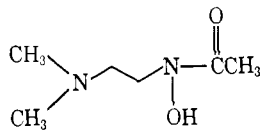

V

$= 2.01 \times 10^{-3}$ *M*, pH 8.26) 25-fold over-all in a pseudo-first-order process. A detailed kinetic investigation will be presented elsewhere.

The data presented here establish that N-alkylhydroxamate ions are true catalysts of the hydrolysis of labile esters. In addition, they function effectively at very low concentration and in the neutral pH range. The catalytic rates that we observed are at least as large as those observed in PNPA hydrolysis catalyzed by imidazole,[17] the only other previously known catalyst of this kind. However, in their ease of synthesis, N-alkylhydroxamate ions hold an important advantage over imidazole. Most carboxylic acid esters may be converted to N-alkylhydroxamic acids under very mild conditions. This affords a convenient means of introducing these functionalities into higher molecular weight systems, such as the cycloamyloses,[18] that exhibit substrate binding. By utilizing the proximity and orientation effects afforded by these systems, we expect to realize the full catalytic potential of the N-alkylhydroxamates. Furthermore, since ester derivatives of the carboxyl groups at the active sites of both lysozyme[19] and pepsin[20] have been prepared, there is the attractive possibility that the active sites of these enzymes may be altered to contain an N-alkylhydroxamic acid, an alteration which should lead to enzymes of new functional group specificity.

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Enhanced Rates due to Apolar Interactions between Polymer and Substrate

Sir:

In previous publications[1-3] it has been shown that polyethylenimine (PEI) and some of its acyl derivatives possess extraordinarily high binding affinities for small organic molecules. This polymer is a highly branched, relatively compact, water-soluble macromolecule.[4] Its aliphatic acyl derivatives provide apolar binding sites in proximity to amine residues of the polymer. One might expect, therefore, to find progressively enhanced rates of aminolysis of substrates with increasingly large apolar substituents. Quantitative measurements of rates of aminolysis indeed reveal enhancements of several orders of magnitude.

Rates of cleavage of three acyl nitrophenyl esters were followed by the appearance of *p*-nitrophenolate ion as reflected by increased absorbances at 400 nm, measured with a Cary Model 14 spectrophotometer. The reaction was followed at pH 9.0, in 0.02 *M* tris(hydroxymethyl)aminomethane buffer, at 25°. Rate constants were determined from measurements under pseudo-first-order conditions, with the residue molarity of primary amine present in approximately tenfold excess. First-order rate graphs were linear for at least 80% of the reaction. With nitrophenyl acetate and nitrophenyl caproate, the initial ester concentration was 6.66×10^{-5} *M*. With nitrophenyl laurate at this concentration, aminolysis by polymer was too fast to follow and, therefore, both substrate and amine were diluted tenfold for rate measurements.

Polyethylenimines of different molecular weight ranges (PEI-6, PEI-18, PEI-600)[4] were obtained from Dow Chemical Co. The lauroyl derivative of PEI-6, containing 10% of the nitrogen residues conjugated to this acyl group, was prepared as described elsewhere.[3]

Table I lists first-order rate constants, corrected for hydrolysis of ester in buffer alone. Propylamine served as a reference amine; in its presence k (in min^{-1}) for aminolysis decreased progressively from 0.98×10^{-2} to 0.51×10^{-2} to 0.05×10^{-2} as the length of the acyl group increased from 2 to 12 carbons (see Table I). The sharp drop for nitrophenyl laurate may be the result of micelle formation[5] even at concentrations of 6×10^{-6} *M*.

Table I. First-Order Rate Constants for Amine Acylation by *p*-Nitrophenyl Esters[a]

Amine	$k \times 10^2$ min[b] *p*-Nitrophenyl acetate	*p*-Nitrophenyl caproate	*p*-Nitrophenyl laurate
Propyl	0.98	0.51	0.053
PEI-6[c]	3.60	1.47	0.11
PEI-18[c]	4.38	1.57	0.11
PEI-600[c]	4.60	1.80	0.17
L(10%)-PEI-6[d]	15.2	68.1	698

[a] Measurements made at pH 9.0 in 0.02 *M* tris(hydroxymethyl)-aminomethane buffer, 25°. Stock solutions of substrate were made in acetonitrile; hence the final buffer also contained 6.7% acetonitrile. [b] $k = k_a - k_0$, where k_a is the measured rate constant in the presence of amine and k_0 is that for the hydrolysis in Tris buffer alone; k_0 is 0.94×10^{-2} min^{-1} for the acetyl ester, 0.61×10^{-2} min^{-1} for the caproyl ester, and 0.023×10^{-2} min^{-1} for the lauroyl ester. [c] The numeral following "PEI" multiplied by 100 is the molecular weight of the polymer sample. [d] This sample of PEI-6 has 10% of its nitrogens acylated with lauroyl groups.

With nonacylated polyethylenimines (Table I) the rate constant is increased by a factor of about 4 over that of propylamine. This small enhancement may be due merely to the fact that a greater fraction of primary amine groups in the polymer are in the basic, NH_2 state. With these polyethylenimines, as with propylamine, k drops with increasing length of the hydrocarbon chain of the acyl nitrophenyl ester.

