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The Crystal and Molecular Structure of *N*-Carboxy-L-leucine Anhydride at −70 °C

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The crystal structure of *N*-carboxy-L-leucine anhydride has been determined by means of X-ray. The crystal is orthorhombic; space group, $P2_12_12_1$; $Z=4$ with $a=6.527(1)$, $b=23.920(4)$, and $c=5.527(1)$ Å. The structure was deduced by the direct method and refined to the final R value 0.049 for 624 observed reflections by the block-diagonal least-squares method. The *N*-carboxy anhydride rings are arranged in a layer form sandwiched by hydrophobic side chains. The highest polymerizability of this compound in the crystalline state is due to the sandwich structure.

The *N*-carboxy anhydrides of α-amino acids (amino acid NCAs) are the most useful for preparing polypeptides with high degree of polymerization. Serial crystal structures of amino acid NCAs[1–3] were reported for interpreting their polymerization in the crystalline state. Of the various amino acid NCAs, the title compound is the most reactive in the solid state, polymerizing more rapidly in the crystalline state than in the heterogeneous system using acetonitrile as the solvent of NCA.[4] In a previous paper,[3] we proposed that the charge distribution of the five-membered ring of the amino acid NCA affects its polymerizability. However, this factor alone is not sufficient to explain the high reactivity. We have examined other factors, especially the crystalline field.

Experimental

The title compound was synthesized in the same way as glycine NCA,[1] and purified by recrystallization several times with ethyl acetate–hexane and diisopropyl ether–hexane. L-Leucine NCA thus obtained (*ca.* 100 mg) was dissolved in an appropriate vessel with 20 ml of diisopropyl ether. The vessel was placed in a covered bottle containing hexane and allowed to stand overnight at about 6 °C. Colorless rod-like crystals suitable for X-ray work were obtained.

Since the crystal decomposes easily owing to polymerization at room temperature, data collection was carried out at −70 °C, using a cooling equipment with liquid nitrogen. The unit cell dimensions were determined by the least-squares refinement with 25 high-angle reflections at −70 °C and room temperature. The crystal data are summarized in Table 1.

TABLE 1. CRYSTAL DATA OF L-LEUCINE NCA AT −70 °C (a) AND AT ROOM TEMPERATURE 20 °C (b)

(a)	(b)
$C_7NO_3H_{11}$	
M.W.: 157	
Orthorhombic	
$a=6.527(1)$ Å	$a=6.518(4)$ Å
$b=23.920(4)$	$b=23.983(14)$
$c=5.527(1)$	$c=5.531(5)$
$U=862.9(3)$ Å^3	$U=864.7(11)$ Å^3
Space group: $P2_12_12_1$	
$Z=4$	
$D_x=1.21$ g·cm^{-3}	$D_x=1.21$ g·cm^{-3}
$D_m=\cdots$	$D_m=1.21$ g·cm^{-3}
μ(Mo $K\alpha$) = 1.13 cm^{-1}	

The intensities for the reflections with $2\theta \leqq 40°$ were collected on a Rigaku automated diffractometer with graphite monochromated Mo $K\alpha$ radiation. The usual ω-2θ scan technique was employed with a scan speed $4°(2\theta)\mathrm{min}^{-1}$. A total of 693 reflections were observed, in which 69 with $|F_o|<3.0\sigma(|F_o|)$ were considered as zero reflections. The usual Lorentz and polarization corrections were made. Absorption was uncorrected.

Structure Determination

The structure was determined by the direct method using the program MULTAN. Refinement of the structure was carried out by the block-diagonal least-squares method. All the hydrogen atoms were revealed on a difference map. The final refinement was performed including these hydrogen atoms with isotropic temperature factors. The weighting scheme, $w=0.3$ for $|F_o|<2.52$ and $|F_o|>37.75$, and $w=(4.0476-0.3027|F_o|+0.00758|F_o|^2)^{-1}$ for the other reflections. The final R value became 0.049 for all the non-zero reflections. At the final stage, no peaks higher than 0.11 eÅ^{-3} were found on the difference map. The atomic scattering factors were taken from "International Tables for X-Ray Crystallography."[5] The final atomic parameters and their standard deviations are given in Table 2. A list of the observed and calculated structure factors is kept in the office of the Chemical Society of Japan (Document No. 7822). Computation was carried out on the HITAC 8700 computer at this institute.

