Acta Cryst. (1970). B27, 1299

The Crystal and Molecular Structure of the Dimeric Copper(II) Chelate of Glycyl-L-leucyl-L-tyrosine*

BY W. A. FRANKS† AND DICK VAN DER HELM‡

University of Oklahoma, Chemistry Department, Norman, Oklahoma, 73069, U.S.A.

(*Received* 24 *July* 1969)

The crystal structure of the copper(II) chelate of glycyl-L-leucyl-L-tyrosine, prepared at neutral *p*H, has been determined and refined by three-dimensional least-squares techniques using 2043 counter data. The crystals are orthorhombic, space group $P2_12_12_1$, with $a=9{\cdot}316$, $b=25{\cdot}76$ and $c=21{\cdot}05$ Å. The unit-cell contains four dimeric units of $Cu_2(glt)_2$, 32 molecules of water and four molecules of diethyl ether; thus, there are two independent peptide units in the asymmetric unit. The final R value is 0·103. The packing of the peptides is very similar to an anti-parallel pleated sheet structure. It is concluded that the conformation of the peptide is only affected slightly by the chelation. There seems to exist a weak interaction of the copper ions with the π-system of the aromatic rings of the tyrosine groups and the suggestion is made that this interaction correlates with the oxidase activity of certain copper-containing proteins and enzymes.

Introduction

The structure of the copper (II) chelate of glycyl-L-leucyl-L-tyrosine (glt) was determined as one in a series of peptide chelate studies to investigate the influence of metal ions on the conformation of peptides and to study the coordination of metal ions in these complexes as well as the hydrogen bonding which occurs. The peptide seemed particularly suitable because both residues contain side chains and one – the tyrosine residue – has an extra functional group. Furthermore the preliminary work indicated the occurrence of two peptides and two copper ions in the asymmetric unit, allowing comparisons of two independent molecules. A note on the unusual coordination geometry observed in this structure has been published (Van der Helm & Franks, 1968).

Experimental results

The method which was most consistent in yielding crystals of the copper complex of glt proceeded by reacting equimolar quantities of $CuSO_4$, $Ba(OH)_2$, both in aqueous solution, and powdered glt. The $BaSO_4$ was removed by filtration and the resulting chelate solution was approximately 0·055 *M* in peptide. The aqueous solution (*p*H 6–7) was equilibrated with diethyl ether and suitable crystals appeared on standing overnight. The crystals of the complex were well-formed blue plates which decomposed within one minute on exposure to the atmosphere.

For the X-ray experiments a crystal was transferred in its mother liquor to a thin-walled (0·01 mm) glass capillary. The mother liquor in the capillary was layered with ether. The capillary was then sealed with a small flame during which it was found useful to immerse the capillary in ice water. The systematic absences, $h00(h\neq 2n)$, $0k0(k\neq 2n)$ and $00l(l\neq 2n)$, showed the space group to be $P2_12_12_1$. The 2θ-values of 49 reflections were measured at room temperature (23° C) and used in a least-squares calculation to obtain the cell dimensions which are $a=9{\cdot}316\pm 0{\cdot}002$, $b=25{\cdot}76\pm 0{\cdot}02$ and $c=21{\cdot}05\pm 0{\cdot}01$ Å. The density of the crystal could not be measured owing to the instability of the compound. The cell dimensions together with a likely value of the density indicated the contents of the asymmetric unit to be $Cu_2(glt)_2.(8–12)H_2O$. It was not until the structure was fully determined that the actual contents of the asymmetric unit were found to be $Cu_2(C_{17}H_{23}N_3O_5)_2.8H_2O.C_4H_{10}O$, with a formula weight of 1044·1. The calculated density is 1·37 g.cm^{-3} for $Z=4$.

The integrated intensities were measured using Ni-filtered Cu $K\alpha$ radiation and the θ–2θ scan technique on a General Electric XRD-5 diffractometer equipped with a single-crystal orienter and a scintillation counter. The background counts were fairly high and few reflections beyond 2θ of 90° had an observable intensity. Of the 2252 independent reflections with a 2θ value below 90°, 2044 were observed and used for the subsequent structure determination. Lorentz, polarization and absorption corrections were applied to the data. The linear absorption factor assumed was 1·80 cm^{-1}, although on the basis of the composition indicated by the final results of the structure determination a value of 17·0 cm^{-1} was calculated. The presence of the

* Work supported by Grant GM-10514 from the National Institutes of Health.

† Present address: Chemistry Department, Harvard University, Cambridge, Mass. 02138, U.S.A. This work was presented in partial fulfillment of the requirements for the degree of Doctor of Philosophy at the University of Oklahoma.

‡ Supported by N.I.H. Development Award 1-K4-GM-42,572. Author to whom inquiries should be made.

Table 1. *Parameters of all carbon, nitrogen and oxygen atoms*

Calculated standard deviations for the last digit are given in parentheses.
Fractional molecule of water, *W*(1), was located from the last difference Fourier synthesis.

	x	*y*	*z*	*B*
Molecule *A*				
N(1)	0·525 (2)	−0·0409 (8)	0·0384 (10)	6·0 (5)
C(1)	0·634 (2)	−0·0280 (8)	0·0847 (9)	3·0 (4)
C(2)	0·566 (2)	−0·0133 (7)	0·1454 (9)	2·2 (4)
O(1)	0·430 (1)	−0·0143 (4)	0·1505 (6)	2·5 (3)
N(2)	0·651 (2)	−0·0004 (6)	0·1918 (7)	3·0 (4)
C(3)	0·598 (2)	0·0099 (7)	0·2598 (9)	2·7 (4)
C(4)	0·663 (2)	0·0611 (7)	0·2776 (9)	2·3 (4)
O(2)	0·791 (2)	0·0714 (6)	0·2632 (7)	4·2 (3)
N(3)	0·583 (2)	0·0934 (6)	0·3087 (7)	3·1 (3)
C(5)	0·648 (2)	0·1423 (9)	0·3214 (10)	4·1 (5)
C(6)	0·585 (3)	0·1627 (10)	0·3861 (10)	5·4 (6)
O(3)	0·470 (2)	0·1442 (6)	0·4059 (7)	4·6 (4)
O(4)	0·653 (2)	0·2013 (7)	0·4112 (9)	7·6 (5)
C(7)	0·620 (3)	0·1859 (10)	0·2676 (10)	6·0 (6)
C(8)	0·460 (3)	0·1923 (10)	0·2557 (10)	6·0 (7)
C(9)	0·382 (3)	0·1611 (10)	0·2212 (10)	5·2 (6)
C(10)	0·229 (3)	0·1669 (10)	0·2125 (10)	6·2 (7)
C(11)	0·166 (3)	0·2039 (10)	0·2557 (10)	7·0 (7)
C(12)	0·241 (3)	0·2371 (10)	0·2837 (10)	7·5 (8)
C(13)	0·381 (3)	0·2313 (10)	0·2940 (10)	7·0 (7)
O(5)	0·013 (2)	0·2047 (8)	0·2499 (10)	8·2 (5)
C(14)	0·649 (2)	−0·0343 (8)	0·3033 (9)	3·4 (5)
C(15)	0·611 (3)	−0·0900 (10)	0·2804 (10)	4·9 (5)
C(16)	0·448 (4)	−0·0944 (10)	0·2847 (20)	9·5 (10)
C(17)	0·683 (4)	−0·1284 (10)	0·3257 (20)	8·6 (9)
Molecule *B*				
N(1)	0·193 (2)	0·1112 (6)	0·3785 (8)	3·7 (4)
C(1)	0·080 (2)	0·0862 (8)	0·3411 (9)	3·1 (4)
C(2)	0·151 (2)	0·0491 (7)	0·2917 (9)	2·9 (4)
O(1)	0·285 (1)	0·0420 (5)	0·2950 (6)	3·3 (3)
N(2)	0·069 (2)	0·0264 (6)	0·2533 (7)	2·9 (4)
C(3)	0·123 (2)	−0·0129 (7)	0·2052 (8)	1·7 (4)
C(4)	0·057 (2)	0·0029 (8)	0·1412 (10)	3·1 (5)
O(2)	−0·076 (1)	0·0169 (5)	0·1398 (6)	3·1 (3)
N(3)	0·139 (2)	−0·0019 (6)	0·0901 (7)	3·3 (4)
C(5)	0·066 (2)	0·0115 (8)	0·0303 (10)	3·5 (5)
C(6)	0·122 (2)	−0·0278 (8)	−0·0181 (9)	2·8 (4)
O(3)	0·243 (2)	−0·0506 (6)	−0·0083 (7)	4·1 (3)
O(4)	0·059 (1)	−0·0308 (6)	−0·0706 (7)	4·4 (3)
C(7)	0·092 (3)	0·0661 (9)	0·0087 (10)	4·8 (6)
C(8)	0·259 (3)	0·0781 (10)	−0·0014 (10)	4·7 (6)
C(9)	0·336 (3)	0·0924 (10)	0·0448 (10)	6·2 (6)
C(10)	0·498 (3)	0·0998 (10)	0·0369 (10)	6·7 (7)
C(11)	0·549 (3)	0·0854 (10)	−0·0211 (10)	6·1 (7)
C(12)	0·467 (3)	0·0700 (10)	−0·0642 (10)	7·1 (7)
C(13)	0·320 (3)	0·0634 (10)	−0·0609 (10)	7·1 (7)
O(5)	0·697 (3)	0·0891 (10)	−0·0256 (10)	11·3 (7)
C(14)	0·060 (3)	−0·0650 (10)	0·2309 (10)	5·3 (6)
C(15)	0·092 (4)	−0·1104 (10)	0·1863 (10)	7·8 (8)
C(16)	−0·031 (4)	−0·1197 (10)	0·1370 (20)	10·0 (10)
C(17)	0·094 (5)	−0·1638 (20)	0·2347 (20)	11·7 (10)
Water molecules				
W(1)	−0·417	−0·159	0·037	19·0
W(2)	0·176 (3)	−0·1875 (10)	0·0034 (10)	13·0 (8)
W(3)	−0·140 (4)	−0·2342 (10)	0·0413 (10)	16·2 (10)
W(4)	−0·180 (3)	0·1564 (10)	0·0362 (10)	13·0 (8)
W(5)	−0·261 (3)	−0·0382 (10)	−0·0612 (10)	12·3 (8)
W(6)	0·356 (4)	−0·1265 (10)	0·0918 (20)	17·9 (10)
W(7)	0·070 (3)	−0·2754 (9)	0·1182 (10)	11·2 (7)
W(8)	−0·110 (3)	0·1338 (10)	0·1602 (10)	11·9 (7)
Ether molecule				
C(18)	0·371 (6)	−0·2406 (20)	0·3782 (30)	17·4 (20)
C(19)	0·365 (6)	−0·2132 (20)	0·4342 (30)	17·9 (20)
O(9)	0·348 (4)	−0·2504 (10)	0·4838 (20)	15·4 (10)
C(20)	0·363 (7)	−0·2327 (20)	0·5393 (30)	19·0 (20)
C(21)	0·397 (7)	−0·2677 (20)	0·5881 (30)	17·7 (20)

capillary and the liquid were neglected in the absorption calculations.

