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# Conformation and Coordination Behavior of Zinc(II) Complex with Cyclo(L-methionyl-L-histidyl)

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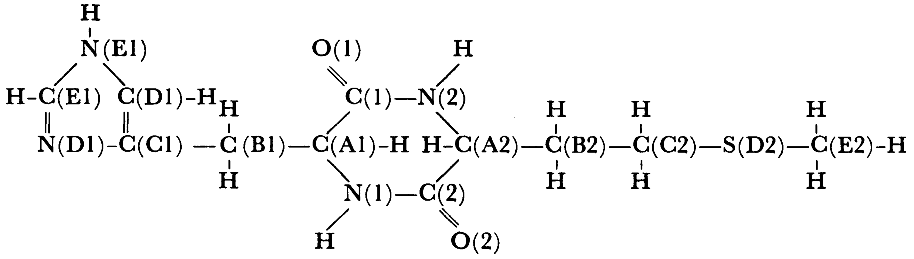
The crystal structure of a zinc(II) complex with cyclo(L-methionyl-L-histidyl) shows that all four ligands coordinated tetrahedrally to a $Zn^{2+}$ ion have a folded conformation, with the imidazole ring facing the piperazine ring in each ligand. The calculation from $\alpha$-$\beta$ coupling constant data (obtained by $^1H$ NMR measurement) of the histidyl residue of cyclo(L-methionyl-L-histidyl) linked to a $Zn^{2+}$ ion shows that the ratio of a folded conformation to an unfolded one is 61:39 in $D_2O$.

Several metal complexes with cyclic dipeptides[1)] have been used as simple model compounds for studying metallo-proteins and -enzymes, in which metal ions link to the functional groups in the side chains of the peptides, but not to the amide groups in the main chain. Especially, the limited conformations[2)] of the peptides with the imidazole rings show the important effects on the formations and conformations of the metal complexes.

In this paper, the conformation and coordination behavior of the $[Zn(CMH)_4]^{2+}$ [abbreviated as CMH for cyclo(L-methionyl-L-histidyl)] ion are examined using X-ray structure analysis, NMR ($^1H$ and $^{13}C$) and molecular-weight measurements. A preliminary communication on the structure of this zinc(II) complex with CMH has already been published.[1b)] The numbering scheme of a CMH moiety is given below:



## Experimental

*Materials.* CMH was prepared according to the methods reported earlier.[1a)] Zinc(II) sulfate (obtained commercially) was used without further purification.

*Preparations of Zinc(II) Complexes.* A mixture of CMH (1.40 mmol) and zinc(II) sulfate (0.35 mmol) was completely dissolved in hot water (5 ml) with stirring. After standing at room temperature for a few days, a formed precipitate was filtered off. mp; 232—234°dec., Yield; 63%. Found: C, 41.51; H, 5.22; N, 17.49%. Calcd for $Zn(C_{11}H_{16}N_4O_2S)_4SO_4\cdot 2H_2O$: C, 41.65; H, 5.24; N, 17.66%.

*X-Ray Measurement.* X-Ray analysis: $Zn(C_{11}H_{16}N_4O_2S)_4\cdot SO_4\cdot 10H_2O$, $M$=1414.9, monoclinic, space group $P2_1$, $a$=15.143(2), $b$=20.586(4), $c$=11.007(2) Å, $\beta$=97.89(1)°. $Z$=2. With the use of 2692 uniquely observed reflections, the structure was solved by MULTAN[3a)] and refined anisotropically to $R$=0.062 ($R_w$=0.071).[3b)] The absolute configuration was determined by reference to the known configuration of L-histidine moiety. Fractional atomic coordinates and $B_{eq}$ values are listed in Table 1. Tables of observed and calculated structure factors, bond distances and angles, anisotropic temperature factors, atomic parameters of hydrogen atoms, and hydrogen bonds are kept at the chemical society of Japan. (Document No. 8526)

*Other Measurements.* $^1H$ and $^{13}C$ NMR data were obtained in $D_2O$ at 50°, using sodium 3-trimethylsilyl-1-propanesulfonate as an internal reference. The $^1H$ NMR investigation of CMH were carried out with the aid of spin decoupling methods. The amide protons were replaced by deuterium in $D_2O$. The molecular weights of zinc(II) complexes with CMH were measured in $H_2O$ at 42°, using urea as a standard material. A JEOL FX-100 (for $^1H$ and $^{13}C$ NMR spectra), a PS-100 (for $^1H$ NMR spectra), and a Knauer Vapor-Pressure Osmometer (for molecular weight) were used for the measurements of the samples.