Description of the Structure

The bond lengths and angles and the numbering of the atoms are shown in Fig. 1. The distances of C(1)–O(2) and C(2)–O(2) are in good agreement with the corresponding ones in glycine NCA and distinct from the corresponding ones in L-alanine NCA and γ-benzyl-L-glutamate NCA (BLG NCA). Other bond distances in the five-membered ring are close to the corresponding ones in these three NCAs. The C(4)–C(5) and C(5)–C(6) bonds are slightly shorter probably due to ignoring of the high-angle reflections. The angles in the five-membered ring are also in good agreement with the corresponding ones of the other three NCAs. The bond lengths and angles involving the hydrogen atoms are

TABLE 2. FINAL ATOMIC COORDINATES AND THERMAL PARAMETERS FOR L-LEUCINE NCA WITH THEIR ESTIMATED STANDARD DEVIATIONS

The anisotropic thermal parameters ($\times 10^5$) are in the form: $\exp\{-(B_{11}h^2+B_{22}k^2+B_{33}l^2+B_{12}hk+B_{13}hl+B_{23}kl)\}$.

Atom	x	y	z	B_{11}(or B)	B_{22}	B_{33}	B_{12}	B_{13}	B_{23}
C(1)	0.4151(6)	0.2272(1)	0.2778(7)	4919(144)	192(7)	3808(153)	−376(57)	1809(310)	−277(64)
C(2)	0.3703(6)	0.1577(1)	0.5481(7)	3712(120)	259(8)	3830(157)	−328(58)	1555(287)	4(66)
C(3)	0.1946(5)	0.1548(1)	0.3764(7)	2670(99)	202(7)	3190(131)	−12(47)	727(249)	111(57)
C(4)	0.1755(5)	0.0979(1)	0.2562(7)	2632(98)	174(6)	4215(152)	39(44)	581(257)	−7(59)
C(5)	0.0003(6)	0.0934(1)	0.0779(7)	2756(93)	209(7)	5716(197)	−113(56)	−640(316)	−34(69)
C(6)	−0.2078(6)	0.1041(2)	0.1873(11)	2636(116)	427(12)	11067(345)	−155(69)	−250(424)	−394(127)
C(7)	0.0031(8)	0.0353(1)	−0.0458(10)	5538(163)	239(9)	8418(273)	−387(76)	−3956(488)	−560(87)
N(1)	0.2482(5)	0.2001(1)	0.2119(5)	4016(94)	159(5)	3739(120)	−273(44)	−370(230)	−70(49)
O(1)	0.5001(5)	0.2669(1)	0.1909(5)	7020(117)	228(5)	6715(143)	−1225(48)	4732(322)	−215(51)
O(2)	0.4933(4)	0.2024(1)	0.4863(5)	3998(79)	319(5)	4713(103)	−815(45)	−29(218)	−350(50)
O(3)	0.4119(5)	0.1282(1)	0.7150(5)	5211(102)	425(7)	4373(122)	−448(51)	−1415(239)	662(57)
H(1)	0.162(6)	0.211(1)	0.084(7)	10.6(11)					
H(2)	0.081(4)	0.167(1)	0.468(6)	5.5(7)					
H(3)	0.304(5)	0.089(1)	0.174(7)	8.0(9)					
H(4)	0.145(5)	0.068(1)	0.407(5)	7.2(8)					
H(5)	0.021(5)	0.122(1)	−0.053(6)	6.7(8)					
H(6)	−0.343(6)	0.091(1)	0.088(7)	11.5(11)					
H(7)	−0.197(5)	0.084(1)	0.342(7)	8.9(9)					
H(8)	−0.200(5)	0.145(1)	0.243(7)	8.6(9)					
H(9)	−0.129(6)	0.038(1)	−0.163(8)	11.5(12)					
H(10)	−0.019(6)	0.005(1)	0.105(6)	9.6(10)					
H(11)	0.146(6)	0.026(1)	−0.089(7)	11.6(13)					