Structure determination

The positions of the two copper atoms were unambiguously located from a sharpened Patterson synthesis. In order to locate more atoms an eightfold superposition map and a Fourier synthesis phased on the copper atom locations were used simultaneously. It was possible to locate the benzene groups and peptide chains. Three side-chain atoms, *i.e.* two from isopropyl groups and one from a benzene ring, were not found in either map. The peaks of some water molecules were present in both maps. A structure factor calculation was made using the peptide atoms which were located and the two copper atoms. The resulting R index ($=\sum||kF_o|-|F_c||/\sum|kF_o|$) was 0·32.

The trial structure was refined by means of block-diagonal least-squares computations (4×4 and 9×9) using anisotropic temperature factors for the copper atoms and isotropic ones for the light atoms. When the R value was 0·17 a difference Fourier synthesis was calculated. It showed the remaining three side-chain carbon atoms and seven water molecules. These atoms were included in subsequent least-squares cycles. This first difference Fourier synthesis also showed four peaks which were clustered together at distances smaller than hydrogen bond lengths In a second difference synthesis, calculated when the R index was 0·125, a fifth peak appeared in close proximity to the four previously observed. The five peaks formed an extended chain with distances of 1·34, 1·50, 1·38 and 1·34 Å. The peak in the middle had a height of 2·1 e.Å^{-3} where the other four ranged between 1·26–1·39 e.Å^{-3}. In addition the peak in the middle was located at a hydrogen bond distance from a water molecule where the distances of the remaining peaks to other atoms in the structure were all larger than 3·5 Å. The group of peaks was therefore identified as diethyl ether, thus explaining the necessity of this solvent for crystallization. All atoms so far located were included in a final series of least-squares calculations. The refinement was terminated when the parameter shifts were smaller than $\frac{1}{3}$ of the calculated standard deviations. The most pronounced features in the subsequent difference synthesis were two peaks with heights of 1·0 and 0·6 e.Å^{-3}, which were close together and at normal hydrogen bond distances from other atoms in the structure. The two peaks had already been observed in the previous difference Fourier synthesis. The peaks were assigned as a disordered molecule of

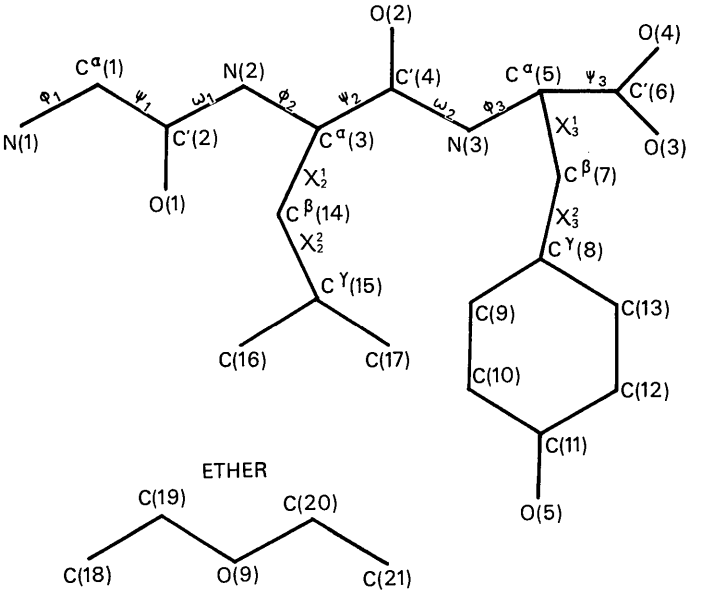


Fig. 1. Numbering of atoms and notation for conformational angles.

Table 2. *Parameters for the copper atoms*

The anisotropic temperature factors are expressed in the form:

$$\exp\left[-(b_{11}h^2+b_{22}k^2+b_{33}l^2+b_{23}kl+b_{13}hl+b_{12}hk)\right].$$

	x	y	z	b_{11}	b_{22}	b_{33}	b_{23}	b_{13}	b_{12}
Cu(1)	0·3268 (3)	−0·0285 (1)	0·0721 (1)	0·0087 (4)	0·0031 (1)	0·0020 (1)	0·0008 (1)	0·0000 (3)	0·0006 (3)
Cu(2)	0·3935 (3)	0·0911 (1)	0·3504 (1)	0·0094 (3)	0·0021 (1)	0·0021 (1)	0·0010 (1)	0·0002 (3)	0·0003 (3)

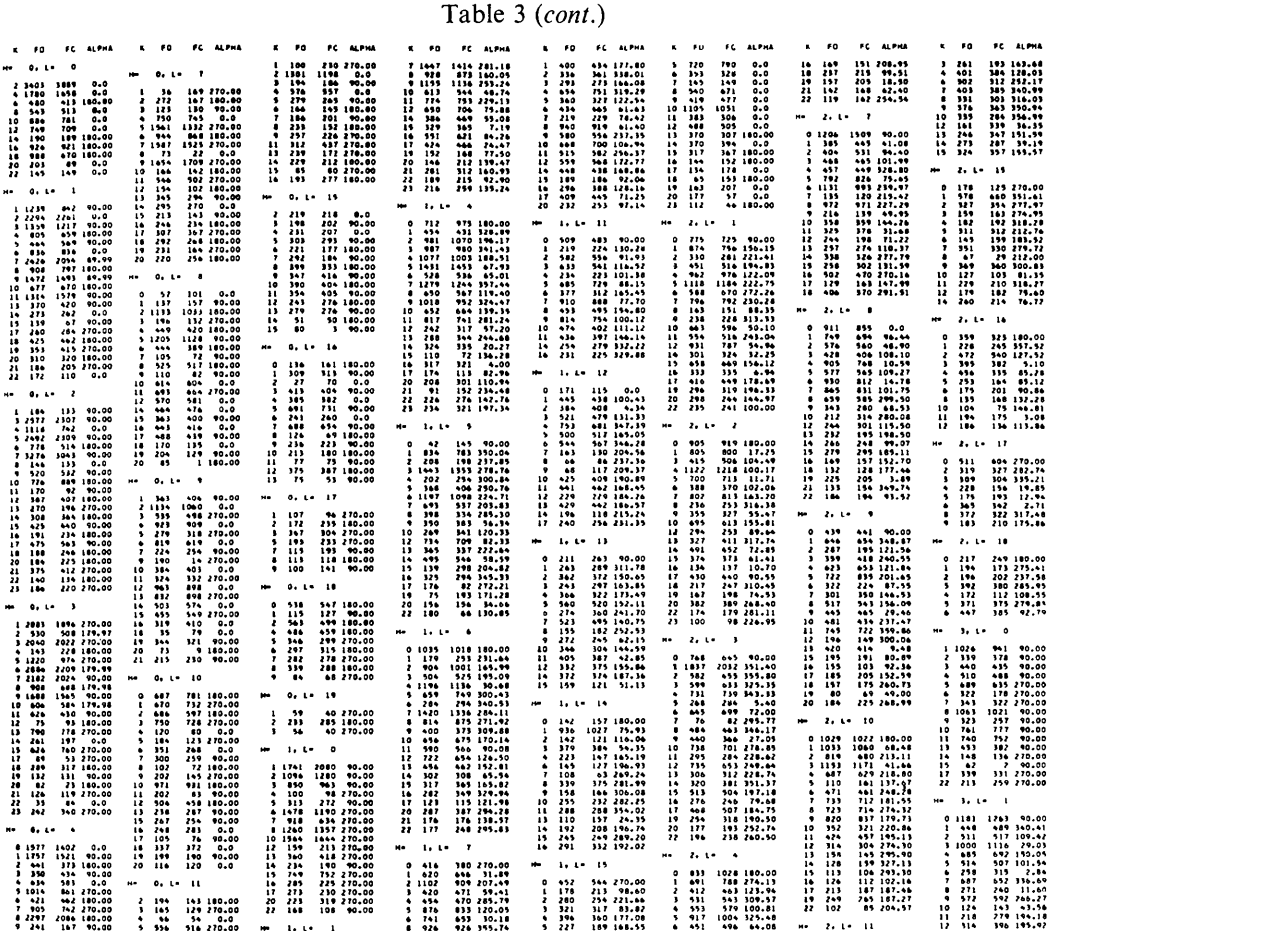
water with an occupancy of $\frac{2}{3}$ and $\frac{1}{3}$ in the two positions. The position with occupancy of $\frac{2}{3}$ is listed in Table 1 [W(1)]. The peak with lower electron density was located at $x=-0.42$, $y=-0.135$, $z=-0.03$. The quantity minimized in the least-squares calculations was $\sum w(|kF_o|-|F_c|)^2$. The weighting scheme used throughout the refinement was $\sqrt{w}=|kF_o|/P$ if $|kF_o|\le P$ and $\sqrt{w}=P/|kF_o|$ if $|kF_o|>P$ ($P=41$ electrons). These weights have a maximum for those intensities which are observed most accurately. The intensities of reflections associated with a structure factor larger than 41 electrons required a filter factor bringing them on

Table 3. *Structure factors*

The values ($\times 10$) for $|kF_o|$ and F_c are listed. All the phase angles have signs opposite to those given in this Table.

