ficulty of obtaining pure samples of the synthetic intermediate N-propargyl-2-pyrrolidone. We now report an improved synthesis of this intermediate which enables oxotremorine to be obtained in an over-all yield of 46% based on the starting material 2-pyrrolidone.

Experimental

N-Propargyl-2-pyrrolidone.—A solution of 28 g. of A.R. KOH in 100 ml. of dry methanol was added to 45 g. of 2-pyrrolidone. The solvent was slowly removed under reduced pressure from the stirred solution at an internal temperature not exceeding 25°. When solid began to separate, 200 ml. of toluene was added and toluene was then continuously added during the course of all subsequent distillations. When 500 ml. of toluene had distilled, the pressure was allowed to rise and distillation was continued at an internal temperature of 90°. Finally 700 ml. of toluene was collected at atmospheric pressure. The suspension was cooled and treated at 40° with 60 g. of propargyl bromide over a period of 1 hr. The reaction mixture was then heated to 67° for 30 min., cooled, and filtered. The toluene was removed under reduced pressure, and the product was distilled. It had b.p. 77° (0.05 mm.), $n^{25}D$ 1.4970, yield 40 g. (61%).

1-(2-Oxopyrrolidino)-4-pyrrolidino-2-butyne, Oxotremorine.—A solution of 30 g. of N-propargyl-2-pyrrolidone, 9.2 g. of paraformaldehyde, and 23 ml. of pyrrolidine in 60 ml. of dioxane was heated at 100° for 12 hr. in an atmosphere of nitrogen. The solvent was removed under reduced pressure, and the residue was dissolved in 60 ml. of 5 *N* HCl. After three extractions with ether, the aqueous solution was made alkaline with 33% aqueous NaOH and extracted with chloroform. Distillation of the chloroform extracts yielded a fraction having b.p. 124° (0.1 mm.), $n^{25}D$ 1.5156, yield 38 g. (76%).

Anal. Calcd. for $C_{12}H_{18}N_2O$: C, 69.87; H, 8.80. Found: C, 69.89; H, 8.59.

Perchloric Acid in the Preparation of 2′,3′-Isopropylidene 6-Thioinosine

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The preparation of the title compound reported in the literature[2-4] can be greatly improved by using perchloric acid[5] as a catalyst for the condensation of 6-thioinosine and acetone. Adequate amounts of this isopropylidene intermediate, required for chemotherapeutic work,[6] can be prepared readily following the procedure described in the Experimental section.

Experimental

The extent of formation of the isopropylidene derivatives was estimated by paper chromatography of an aliquot of the reaction mixture on Schleicher and Schuell No. 589 paper (orange ribbon) by the descending method. The solvent system used throughout consisted of the standard isopropyl alcohol–water–ammonia mixture (70:20:10). For each isolated product, the R_f value was compared with an authentic sample of the same material prepared by previously reported procedures.

2′,3′-O-Isopropylidene 6-Thioinosine. A. Small Scale.—To 100 ml. of acetone (dried with $MgSO_4$), 0.35 ml. of 2,2-dimethoxypropane was added, followed by 0.45 ml. of 70% aqueous perchloric acid. After 5 min. 244 mg. (1 mmole) of 6-thioinosine was added rapidly, and the reaction flask was stoppered and swirled for approximately 5 min. until the solids had dissolved. After 10 min. the solution was neutralized with 0.35 ml. of pyridine and concentrated under diminished pressure to approximately 20 ml. A 10% Na_2CO_3 solution (5 ml.) was added and the remaining acetone was removed. The aqueous solution was extracted twice with 5-ml. portions of dichloromethane and acidified with glacial acetic acid (about 0.7 ml.) to pH 6. After 2 hr. at 4°, the crystalline product was filtered off, washed with water, and dried *in vacuo* at room temperature. The material obtained showed a single spot on paper chromatography (R_f 0.72) and weighed 230 mg. (yield, 80%). Calculated on the basis of the ultraviolet absorption, the aqueous filtrate contained another 30 mg. of product, but no starting material (single spot).

B. "Large" Scale.—To avoid foaming, it was necessary to replace the Na_2CO_3 with an NH_4OH solution. To 2400 ml. of dry acetone was added 65 ml. of 2,2-dimethoxypropane followed by 87.2 ml. of 70% perchloric acid solution. After 5 min., 47.3 g. (0.166 mole) of 6-thioinosine was added quickly with continuous stirring. After 20 min., the clear, yellow reaction mixture was quenched by the addition of 194 ml. of pyridine which had been dried with calcium hydride. A white precipitate formed immediately. Water (920 ml.) was added, followed by 65 ml. of 15 *N* NH_4OH solution, and the suspension was concentrated under reduced pressure until all of the acetone had been removed. Another 65-ml. portion of 15 *N* NH_4OH was added, and the clear solution obtained was extracted with two 325-ml. portions of dichloromethane. The aqueous layer was filtered, chilled, and acidified to pH 6 with glacial acetic acid. After standing for 18 hr. at 4°, the white cyrstalline solid was collected by filtration, washed with water, and dried *in vacuo* at 78°. The product weighed 46.5 g. 86%; m.p. 238–240° dec.; λ_{max} ($\epsilon \times 10^3$): pH 1, 322 (24.0); pH 7, 318 (22.7); pH 13, 310 (23.4). The material showed a single spot on paper chromatography (R_f 0.72).

