Keyphrases

3-Quinuclidinol dibasic acid esters—synthesis
Alcohol-anhydride acylation reaction
Anesthetic activity, local—3-quinuclidinol esters
Vasomotor activity—3-quinuclidinol esters
Respiratory activity—3-quinuclidinol esters

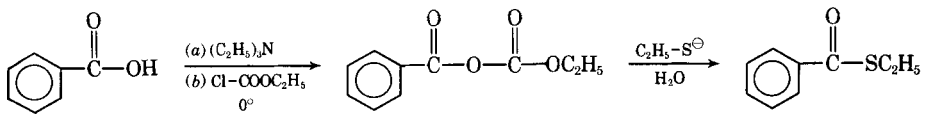
α-Alkyloximino Acid Thiol Esters: New Class of Compounds Showing Antimalarial Activity

By DOMINICK A. COVIELLO and NORMAN L. HINES

New derivatives of α-alkyloximino acids, the thiol esters, were prepared by modification of an application of mixed carboxylic-carbonic anhydrides. In general, the compounds show modest antimalarial activity, though one prolongs the lives of infected mice for more than 3 days longer than controls.

IT IS THE PURPOSE of this communication to report the antimalarial activity of α-alkyloximinocarboxylic acid thiol esters prepared by a modification of a reported method (1). The method utilizes mixed carboxylic-carbonic anhydrides (acyl alkyl carbonates) which are decomposed by a mercaptan in the presence of triethylamine. The authors have found that aqueous solutions of mercaptides are equally efficient in the anhydride decomposition. The modification was inspired by a report that carboxamides are produced by reacting the acyl alkyl carbonates with aqueous ammonia (2) indicating that anhydrous conditions are not essential for success in the anhydride reaction.

In a model reaction, ethyl thiolbenzoate was prepared as follows:



It was found that a better elemental analysis for the product could be obtained in the case of the title compounds through the modified procedure.

The compounds prepared are shown in Table I.

Biological Testing—Groups of five mice were infected with *Plasmodium berghii*. Three days after challenge test compounds were administered subcutaneously The data are reported in Table II.

EXPERIMENTAL

α-Ethyloximinopropionic Acid—This was obtained in 50% yield according to the procedure described by Ferris (3). It melted at 68–69°. Reported: 68–70°.

α-Benzyloximinopropionic Acid—The product was obtained by hypochlorite oxidation of the previously reported α-benzyloximinobutanone (3). The product melting at 84–85° was obtained in 52% yield. Reported melting point 84–85° (3).

α-Methyloximinobutyric Acid—Oxidation of α-methyloximino-2-pentanone in the manner described above gave a 13% yield of acid melting at 75–77°. Its confirmation was by elemental analysis of the thiol ester derivative (see Table I).

α-Ethyloximinobutyric Acid—The acid was prepared according to the procedure of Waters and Hartung (4). The yield was 48% and the melting point 58–59° was in agreement with the reported value.

α-Benzyloximinobutyric Acid—The product was obtained in 55% yield and had a melting point of 89–94° which was in agreement with the reported value (5).

α-Ethyloximinocaproic Acid—This was prepared in 82% yield according to the method of Waters and Hartung (5) and its boiling point of 76–80°/0.05

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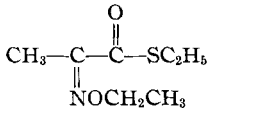
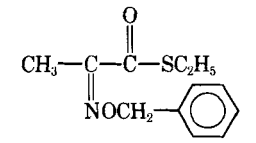
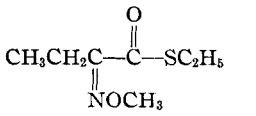
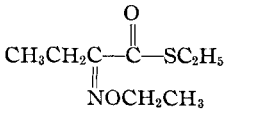
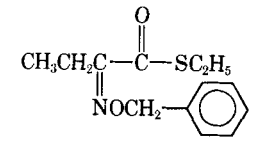
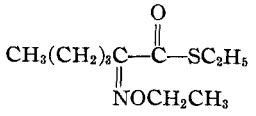
Antimalarial testing and test data have been made available through the courtesy of Dr. David Jacobus, Walter Reed Army Institute of Research, Washington, D. C.