K	FO	FC	ALPHA
H= 3, L= 4			
0	235	284	0.0
1	202	250	277.14
2	621	584	323.28
3	263	303	198.69
4	691	621	1.63
5	822	917	119.85
6	364	448	347.55
7	916	976	157.01
8	551	590	266.83
9	606	630	192.99
10	681	719	232.36
11	822	369	218.28
12	351	454	194.14
13	393	360	214.41
14	279	344	151.85
15	155	133	137.85
16	194	184	127.87
17	254	198	114.82
21	388	468	269.04
H= 3, L= 5			
0	112	90	270.00
1	1134	1165	177.32
2	394	437	87.51
3	324	399	251.12
4	759	744	71.74
5	330	278	102.70
6	364	340	81.78
7	926	1024	114.91
8	338	265	4.08
9	1030	1148	78.27
11	670	761	35.21
12	340	406	249.06
13	428	531	344.40
14	339	309	229.20
15	496	479	356.26
17	290	368	25.36
18	140	169	219.53
19	395	396	39.55
20	215	254	243.15
21	151	161	35.87
H= 3, L= 6			
0	547	678	180.00
1	110	81	290.65
2	921	905	236.38
3	172	91	194.00
4	930	957	297.63
5	360	349	289.87
6	584	664	308.50
7	335	335	220.53
8	501	568	269.22
9	712	669	121.12
10	516	520	231.76
11	602	657	99.46
12	551	546	231.32
13	185	175	96.29
14	444	451	265.24
15	264	259	275.61
16	253	254	306.18
18	221	233	20.31
20	211	229	117.34
21	166	154	137.06
H= 3, L= 7			
1	1086	1358	171.71
2	507	514	148.01
3	516	543	193.98
4	431	482	201.79
6	313	234	19.51
7	170	104	191.57
8	696	679	29.11
9	164	194	314.28
10	242	238	36.46
12	479	537	158.63
13	257	274	54.10
14	574	618	164.46
15	361	395	9.95
16	252	295	169.40
17	419	466	351.43
18	249	186	346.47
19	231	262	17.12
20	339	299	12.66
H= 3, L= 8			
0	795	965	0.0
2	243	249	330.43
3	335	313	178.03
4	548	538	274.74
5	576	609	340.74
6	1012	959	232.22
7	475	577	340.17
8	693	687	268.38
9	405	365	340.35
10	656	644	313.18
11	347	337	31.74
12	586	586	308.96
14	293	340	243.18
15	227	173	188.59
16	341	336	200.14
17	252	244	289.67
18	143	153	167.93
19	114	165	320.56
H= 3, L= 9			
0	580	626	270.00
1	468	432	120.42
2	528	548	322.55
3	963	996	135.54
4	439	461	331.72
5	658	723	167.38
6	336	311	278.71
7	499	463	266.11
8	219	174	149.91
9	617	696	278.20
10	186	235	77.33
12	494	489	67.08
13	334	341	62.04
14	279	294	68.44
15	256	240	68.77
16	170	227	141.75
18	191	219	144.72
19	213	244	294.72
H= 3, L= 10			
0	648	568	180.00
1	903	906	283.41
2	284	266	136.76
3	143	194	355.32
4	394	326	107.39
5	618	654	51.08
6	202	223	229.69
7	786	805	60.30
8	97	98	274.36
9	467	439	6.35
10	88	118	335.99
11	583	583	312.85
12	57	46	90.00
13	328	267	340.67
14	190	162	85.75
15	354	370	70.59
17	389	358	87.93
18	125	166	246.64
H= 3, L= 11			
0	615	681	278.00
1	900	498	292.43
2	517	571	305.90
3	618	630	289.26
4	473	534	1.03
5	331	337	265.80
6	211	188	342.81
7	71	140	88.43
8	178	206	206.49
9	250	262	110.34
10	274	305	209.37
11	141	128	244.10
12	92	116	46.70
13	330	341	272.19
14	288	278	57.13
15	239	217	203.60
16	320	290	67.60
17	233	249	119.68
H= 3, L= 12			
0	884	891	180.00
1	325	366	66.26
2	464	498	170.61
3	378	404	346.88
4	426	471	49.36
5	298	327	333.21
6	673	656	35.16
7	193	150	317.33
8	400	445	73.41
9	147	166	13.23
10	373	390	138.18
11	186	179	23.91
12	153	187	172.29
13	215	199	342.28
15	228	199	282.29
16	306	334	16.83
H= 3, L= 13			
0	175	212	270.00
1	255	299	1.40
2	655	682	195.24
3	377	390	349.57
4	444	455	182.23
5	410	400	13.99
6	152	138	6.62
7	226	256	54.14
8	405	415	3.16
9	321	309	146.87
10	145	197	350.46
11	504	496	166.81
12	262	227	215.45
13	272	281	166.16
14	378	401	188.31
15	268	259	170.51
H= 3, L= 14			
0	257	238	0.0
1	152	175	173.17
3	284	264	202.55
4	472	401	148.80
5	330	366	237.39
6	566	498	127.81
7	143	156	250.77
8	459	492	87.71
9	174	124	85.91
10	524	433	70.60
11	288	329	125.57
12	170	156	85.93
13	206	237	118.44
14	82	186	169.31
H= 3, L= 15			
0	409	382	90.00
1	489	494	33.90
2	176	226	56.29
3	534	569	54.37
5	278	218	1.29
6	280	257	256.04
7	419	432	297.05
8	213	232	237.19
9	331	341	288.07
12	104	136	222.08
H= 3, L= 16			
0	214	249	0.0
1	354	416	249.85
2	217	190	80.48
3	228	247	187.48
4	309	279	102.74
5	314	335	137.32
6	244	229	89.58
7	172	125	171.99
9	249	224	249.27
10	228	164	47.56
H= 3, L= 17			
0	398	409	90.00
1	267	264	349.74
2	478	484	54.17
3	147	177	316.68
4	438	408	75.39
6	316	311	108.02
7	225	273	113.00
H= 3, L= 18			
0	174	224	180.00
H= 4, L= 0			
0	1892	2330	180.00
1	648	470	180.00
2	466	515	180.00
4	378	402	180.00
5	335	426	180.00
6	218	116	180.00
7	280	229	180.00
8	640	689	180.00
9	519	530	180.00
10	757	759	180.00
11	558	490	180.00
12	684	632	180.00
13	167	125	180.00
16	454	383	0.0
18	219	243	0.0
19	193	139	0.0
H= 4, L= 1			
0	1482	1498	90.00
1	316	303	243.84
2	836	927	120.84
3	714	841	223.19
4	214	282	134.92
5	126	95	218.74
6	451	458	9.24
7	229	169	80.13
8	547	601	13.47
9	468	548	51.85
10	410	462	346.15
11	279	320	271.17
12	333	281	267.86
13	576	565	271.23
14	280	253	217.34
15	424	448	288.14
16	381	325	260.33
18	246	264	302.84
20	150	140	328.51
H= 4, L= 2			
0	757	605	0.0
1	206	134	315.14
2	787	806	8.16
3	798	762	247.86
4	598	670	212.97
5	1045	1056	233.44
6	517	512	253.93
7	948	977	254.29
8	735	757	264.70
9	660	738	174.34
10	337	312	298.60
11	735	741	140.80
12	348	325	323.60
13	325	313	160.00
15	283	309	238.94
17	237	264	270.43
18	162	117	325.51
20	147	202	0.55
H= 4, L= 3			
0	270	189	90.00
1	375	425	104.16
2	690	643	79.53
3	592	642	116.46
4	459	443	87.41
5	539	522	145.60
6	540	538	77.27
7	284	369	227.79
8	597	590	4.44
9	517	530	292.75
10	459	470	346.17
11	182	159	302.43
12	175	101	108.72
13	319	294	52.32
15	206	215	40.70
16	107	115	227.94
17	194	140	282.42
19	183	141	267.98
20	115	139	287.55
H= 4, L= 4			
0	829	838	0.0
1	922	912	291.37
2	544	469	56.86
3	584	653	350.92
4	308	369	226.89
5	499	581	47.91
6	763	793	214.94
7	236	288	75.90
8	320	411	230.15
10	466	435	323.53
11	398	390	345.73
13	344	313	347.84
15	427	416	42.35
16	308	254	174.39
17	282	320	75.15
18	197	157	185.51
19	183	140	108.35
H= 4, L= 5			
0	749	678	270.00
1	1219	1343	183.83
2	938	935	346.49
3	568	618	174.63
4	467	618	321.57
5	400	393	94.59
6	222	190	230.26
7	438	409	120.38
8	466	479	191.75
9	294	272	146.03
10	100	123	247.77
12	326	288	33.09
14	488	468	31.21
15	348	320	332.48
16	250	272	64.16
17	333	284	335.98
18	275	295	146.48
20	244	252	178.52
H= 4, L= 6			
0	302	284	180.00
1	505	502	315.74
2	529	486	284.88
3	126	229	344.56
4	792	790	326.19
5	297	370	25.81
6	619	563	346.08
7	702	736	14.91
8	164	168	128.93
9	293	377	336.13
10	272	297	216.88
11	353	428	318.17
12	319	341	215.01
13	245	240	265.84
14	126	126	220.65
16	239	207	27.87
18	231	226	34.24
H= 4, L= 7			
0	652	679	270.00
1	182	139	270.00
2	326	593	224.12
3	468	497	299.61
4	661	670	254.27
5	62	73	53.73
6	531	550	289.72
7	634	649	73.32
8	177	179	0.76
9	571	638	106.50
10	373	391	84.64
11	253	214	104.61
12	558	507	81.52
13	304	322	285.69
14	380	324	112.10
15	312	317	278.42
16	161	192	128.98
18	238	249	61.30
19	346	355	104.90
H= 4, L= 8			
0	331	364	0.0
1	744	718	92.66
2	407	425	64.12
3	289	288	47.91
4	331	431	69.31
5	494	508	312.52
6	149	108	41.51
7	690	706	11.81
8	146	154	40.94
9	537	588	25.20
11	351	360	94.07
12	305	329	126.92
13	203	169	39.44
14	260	318	80.59
17	288	341	288.50
H= 4, L= 9			
0	377	422	270.00
1	191	264	48.90
2	651	692	212.36
3	397	410	83.11
4	628	688	196.93
5	320	291	144.02
6	320	339	246.11
7	327	391	239.21
8	389	234	199.82
9	208	171	319.53
10	235	230	172.16
12	406	409	177.42
13	339	344	154.52
14	494	507	157.10
15	341	350	164.11
16	246	291	122.55
17	141	270	206.19
H= 4, L= 10			
0	188	223	0.0
1	438	431	76.23
2	238	268	74.45
3	422	421	56.47
4	564	587	149.12
5	290	285	45.68
6	459	477	161.27
7	204	156	73.50
8	134	113	147.12
9	170	162	171.60
10	313	339	71.30
12	331	342	40.69
14	165	65	148.19
16	254	258	189.39
H= 4, L= 11			
0	118	72	270.00
1	243	315	8.24
2	241	271	358.61
3	341	411	18.60
4	137	186	18.67
5	279	279	6.25
6	381	407	146.41
7	140	168	315.36
8	193	377	169.64
10	313	293	232.24
11	194	224	219.80
12	294	277	297.71
14	170	179	341.91
H= 4, L= 12			
0	199	207	180.00
1	240	267	256.88
2	181	172	188.48
3	186	119	128.98
4	41	23	83.73
5	370	409	92.88
6	318	301	104.36
7	323	336	132.58
8	503	486	106.87
9	171	262	229.40
10	279	318	113.83
11	398	447	248.95
12	78	64	28.03
13	254	223	249.68
14	116	188	12.78
H= 4, L= 13			
0	368	416	90.00
1	174	173	241.94
2	351	366	73.73
3	386	390	260.66
4	119	153	78.14
5	299	334	303.63
6	181	99	190.44
7	159	171	347.12
8	404	239	214.35
10	185	197	188.35
12	98	157	190.72
13	340	293	260.92
H= 4, L= 14			
0	397	427	180.00
1	175	172	141.64
2	293	273	166.90
3	395	414	175.87
5	421	432	191.01
6	220	202	15.56
7	326	347	194.15
8	110	106	36.57
9	304	347	142.26
10	211	200	165.78
11	395	364	132.36
16	222	201	12.18
H= 4, L= 15			
0	253	247	90.00
2	386	430	124.07
4	292	305	120.11
5	141	167	191.53
10	210	219	0.11
H= 4, L= 16			
0	307	259	0.0
1	160	141	298.95
3	231	249	290.00
4	148	152	201.81
5	286	312	261.80
6	298	310	214.26
7	308	264	239.58
8	313	273	241.21
H= 4, L= 17			
0	32	53	270.00
1	265	316	197.87
2	147	121	355.19
H= 5, L= 0			
1	92	24	90.00
2	680	720	270.00
3	268	268	270.00
4	141	211	270.00
5	118	63	90.00
8	229	196	90.00
9	216	268	270.00
12	417	495	270.00
13	194	157	90.00
14	544	474	270.00
17	182	130	90.00
18	210	205	90.00
H= 5, L= 1			
