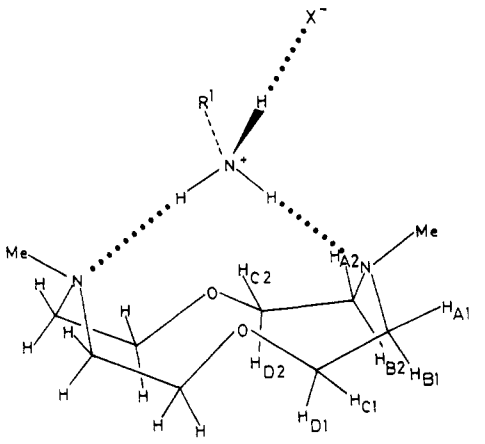

R^1, Achiral — C_s symmetry
R^1, Chiral — C_1 symmetry

16

likely to occur when the anion is SCN^- and the "available" hydrogen on the positively charged nitrogen is sterically accessible and relatively more acidic (i.e., when R^1 is Me, CH_2Me, and CH_2Ph rather than when R^1 is $CHMe_2$, CMe_3, and CHMePh).

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(8) Compound **3** was obtained pure as an oil after vacuum distillation (bp 79 °C at 0.02 mmHg). This compound and all of the salts derived from the secondary dialkylamines **5–8** and the primary alkylamines **9**–(*S*)-**14** gave satisfactory results for their elemental analyses.

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(10) The Eschweiler Clarke methylation procedure has been used successfully in related work; cf. E. Graf and J.-M. Lehn, *J. Am. Chem. Soc.*, **98**, 6403 (1976).

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(12) Complex formation (1:1) with the perchlorate salts at 20 °C resulted in the following shifts (parts per million) of the signals for the NMe, NCH_2, and OCH_2 protons, respectively, in **3**: **5**·$HClO_4$, 0.15, 0.16, 0.08; **6**·$HClO_4$, 0.38, 0.42, 0.24; **7**·$HClO_4$, 0.42, 0.47, 0.28; **8**·$HClO_4$, 0.13, 0.12, 0.07; **9**·$HClO_4$, 0.10, 0.07, 0.03; **10**·$HClO_4$, 0.11, 0.19, 0.05; **11**·$HClO_4$, 0.14, 0.14, 0.08; **12**·$HClO_4$, 0.20, 0.21, 0.12; **13**·$HClO_4$, −0.19, 0.06, 0.06; (*S*)-**14**·$HClO_4$, −0.17, 0.10, 0.09. Similar results were obtained for the thiocyanate salts. It will be noted that all of the signals are shifted downfield except for the upfield shifts experienced by the NMe protons when **3** is complexed with either $PhCH_2NH_3^+ClO_4^-$ or (*S*)-$PhCHMeNH_3^+ClO_4^-$. At first glance there appears to be a major discrepancy between the influence of these salts on the chemical shift of the NMe protons in **3** and the significant downfield shift witnessed by the same protons in **3** when it complexes with $(PhCH_2)_2NH_2^+ClO_4^-$. However, the chemical shifts are markedly dependent on temperature (cf. ref 5c). For example, at −90 °C, the presence of 1 molar equiv of **7**·$HClO_4$ causes an upfield shift of 0.30 ppm in the signal for the NMe protons of **3**.

(13) Variable temperature ^1H NMR spectroscopy of solutions containing equimolar amounts of crown and 1:1 complex (i.e., a molar ratio of crown to salt of 2:1) has proved to be an extremely valuable technique in the investigation of the kinetics of complexation–decomplexation of ligands with metal cations (J.-M. Lehn, J.-P. Sauvage, and B. Dietrich, *J. Am. Chem. Soc.*, **92**, 2916 (1970); J.-M. Lehn, *Struct. Bond.*, **16**, 1 (1973); A. C. Coxon and J. F. Stoddart, *Carbohydr. Res.*, **44**, C1 (1975); *J. Chem. Soc., Perkin Trans. 1*, 767, (1977)). In this approach, signals for complexed and uncomplexed crown become evident in the spectra under conditions of slow exchange. In principle, the kinetics of complexation–decomplexation of crowns with cations can also be investigated by dynamic ^1H NMR spectroscopy for a molar ratio of crown-to-salt of 1:2. In this approach, which is dependent upon the solubility of the salt in organic solvents, signals for complexed and uncomplexed salt become evident in the spectra under conditions of slow exchange (cf. ref 5b). Attempts to employ this approach in the present investigation resulted in the protonation of the crown by the excess of salt with consequent release of free amine into solution. This observation is being studied in more detail at the moment.

