44. *Amino-oxy-derivatives. Part I. Some α-Amino-oxy-acids and α-Amino-oxy-hydrazides.*

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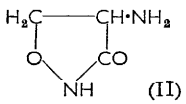
The preparation of some α-amino-oxy-acids and some α-amino-oxy-hydrazides is described.

Certain naturally occurring compounds containing the amino-oxy-group are known to have antibacterial properties. Thus, canavanine (α-amino-γ-guanidino-oxybutyric acid) (I), an amino-acid isolated from the jack bean by Kitagawa and Tomiyama,[1] has been shown to be a potent growth-inhibitor of many organisms [2] and certain viruses.[3] The

(I) $NH{:}C(NH_2)\cdot NH\cdot O\cdot[CH_2]_2\cdot CH(NH_2)\cdot CO_2H$

(III) $R''NH\cdot O\cdot CRR'\cdot CO_2H$



(II)

antibiotic cycloserine (D-4-amino-3-isoxazolidone) (II), a cyclised derivative of α-amino-β-amino-oxypropionic acid, possesses broad-spectrum antibacterial activity.[4] Less complex structures containing the amino-oxy-group, such as hydroxylamine,[5] the simple amino-oxyalkanes,[6] and amino-oxyacetic acid,[7] also exhibit distinct bacteriostatic activity against several Gram-positive and Gram-negative organisms. These facts indicated that it might be worth while to investigate other amino-oxy-compounds as potential antibacterials. This paper describes the synthesis of some α-amino-oxy-acids and -hydrazides.

Although the lower α-amino-oxy-acids (III; R = R′ = R″ = H; R = R″ = H, R′ = Me; R = R″ = H, R′ = Et; R″ = H, R = R′ = Me) have been prepared by various routes,[8–15] none of the higher α-amino-oxy-acids has been described. In general, the syntheses have consisted of the condensation of an α-halogeno-ester with a suitably blocked hydroxylamine derivative, hydrolysis of the resulting intermediate, and isolation as the amino-oxy-acid hydrochloride.

Kitagawa's method,[12,14,15] which used benzhydroxamic acid as the substituted hydroxylamine derivative, was selected for the present work but, in order to simplify the working-up, the α-halogeno-ester was replaced by the α-halogeno-acid. The α-benzamido-oxy-acids were readily isolable solids, which were easily hydrolysed by hydrochloric acid to the α-amino-oxy-acids (III; R′ = R″ = H, R = H·$[CH_2]_n$, where n = 0—8, 10, 12, 14, and 16; R″ = H, R = R′ = Me; R′ = R″ = H, R = Pr^i; R′ = R″ = H, R = $C_6H_{11}\cdot[CH_2]_4$; R′ = R″ = H, R = $[CH_2]_3\cdot CO_2H$).

[1] Kitagawa and Tomiyama, *J. Biochem., Japan*, 1929, **11**, 265.
[2] Volcani and Snell, *J. Biol. Chem.*, 1948, **174**, 893; Horowitz and Srb, *ibid.*, p. 371; Suzuki and Muraoka, *J. Pharm. Soc., Japan*, 1954, **74**, 534; Suzuki, Muraoka, and Konobu, *ibid.*, p. 537; Walker, *J. Biol. Chem.*, 1955, **212**, 207.
[3] Pearson, Lagerborg, and Winzler, *Proc. Soc. Exp. Biol. Med.*, 1952, **79**, 409; Pilcher, Soike, Smith, Trosper, and Folston, *ibid.*, 1955, **88**, 79.
[4] Cuckler, Frost, McClelland, and Solotorovsky, *Antibiotics and Chemotherapy*, 1955, **5**, 191; Welch, Putnam, and Randall, *Antibiotic Medicine*, 1955, **1**, 72.
[5] Gray and Lambert, *Nature*, 1948, **162**, 733.
[6] Andrews, King, and Walker, *Proc. Roy. Soc.*, 1946, *B*, **133**, 29; Fuller and King, *J.*, 1947, 963.
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[11] Werner and Bial, *Ber.*, 1895, **28**, 1374.
[12] Kitagawa and Takani, *J. Biochem. (Japan)*, 1936, **23**, 181.
[13] Borek and Clarke, *J. Amer. Chem. Soc.*, 1936, **58**, 2020.
[14] Kitagawa and Takani, *J. Agric. Chem. Soc., Japan*, 1935, **11**, 1077.
[15] Kitagawa, *J. Biochem., Japan*, 1936, **24**, 107.