## Results and Discussion

A $Zn^{2+}$ ion is coordinated tetrahedrally by four N (E1) atoms of the imidazolyl groups of the histidyl residues of peptides exclusively (see Fig. 1). The N-Zn-N angles are all within 3.5° from 109.5° and the average distance of four independent Zn-N bonds is 2.01(1) Å, which is comparable to the values found in other tetrahedral complexes.[4] The complex ion, except for the terminal -$CH_2SCH_3$ groups, has an approximate $D_2$ symmetry since three pseudo two-fold axes bisect the N-Zn-N angles.

Selected dihedral angles of the CMH moieties of the complex and the free CMH molecule are given in Table 2.[5] The orientation of imidazole rings to the piperazine rings is determined by the dihedral angles around C(A1)-C(B1) and C(B1)-C(C1) bonds. The values of $\chi_1^1$ and $\chi_1^2$ show that all CMH moieties have a same conformation concerning the piperazine and imidazole rings. The imidazole rings are folded back over the piperazine rings to have the intimate contacts between these two rings. Recent X-ray crystallographic studies of cyclic dipeptides such as cyclo(L-histidyl-L-histidyl),[6] CMH,[5] and cyclo(L-

TABLE 1. FRACTIONAL ATOMIC COORDINATES AND ISOTROPIC TEMPERATURE FACTORS FOR NON-HYDROGEN ATOMS WITH ESTIMATED STANDARD DEVIATIONS IN PARENTHESES[a)]