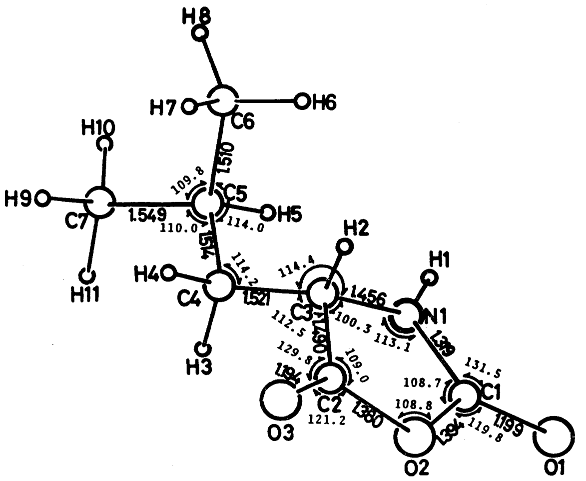

Fig. 1. A perspective drawing of L-leucine NCA viewed along the c axis with the numbering of the atoms, the bond distances (l/Å) and angles (ϕ/°). The estimated standard deviations are 0.004—0.006Å in distances and 0.2—0.4° in angles.

given in Table 3.

Both conformations around C(3)–C(4) and C(4)–C(5) are of stable staggered forms (Fig. 2).

The mean plane consisting of four atoms, C(1), C(2), C(3), and N(1), and the deviations of the atoms from the plane are given in Table 4. The three oxygen atoms, O(1), O(2), and O(3), are significantly displaced from the plane. These features are close to those of the other NCAs.

A stereoscopic drawing of the crystal structure viewed along the c axis is shown in Fig. 3. The hydrogen bonds, N(1)–H(1)···O(1′), connect the molecules to form a ribbon along the a axis (broken lines). The distance of N(1)···O(1′) and the angle of N(1)–H(1)···O(1′) are 2.864(5) Å and 176(3)°, respectively. The ribbons are stacked along the c axis. A pile of ribbons looks like a hydrophilic layer of the five-membered rings sandwiched by hydrophobic side chain layers. These sandwiches are in contact with each other at $y=0$ and $y=1/2$. This

TABLE 3. BOND LENGTHS (l/Å) AND ANGLES (ϕ/°) INVOLVING HYDROGEN ATOMS

The standard deviations are 0.03—0.04 Å in lengths and 2—3° in angles.

Bond lengths			
N(1)–H(1)	0.94	C(6)–H(7)	0.99
C(3)–H(2)	0.95	C(6)–H(8)	1.02
C(4)–H(3)	0.98	C(7)–H(9)	1.08
C(4)–H(4)	1.12	C(7)–H(10)	1.12
C(5)–H(5)	1.01	C(7)–H(11)	0.99
C(6)–H(6)	1.09		
Bond angles			
C(1)–N(1)–H(1)	125	C(5)–C(6)–H(6)	119
C(3)–N(1)–H(1)	122	C(5)–C(6)–H(7)	102
C(2)–C(3)–H(2)	104	C(5)–C(6)–H(8)	104
C(4)–C(3)–H(2)	116	H(6)–C(6)–H(7)	111
N(1)–C(3)–H(2)	107	H(6)–C(6)–H(8)	117
C(3)–C(4)–H(3)	109	H(7)–C(6)–H(8)	102
C(3)–C(4)–H(4)	106	C(5)–C(7)–H(9)	101
C(5)–C(4)–H(3)	109	C(5)–C(7)–H(10)	105
C(5)–C(4)–H(4)	108	C(5)–C(7)–H(11)	108
H(3)–C(4)–H(4)	111	H(9)–C(7)–H(10)	113
C(4)–C(5)–H(5)	109	H(9)–C(7)–H(11)	128
C(6)–C(5)–H(5)	107	H(10)–C(7)–H(11)	99
C(7)–C(5)–H(5)	108		

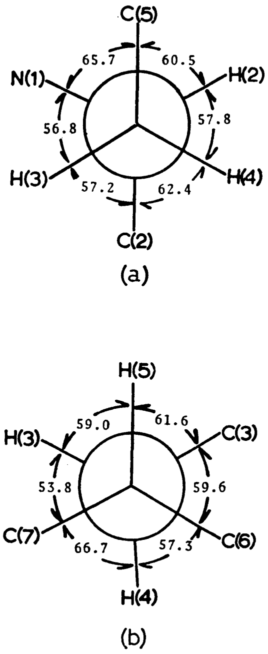

Fig. 2. Newman projection along the bonds (a) C(4)–C(3) and (b) C(5)–C(4).

explains the fact that polymerization cannot proceed along the b axis, the cleavage plane of the crystal being perpendicular to the b axis.