Two α-benzamido-oxy-acids have been described previously, namely, benzamido-oxyacetic acid and α-benzamido-oxypropionic acid, the latter as a hygroscopic solid by Kitagawa.[15] There appears to be some confusion in the literature as to the melting point of benzamido-oxyacetic acid: Borek and Clarke [16] gave a value of 123° whilst other workers [8,12,14] reported values closer to 140°. The first preparation of benzamido-oxyacetic acid in this laboratory gave a material that was recrystallised to a constant melting point of 119—121°; however, after several weeks this material melted at 144—145°. All subsequent preparations gave benzamido-oxyacetic acid of m. p. 145—146°. A similar behaviour was noted in the preparation of α-benzamido-oxybutyric acid: two compounds, both analysing correctly but with different melting points and infrared spectra, were obtained.

The lower α-amino-oxy-acids were isolated as hydrochlorides. The nature of the hydrochloride of amino-oxyacetic acid has previously been in doubt: Werner and Sonnenfeld [9] and later, Kitagawa and Takani,[12] reported a monohydrochloride, m. p. 156°; Borek and Clarke [13] reported a half-hydrochloride, m. p. 151°. The present work has shown that the monohydrochloride is the initial product of the hydrolysis of benzamido-oxyacetic acid; however, hydrogen chloride is lost on crystallisation, giving the half-hydrochloride. On treatment with ethereal hydrogen chloride, the half-hydrochloride is converted into the unstable monohydrochloride, m. p. 115—116°. The hydrochlorides of the other amino-oxy-acids gradually lose hydrogen chloride.

The α-amino-oxy-acids (III; R′ = R″ = H, R = H·$[CH_2]_n$, where n = 6, 7, 8, 10, 12, and 14; R′ = R″ = H, R = C_6H_{11}·$[CH_2]_4$) were esterified by diazomethane and converted into hydrazides.

The α-amino-oxy-acids possessed weak antibacterial activity *in vitro* against *Staph. aureus* and *E. coli* (inhibiting them at a concentration of 300 p.p.m. compared with sulphathiazole at 5 p.p.m.) and stronger activity against *M. tuberculosis* (inhibiting it at 37 p.p.m. compared with isoniazid at 0·2 p.p.m.); the activity did not vary with increasing molecular weight. Solubility difficulties precluded the testing of α-amino-oxytetradecanoic acid and higher acids. The α-amino-oxy-hydrazides were in general more active than the corresponding acids; the activity varied with molecular weight and reached a maximum in the C_{12} and C_{14} compounds (19 p.p.m. inhibiting the growth of *Staph. aureus* and *E. coli*). The hydrazides were more soluble than the acids and thus it was possible to test α-amino-oxyhexadecanohydrazide. α-Amino-oxytetradecanohydrazide proved to be strongly active against several Gram-positive organisms (*e.g.*, *Strept. pyrogenes* was inhibited by *ca.* 5 p.p.m.) and was more active than tetradecanohydrazide. The complete results of these antibacterial tests will be published elsewhere.

Experimental

Light petroleum had b. p. 40—60° except where otherwise stated. Infrared spectra were measured with a Grubb–Parsons D.B.1/S.4. spectrometer.

α-Bromo-acids.—When not available commercially, these were prepared by the method of von Auwers and Bernhardi.[17] After removal of the excess of bromine, the bromo-acyl bromide was poured on crushed ice and allowed to come to room temperature: hydrolysis was completed on a steam-bath. The product was extracted into benzene, and the extract washed free from hydrobromic acid. Evaporation gave the bromo-acid which was purified by crystallisation or distillation.

