Mononuclear and Binuclear Copper(II) Complexes of the Antiinflammatory Drug Ibuprofen: Synthesis, Characterization, and Catecholase-Mimetic Activity

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ABSTRACT

Two mononuclear copper(II) ibuprofenate adducts with imidazole or 2-methylimidazole and two binuclear copper(II) ibuprofenate adducts with metronidazole or caffeine have been prepared and characterized. Elemental analyses, UV-VIS, IR, EPR, and magnetic moment data for imidazole or 2-methylimidazole adducts are consistent with mononuclear square planar complexes that contain two ibuprofenato ligands and two N-containing imidazole ligands to give essentially a CuO_2N_2 chromophore. The above data for metronidazole or caffeine adducts are consistent with a binuclear structure as found for copper(II) acetate monohydrate and other copper(II) carboxylate dimers. In these complexes four carboxylate groups are bridging two copper(II) atoms, and two added bases coordinated at axial positions to form CuO_4N chromophore around each copper. The catecholase-mimetic catalytic activities of the complexes have been determined by monitoring the formation of o-quinone from catechol. The catalytic activities of the mononuclear complexes are lower than those of the binuclear copper(II) ibuprofenate or its metronidazole or caffeine mono-adducts.

INTRODUCTION

Ibuprofen (2-(4-isobutylphenyl)propionic acid) is a nonsteroidal antiinflammatory drug and has been used for many years in treatment of inflammatory diseases. Copper(II) complexes of antiinflammatory drugs have been found to be more potent and desirable drugs than the parent ligands themselves [1]. Physical studies of copper(II) ibuprofenate [2, 3] have shown that it contains binuclear units with bridging carboxylate ligands similar to other copper(II) carboxylates[4].

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Journal of Inorganic Biochemistry, **55**, 255–262 (1994)

0162-0134/94/$7.00

The only known adduct of copper(II) ibuprofenate is that with pyridine, which has been shown to be a mononuclear complex of the type $[Cu(O_2CR)_2.L_2.H_2O]$ [5].

The coordination chemistry of imidazoles and purines-type ligands have been subjected to intensive studies because of the importance of the interactions between these ligands and metal ions in many biological system [6]. In addition, copper(II) carboxylate adducts with imidazole type ligands have been found to have a variety of pharmacological effects such as anticancer [7], superoxide dismutase [8], and catecholase mimetic activities [9]. Since nitroimidazoles are used as chemotherapeutic agents in the treatment of bacterial infections and as radiosensitizers [10, 11], metronidazole (1-(2-hydroxyethyl)-2-methyl-5-nitroimidazole) is used in this study in addition to other ligands, imidazoles, and caffeine to form copper(II) ibuprofenate adducts.

Interest in developing small molecular weight copper(II) complexes as model copper oxidase enzymes had led to the synthesis of many binuclear and mononuclear copper(II) complexes [9, 12–19]; the idea being that these complexes might mimic the behavior of various metalloproteins such as the copper containing protein tyrosinase. This enzyme is found in many plants and animals and whose primary function is to catalyze the oxidation of phenols to o-diphenols (cresolase) and o-diphenols to o-quinones (catecholase) [20]. Recently, we reported the synthesis and characterization of several mononuclear copper(II) carboxylates and studied the catecholase mimetic activities of some of these adducts [9, 21]. We found the dependence of the structure of copper(II) carboxylates on the electronic property of the carboxylate groups, and the electronic and steric properties of added bases [9, 21–23]. In addition, the rate of oxidation of catechol to o-benzoquinone by copper(II) complexes studied is dependent on the nature of nitrogen containing ligands [9, 21]. Therefore, structural and electronic factors that might impact the properties of copper(II) complexes as metalloprotein models are of interest. This article reports the synthesis and spectroscopic characterization of mononuclear and binuclear copper(II) ibuprofenate complexes and examines their catalytic properties in the aerobic oxidation of catechol to the corresponding o-quinone as models for the catecholase function of the copper containing enzyme tyrosinase.

