(0.016 mole) was added a cooled solution of 0.43 g. of sodium in 15 ml. of absolute ethyl alcohol. The mixture was kept overnight (at temperature not above 20°). Orange crystals of 2,6-distyryl-γ-pyrone 0.75 g. (0.0025 mole) m.p. 166°, separated. After crystallization from ethyl alcohol, yellow flakes, m.p. 168°, no depression on admixture with an authentic sample,[8] were obtained.

Anal. Calcd. for $C_{21}H_{16}O_2$: C, 84.0; H, 5.33. Found, C, 83.5; H, 5.6.

The *trans*-diaroylethylenes were prepared according to the method of Conant and Lutz.[10]

(A). Experiments with 2,6-Distyryl-γ-pyrone. (i). **Action of *trans*-Dibenzoylethylene on 2,6-Distyryl-γ-pyrone.**—A mixture of 2,6-distyryl-γ-pyrone, 1 g. (0.0033 mole), and *trans*-dibenzoylethylene, 1.55 g. (0.0066 mole), in anisole, 50 ml., was refluxed for 25 hr. Anisole was distilled in vacuum. The oily residue was dissolved in the least amount of chloroform and petroleum ether (b.p. 60–80°) was added dropwise till a slight turbidity appeared, kept overnight, whereby colorless crystals 0.25 g. (0.0004 mole) of **4a,5,6,7-tetrahydro-5,6-dibenzoyl-7-phenyl-2-styrylchromone (IVa)** m.p. 238° separated. After recrystallization from a mixture of alcohol and benzene, colorless crystals, m.p. 242°, were obtained.

Anal. Calcd. for $C_{37}H_{28}O_4$: C, 82.83; H, 5.22. mol. wt. 536. Found: C, 82.3; H, 5.4. mol. wt. 532.

(ii). **The above experiment was repeated using 2,6-distyryl-γ-pyrone**, 1 g. (0.0033 mole), and *trans*-dibenzoylethylene, 3.1 g. (0.0132 mole), and heating was continued for 35 hr. A 0.3-g. sample (0.00055 mole) of the adduct (IVa), m.p. 242°, no depression on admixture with an authentic sample, was obtained.

(iii). **Attempted Condensation with Hydroxylamine.**—To 0.3 g. (0.0057 mole) of IVa in 200 ml. of ethyl alcohol, a mixture of 0.78 g. (0.00114 mole) of hydroxylamine hydrochloride, and sodium acetate, 1.2 g. (0.0114 mole), was added. The mixture was refluxed for 3 hr., cooled, and the precipitate which formed filtered, washed with water, dried, and crystallized from a mixture of alcohol and benzene, whereby, colorless crystals of IVa, no depression on admixture with an authentic sample, were obtained.

(iv). **Attempted Condensation with Other Ketonic Reagents.**—When repeating experiment iii using phenylhydrazine hydrochloride and sodium acetate or 2,4-dinitrophenylhydrazine, IVa was recovered unchanged. Table II shows the experimental details of other Diels-Alder reactions with 2,6-distyryl-γ-pyrone.

(B). Experiments with 2-Styrylchromone (IX), 2-Styrylkhellin (XIa), and 2-Stryrylvisnagin (XIb). Action of *p,p*-Dichlorodibenzoylethylene (IIIc) on 2-styrylchromone.—A mixture of 2-styrylchromone,[11] 1 g. (0.004 mole), and *trans*-*p,p'*-dichlorodibenzoylethylene, 1.22 g. (0.004 mole), dissolved in 40 ml. of anisole was heated for 30 hr. Anisole was distilled in vacuum, the oily residue crystallized from benzene–petroleum ether (60–80°), whereby, 0.05 g. (0.0004 mole) of 2-styrylchromone (IX), m.p. and mixed m.p. with an authentic sample 131°, separated. On concentrating the mother liquor, almost colorless crystals 0.15 g. (0.00027 mole) of **1,2,3,9a-tetrahydro-1,2-*p,p'*-dichlorodibenzoyl-9-oxo-3-phenyl-xanthene (X)**, m.p. 212°, separated, which after recrystallization from a mixture of alcohol–benzene, gave colorless crystals, m.p. 215°.

Anal. Calcd. for $C_{33}H_{22}O_4Cl_2$: C, 71.6; H, 3.99; Cl, 12.87. Found: C, 71.1; H, 4.1; Cl, 12.4.

Table III shows the experimental details of other Diels-Alder reactions with 2-styrylfurochromones. For the preparation of 2-styrylkhellin, and 2-styrylvisnagin see Schönberg, Mustafa, and Aziz.[7]

(10) J. B. Conant and R. E. Lutz, *J. Am. Chem. Soc.*, **45**, 1303 (1923).

(11) U. S. Cheema, K. C. Gulati, and K. Venkataraman, *J. Chem. Soc.*, 925 (1932).