2-Bromo-6-cyclohexylhexanoic Acid.—An intimate mixture of 6-cyclohexylhexanoic acid (50 g.) and red phosphorus (5 g.) was treated with bromine (40 ml.) during 4 hr. The product, isolated as above, gave *2-bromo-6-cyclohexylhexanoic acid* (51 g.), b. p. 150°/0·1 mm. (Found: C, 51·1; H, 7·4. $C_{12}H_{21}O_2Br$ requires C, 50·8; H, 7·6%).

Dimethyl α-Bromoadipate.—This was prepared by the method of Buu-Hoï and Demerseman.[18]

[16] Borek and Clarke, *J. Biol. Chem.*, 1938, **125**, 479.
[17] von Auwers and Bernhardi, *Ber.*, 1891, **24**, 2209.
[18] Buu-Hoï and Demerseman, *J. Org. Chem.*, 1953, **18**, 649.

α-Benzamido-oxy-acids.—Benzhydroxamic acid [19] (0·1 mole) and sodium hydroxide (0·2 mole) in aqueous ethanol were refluxed for 5 hr. with the α-bromo-acid (0·1 mole). After evaporation of the solvent, the residue was dissolved in water, acidified, and extracted with ethyl acetate. The organic layer was dried and evaporated and the resulting solid crystallised. Compounds prepared in this way are given in Table 1.

The initial preparation of benzamido-oxyacetic acid gave a solid which on crystallisation from ethyl acetate–light petroleum had m. p. 119—121°. After several weeks the m. p. was 144—145°. Subsequent preparations gave benzamido-oxyacetic acid, m. p. 145—146° (from ethyl acetate).

Anomalous results were noted in the preparation of α-benzamido-oxybutyric acid. One preparation gave a product (1), m. p. 125—127° (from ethyl acetate–light petroleum), $\nu_{max.}$ 3245s, 3005s, 2565m, 2510m, 1720vs, 1630vs, 1570s, 1505s, 1480s, 1430m, 1380w, 1305m, 1290m, 1260m, 1215s, 1150m, 1135m, 1100m, 1085m, 1075w, 1050m, 1035m, 1020s, 962m, 943m, 922m, 893m, 800m, 774m, 711m, 686s cm.$^{-1}$ (KBr disc). The product (2) from a similar preparation had m. p. 134—135°, $\nu_{max.}$ 3300m, 2985m, 2930m, 2595m, 2480w, 1695s, 1625s, 1575s, 1505s, 1475s, 1460m, 1430m, 1385w, 1340w, 1325m, 1305s, 1280s, 1240s, 1150m, 1135m, 1100w, 1085m, 1055m, 1035w, 1020m, 948m, 942m, 911m, 898m, 894w, 790m, 789m, 742m, 717s, 692s, 680m cm.$^{-1}$ (KBr disc).

α-Benzamido-oxyadipic Acid (III; R = H, R′ = $[CH_2]_3{\cdot}CO_2H$, R″ = Bz).—Benzhydroxamic acid (2·8 g.) and sodium hydroxide (0·8 g.) in aqueous ethanol were refluxed for 5 hr. with

TABLE 1. *α-Benzamido-oxy-acids*, $Bz{\cdot}NH{\cdot}O{\cdot}X{\cdot}CO_2H$.