Discussion

The hydrogen bonding schemes differ for the four amino acid NCA crystals so far determined. The distances and angles are summarized in Table 5. The hydrogen bonds are fairly strong.[6] In the crystal of glycine NCA, the molecules form dimers through two hydrogen bonds around the center of symmetry. In the crystals of L-alanine NCA, L-leucine NCA and BLG NCA, the hydrogen bonds extend as linear chains along the b, a, and c axes, respectively.

When the amino acid NCA molecules polymerize progressively, N(1) should attack the C(2) atom of the neighboring molecule nucleophilically.[7] This indicates that in the crystal the C(2) atom of the amino acid NCA molecule contiguous to the terminal amino group of the polymer comes close to the amino group. The distance of the movement required to make a bond is 2.0—5.0 Å for these amino acid NCA crystals.

The dimer structure, as found in the glycine NCA crystal, is unfavorable for progressive polymerization, since two hydrogen bonds should be broken or two molecules as a dimer should move in the crystal. Low reactivity was thus observed for the glycine NCA crystal.

Table 4. The equation of the least-squares plane

Equation	Atom	Deviation/Å
$0.5342X-0.6218Y-0.5727Z$	C(1)	−0.012
$+2.799=0$	C(2)	0.010
($X=ax$, $Y=by$, $Z=cz$)	C(3)	−0.017
	N(1)	0.019
	*O(1)	−0.030
	*O(2)	−0.030
	*O(3)	0.066
	*C(4)	1.145
	*H(1)	−0.035

* Atoms not included in the least-squares calculation.

Table 5. Hydrogen-bond distances and angles of four NCAs

Standard deviations are in parentheses.

NCA	D–H···A	Distance (D···A)	Angle (D–H–A)
Glycine	N(1)–H(1)···O(1′)	2.915(2) Å	162(2)°
L-Alanine	N(1)–H(1)···O(3′)	3.034(5)	170(3)
L-Leucine	N(1)–H(1)···O(1′)	2.864(5)	176(3)
BLG[a)]	N(1)–H(1)···O(4′)[b)]	2.898(4)	174(3)

a) γ-Benzyl-L-glutamate NCA. b) An oxygen atom of carbonyl group in the side chain of γ-benzyl-L-glutamate NCA.

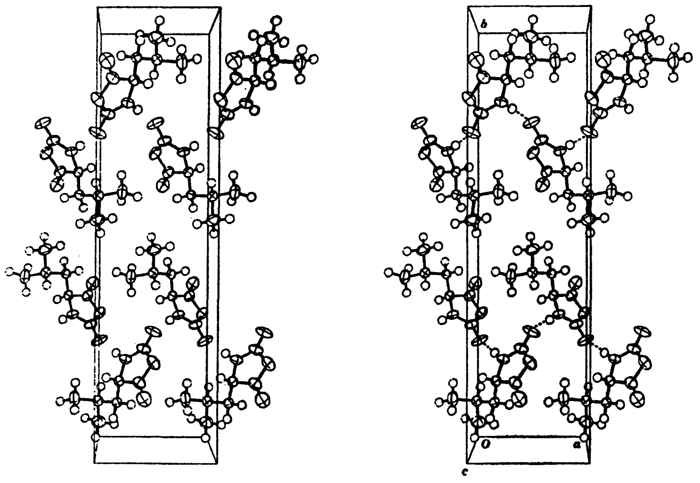

Fig. 3. Stereoscopic drawing of the crystal structure of L-leucine NCA viewed along the c axis. The thermal ellipsoids are drawn at a 50% probability level and the hydrogen atoms are shown in circles.

