those prepared by the procedure recommended above, the sample is subjected to an additional reprecipitation from 6 *M* potassium hydroxide solution. In this Laboratory, such additional purifications of samples obtained by the recommended procedure gave products ranging in purity from 98.5 to 99% potassium ferrate, as analyzed by the chromite method,[11] although a great reduction in the quantity of potassium ferrate resulted.

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Phosphorus Oxychloride in the Preparation of Ketimines

BY ARTHUR W. WESTON AND R. J. MICHAELS, JR.

It has been found that phosphorus oxychloride is an excellent agent for promoting the formation of anils derived from 2-thienyl aryl and 2-thienyl alkyl ketones. Recently Hartough[1] reported that propyl 2-thienyl ketone yielded 28% of the Schiff base when condensed with aniline in the presence of iodine. However, by adding a small amount of phosphorus oxychloride to a refluxing solution of the reactants we were able to isolate 88% of the desired condensation product. Similarly, an improved yield of N-phenyl methyl 2-thienyl ketimine[1] resulted when phosphorus oxychloride was used. High yields were obtained with the other thienyl ketones employed.

The use of phosphorus oxychloride is particularly indicated for the 2-acylthiophenes which form insoluble complexes[1] with zinc chloride, a commonly employed condensing agent, and for other compounds that are sensitive to zinc chloride.

In addition, it was observed that benzophenone, which does not condense directly with aniline, readily produced the anil when these reactants were treated with phosphorus oxychloride. On the other hand, the condensation of acetophenone with aniline was complicated by the formation of both acetophenone anil and dypnone anil. The latter product is to be expected in view of the tendency of acetophenone to undergo molecular condensation in the presence of acid. It is interesting to note, however, that the isostere, methyl 2-thienyl ketone, yielded little or no self condensation product under the same reaction conditions.

The addition of 0.5 cc.[2] of phosphorus oxychloride to a mixture of 0.25 mole of ketone and 0.3 mole of aniline was sufficient to ensure complete condensation, except in the case of phenyl 2-thienyl ketone. In this instance it was necessary to use twice the quantity of catalyst in order to obtain a good yield of N-phenyl phenyl 2-thienyl ketimine. In all condensations a slight excess of aniline was employed to compensate for the material removed by salt formation.

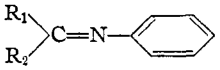
If the phosphorus oxychloride added at the start of the reaction proved to be inadequate, more could be added later without impairing the yield, as illustrated in the experimental section. An amount of concentrated hydrochloric acid, sirupy phosphoric acid, or a mixture of the two equivalent to the quantity of phosphorus oxychloride employed gave inferior results in the preparation of N-phenyl methyl 2-thienyl ketimine. With benzophenone and aniline, comparable results were realized when the phosphorus oxychloride was replaced by phosphorus oxybromide, phosphorus trichloride or phosphorus pentachloride. Phosphorus pentoxide was not effective, indicating that a halogen-containing catalyst is probably required.

Experimental

N-Phenyl Ethyl 2-Thienyl Ketimine.—A mixture of 27.9 g. (0.30 mole) of aniline, 35 g. (0.25 mole) of ethyl 2-thienyl ketone[3] and 0.5 cc. (0.0055 mole) of phosphorus oxychloride in 150 cc. of toluene was refluxed overnight in a flask equipped with a reflux condenser and a conventional water separator. At the end of this time, the theoretical amount (4.5 cc.) of water had been collected. After the reaction mixture was filtered to remove the aniline salt which had separated, the toluene was removed under reduced pressure and the residue distilled. There was obtained 10.2 g. of forerun followed by 40.4 g. (75%) of product, b.p. 133° at 1 mm., n^{24}D 1.6347.

TABLE I

N-PHENYL KETIMINES



R₁	R₂	B.p. °C.	Mm.	M.p., °C.	Yield,[g] %	Formula	Nitrogen, %[h] Calcd.	Nitrogen, %[h] Found
2-Thienyl	Methyl	137–138	1.5	69–70[a,b]	68	$C_{12}H_{11}NS$	6.96	6.87
2-Thienyl	Ethyl	130–133	1.0	[c]	75	$C_{13}H_{13}NS$	6.51	6.42
2-Thienyl	*n*-Propyl	130–132	0.8	[d]	88	$C_{14}H_{15}NS$	6.11	6.04
2-Thienyl	Phenyl	158–162	1.7	123–124[b]	80	$C_{17}H_{13}NS$	5.32	5.35
Phenyl	Phenyl	160–163	0.8	113–114[e,f]	86	$C_{19}H_{15}N$	..	..