Parent acid, $HX{\cdot}CO_2H$	M. p.	Formula	Found (%) C	Found (%) H	Found (%) N	Reqd. (%) C	Reqd. (%) H	Reqd. (%) N
Acetic [a] (1st prep.)	119—121° [d]	$C_9H_9O_4N$	55·8	4·4	7·0	55·4	4·6	7·2
Acetic [b]	145—146	$C_9H_9O_4N$	55·1	4·3	6·8	55·4	4·6	7·2
Propionic [b]	132—133	$C_{10}H_{11}O_4N$	57·4	5·1	6·6	57·4	5·3	6·7
Butyric [a] (1)	125—127	$C_{11}H_{13}O_4N$	59·3	5·9	6·5	59·2	5·9	6·3
Butyric [a] (2)	134—135	$C_{11}H_{13}O_4N$	59·3	5·8	5·8	59·2	5·9	6·3
Isobutyric [a]	79—81	$C_{11}H_{13}O_4N$	59·4	5·6	6·5	59·2	5·9	6·3
Valeric [a]	119—120	$C_{12}H_{15}O_4N$	60·8	6·5	5·8	60·8	6·4	5·9
Isovaleric [b]	159—160	$C_{12}H_{15}O_4N$	61·1	6·2	5·5	60·8	6·4	5·9
Hexanoic [a]	125—126	$C_{13}H_{17}O_4N$	62·2	6·9	5·6	62·1	6·8	5·6
Heptanoic [b]	117—118	$C_{14}H_{19}O_4N$	63·2	7·6	5·2	63·4	7·2	5·3
Octanoic [b]	118—120	$C_{15}H_{21}O_4N$	64·1	7·2	5·0	64·5	7·6	5·0
Nonanoic [b]	106—107	$C_{16}H_{23}O_4N$	65·2	7·9	4·8	65·5	7·9	4·8
Decanoic [a]	108—109	$C_{17}H_{25}O_4N$	66·8	8·7	5·0	66·3	8·3	4·6
Dodecanoic [c]	108—109	$C_{19}H_{29}O_4N$	67·6	8·5	3·9	68·0	8·7	4·2
Tetradecanoic [c]	115—116	$C_{21}H_{33}O_4N$	69·6	9·1	3·8	69·4	9·2	3·9
Hexadecanoic [c]	115—116	$C_{23}H_{37}O_4N$	70·2	9·4	4·0	70·6	9·5	3·6
Octadecanoic [c]	104—105	$C_{25}H_{41}O_4N$	70·6	9·7	3·2	71·6	9·9	3·3
6-*Cyclohexylhexanoic* [b]	148—149	$C_{19}H_{27}O_4N$	68·2	8·1	4·1	68·2	8·2	4·2

Recrystallised from: (*a*) ethyl acetate–light petroleum, (*b*) ethyl acetate, (*c*) light petroleum (b. p. 100—120°). (*d*) Later 144—145°.

dimethyl α-bromoadipate (5·1 g.). The mixture was cooled, treated with N-sodium hydroxide (40 ml.), and left overnight; the inorganic solid was filtered off and the filtrate evaporated. The residue was taken up in water and extracted with ethyl acetate; the aqueous solution was acidified and extracted with ethyl acetate. Evaporation of the latter extract gave a solid *acid* (1·2 g.) which after crystallising from ethyl acetate–light petroleum had m. p. 163—164° (Found: C, 55·5; H, 5·2; N, 4·7. $C_{13}H_{15}O_6N$ requires C, 55·5; H, 5·4; N, 5·0%).

Hydrolysis of α-Benzamido-oxy-acids.—The benzamido-compound (2 g.) was refluxed for 2 hr. with 5N-hydrochloric acid (20 ml.) [it was necessary to add acetic acid (10 ml.) to dissolve α-benzamido-oxyoctanoic and higher acids]. After cooling, the benzoic acid was filtered off and rejected, the filtrate was evaporated, and the resulting hydrochloride was recrystallised. Compounds prepared in this way are described in Table 2.

Amino-oxyacetic Acid Half-hydrochloride.—The crude hydrolysis product, m. p. 103—107°, from benzamido-oxyacetic acid was dissolved in an equal weight of water, treated with propan-2-ol, and left at 0°; crystals of amino-oxyacetic acid half-hydrochloride were deposited, having

[19] *Org. Synth.*, Coll. Vol. II, p. 67.

TABLE 2. *α-Amino-oxy-acids*, $NH_2{\cdot}O{\cdot}X{\cdot}CO_2H$.

