the toluene used as solvent in the azide decomposition.[4a] The preparation of pure butyl isocyanate by the phosgenation of butylamine has been reported only recently.[5]

### Experimental

**Ethyl Isocyanate.**—In a 500-ml. distilling flask were placed 98 g. of triethyl phosphate and 30 g. of potassium cyanate (along with a few glass beads). Potassium cyanate does not dissolve appreciably in triethyl phosphate, even at higher temperatures. The mixture was strongly heated until distillation occurred. For the next ten minutes heat was applied on and off to maintain distillation at a fairly even rate. The reaction mixture became quite viscous by the end of this time and heating was discontinued when strong white fumes due to the condensation of water vapor started to appear, the water being a result of decomposition within the viscous mass. A parallel experiment had shown that when these fumes were allowed to distil, water droplets appeared in the distillate and caused the evolution of carbon dioxide from the latter (hydrolysis of ethyl isocyanate). The viscous residue in the distilling flask set to a gel on cooling. It consisted chiefly of potassium diethyl phosphate along with some triethyl phosphate.

The distillate, a mixture of ethyl isocyanate and triethyl phosphate, was subjected to distillation, and 16.5 g. of ethyl isocyanate (a 63% yield based on potassium cyanate) was collected at 60–63°. The high-boiling liquid residue from the distillation was triethyl phosphate (16.3 g.).

An experiment employing a smaller relative amount of triethyl phosphate than used above gave a less satisfactory result, since the reaction mixture became viscous and gave rise to water vapor before much product had been collected.

**Butyl Isocyanate.**—The procedure is very similar to that used for ethyl isocyanate. There were employed 73 g. of tributyl phosphate and 20 g. of potassium cyanate. In this case also, when heating of the reaction mixture was continued after it had become viscous, decomposition with formation of water vapor occurred. The reaction was therefore stopped at this stage. The residue in the distilling flask, consisting chiefly of potassium dibutyl phosphate, solidified on cooling. The distillate contained butyl isocyanate and tributyl phosphate, separable by fractional distillation. After three distillations 7 g. of butyl isocyanate (a 29% yield based on potassium cyanate) boiling at 115–117° was obtained; reported b.p. 113–116°.[5]

Reaction of the butyl isocyanate prepared in this manner with *p*-toluidine in benzene solution yielded 1-butyl-3-(*p*-tolyl)-urea in very good yield, m.p. 118° after recrystallization from an alcohol–water mixture; reported m.p. 119°.[4b]

(4) (a) J. W. Boelmer, *Rec. trav. chim.*, **55**, 382 (1936); (b) **55**, 386 (1936).

(5) W. Siefken and A. Doser, U. S. Patent 2,326,501 (1943); R. J. Slocombe, E. E. Hardy, J. H. Saunders and R. L. Jenkins, THIS JOURNAL, **72**, 1890 (1950).

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## Optical Enantiomorphs of Isovaline

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A levorotatory isomer ($[\alpha]^{20}D$ −9.10° in $H_2O$) was isolated by Ehrlich and Wendel[1] from the yeast fermentation of racemic isovaline (I). Fischer and

$(C_2H_5)(CH_3)C(NH_2)COOH$
I

von Grävenitz prepared the dextrorotatory isomer ($[\alpha]^{19}D$ + 11.0° in water) from the formyl compound with brucine.[1a] They did not prepare the enantiomorph. Because of this lack of concordance, there exists some uncertainty in regard to the rotation value of the isolated isomers and to their optical configuration. For these reasons, racemic isovaline was resolved into its optical enantiomorphs by the general enzymatic procedure developed in this Laboratory.[2–7] This procedure essentially depends upon the asymmetric enzymatic hydrolysis of the N-acylated derivative of the racemic amino acid, followed by the separation of the resulting L-amino acid and acyl-D-amino acid in different solvents.