[a] Reported (ref. 1) m.p. 69–70°. [b] Crystallized from absolute alcohol. [c] n^{24}D 1.6347. [d] n^{25}D 1.6199; reported (ref. 1) b.p. 128–130 at 1 mm. [e] C. M. Rosser and J. J. Ritter, THIS JOURNAL, 59, 2179 (1937), report m.p. 113–114°. [f] Crystallized from acetone. [g] In most instances, some of the original ketone was recovered so that the yields are actually higher than reported. [h] We are indebted to Mr. E. F. Shelberg and members of the Microanalytical Department for these microanalyses.

N-Phenyl Phenyl 2-Thienyl Ketimine.—By refluxing a mixture of 47 g. (0.25 mole) of phenyl 2-thienyl ketone,[4] 27.9 g. (0.30 mole) of aniline and 0.5 cc. (0.0055 mole) of phosphorus oxychloride for 17 hours, 2 cc. of water was liberated. After the addition of another 0.5 cc. (0.0055 mole) of phosphorus oxychloride along with 4.7 g. (0.05 mole) of aniline to combine with the acid subsequently formed, the mixture was again refluxed overnight. The theoretical amount of water (4.5 cc.) had then been liber-

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(1) H. D. Hartough, THIS JOURNAL, 70, 1282 (1948).

(2) The importance of this quantity was not determined but it was noted that the use of appreciably smaller amounts of the agent lowered the yield considerably.

(3) H. D. Hartough and A. I. Kosak, THIS JOURNAL, 69, 3099 (1947).

(4) The phenyl 2-thienyl ketone and methyl 2-thienyl ketone were furnished through the courtesy of Dr. G. A. Harrington of the Socony-Vacuum Oil Company.

ated. The crude product was isolated as described above and then distilled. A forerun of 6.6 g. was followed by the main fraction b.p. 158–162° at 1.7 mm. The 52.5 g. (80%) of ketimine thus obtained melted at 121–123°. Crystallization from absolute alcohol gave pure material m.p. 123–124°.

Data pertaining to the anils prepared in this study are recorded in Table I.

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Trypsin Hydrolysis of Lysine Ethyl Ester

BY HAROLD WERBIN AND ANN PALM

In a recent review Neurath and Schwert[1] have discussed the specificity requirements of synthetic substrates for trypsin. L-Lysine ethyl ester (LyEE) which possesses the necessary configuration, (1) a lysyl side chain, (2) a susceptible ester linkage and (3) a polar group (NH_2) alpha to the carbonyl of the susceptible bond, has been found to be hydrolyzed by trypsin. The rate of hydrolysis was measured by using the Hestrin technique[2] for the quantitative determination of esters. Figure 1 shows that the hydrolysis follows zero order kinetics at *p*H 7.88 and 25.0°. The calculated rate constant was found to be a linear function of the enzyme concentration as illustrated in Fig. 2.

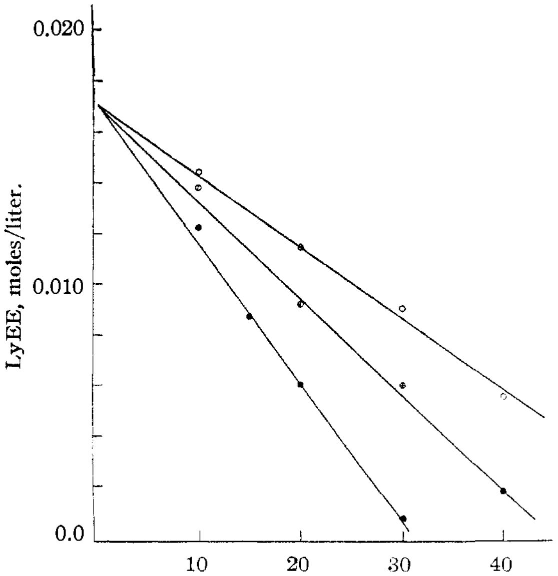

Fig. 1.—Zero order hydrolysis of 0.017 *M* lysine ethyl ester by trypsin at 25° and *p*H 7.88: ○, 2.73 × 10^{-3} mg. TPN/ml.; ◐, 3.66 × 10^{-3} mg. TPN/ml.; ●, 5.46 × 10^{-3} mg. TPN/ml.