| Atom | $x$ | $y$ | $z$ | $B_{eq}$ | Atom | $x$ | $y$ | $z$ | $B_{eq}$ |
|---|---|---|---|---|---|---|---|---|---|
| Zn | 0.1869(1) | 0.5000(1) | 0.0302(2) | 2.34(5) | C(B1)C | 0.437(1) | 0.3002(8) | 0.104(2) | 3.1(6) |
| S(D2)A | 0.2378(4) | 0.3869(4) | −0.3291(8) | 8.3(3) | C(C1)C | 0.366(1) | 0.3488(8) | 0.117(2) | 3.5(6) |
| N(1)A | 0.4501(9) | 0.6011(7) | −0.384(1) | 3.6(5) | C(D1)C | 0.329(1) | 0.3977(9) | 0.045(2) | 3.4(6) |
| N(D1)A | 0.2809(9) | 0.6108(6) | −0.235(1) | 2.8(4) | C(E1)C | 0.268(1) | 0.4001(9) | 0.204(2) | 3.6(6) |
| N(E1)A | 0.2530(8) | 0.5489(7) | −0.082(1) | 2.7(4) | C(2)C | 0.577(1) | 0.4124(8) | 0.302(2) | 2.7(5) |
| N(2)A | 0.5181(9) | 0.4892(7) | −0.266(1) | 3.0(4) | C(A2)C | 0.585(1) | 0.4658(8) | 0.210(2) | 2.5(5) |
| C(1)A | 0.533(1) | 0.5436(8) | −0.206(2) | 3.1(6) | C(B2)C | 0.528(1) | 0.5229(8) | 0.237(2) | 3.8(6) |
| C(A1)A | 0.500(1) | 0.6092(9) | −0.258(2) | 3.3(6) | C(C2)C | 0.576(2) | 0.570(2) | 0.303(4) | 15. (2) |
| C(B1)A | 0.442(1) | 0.6443(9) | −0.176(2) | 3.3(6) | C(E2)C | 0.650(2) | 0.657(1) | 0.145(2) | 8. (1) |
| C(C1)A | 0.358(1) | 0.6083(8) | −0.152(2) | 2.4(5) | O(1)C | 0.5305(8) | 0.3777(6) | −0.071(1) | 4.5(4) |
| C(D1)A | 0.341(1) | 0.5708(9) | −0.055(2) | 2.9(5) | O(2)C | 0.6011(8) | 0.4214(6) | 0.411(1) | 4.8(4) |
| C(E1)A | 0.221(1) | 0.5766(9) | −0.188(1) | 3.1(5) | S(D2)D | 0.0392(4) | 0.3479(3) | 0.2761(5) | 4.6(2) |
| C(2)A | 0.430(1) | 0.5463(9) | −0.446(2) | 3.9(6) | N(1)D | 0.0133(9) | 0.6276(7) | 0.503(1) | 3.2(4) |
| C(A2)A | 0.469(1) | 0.4808(9) | −0.388(2) | 4.4(7) | N(D1)D | 0.1835(9) | 0.6136(7) | 0.336(1) | 3.3(5) |
| C(B2)A | 0.403(1) | 0.428(1) | −0.395(2) | 6.9(9) | N(E1)D | 0.1607(8) | 0.5572(7) | 0.172(1) | 2.8(4) |
| C(C2)A | 0.333(2) | 0.444(1) | −0.320(2) | 6.3(8) | N(2)D | −0.0931(8) | 0.5515(7) | 0.335(1) | 3.0(4) |
| C(E2)A | 0.283(2) | 0.314(2) | −0.281(3) | 12. (1) | C(1)D | −0.088(1) | 0.6111(9) | 0.307(2) | 3.9(6) |
| O(1)A | 0.5779(8) | 0.5451(6) | −0.099(1) | 4.2(4) | C(A1)D | −0.026(1) | 0.6592(8) | 0.390(2) | 3.3(6) |
| O(2)A | 0.3867(9) | 0.5449(8) | −0.546(1) | 6.9(5) | C(B1)D | 0.047(1) | 0.6857(8) | 0.320(2) | 3.3(6) |
| S(D2)B | −0.1672(4) | 0.7351(3) | −0.1001(6) | 6.6(2) | C(C1)D | 0.104(1) | 0.6341(8) | 0.273(2) | 2.9(5) |
| N(1)B | −0.2490(8) | 0.4724(6) | −0.192(1) | 3.1(4) | C(D1)D | 0.093(1) | 0.5986(8) | 0.172(1) | 2.8(5) |