EXPERIMENTAL

Reagents and Materials

All chemicals were of high purity grades (Aldrich or Sigma Chemical Co.) and were used without further purification. Tetrakis-μ-ibuprofenato dicopper (II), $[Cu_2(Ibup)_4]$, (*1*), (Ibup = ibuprofenato ion) was prepared according to a published procedure [2] and recrystallized from anhydrous diethylether and dichloromethane (1 : 1)

Preparation of Complexes

Bis(ibuprofenato) bis(imidazole) copper(II), $[Cu(Ibup)_2(Im)_2]$, (2). A solution of 0.089 g (1.30 mmol) of imidazole in 15 mL methanol was added to 0.303 g (0.32 mmol) of $Cu_2(Ibup)_4$. The mixture was stirred at about 60°C for 1 hr. The blue solution was filtered and left in the hood to evaporate. The blue precipi-

tate that formed recrystallized from methanol and air dried. Anal. Calc. for $C_{32}H_{42}N_4O_4Cu$: C, 63.00; H, 6.89; N, 9.19. Found: C, 63.10; H, 7.10; N, 9.25%.

Bis(ibuprofenato) bis(2-methylimidazole) copper(II), [Cu(Ibup)$_2$(2-Mim)$_2$], (3). A solution of 0.192 g (2.33 mmol) 2-methylimidazole in 30 mL methanol was added to 0.538 g (0.568 mmol) of $Cu_2(Ibup)_4$. The mixture was stirred at about 60°C for 1 hr. The blue solution was filtered and left in the hood to evaporate. The bluish purple precipitate that formed was washed with anhydrous diethylether and air dried. Anal. Calc. for $C_{34}H_{46}N_4O_4Cu$: C, 64.00; H, 7.21; N, 8.89. Found: C, 64.15; H, 7.35; N, 8.69%.

Bis(metronidazole) tetrakis (μ-ibuprofenato) dicopper(II), [Cu$_2$(Ibup)$_4$(Mtnd)$_2$], (4). A solution of 0.387 g (2.26 mmol) metronidazole in 50 mL hot absolute ethanol was added to 0.523 g (0.552 mmol) of $Cu_2(Ibup)_4$. The greenish blue solution was filtered and left in the hood of evaporate. The green precipitate that formed was recrystallized from chloroform and air dried. Anal. Calc. for $C_{64}H_{86}N_6O_{14}Cu_2$: C, 59.85, H, 6.67; N, 6.52. Found: C, 60.15; H, 6.77; N, 6.65%.

Bis(caffeine) tetrakis (μ-ibuprofenato) dicopper(II), [Cu$_2$(Ibup)$_4$(Cafn)$_2$], (5). 0.458 g (2.36 mmol) caffeine dissolved in 80 mL hot absolute ethanol was added to 0.545 g (0.576 mmol) $Cu_2(Ibup)_4$ in 20 mL CH_2Cl_2. The mixture was stirred at about 60°C for 3 hr. The greenish blue solution was filtered and left in the hood to evaporate. The bluish green precipitate that formed was recrystallized from $CHCl_3$ and the green precipitate was air dried. Anal. Calc. for $C_{68}H_{88}N_8O_{12}Cu_2$: C, 61.12; H, 6.59; N, 8.39. Found: C, 60.94; H, 6.60; N, 8.48%.

Physical Measurements

Magnetic susceptibility measurements at 298 K of powdered samples were determined by the Gouy method, with $HgCo(NCS)_4$ as calibrant, and corrected for diamagnetism with appropriate Pascal constants. The effective magnetic moment was calculated from the expression: $\mu_{eff} = 2.84\ (X.T)^{\frac{1}{2}}$.

Electronic spectra of methanol solutions were obtained with a Bauch and Lomb spectronic 2000. Nujol mulls sealed between a polyethylene sheet were used to obtain IR spectra of the complexes in the 4000 to 200 cm^{-1} region with a Perkin-Elmer model 843 infrared spectrophotometer. X-band ESR spectra of polycrystalline material and of methanol/toluene solutions were obtained at room temperature and 77 K with an ESPIT-330 VO1.501 spectrometer. Diphenylpicrylhydrazide (DPPH, g = 2.0036) was used as the calibrating field marker. The catecholase-mimetic activities in air were followed spectrophotometrically by monitoring the increase in the o-quinone absorbance at 390 nm as a function of time. 0.3 ml of methanol solutions (1×10^{-3} M) of the copper(II) complexes *2*, *3*, *4*, and *5* or methanol/CH_2Cl_2 (4:1) in the case of complex *1* (*1* is insoluble in methanol alone) and 2.0 mL of a methanol solution (0.1 M) catechol were combined in a 1 cm quartz cell at 298 K and the absorbance change at 390 nm is recorded.

