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Electrochemical Reduction Of Zn (II)- Amino And Nucleic Acid Complexes At Dropping Mercury Electrode

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ABSTRACT: The dropping mercury electrode (DME) is a working electrode made of mercury and used in polarography. Experiments run with mercury electrodes are referred to as forms of polarography even if the experiments are identical or very similar to a corresponding voltammetry experiment which uses solid working electrodes. Like other working electrodes these electrodes are used in electrochemical studies using three electrode systems when investigating reaction mechanisms related to redox chemistry among other chemical phenomena. A flow of mercury passes through an insulating capillary producing a droplet which grows from the end of the capillary in a reproducible way. Each droplet grows until it reaches a diameter of about a millimeter and releases. The released droplet is no longer in contact with the working electrode whose contact is above the capillary. As the electrode is used mercury collects in the bottom of the cell. In some cell designs this mercury pool is connected to a lead and used as the cell's auxiliary electrode. Each released drop is immediately followed by the formation of another drop. The drops are generally produced at a rate of about 0.2 Hz. The present review is based on electrochemical reduction of Zn (II)-amino and nucleic acid complexes at dropping mercury electrode.

KEYWORDS: dropping mercury electrode, polarography, voltammetry, workin, auxiliary, electrochemical

I. INTRODUCTION

Zinc is an essential element playing numerous crucial roles in organisms. It is involved especially in the synthesis of proteins and DNA [1], because zinc stabilizes the structure of chromatin and affects replication of DNA and transcription of RNA by regulating the activity of transcription factors for RNA and DNA polymerases [2]. Zinc is also essential to stabilize the structure of proteins containing zinc finger motifs [3]. Zinc is further closely connected with the production of insulin [4], and in light of this fact, zinc complexes could find an application in the treatment of diabetes [5]. Zn(II) complexes are able to modulate an inflammatory response by influencing the secretion and activity of several inflammation-related cytokines and enzymes [6]. Moreover, xylan-chitooligomer-zinc complex exhibited antioxidant and antimicrobial activity [7]. Transition metal complexes that are capable of cleaving DNA under physiological conditions are of interest in the development of anticancer drugs [8]. Cisplatin and related platinum-based drugs bind covalently to DNA, but they have side effects, especially, toxicity and acquired drug resistance, that requires the development of new drugs, which bind non-covalently to DNA, are less toxic and are target-specific. Among the non-platinum complexes for metal based chemotherapy, copper and zinc complexes have been much explored due to the fact that both copper and zinc are bioessential elements responsible for numerous bioactivities in living organisms [9–11]. Role of zinc and copper complexes as potential chemotherapeutic compounds have been confirmed, both complexes were able to bind and cleave DNA [12]. Zinc sulphide (ZnS) is one of the first semiconductors discovered and it has shown remarkable properties, versatility and a promise for novel diverse applications, including light-emitting diodes (LEDs), electroluminescence, flat panel displays, infrared windows, sensors, lasers, and biodevices, etc. [13]. Its atomic structure and chemical properties are comparable to more popular and widely known ZnO [14]. In the past decade, numerous results have been reported on the synthesis of nanometer scale semiconductor crystals (quantum dots, nanowires, nanorods, etc.) because their properties, due to quantum confinement effect, dramatically change and, in most cases, improve as compared with their bulk counterparts [15–17]. Among them, ZnS quantum dots (QDs) as semiconductor nanocrystals with a typical size of 2–10 nm have been attracting much interest [18]. An advantage of ZnS QDs is that they can be analysed electrochemically [19].

There is a wide range of well-established techniques for detection of metals, including the most widely used mass spectrometry and atomic absorption spectrometry. These methods are reliable and highly sensitive. On the other hand, they require expensive instrumentation and involve time-consuming procedures. Electrochemical methods represent another class of widely used techniques for the detection of metal ions. Anodic stripping voltammetry has become one

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of the most important techniques [20–22] in this field, together with hanging mercury drop electrode (HMDE) [23–25]. The disadvantage of this method is the difficulty of miniaturization, especially due to the hanging drop, which needs the supply of gas. Another disadvantage of the mercury electrode is its limited modification possibilities, a small anodic range (limited by the oxidation of mercury) and the high toxicity of mercury. Mercury electrodes also cannot be used in a flow system.