0	717	578	90.00
1	1198	1210	173.54
2	309	274	316.98
3	629	588	195.10
4	595	619	299.63
5	336	320	345.51
6	637	660	284.77
7	467	483	55.12
8	363	361	319.95
9	440	413	125.33
10	644	603	70.79
11	344	365	158.99
12	650	631	84.47
14	227	184	19.87
15	373	327	356.70
16	269	188	273.05
17	315	288	33.30
H= 5, L= 2			
0	519	524	0.0
1	719	667	240.13
2	734	739	55.38
3	563	567	203.97
4	282	215	311.68
5	329	317	294.12
6	390	417	319.67
7	481	714	347.93
8	713	730	271.77
9	515	484	355.90
10	561	571	249.55
11	209	113	187.40
12	302	354	201.67
13	383	452	158.48
14	186	136	139.60
15	264	257	150.35
16	191	167	138.44
18	262	272	159.59
19	165	148	70.21
H= 5, L= 3			
0	447	437	270.00
1	261	275	37.21
3	616	611	139.07
4	468	466	257.76
5	555	623	158.15
6	324	311	300.41
7	383	374	121.82
8	402	386	345.92
9	408	477	40.80
10	145	101	334.42
11	526	560	21.26
13	248	175	352.39
15	259	178	154.04
16	143	221	7.36
18	190	179	26.98
H= 5, L= 4			
0	556	573	0.0
1	505	579	53.64
2	696	684	36.77
3	451	490	24.15
4	651	704	64.61
5	283	273	4.00
6	326	369	306.38
7	183	184	249.67
8	529	539	274.73
10	288	351	297.65
11	199	173	5.49
12	234	181	99.55
13	244	212	354.26
14	84	117	119.60
15	210	263	343.83
16	128	201	248.00
18	276	293	272.86
H= 5, L= 5			
0	564	606	270.00
1	233	229	73.52
2	594	583	272.78
3	307	272	54.82
4	128	201	144.85
5	502	492	55.99
6	333	335	101.92
7	583	581	69.84
8	185	169	241.01
9	396	450	128.65
10	241	288	264.79
11	425	465	112.56
13	411	395	126.26
14	313	330	73.35
15	150	146	138.19
16	371	404	70.06
18	253	142	97.16
H= 5, L= 6			
0	261	284	0.0
1	471	500	46.79
3	826	874	35.16
5	283	254	49.72
6	620	621	96.18
7	307	339	59.84
8	566	624	97.66
10	279	286	106.47
11	134	174	320.17
13	338	324	17.21
14	259	215	203.00
15	140	168	25.65
16	172	206	145.11
H= 5, L= 7			
0	193	177	270.00
3	446	453	1.93
4	414	373	180.97
5	605	675	359.78
6	270	322	192.67
7	151	251	2.02
8	382	400	181.28
10	262	283	184.02
11	285	276	218.28
12	231	225	155.12
15	142	122	19.58
16	160	171	274.40
17	170	121	2.61
H= 5, L= 8			
0	650	631	180.00
1	419	458	142.14
2	269	282	198.39
3	666	791	153.10
4	236	264	152.17
5	156	232	153.42
6	308	312	128.88
8	425	445	116.48
10	448	451	110.07
11	171	239	192.92
12	166	206	113.50
13	315	428	197.45
25	110	130	198.19
H= 5, L= 9			
0	798	802	90.00
1	298	320	337.96
2	360	430	100.54
4	209	184	187.50
5	177	114	215.19
6	195	224	202.42
7	176	180	245.31
8	202	216	173.47
9	214	218	262.30
10	280	270	107.71
11	209	158	250.27
15	313	290	216.16
H= 5, L= 10			
0	477	499	180.00
1	217	233	107.36
2	412	471	125.09
4	343	367	125.71
5	257	310	209.46
6	178	203	162.66
7	163	227	208.18
8	303	310	252.47
9	150	180	210.89
10	325	277	287.54
11	196	250	204.74
H= 5, L= 11			
0	139	141	270.00
1	224	296	345.09
2	159	102	240.32
5	325	314	220.29
6	330	340	66.87
7	320	371	238.71
9	305	304	291.64
10	267	247	258.06
11	295	347	308.36
H= 5, L= 12			
0	276	284	180.00
2	204	258	198.86
3	190	263	322.04
4	312	348	220.00
5	55	88	267.39
6	138	186	236.02
7	342	320	196.66
9	300	281	192.55
10	99	117	241.89
H= 5, L= 13			
0	37	60	270.00
1	295	417	161.93
3	217	183	159.02
4	309	339	77.17
5	108	129	328.27
7	153	206	338.48
10	146	22	338.34
H= 5, L= 14			
0	132	66	0.0
2	140	198	236.79
3	236	207	229.70
4	237	188	259.85
5	271	279	232.11
7	263	248	257.84
H= 5, L= 15			
0	18	128	90.00
4	174	241	306.69
5	102	109	121.79
H= 6, L= 0			
0	244	206	180.00
2	250	192	180.00
3	386	454	0.0
4	144	291	0.0
6	353	403	0.0
7	355	422	180.00
9	114	129	180.00
10	342	370	180.00
11	331	392	0.0
12	753	703	180.00
13	690	724	0.0
14	156	135	180.00
15	373	386	0.0
H= 6, L= 1			
0	138	256	270.00
1	341	250	338.85
2	402	411	215.98
3	387	383	344.91
4	495	473	186.51
5	535	506	16.66
6	177	230	131.35
7	562	577	67.31
8	140	161	227.60
9	537	467	107.36
10	301	306	239.09
11	346	339	118.66
12	230	380	206.57
13	106	142	193.43
14	482	481	171.53
15	196	176	6.77
H= 6, L= 2			
0	577	617	0.0
1	517	570	110.94
2	306	271	338.08
3	206	205	72.25
4	287	249	285.99
5	264	265	293.01
6	355	317	80.63
7	405	388	304.66
8	757	721	69.13
10	400	408	54.41
11	201	186	71.98
12	296	262	325.64
14	381	329	213.71
15	499	463	259.06
16	325	335	165.53
H= 6, L= 3			
0	271	274	90.00
1	289	247	119.69
2	400	402	158.75
3	317	355	88.40
4	606	560	198.97
5	442	420	39.74
7	348	355	331.94
8	386	384	155.03
9	259	254	265.77
10	547	516	113.83
11	303	274	151.22
12	286	289	111.98
13	424	414	92.50
14	118	61	221.04
15	317	335	108.22
H= 6, L= 4			
0	173	180	0.0
1	649	649	105.58
3	809	710	130.11
4	243	267	104.06
5	639	607	110.84
6	145	182	151.93
7	468	456	69.58
8	263	202	270.65
9	361	402	330.08
10	351	340	6.29
11	492	456	274.45
12	330	288	33.68
13	317	351	263.77
14	255	235	75.27
15	243	207	297.70
16	179	143	139.56
H= 6, L= 5			
0	404	369	90.00
1	610	590	347.18
2	445	415	44.92
3	174	192	269.51
4	213	157	198.20
5	406	425	215.96
6	518	518	190.57
7	181	231	208.75
8	917	930	194.11
9	386	388	299.70
10	639	632	185.67
11	578	581	315.10
12	196	178	191.63
13	225	218	288.02
14	148	132	271.79
H= 6, L= 6			
0	1123	1030	180.00
1	409	402	43.11
2	699	741	157.04
3	544	533	47.45
4	266	290	125.50
5	395	361	80.24
6	215	196	20.27
7	411	400	125.77
8	130	126	27.55
9	451	462	139.90
10	312	269	196.70
12	172	171	101.92
H= 6, L= 7			
0	452	491	270.00
1	305	282	238.87
2	258	207	203.46
3	427	440	275.47
4	451	498	102.42
5	346	356	264.15
6	132	421	99.43
7	298	251	281.27
8	398	358	53.07
11	182	209	244.28
13	229	241	257.92
14	169	197	60.87
H= 6, L= 8			
0	864	870	180.00
2	974	953	212.86
3	511	534	290.64
4	626	573	214.18
5	547	641	267.00
6	395	347	215.36
7	590	550	255.40
9	219	207	261.10
12	194	206	251.73
H= 6, L= 9			
0	414	405	90.00
1	666	698	167.85
2	178	129	111.64
3	546	513	141.37
4	155	209	279.91
6	289	332	325.55
7	390	381	244.99
8	409	378	322.68
9	512	493	248.84
10	292	268	13.77
11	138	147	238.02
12	101	200	44.13
H= 6, L= 10			
0	339	346	0.0
1	488	513	255.53
2	401	412	334.29
3	671	605	242.84
4	281	279	302.30
5	253	287	273.48
6	228	180	336.61
7	205	250	330.81
8	164	248	346.54
9	161	164	356.28
10	325	324	324.57
12	225	220	334.87
H= 6, L= 11			
0	113	92	270.00
1	115	231	39.66
2	360	332	330.39
3	231	125	104.80
4	590	574	330.20
5	263	329	171.70
6	268	244	304.68
7	216	197	185.51
10	157	155	355.83
H= 6, L= 12			
1	220	232	248.93
2	120	151	132.89
4	200	211	19.49
5	245	230	43.34
6	376	361	323.62
8	288	280	289.27
H= 6, L= 13			
0	226	196	270.00
2	204	206	292.03
3	182	203	179.22
5	104	152	138.32
H= 7, L= 0			
3	274	307	90.00
4	156	127	270.00
5	682	667	90.00
7	474	425	90.00
11	335	273	270.00
13	298	336	270.00
H= 7, L= 1			
0	306	269	270.00
1	250	229	220.53
2	155	48	294.71
3	498	453	243.51
5	340	346	263.61
6	196	217	169.63
7	193	170	101.56
8	517	498	178.26
9	271	248	60.09
10	389	309	165.04
13	237	224	299.78
H= 7, L= 2			
0	537	573	180.00
1	190	258	77.75
2	411	428	184.60
3	166	154	114.58
4	176	119	210.83
5	97	128	162.37
6	227	268	353.79
7	250	256	134.72
8	116	139	282.90
10	233	223	187.34
13	246	226	171.55
H= 7, L= 3			
0	118	120	270.00
1	165	107	231.80
2	435	407	181.19
3	136	177	254.76
4	327	334	185.74
5	320	381	265.08
7	400	392	264.84
8	435	410	356.18
10	209	172	327.05
11	398	376	274.76
12	293	259	130.71
13	247	232	264.97
H= 7, L= 4			
0	80	142	180.00
1	111	49	277.75
2	243	307	186.38
3	244	234	294.04
4	403	439	179.87
5	307	317	309.10
6	427	426	184.26
7	410	423	323.46
9	392	362	48.38
11	493	474	61.74
12	177	161	330.20
H= 7, L= 5			
0	277	254	270.00
1	217	135	106.87
2	338	263	302.64
3	113	124	67.10
4	248	246	350.37
7	480	414	274.74
8	303	331	13.47
9	595	552	262.17
10	400	385	8.74
H= 7, L= 6			
0	225	161	0.0
1	921	955	265.88
3	314	318	252.88
4	163	178	165.03
7	204	151	66.97
8	260	239	110.48
10	312	326	44.77
H= 7, L= 7			
1	270	290	330.93
2	447	421	0.35
3	183	117	3.75
4	476	407	0.0
6	323	265	333.78
7	53	26	358.38
8	233	278	334.67
10	204	246	334.75
H= 7, L= 8			
0	459	433	180.00
1	462	451	261.75
2	143	155	124.45
3	388	354	261.94
4	513	519	39.92
6	315	327	57.66
7	203	184	40.99
8	290	222	104.97
9	234	176	91.72
H= 7, L= 9			
0	147	151	270.00
2	333	315	222.64
3	258	212	289.09
6	306	316	24.61
7	372	372	95.90
H= 7, L= 10			
0	337	295	0.0
2	168	219	318.42
3	347	251	258.64
4	155	317	299.57
5	516	489	271.24
H= 7, L= 11			
1	188	130	93.27
H= 8, L= 0			
0	202	210	180.00
1	211	206	0.0
2	370	381	180.00
3	373	371	0.0
4	360	330	180.00
5	347	337	0.0
6	229	288	180.00
8	78	123	180.00
H= 8, L= 1			
0	375	331	270.00
1	257	250	178.37
2	293	269	305.16
6	176	174	93.35
7	307	248	374.14
8	113	144	110.89
H= 8, L= 2			
0	139	156	180.00
1	385	370	254.09
3	405	330	237.35
4	128	98	241.13
5	158	130	265.23
6	156	145	146.12
7	148	124	336.77
8	349	345	105.99
H= 8, L= 3			
0	114	141	90.00
1	186	196	344.76
2	99	99	355.11
4	413	373	325.72
6	345	322	305.19
7	191	165	80.78
H= 8, L= 4			
0	175	167	0.0
2	252	250	65.90
3	197	172	225.69
4	410	348	43.49
6	326	297	78.71
H= 8, L= 5			
0	115	68	270.00
1	402	378	9.17
2	89	64	187.30
3	178	152	16.29
4	91	106	125.93
5	208	152	88.78
H= 8, L= 6			
0	315	310	0.0
1	197	148	59.15
2	276	248	357.72
H= 9, L= 3			
9	336	278	126.97
H= 10, L= 2			
10	250	233	245.84

