

Structures and Catalytic Activities of Carboxylate-Bridged Dinickel(II) Complexes as Models for the Metal Center of Urease

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Urease, a Ni(II)-containing enzyme isolated from various bacteria and plants hydrolyzes urea to form carbamate and ammonia: the former compound spontaneously decomposes to yield carbonic acid and another ammonia.¹ The recent paper by Park and Hausinger has described that carbon dioxide is required for the activation of apo-urease in the presence of Ni(II) ion.² Carbon dioxide reacts with the ϵ -amino group of Lys at the active site to form a carbamate group. The X-ray crystal structure analysis of a bacterial urease demonstrated the presence of a binuclear Ni(II) active site: two Ni ions are 3.5 Å apart and bridged by a carbamylated Lys residue.³ One of the Ni ions has two His nitrogens, one oxygen of bridging carbamylated Lys, and one water oxygen. The geometry at the Ni site is pseudotetrahedral. The pentacoordinate sphere of the second Ni atom is approximately trigonal bipyramidal. The ligand atoms comprise one oxygen of the bridging residue, two His nitrogens, one carboxylate oxygen of Asp, and one water oxygen. The relationship between the structure and function of the dinickel active site has not been substantiated, although there are many studies of urease¹ and model dinickel complexes.⁴ Moreover, it is important to shed light on the activation of apo-urease by carbon dioxide and a role of the carbamylate bridge on the catalytic reaction of urease. We report herein the structures and the catalytic features of carboxylate-bridged dinickel(II) complexes which are able to catalyze the ethanolysis of urea to form ethyl carbamate. Although many dinickel complexes containing carboxylate chelate ligand(s) have previously reported as structural models for urease,⁵ their urease-like activities have not been investigated.

$[\text{Ni}_2(\text{Me}_4\text{-tpdp})(\text{CH}_3\text{CO}_2)(\text{ClO}_4)(\text{CH}_3\text{OH})]\text{ClO}_4$ (**1**) is prepared by the reaction of 2 equiv of Ni(CH₃CO₂)₂·4H₂O and NaClO₄ with 1 equiv of Me₄-Htpdp⁶ in methanol at room temperature, being recrystallized from a methanol/diethyl ether solution as light green crystals. As shown in Figure 1a, the

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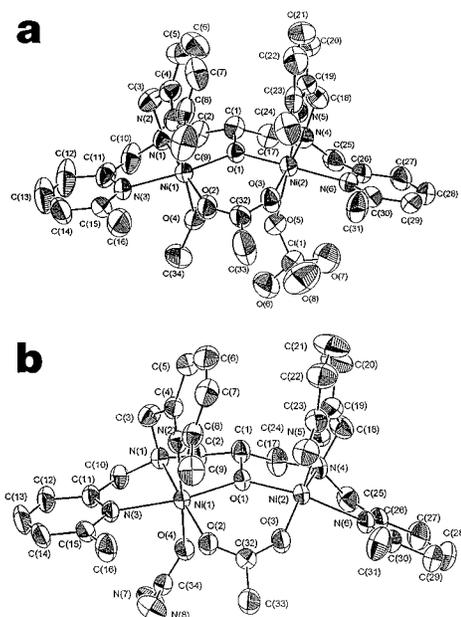


Figure 1. ORTEP drawings of $[\text{Ni}_2(\text{Me}_4\text{-tpdp})(\text{CH}_3\text{CO}_2)(\text{ClO}_4)(\text{CH}_3\text{OH})]^+$ (a) and $[\text{Ni}_2(\text{Me}_4\text{-tpdp})(\text{CH}_3\text{CO}_2)(\text{urea})]^{2+}$ (b) showing thermal ellipsoids and atom labels. Hydrogen atoms are omitted for clarity. Selected distances (Å) and angles (deg) for **1** are as follows: Ni(1)···Ni(2), 3.622(1); Ni(1)–O(1), 1.990(3); Ni(1)–O(2), 1.991(3); Ni(1)–O(4), 2.174(4); Ni(1)–N(1), 2.075(4); Ni(1)–N(2), 2.188(4); Ni(1)–N(3), 2.169(4); Ni(2)–O(1), 1.982(3); Ni(2)–O(3), 1.969(3); Ni(2)–O(5), 2.506(4); Ni(2)–N(4), 2.058(4); Ni(2)–N(5), 2.122(4); Ni(2)–N(6), 2.168(4); O(1)–Ni(1)–O(2), 92.7(2); O(1)–Ni(1)–O(4), 85.4(2); O(1)–Ni(1)–N(2), 90.0(2); O(1)–Ni(1)–N(3), 160.4(2); O(2)–Ni(1)–N(1), 174.4(2); O(4)–Ni(1)–N(2), 171.0(2); O(1)–Ni(2)–O(3), 93.7(2); O(1)–Ni(2)–O(5), 81.9(2); O(1)–Ni(2)–N(5), 94.2(2); O(1)–Ni(2)–N(6), 160.2(2); O(3)–Ni(2)–N(4), 170.3(2); O(5)–Ni(2)–N(5), 170.6(2). Selected distances (Å) and angles (deg) for **2** are as follows: Ni(1)···Ni(2), 3.647(1); Ni(1)–O(1), 2.019(2); Ni(1)–O(2), 2.016(2); Ni(1)–O(4), 2.134(2); Ni(1)–N(1), 2.071(2); Ni(1)–N(2), 2.183(2); Ni(1)–N(3), 2.183(2); Ni(2)–O(1), 1.954(2); Ni(2)–O(3), 1.971(2); Ni(2)–N(4), 2.072(2); Ni(2)–N(5), 2.062(2); Ni(2)–N(6), 2.148(3); O(1)–Ni(1)–O(2), 92.9(1); O(1)–Ni(1)–O(4), 88.0(1); O(1)–Ni(1)–N(2), 89.0(1); O(1)–Ni(1)–N(3), 162.2(1); O(2)–Ni(1)–N(1), 173.3(1); O(4)–Ni(1)–N(2), 170.5(1); O(1)–Ni(2)–O(3), 92.1(1); O(1)–Ni(2)–N(5), 99.7(1); O(1)–Ni(2)–N(6), 155.7(1); O(3)–Ni(2)–N(4), 155.3(1). Numbers in parentheses are estimated standard deviations in the last digit.