Despite their sensitivity issues, screen printed electrodes (SPEs) are a suitable alternative to HMDE. The low acquisition costs of lithographic equipment have enabled the widespread use of disposable SPEs as biosensors and chemical sensors in microfluidic systems. Microfluidics is a technology that requires lower volumes of sample, increases the speed of analysis and response time, allowing a massive parallelization for high-throughput analysis, and reducing the cost of fabrication of biosensors [26–28]. In recent years, methods involving the coupling of microfluidics with electrochemical techniques have been increasing because of the benefits associated with miniaturization, automation, sensitivity and specificity [29–35].

Based on the abovementioned facts we investigated the combination of zinc as a central atom, 1,10-phenanthroline (phen) as a versatile N-N chelating aromatic ligand that can interact with DNA by π-π interaction and histidine as an amino acid with a side chain aromatic ring. Aromatic ligands also play an important role in enhancing DNA binding and cleavage activity. We also selected histidine as a ligand because it is known that amino acids/peptides recognize a specific base sequence of DNA and that aromatic ring contributes to the stabilization of proteins through hydrophobic interactions and the formation of hydrophilic environments [36].

Since the metal ion complexes play an important role in life science, pharmacology, complexometric titration, colorimetric analysis, precipitation and solvent extraction, the determination of the stability constants of metal ion complexes with different ligands is very important[1]. Azo dyes are a very important class of organic compounds receiving attention in the scientific literature. Recently, azo metal chelates have drawn the attention of some research due to their excellence in sensitivity and stability as optical recording medium Moreover, the complexes of some azo dyes with metal ion Zn(II). Among the electrochemical techniques, voltammetry has been widely used to study the interactions between metal ions and ligands . The voltammetric and polarographic studies onthe metal complexes of the some azo-dye ligands were done in the literature .

AR 1 is an azo dye, used in light and fluorescence microscopy as a real acid counterstain. In addition, AR 1 is a reactive azo dye belonging to the largest class of dyes commonly employed in textile industry. No information is available about the voltammetric behaviour of Zn(II) complexes with AR 1. [35-41]

II. DISCUSSION

The square-wave voltammogram of $4.76x10^{-5}$ M Zn(II) ions in NH_3/ NH_4C1 buffer solution pH 9.20 exhibited only one cathodic peak at -1.184 V (Fig. 7). In the literature[20], it was indicated that $[Zn(NH_3)]^{2+}$ reduced at half-wave potential of-1.33 V (versus satured calomel electrode) with a cathodic peak of two-electrons (2 —> 0). So, the peak at -1.184 V can be sourced from the reduction of $[Zn(NH_3)]^{2+}$ ions to the amalgam (Zn(II) + $2e^-$ $\rightleftharpoons$ Zn(Hg)). After adding AR 1 into the cell containing $4.76x10^{-55}$ M Zn(II), a new peak was no observed except for the $[Zn(NH_3)]^{2+}$ and AR 1. With increasing AR 1 concentration, the peak current of $[Zn(NH_3)]^{2+}$ decreases (Fig. 8) but its peak potential remains practically unchanged, which verifies the inert character of Zn(II)-AR 1 complex under the experimental conditions employed. The similar results were obtained for the Zn(II)-nitrilotriacetic acid (NTA)[35] and Zn(II)-cysteine[11] systems.