| N(D1)B | −0.0304(9) | 0.4172(7) | −0.189(1) | 3.4(5) | C(E1)D | 0.216(1) | 0.5662(8) | 0.272(2) | 2.8(5) |
| N(E1)B | 0.0708(8) | 0.4655(7) | −0.055(1) | 2.9(4) | C(2)D | 0.010(1) | 0.5647(9) | 0.529(2) | 4.1(6) |
| N(2)B | −0.1979(8) | 0.5544(7) | −0.001(1) | 3.3(5) | C(A2)D | −0.039(1) | 0.5151(9) | 0.439(2) | 4.9(7) |
| C(1)B | −0.2059(9) | 0.4933(9) | 0.030(1) | 2.8(5) | C(B2)D | 0.022(1) | 0.4637(9) | 0.398(2) | 4.0(6) |
| C(A1)B | −0.229(1) | 0.4425(8) | −0.072(2) | 2.6(5) | C(C2)D | −0.031(1) | 0.4087(9) | 0.340(2) | 3.4(6) |
| C(B1)B | −0.154(1) | 0.3925(8) | −0.067(1) | 2.7(5) | C(E2)D | 0.098(2) | 0.312(1) | 0.412(2) | 7.3(9) |
| C(C1)B | −0.064(1) | 0.4210(8) | −0.084(1) | 2.3(5) | O(1)D | −0.1280(8) | 0.6382(6) | 0.209(1) | 3.7(4) |
| C(D1)B | −0.002(1) | 0.4498(8) | 0.002(2) | 2.7(5) | O(2)D | 0.0495(9) | 0.5410(7) | 0.626(1) | 6.5(5) |
| C(E1)B | 0.054(1) | 0.4455(9) | −0.171(2) | 3.2(6) | S | 0.2837(3) | 0.7419(2) | 0.5324(4) | 3.0(1) |
| C(2)B | −0.248(1) | 0.5342(9) | −0.221(2) | 4.0(6) | O(1) | 0.2342(9) | 0.7846(6) | 0.600(1) | 5.7(5) |
| C(A2)B | −0.209(1) | 0.5824(8) | −0.125(1) | 2.7(5) | O(2) | 0.3783(8) | 0.7456(6) | 0.582(1) | 5.0(4) |
| C(B2)B | −0.120(1) | 0.6123(8) | −0.152(2) | 3.9(6) | O(3) | 0.2544(8) | 0.6756(6) | 0.5452(9) | 3.4(4) |
| C(C2)B | −0.087(1) | 0.669(1) | −0.084(2) | 5.3(7) | O(4) | 0.2666(9) | 0.7589(6) | 0.403(1) | 4.9(4) |
| C(E2)B | −0.126(2) | 0.785(1) | 0.026(2) | 10. (1) | OW(1) | −0.3554(8) | 0.3765(6) | −0.337(1) | 4.7(4) |
| O(1)B | −0.1956(8) | 0.4742(5) | 0.137(1) | 3.6(4) | OW(2) | −0.2550(8) | 0.3445(7) | 0.174(1) | 5.6(5) |
| O(2)B | −0.2704(9) | 0.5542(7) | −0.326(1) | 6.3(5) | OW(3) | 0.532(1) | 0.2499(7) | −0.176(2) | 7.3(6) |
| S(D2)C | 0.6766(4) | 0.6049(3) | 0.2716(6) | 6.1(2) | OW(4) | 0.0920(9) | 0.7193(6) | 0.694(1) | 5.6(5) |
| N(1)C | 0.5541(9) | 0.3518(7) | 0.257(1) | 3.2(5) | OW(5) | 0.7663(9) | 0.4697(8) | 0.492(1) | 6.7(5) |
| N(D1)C | 0.3257(8) | 0.3495(7) | 0.218(1) | 3.3(5) | OW(6) | −0.1314(9) | 0.3701(7) | −0.394(1) | 6.1(5) |
| N(E1)C | 0.2684(8) | 0.4296(6) | 0.104(1) | 2.9(5) | OW(7) | 0.357(1) | 0.2568(9) | 0.396(2) | 9.6(7) |
| N(2)C | 0.5622(9) | 0.4484(6) | 0.080(1) | 3.0(4) | OW(8) | 0.507(1) | 0.747(1) | 0.425(2) | 10.2(8) |
| C(1)C | 0.546(1) | 0.3898(8) | 0.042(2) | 2.7(5) | OW(9) | −0.235(1) | 0.6649(7) | −0.465(1) | 7.0(6) |
| C(A1)C | 0.536(1) | 0.3338(8) | 0.127(1) | 2.7(5) | OW(10) | 0.199(2) | 0.733(1) | 0.953(2) | 13.2(9) |