X-ray crystal structure⁷ of **1** reveals that the two hexacoordinate Ni(II) ions bridged by alkoxide and acetate anions are 3.62 Å apart. This dinickel site is comparable with that in urease (3.5 Å). Both of the geometries of Ni(1) and Ni(2) bound the oxygen atoms of methanol and perchlorate at the sixth coordination site, respectively, are octahedral. The average value of the Ni–N,O distances is 2.11 Å.

When 2 equiv of urea is added to the acetone solution of **1** at -80°C , the absorption spectrum is changed (data not shown), suggesting the formation of a urea adduct. From the reaction mixture, the green complex **2** is isolated and recrystallized from an acetone/diethyl ether solution at -80°C . The X-ray crystal structure analysis of **2** reveals that only Ni(1) binds to a urea

(6) Me₄-Htpdp is *N,N,N',N'*-tetrakis[6-(methyl-2-pyridyl)methyl]-1,3-diaminopropan-2-ol (Hayashi, Y.; Kayatani, T.; Sugimoto, H.; Suzuki, M.; Inomata, K.; Uehara, A.; Mizutani, Y.; Kitagawa, T.; Maeda, Y. *J. Am. Chem. Soc.* **1995**, *117*, 11220–11229).

(7) Anal. Calcd for C₃₄H₄₄N₆O₁₂Cl₂Ni₂ (**1**, $[\text{Ni}_2(\text{Me}_4\text{-tpdp})(\text{CH}_3\text{CO}_2)(\text{ClO}_4)(\text{CH}_3\text{OH})]\text{ClO}_4$): C, 44.52; H, 4.85; N, 9.17. Found: C, 44.30; H, 4.85; N, 9.14. Crystal data for **1**: $M_r = 939.00$, monoclinic, $P2_1/c$, $a = 17.707(6)$ Å, $b = 13.780(5)$ Å, $c = 16.267(8)$ Å, $\beta = 94.72(3)^\circ$, $V = 3955.9(26)$ Å³, $Z = 4$; for 5072 unique observed reflections collected with $F > 3\sigma(F)$, $R = 0.0696$, and $R_w = 0.0690$.

molecule through its oxygen atom, while Ni(2) has no ligand at the sixth coordination site and is pentacoordinate (Figure 1b).⁸ The structures of **1** and **2** suggest that these complexes are useful models for an intermediate in the catalytic cycle in urease, that is, one Ni activates the carbonyl carbon of urea and the other one has a vacant coordination site available to generate a nucleophile (hydroxide in urease) which is sterically in a suitable position to attack the carbonyl carbon of the coordinated urea, as proposed in the mechanism of urease by some investigators.^{1d,9,10}

As a model reaction of urease, we have examined ethanolysis of urea with **1**. An ethanol solution containing **1** (24 mM) and 200 equiv of urea is stirred at 80 °C.¹¹ In the presence of **1**, urea gives rise to ethyl carbamate by ethanolysis, whereas no reaction occurs in the absence of the dinickel complex and even in the presence of nickel(II) ion or the ligand only. Moreover, urea is hardly converted into ethyl carbamate in a ethanol solution containing 1:1 nickel(II) ion/a tridentate ligand (bis-[(6-methyl-2-pyridyl)methyl]amine), and hence, a binuclear nickel center is essential for the ethanolysis. When the reactions are carried out for 2, 6, and 12 h, the yields of the product (ethyl carbamate) are 96, 229, and 409% vs the complex, respectively. Complex **1** obviously acts as a catalyst in the ethanolysis.