RESULTS AND DISCUSSION

Magnetic and Spectroscopic Characterization

The effective magnetic moments and electronic and IR spectral data for the copper(II) complexes under investigation are summarized in Table 1. The

room temperature magnetic moments for complexes *2* and *3* are in the range 1.87–1.90 BM. These values are consistent with the presence of one unpaired electron in mononuclear copper(II) complexes. The room temperature magnetic moments for complexes *4* and *5* are in the range 1.42–1.45 BM. These subnormal values are lower than the spin-only magnetic moment of 1.73 BM, suggesting that coupling between copper atoms occurs. These values are comparable to the magnetic moment values of binuclear copper(II) carboxylate adducts of the type $[Cu(RCOO)_2L]_2$ [3, 4].

The electronic spectra for the mononuclear adducts *2* and *3* in methanol solutions exhibit one broad asymmetric band due to the copper(II) d-d transitions (Table 1). The position of the band and the value of its molar absorbitivity are in the range expected for mononuclear copper(II) carboxylate adducts [8, 9, 21–24]. The bis-adducts, $Cu(Ibup)_2(Im)_2$,*2* and $Cu(Ibup)(2\text{-}Mim)_2$,*3* have an absorption maximum which is comparable with that found for structurally known mononuclear $Cu(CH_3COO)_2(Im)_2$ and $Cu(CH_3COO)_2(2\text{-}Mim)_2$ [21, 22, 24], and other mononuclear copper(II) complexes that contain the *trans-* or *cis*-$CuN_2O_2\ldots O_2$ chromophore [4c, 8, 9, 21–24].

The electronic spectra for complexes *4* and *5* obtained in methanol solutions exhibit one broad band near 700 nm (Table 1). This band is assigned to the copper(II) d-d transitions. These complexes do not show the second band (shoulder) at about 375 nm, the charge transfer band that is characteristic of binuclear copper(II) adducts with bridging carboxylates [3, 4]. This band (shoulder) may be obscured by the very intense ligand-to-metal charge transfer band at about 300–330 nm due to the presence of metronidazole or caffeine ligands in these binuclear adducts. The position of the band of the copper(II) d-d transitions and the magnitude of its molar absorpitivity are comparable to those found for binuclear copper(II) carboxylate adducts having bridging carboxylate groups and axially coordinated amine ligands [4, 25].

The assignments of IR frequencies for the antisymmetric $\nu_{asym}(CO_2)$ and symmetric, $\nu_{sym}(CO_2)$, stretching vibrations for ibuprofenato group in these complexes are given in Table 1. The antisymmetric carboxyl vibration in complex *3* is not resolved but overlaps with the 2-methylimidazole band to give intense and broad absorption band centered at 1565 cm^{-1}. The positions of the $\nu_{asym}(CO_2)$ and $\nu_{sym}(CO_2)$ and the separation between them in complexes *2* and *3* are consistent with a carboxylate group that acts as an unsymmetrical

TABLE 1. Magnetic Moments and Electronic and IR Spectral and Kinetic Data for the Oxidation of Catechol by Cu(II) Complexes

Compound	μ_{eff} (BM) (298 K)	λ_{max} (nm) ($\epsilon = dm^3.mol^{-1}\ cm^{-1}$)	ν_{asym} (CO_2) (cm^{-1})	ν_{sym} (CO_2) (cm^{-1})	Activity[a]
1[b]	1.34	644	1588	1409	0.280
2	1.90	686 (82)	1595	1395	0.120
3	1.87	684 (75)	1565 (br)[c]	1400	0.065
4	1.45	700 (320)	1620	1408	0.250
5	1.42	690 (270)	1618	1410	0.220

[a]The activity is reported as micromoles of o-quinone produced per mg catalyst per min.
[b]Magnetic moment and spectral data are taken from Ref. [2].
[c]br is broad band overlaps with 2-methylimidazole bands.

bidentate ligand [21–24, 26]. They are comparable to those reported for mononuclear copper(II) carboxylate complexes that contain imidazoles having the $CuN_2O_2...O_2$ chromophore [9, 21–24]. The asymmetric $\nu_{asym}(CO_2)$ and the symmetric $\nu_{sym}(CO_2)$ frequencies for complexes *4* and *5* occur at about 1620 and 1410 cm^{-1}, respectively. The positions and the separation between these frequencies are similar to those reported for other binuclear copper(II) carboxylate adducts in which the carboxylate acts as a "bridging" bidentate ligand [3, 4b, 26].