a) $B_{eq}$ is the equivalent temperature factor calculated from corresponding anisotropic temperature factors.*
* W. C. Hamilton, *Acta Crystallogr.*, **12**, 609 (1959).

leucyl-L-histidyl)[7] with at least one histidyl residue indicate that these dimers also have the folded conformation, although the $\chi_1^2$ values are nearly +90° and are different by about 180° from those in the $[Zn(CMH)_4]^{2+}$ ion.

The piperazine rings (A and B) are intramolecularly connected to the C and D piperazines through two -NH···O=C hydrogen bonds, respectively, to give the hydrogen-bonded dimers which may behave like bidentate ligands. The formation of these dimers forces $\chi_1^2$ to have nearly −90° when the folded conformations between the imidazole and the piperazine rings are maintained, since $\chi_1^2$=+90° causes a large conformational change of the complex ion, which make the intramolecular hydrogen bonds between the piperazine rings impossible. Interestingly, the conformations of the $-CH_2-CH_2-S-CH_3$ groups are not only different from each other but also different from that of free CMH as given in Table 2, reflecting a large conformational flexibility of this group.

The crystal structure of $[Zn(CMH)_4]$ $SO_4 \cdot 10H_2O$ is shown in Fig. 1. The complexes are linked by a complex system of many hydrogen bonds. There is no hydrogen bond which directly connects the complex ions. All of N(1), N(2), N(D1), O(1), and O(2) atoms except O(2)A and O(2)D participate in hydrogen bonds as donors or acceptors. There are two >C-H···O=C< type short contacts other than typical hydrogen bonds described above: C(E1)B···O(2)D of

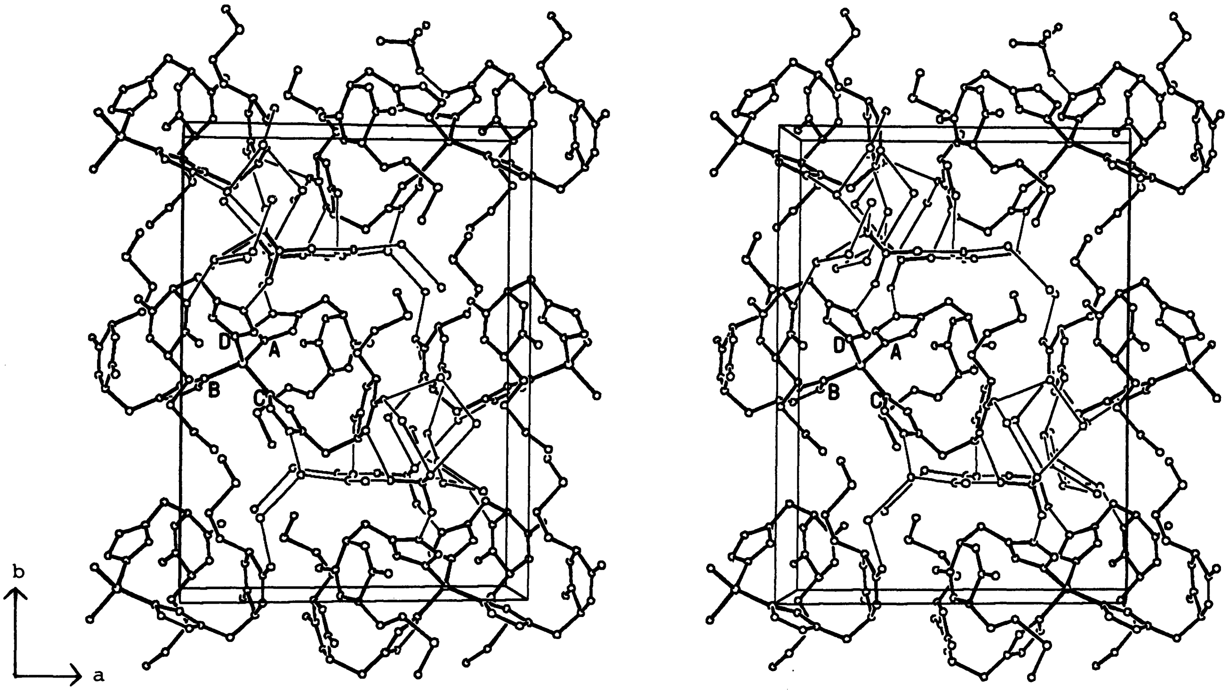


Fig. 1. Crystal structure of $Zn[cyclo(L\text{-methionyl-}L\text{-histidyl})]_4SO_4 \cdot 10H_2O$.

TABLE 2. SELECTED DIHEDRAL ANGLES (°) OF THE CYCLO(L-METHIONYL-L-HISTIDYL) MOIETIES IN THE ZINC(II) COMPLEX AND THE CYCLO(L-METHIONYL-L-HISTIDYL) MOLECULE