Recently Iselin, *et al.*,[3] have employed a colorimetric procedure similar to the one described below to measure the hydrolysis of hydroxamides by chymotrypsin, while the use of the Hestrin method to measure the extent of hydrolysis of 1-arginine methyl ester[4] by trypsin and of 1-tyrosine ethyl ester[4] by chymotrypsin has been described by Werbin.[5] Financial aid from the Dazian Foundation for Medical Research is acknowledged.

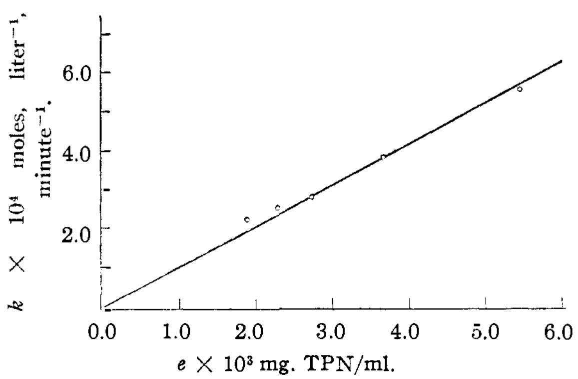

Fig. 2.—Specific rate constant for the hydrolysis of lysine ethyl ester by trypsin.

Experimental

Substrate.—LyEE was prepared by passing dry hydrogen chloride gas through a mixture of 6.0 g. of lysine monohydrochloride and 210 ml. of absolute ethanol until all the lysine had dissolved. The addition of hydrogen chloride was stopped, the solution was refluxed for 2 hours, and placed in the refrigerator overnight. On the following day it was distilled under reduced pressure and the residual oil was reësterified. After removal of the ethanol, crystals appeared and recrystallization from 25 ml. of absolute ethanol yielded 5.0 g. of white crystals, decomposing at 144°. Akabori and Kaneko[6] report 143.5–144.5°.

Kinetic Study.—To 2.0 ml. of 0.06 *M* LyEE and 2.0 ml. of 0.3 *M* phosphate buffer incubated for 10 minutes at 25° ± 0.02 was added 2.0 ml. of crystalline trypsin dissolved in hydrochloric acid solution of *p*H 3.0. The stopwatch was started at the time of half-addition of the trypsin. At the desired time 1.0 ml. of reaction mixture was added to 2.0 ml. of alkaline hydroxylamine solution following the procedure employed by Hestrin.[2] The ferric chloride solution used to develop the color was 0.4 *M*. The transmission of the solution at 520 mμ was read on a brociner-mass photoelectric colorimeter 5 minutes after the addition of the ferric chloride. A blank run demonstrated that there was no spontaneous hydrolysis of the ester.

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[1] H. Neurath and G. W. Schwert, *Chem. Revs.*, **46**, 69 (1950).

[2] S. Hestrin, *J. Biol. Chem.*, **180**, 249 (1949).

[3] B. M. Iselin, H. T. Huang and C. Niemann, *ibid.*, **183**, 403 (1950).

[4] These substrates were suggested by Dr. Harry Goldenberg.

[5] H. Werbin, Ph.D. Thesis, Polytechnic Institute of Brooklyn, June, 1950.

[6] S. Akabori and T. Kaneko, *Bull. Chem. Soc. Japan*, **11**, 208 (1936).

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Preparation of 1,1,3,3-Tetrachloropropane

BY A. M. WHALEY[1] AND H. W. DAVIS

There are twenty-nine possible chloropropanes, two of which do not have physical properties listed in the literature. Both of these compounds, namely, 1,1,3,3-tetrachloropropane and 1,1,1,3,3,3-hexachloropropane, have all the halogen atoms located on the end carbon atoms, so that it is not easy, if at all possible, to make these chlorides by processes involving addition of chlorine to a double bond; of interest, however, is the fact that the 1,1,1,3,3-pentachloropropane has been pro-

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[1] Halogen Chemicals Inc., 616 King St., Columbia, S. C.