Synthesis, Structural Characterization, and Biological Activity of Two Different Nickel(II) Complexes Derived from *N'*-[1-(2-pyridyl)ethylidene]morpholine-4-carbothiohydrazide

Noriko Chikaraishi Kasuga, Atsushi Ohashi, Chisa Koumo, Jun Uesugi, Munehiro Oda,† and Kenji Nomiya*
Department of Materials Science, Faculty of Science, Kanagawa University, Tsuchiya, Hiratsuka, Kanagawa 259-12
†Meiji Cell Technology Center, Meiji Milk Products Co., Ltd. 540, Naruda, Odawara, Kanagawa 250

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The reaction of $Ni(OAc)_2$ with *N'*-[1-(2-pyridyl) ethylidene]morpholine-4-carbothiohydrazide (HL) afforded two nickel(II) complexes, $[Ni(L)_2]$ (**1**) and [Ni(L)(OAc)] (**2**), depending on reaction temperature and starting molar ratio; the former was a 6-coordinate, paramagnetic complex and the latter a 4-coordinate, diamagnetic complex. X-ray analysis of the complex **2** revealed that the ligands, L and OAc, were coordinated to the central metal ion to form a square-planar geometry. The complex **2** showed enhanced antimicrobial activities against selected Gram-positive bacteria, compared with those of the ligand alone, whereas the other complex **1** showed no activity.

Heterocyclic thiosemicarbazones, as well as their metal complexes, are an important series of compounds as they possess potentially beneficial, biological activities, such as antitumor, antibacterial, antiviral, antifungal, and antimalarial properties.[1] The thiosemicarbazones are also versatile ligands which can coordinate to the metal either as a neutral tridentate ligand HL or as a deprotonated ligand L through the sulfur and two nitrogen atoms.[2] In our continuing studies of the relationship between the biological properties and structures of metal complexes, we found that two novel nickel(II) complexes **1** and **2** were formed from the same starting materials: $Ni(OAc)_2$ and HL. These two nickel(II) chelates with a common thiosemicarbazone ligand gave more informative views to such a relationship, compared with the studies of the previously reported, nickel(II) complexes, [3] by the unequivocal characterization and the biological test using the wide range of organisms in this work. Herein we report the synthesis of these nickel(II) complexes and the characterization by elemental analysis, solid FT-IR, magnetic susceptibility measurements, UV-VIS absorption spectra and NMR (^1H and ^{13}C) measurements. We describe the molecular structure of **2** determined with a single-crystal X-ray analysis. Also reported are antimicrobial activities of the two nickel(II) complexes and those of the ligand alone, evaluated by MIC (minimum inhibitory concentration: μg/mL), against the wide range of organisms.

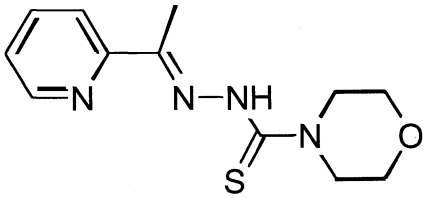

Figure 1. *N'*-[1-(2-pyridyl)ethylidene]morpholine -4-carbothiohydrazide (HL).

The ligand, HL, was prepared according to the literature.[4] To a solution of 1.24 g (5 mmol) of $[Ni(OAc)_2]\cdot 4H_2O$ dissolved in 30 mL ethanol was added 2.64 g (10 mmol) of the ligand (HL) in 30 mL $CHCl_3$ with stirring for 3 h at room temperature. From the mixture, ocher powder **1** was obtained by filtration through a membrane filter (0.2 μm) in 2.22 g yield (76%).[5] The complex **1** was readily soluble in $CHCl_3$, soluble in DMSO, slightly soluble in Et_2O, MeOH, CH_3CN and H_2O. The magnetic susceptibility at 23 °C measured by Gouy method showed μ_{eff} = 3.02 BM, indicating that **1** was paramagnetic due to two unpaired electrons. The elemental analysis of **1** revealed the composition with Ni^{II} : L = 1 : 2 ratio. These results exhibit a 6-coordinate, octahedral geometry for **1**, $[Ni(L)_2]$.