Two special problems arose in the application of this procedure to the resolution of isovaline. The first of these was whether the N-acylated derivative of an amino acid which lacked a hydrogen atom on the α-carbon atom would be susceptible to the action of hog kidney acylase I.[4] N-Chloroacetyl-DL-isovaline was prepared, and found to be asymmetrically hydrolyzed at the L-isomer at a rate which, although considerably lower than that observed with either chloroacetyl-DL-valine or chloroacetyl-DL-norvaline,[4] was sufficient to serve the present purpose. The second problem was to isolate the extremely soluble L-isovaline from the resolution mixture at the end of the enzymatic reaction. This was solved by the employment of a chromatographic procedure involving a cationic exchange resin.

By these means no difficulty was encountered in preparing L-isovaline ($[\alpha]^{25}D$ +11.13°) and D-isovaline ($[\alpha]^{25}D$ −11.28°) in satisfactory yield. It would have been expected that the levorotatory isomer isolated by Ehrlich and Wendel from the fermentation mixture was D-isovaline, and our results are in accord with this assignment of configuration. The fact that our rotation values are higher in magnitude than that reported by Ehrlich and Wendel is not surprising in view of the difficulties which they encountered in their isolation and purification procedures. Our values are in good agreement with that obtained for the dextrorotatory isomer by Fischer and von Grävenitz.[1a]

### Experimental Part

**N-Chloroacetyl-DL-isovaline.**—DL-Isovaline[8] was treated with chloroacetyl chloride and chilled NaOH in the usual manner. On acidification with concd. HCl to *p*H 1.7, N-chloroacetyl-DL-isovaline crystallized in 82% yield. It was recrystallized from water; m.p. 161.5–163.0° (cor.). A m.p. of 162° has been reported for this compound.[9]

*Anal.*[10] Calcd. for $C_7H_{12}O_3NCl$: C, 43.4; H, 6.3; N, 7.2; Cl, 18.3. Found: C, 43.4; H, 6.4; N, 7.2; Cl, 18.2.

**Enzymatic Resolution of Chloroacetyl-DL-isovaline.**—Fifty-three grams of N-chloroacetyl-DL-isovaline was dissolved in 2 liters of water and the solution brought to *p*H 7.5 with 2 *N* LiOH. Three grams of acylase I powder[4] was dissolved in the solution, and water added to bring the concentration of the racemic compound to 0.1 *M*. The enzymatic hydrolysis of the substrate could not be followed by the usual manometric ninhydrin procedure, because the

(1) F. Ehrlich and A. Wendel, *Biochem. Z.*, **8**, 438 (1908).

(1a) E. Fischer and von Grävenitz, *Ann.*, **406**, 5 (1914).

(2) J. P. Greenstein, L. Levintow, C. G. Baker and J. White, *J. Biol. Chem.*, **188**, 647 (1951).

(3) L. Levintow and J. P. Greenstein, *ibid.*, **188**, 643 (1951).

(4) S. M. Birnbaum, L. Levintow, R. B. Kingsley and J. P. Greenstein, *ibid.*, **194**, 455 (1952).

(5) D. Rudman, A. Meister and J. P. Greenstein, THIS JOURNAL, **74**, 551 (1952).

(6) D. Hamer and J. P. Greenstein, *J. Biol. Chem.*, **193**, 81 (1951).

(7) S. M. Birnbaum and J. P. Greenstein, *Archiv. Biochem. Biophys.*, **39**, 108 (1952).

(8) P. A. Levene and R. Steiger, *J. Biol. Chem.*, **76**, 299 (1928).

(9) K. W. Rosenmund, *Ber.*, **42**, 4473 (1909).

