The tissue and subcellular distribution of mammalian aldehyde oxidizing enzyme has recently been studied by Deitrich (11). Activity was found in liver, adrenal, intestine, kidney, ovary, testis, adipose tissue, uterus, heart, lung, brain, spleen, skeletal muscle, seminal vesicles, and bladder. Liver exhibited the greatest degree of activity. Thus, the placenta appears to be unique in not having the capacity to oxidize aldehydes.

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Identification of Low Molecular Weight Aliphatic Esters from Rates of Alkaline Hydrolysis

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Abstract □ The second-order rate constants for the alkaline hydrolysis of 27 low molecular weight aliphatic esters have been determined at 25° in 37.27% acetone–water solution. These rate constants are characteristic of the entire ester molecule, and therefore provide more information for identification purposes than do other chemical methods for the characterization of esters.

Keyphrases □ Aliphatic esters, low molecular weight—identification □ Alkaline hydrolysis rates—aliphatic ester identification □ Rate constants, second-order—ester hydrolysis

Kinetic measurements provide a potentially powerful approach to the characterization of organic compounds because rate constants can be very sensitive to minor structural alterations. Moreover, the technique is simple, inexpensive, and it seldom requires large samples. Earlier papers in this series have described the identification of alcohols from rates of alkaline hydrolysis of their 3,5-dinitrobenzoate esters (1), of sugars from their rates of oxime formation (2), and of aliphatic amines from rates of acylation by cinnamic anhydride (3).[1] For this method to be practicable, first-order or pseudo-first-order kinetics must be observed, reaction conditions and the method of analysis must be common to all members of a class of compounds, and a large number of rate constants must be determined under these common conditions. This note reports rate constants for the alkaline hydrolysis of 27 aliphatic carboxylic acid esters of low molecular weight.

[1] This paper is part IV in the series "Precise Kinetic Measurements Applied to the Identification of Organic Compounds." For Part III, see *Reference 3*.

The reactions were followed by the pH-stat method, using a pH meter as a manually operated pH-stat (4). The solvent was an acetone–water mixture (see *Experimental*), and the esters studied include all of those that are soluble to the required extent in this solvent. First-order rate constants were obtained from Guggenheim plots (5), and were converted to second-order constants by dividing by the essentially constant concentration of hydroxide ion; the pH meter was used merely as an indicator to keep the pH constant, rather than as a source of information about hydroxide-ion activity (which would be questionable in this solvent). It is extremely important to bear in mind the difference between second-order rate constants for ester hydrolysis as calculated on the concentration and activity bases (6).

Table I gives the esters, their observed atmospheric boiling points, the mean second-order rate constants, and the standard deviations (of a single observation). The means are based on three to five determinations. It is seen that the reproducibility is about 1–2% for most esters.

These 27 esters represent only seven different values of saponification equivalent, and many pairs of them (especially isomers) have similar boiling points. But despite these similarities, most of the esters can be differentiated by a combination of boiling point and rate constant. Of course, one should submit authentic samples to measurement to check these values under one's own conditions, especially when a comparison of very similar constants is to be made. The method described here is unusual in that it is probably only the second general chemical method for characterizing

Table I—Second-Order Rate Constants for the Alkaline Hydrolysis of Aliphatic Esters[a,b]