To a solution of 1.24 g (5 mmol) of $[Ni(OAc)_2]\cdot 4H_2O$ dissolved in 30 mL ethanol was added 1.32 g (5 mmol) of HL in 30 mL $CHCl_3$. The mixture was refluxed for 3 h, then cooled to room temperature, followed by filtering through a membrane filter

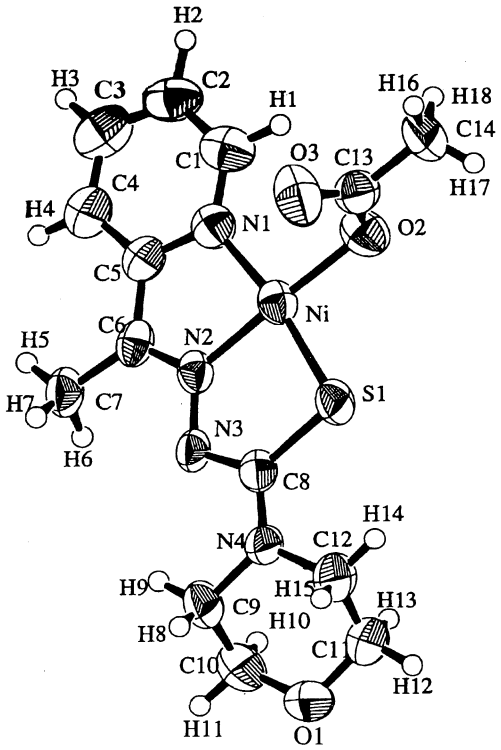

Figure 2. Molecular structure of complex **2**. Solvated molecule is omitted for clarity. (thermal ellipsoids at 50% probability level). The selected bond distances(Å) and angles(°) are follows; Ni-O2 1.903(4), Ni-N1 1.913(5), Ni-N2 1.845(4), Ni-S1 2.151(2), C5-C6 1.458(8), C6-C7 1.492(7), C6-N2 1.314(6), N2-N3 1.357(6), N3-C8 1.322(6), C8-S1 1.758(6), C8-N4 1.341(7), O2-C13 1.257(7), O3-C13 1.239(7), C13-C14 1.496(8). O2-Ni-N1 96.0(2), O2-Ni-S1 93.0(1), N1-Ni-N2 84.2(2), N2-Ni-S1 86.7(2), C5-C6-N2 112.9(6), C5-C6-C7 123.6(6), C6-N2-N3 118.0(5), N2-N3-C8 110.9(5), N3-C8-S1 122.2(5), N3-C8-N4 118.8(5).

Table 1. Antimicrobial activities of nickel complexes and ligand evaluated by MIC[a]

Test organisms	MIC		
	HL	1	2
Staphylococcus aureus	250	>1000	31.3
Bacillus subtilis	125	1000	31.3
Pseudomonas aeruginosa	125	>1000	>1000
Escherichia coli	250	>1000	1000.
Saccharomyces cerevisiae	31.3	>1000	500.
Candida albicans	31.3	>1000	1000.
Aspergillus niger	125	>1000	>1000.
Penicillium citrinum	125	>1000	>1000.

[a] Minimum inhibitory concentration (μg/mL).

(0.2 μm). The clear filtrate was slowly evaporated. About a few weeks later, dark-red crystals **2**[6] suitable for X-ray analysis were obtained in 0.97 g yield (51%). From the starting solution without refluxing, **1** was obtained in 0.75 g (26%) yield, and from its filtrate **2** was isolated in 0.42 g (22%) yield. Thus, the formation of **1** and **2** is strongly depending on the molar ratio and the reaction temperature, respectively. The complex **2** was readily soluble in $CHCl_3$, slightly soluble in DMSO, MeOH, CH_3CN, and H_2O, but insoluble in Et_2O. In contrast to **1**, the μ_{eff} value for **2** at 23 °C estimated by Gouy method was 0.26 BM, *i.e.*, it was diamagnetic. The electronic absorption spectrum in visible region for **2** in $CHCl_3$ showed one weak and two intense bands,[6] which correspond to those of other planar nickel(II) complexes. ^1H and ^{13}C NMR spectra for **2** also supported it was diamagnetic. In ^1H NMR spectrum, the signal from the ethanol used in the preparation was also observed. The solution for ^1H NMR measurement, prepared using the crystals standing at room temperature for several months, showed much smaller intensity of EtOH signals. This indicated that the ethanol solvated in the crystal was volatile. The full elemental analysis of **2** revealed the composition with L : OAc : Ni^{II} = 1 : 1 : 1 ratio.

The crystal structure of **2** was determined with X-ray analysis.[7] The molecular structure of **2** with partial atoms labeling scheme is shown in Figure 2. The deprotonated thiosemicarbazone as a tridentate ligand coordinates to nickel(II) via the pyridyl nitrogen N1, the azomethine nitrogen N2, and the thiolato sulfur atom. From the bond angles around nickel(II), the molecular structure of **2** is described as square planar geometry, consistent with the magnetic susceptibility measurements. The distances of Ni-S, Ni-N, and Ni-O are comparable to those of nickel compounds derived from other thiosemicarbazones.[3] In comparison with bond distances of 2-acetylpyridine N4-substituted thiosemicarbazones ligands:[3a] C5-C6 in the complex **2** is shorter (1.458(8) vs 1.48), C6-N2 is longer (1.314(6) vs 1.27), N2-N3 is shorter (1.357(6) vs 1.36) , C8-N4 is shorter (1.341(7) vs 1.46).

