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Electrochemical recognition of anions by 1,1′-*N*,*N*′-ferrocenoylbisamino acid esters

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Abstract

A series of novel ferrocene-based receptors, 1,1′-*N*,*N*′-ferrocenoylbisamino acid methyl esters **2**–**5** have been prepared and their electrochemical properties determined. The amino acids employed were glycine (**2**), β-alanine (**3**), γ-aminobutyric acid (**4**) and L-norleucine (**5**). These receptors are composed of an electroactive core and two parallel strands of amino acids that can interact with anions *via* electrostatic interactions in the oxidized state as well as secondary interactions, such as hydrogen bonding and hydrophobic interactions. Furthermore, the semi-rigid molecular clefts between the two strands of amino acids in these receptors are capable of discerning anions of different geometries and sizes. The anion sensing capabilities of receptors **2**–**5** were studied using cyclic voltammetry (CV). The anions studied were chloride (Cl^-), nitrate (NO_3^-), dihydrogen phosphate ($H_2PO_4^-$), hydrogen sulfate (HSO_4^-), acetate (CH_3COO^-) and neurologically important anions such as lactate ($CH_3CH(OH)COO^-$), pyruvate (CH_3COCOO^-) and glutamate ($HOOC–CH(NH_2)CH_2CH_2COO^-$). The receptors **2**–**5** exhibit selectivity towards chloride, dihydrogen phosphate and acetate, over hydrogen sulfate and nitrate ions and generate a redox response in organic media. Also, binding studies of receptor **3** with neurologically important anions show it displays selectivity towards lactate and pyruvate, over glutamate ions and generates a redox response. However the response is ill defined in all cases, with poorly separated "free" and "ion-pair" peaks, which preclude accurate measurement of the response by CV. These results emphasize the considerations required for the design of ferrocene-based receptors and the necessary parameters for efficient electrochemical recognition of small anions by CV.

Keywords: Ferrocene; Amino acids; Anion receptors; Molecular recognition; CV

1. Introduction

Anions play important roles in biological, chemical and environmental processes. Thus, the design and synthesis of molecular receptors with the ability to selectively bind anions and provide a quantifiable physical response is a rapidly growing area of interest (Beer and Hayes, 2003; Beer and Gale, 2001). The excellent stability of ferrocene and its derivatives coupled with their favorable electrochemical properties make these derivatives suitable as receptors for sensor devices with electrochemical detection. The ferrocene-based receptors can interact with anions *via* electrostatic interactions, hydrogen bonding, hydrophobic interactions, or a combination of two or more of these interactions. The electrostatic interactions can be switched on by the electrochemical oxidation of the ferrocene to the ferricinium ion; hydrogen bond donor groups such as amide groups can provide the hydrogen bonding interactions, and the hydrophobic environment can be created by appending phenyl or alkyl groups to the ferrocene moiety. Furthermore, ferrocene-based receptors with molecular clefts or cavities can discern anions with different geometries or hydrogen bonding requirements.

We have previously reported the syntheses and structural characterization of various novel *N*-ferrocenoyl and *N*-ferrocenyl benzoyl amino acid and peptide derivatives (Savage et al., 2006a,b, 2005a,b, 2002). Herein, we report the synthesis and the anion sensing ability of a series of 1,1′-*N*,*N*′-ferrocenoylbisamino acid methyl esters **2**–**5**. These receptors are composed of an electroactive core and two parallel strands of amino acids that can interact with other molecules *via* electrostatic interactions in the oxidized state as well as secondary interactions, such as hydrogen bonding with the amide groups

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and hydrophobic interactions with the alkyl groups/chains of the amino acids. Moreover, these receptors have an ordered structure in organic solvents (Herrick et al., 1996), stabilized by two intra-molecular hydrogen bonds N–H···O=C between the amide group N–H of one strand and the ester group O=C of the other strand, that creates a semi-rigid molecular cleft between the two strands. The molecular clefts of these receptors have specific shapes and sizes depending on the length of the amino acid strands, and thus these receptors are capable of discerning anions of different geometries and sizes. We evaluated the anion recognition capacity of receptors **2–5** using cyclic voltammetry. These receptors are composed of amino acids with different alkyl chains and thus hydrophobicity, the size and shape of the molecular cleft is unique to each receptor. The anions studied were of different shapes and sizes as well, *i.e.*, Cl^- (spherical), $H_2PO_4^-$ (tetrahedral), HSO_4^- (tetrahedral), CH_3COO^- (trigonal planar) and NO_3^- (trigonal planar). In addition, the neurologically important anions (Georganopoulou et al., 2000), such as lactate ($CH_3CH(OH)COO^-$), pyruvate (CH_3COCOO^-) and glutamate ($HOOC–CH(NH_2)CH_2CH_2COO^-$) were also studied and results from the anion binding studies for receptor **3** are presented.

2. Experimental

2.1. Synthesis

The compounds **2–5** were synthesized by reacting stoichiometric amounts of the amino acid methyl ester hydrochloride salts of glycine (**2**), β-alanine (**3**), γ-aminobutyric acid (**4**) and L-norleucine (**5**) with 1,1′-ferrocenedicarboxylic acid **1** using equimolar amounts of the coupling agents 1,3-dicyclohexylcarbodiimide (DCC), 1-hydroxybenzotriazole (HOBt) and excess of triethyl amine in dichloromethane. Purification by silica gel column chromatography furnished the pure products in yields of 68–72% and all gave analytical and spectroscopic data, including ^{1}H NMR, ^{13}C NMR, HMQC, DEPT-135 and ESI-MS, in accordance with the proposed structures.