Table 3 (*cont.*)



a common scale with the standard reflection while those with low intensities were affected by adverse counting statistics. The final positional and thermal parameters and their estimated standard deviations, as calculated from the inverse of the least-squares matrix, are given in Tables 1 and 2. No attempt was made to locate the hydrogen atoms in view of the stated purposes of the structure determination, the high temperature factors observed and the large size of the asymmetric unit. The final R value for the 2043 observed amplitudes used in the refinement is 0·103. When the 311 unobserved reflections within the 2θ

limit of 90° are included, the R value is 0·108. A list of the observed and calculated structure factors is given in Table 3. The absorption corrections were calculated on an IBM 1410 computer with a program written by P. J. Schapiro. All other computations were carried out on an IBM 360-40. The structure factor-least squares and Fourier programs used were written by Ahmed (1966*a*,*b*). Table 3 was prepared with a program written by Pippy (1967). The atomic scattering factors used for Cu^{2+}, O, N and C were those listed in *International Tables for X-ray Crystallography* (1962).

Description and discussion of the structure

General, chelation sites and metal surrounding

A schematic drawing of a peptide and ether molecule, specifying the atom numbering used, is shown in Fig. 1. Fig. 2 shows a projection of a part of the structure down the *b* axis.

The atomic positions of Table 1 correspond to the atoms of peptide chains *A* and *B*. Some of the carbon atoms in these peptide units are identified in Fig. 2 for clarification. Some of the water molecules are drawn but none of the ether molecules. The reason for the limitations is the length of the projection axis, 25·76 Å. This particular projection, however, shows most clearly the basic building blocks of the structure which are copper–peptide dimers. One dimer is identified in Fig. 2 by labels *A* and *B*, for the peptides, Cu(1) and Cu(2). The modes of chelation of the two peptide groups to the metal ions are quite similar. Two five-membered chelate rings are formed by means of the terminal nitrogen atoms of the glycyl residues, N(1), and the carbonyl oxygen atoms of the glycyl–leucyl peptide groups O(1) of molecules *A* and *B* with Cu(1) and Cu(2) respectively. Two other five-membered chelate rings are formed by the deprotonated amide nitrogen atom of the leucyl–tyrosine peptide groups, N(3), and an oxygen atom of the terminal acid groups of *A* and *B* with Cu(2) and Cu(1) respectively. Another proof that these particular peptide nitrogen atoms are deprotonated in this complex, formed at *p*H 6–7, is the trigonal surrounding of both N(3) atoms by Cu, C(4) and C(5). The sum of the bond angles is 360° for peptide N(3*A*) and 359° for N(3*B*) (see Tables 4 and 7). It should be noted that both peptides are doubly deprotonated on the acid end of the molecule.

The five-membered chelate rings form the bases for the square-pyramidal surroundings of Cu(1) and Cu(2). The tops of the pyramids are *W*(6) for Cu(1) and O(4*B*) (*e*) for Cu(2). These fifth bonds are somewhat weaker than those in the basal plane (Van der Helm & Franks, 1968). The atoms forming the bases of the pyramids are tetrahedrally distorted (planes 1 and 2, Table 4), while the copper ions are displaced from the base plane toward the top of the pyramid. Angles and distances in the coordination of the two independent copper ions are given in Table 5. None of the four 5-membered chelate rings are planar (planes 3, 4, 5 and 6, Table 4). The rings involving the free acid group are in the half-chair conformation with the peptide nitrogen and copper ion on opposite sides of the least-squares planes through the acid groups. The chelate

Table 4. *Least-squares planes*

The equations of the planes are expressed in the form:

$$Ax + By + Cz = D$$

where x, y and z are fractional coordinates and D is the distance from the origin in Å. The method of Schomaker, Waser, Marsh & Bergman (1959) was used to calculate the least-squares planes.

1	N(1*A*), O(1*A*), O(3*B*) N(3*B*)	1·481	23·887	−7·140	−0·631
2	N(1*B*), O(1*B*), O(3*A*), N(3*A*)	1·731	−18·412	14·193	3·789
3	C(1*A*), C(2*A*), O(1*A*), N(2*A*)	0·374	−24·573	6·269	1·454
4	C(5*A*), C(6*A*), O(3*A*), O(4*A*)	5·017	−17·121	10·902	4·327
5	C(1*B*), C(2*B*), O(1*B*), N(2*B*)	1·184	19·556	−13·438	−2·798
6	C(5*B*), C(6*B*), O(3*B*), O(4*B*)	4·929	19·242	−8·476	0·276
7	C(3*A*), C(4*A*), O(2*A*), N(3*A*)	2·913	−10·135	18·195	6·368
8	C(3*B*), C(4*B*), O(2*B*), N(3*B*)	2·768	24·374	2·720	0·590

	Δ (1)	Δ (2)
N(1)	+0·16 Å	−0·13 Å
O(1)	−0·15	+0·12
O(3)	−0·16	+0·13
N(3)	+0·15	−0·12
Cu	−0·08	+0·19

	Δ (3)	Δ (5)
C(1)	−0·00 Å	−0·01 Å
C(2)	−0·00	+0·02
O(1)	+0·00	−0·01
N(2)	+0·00	−0·01
N(1)	−0·01	+0·12
Cu	−0·18	+0·34
C(3)	+0·16	−0·07

	Δ (4)	Δ (6)
C(5)	−0·01	+0·02
C(6)	+0·03	−0·06
O(4)	−0·01	+0·02
O(3)	−0·01	+0·02
N(3)	+0·37	−0·39
Cu	−0·09	+0·18

	Δ (7)	Δ (8)
C(3)	0·00	−0·01
C(4)	−0·00	+0·02
O(2)	0·00	−0·01
N(3)	0·00	−0·01
N(2)	−0·98	+0·93
C(5)	−0·08	−0·04

rings involving the free amino group are in the envelope conformation.

It was pointed out (Van der Helm & Franks, 1968) that close approaches (between 3·17 and 3·34 Å) (Table 5) occur between the copper atoms and C(8) and C(9) of both tyrosine groups. A similar and even closer approach has now been found in the structure of bis(L-tyrosinato)copper (II) (Tatsch & Van der Helm, 1969). In all three cases the side chains of L-tyrosine fold back so that they are situated below the basal planes of the square pyramidal copper coordination (Fig. 2; Van der Helm & Franks, 1968: Fig. 1). This is a result of the fact that the C^{β}–C^{γ} [*i.e.* C(7)–C(8)] bonds are skewed with respect to both N–C^{α} [N(3)–C(5)] and C^{α}–C′ [C(5)–C(6)] bonds. For an L-amino acid that means that the N–C^{α}–C^{β}–C^{γ} and C′–C^{α}–C^{β}–C^{γ} configurational angles have to be approximately 60° and 300°, respectively. The fact, however, that both these angles are skewed does not necessarily imply that close approaches must occur between the metal atom and the ring in the side chain. For instance in the structure of (β-alanyl-L-histidinato)copper(II) dihydrate (Freeman & Szymanski, 1967), the C^{β}–C^{γ} bond of the histidine residue is skewed with respect to the N–C^{α} and C^{α}–C′ bonds, but the closest metal imidazole distance is 3·52 Å. It is therefore believed that an interaction exists between the Cu^{2+} ions and the aromatic rings of the tyrosine groups both in the present structure as in the bis(L-tyrosinato)copper(II) structure. The weakness of the interaction allows for rapid kinetics of reaction. Both the enzymes tyrosinase and laccase, and the protein ceruloplasmin contain copper and have oxygenase activity. It is interesting to note that Levine & Peisach (1962) suggested, on the basis of chemical data, that the enzyme binding in ceruloplasmin to substrate was not through the amine or phenolic group but instead directly to the π-electrons of the aromatic ring. Broman, Malmström, Aasa & Vänngård (1963) hypothesized that the Cu^{+} ion, which is also present in ceruloplasmin, partakes in substrate binding by interaction with the π-system of the substrate. The present suggestion, however, is the involvement of Cu^{2+}, rather than Cu^{+}, in this type of binding, or interaction, with the substrate. This interaction, subsequently, allows an electronic reduction-oxidation reaction of the Cu^{2+} ion with the phenolic group.