(14) Below −100 °C, the signal for the NCH_2 protons of **3** in CD_2Cl_2 separated into two signals with $\Delta\nu = 76$ Hz. A value of 8.4 kcal/mol for $\Delta G^{\ddagger}_{rni}$ was deduced from a calculation of k_c (170 s^{-1}), and hence $\Delta G^{\ddagger}_c$, at T_c (−96 °C).

(15) In principle, a total fo four ABCD systems could be observed for the NCH_2CH_2O protons in the asymmetric complexes at low temperatures. In practice, only two ABCD systems were identified in the low temperature spectra.

(16) In rigid 18-crown-6 systems which cannot undergo ring inversion there is some evidence (D. A. Laidler and J. F. Stoddart, *J. Chem. Soc., Chem. Commun.*, 481 (1977)) that this assumption does not hold when the crown is highly substituted and contains a secondary binding site for the cation.

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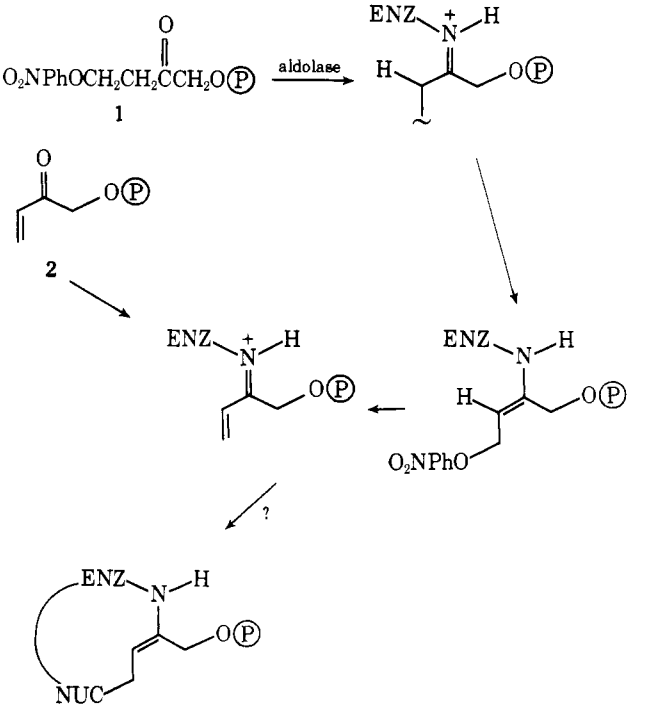
An Inhibitor for Aldolase

Sir:

Compound **1** was synthesized in order to determine if rabbit muscle aldolase would catalyze the elimination reaction shown in Scheme I in a manner similar to that found for primary amines in aqueous solution with a α-acetoxy or β-hydroxy

Scheme I



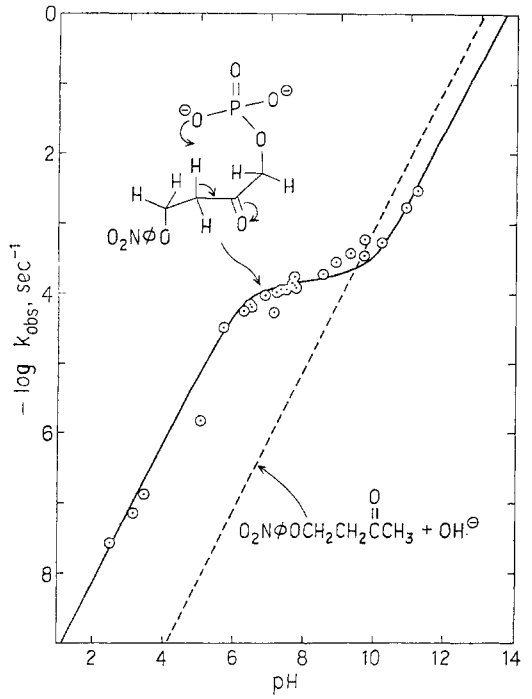
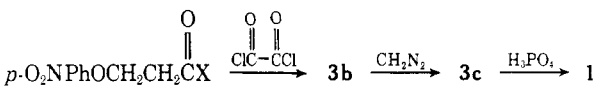


Figure 1. A pH rate profile for the production of O_2NPhOH from **1**, which is fit by a line computed from $v = k_1[1^{2-}] + k_2[OH^-][1^{2-}]$ where $k_1 = 1.30 \times 10^{-4}$ s^{-1} and $k_2 = 2.13$ M^{-1} s^{-1}. A value of pK_a ($ROPO_3H^-$) = 6.80 was used for the computation. The dashed line represents the corresponding pH rate profile for 4-*p*-nitrophenoxy-2-butanone.[7] The term proportional to the dianion of **1** is ascribed to an intramolecular proton abstraction as shown.