| | | Complex | | | | CMH molecule |
|---|---|---|---|---|---|---|
| | | A | B | C | D | |
| C(2)-N(1)-C(A1)-C(1) | $\phi_1$ | 3 | 1 | 1 | 9 | −2 |
| N(1)-C(A1)-C(1)-N(2) | $\psi_1$ | 0 | −7 | −5 | −6 | 3 |
| C(A1)-C(1)-N(2)-C(A2) | $\omega_1$ | −1 | 1 | 9 | −6 | 1 |
| C(1)-N(2)-C(A2)-C(2) | $\phi_2$ | −1 | 10 | −8 | 14 | −7 |
| N(2)-C(A2)-C(2)-N(1) | $\psi_2$ | 4 | −15 | 4 | −10 | 8 |
| C(A2)-C(2)-N(1)-C(A1) | $\omega_2$ | −6 | 11 | −1 | −1 | −4 |
| N(1)-C(A1)-C(B1)-C(C1) | $\chi_1^1$ | 64 | 65 | 68 | 66 | 56 |
| C(A1)-C(B1)-C(C1)-N(D1) | $\chi_1^2$ | −83 | −102 | −104 | −94 | 89 |
| N(2)-C(A2)-C(B2)-C(C2) | $\chi_2^1$ | 65 | −68 | −134 | −70 | 70 |
| C(A2)-C(B2)-C(C2)-S(D2) | $\chi_2^2$ | 174 | −57 | 49 | 175 | −163 |
| C(B2)-C(C2)-S(D2)-C(E2) | | 60 | 162 | 75 | 71 | 79 |

Estimated standard deviations are 1—2°.

TABLE 3. $^{1}$H NMR DATA OF CYCLO(L-METHIONYL-L-HISTIDYL) AND ITS ZINC(II) COMPLEX IN $D_2O$ AT 50 °C

| Compound | Chemical shifts (ppm), $\delta(\Delta\delta)$[a] | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | C(A1)-H | C(B1)-H[b] high | C(B1)-H[b] low | C(D1)-H | C(E1)-H | C(A2)-H | C(B2)-H | C(C2)-H | C(E2)-H |
| CMH | 4.40 | 3.01 | 3.28 | 6.97 | 7.71 | 4.08 | 1.11—1.84[c] | 2.15 | 2.03 |
| $Zn(CMH)_4{}^{2+}$,[d] | 4.47<br>(0.07) | 3.13<br>(0.12) | 3.34<br>(0.06) | 7.06<br>(0.09) | 8.04<br>(0.33) | 4.16<br>(0.08) | 1.30—1.85[c] | 2.13<br>(−0.02) | 1.99<br>(−0.04) |

a) $\Delta\delta=\delta$[zinc(II) complex ion]$-\delta$(free ligand). b) C(B1)-H(β-protons) and C(A1)-H(α-proton) fragments of histidyl residues exhibit an ABX spin system, in which A and B are the β-protons and X is the α-proton. c) Complex and unresolved signals were given for C(B2)-H at this region. d) $Zn(CMH)_4SO_4\cdot 2H_2O$ was used for the measurement.

2.97(2) Å and C(E1)D···O(2)A of 3.08(2) Å.[8] It is noticeable that O(2)A and O(2)D atoms free from hydrogen bonds take part in this interaction as proton acceptors.

Table 3 shows the proton chemical shifts of CMH and its zinc(II) complex ion. β-Methylene protons [C(B2)-H; $\delta$=1.11—1.84] of the methionyl residue of CMH absorbs at higher fields than those ($\delta$= 2.10—3.00) of cyclo(L-methionyl-L-methionyl)[1a] because of the magnetic anisotropy of imidazole ring, suggesting that CMH prefers a folded form.[2a] Although the peaks of β-methylene protons [C(B2)-H] of CMH and $[Zn(CMH)_4]^{2+}$ ion are complex and unresolved, the values of the chemical shifts are used for explaining the conformation of $[Zn(CMH)_4]^{2+}$ ion. The small difference between the chemical shift ($\delta$=1.11—1.84) of β-methylene protons [C(B2)-H] of CMH and those ($\delta$=1.30—1.85) of $[Zn(CMH)_4]^{2+}$ ion suggests a possibility for the complex ion to have a folded form.

Table 4 shows α-β coupling constants and side-chain conformations for CMH and its zinc(II) complex in $D_2O$. These data indicate that the side-chain conformations of CMH and its zinc(II) complex are 67 and 61% for a folded form, respectively, showing the preference of a folded one in aqueous solution.