Table 5. *Angles and distances in the metal coordination*

The literature values are from Freeman (1967). Standard deviations for the last digit are given in parentheses. O(*t*) is *W*(6) and O(4*B*) for the Cu(1) and Cu(2) coordination respectively.

	Cu(1)	Cu(2)	Literature
Cu–N(1)	2·00 (2) Å	2·02 (2) Å	2·00 (1) Å
Cu–O(1)	1·95 (2)	2·00 (2)	1·99 (1)
Cu–N(3)	1·92 (2)	1·98 (2)	1·92 (1)
Cu–O(3)	1·95 (2)	1·94 (2)	1·98 (1)
Cu–O(*t*)	2·57 (2)	2·32 (2)	
Cu–C(8)	3·21 (3)	3·34 (3)	
Cu–C(9)	3·17 (3)	3·27 (3)	

Table 5 (*cont.*)

	Cu(1)	Cu(2)	Literature
O(1)–Cu–N(1)	83 (1)°	82 (1)°	
O(3)–Cu–N(3)	85 (1)	85 (1)	
O(3)–Cu–N(1)	91 (1)	89 (1)	
O(1)–Cu–N(3)	103 (1)	102 (1)	
O(*t*)–Cu–O(1)	90 (1)	95 (1)	
O(*t*)–Cu–N(1)	79 (1)	98 (1)	
O(*t*)–Cu–N(3)	114 (1)	100 (1)	
O(*t*)–Cu–O(3)	84 (1)	88 (1)	
Cu–N(1)–C(1)	112 (1)°	113 (1)°	111 (1)°
Cu–O(1)–C(2)	115 (1)	116 (1)	113
Cu–O(3)–C(6)	111 (1)	113 (2)	115 (1)
Cu–N(3)–C(5)	109 (1)	109 (1)	116 (1)
Cu–N(3)–C(4)	136 (1)	136 (1)	120 (1)
O(1*A*)–N(3*B*)	3·02 (3) Å	O(3*B*)–N(1*A*)	2·81 (3) Å
O(1*B*)–N(3*A*)	3·09 (3)	O(3*A*)–N(1*B*)	2·78 (3)

Some of the bond angles involving the chelate rings are shown in Table 5 and compared with literature values. The significant differences are in the angles around the N(3) atom. The Cu–N(3)–C(4) angles deviate 16° from the average values (for 5 angles) reported previously (Freeman, 1967). Both in the present structure and in those to which the bond angles are compared, the hydrogen atom on the amide nitrogen atom has been ionized. The important difference between this structure and those with which it is compared, *i.e.* diaquoglycylglycinatocopper(II) hydrate (Freeman, 1967), sodiumglycylglycylglycinato cuprate(II) (Freeman, Schoone, & Sime, 1965) and disodium glycylglycylglycylglycinato cuprate(II) decahydrate (Freeman, & Taylor, 1965), is the fact that more than one chelate ring is formed between the peptide and one particular metal ion in the latter three structures, compared with only one in the present structure. This same feature is the probable cause for the observed differences, from the literature values, of the C′–N–C^{α} angles. Freeman (1967) lists an average value of 123° for this angle when the N atom is a part of a chelate ring, which is the same as observed in free peptides (Marsh & Donohue, 1967). In the present structure, however, the C′–N–C^{α} angles are 114 and 115° for the peptide nitrogen atoms involved in chelate rings, but 123° (twice) for those which are not a part of a chelate ring (Table 7). It is therefore suggested that when a peptide forms only one chelate ring with a particular metal ion the following angles can be expected around the deprotonated nitrogen atom: Cu–N–C^{α}, 109°, Cu–N–C′, 136° and C′–N–C^{α}, 115°, rather than 116, 120 and 123° respectively.

Bond distances and bond angles

The bond angles and bond distances in the backbone of the peptides, the phenyl and isopropyl groups, and the ether molecule, are shown in Tables 6, 7 and Fig. 3 respectively. In general these values are close to those expected. There are some relatively large deviations in the side groups, but the temperature movement is

large for those atoms (Table 1) and the estimated standard deviations are therefore rather large. The bond distances in the backbone of the peptides are similar for the same bonds in the two peptides. The average bond distances compare favorably with those of copper complexes of amino acids and peptides summarized by Freeman (1967). There is one notable exception, namely the bond distances N(2)–C(3) which are 1·54 and 1·52 Å and deviate by 4σ and 3σ from the literature value of 1·455 (Marsh & Donohue, 1967) for a free peptide or 1·46 Å (Freeman, 1967) for a chelated peptide. The other N–C^{α} distances in the present structure are normal.

Table 6. *Bond distances in side-chains and ether molecule*

Standard deviations for the last digit are given in parentheses.

C(5)—C(7)	1·62 (3) Å	1·50 (3) Å
C(7)—C(8)	1·52 (4)	1·60 (4)
C(8)—C(9)	1·30 (4)	1·26 (4)
C(9)—C(10)	1·44 (4)	1·53 (4)
C(10)–C(11)	1·44 (4)	1·36 (4)
C(11)–C(12)	1·25 (4)	1·25 (4)
C(12)–C(13)	1·33 (4)	1·39 (4)
C(13)–C(8)	1·48 (4)	1·43 (4)
C(11)–O(5)	1·43 (4)	1·39 (4)
C(3)—C(14)	1·53 (3)	1·56 (3)
C(14)–C(15)	1·56 (3)	1·53 (4)
C(15)–C(16)	1·52 (4)	1·56 (5)
C(15)–C(17)	1·53 (4)	1·71 (5)
C(18)–C(19)	1·38 (8)	
C(19)–O(9)	1·42 (7)	
O(9)—C(20)	1·26 (6)	
C(20)–C(21)	1·40 (8)	

Table 7. *Bond angles*

Standard deviations for the last digit are given in parentheses.

Angle	Molecule *A*	Molecule *B*
N(1)—C(1)—C(2)	110 (2)°	109 (2)°
C(1)—C(2)—O(1)	120 (2)	118 (2)
C(1)—C(2)—N(2)	117 (2)	117 (2)
O(1)—C(2)—N(2)	123 (2)	125 (2)
C(2)—N(2)—C(3)	123 (2)	123 (2)
N(2)—C(3)—C(4)	105 (2)	106 (1)
N(2)—C(3)—C(14)	109 (2)	103 (1)
C(14)–C(3)—C(4)	112 (2)	112 (2)
C(3)—C(4)—O(2)	121 (2)	119 (2)
C(3)—C(4)—N(3)	117 (2)	117 (2)
O(2)—C(4)—N(3)	122 (2)	124 (2)
C(4)—N(3)—C(5)	115 (2)	114 (2)
N(3)—C(5)—C(6)	107 (2)	105 (2)
N(3)—C(5)—C(7)	115 (2)	114 (2)
C(6)—C(5)—C(7)	108 (2)	112 (2)
C(5)—C(6)—O(3)	119 (2)	119 (2)
C(5)—C(6)—O(4)	115 (2)	118 (2)
O(3)—C(6)—O(4)	126 (2)	121 (2)
C(5)—C(7)—C(8)	110 (2)	112 (2)
C(7)—C(8)—C(9)	125 (2)	120 (2)
C(7)—C(8)—C(13)	118 (2)	117 (2)
C(9)—C(8)—C(13)	116 (3)	122 (3)
C(8)—C(9)—C(10)	124 (2)	121 (2)
C(9)—C(10)–C(11)	113 (2)	114 (2)
C(10)–C(11)–C(12)	121 (3)	122 (3)
C(10)–C(11)–O(5)	111 (2)	113 (2)
C(12)–C(11)–O(5)	126 (3)	125 (3)
C(11)–C(12)–C(13)	123 (3)	127 (3)
C(8)—C(13)–C(12)	118 (3)	114 (3)
C(3)—C(14)–C(15)	115 (2)	112 (2)
C(14)–C(15)–C(16)	106 (2)	112 (3)
C(14)–C(15)–C(17)	108 (2)	105 (2)
C(16)–C(15)–C(17)	111 (2)	106 (3)
C(18)–C(19)–O(9)	107 (4)	
C(19)–O(9)—C(20)	115 (4)	
O(9)—C(20)–C(21)	118 (5)	

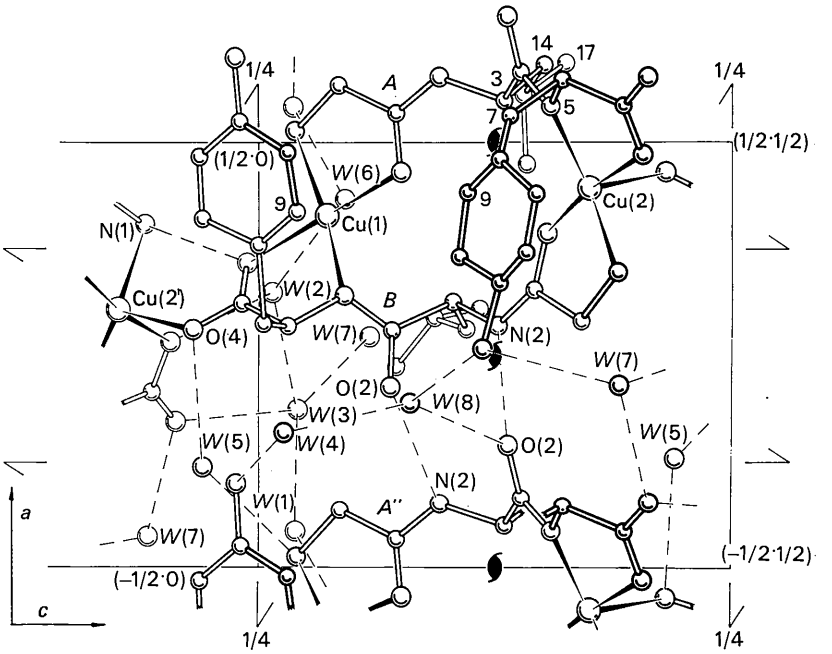


Fig. 2. Projection of a part of the structure down the *b* axis. The dashed lines are hydrogen bonds. The numbers indicate carbon atoms.

Conformational angles and sheet structure

Least-squares planes were calculated through the four atoms of each of four peptide groups $C^{\alpha}C'ON$ (planes 3, 5, 7 and 8, Table 4) and the acid groups (planes 4 and 6, Table 4). These groups are planar. Due to the partial double-bond character of the C′–N bond the $C^{\alpha+1}$ atom is supposed to be in the plane of $C^{\alpha}C'ON$. In the present structure the largest deviation occurs for C(3*A*) (plane 3), 0·16 Å. If least-squares plane 3 is recalculated including the parameters of C(3*A*), the deviations from the plane are 0·04 Å for C(3*A*), while the largest deviation is for N(2*A*) 0·06 Å.

