}$  is the trademark band of fragrant  $\text{C}-\text{N}$  extending vibration and a powerless crest at  $3462\text{ cm}^{-1}$  is allocated to extending method of  $\text{N}-\text{H}$  [8]. The assimilation band shows

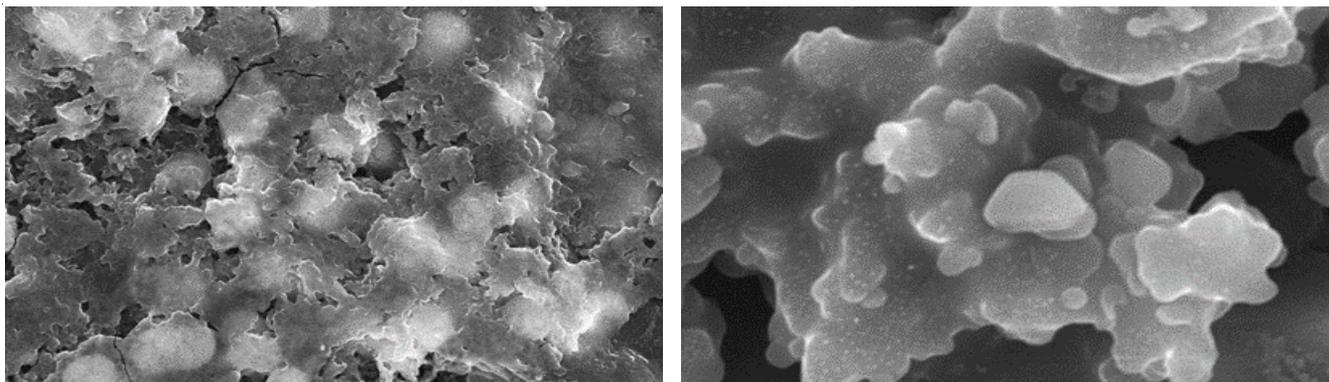


Fig. 1. SEM of (a) PANI and (b) POA (Mag.25KX)