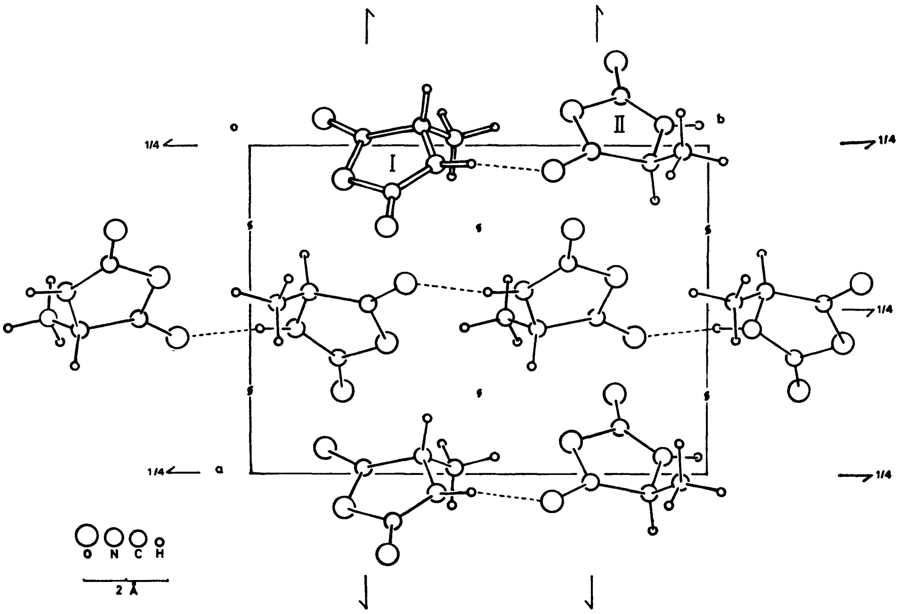

Fig. 4. Projection of the crystal structure of L-alanine NCA viewed along the c axis. I,(x,y,z); II, $(-x,1/2+y,1/2-z)$. Hydrogen bonds are shown by broken lines.

In the BLG NCA crystal, the hydrogen bond is formed between N(1)–H(1) and the oxygen atom of the carbonyl group in the side chain of the neighboring molecule. Since the five-membered ring would drag the side chain of the neighboring molecule, polymerization cannot occur easily. The reactivity of BLG NCA in the crystalline state is not so high as in the heterogeneous system.[4]

On the other hand, the crystal of L-leucine NCA has the following favorable conditions:

(1) because of the sandwiched layer structure, the polymerization proceeds within one sandwich and would not destroy the molecular arrangement in the neighboring sandwich, (2) within a sandwich, the side chains do not hinder the five-membered rings from approaching each other, (3) the side chain can move easily since it does not entwine with side chains of the neighboring molecules and relatively loose contacts are observed between them (compare the structure and the density of the L-leucine NCA crystal with the corresponding ones of the BLG NCA crystal), and (4) in the process of polymerization, the crystal should be easily cloven between sandwichs, leaving a gap between them, and carbon dioxide goes out through the gap without deteriorating the alignment of the amino acid NCA molecules.

The crystal of L-alanine NCA, whose structure viewed along the c axis is shown in Fig. 4, has no such favorable conditions as a result of the N(1)–H(1)···C(3′) hydrogen bond. For example, the shortest distance 3.591 Å of N(1)···C(2), is found between molecules I and II. However, the methyl group prevents molecule II from approaching I. Polymerization does not seem to proceed without destroying the arrangement of the monomers. The L-alanine NCA crystal is very stable in the air, having very low reactivity.

The type of hydrogen bond, N(1)–H(1)···O(1′), is the most favorable for polymerization, taking into account the charge distribution of the five-membered ring and facility of the ring opening.[3] However, high reactivity cannot be attained only by this factor, as suggested by the low reactivity of the glycine NCA crystal. Thus, it is quite reasonable that the sandwich structure accelerates the reactivity in the crystalline state. Because of the sandwich structure and the charge distribution in the NCA ring, L-leucine NCA is more reactive in the crystalline state than in the solution state, and even in the molten state.

Recently a Weissenberg photograph of a polymer crystal formed by keeping an L-leucine NCA crystal on the goniometer head for a few days, indicated that the polymer has the α-helical structure elongated along the c axis. This can be interpreted easily on the basis of the present crystal structure.

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