The hydroxo-bridged dinickel complex, [Ni₂(Me₄-tpdp)(OH)]-(ClO₄)₂·acetone (**3**),¹² is obtained by the reaction of Me₄-Htpdp with 2 equiv of triethylamine and Ni(ClO₄)₂·6H₂O in an acetone solution under N₂ atmosphere. Complex **3** does not catalyze the ethanolysis of urea.¹³ When the acetone solution is exposed to carbon dioxide, a spectral change of **3** is observed (Figure 2). The absorption spectrum of the reaction solution is very similar to that of **1** (Figure 2, inset). The resulting complex **4** is isolated from acetone/hexane mixture, and its IR spectrum (nujol) displays a new peak at 1532 cm⁻¹. According to the elemental analysis¹⁴ and visible absorption and IR spectroscopies, **4** is characterized as a dinickel complex bridged by

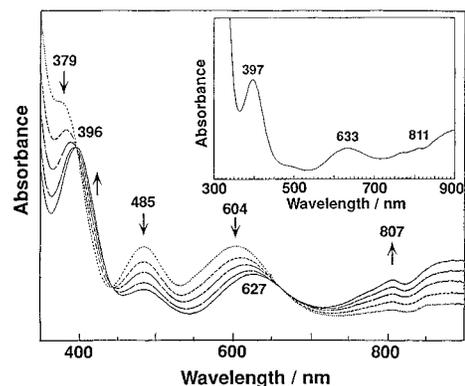


Figure 2. Time course of the conversion of **3** to **4** under CO₂ atmosphere in acetone at 23 °C. The spectra were recorded at 30 s intervals. The inset shows the absorption spectrum of **1** in acetone.

bidentate hydrogencarbonate (HCO₃⁻).¹⁵ The ethanolysis of urea is catalyzed by **4**. After the reactions for 2, 6, and 12 h, the amount of ethyl carbamate as the product is 48, 84, and 205% vs the complex, respectively. It is noted that **3** has no reactivity for the ethanolysis of urea, but it hydrates carbon dioxide to form **4** which exhibits the ethanolysis activity of urea. This finding resembles the activation of apo-urease in the presence of Ni ions by carbon dioxide.² The bridging ligands such as acetate and hydrogencarbonate play an essential role for the ethanolysis of urea. Possible roles of these bridging groups would control the acidity of Ni ions for the activation of both urea and ethanol and the stereochemical arrangement of the two molecule for the ethanolysis.

In conclusion, we have demonstrated that the RCO₂⁻-bridged dinickel complexes (R = CH₃- or HO-) containing Me₄-tpdp catalyze the ethanolysis of urea. The X-ray crystal structure analyses of the μ -acetato Ni(II) complexes have shown that one of the two Ni ions is capable of binding urea and the another one would activate ethanol by coordination. Furthermore, the RCO₂⁻-bridging ligands between two Ni ions are important to the catalytic reaction. The structures and catalytic reactions of the dinickel model complexes show a striking resemblance to those of the dinickel site in urease. They would be also helpful in an understanding of the requirement of CO₂ for the reconstitution of apo-urease with Ni(II) ions.

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Supporting Information Available: Crystallographic details in CIF format (40 pages). See any current masthead page for ordering and Internet access instructions.

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(15) The coordination of HCO₃⁻ ion as a bridging ligand has been confirmed in a binuclear zinc(II) complex containing Me₄-tpdp by X-ray crystal structure analysis (to be submitted).

(8) Crystal data for [Ni₂(Me₄-tpdp)(CH₃CO₂)(urea)](ClO₄)₂·urea (**2**): *M*_r = 981.1, triclinic, *P*1, *a* = 13.971(7) Å, *b* = 14.474(5) Å, *c* = 12.479(6) Å, α = 93.47(3)°, β = 116.46(4)°, γ = 92.49(4)°, *V* = 2247.8(16) Å³, *Z* = 2; for 7280 unique observed reflections collected with *F* > 3 σ (*F*), *R* = 0.0604, and *R*_w = 0.0641.

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(11) The reaction mixtures were analyzed by a Shimadzu GCMS2200 GC-MS spectrometer equipped with a nonpolar C-18 column (15 m). The mass peaks of ethyl carbamate (89, 74, 62, 44) and urea (60, 44) were observed at 8.28 and 17.7 min, respectively. The column temperature for the analysis was maintained at 40 °C during the first 10 min and then raised to 240 °C for 10 min. The amounts of ethyl carbamate and urea were estimated by integration of the mass peaks of 62 (ethyl carbamate) and 60 (urea).

(12) The X-ray crystal structure analysis of **3** has been carried out. Two Ni(II) sites are pentacoordinate (unpublished results).

(13) The tdpdp ligand without four methyl groups has given rise to the mononuclear Ni(tpdp)ClO₄ complex, which shows no ethanolysis of urea. It is important that there is the cooperative effect between the two Ni(II) centers for the catalytic activity.

(14) Anal. Calcd for C₃₂H₄₂N₆O₁₄Cl₂Ni₂ (**4**, [Ni₂(Me₄-tpdp)(HCO₃)](ClO₄)₂·2H₂O): C, 41.63; H, 4.60; N, 9.11. Found: C, 41.20; H, 4.70; N, 9.10.