Scheme II



3a, X = OH
b, X = Cl
c, X = CH═N═N

ketone.[1,2] A reaction of this type would provide an interesting mechanistic probe of the active site and could also be a simple assay for this enzyme because of the generation of chromophoric *p*-nitrophenoxide. Although no aldolase dependent production of *p*-nitrophenoxide has been observed with **1**, it has been found that its elimination product **2** is a potent inhibitor for this enzyme.

The synthesis of compound **1** involved the conversion of **3a**, prepared from *p*-nitrophenol and β-propiolactone,[3] to the corresponding acid chloride **3b**, using oxalyl chloride in benzene (Scheme II).[4] After removal of solvent, this material was added, without purification, to a 4 M excess of distilled diazomethane solution in diethyl ether and stirred for 10 min before removal of solvent to yield stable, crystalline **3c**.[5] This material was quantitatively converted to **1** by stirring a 4×10^{-3} M solution of **3c** in 1.5×10^{-2} M H_3PO_4 in benzene for 48 h, and then extracting this solution with 30 mL of water to give an aqueous solution of **1**. The addition of sodium hydroxide to this solution generated *p*-nitrophenoxide which allowed the concentration to be measured spectrophotometrically. Removal of phosphate from **1** was accomplished by passing 30 mL of a 4×10^{-3} M aqueous solution of **1** through a 35 × 1.5 cm column of Sephadex G-15 and collecting the early fractions. Freezing and lyophilizing this solution yielded stable, crystalline **1** as the acid.[6]

As shown in Figure 1, the pH-rate profile for the rate of formation of *p*-nitrophenol from **1** differs substantially from 4-*p*-nitrophenoxy-2-butanone, **5**, which has been shown to

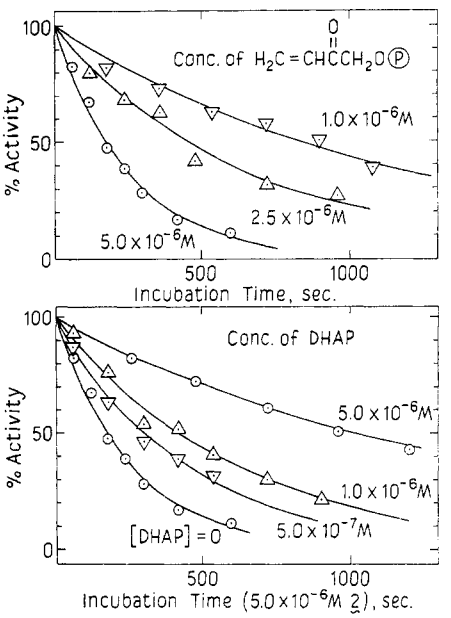
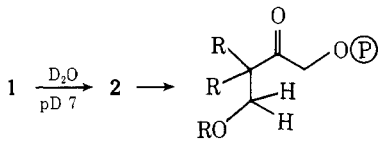


Figure 2. The upper plot shows the dependence of percent inactivation of aldolase on the concentration of **2** and the length of the incubation time with **2**, consistent with a second-order rate constant for inactivation of 7.6 × 10^{-2} M^{-1} s^{-1}. The lower plot demonstrates the protection afforded aldolase by various concentrations of DHAP, consistent with a K_m of 1.4 × 10^{-6} M for this natural substrate. The percent activity measurements were performed on aliquots of incubation mixture at 37 °C.

Scheme III



4a, R = D
b, R = H

undergo rate-determining α-proton abstraction by general bases.[7] In addition to hydroxide ion catalysis of enolization, a term proportional to the concentration of the dianion of **1** is apparent which does not appear in the pH-rate profile of **5**. This term may most reasonably be ascribed to an intramolecular proton abstraction by the phosphate dianion as pictured in Figure 1. By dividing the first-order rate constant for this process (1.30×10^{-4} s^{-1}) by the second-order rate constant for the reaction of HPO_4^{-2} with **5** (3.5×10^{-4} M^{-1} s^{-1}), which was measured under the same conditions, an effective molarity[8] of approximately 0.4 M may be computed.