Antimicrobial activities (Table 1) of the ligand and the two nickel complexes were estimated by MIC, as usual.[8] The complex **1** did not inhibit the growth of the test organisms. On the contrary, **2** showed enhanced activites against selected Gram-positive bacteria (*S. aureus* and *B. subtilis*), compared with those of the ligand alone. The inhibition by **2** against Gram-negative bacteria (*E. coli* and *P. aeruginosa*), yeasts (*S. cerevisiae* and *C. albicans*), and molds (*A. niger* and *P. citrinum*) was not high. In antifungal studies with ^{4}N-dialkyl thiosemicarbazones, octahedral complexes did not show higher activities than those of planar complexes.[9] Similar tendency was seen in our antimicrobial studies with **1** and **2**. The differences observed in antimicrobial activities of **1** and **2** should reflect the difference in their molecular structure.

In conclusion, the reaction of $Ni(OAc)_2$ with HL afforded two different chelates, **1** and **2**, depending on the reaction temperature and the starting molar ratio. The 4-coordinate, planar complex **2** inhibited growth of selected Gram-positive bacteria, but the 6-coordinate, octahedral complex **1** did not. These antimicrobial activities are significantly influenced by the molecular structure of the complex, despite the fact that the two chelate compounds comprise the common thiosemicarbazone ligand.

References and Notes

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5 **1**; Anal. Found: C, 49.09; H, 5.04; N, 18.94%. Calcd for $\{C_{24}H_{30}N_8O_2S_2Ni\}$: C, 49.24; H, 5.17; N, 19.14%. IR(KBr disk): 1414(s), 1298(s), 1255(s), 1227(m), 1113(m) cm^{-1}. UV-VIS ($CHCl_3$) λ_{max} with ε in parenthesis: 242(33300), 357(33700), and 428(34600) nm ($mol^{-1}dm^3cm^{-1}$).

6 **2**; Anal. Found: C, 44.32; H, 5.26; N, 13.8; O, 13.5; S, 8.12; Ni, 14.7%. Calcd for $\{(C_{14}H_{18}N_4O_3SNi)\cdot 0.25\ (C_2H_6O)\}$: C, 44.36; H, 5.01; N, 14.27; O, 13.24; S, 8.17; Ni, 14.95% (Mikroanalytisches Labor Pascher). IR (KBr): 1632(s), 1499(s), 1469(s), 1432(s), 1359(m), 1315(s), 1267(s), 1240(s), 1158(m), 1109(m) cm^{-1}. UV-VIS ($CHCl_3$) λ_{max} with ε in parenthesis: 243(22400), 338(15000), 400(20400), and 523(1100) nm ($mol^{-1}dm^3cm^{-1}$). ^1H NMR (400MHz, $CDCl_3$ at 23 °C): 1.94(3H, s, CH_3CO_2), 2.10(3H, s, CH_3C=N), 3.70(4H, m, OCH_2), 3.78(4H, m, NCH_2), 7.22(1H, d, H4) 7.28(1H, t, H3), 7.81(1H, d, H1), 7.84(1H, t, H2) ppm: ^{13}C NMR (100MHz, $CDCl_3$ at 23 °C): 13.04(C7), 23.72(C14), 48.48(C9, C12), 66.51(C10, C11), 120.96 (C4), 124.47(C3), 139.87(C2), 147.97(C1), 152.32(C5), 156.42 (C6), 177.91(C13), 180.45(C8) ppm. These assingments were done using DIFNOE, CH-COSY, and HMBC NMR.

7 Crystal data of **2**: $C_{14}H_{18}O_4N_4SNi\cdot 0.6C_2H_5OH$. M = 394.72, triclinic, a = 8.854(6)Å, b = 11.985(5)Å, c = 8.833(2)Å, α = 105.32(3)°, β = 93.04(4)°, γ = 75.56(5)°, V = 875.4(7)Å^3, space group P1(#2), Z = 2, F(000) = 413.2, MoKα radiation, room temperature. d_{calc} = 1.50 $g\cdot cm^{-3}$. μ = 12.39 cm^{-1}. A red plate crystal was sealed in a glass capillary. All crystallographic measurements were made using RIGAKU AFC5S diffractometer. The number of solvated molecule was determined with ^1H NMR. The solvated molecule was highly disordered. Empirical absorption correction (PSI scan) was applied (trans. factor 0.826 - 0.999). Data were measured in the range of 6<2θ<55° in the ω-2θ scan. The structure was solved by direct method and refined by full-matrix least-squares using 1930 reflections (I>3σ(I)) among 4036 unique reflections. Final R and R_w are 0.050 and 0.041, respectively. Non-hydrogen atoms were refined with anisotropic thermal factors. Hydrogen atoms derived calculated geometrically (C-H 0.95Å) were refined with isotropic temperature factor.

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