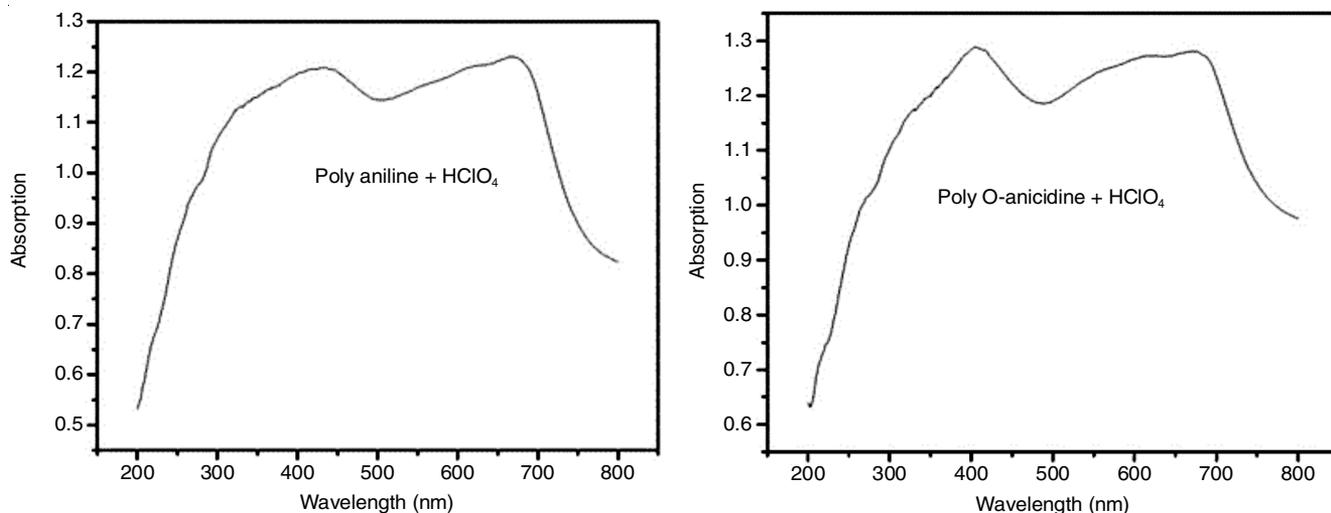


Fig. 2. UV-visible study of (a) PANI (b) POA

up at  $1101\text{ cm}^{-1}$  is observed as vibration band of nitrogen quinine ( $\text{N}=\text{Q}=\text{N}$ ). Likewise, the band at  $860\text{ cm}^{-1}$  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  $2968\text{ cm}^{-1}$  are because of lopsided C-H extending and symmetric C-H extending vibrations [8]. These groups relates to the attributes groups of aniline.

Fig 3b demonstrates the atomic structure of POA integrated examples in the  $4000\text{-}400\text{ cm}^{-1}$  region by FTIR spectra. The bands  $1689\text{ cm}^{-1}$  compares due to presence of carbonyl group [1] and extending vibrations of quinonoid ring. The band at  $1438\text{ cm}^{-1}$  relates to C-O and the extending vibrations of benzenoid ring. The assimilation band shows up at  $929\text{ cm}^{-1}$  compares to O-C=O in the plane disfigurement. The bands at  $626$  and  $1259\text{ cm}^{-1}$  is due to C-C-O in plane twisting. The band region  $3631\text{-}3446\text{ cm}^{-1}$  relates to N-H, while the band at  $1489\text{ cm}^{-1}$  is due to  $\text{CH}_3$  gathering. The band at  $1489\text{ cm}^{-1}$  might be because of  $\text{CH}_3$  gathering or because of aromaticity [4]. The C-H extending vibrations relate to  $2945\text{ cm}^{-1}$  bonds.

**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's Law and Debye Scherrer equation:

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

where,  $\beta$  is (FWHM) full width half maxima.

**Current response:** Fig.5 demonstrates the present reaction for different convergence of urea. Fig. 5a demonstrates amperometric reaction for PANI and Fig.5b indicates reaction for POA. At point when the capability of compound anode was set at  $0.5\text{ V}$  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  $0.1\text{ M}$  phosphate support pH 7 is appeared. It was discovered that present increments with expanding urea focus in the scope of  $0.1 \times 10^{-9}$  to  $1.1 \times 10^{-9}\text{ M}$  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.