TABLE 4. α-β COUPLING CONSTANTS AND SIDE CHAIN CONFORMATIONS OF CYCLO(L-METHIONYL-L-HISTIDYL) AND ITS ZINC(II) COMPLEX ION IN $D_2O$ AT 50 °C

| Compound | α-β Coupling constant, Hz | | Side chain conformation[a] | | |
|---|---|---|---|---|---|
| | $J_{\alpha h\beta}$ | $J_{\alpha l\beta}$ | F | $U_I$ | $U_{II}$ |
| CMH | 4.7 | 4.1 | 67 | 14 | 19 |
| $Zn(CMH)_4{}^{2+}$,[b] | 4.9 | 4.6 | 61 | 18 | 21 |

a) Expressed in %; for notations see Ref. 2. b) $Zn(CMH)_4SO_4\cdot 2H_2O$ was used for the measurement.

Also, Table 3 indicates the downfield shift ($\Delta\delta$= 0.09 and 0.33) for C(D1)-H and C(E1)-H of $[Zn(CMH)_4]^{2+}$ ion from those of CMH, respectively. Table 5 shows the up- or down-field shifts ($\Delta\delta$=−1.4, −2.1, 3.4, and 1.0) for C(B1), C(C1), C(D1), and C(E1) carbons of $[Zn(CMH)_4]^{2+}$ ion from those of CMH, respectively. Moreover, Tables 3 and 5 show no distinct shifts for the methine protons [C(A1)-H and C(A2)-H], the methionyl residue protons [C(B2)-H, C(C2)-H, and C(E2)-H], the piperazine ring carbons [C(A1), C(A2), C(1), and C(2)], and the methionyl residue carbons [C(B2), C(C2), and C(E2)] of zinc(II) complex ion from those of CMH. These results reveal that the $Zn^{2+}$ ion links exclusively to the imidazolyl groups of the histidyl residues.

We would also like to infer which N(D1 or E1) atom of the imidazolyl group coordinates to the $Zn^{2+}$ ion in aqueous solution. Iyer *et al.*[9] previously reported that the N(D1) atom of the imidazolyl group coordinates to the $Zn^{2+}$ ion in DMSO-$d_6$ from their NMR studies of the zinc(II) complexes with cyclohepta- and cyclooctapeptides though their results are similar to those for our zinc(II) complex. As shown in Fig. 1, the X-ray structure of the $[Zn(CMH)_4]^{2+}$ ion indicates that the N(E1) atom of the imidazolyl group of CMH links to the $Zn^{2+}$ ion. Speculation regarding the CPK model also supports a possibility for the N(E1) atom of imidazolyl groups to coordinate to the $Zn^{2+}$ ion. These results introduce a conclusion that the N(E1) atom of CMH coordinates to the $Zn^{2+}$ ion in an aqueous solution (as

TABLE 5. $^{13}$C NMR DATA OF CYCLO(L-METHIONYL-L-HISTIDYL) AND ITS ZINC(II) COMPLEX ION IN $D_2O$ AT 50 °C

| Compound | Chemical shifts (ppm), $\delta(\Delta\delta)$[a] | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | C(1) | C(A1) | C(B1) | C(C1) | C(D1) | C(E1) | C(2) | C(A2) | C(B2) | C(C2) | C(E2) |
| CMH | 171.9 | 57.6 | 33.1 | 133.8 | 121.2 | 138.5 | 171.4 | 56.1 | 35.3 | 30.4 | 16.7 |
| $Zn(CMH)_4{}^{2+}$,[b] | 172.0<br>(0.1) | 57.3<br>(−0.3) | 31.7<br>(−1.4) | 131.7<br>(−2.1) | 124.6<br>(3.4) | 139.5<br>(1.0) | 171.2<br>(−0.2) | 56.2<br>(0.1) | 35.2<br>(−0.1) | 30.4<br>(0.0) | 16.7<br>(0.0) |

a) $\Delta\delta=\delta$[zinc(II) complex ion]$-\delta$(free ligand). b) $Zn(CMH)_4SO_4\cdot 2H_2O$ was used for the measurement.

opposed to that of Iyer *et al.*). However, further studies of similar complexes are necessary for determining which N(D1 or E1) atom of the imidazolyl group coordinates to the $Zn^{2+}$ ion in an aqueous solution.