(10) Analyses by R. J. Koegel and staff of this Laboratory.

liberated amino acid does not yield quantitative amounts of carbon dioxide, and therefore the nitrous acid method was employed. The rate of hydrolysis of the susceptible L-isomer of N-chloroacetyl-DL-isovaline by acylase I is 38 micromoles per hour per mg. protein N. The digest was treated with a few drops of toluene, and allowed to incubate at 38° for 24 hours. Analyses on an aliquot of the digest revealed that the hydrolysis of the compound had proceeded to 50%. Another gram of the enzyme was added, and the digest allowed to stand for 12 hours longer. Analysis again revealed 50% hydrolysis. Acetic acid was added to *p*H 5, and the protein filtered off with the aid of Norit. The filtrate was evaporated at 40° *in vacuo*, and the small amount of protein which flocculated was again removed by filtration. The filtrate contained L-isovaline, chloroacetic acid and chloroacetyl-D-isovaline. Treatment with excess ethanol in the usual manner[2-7] failed to bring about the separation of the highly soluble L-isovaline. Treatment with concd. HCl to *p*H 1.7 led to the separation in 50% yield of chloroacetyl-D-isovaline. After recrystallization from acetone-ether, the m.p. was 158° (cor.), and $[\alpha]^{25}D$ −9.0° for a 2% solution in absolute ethanol. In 2% aqueous solution, the rotation of the compound was imperceptible.

*Anal.* Calcd. for $C_7H_{12}O_3NCl$: N, 7.2. Found: N, 7.1.

**Chromatographic Separation of the Enzymatic Products.**—A general chromatographic procedure for the separation of the amino acid products obtained by the enzymatic resolution method has been developed in this Laboratory and will be described more fully in a subsequent publication.[11] A brief description of the procedure as it applied to the present problem is as follows. A 100-ml. aliquot of a deproteinized and concentrated isovaline resolution mixture (corresponding to 25.8 g. of chloroacetyl-DL-isovaline in the original digest) was poured onto the top of a column 87 cm. high and 6.5 cm. in diameter composed of 20 to 50 mesh Dowex 50 resin in the acid phase.[12]

Elution with water was carried out at a flow rate of 40 to 60 ml. per hour. Chloroacetyl-D-isovaline appeared in the effluent after approximately 250 ml. of water had passed through the column, as indicated by a fall in *p*H from about 7 to about 3. Aliquots taken from the hour-long fractions were hydrolyzed in 2 *N* HCl for 2 hours and tested for color development with ninhydrin. By this means it was demonstrated that the N-acyl derivative was eluted in approximately 3 liters of effluent. No free isovaline was present in the fractions collected during this interval since ninhydrin tests on unhydrolyzed aliquots were all negative. After further washing of the column with an additional 1.5 liters of water, elution was begun with 2.5 *N* HCl. L-Isovaline began to appear after about 4 liters of solution had passed through the column, as shown by positive ninhydrin tests. The entire L-isovaline was eluted after an additional 3800 ml. of solution had passed through the column.

All the fractions containing chloroacetyl-D-isovaline were combined and evaporated to dryness *in vacuo*, and the residue was taken up in absolute ethanol to remove sodium chloride[13] and any residual protein. The ethanol was evaporated and the residue taken up in acetone and filtered to ensure further the absence of any L-isovaline or sodium chloride. The chloroacetyl-D-isovaline was then isolated by evaporation of the acetone and crystallization from acetone–ether; m.p. 158° (cor.); yield 55% of theory, based on the original amount of chloroacetyl-DL-isovaline; $[\alpha]^{25}D$ −9.0° for a 2% solution in absolute ethanol.

*Anal.* Calcd. for $C_7H_{12}O_3NCl$: N, 7.2. Found: N, 7.2.

Thus the chloroacetyl-D-isovaline isolated from the column was identical in properties with that obtained by acidification of the resolution mixture. Five grams of chloroacetyl-D-isovaline was refluxed for 2 hours with 100 cc. of 2 *N* HCl. The solution was decolorized with Norit, and the filtrate evaporated *in vacuo* to dryness. The residue was dissolved in 100 cc. water and the solution treated with a slight excess of silver carbonate. The silver chloride was filtered off, and the filtrate saturated with hydrogen sulfide gas. The final filtrate was evaporated to dryness *in vacuo* and the residual D-isovaline taken up in a little water, the solution filtered, and acetone added in excess to the clear filtrate. The D-isovaline crystallized as long needles in nearly quantitative yield, $[\alpha]^{25}D$ −11.28° for a 5% solution in water.