The EPR parameters for the frozen solution and the polycrystalline forms of the complexes studied are summarized in Table 2. A representative frozen EPR spectrum of the mononuclear complexes is that of *2* shown in Figure 1(A). Frozen solution EPR spectra of mononuclear complexes *2* and *3* exhibit resolved structure with $g_{\|} > g_{\perp}$ and are consistent with a tetragonally structure [27]. While the $g_{\|}$ regions exhibit Cu hyperfine coupling, the $g_{\perp}$ regions show substantial ^{14}N-superhyperfine structure consisting of five equally-spaced peaks. The splitting in the $g_{\perp}$ region is attributed to the presence of two nitrogen atoms in the copper(II) ion plane. Spectral parameters for the mononuclear complexes *2* and *3* are comparable to those of previously reported complexes that contain essentially the CuN_2O_2 chromophore in a *trans* or *cis* square-planar arrangement, including those reported for mononuclear bis-adducts of copper(II) carboxylates with imidazoles [8, 9, 21–24]. These complexes contain a copper(II) atom in a *trans* or *cis* arrangement of two ligand nitrogen atoms and one oxygen atom from each of the two carboxylate ligands. The second oxygen atom of each carboxylate ligand is weakly bonded in a pseudo axial arrangement [21–23, 28]. Solid-state EPR spectra of *2* and *3* are anisotropic and contain $g_{\perp}$ and broad $g_{\|}$ components. The lack of copper(II) hyperfine coupling in these complexes is likely due to dipole-dipole interactions between copper(II) atoms of neighboring molecules.

The EPR spectra for the polycrystalline forms of the binuclear complexes *4* and *5* were recorded at room temperature. A representative spectrum is that of *5* shown in Figure 1(B). The spectra exhibit three absorption lines typical of triplet-state binuclear copper(II) complexes with axial symmetry [3, 4, 25]. The

TABLE 2. EPR data of Copper(II) Ibuprofenate Complexes

Compound	State (temperature)	g_o[a]	$g_{\|}$	$g_{\perp}$	$A_{\|}$ Cu ($\times 10^4$ cm^{-1})	$A_{\perp}$ N ($\times 10^4$ cm^{-1})	$\|D\|$ (cm^{-1})
2	Solid (Room)	2.138	2.278	2.069	—	—	—
	Frozen (77 K)	2.128	2.275	2.055	183	14.7	—
3	Solid (Room)	2.147	2.314	2.064	—	—	—
	Frozen (77 K)	2.142	2.311	2.058	173	14.0	—
4	Solid (Room)	2.217	2.424	2.114	—	—	0.368
5	Solid (Room)	2.211	2.419	2.109	—	—	0.370

[a] g_o Values are calculated from the equation $g_o = \frac{1}{3}(g_{\|} + 2g_{\perp})$.

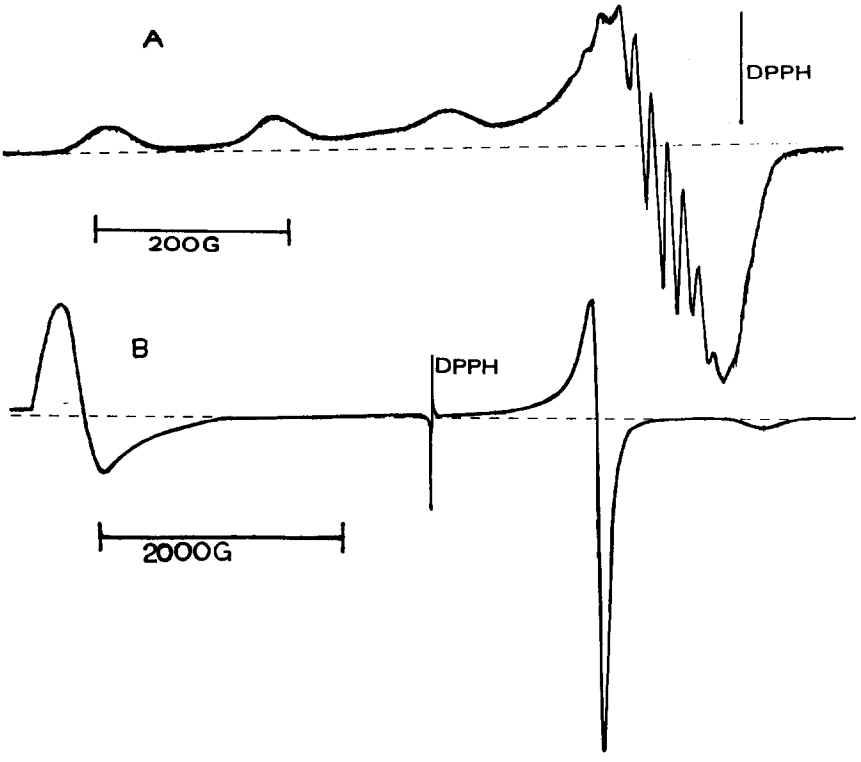

FIGURE 1. (A) Frozen-solution EPR spectrum of compound *2*. (B) solid-state EPR spectrum of compound *5* at room temperature.

