Acidity and complex formation studies of 3-(adenine-9-yl)-propionic and 3-(thymine-1-yl)-propionic acids in ethanol–water media

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HIGHLIGHTS

- Synthesis and characterization of adenine and thymine propionic acid derivatives.
- Determination of acid dissociation constants and metal stability constants.
- Determination of thermodynamic parameters.

ARTICLE INFO

ABSTRACT

The ligands 3-(adenine-9-yl)propionic acid (AA) and 3-(thymine-1-yl)propionic acid (TA) were prepared by N9-alkylation of adenine and N1-alkylation of thymine with ethylacrylate in presence of a base catalyst, followed by acid hydrolysis of the formed ethyl esters to give the corresponding propionic acid derivatives. The products were characterized by spectral methods (FTIR, ^{1}H NMR and ^{13}C NMR), which confirm their structures. The dissociation constants of ligands, were potentiometrically determined in 0.3 M KCl at 20–50 °C temperature range. The work was extended to study complexation behavior of AA and TA with various biologically important divalent metal ions (Co^{2+}, Ni^{2+}, Cu^{2+}, Zn^{2+}, Cd^{2+}, Mn^{2+} and Pb^{2+}) in 50% v/v water–ethanol medium at four different temperatures, keeping ionic strength constant (0.3 M KCl). The order of the stability constants of the formed complexes decreases in the sequence Cu^{2+} > Pb^{2+} > Zn^{2+} > Ni^{2+} > Co^{2+} > Mn^{2+} > Cd^{2+} for both ligands. The effect of temperature was also studied and the corresponding thermodynamic functions (ΔG, ΔH, ΔS) were derived and discussed. The formation of metal complexes has been found to be spontaneous, and the stability constants were dependant markedly on the basicity of the ligands.

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Introduction

After the discovery of the famous DNA double-stranded helix by Watson and Crick, many research efforts have been dedicated to the rational design and elaboration of biomimetic systems based on the interaction of nucleobases and their derivatives with a wide range of metal ions. Such interactions depend on multiple factors that become further understood due to solution and solid state studies. The effect of platinum-based chemotherapeutic drugs probably originates from their attack on DNA. Studies into metal–nucleobase bindings are of great interest since metal ions play a crucial role in the structure and function of nucleic acids and genetic information transfer [1]. Medicinal inorganic chemistry study ligands as drugs which target free or protein-bound metal ions; and secondly, metal-based drugs and imaging agents. Currently, the main way to fight cancer is still chemotherapy. Metal complexes have already been suggested as possible means of exploiting the difference between normal and cancer cells and thereby destroying the latter. Numerous structural analogs to cisplatin entered clinical trials but most of them have been abandoned due to unacceptable toxicity profile and/or lack of improved anticancer efficacy. The need for new agents in cancer chemotherapy is apparent from the inability to predictably cure or induce remissions in tumors [2,3]. Nucleobases are very versatile ligands with large number of heteroatoms available for metal binding; their derivatives as well as their metal complexes have

proved to be an important group of compounds with anticancer activity. Among the different nucleobases, adenine (6-aminopurine) shows the widest range of binding possibilities because it exhibits at least five donor sites (N9, N7, N3, N1 and N6), and a great variety of complexes with different metal-ion binding patterns have been reported [4]. It is surprising that exocyclic amino groups of the nucleobases are not metal binding sites due to delocalization of the lone electron pair into the heterocyclic ring. Coordination is only possible when the amino group is deprotonated or adopts a rare imino tautomer structure. By the judicious choice of the type of substituents that can be attached on N1, N3, N6, N7 and N9 centers, the biological function displayed by adenine could be tuned. This heterocyclic adenine also has an ability to coordinate a variety of metal ions, that could be used for the stabilization of superstructures as well as to support metal-aided catalytic transformations [5]. Another important nucleobase is thymine, which is pyrimidine like and is unique among the nucleic acid constituents because it does not have nitrogen lone pairs available for metal complexation. Thus it can act as a bidentate ligand through O4 and O2 [6].

Due to biological importance of adenine and thymine, considerable research work has been done on the study of their metal complexes in solution. The interest in such complexes continues to increase due to the possibility of their use as models to explain some intricate reaction in biological systems such as anticancer, antiviral and antimalarial [7–11]. Literature survey shows that Hamada et al. reported the potentiometric study of some divalent metal complexes of adenine in aqueous phase at 25 °C and 1 M NaNO$_3$ [12]. Ammar et al. investigated the formation constants of mixed ligand complexes Cu(II)-adenine-amino acids at 25 °C and ionic strength (I) 0.1 M NaNO$_3$ by pH-metric titration [13]. In another work, the authors studied potentiometrically the formation equilibria and stability constants of binary and ternary complexes of Ni(II) involving adenine and various biologically relevant ligands [14]. Shukla et al. also used potentiometric techniques in order to determine the formation constants and complexation equilibria at 30 °C and at I = 0.1 M NaNO$_3$ for quaternary metal complexes of some divalent metal ions containing thymine [15]. The formation constants of the ternary (1:1:1) and quaternary (1:1:1:1) complexes of some divalent metals with glutamic acid/L-cysteine as primary ligand and thymine as secondary ligand were determined potentiometrically in aqueous medium by Krishna et al. [16]. Concerning ligand synthesis, literature reveals that carboxylic acid derivatives of adenine and thymine had been prepared via N9 and N1 alkylation, respectively using ethylacrylate followed by acid hydrolysis [17–19] or by reaction with halogenated acetic acid [20].

