Vitamin K₃ encapsulated poly(vinylpyrrolidone) film modified glassy carbon electrode for studying the vitamin K₃ – caffeine interaction

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The cathodic and anodic peak currents in cyclic voltammogram of vitamin K₃ encapsulated polyvinyl pyrrolidone film modified glassy carbon electrode decrease with addition of caffeine (from 0.02 μM to 0.2 μM) in the electrolytic solution. Caffeine converts vitamin K₃ into diphenylenetetranol, which is electrochemically inactive. This interaction between vitamin K₃ and caffeine is not observed when both are in solution.

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Molecular recognition has become a prominent field in modern chemistry. A change in a property of the probe molecule observable by various instrumental techniques upon interaction with the target molecule is the basic principle for molecular recognition. H-bonding is well exploited as a mean of interaction between probe and the target. Study of the change in redox potential and redox currents of the probe on its interaction with the target is well studied for qualitative and quantitative detection of the target.

Caffeine is an alkaloid present in tea, coffee, kola nuts, yerba mate and guarana berries. It stimulates central nervous system, cardiac muscle, respiratory system and delay fatigue. Caffeine can interfere with adenosine, which is a neurotransmitter present in brain. There are only a few reports of electrochemical methods for the detection of caffeine.

Vitamin K₃ (2-methyl-1,4-naphthoquinone) is a synthetic vitamin K analogue that plays important role in blood coagulation as an antihemorrhagic agent, in bone mineralization and has gained attention as anti carcinogens. Vitamin K₃ shows well defined cyclic voltammograms due to the reversible conversion of the two ketone groups into hydroxyls.

We show herein that caffeine inactivates electrochemical property of vitamin K₃ while the later is inside film. This electrochemical inactivation of vitamin K₃ by caffeine has been explored to determine caffeine in micromolar concentration. Determination of caffeine in tablet form in presence of vitamin D₃ and Ca²⁺ ion is also reported.

Experimental

All the chemicals were procured from Loba Chemie and used without any further purification. Double distilled water (prepared using Rieviera quartz double distillation apparatus) was used for all electrochemical experiments. CHI 600B Electrochemical Analyzer (USA) with a three-electrode cell assembly was used for electrochemical studies. Electrochemical experiments were carried out under a blanket of nitrogen gas after passing the gas through the solution for 10 min. Glassy carbon (GC) disc working electrode, Ag-AgCl (3 M NaCl) reference electrode and sodium nitrate (0.1 M) supporting electrolyte were used during the studies. In the Osteryoung Square Wave Voltammetry (OSWV) experiments, the square wave amplitude was 25 mV, the frequency 15 Hz and the potential height for base stair case wave front 4 mV. The working electrode was cleaned by polishing with 0.1 micron alumina slurry using a polishing kit (CHI) followed by sonication in distilled water for 5 min.

Vitamin K₃ incorporated polyvinylpyrrolidone (PVP) film on GC electrode is prepared by dissolving 0.02 g PVP and 0.2 g vitamin K₃ in 10 mL chloroform. 20 μL of the solution is then placed on the tip of the pre cleaned GC electrode surface using Hamilton micro syringe. Chloroform is allowed to evaporate (in dark) resulting in the formation of vitamin K₃ encapsulated PVP film. Cyclic voltammograms are recorded by dipping the tip of
this modified electrode in 0.1 M Tris buffer solution at pH 7.0 and containing 0.1 M sodium nitrate.

Results and discussion

Cyclic voltammetric response of the vitamin K$_3$ incorporated PVP film modified GC electrode in the tris buffer (pH 7.0) shows quasi-reversible behaviour with redox potential value --0.220 V, peak separation 0.136 V at scan rate 0.100 Vsec$^{-1}$. The redox potential is also confirmed by OSWV measurements. Chrono coulometry measurements show the charge to decrease sharply against time indicating adsorption.

An approximate estimation of surface coverage of the electrode is made by using the method proposed by Sharp et al.\textsuperscript{17}. In this method, the relation between peak current ($I_p$) and surface coverage ($\tau$) is given by:

$$I_p = n^2 F^2 A \frac{\nu \tau}{4RT}$$

where $n$ is the number of electrons involved in the reaction which is 2 here, $A$ is the geometric area of the electrode (0.0314 cm$^2$), $\tau$ (mol cm$^{-2}$) is the surface coverage and $\nu$ is the scan rate. At scan rate 0.1 Vsec$^{-1}$, the surface coverage is calculated to be $15.8 \times 10^{-10}$ mol cm$^{-2}$.

The cyclic voltammetric response of vitamin K$_3$ incorporated PVP film modified GC electrode at different caffeine concentration in the electrolytic solution is shown in Fig. 1. Both the cathodic and anodic peak currents decrease as caffeine concentration is changed from 0.02 $\mu$M to 0.2 $\mu$M and remains same thereafter. No change is however observed in peak potentials. The decrease only in peak currents and not in peak potentials indicates formation of electroinactive product due to interaction between vitamin K$_3$ and caffeine.

When the cyclic voltammogram of 1 mM vitamin K$_3$ in 10% (v/v, Tris buffer: methanol) solution is recorded at different added concentration of caffeine, change neither in peak currents nor in peak potentials is observed. Thus, the interaction between vitamin K$_3$ and caffeine leading to electroinactive product is possible only when the former is inside film.

Scheme 1 shows the oxidation of caffeine by electrode releasing two electrons and two protons as reported elsewhere\textsuperscript{9}. Vitamin K$_3$, in absence of caffeine, undergoes reversible redox process through intermediate X as shown in Scheme 2. Caffeine is oxidized by X forming the intermediate Y and two protons are released to the solvent. Y dimerises irreversibly in presence of light\textsuperscript{15} and subsequent protonation leads to the formation of diphenylenetetranol.

Analytical applications

The vitamin K$_3$/PVP/GC electrode has been tested for the determination of caffeine in well known analgesic Saridon. One tablet of Saridon is dissolved in water and diluted to get a final concentration of 0.1 mM. A 0.1 mM standard solution of caffeine is also prepared. The relative decrease in cathodic currents (decrease in current divided by initial current) of the modified electrode was determined at...
caffeine concentration 0.05, 0.1, 0.15, 0.2 and 0.25 μM for both the standard caffeine solution and Saridon tablet solution. The relative decrease in current at a particular caffeine concentration is found to be same for both the solutions. This confirms the applicability of the electrode in determination of caffeine in tablet solution.

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References