Synthesis of Some Amino Acid Thiol Esters[1a]

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In connection with a study of certain aspects of the mechanism of action of sulfhydryl enzymes, several amino acid thiol esters have been prepared in this laboratory. These compounds are of particular interest since amino acid thiol esters serve as important model compounds for the study of amino acid–sulfhydryl enzyme interaction. Certain of the N-acyl compounds have been shown to be active substrates for proteolytic enzymes.[2,3]

The syntheses of amino acid thiol esters which have been reported have been for the most part restricted to N-acylamino acid thiol esters. Thiol esters of hippuric acid have been reported by Jeger, *et al.*,[4] by Johnston,[2] and by Schwyzer and Hürlimann.[5] A number of amino acid thiolesters of thiophenol and cysteamine have been prepared by Wieland.[6] Thiol ester

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TABLE I

AMINO ACID THIOLESTER HYDROCHLORIDES, $RCH(NH_2{-}HCl)COSR'$

RCHCO-NH_2	R′	Infrared ν_{CO}, $cm.^{-1}$	Ultraviolet λ_{max}, mμ	Ultraviolet $E \times 10^3$	M.p., °C.	Formula	%C Calcd.	%C Found	%H Calcd.	%H Found	%N Calcd.	%N Found
DL-Alanyl	Et	1684	235	5.0	130	$C_5H_{12}ClNOS$	36.24	36.21	7.30	7.26	8.46	8.25
Glycyl	Et	1698	234	4.3	117–119	$C_4H_{10}ClNOS$	30.08	31.11	6.31	6.34	8.78	8.49
DL-Leucyl	Et	1674	237	5.0	150, 157–159	$C_8H_{18}ClNOS$	45.38	45.54	8.57	8.61	6.62	6.40
DL-Leucyl	Me	1675	236	5.0	168, 175–177.5	$C_7H_{16}ClNOS$	42.53	42.69	8.16	7.97	7.09	7.08
DL-Methionyl	Et	1673	237	4.6	105, 110–111	$C_7H_{16}ClNOS_2$	36.59	36.63	7.03	6.86	6.10	5.96
DL-Methionyl	*i*-Pr	1672	237	4.7	102, 121–123	$C_8H_{18}ClNOS_2$	39.41	39.41	7.44	7.23	5.75	5.96
DL-Methionyl	Me	1691	235	4.7	145, 155–156	$C_6H_{14}ClNOS_2$	33.39	33.18	6.54	6.49	6.49	6.22
DL-Valyl	Et	1688	238	5.0	158–160	$C_7H_{16}ClNOS$	42.52	42.35	8.16	8.13	7.09	7.08

derivatives of glutathione and acetylcysteine have been prepared by Strecker, *et al.*[7]

Experimental

Melting Points.—The melting points were determined in an electrically heated, mechanically stirred oil bath. The temperatures are uncorrected. For the cases where three values are reported the first figure designates where the first shrinkage of the sample occurred. Most of these compounds undergo a gradual shrinkage before actual melting begins. The second and third figures designate the typical melting range. All of these compounds melt with decomposition giving bubbles in the melt but little if any charring occurs.

Spectra.—The infrared spectra were determined as the KBr pellets on a Perkin-Elmer Model 21 recording spectrophotometer. They were calibrated against the 1700 peak of water and corrected as needed. The wave length of the maximum of the carbonyl peak is tabulated in Table I. The ultraviolet spectra were obtained on a Beckman DB spectrophotometer with a Photovolt-Varicord Model 43 automatic recorder. The values of E_{max} were corrected for the absorbtion of water at 236.5 mμ.

Alanine Thiol Ethyl Ester Hydrochloride.—Five grams of DL-alanine was suspended in 150 ml. of acetyl chloride and cooled to −5° in an ice–salt bath. To this suspension was added 12.5 g. of PCl_5 slowly with vigorous shaking over a period of about 30 min. The resulting suspension was transferred to a 200-ml. centrifuge bottle and centrifuged down at 5°. The centrifugate was discarded and the residue was washed once with 100 ml. of acetyl chloride. The residue was then washed five times with 100 ml. of anhydrous petroleum ether.[8] Liquid ethyl mercaptan (25 ml.) was then added directly to the centrifuge bottle which was then fitted with a pressure stopper. After allowing the reaction mixture to stand for 24 hr., the excess mercaptan was evaporated leaving a solid white residue. The crude product was recrystallized by dissolving in a minimum of methanol, adding anhydrous ether to turbidity, and cooling in the deep freeze, m.p. 130, 139–140°, 50% yield.

Acknowledgment.—The infrared spectra were performed by Mrs. M. Wheeler to whom the authors are very thankful. The carbon–hydrogen analyses were performed by A. Bernhardt and associates of Mülheim, West Germany.

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