The elimination reaction of a 0.02 M solution of **1** in D_2O adjusted to pD 7.0 was followed by ^1H NMR and the disappearance of **1** over the course of 24 h was coupled with the appearance of enone **2** (Scheme III).[9] This material hydrated to the β-hydroxy ketone **4a**[10] over a period of 2 weeks and did not decompose via retro-aldol reaction under these conditions even after 1 month, as shown by the lack of appearance of formaldehyde hydrate. The nondeuterated compound **4b** was prepared in H_2O and lyophilized and the spectrum taken in D_2O showing the expected pair of triplets.[11] The UV spectrum of **2** (λ_{max} 215 nm, ϵ 6.0×10^3) was obtained by the extraction of a 10-mL, 10^{-3} M aqueous solution of **2** adjusted to pH 5.8, with 3 × 10 mL of Et_2O, followed by bubbling nitrogen through the solution to remove the remaining Et_2O. The peak due to the enone disappears if the solution is made basic, as is the case for methyl vinyl ketone and other enones.[12,13]

When **1** is allowed to react with increasing concentrations of aldolase, no increase in the rate of production of *p*-nitrophenol is observed over the background rate. This may mean either that **1** does not undergo enzyme-catalyzed elimination

or that the elimination is followed by inactivation in the first few turnovers. The concentration of enzyme was not great enough to allow an initial burst to be seen.

Compound **2** behaves as though it were an irreversible inhibitor since it shows time-dependent inactivation[14] with a rate constant of 7.6×10^2 M^{-1} s^{-1}, as shown in Figure 2. The percent inhibition of aldolase reactivity, measured by standard assay techniques,[15] depends on both the concentration of **2** and the length of time with which it is incubated with enzyme. The reaction may in fact be reversible under appropriate experimental conditions even though the equilibrium constant for formation of inhibited enzyme is quite large.[14] As is also shown in Figure 2, the enzyme is protected from inhibition by the natural substrate, dihydroxyacetone phosphate, DHAP. The K_m for DHAP computed from these data, 1.5×10^{-6} M, is smaller than the K_m measured by fluorescence,[16] 4.5×10^{-6} M. Since the literature value was determined in the presence of Cl^-, a known reversible inhibitor of aldolase, these values are not inconsistent. The disappearance of inhibitory ability in solutions of **2** kept at 25 °C and pH 7.0 parallels the slow conversion of **2** to **4b** as followed spectrophotometrically. In contrast to **2**, methyl vinyl ketone exhibits a much slower inactivation of aldolase and that inactivation is not completely prevented by high concentrations of DHAP. A possible mechanism for the inactivation of aldolase by **2** is that shown in Scheme I.

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(4) To a solution of 5.2 g (2.46×10^{-2} mol) 3a in 300 mL of benzene at 60 °C was added 4.2 mL of oxalyl chloride (4.92×10^{-2} mol) and the mixture was refluxed for 90 min to yield a solution of **3b:** ^1H NMR ($CDCl_3$) δ 3.40 (t, 2 H), 4.33 (t, 2 H), 6.88 (d, 2 H), 8.05 (d, 2 H).
(5) The overall yield from **3a** to **3c** is 75%. Compound **3c:** mp 94–95 °C; ^1H NMR ($CDCl_3$) δ 2.83 (t, 2 H), 4.40 (t, 2 H), 5.40 (s, 1 H), 6.95 (d, 2 H), 7.84 (d, 2 H); IR 1740, 2130 cm^{-1}; analysis satisfactory.
(6) Compound **1:** mp 84–86 °C; ^1H NMR (D_2O, pD 1.6) δ 3.11 (t, 2 H), 4.44 (t, 2 H), 4.65 (d, J = 7.93 Hz, 2 H, CH_2OP), 7.09 (d, 2 H), 8.23 (d, 2 H); IR (KBr) 1730 cm^{-1}.
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(9) Compound **2:** ^1H NMR (D_2O, pD 7.1) δ 4.49 (d, 2 H, J = 6.35 Hz, CH_2OP), 5.94–6.68 (m, 3 H).
(10) Compound **4a:** ^1H NMR (D_2O, pD 7.1) δ 3.86 (s, 2 H, CH_2OD), 4.49 (d, 2 H, J = 6.35 Hz, CH_2OP).
(11) Compound **4b:** ^1H NMR (D_2O, pD 7) δ 2.78 (t, 2 H). 3.85 (t, 2 H), 4.48 (d, 2 H, J = 6.25 Hz, CH_2OP).
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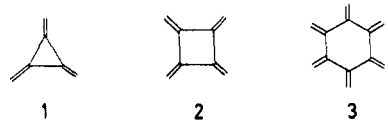
Hexaradialene