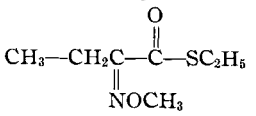
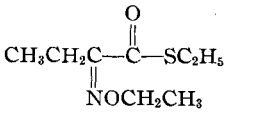
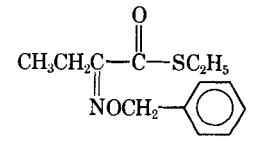
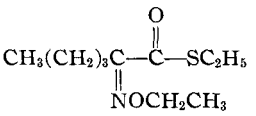
The technical assistance of J. C. Ziebell, W. L. Jacobson, and L. Sternson is also acknowledged.

TABLE I—α-ALKYLOXIMINO ACID THIOL ESTERS

Compd.	B.p., °C.	Yield, %	$\eta_{D/t}$	Anal.—Calcd.	Anal.—Found
	49–50/0.03 mm.	67	1.4864/24°	C, 48.00 H, 7.43 N, 8.00 S, 18.30	47.85[a] 7.61 8.09 18.21
	110–112/0.01 mm.	47	1.4861/28°	C, 60.72 H, 6.39 N, 5.91 S, 13.49	60.95[a] 6.29 5.95 13.38
	48–49/0.01 mm.	66	1.4851/30°	C, 48.00 H, 7.43 N, 8.00 S, 18.30	48.24[b] 7.60 8.00 18.41
	50–52/0.01 mm.	63	1.462/25°	C, 50.79 H, 7.94 N, 7.41 S, 16.93	51.23[b] 8.14 7.56 16.79
	114–16/0.07 mm.	71	1.541/21°	C, 62.15 H, 6.77 N, 5.58 S, 12.75	62.18[b] 6.86 5.51 12.73
	67–68/0.02 mm.	37	1.4756/25°	C, 55.23 H, 8.84 N, 6.45 S, 14.76	55.35[a] 8.87 6.38 14.71

[a] Analyses performed by Kurt Eder, Geneva, Switzerland. [b] Analyses performed by Micro-Tech Laboratories, Skokie, Ill.

TABLE II—EFFECT OF α-ALKYLOXIMINO THIOL ESTERS ON MALARIA-INFECTED MICE

Compd.	Dose, mg./Kg.	Mean Survival Time, Days		Change in Survival Time, Days	Toxic Deaths
	40	7.2	6.4	+0.8	
	160	7.4	6.4	+1.0	
	640	7.8	6.4	+1.4	
	40	6.8	6.4	+0.4	
	160	6.8	6.4	+0.4	
	640	6.8	6.4	+0.4	
	40	7.8	6.7	+1.1	
	160	10.0	6.7	+3.3	
	640				5[a]
	40	7.0	6.4	+0.6	
	160	7.0	6.4	+0.6	
	640	8.2	6.4	+1.8	

[a] Deaths occurring on days 2, 3, 4, and 5, after infection, are attributed to drug action and counted as toxic deaths. Control animals do not die before day 6.

mm. was in agreement with the reported value.

Ethyl Thiolbenzoate—In a 1-L., round-bottom flask was placed 200 ml. of anhydrous ether and 12.2 Gm. (0.1 mole) of benzoic acid. The solution was then cooled to 0° with an ice bath and 10.12 Gm. (0.1 mole) of triethylamine was added followed by 10.85 Gm. (0.1 mole) of ethyl chlorocarbonate, at such a rate as to maintain the temperature at 0–5°. The solution was then stirred for 30 min. in the ice bath. The triethylamine hydrochloride was

filtered off and 50 ml. of a 4 *M* aqueous solution of sodium ethylmercaptide was added. The solution was allowed to stir for 1 hr. at room temperature. The ether layer was separated and the aqueous layer was extracted with two 50-ml. portions of ether. The ethereal layers were combined and dried overnight with anhydrous magnesium sulfate. The magnesium sulfate was removed by filtration and the ether removed by distillation. The residual oil was then distilled under reduced pressure and the fraction distilling between 59–60°/.075 mm. was collected. The product weighed 11.95 Gm. (71.98%) and its refractive index was 1.5690 at 20.5°. The reported boiling point is 83–84° at 2.5 mm. and refractive index is 1.5678 at 25° (5).