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β-Aminoxy-D-alanine

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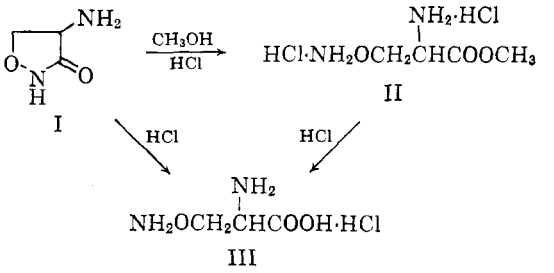
In the course of our work on cycloserine (I), we became interested in the preparation of its hydrolysis product, β-aminoxy-D-alanine (III). This amino acid is related structurally to another aminoxy amino acid, canaline,[1] which was obtained from the interesting guanidinoxy amino acid, canavanine.[2] In 1957, Nyberg and Christensen,[3] in their paper describing the synthesis of both DL-canaline and DL-canavanine, reported an unsuccessful attempt to synthesize β-aminoxy-DL-alanine. They concluded that the aminoxy group was rapidly degraded to a hydroxyl group in hot acid. Under the conditions which we used, the aminoxy group remained intact.

This note describes the preparation of β-aminoxy-D-alanine (III) by two routes. The methanolysis of I giving the ester II has been described by



Kuehl, *et al.*,[4] and is analogous to reactions used in the synthesis[5] of I. The hydrolysis of the ester II in 6 *N* hydrochloric acid at 90° gave III in 68% yield. Direct hydrolysis of cycloserine using 6 *N* hydrochloric acid at 60° gave III in 41% yield. The conditions (time, temperature, and acid concentrations) used in both hydrolyses were approxi-

(1) M. Kitagawa, *J. Agr. Chem. Soc. Japan*, **15**, 267 (1939); *Chem. Abstr.*, **34**, 1196 (1940).

(2) M. Kitagawa and T. Tomiyama, *J. Biochem.* (Japan), **11**, 265 (1929).

(3) D. D. Nyberg and B. E. Christensen, *J. Am. Chem. Soc.*, **79**, 1222 (1957).

(4) F. A. Kuehl, Jr., F. J. Wolf, N. R. Trenner, R. L. Peck, Eugene Howe, B. D. Hunnewell, G. Downing, E. Newstead, K. Folkers, R. P. Buhs, I. Putter, R. Ormond, J. E. Lyons, and L. Chaiet, *ibid.*, **77**, 2344 (1955).

(5) C. H. Stammer, A. N. Wilson, C. F. Spencer, F. W. Bachelor, F. W. Holly, and K. Folkers, *ibid.*, **79**, 3236 (1957).

mately optimum as indicated by paper chromatographic studies. The structure of III was confirmed by its hydrogenolysis to D-serine and its conversion to an isopropylidene derivative.

The antibacterial activity of III was considerably lower than that of cycloserine.

Experimental

All melting points were taken on a Kofler micro hot stage. Paper chromatography data is reported using the system previously described.[6] Radial chromatograms using the methyl ethyl ketone–pyridine–water system (MPW, 20:5:8) were run and the spots were detected with ninhydrin (N) reagent.

β-Aminoxy-D-alanine Methyl Ester Dihydrochloride (II).—Hydrogen chloride was passed for 20 min. into 300 ml. of methanol contained in a 500-ml. round-bottomed flask immersed in an ice bath. To the resulting solution was added 25 g. of cycloserine and the solution was refluxed 3 hr. After standing overnight at room temperature, the solution was poured slowly into 1500 ml. of ethyl acetate. The precipitated oil crystallized when the mixture was cooled in an ice bath for about 1 hr. The solid was collected on a filter and washed with ethyl acetate and ether. The crude β-aminoxy-D-alanine methyl ester dihydrochloride, which weighed 39.9 g., was dissolved in 150 ml. of hot methanol. The solution was filtered and 200 ml. of ethyl acetate was added to the filtrate. The mixture was placed in the refrigerator for several hours. Twenty-nine grams (52%) of β-aminoxy-D-alanine methyl ester dihydrochloride, m.p. 135–139°, $[\alpha]^{25}D$ −11.7° (*c*, 2 in 1 *N* HCl), was obtained.

Anal. Calcd. for $C_4H_{12}N_2O_3Cl_2$: C, 23.2; H, 5.84; N, 13.5; Cl^-, 34.3. Found: C, 23.4; H, 5.71; N, 13.7; Cl^-, 34.3.

β-Aminoxy-D-alanine Monohydrochloride (III). (a) **By Ester Hydrolysis.**—A solution of 24 g. of β-aminoxy-D-alanine methyl ester dihydrochloride in 120 ml. of 6 *N* hydrochloric acid was heated at 85–90° in an oil bath for 4 hr. The solution was then evaporated *in vacuo*. The residue was dissolved in absolute ethanol and evaporated to remove residual hydrochloric acid. This operation was repeated twice. After 4 hr. at *ca.* 0.1 mm., the residue was dissolved in 100 ml. of boiling absolute ethanol, and the solution was cooled to room temperature. Ten milliliters of pyridine was added slowly to the stirred solution. The precipitated product, which weighed 24.3 g., was collected on a filter, washed with absolute ethanol, and dried. Elemental analysis of this material, m.p. 134–136°, R_f^{MPW} 0.74 (N), indicated it to be a mixture of pyridine hydrochloride and the desired aminoxy compound.