Typically, γ-aminobutyric acid methyl ester hydrochloride (1.05 g, 7.5 mmol) was added to a solution of 1,1′-ferrocenedicarboxylic acid **1** (1.02 g, 3.75 mmol), 1-hydroxybenzotriazole (1.02 g, 7.5 mmol), triethylamine (0.5 ml), and dicyclohexylcarbodiimide (1.5 g, 7.5 mmol) in 40 ml of dichloromethane at 0 °C. After 30 min, the temperature was raised to room temperature and the reaction was allowed to proceed for 48 h. The precipitated *N*,*N*′-dicyclohexylurea was removed by filtration and the solvent was removed *in vacuo*. The product was purified by column chromatography {eluant 1:3 petroleum ether (40–60 °C): ethyl acetate}. Recrystallization from petroleum ether (40–60 °C): ethyl acetate furnished compound **4** as an orange powder (1.2 g, 68%), *m/z*: 473 $[M+H]^+$, 495 $[M+Na]^+$, mp 131–132 °C, $E'^0 = 0.28$ V (versus Fc/Fc^+), IR, ν_{max} (KBr): 3327, 2929, 2851, 1729, 1629, 1538, 1437, 1229, 1103 cm^{-1}. UV–vis, λ_{max} EtOH; 344, 444 (ε190) nm. ^{1}H NMR (400 MHz), δ (DMSO-d_6): 7.94 (2H, t, $J = 5.2$ Hz, –CONH– x2), 4.71 {4H, s, *ortho* on (η^5-C_5H_4)}, 4.28 {4H, s, *meta* on (η^5-C_5H_4)}, 3.60 (6H, s, $–OCH_3$ x2), 3.19 (4H, m, $–NHCH_2–$ x2), 2.39 (4H, t, $J = 6.8$ Hz, $–CH_2CO–$ x2), 1.77 (4H, m, $–NHCH_2CH_2–$ x2). ^{13}C NMR (100 MHz), δ (DMSO-d_6): 173.57, 168.78, 78.28, 71.80, 69.82, 51.64, 38.43 (−ve DEPT), 31.09 (−ve DEPT), 25.01 (−ve DEPT). Elemental analysis: $C_{22}H_{28}N_2O_6Fe$ requires: C 55.95%, H 5.98%, N 5.93%; Found: C 55.81%, H 5.94%, N 5.84%.

2.2. Electrochemical measurements

Cyclic voltammograms were recorded in acetonitrile (Sigma–Aldrich), with 0.1 M tetrabutylammonium perchlorate (TBAP) as a supporting electrolyte, using a CH Instruments electrochemical analyzer (Pico-Amp Booster and Faraday Cage). The experiments were carried out at room temperature. A three-electrode cell consisting of a glassy carbon working-electrode, a platinum wire counter-electrode and an Ag|AgCl reference electrode was used. The glassy carbon electrode was polished with 0.3 μm alumina followed by 0.05 μm alumina, between each experiment to remove any surface contaminants. Receptor solutions (1 mM) containing tetrabutylammonium perchlorate (0.1 mol, TBAP) were prepared in acetonitrile. The anions were added as either their tetrabutylammonium or sodium salts and were prepared in acetonitrile; acetonitrile–water (1:1) for lactate, pyruvate and glutamate. Typically, the receptor solution (5 ml) was pipetted into an electrochemical cell, the electrodes were inserted into the receptor solution and the cyclic voltammograms was recorded by scanning voltage in a pre-defined range (*e.g.*, 0.0–1.2 V) at a scan rate of 100 mVs^{-1}. A CV scan of a sample of ferrocene (1 mM) with tetrabutylammonium perchlorate (0.1 mol) in acetonitrile was also recorded before each experiment to obtain $E^{0\prime}(Fc/Fc^+)$, and the $E^{0\prime}$ values obtained for the receptors were referenced to the Fc/Fc^+ couple. The anion binding studies were carried out by sequential additions of the anion solutions (25 μl, 1 mmol) to the receptor solutions containing TBAP, and the CV scans were recorded after each addition.

3. Results and discussion

The CV curves of receptors **2–5** exhibit quasi-reversible redox behavior, similar to the Fc/Fc^+ redox couple, and display higher oxidation potential values with respect to ferrocene itself; the $E^{0\prime}(Fc/Fc^+)$ being 0.0 V by definition. The key features of their redox behavior are summarized in Table 1.

Table 1
All potentials are referenced to the ferrocene/ferrocenium redox couple ($E^{0\prime} = 0.45$ V, under the experimental conditions)

Compound	$E^{0\prime}$ (V)	$\Delta E^{0\prime}$ (V)	I_a/I_c
2	0.45	0.09	1.08
3	0.34	0.07	1.05
4	0.28	0.08	1.06
5	0.33	0.08	1.06

$E^{0\prime}$ refers to the formal potential and is calculated by taking the mean of the peak potentials for the anodic (a) and the cathodic wave (c); I_a and I_c refer to the maximum peak currents of the anodic and the cathodic waves, respectively.