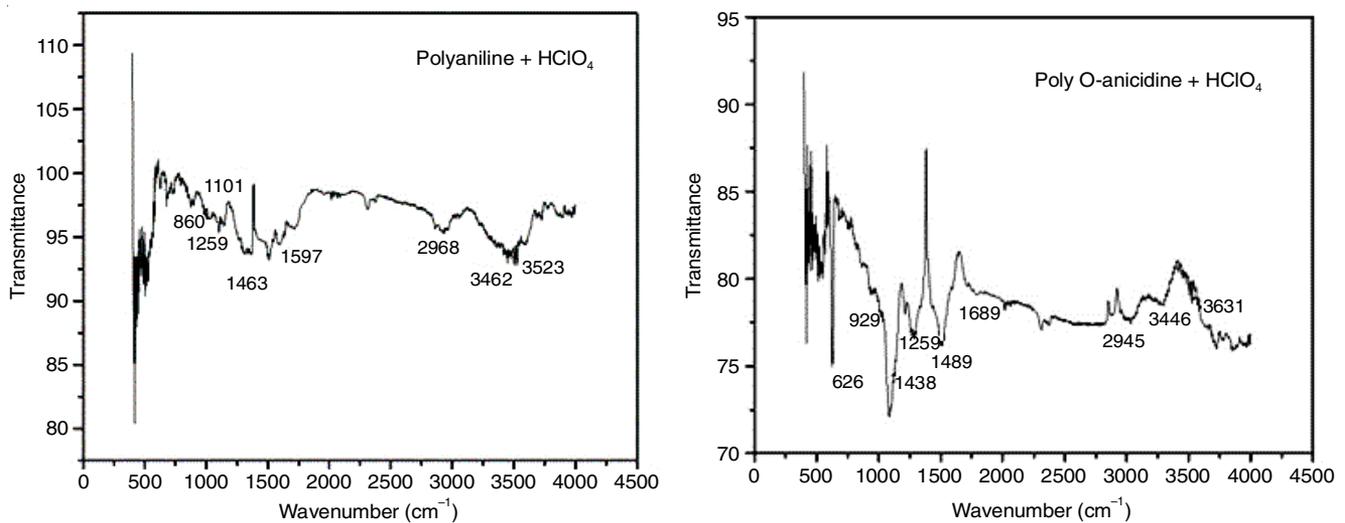


Fig. 3. FTIR study of (a) PANI (b) POA

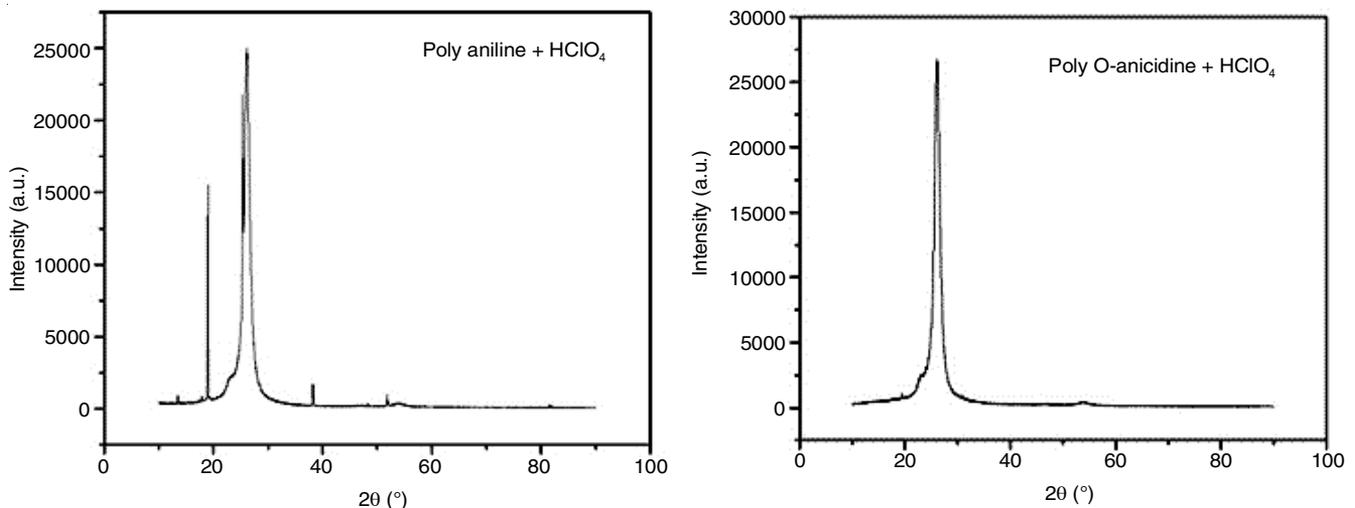


Fig. 4 XRD study of (a) PANI (b) POA

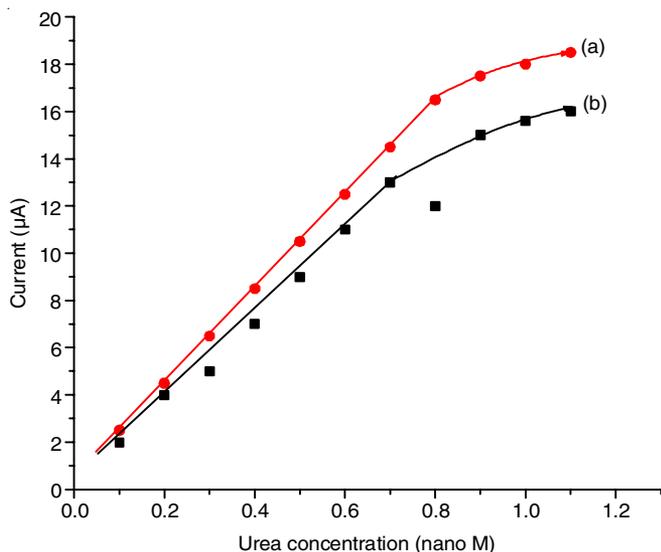


Fig. 5. (a-b) Current-concentration curve a) PANI (b) POA at 0.7 V and pH 7 in 0.1 M PBS for different urea solution of  $0.1 \times 10^{-9}$  to  $1.1 \times 10^{-9}$  M