An alternative way to describe the peptide backbone is by the use of conformational angles. The conventions and nomenclature adopted for this purpose (Edsall, Flory, Kendrew, Liquori, Némethy, Ramachandran & Scheraga, 1966) will be used. The angles used are shown in Fig. 1. The calculated values of these angles in the backbone of the peptides for the present structure are given in Table 8. The standard deviation of all conformational angles is estimated to be 2°. The similarity of the φ, ψ and ω angles for peptides *A* and *B* illustrates that both have the same conformation.

Table 8. *Conformational angles*

The ψ_3^1 and ψ_3^2 are the conformational angles involving O(3) and O(4) respectively. The φ_1 is the rotational angle of C(1)–C(2) with respect to Cu–N(1). The hydrogen atoms are not located in the structure and ω and φ angles are therefore calculated on the assumption that the bonds C′–N, N–H and N–C^{α} form a planar set. The columns APPS and PPS give the conformational angles for the antiparallel pleated sheet and parallel pleat structure respectively which were originally proposed by Pauling & Corey (1953). The models proposed by Pauling & Corey were built from D-amino acids. The values given in the Table are for the same models but now built from L-amino acids. The original use of D-amino acids has resulted in confusion in the scientific literature and textbooks (Day & Ritter, 1967). For example the φ angles for pleated sheet structures in Table 3 of Edsall *et al.* (1966) are incorrect.

Backbones

	A	*B*	APPS	PPS
φ_1	5°	2°		
ψ_1	1	7		
ω_1	6	0		
φ_2	50	49	39°	59°
ψ_2	318	317	318	295
ω_2	4	1	0	0
φ_3	31	38		
ψ_3^1	341	337		
ψ_3^2	167	168		

Side chains

		A	*B*
N(2)–C(3)—C(14)–C(15)	(X_2^1)	307°	176°
C(4)–C(3)—C(14)–C(15)		192	62
C(3)–C(14)–C(15)–C(16)	(X_3^{22})	293	268
C(3)–C(14)–C(15)–C(17)	(X_3^{21})	174	153
N(3)–C(5)—C(7)—C(8)	(X_3^1)	54	58
C(6)–C(5)—C(7)—C(8)		295	300
C(5)–C(7)—C(8)—C(9)	(X_3^{22})	281	274
C(5)–C(7)—C(8)—C(13)	(X_3^{21})	90	84

The subscript and superscript notation used for *X* is that suggested by Ramachandran (1968).

All peptide linkages are *trans*, *i.e.* the C′–O bonds are *trans* to the N–H bonds, which is shown by the values for the ω angles: 6, 0, 4 and 1°. The ideal value for a planar *trans* peptide linkage is 0°.

Ramachandran, Ramakrishnan & Sasisekharan (1963) worked out two-dimensional conformational maps correlating the allowed ranges of the dihedral angles around N–C and C –C′ in peptides containing residues with a β-carbon atom. New conventions for these angles were established by Edsall *et al.* (1966), while the subject of conformation of polypeptides and proteins has recently been reviewed (Ramachandran, 1968). This map shows small allowed areas for the right- and left-handed α-helices, and a much larger area with φ approximately between 20 and 120°, and ψ between 270 and 360°, in which the peptide chain is stretched, which is the β-structure. The conformational angles of the parallel pleated sheet structure (PPS) and anti-parallel pleated sheet structure (APPS), originally proposed by Pauling & Corey (1953) fall within the area of β-structure on the conformational map.

The φ_1 and ψ_1 values involve a glycyl residue and the φ_3 and ψ_3 angles the end-acid group. Only the φ_2 and ψ_2 angles can therefore be properly compared with standard values.

It was pointed out that in the present structure peptide dimers can be recognized. Peptide *A* is pointed in the positive **c** direction while peptide *B* is pointed in the negative **c** direction. This general feature is the basic pattern of the antiparallel pleated sheet structure. If this is the basic feature of the structure one would expect the appropriate peptide–peptide hydrogen bonds. These exist in this structure and are shown in Figs. 2 and 3: the H-bonds N(2*B*)–O(2*A*″) and O(2*B*)–N(2*A*″) are formed by the second amino acid residue. The H-bonds of the neighboring residues should then be pointed in the opposite direction and be formed by N(1), O(1), N(3) and O(3) of peptides *A* and *B*. These atoms do not form H-bonds but instead form the coordination bonds, in the expected direction, with the copper ions. One generalization of the structure therefore is that it is an APPS structure in which coordination bonds replace hydrogen bonds. The following points conform with this generalization. (*a*) The ψ_2 angles are very close to those calculated for the APPS model, although the φ_2 angles deviate by about 10°. (*b*) The translation distance between chains running in the same direction was proposed to be 9·50 Å by Pauling & Corey (1953); this compares well with the dimension of the *a* axis in this structure of 9·316 Å. (*c*) Pauling & Corey predicted a repeat distance of 6·68 Å along the chain; that same repeat distance in this structure is the distance between C(1) and C(5), which is 6·64 Å in molecule *A* and 6·59 Å in molecule *B*. (*d*) The φ_3 and ψ_3' angles are comparable with those for an APPS structure as they should be with the third residue having a β-carbon atom and the complex bonds replacing the hydrogen bonds of N(3)

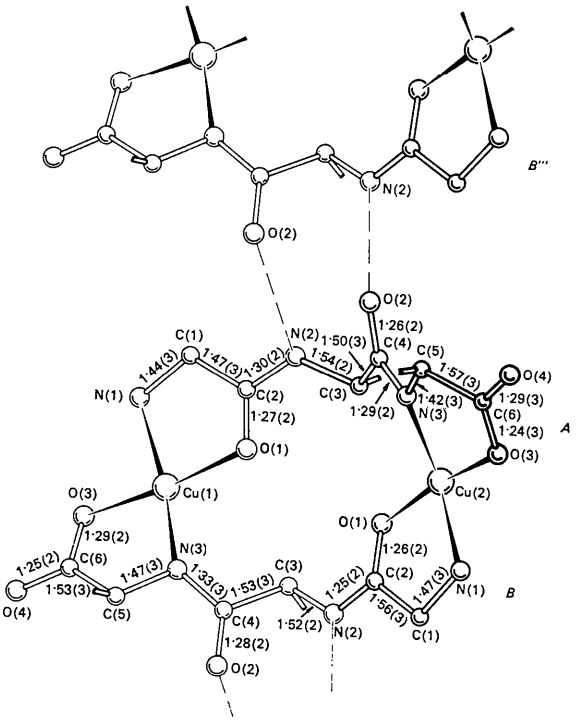


Fig. 3. Bond distances in the backbones of the peptides. The standard deviation for the last digit is given in parentheses.

and O(4). (*e*) The peptide itself can be crystallized (Hossain & van der Helm, 1969) and the observations obtained so far indicate that it is an APPS structure. On the basis of these observations we suggest that in general the conformational changes in peptides as a result of chelation can be expected to be small when peptide chelates are formed at neutral *p*H values.

The only large difference between the conformations of the two peptide units is found in the orientations of the isopropyl groups of the leucyl residues: C(14)–C(15) and N(2)–C(3) are *trans* for *B* and skewed for *A*. As noted before the C(7)–C(8) bond has to be skewed both with respect to N(3)–C(5) and C(5)–C(6) for the aromatic ring to be located below the basal plane of the square-pyramidal copper coordination. Table 8 shows that this is the case for both tyrosyl residues.

Hydrogen bonding

The hydrogen bonds involved in the formation of the sheet structure have been mentioned. The sheets are linked in the **c** direction by the complex bond O(4*B*)–Cu(2′) and another peptide–peptide hydrogen bond: N(1*B*) (*b*)–O(3*B*) (Fig. 2, Table 9).

Fig. 4 shows a projection of structure down the *a* axis and is therefore a view approximately parallel to the peptide chains. The cavities between dimers are either filled by ether molecules or water molecules.

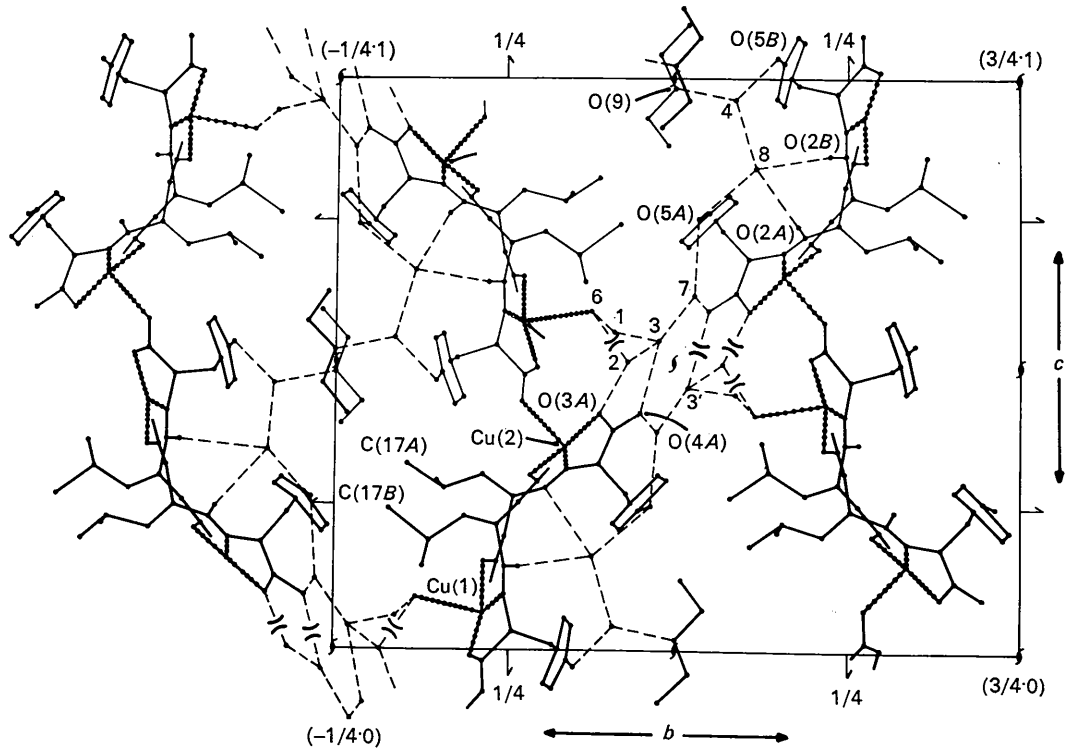


Fig. 4. Projection of the structure down the *a* axis. Atoms N(1) and C(1) for both independent peptides are not shown in this projection. The numbers indicate water molecules. Dashed lines are hydrogen bonds.

