New Photoactivated Protecting Groups. 6. *p*-Hydroxyphenacyl: A Phototrigger for Chemical and Biochemical Probes[1,2]

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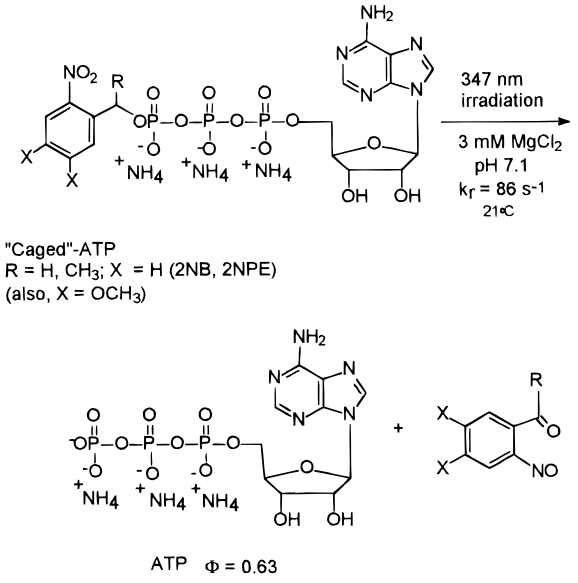
Abstract: *p*-Hydroxyphenacyl, a new photoactive, aqueous soluble protecting group is proposed as a second generation α-*keto "cage" reagent*, a phototrigger for the efficient, rapid release of bioactive phosphates, *e.g.*, inorganic phosphate (P_i) and ATP (Givens, R. S.; Park, C.-H. *Tetrahedron Lett.* **1996**, *37*, 6259−6262). *p*-Hydroxyphenacyl esters **6c** and **7** trigger the release of P_i and ATP when irradiated at wavelengths between 300−350 nm also yielding *p*-hydroxyphenylacetic acid (**8**) from the rearrangement of the intermediate α-keto carbocation or its equivalent. In contrast, unsubstituted and *m*-substituted phenacyl esters yield only photoreduction and radical coupling products and none of the rearrangement product. Quantum efficiencies of 0.38 ± 0.04 were measured for the disappearance of the *p*-hydroxyphenacyl phosphate esters **6c** and **7**; the appearance efficiencies for **8** and ATP were 0.30 ± 0.03. Rates of release of $\sim 10^7$ s^{-1} or better are observed for these esters with only minor variations in efficiencies and rate constants between these two examples of the *p*-hydroxyphenacyl phototrigger. Just as was found for the desyl "cage" series reported earlier (Givens, R. S.; Athey, P. S.; Kueper, L. W., III; Matuszewski, B.; Xue, J.-y.; Fister, T. J. Am. Chem. Soc. **1993**, *115*, 6001−6010), the *p*-hydroxyphenacyl derivatives react via their triplet states. Amino substituents, *i.e.*, *p*-amino-, *p*-acetamido-, and *p*-(carbomethoxyamino)phenacyl phosphates **6f**−**h**, were also investigated, but these analogues proved to be inferior as photoreleasers when compared with *p*-hydroxyphenacyl.

Photoactivated release of biochemical substrates has received considerable attention since Engels and Schlaeger's[3] first reported that the *o*-nitrobenzyl (2-nitrobenzyl or 2NB) ester of cAMP released free cAMP upon photolysis, thus providing the potential for controlling both the temporal and spatial release of the nucleotide in viscous or structured environments. Subsequently, Kaplan, Forbush, and Hoffman[4] reported the efficient photorelease of ATP and inorganic phosphate (P_i) from the corresponding 2NB and the *o*-nitrophenethyl (1-(2-nitrophenyl)ethyl or 2NPE) esters (eq 1). These authors assigned the general term "cage" to the 2-nitrobenzyl group to emphasize its application as a photoactivated protecting group. Previous studies[5] had clearly demonstrated that the 2-nitrobenzyl group could serve the function of a photoprotecting group in synthesis of phosphates, amines, alcohols, carbamates, and carboxylic acids. However, the reports of Engels and Schlaeger[3] and Kaplan *et al.*[4] were among the first to demonstrate the photorelease of a nucleotide from a "caged" substrate which then could elicit a biochemical response, thus opening the way for future exploitation of photoprotecting groups for biological investigations. The release of biochemical substrates from 2-nitrobenzyl esters of nucleotides, oligopeptides, proteins, and



(1)

amino acids has since rapidly gained wide use among biochemists and physiologists.[6−14]

[⊗] Abstract published in *Advance ACS Abstracts*, February 15, 1997.

(1) For a recent study on α-keto "caged" amino acids, see: Gee, K. R.; Kueper, L. W., III; Barnes, J.; Dudley, G.; Givens, R. S. *J. Org. Chem.* **1996**, *61*, 1228−1233.