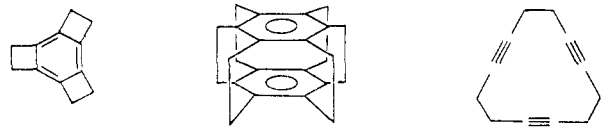
Sir:

Of the parent radialenes[1] only [3]-[2] and [4]radialene[3] (**1** and **2**) have been isolated as air-sensitive, reactive hydrocarbons. Several stable, substituted radialenes are known[4] including hexaalkylated hexaradialenes.[1,5] The latter appear to minimize nonbonded interactions by adopting a nonplanar cyclohexane chair-type conformation[6] devoid of 6π-electronic delocalization. The parent hexaradialene (hexamethylenecyclohexane)



1 2 3

(**3**) is of considerable theoretical[7] and synthetic interest because of its potentially stabilizing benzenoid topology, its potential as a synthon for the construction of polycyclic nuclei, and as a possible precursor to the strained tricyclobutabenzene **4**[8] and the elusive cyclophane **5,** respectively.



4 5 6

Recently we reported that 1,5,9-cyclododecatriyne (**6**)[9] reacted with dimethyl maleate at 230 °C to give a triadduct formally derived from **4** via sequential four-ring opening or, alternatively, from **3** via threefold cycloaddition.[10] Since in the absence of trapping agent extensive polymerization occurred we decided to investigate the gas phase thermochemistry of **6.**

Sublimation of **6** through a hot quartz tube (650 °C (10^{-3} Torr)) and collection of the pyrolysate at −196 °C gave a colorless brittle solid which turned dark on warming. Vacuum transfer of solvent (toluene-d_8) onto the pyrolysis product or, alternatively, collection of the pyrolysate on a frozen solvent covered glass surface allowed on warming the rapid extraction of nonpolymerized materials and transfer into an NMR tube which was vacuum sealed. In addition to starting material (τ 7.62, toluene-d_8) a new sharp singlet was observed in the *olefinic* region (τ 4.69). This signal disappeared after several hours at room temperature or instantly on exposure of the sample to air with concomitant deposition of a brown flocculent precipitate. No new absorptions appeared in the NMR spectrum. Gas phase pyrolysis of **6** at 850 °C led to complete disappearance of starting material (NMR). A mass spectrum of this product gave a parent ion at *m/e* 156.

Hydrogenation of the pyrolysate prepared at 650 °C (Pd/C, ether, −65 °C to room temperature) gave[11] nearly equal amounts of naphthalene,[12] hexamethylbenzene,[12] and cyclododecane,[12] in addition to several unidentified products. Analysis[11] of the air-oxidized crude pyrolysate of **6** (generated at 650 °C) indicated a multitude of products including starting material and naphthalene, but no hexamethylbenzene. Deuteration of the pyrolysis product gave hexakis(deuteriomethyl)benzene.[11]

Gas phase pyrolysis of **6** at temperatures above 850 °C gave rise to increasing amounts of volatile but stable butatriene, identified by its singlet NMR singlet at τ 4.65 and by comparison with authentic material.[13,14] Pyrolysis of butatriene[3] under the above conditions gave recovered starting material.

The observed experimental data are most easily accommodated by the assumption that cyclododecatriyne **6** undergoes pyrolytic conversion to hexaradialene (**3**), a highly reactive compound seemingly devoid of any stabilizing "aromatic" features and comparable in its properties with the other known radialenes **1** and **2**.[15] If one invokes **4** as a likely intermediate in this transformation, then the reported data lead to the somewhat surprising conclusion that radialene **3** enjoys higher thermal stability than benzene **4.** Thermochemical calculations[16,17] (Figure 1) seem to support this view. Thus, even a hypothetical, fully "aromatic" **4** ($\Delta H_f°$ = 93 kcal/mol) is estimated to be at least 11 kcal/mol higher in energy than (nonplanar) **3**.[18]

An alternative approach to rationalize the observed data