Parent acid, $H{\cdot}X{\cdot}CO_2H$	M. p.	Formula	Found (%) C	H	N	Cl	Reqd. (%) C	H	N	Cl
Propionic, HCl [a] ...	163—164° † [d]	$C_3H_8O_3NCl$	26·0	5·7	9·9	—	25·4	5·7	9·9	—
Butyric, HCl [a] * ...	142 † [e]	$C_4H_{10}O_3NCl$	30·4	6·5	9·1	23·1	30·9	6·5	9·0	22·8
Valeric, HCl [a]	121—123 †	$C_5H_{12}O_3NCl$	34·9	7·3	8·2	20·8	35·4	7·1	8·4	20·9
Isovaleric, HCl [a]	151—152 †	$C_5H_{12}O_3NCl$	34·9	7·1	8·0	21·9	35·4	7·1	8·4	20·9
Hexanoic, HCl [a]	112 †	$C_6H_{14}O_3NCl$	39·4	7·3	7·5	19·1	39·3	7·7	7·6	19·3
Heptanoic, HCl [b] ...	123 †	$C_7H_{16}O_3NCl$	42·4	8·1	6·8	17·6	42·5	8·2	7·1	17·9
Heptanoic [c]	145	$C_7H_{15}O_3N$	52·2	9·4	8·8	—	52·2	9·4	8·7	—
Octanoic [c]	145—146	$C_8H_{17}O_3N$	54·8	9·9	7·7	—	54·9	9·8	8·0	—
Nonanoic, HCl [b]	113—114 †	$C_9H_{20}O_3NCl$	48·1	8·9	6·1	15·3	47·9	8·9	6·2	15·7
Nonanoic [c]	147—148	$C_9H_{19}O_3N$	57·4	9·8	7·2	—	57·1	10·1	7·4	—
Decanoic [c]	149	$C_{10}H_{21}O_3N$	59·2	10·3	7·2	—	59·1	10·4	6·9	—
Dodecanoic, HCl [b] ...	112 †	$C_{12}H_{26}O_3NCl$	54·5	9·7	5·4	13·2	53·8	9·8	5·2	13·2
Dodecanoic [c]	145—147	$C_{12}H_{25}O_3N$	62·3	11·3	5·8	—	62·3	10·9	6·1	—
Tetradecanoic [c]	145—146	$C_{14}H_{29}O_3N$	64·5	11·8	5·3	—	64·8	11·3	5·4	—
Hexadecanoic [c]	147—148	$C_{16}H_{33}O_3N$	66·9	12·0	5·1	—	66·8	11·6	4·9	—
Octadecanoic, HCl [b]	93 †	$C_{18}H_{38}O_3NCl$	60·9	10·5	4·2	10·2	61·4	10·9	4·0	10·1
Octadecanoic [c]	144—146	$C_{18}H_{37}O_3N$	68·4	11·6	4·7	—	68·5	11·8	4·4	—
6-*Cyclohexylhexanoic* [c]	156	$C_{12}H_{23}O_3N$	62·5	10·1	5·8	—	62·8	10·1	6·1	—

Recrystallised from: (*a*) ethanol–ether; (*b*) ethyl acetate; (*c*) aqueous ethanol. (*d*) Lit.,[9,15] 168°. (*e*) Lit.,[10] 148°. * Both α-benzamido-oxybutyric acids gave this compound. † With decomp.

m. p. 149—150° [Found: C, 21·9; H, 4·9; N, 13·2; Cl, 16·6. Calc. for $(C_2H_5O_3N)_2$,HCl: C, 22·0; H, 5·0; N, 12·8; Cl, 16·2%].

Amino-oxyacetic Acid Hydrochloride.—Amino-oxyacetic acid half-hydrochloride (1 g.) was suspended in dry ether and treated with dry hydrogen chloride until the solution was saturated. The *hydrochloride* was collected and washed with dry ether and, after drying *in vacuo* over phosphoric oxide, had m. p. 115—116° (Found: C, 18·1; H, 4·6; N, 11·1; Cl, 28·7. $C_2H_6O_3NCl$ requires C, 18·8; H, 4·7; N, 11·0; Cl, 27·8%). This material gradually reverted to the half-hydrochloride.