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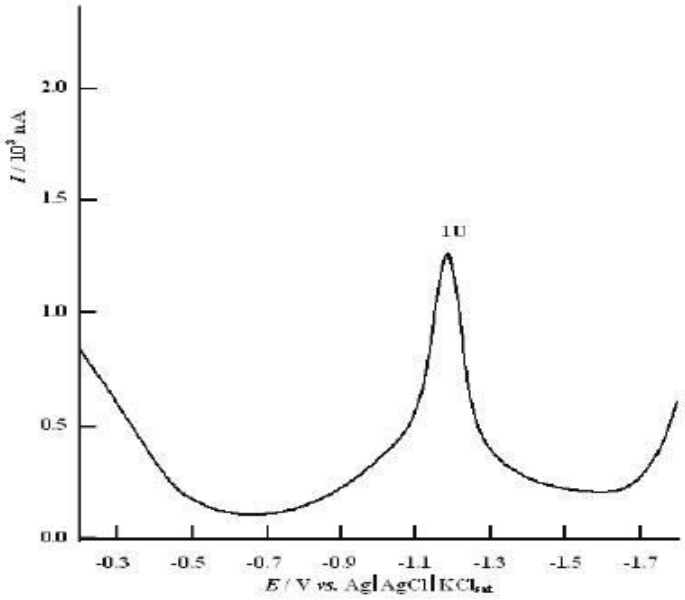
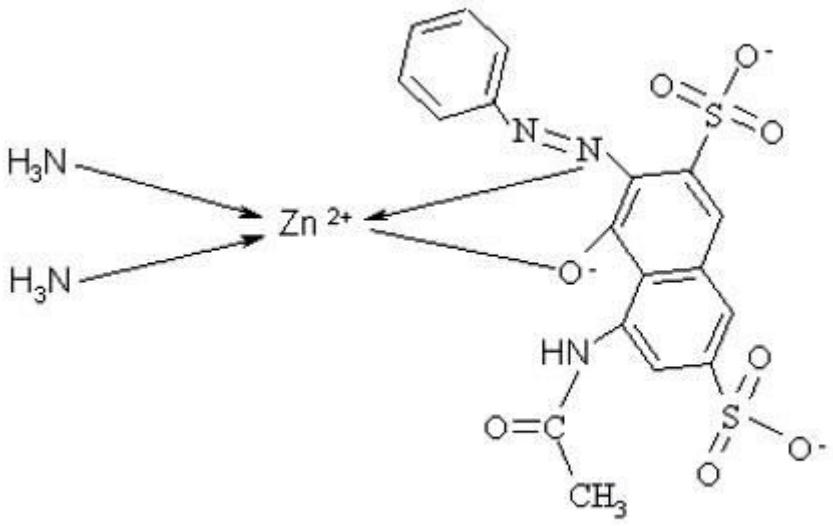


Fig. 7. Square-wave voltammogram of 4.76×10^{-5} M Zn(II) solution at NH_3/NH_4Cl buffer solution pH 9.20. **1U**, the reduction of free Zn(II). *Other conditions as in Fig. 1.*

At a similar manner to the binding of Zn(II) ions with some azo-compounds, the coordination of AR 1 to Zn(II) ions may occur mainly by means of its azo-nitrogen and phenolic oxygen. Treatment of $[Zn(NH_3)]^{2+}$ species with AR 1 may cause displacement of ammonia molecule by AR 1 [25-30]



Scheme 3. The possible structure of Zn(II)-AR 1 complex.

In a health perspective the need for the analysis of gene sequences, oxidative damage to DNA and the understanding of DNA interactions with molecules or ions led to the development of DNA-based biosensors. The DNA-based biosensor is a device that incorporates immobilized DNA as molecular recognition element in the biological active layer on the surface, and measures specific binding processes with DNA mainly using electrochemical, optical and piezoelectric transducers. The fact that the DNA sequences are unique to each organism means that any self-replicating biological organism can be discriminated. The DNA-based biosensor is also a complementary tool for the study of biomolecular