α-Alkyloximinocarboxylic Acid Thiol Esters—The title compounds were prepared in the manner described for the preparation of ethyl thiolbenzoate. Pertinent data on the compounds are summarized in Table I.

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(2) Woolley, D. W., Hershey, J. W. B., and Jodlowski, H. A., *J. Org. Chem.*, **28**, 2012(1963).

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(4) Waters, K. C., and Hartung, W. H., *ibid.*, **12**, 469 (1947).

(5) Meyer, E., and Zublin, J., *Ber.*, **11**, 320(1878).

(6) "Handbook of Chemistry and Physics," Chemical Rubber Publishing Co., Cleveland, Ohio, 1959.

Keyphrases

α-Alkyloximino acid thiol esters—synthesis
Mercaptan decomposition of acyl alkyl carbonates
Refractive index
Antimalarial activity

Molecular Association of the Antibiotic Prasinomycins

By JOEL KIRSCHBAUM

The molecular weight of the antibiotic prasinomycins in ethanol is 1600–2300. In aqueous solvents the prasinomycins associate, forming two aggregates with molecular weights of approximately 31,000 and 61,000. The aggregates may be disrupted by organic solvents, dilution below 0.012 percent, or the addition of salts.

THE prasinomycins are a family of closely related antibiotics isolated from *Streptomyces prasinus* (1). Although the molecular weight of one major component of the mixture, as calculated from phosphate group analysis (1), was 1550 ± 10%, the inability to dialyze these antibiotics through a cellulose membrane indicated the possibility of molecular association. The molecular weight in ethanol, determined by means of an analytical ultracentrifuge,[1] ranged between 1600 and 2300. In aqueous solvents the apparent molecular weights were 31,000 and 61,000. These solvent-induced differences in molecular weight indicate association or aggregation. Some of the properties of these aggregates will be described in this paper.

EXPERIMENTAL

Sedimentation coefficients of prasinomycin solutions of concentrations from 10 mg./ml. to 1.25 mg./ml. were measured in 4°, 12-mm. cells centrifuged in an An-D rotor at 42,040 r.p.m. and 20.7°. The phaseplate angle of the schlieren optical system was 75°. For solutions with concentrations of 1.25 mg./ml. to 0.25 mg./ml., the sedimentation coefficients (s) were determined in a 12-mm. synthetic boundary centerpiece in an interference cell.

Molecular weights were determined using equilibrium and Archibald approach-to-sedimentation equilibrium methods.

For the Archibald (2) method, 5–10 mg./ml. solutions in 12-mm. interference cells were centrifuged in an An-D rotor at 20.7°. Concentration gradients were formed at speeds of 6995, 8766, 12,590, 20,410, 29,500, and 42,040 r.p.m. to test for any dependence of molecular weight on speed, which, in turn, affects concentration of the sedimenting material. The schlieren optical system was used with a phaseplate angle of either 75° or 80°. The concentration gradients at the meniscus and cell bottom were measured from data recorded on Kodak metallographic plates magnified 10-fold on a Nikon magnifier. Engelberg's technique (3) for evaluating the concentration gradient integral was used. Total concentrations were determined in separate experiments using a synthetic boundary centerpiece. An IBM 1620 computer was used for some of the repetitive calculations.

Equilibrium centrifugation (4) aided in obtaining the molecular weights of prasinomycin solutions of 0.15 mg./ml. and lower in concentration. Using a calibrated syringe, 0.03 ml. of prasinomycin solution was injected into one compartment of an interference cell fitted with sapphire windows and layered with fluorochemical FC 43 (Beckman Instruments, Inc.). Solvent was injected into the reference compartment. The final percentage

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[1] Model E Analytical Ultracentrifuge, Spinco Division, Beckman Instruments, Inc.