α-Amino-oxy-α-methylpropionic Acid Hydrochloride.[11]—Hydrolysis of the benzamido-compound and fractional crystallisation of the product from ethanol–ether gave sparingly soluble hydroxylamine hydrochloride, m. p. and mixed m. p. 151—152°, together with α-amino-oxy-α-methylpropionic acid hydrochloride, m. p. 164—166° (Found: C, 30·4; H, 6·5; N, 8·8; Cl, 22·8. Calc. for $C_4H_{10}O_3NCl$: C, 30·9; H, 6·5; N, 9·0; Cl, 22·8%).

α-Amino-oxyadipic Acid Hydrochloride.—It was not possible to crystallise this compound but a partial purification was achieved by digesting it with boiling light petroleum (b. p. 80—100°). It then had m. p. 130—131° (Found: C, 34·2; H, 6·0; N, 6·6; Cl, 15·3. Calc. for $C_6H_{12}O_5NCl$: C, 33·7; H, 5·7; N, 6·6; Cl, 16·6%).

α-Amino-oxy-hydrazides.—The α-amino-oxy-acid (2 g.) was suspended in ethyl acetate and treated with excess of ethereal diazomethane. The resulting solution was filtered and evaporated to an oil which was taken up in a minimum of methanol, treated with hydrazine hydrate (1 ml.), and left for 2 hr. The product was diluted with water and extracted into ethyl acetate. Evaporation and crystallisation gave the hydrazide. Compounds prepared in this way are given in Table 3.

TABLE 3. *α-Amino-oxy-hydrazides*, $NH_2{\cdot}O{\cdot}X{\cdot}CO{\cdot}NH{\cdot}NH_2$.

Parent acid, $H{\cdot}X{\cdot}CO_2H$	M. p.	Formula	Found (%) C	H	N	Reqd. (%) C	H	N
Octanoic [a]	85—86°	$C_8H_{19}O_2N_3$	51·0	10·1	22·4	50·8	10·1	22·2
Nonanoic [a]	80—81	$C_9H_{21}O_2N_3$	53·3	10·6	20·8	53·2	10·4	20·7
Decanoic [a]	80—81	$C_{10}H_{23}O_2N_3$	55·1	10·7	19·7	55·3	10·7	19·5
Dodecanoic [b]	77·5—78·5	$C_{12}H_{27}O_2N_3$	59·2	10·6	16·9	58·7	11·1	17·1
Tetradecanoic [b]	84—85	$C_{14}H_{31}O_2N_3$	61·0	11·4	15·0	61·5	11·4	15·4
Hexadecanoic [b]	87—88	$C_{16}H_{35}O_2N_3$	64·1	11·5	13·8	63·7	11·7	13·9
6-*Cyclohexylhexanoic* [b] ...	100—101	$C_{12}H_{25}O_2N_3$	59·2	10·1	16·8	59·2	10·4	17·3

Recrystallised from: (*a*) ethyl acetate–light petroleum; (*b*) ethyl acetate.

In the preparation of α-amino-oxydecanohydrazide the intermediate methyl ester was isolated as the hydrochloride. The esterified oil when treated with ethereal hydrogen chloride

failed to give a precipitate; evaporation and crystallisation of the residue from ether–light petroleum gave *methyl α-amino-oxydecanoate hydrochloride*, m. p. 89—90° (Found: C, 51·7; H, 9·6; N, 5·3; Cl, 14·2. $C_{11}H_{24}O_3NCl$ requires C, 52·1; H, 9·5; N, 5·5; Cl, 14·0%).

Tetradecanohydrazide.—This compound prepared in a similar manner and crystallised from ethyl acetate had m. p. 108—109° (lit.,[20] m. p. 108—109°).

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[20] Pajari, *Fette u. Seifen*, 1944, **51**, 347.