The ether molecules are stacked as a column around the 2_1 axis parallel to **a**. This column is used to fill the hole that is surrounded by groups of similar polarity, *i.e.* isopropyl groups and phenolic groups. In Fig. 4 this can be seen for instance for the column located at $(-\frac{1}{4},\frac{1}{2})$. This explains the necessity of ether during the crystallization process. The ether oxygen atom forms an H-bond with *W*(4). The other cavity in the structure, located at $(\frac{1}{4},\frac{1}{2})$, is filled by water molecules.

Two hydrogen-bond spirals are present in the structure. One of these is formed by *W*(7), *W*(3) and the peptide oxygen atom O(4*A*) and their symmetry equivalents by a two fold screw axis. The first spiral is connected to a second one formed by *W*(6), *W*(1), *W*(3) and *W*(2). It is doubtful if *W*(1)–*W*(3) is indeed a hydrogen bond. Although the spirals are primarily used to fill the cavity, the hydrogen bond *W*(2)–O(3*A*) and the participation of O(4*A*) and *W*(6), coordinated to Cu(1), indicates their packing function. The packing is also aided by a hydrogen-bond chain which extends between the ether column and the first hydrogen-bond spiral, *i.e.* O(9), *W*(4), *W*(8), O(5*A*) and *W*(7). This chain has several branches: *W*(4)–O(5*B*), *W*(8)–O(2*B*) and *W*(8)–O(2*A*). The water molecule that has not been considered yet, *W*(5), forms two hydrogen bonds: *W*(5)–O(4*B*) and *W*(5)–N(1*A*). The hydrogen bonds are listed in Table 9, parts *A*, *B* and C, and shown in Figs. 2 and 4. The bond not shown in the Figures is *W*(1)–N(1*A*), because the angle C(1*B*)–N(1*B*)–*W*(1) makes it doubtful that this is indeed hydrogen bond. The alternative location of *W*(1), *i.e.* *W*(1*), with

Table 9. *Intermolecular distances less than* 3.50 Å

The atoms in the first column have the coordinates given in Table 1. The small letters following atoms in the second column indicate the operations to be performed on the coordinates of those atoms as given in Table 1.

a	$x-1$,	y,	z
b	$-x+\frac{1}{2}$,	$-y$,	$z-\frac{1}{2}$
c	$-x$,	$y-\frac{1}{2}$,	$-z+\frac{1}{2}$
d	$-x+1\frac{1}{2}$,	$-y$,	$z+\frac{1}{2}$
e	$-x+\frac{1}{2}$,	$-y$,	$z+\frac{1}{2}$
f	$x-\frac{1}{2}$,	$-y-\frac{1}{2}$,	$-z$
g	$-x+1$,	$y-\frac{1}{2}$,	$-z+\frac{1}{2}$
h	$-x-\frac{1}{2}$,	$-y$,	$z-\frac{1}{2}$

A Peptide–peptide H-bonds

N(2*B*)–O(2*A*)	(*a*)	2·85 Å
O(2*B*)–N(2*A*)	(*a*)	2·80
O(3*B*)–N(1*B*)	(*b*)	2·91

B H_2O–H_2O H-bonds

W(6)–*W*(2)		2·96 Å	*W*(3)–*W*(7)	2·75 Å
W(8)–*W*(4)		2·75	*W*(3)–*W*(2) (*f*)	2·81
W(1)–*W*(3)		3·23	*W*(1)–*W*(6) (*a*)	2·55

C H_2O–peptide and ether H-bonds

W(4)–O(5*B*)	(*a*)	2·45 Å	*W*(2)–O(3*A*)	(*b*)	2·70 Å
W(7)–O(5*A*)	(*c*)	2·93	*W*(3)–O(4*A*)	(*b*)	2·87
W(8)–O(2*B*)		3·06	*W*(7)–O(4*A*)	(*g*)	2·72
W(8)–O(5*A*)		2·86	*W*(8)–O(2*A*)	(*a*)	2·85
W(5)–O(4*B*)		2·99	*W*(1)–N(1*A*)	(*a*)	3·09
W(5)–N(1*A*)	(*a*)	2·90	O(9) –*W*(4)	(*c*)	2·90

D *W*(1*) H-bonds

W(1*)–*W*(7)	(*f*)	2·96 Å
W(1*)–*W*(5)		2·98
W(1*)–N(1*A*)	(*a*)	2·87

E All other van der Waals distances less than 3·50 Å

N(1*B*)—O(4*B*)	(*e*)	3·28 Å	C(17*A*)–O(5*B*)	(*d*)	3·48 Å
W(7) —*W*(2)		3·46	*W*(4) —C(11*B*)	(*a*)	3·34
W(7) —C(11*A*)	(*c*)	3·48	*W*(4) —C(10*B*)	(*a*)	3·33
O(1*B*)—C(3*A*)		3·12	O(3*A*)—O(4*B*)	(*e*)	2·98
O(1*B*)—O(4*B*)	(*e*)	3·19	*W*(6) —C(15*B*)		3·19
C(3*B*)—O(1*A*)		3·09	*W*(8) —C(10*A*)		3·45
C(11*B*)–N(1*A*)		3·49	*W*(4) —C(7*B*)		3·49
N(3*A*) –O(4*B*)	(*e*)	3·29	*W*(5) —O(5*B*)	(*a*)	3·39
O(2*B*)—C(1*A*)	(*a*)	3·16	*W*(5) —C(1*A*)	(*a*)	3·23
O(2*B*)—C(2*A*)	(*a*)	3·43	*W*(5) —N(3*A*)	(*b*)	3·50
C(1*B*)—O(2*A*)	(*a*)	3·17	*W*(1*) –*W*(6)	(*a*)	3·31
C(2*B*)—O(2*A*)	(*a*)	3·45	*W*(1*) –N(1*B*)	(*h*)	3·25
W(3) —*W*(2)		3·28	*W*(1*) –C(1*B*)	(*h*)	3·34
O(2*B*)—O(2*A*)	(*a*)	3·20	*W*(1*) –O(4*A*)	(*b*)	3·26

occupancy of $\frac{1}{3}$, has better hydrogen bonds than $W(1)$ itself (Table 9, *D*). No attempt has been made to indicate donors and acceptors in the hydrogen bonding, because the hydrogen atoms were not located and several bonding schemes are feasible. The large temperature motion of the water molecules (Table 1) is probably the reason that only few reflections with $2\theta > 90°$ could be observed. All other intermolecular distances less than 3·50 Å are shown in Table 9, *E*.

In summary it is interesting to note that the structure is sufficiently large and complicated for it to be possible to recognize so called hydrophilic and hydrophobic regions.

The authors extend their gratitude to Dr F. R. Ahmed who supplied them with copies of his IBM 360 programs, and they are indebted to the Computing Center of the University of Oklahoma for putting computer time at their disposal.

References

AHMED, F. R. (1966*a*). *S.F.L.S. program*, NRC-10. Ottawa: National Research Council.
AHMED, F. R. (1966*b*). *Fourier program*, NRC-8. Ottawa: National Research Council.
BROMAN, L., MALMSTRÖM, B. G., AASA, R. & VÄNNGÅRD, T. (1963). *Biochim. Biophys. Acta*, **75**, 365.
DAY, R. A. & RITTER, E. J. (1967). *J. Chem. Educ.* **44**, 761.
FREEMAN, H. C. (1967). *Advanc. Protein Chem.* **22**, 257.
FREEMAN, H. C., SCHOONE, J. C. & SIME, J. G. (1965). *Acta Cryst.* **18**, 381.
FREEMAN, H. C. & SZYMANSKI, J. T. (1967). *Acta Cryst.* **22**, 406.
FREEMAN, H. C. & TAYLOR, M.R. (1965). *Acta Cryst.* **18**, 939.
HOSSAIN, M. B. & VAN DER HELM, D. (1969). Unpublished results.
EDSALL, J. T., FLORY, P. J., KENDREW, J. C., LIQUORI, A. M., NÉMETHY, G., RAMACHANDRAN, G. N. & SCHERAGA, H. A. (1966). *Biopolymers*, **4**, 121.
International Tables for X-ray Crystallography (1962). Vol. III, p. 202. Birmingham: Kynoch Press.
LEVINE, W. G. & PEISACH, J. (1962). *Biochim. Biophys. Acta*, **63**, 528.
MARSH, R. E. & DONOHUE, J. (1967). *Advanc. Protein Chem.* **22**, 235.
PAULING, L. & COREY, R. (1953). *Proc. Nat. Acad. Sci. Wash.* **39**, 253.
PIPPY, M. E. (1967). *Structure Factor Tabulation*, NRC-23. Ottawa: National Research Council.
RAMACHANDRAN, G. N. (1968). *Advanc. Protein Chem.* **23**, 283.
RAMACHANDRAN, G. N., RAMAKRISHNAN, C. & SASISEKHARAN, V. (1963). *J. Mol. Biol.* **7**, 95.
SCHOMAKER, V., WASER, J., MARSH, R. E. & BERGMAN, G. (1959). *Acta Cryst.* **12**, 600.
TATSCH, C. E. & VAN DER HELM, D. (1969). VIIIth General Assembly and International Congress of the International Union of Crystallography. Abstract XV-49.
VAN DER HELM, D. & FRANKS, W. A. (1968). *J. Amer. Chem. Soc.* **90**, 5627.

Acta Cryst. (1971). B**27**, 1310

The Crystal and Molecular Structure of 1,8-Dinitrosonaphthalene

BY C. K. PROUT, T. S. CAMERON, R. M. A. DUNN, O. J. R. HODDER AND D. VITERBO*
Chemical Crystallography Laboratory, South Parks Road, Oxford, England

(*Received* 6 *July* 1970)

1,8-Dinitrosonaphthalene crystallizes in space group $P2_1/b$ with $a=13·13$, $b=16·01$, $c=3·89$ Å, $=92·7°$ and with $Z=4$. The structure was determined from photographic X-ray data and refined by the least-squares method to $R=0·078$ for 1009 reflexions. The molecule is in the form of an internal nitroso dimer with an N–N bond 1·38 Å long. The naphthalene nucleus shows similar distortions to those found in the acenaphthenes.

Introduction

It is possible to devise a number of plausible molecular structures for 1,8-dinitrosonaphthalene. Data from chemical and spectroscopic measurements do not allow an unambiguous distinction between these structures. Therefore the structure of the crystal was determined by X-ray diffraction.

* Permanent address: Istituto di Chimica Fisica della Università di Torino, Corso Massimo d'Azeglio 48, Torino, Italy.

Experimental

Red-brown crystals of the compound were supplied by Professor M. C. Whiting of Bristol University (Whiting, 1969). The crystal data are: $C_{10}H_6N_2O_2$, $M=186·2$, monoclinic,

$a=13·13\pm0·03$, $b=16·01\pm0·03$, $c=3·89\pm0·01$ Å,
$\alpha=92·7\pm0·3°$,
$Z=4$,
$D_m=1·51$ g. cm^{-3} by flotation,
$D_c=1·513$ g. cm^{-3} Cu $K\alpha$ 1·5418 Å,
$\mu=9·21$ cm^{-1}.