*Anal.* Calcd. for $C_5H_{11}O_2N$: C, 51.2; H, 9.4; N, 12.0. Found: C, 51.0; H, 9.5; N, 12.0.

The combined fractions containing the L-isovaline were evaporated to dryness *in vacuo*, and the residue taken up in absolute ethanol and filtered to remove sodium chloride.[13] The ethanol was evaporated, and the residue treated successively with silver carbonate and hydrogen sulfide as described for the D-enantiomorph. The yield after crystallization from water with excess acetone was 77% of the theoretical, based on the original amount of chloroacetyl-DL-isovaline; $[\alpha]^{25}D$ +11.13° for a 5% solution in water.

*Anal.* Calcd. for $C_5H_{11}O_2N$: C, 51.2; H, 9.4; N, 12.0. Found: C, 51.0; H, 9.5; N, 12.2.

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(11) C. G. Baker and H. A. Sober, in preparation.

(12) Cationic exchange resin from the Dow Chemical Company. The resin was regenerated by two cycles of washing with 5 *N* HCl, water, 1 *N* NaOH and water, followed by a final 5 *N* HCl and water wash.

(13) Large volumes of water were used for the final wash of the resin during its regeneration. However, even after the effluent was neutral to phenolphthalein, additional sodium chloride was obtained. The coarse mesh resin employed probably requires a longer equilibration period than does the resin of a finer mesh.

## Some Alkyl Benzenesulfonates[1,2]

BY BERTIN L. EMLING

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Six alkyl benzenesulfonates and their pyridinium salts have been synthesized. Their physical properties and yields are given in Tables I and II. None of these compounds has been reported previously in the chemical literature, although a commercial grade of *n*-butyl benzenesulfonate is produced by the Wyandotte Chemicals Corporation.

Attempts to synthesize *t*-butyl arylsulfonates were unsuccessful. Besides the method used to make the normal alkyl sulfonates, the addition reaction of isobutylene with a sulfonic acid was tried, but the *t*-butyl esters could not be isolated. *t*-Butyl alcohol was produced when water was present, while dimers and trimers of isobutylene were obtained under anhydrous conditions.

### Experimental

*n*-Alkyl Benzenesulfonates.—The sulfonates listed in Table I were prepared from benzenesulfonyl chloride, pyridine and the appropriate alcohol according to the procedure of Sekera and Marvel.[3] The pyridinium salts, listed

TABLE I

*n*-ALKYL BENZENESULFONATES, $C_6H_5SO_3$-R

| R | B.p. °C. | Mm. | Yield, % | $n^{25}D$ | $d^{25}_4$ | S analyses, % Calcd. | Found |
|---|---|---|---|---|---|---|---|
| $C_4H_9$ | 147–149 | 4 | 65 | 1.4997 | 1.148 | 14.97 | 15.15 |
| $C_5H_{11}$ | 136–138 | 1 | 75 | 1.4969 | 1.119 | 14.04 | 13.96 |
| $C_6H_{13}$ | 135–136 | 0.5 | 58 | 1.4952 | 1.099 | 13.23 | 13.36 |
| | M.p. | | | | | | |
| $C_{14}H_{29}$ | 25–25.5 | | 63 | | | 9.04 | 8.82 |
| $C_{18}H_{33}$ | 35–36 | | 79 | | | 8.38 | 8.45 |
| $C_{18}H_{37}$ | 45–46 | | 85 | | | 7.81 | 7.80 |

(1) Based on a paper presented, March 26, 1952, at the 121st Meeting of the American Chemical Society, Buffalo, N. Y.

(2) The author wishes to acknowledge the assistance of John Palkiewicz and Carl Miskowicz of King's College; of Jane Furikawa, Mary Wassel and Joan Boersig of Marian College; of Maragret Jevnik of Caldwell College; and of Paul Mosso and Arthur Marcozzi of St. Vincent College.

(3) V. C. Sekera and C. S. Marvel, THIS JOURNAL, **55**, 346 (1933).