A solution of 23.8 g. of this mixture in 50 ml. of water (pH 2) was filtered and then diluted slowly with 125 ml. of absolute ethanol. After 6 hr. at 5°, the solution was filtered giving 12.5 g. (68%) of β-aminoxy-D-alanine monohydrochloride, m.p. 144–145°. The analytical sample, m.p. 146–148°, $[\alpha]^{25}D$ −19.5° (*c*, 2 in 1 *N* HCl), R_f^{MPW} 0.78 (N), was dried at 52° for 5 hr. *in vacuo*.

Anal. Calcd. for $C_3H_9N_2O_3Cl$: C, 23.0; H, 5.79; N, 17.9, Cl, 22.7. Found: C, 23.5; H, 5.35; N, 17.3; Cl, 23.7.

(b) **By Cycloserine Hydrolysis.**—A solution of 20 g. of D-cycloserine in 100 ml. of 6 *N* hydrochloric acid was heated at 60° in an oil bath for 3 hr. The solution was evaporated to dryness *in vacuo*. The residue was twice dissolved in absolute ethanol and evaporated to dryness to remove residual hydrochloric acid. After 3 hr. *in vacuo*, the residue was dissolved in 150 ml. of boiling absolute ethanol, and the solution was allowed to cool to room temperature. Twelve milliliters of pyridine was added dropwise to the stirred solution. The pink precipitate was collected on a filter, washed with ethanol, and dried. The product, which weighed 32.9 g., m.p. 124–128° dec., was apparently the same pyridine hydrochloride–aminoxyalanine complex isolated in part a. This material was dissolved in 60 ml. of water and, after filtration, the solution was slowly (2 hr.) diluted with 150 ml. of absolute ethanol. After standing at 5° overnight, the β-aminoxy-D-alanine monohydrochloride, 12.5 g. (41%), m.p. 143–145°, $[\alpha]^{25}D$ −18.3° (*c*, 2.1 in 1 *N* HCl), was collected on a filter and dried. Its paper chromatographic behavior and infrared spectrum were the same as that of the product obtained in part a.

β-Aminoxy-D-alanine.—A solution of 500 mg. of β-aminoxy-D-alanine monohydrochloride in 3.5 ml. of 3 *N* ammonium hydroxide was cooled in an ice bath and 10 ml. of a 1:1 2-propanol–ethanol mixture was added. The dropwise addition of acetic acid to the solution caused crystallization of the zwitter ion. Another 10 ml. of the 2-propanol–ethanol solution was added and the mixture stood at 5° for 1 hr. The β-aminoxy-D-alanine, m.p. 166–168° dec., $[\alpha]^{25}D$ −24.4° (*c*, 2.13 in 1 *N* HCl), weighed 296 mg. (77%). A sample was dried for analysis at 52° *in vacuo*.

Anal. Calcd. for $C_3H_8N_2O_3$: C, 30.0; H, 6.71; N, 23.3. Found: C, 30.2; H, 6.48; N, 23.2.

Hydrogenation of β-Aminoxy-D-alanine Monohydrochloride.—A solution of 250 mg. of β-aminoxy-D-alanine monohydrochloride in 10 ml. of water was shaken under hydrogen for 16 hr. at 40 p.s.i. and room temperature using 0.25 g. of platinum oxide as catalyst. After removal of the catalyst, the solution was evaporated to a 1-ml. volume and diluted slowly to 5 ml. with absolute ethanol. The crystalline precipitate weighed 145 mg., R_f^{MPW} 0.46 (N); $[\alpha]^{25}D$ −13.2° (*c*, 5 in 1 *N* HCl). L-Serine, R_f^{MPW} 0.46 (N), $[\alpha]^{25}D$ +14.45° (*c*, 8.9 in 1 *N* HCl).

Reaction of Acetone with β-Aminoxy-D-alanine Monohydrochloride.—A solution of 500 mg. of β-aminoxy-D-alanine monohydrochloride in 50 ml. of boiling acetone containing 1 ml. of water was refluxed 4 hr. and evaporated to dryness. The residue was dissolved in a few drops of water and acetone was added to a total volume of 10 ml. After 16 hr. in the refrigerator, the solution yielded 265 mg. of crystalline β-(isopropylideneaminoxy)-D-alanine; m.p. 160–162° dec., R_f^{MPW} 0.69 (N), $[\alpha]^{25}D$ +4.1° (*c*, 2.2 in 1 *N* HCl). A sample was dried for analysis at 52°, *in vacuo* for 2 hr.

Anal. Calcd. for $C_6H_{13}N_2O_3Cl$: C, 36.6; H, 6.66; N, 14.3; Cl^-, 18.0. Found: C, 37.0; H, 6.76; N, 14.1; Cl^-, 18.4.

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N-Carbobenzoxyamino Acyl Derivatives of D-Glucosamine[1,2]

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Prior to our attempt to synthesize "model glycoproteins," we carried out a preliminary study of

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