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  $2 \times 10^{-9}$  to  $8 \times 10^{-9}$  M and for Fig. 6b POA it turns out to be short  $3 \times 10^{-9}$  to  $6 \times 10^{-9}$  M. This linearity goes in well similarity with that got in the amperometric reaction of sensor is legitimate in extent to urea focus.

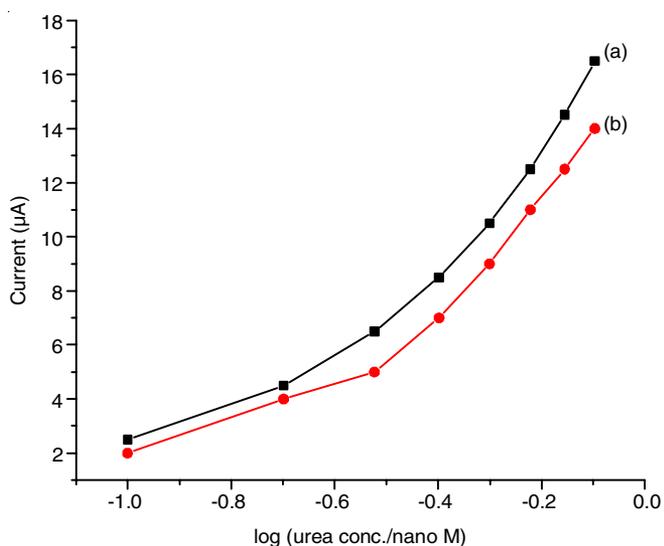


Fig. 6. (a-b) Steady-state potential dependence calibration curve of biosensor (a) PANI (b) POA

**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].

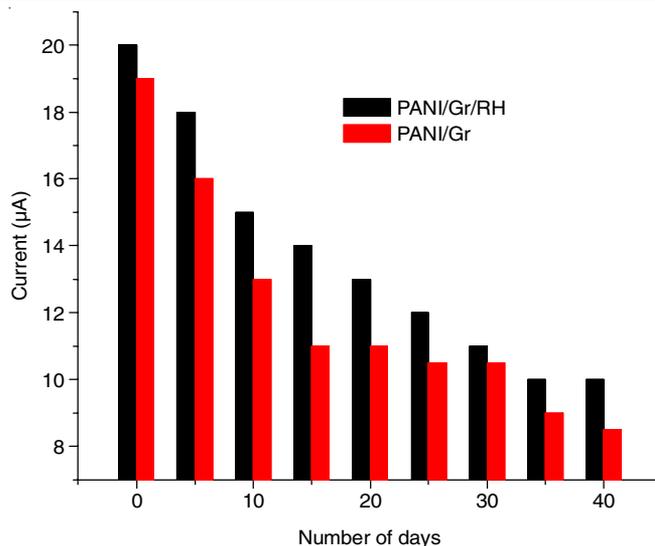


Fig. 7. Stability of the a) PANI (b) POA electrode on storage in 0.1 M PBS (pH 7) for 40 days

## 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  $0.1 \times 10^{-9}$  M for urea was accomplished with the utilization of PANI. This strategy gives advantages such as favourable circumstances as high affectability, low location restrict, simple taking care of opposition against surface fouling and minimal effort. Therefore, this strategy is suggested for the examinations of phosphate, antimony, glucose, creatinine in clinical and in addition quality control research facilities.

## CONFLICT OF INTEREST

The authors declare that there is no conflict of interests regarding the publication of this article.

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