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interaction mechanisms of compounds with DNA, enabling the screening and evaluation of the effect caused to DNA by health hazardous compounds and oxidizing substances. There are hundreds of compounds which bind and interact with DNA. Exposure to toxic chemicals is the cause of many human cancers; these carcinogens act by chemically damaging the DNA[27,28]. Thus it is very important to explain the factors that determine affinity and selectivity in binding molecules to DNA, identify these chemicals and ascertain their potency so that human exposure to them can be minimized. The reactions with chemicals cause changes in the structure of DNA and the base sequence leading to perturbations in DNA replication. A quantitative understanding of the reasons that determine selection of DNA reaction sites is useful in designing sequence-specific DNA binding molecules for application in chemotherapy and in explaining the mechanism of action of neoplasic drugs. Electrochemical techniques have the advantage in DNAbiosensor design of having a rapid response time, being quantitative, sensitive, suitable for automation, cost effective, disposable, enabling in situ generation of reactive intermediates and detection of DNA damage and solving analytical problems in a wide range of contexts in order to be commercially viable. Comprehensive descriptions of research on DNA and DNA sensing show the great possibilities of using electrochemical transduction in DNA diagnostics.[29,30,31]

III. RESULTS

Electrochemical oxidation on carbon electrodes [17–24] showed that all bases–guanine (G), adenine (A), cytosine (C) and thymine (T)–can be oxidized, following a pH dependent mechanism. The voltammetric studies on DNA shown in this figure include all four bases—for the first time equimolar mixtures of all DNA bases, nucleosides and nucleotides have been quantified by differential pulse voltammetry [18]. Electrochemical pre-conditioning of the glassy carbon electrode enabled a better peak separation and an enhancement of the current of the oxidation peaks for all four DNA bases in pH 7.4 phosphate buffer supporting electrolyte, close to physiological pH. Detection limits in the nano- and micromolar range were obtained for purine and pyrimidine bases, respectively, together in solution. The results showed for the first time that the pyrimidine nucleosides and nucleotides are electroactive on glassy carbon electrodes and that, besides the easy detection of the purines, it was also possible to detect simultaneously the oxidation of pyrimidine residues in ssDNA [18]. Electrochemical reduction of natural and biosynthetic nucleic acids at a dropping mercury electrode (DME) [1, 6, 12, 13] showed that adenine and cytosine residues as well as guanine residues in a polynucleotide chain are reducible. The cyclic voltammogram of ssDNA at a hanging mercury drop electrode (HMDE) showed a cathodic peak due to irreversible reduction of cytosine (C) and adenine (A). The reduction of guanine (G) occurs at very negative potentials but a peak due to the oxidation of the reduction product of (G) could be detected in the reverse scan [6].

Electrochemical research on DNA is of great relevance to explain many biological mechanisms. The DNA-modified electrode is a very good model for simulating the nucleic acid interaction with cell membranes, potential environmental carcinogenic compounds and to clarify the mechanisms of action of drugs used as chemotherapeutic agents. The use of DNA-electrochemical biosensors for the understanding of DNA interactions with molecules or ions exploits the use of voltammetric techniques for in situ generation of reactive intermediates and is a complementary tool for the study of biomolecular interaction mechanisms. Voltammetric methods are an inexpensive and fast detection procedure. Additionally, the interpretation of electrochemical data can contribute to elucidation of the mechanism by which DNA is oxidatively damaged by such substances, in an approach to the real action scenario that occurs in the living cell. The development of the DNA-electrochemical biosensor has opened wide perspectives using a particularly sensitive and selective method for the detection of specific interactions. The possibility of foreseeing the damage that these compounds cause to DNA integrity arises from the preconcentration of either the starting materials or the redox reaction products on the DNA-biosensor surface, thus permitting the electrochemical probing of the presence of shortlived intermediates and of their damage to DNA.[36-41]

IV. CONCLUSIONS

Kinetic parameters of Zn (II) in presence of ligand l-lysine, l-aspartic acid, l-glutamic acid, larginine, l-tryptophan and l-tyrosine at pH 7.50 ± 0.02 and at constant ionic strength $\mu = 0.1$ $NaClO_4$ have been evaluated. The reductions in all these cases were found to be quasi reversible and diffusion controlled. The values of kinetic parameters for the electrode processes viz and ks have been evaluated using Gelling's treatment1 and Er 1/2 values were also calculated for quasi reversible electrode processes.[38-41]

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