Ester	B.p., °C.	$-10^2k_{OH}(M^{-1}\ sec.^{-1})-$ Mean	*SD*
Methyl formate	32	1690	32
Ethyl formate	53	976	18
Methyl acetate	56.5	15.1	0.3
Isopropyl formate	66.5	352	8
Ethyl acetate	77	6.61	0.13
Methyl propionate	79.5	10.82	0.08
n-Propyl formate	80	770	13
Isopropyl acetate	88	1.41	0.02
Methyl isobutyrate	91	3.88	0.05
sec-Butyl formate	94	202	8
Isobutyl formate	97	646	9
Ethyl propionate	97	4.65	0.04
n-Propyl acetate	100.5	4.69	0.04
Methyl butyrate	101	5.09	0.08
n-Butyl formate	105.5	697	10
Isopropyl propionate	108	0.785	0.010
sec-Butyl acetate	110	0.640	0.004
Isobutyl acetate	116	3.37	0.05
Ethyl butyrate	120	2.02	0.04
n-Propyl propionate	121	2.96	0.07
Isoamyl formate	128	528	12
n-Butyl acetate	126	4.26	0.09
n-Pentyl formate	127	556	2
Isoamyl acetate	139	3.34	0.07
n-Butyl propionate	142	2.37	0.04
n-Amyl acetate	147	3.56	0.05
n-Hexyl acetate	169	3.15	0.06

[a] At 25.0° in 37.27% v/v acetone–water; initial ionic strength 0.32–0.37 *M*. [b] Calculated on basis of hydroxide-ion concentration.

esters that gives a result dependent upon the entire ester molecule (the saponification equivalent is the other).

The authors believe that Table I gives the most complete set presently available of rate constants for the alkaline hydrolysis of aliphatic esters under common conditions. As such, it should be useful in generating substituent parameters for polar and steric effects in aliphatic systems, particularly since variations have been made in both the acyl and alkyl portions of the molecule.

EXPERIMENTAL

Materials—Esters were from commercial sources and were redistilled before use; boiling points are reported in Table I. Water was redistilled from alkaline permanganate through an all-glass apparatus. All other chemicals were reagent grade.

Apparatus (4)—The reaction vessel was a glass-jacketed beaker of about 90-ml. capacity. Water thermostatted at 25.0 ± 0.05° was pumped through the jacket. The vessel was supported over a magnetic stirrer. The mouth of the vessel was closed with a rubber stopper containing four holes; through one hole passed a 2-ml. micrometer buret (Roger Gilmont Instruments), the tip of which extended into the reaction solution. Another hole accommodated a high-alkaline range combination glass-calomel pH electrode (Sargent). The third hole held a thermometer. The fourth hole was used for flushing the vessel with nitrogen gas before reaction and for introducing the ester sample; this hole was stoppered during reactions. A pH meter was used (Sargent model DR).

Procedure—A typical experiment was conducted as follows: 3.0 ml. of standard 1 *N* aqueous potassium hydroxide, 50.0 ml. of 0.50 *N* aqueous potassium chloride, and 30.0 ml. of reagent grade acetone were delivered into the reaction vessel, which was flushed with nitrogen gas for about 30 sec. The system was allowed to reach temperature equilibrium. The reaction was initiated by adding the neat ester with a syringe. Timing began when the measured pH dropped 0.01 unit below the initial value. When the pH had dropped 0.01 unit lower, sufficient 1.0 *N* KOH was added from the buret to return the pH to its initial value. The time and volume were recorded. This process was continued, the pH being held at ±0.01 unit.

The initial composition of the reaction solution depended upon the reactivity of the ester; with all acetates, propionates, and butyrates, 1 to 6 (x) ml. of 1 *N* KOH, (53 − x) ml. of 0.5 *N* KCl, and 30 ml. of acetone were taken. For formates, which are faster reacting, 2 ml. of 0.01 *N* KOH, 51 ml. of 0.5 *N* KCl, and 30 ml. of acetone were used. The sample size was about 0.002 ml. of pure ester for formates and about 0.2 ml. of ester for all other esters. The titrant was 0.01 *N* KOH for formates and 1 *N* KOH for the others. The initial ionic strength was 0.32 *M* for the formate studies, and it was 0.34–0.37 *M* for the other esters. The final ionic strength was 0.32–0.39 *M*.

The volume of a solution prepared by mixing 53.0 ml. of 0.50 *N* KCl and 30.0 ml. of acetone, at 25°, was determined to be 80.50 ml. This value was used in calculating the hydroxide-ion concentration in the reaction mixture.

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