However, to the best of our knowledge, a detailed study of complexation of nucleobase-carboxylic acid derivatives with various metal ions and the determination of their stability constants is still lacking. The coordinating properties of ligands of this type are largely unknown at the present time. In this paper the binding ability of functionalized 3-(thymine-1-yl)propionic acid and 3-(adenine-9-yl)propionic acid towards some divalent heavy metals, Cd, Co, Cu, Pb, Ni, Mn and Zn, was studied. The formation constants of the metal complexes have been determined potentiometrically adopting the modified Irving and Rossotti technique.

Experimental part

Materials and methods

All chemicals and reagents were of analytical grade, purchased from commercial sources. The progress of reactions was monitored by TLC using aluminum silica gel plates 60 F254. Melting points were measured with a Gallenkamp apparatus. IR spectra were recorded in KBr pellet, on a Nicolet iS 10 FT/IR spectrophotometer. ^{1}H NMR and ^{13}C NMR spectra were recorded on a Bruker 500 MHz NMR spectrometer in CDCl$_3$ and D$_2$O, using TMS and DSS as references; chemical shifts are reported in ppm, and signals are expressed as s (singlet), d (doublet), t (triplet), q (quartet) and m (multiplet).

In potentiometric titration, deionized water was boiled for two hours to minimize atmospheric carbon dioxide contamination. There after it was cooled to ambient room temperature, 25 °C, in a closed vessel leaving no headspace. All pH measurements were made using a pH-meter Eutech pH 700 in well-stirred solutions. The temperature of the systems was maintained at 25 °C ± 0.1 °C during titration, with a water bath (VWR, model 12101-10) and a glass titration cell with a double jacket. Stock solutions of metal ions, **AA**, **TA** and KCl for pH-metric titrations were prepared in deionized water. Solutions of Zn(II), Co(II), Mn(II), Ni(II), and Cu(II) were prepared from their chlorides and Cd (II) and Pb(II) from their nitrates. NaOH solution was prepared by dissolving Analar pellets in water, and the solution was standardized with a standard potassium hydrogen phthalate solution. A total concentration of 0.3 M KCl was used as a supporting electrolyte.

Calibration of the glass electrode

Before starting pH measurements of each set, the pH meter scale was calibrated using two standard aqueous buffer solutions. The term pH has significance only in aqueous media. The glass electrode potential in an aqueous solution differs from that in a solution of mixed solvents, and a liquid-junction potential of an uncertain value can affect the results. To overcome this difficulty, it was necessary to calibrate the glass electrode in different solvent mixtures.

The pH-meter readings in ethanol–water media were corrected according to the Van Uitert and Hass relation for dioxane–water mixtures later validated by Bates et al. [21] for use in alcohol–water system. The pH-meter reading B in ethanol + water media was converted into [H$^+$] using Uitert–Hass equation:

$$-\log[H^+] = B + \log U_H$$

where $\log U_H = \log U_H^\circ + \log \gamma_\pm$

$\gamma_\pm$ is the activity coefficient of the hydrogen ions in the solvent mixture under consideration at the same temperature and ionic strength, and U_H° is a correction factor at zero ionic strength, which depends only on the solvent composition.

In this work, the values of B were recorded in various solvent mixtures at different temperatures containing known concentration of hydrochloric acid (1.00×10^{-3} M) and sufficient potassium chloride to give a constant ionic strength of 0.3 M. The difference between the logarithm of known hydrogen ion concentrations and the corresponding values of B was used to calculate values of the correction term $\log U_H = \log(U_H^\circ \gamma_\pm)$ [21].

Synthesis of ligands

The ligands were synthesized according to the reaction in Scheme 1, shown below:

3-(6-Aminopurine-9-yl)-propionic acid ethylester, 1(a)

To a suspension of adenine (11.0 g, 81.4 mmol) in absolute ethanol (280 ml) and dry benzene (35 ml), sodium metal (120 mg, 5.2 mmol, 0.06 eq) was added carefully at room temperature. Upon the disappearance of sodium, ethyl acrylate (26 ml, 244 mmol, 3 eq) was added dropwise. The resulting mixture was refluxed overnight, and then cooled to room temperature. The solvent was