EPR parameters were calculated by the method of Wasson et al. [29] by using the following equations [4b]:

$$h\nu = D - g_{\|}\beta H_1, \tag{1}$$

$$h\nu = -D + g_{\|}\beta H_2, \tag{2}$$

$$h\nu = -0.5D + (0.25D^2 + g_{\perp}^2\beta^2 H_3^2)^{\frac{1}{2}}, \tag{3}$$

where h is the Plank's constant, ν is microwave frequency, H_1, H_2 are the low and the high field parallels, respectively, H_3 is the perpendicular field, and D is the axial zero-field splitting. The parameters $g_{\|}$, $g_{\perp}$, and D values obtained for binuclear complexes *4* and *5* are comparable to those found in other binuclear copper(II) carboxylate adducts [4].

Catecholase-Mimetic Activity

Since o-quinone shows a characteristic absorption band at 390 nm, the catalytic oxidation of catechol to o-quinone by copper(II) complexes can be easily followed spectrophotometrically. The change in absorbance at 390 nm versus time for the first 15 minutes of the reaction with complexes *1–5* was obtained. The activities of these complexes were determined as micromoles of o-quinone

produced per mg of catalyst per minute. These values are shown in Table 1. Although o-quinone is produced for all complexes, the rate at which it is produced varies from binuclear to mononucler catalyst. The rate at which o-quinone is produced is also dependent on the nature of the N-containing ligand.

The relatively high catalytic activities of binuclear complexes *1*, *4*, and *5* compared to mononuclear complexes *2* and *3* (Table 1) is consistent with that previously studied which, generally, showed that binuclear copper(II) complexes exhibit catecholase-mimetic activities higher than those of mononuclear copper(II) complexes [30]. In tyrosinase and in synthetic copper(II) binuclear models, it is believed that two proximate metal atoms are needed to bond to the two hydroxyl oxygen atoms of catechols in the oxidation to quinones [14, 31]. In nonplanar mononucler copper(II) models, it has been proposed that the two copper(II) atoms must be located at a distance of less than 5 Å for bonding to the catechols hydroxyl groups, a mode which should facilitate electron transfer to dioxygen [19, 30a, 31]. The lower catalytic activities of metronidazole and caffeine adducts, *4* and *5*, respectively, compared to that of the binary complex *1* may be attributed to the presence of these axially coordinated ligands in the former adducts. The presence of these ligands could render the approach of catechol to copper(II) sites more difficult in these two binuclear adducts when compared to *1*. In addition, the axially coordinated ligands in *4* and *5* are likely to dissociate to provide sites on copper for catechol bonding and also to facilitate any necessary ligand rearrangement induced by this bonding. Such dissociation is not required in the binary complex *1* which is axially free. The relatively high catalytic activity of *4* compared to *5* may be due to easier dissociation of metronidazole ligands in *4* relative to caffeine ligands in *5*. The dissociation ability of metronidazole may be attributed to the presence of the electron withdrawing nitro group in this ligand. The catecholase-mimetic activity of the imidazole adduct *2* is higher than that of 2-methylimidazole adduct *3* (Table 1). This may be due to steric hindrance caused by the proximity of the methyl group to the nitrogen donor atom in *3*. The presence of a methyl group could render the approach of catechol to copper(II) sites more difficult in this adduct when compared to the imidazole adduct *2*. These results are consistent with our previous studies which revealed that the catecholase-mimetic activity of mononuclear copper(II) carboxylate adducts with benzimidazole or imidazole is higher than those of 2-methyl substituted derivatives [9, 21].

I am grateful to Professor Jan Reedijk and to Mr. G. A. Van Albada of Leiden University, The Netherlands, for obtaining the EPR spectra and to Birzeit University for the support of this research under Grant No. 86 / 68 / 97 / 8.

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Received September 9, 1993; accepted October 29, 1993