Markedly different trends are seen in the rate constants for aminolysis by lauroylpolyethylenimine (con-

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taining 10 residue per cent lauroyl groups). For each nitrophenyl ester the rate is substantially greater with lauroylpolyethylenimine than with polymer containing no acyl group. Furthermore the trend in k is now markedly *upward* as the acyl group is increased from 2 to 12 carbons (see Table I). Compared to k for propylamine with nitrophenyl laurate, the corresponding k for lauroylpolyethylenimine is 10^4 times greater. Such a comparison may not be fully appropriate if the low rate with reference amine is due primarily to the micellar state of the lauroyl nitrophenyl ester. If one assumes that in the absence of micelle formation the long-chain ester would show a rate comparable to that of acetyl nitrophenyl ester, then the enhancement factor in the presence of lauroylpolyethylenimine still is of the order of 10^3. In any event it is clear that the introduction of strong binding sites on the polymer leads to marked rate enhancements. This polymer with binding sites thus should provide a suitable framework for the further introduction of catalytic functional groups to produce a macromolecule with capacity for enhanced rates combined with true turnover of substrate.

(6) Postdoctoral Fellow, National Institute of General Medical Sciences, U. S. Public Health Service, 1968–1970.
(7) This investigation was supported in part by a grant from the National Science Foundation.

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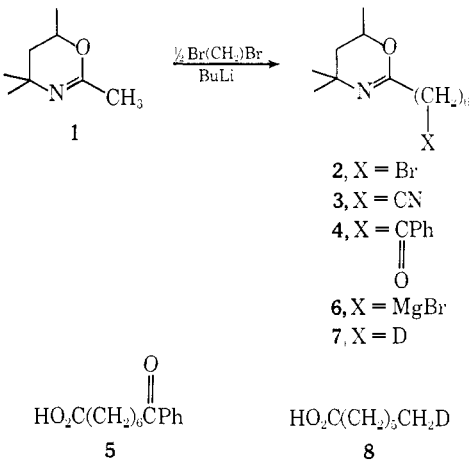
Syntheses *via* Dihydro-1,3-oxazines. VI. A Carboxyl Protecting Group Stable to the Grignard Reagent. A New Synthesis of Carboxylic Acids

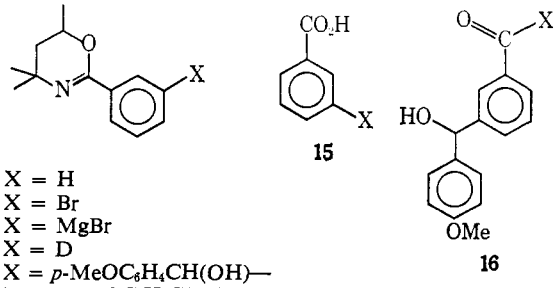
Sir:

It was found early in our studies that the 2-methyl-5,6-dihydro-1,3-oxazine (**1**),[1] though converted to its anion by alkyllithium reagents, is totally inert to various Grignard reagents. We now report that this behavior has indeed proved useful in obtaining alkyl and aryl carboxylic acids,[2] from appropriately substituted dihydro-1,3-oxazines by use of usual Grignard techniques. The value of this approach is obvious in light of the rarity of carboxyl protecting groups toward RMgX.[3] Thus, the methods now known[1] to alkylate **1** coupled with the facile regeneration of carboxylic acids from dihydro-1,-3-oxazines[4] provides substituted acids hitherto obtained only with difficulty.

The readily available[5] 2-methyl-5,6-dihydro-1,3-oxazine (**1**) was alkylated with 1,5-dibromopentane producing the 6-bromohexyl derivative **2** in 90% yield (oil; 1658 cm^{-1}; τ 6.68, t, 2, CH_2Br) which was transformed into the corresponding nitrile **3** in 95% yield with sodium cyanide in DMSO[6] (bp 108° (0.25 mm); 2240, 1658 cm^{-1}). The reaction of **3** with phenylmagnesium bromide in ether followed by hydrolysis gave the keto oxazine derivative **4** (90%; oil, 1675 and 1658 cm^{-1}) which was hydrolyzed with aqueous hydrobromic acid saturated with sodium bromide (reflux, 12 hr) to 7-benzoyl heptanoic acid **5** (90% mp 85°).[7] The 6-bromohexyl oxazine was also converted to the corresponding Grignard reagent **6** in THF (reflux, 3 hr) and hydrolyzed in deuterium oxide to the 6-deuteriohexyl derivative **7** in 80% yield (bp 64° (1.2 mm); 1659 cm^{-1}; *m/e* calcd 212, found 212). Acidic cleavage of **7** in aqueous hydrobromic acid resulted in the formation of 7-deuterioheptanoic acid **8**.



Aromatic acids **15** were likewise formed by this sequence, employing as starting material the 2-phenyldihydro-1,3-oxazine **9**.[8] Bromination[9] of the latter gave, in 95% yield, exclusively *m*-bromo isomer, **10** (bp 120° (0.075 mm), 1640 cm^{-1}) which was transformed into the Grignard reagent **11** in THF (reflux, 5 hr). Both the bromophenyl derivative **10** and the Grignard reagent **11**, after hydrolysis in deuterium oxide, were cleaved to the *meta*-substituted benzoic acids **15** (X = Br) and **15** (X = D) in 99 and 98% yields, respectively. The Grignard reagent **11** was also found to react normally with *p*-anisaldehyde affording **13** in quantitative yield (glass, purified *via* tlc; 1640 and 3400 cm^{-1}). Attempts to hydrolyze **13** to the acid **16** (X = OH) gave tarry products due to polymerization of the benzhydryl cation. However, borohydride reduction of **13** and



9, X = H
10, X = Br
11, X = MgBr
12, X = D
13, X = *p*-$MeOC_6H_4CH(OH)$—
14, X = *p*-$MeOC_6H_4C(=O)$—

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