The values (381 and 412 for 0.0246 and 0.0591M, respectively) of the molecular weight obtained for $[Zn(CMH)_4]SO_4 \cdot 2H_2O$ are smaller than the theoretical value (1233/2=617) given in the dissociation of this complex to a monomeric complex ion and a sulfate one. These results suggest that the above complex ion has the following dissociative equilibrium;

$$[Zn(CMH)_4]^{2+} \rightleftharpoons [Zn(CMH)_3]^{2+} + CMH \rightleftharpoons [Zn(CMH)_2]^{2+} + 2CMH \textit{ etc.}$$

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### References

1) a) Y. Kojima, T. Yamashita, Y. Ishino, T. Hirashima, and T. Miwa, *Bull. Chem. Soc. Jpn.*, **56**, 3841 (1983); b) Y. Kojima, N. Ishio, T. Yamashita, and K. Hirotsu, *Chem. Lett.*, **1983**, 1365; c) Y. Kojima, *Transition Met. Chem.*, **4**, 269 (1979). d) Y. Kojima, T. Yamashita, Y. Ishino, T. Hirashima, and K. Hirotsu, *Chem. Lett.*, **1983**, 453; e) E. Ettore, V. Guantieri, A. M. Tamburro, and L. de Nardo, *Inorg. Chim. Acta*, **32**, L39 (1979).

2) a) K. D. Kopple and M. Ohnishi, *J. Am. Chem. Soc.*, **91**, 962 (1969); b) Y. Imanishi, *Adv. Polymer Sci.*, **20**, 1 (1976).

3) a) G. Germain, P. Main, and M. M. Woolfson, *Acta Crystallogr., Sect. B.*, **26**, 274 (1970); M. M. Woolfson, *ibid., A.*, **33**, 219 (1977); b) W. R. Busing, K. O. Martin, and H. S. Levy, "A Fortran Crystallographic Least Square Program," USAEC Report ORNL-TM-305, Oak Ridge National Laboratory, Oak Ridge, TN, (1965).

4) B. K. S. Lundberg, *Acta Crystallogr.*, **21**, 901 (1966); M. M. Harding and S. J. Cole, *ibid.*, **16**, 643 (1963); C. A. Bear, K. A. Duggan, and H. C. Freeman, *ibid., Sect. B.*, **31**, 2713 (1975); W. D. Horrocks, Jr., J. N. Ishley, and R. R. Whittle, *Inorg. Chem.*, **21**, 3265 (1982); C. I. H. Ashby, C. P. Cheng, E. N. Duesler, and T. L. Brown, *J. Am. Chem. Soc.*, **100**, 6063 (1978).

5) M. Bressan, F. Marchiori, and G. Valle, *Int. J. Pept. Protein Res.*, **23**, 104 (1984). The crystal structure of cyclo-(L-mehionyl-L-histidyl)·monohydrate was independently solved by us and will be published elsewhere. The dihedral angles listed in Table 2 are from our data with two times smaller standard deviations than those of Bressan *et al.*

6) Y. Kojima, T. Yamashita, S. Nishide, K. Hirotsu, and T. Higuchi, *Bull. Chem. Soc. Jpn.*, **58**, 409 (1985).

7) a) I. Tanaka, T. Iwata, N. Takahashi, and T. Ashida, *Acta Crystallogr., Sect. B.*, **33**, 3902 (1977); b) M. Cortrait and M. Ptak, *ibid., Sect. B.*, **34**, 528 (1978); c) R. Ramani, K. Venkatesan, and R. E. Marsh, *J. Am. Chem. Soc.*, **100**, 949 (1978); d) C-F. Lin and L. E. Webb, *ibid.*, **95**, 6803 (1973).

8) a) J. Donohue in "Structural Chemistry and Molecular Biology," ed by A. Rich, N. Davidson, and W. H. Freeman, San Francisco, **1968**, p. 443; b) B. C. Wang and B. M. Craven, *J. Chem. Soc., Chem. Commun.*, **1971**, 290.

9) K. S. Iyer, J.-P. Laussac, S.-J. Lau, and B. Sakar, *Int. J. Pept. Protein Res.*, **17**, 549 (1981); K. S. Iyer, J.-P. Laussac, and B. Sarkar, *ibid.*, **18**, 468 (1981).