(2) For previous studies on α-keto phosphates, see: (a) Givens, R. S., Kueper, L. W., III *Chem. Rev.* **1993**, *93*, 55−66. (b) Givens, R. S.; Athey, P. S.; Kueper, L. W., III; Matuszewski, B.; Xue, J.-y. *J. Am. Chem. Soc.* **1992**, *114*, 8708−8709. (c) Givens, R. S.; Athey, P. S.; Kueper, L. W., III; Matuszewski, B.; Xue, J.-y.; Fister, T. *J. Am. Chem. Soc.* **1993**, *115*, 6001−6010. (d) Givens, R. S.; Matuszewski, B. *J. Am. Chem. Soc.* **1984**, *106*, 6860−6861.

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The major advantages of a photoactivated process for the release of nucleotides, as summarized in many reviews,[6−8] are control of the spatial and temporal distribution of a substrate and the ability to effect significant increases in the concentration of a released substrate. An additional factor of significance is the convenience of a "noninvasive" photochemical activation process.

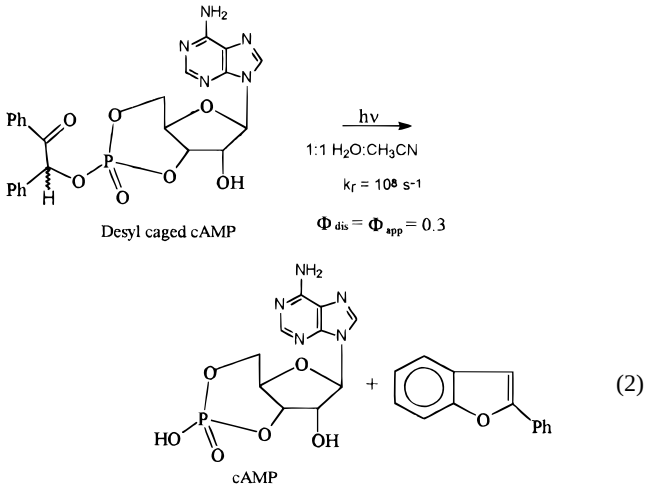
The substantial number of studies that apply some variation of the 2-nitrobenzyl strategy[6−8] for the investigation of biochemical mechanisms have established the 2NB cage as the archetypal chromophore. The reactions are generally unaffected by modest changes in the temperature and media, occur with high quantum efficiencies (∼0.1−0.6), and are activated by UV-vis wavelengths greater than 300 nm. While 2NB and its derivatives would appear to be ideal cages, the photoactivated release of a substrate from 2-nitrobenzyl cages typically is limited to the millisecond to second time regime ($k_r = 1-10^3$ s^{-1}; see eq 1).

Nevertheless, the effectiveness of the 2NB strategy to probe biochemical mechanisms has been abundantly demonstrated[3,4,6−13] over the past decade, *e.g.*, the mechanism of release of inorganic phosphate in skeletal muscle,[9] the action of actomyosin in muscle contraction induced by ATP,[10] the role of cAMP in the relaxation of distal muscle,[11] and the action of Ca−ATPase in the sarcoplasmic reticulum in active calcium transport during ATP hydrolysis.[12] In a recent example, Corrie and Trentham *et al.*[13] reported the use of rapid scan time-resolved Fourier transform infrared (TR-FTIR) spectroscopy to measure the rate constant of ATP release at 1251 cm^{-1} and the appearance of the free phosphate group of ATP at 1119 cm^{-1}. The decay of the signal for the *aci*-nitro anion intermediate and for the formation of ATP were fitted to a single exponential to give a mean rate constant of 218 ± 33 s^{-1} at pH 7.0 and 22 °C. Among the more recent applications of 2-nitrobenzyl caged nucleotides have been the time-resolved X-ray crystallographic investigations of a substrate residing in the active sites of an enzyme.[14]

There have been other investigations which have focused from modifying 2-nitrobenzyl cages to optimizing the conditions for photorelease of substrate.[15] Substituted 2-nitrobenzyl cages have been designed to improve the photochemical efficiencies and to increase the rates of release of several caged neurotransmitters.[16] However, few reports have appeared that describe the discovery, development, and application of entirely new photoactive cages. This is surprising, since there are several disadvantages which limit the application of the nitrobenzyl chromophore. An especially significant shortcoming is the relatively slow, rate-limiting hydrolysis of the *aci*-nitro intermediate formed by the initial rapid hydrogen atom abstraction by the excited 2-nitrobenzyl moiety (eq 1).

The relatively modest microscopic rate constant ($k \approx 1-10^3$ s^{-1}) limits its application to the study of rapid biochemical events and, for example, in at least one case has prevented a complete kinetic analysis of an enzymatic hydrolysis reaction. Niggli and Lederer[17] found that enzymatic-catalyzed formation of ADP from ATP *was more rapid* than the rate of photoinitiated release of ATP from its caged precursor in a study of myocardial muscle contraction. Additional disadvantages encountered with the 2NB and 2NPE cage release strategy for nucleotides have included: (1) the high UV absorptivity of the *aci*-nitro and arylnitroso intermediates, (2) the reactivity of the nitroso functional group with primary amines and other nucleophiles, (3) the instability of the caged derivative which occasionally gives rise to premature hydrolysis and biochemical reaction prior to photorelease, and (4) the competing pathways for energy migration and electron transfer prior to C−O bond lysis.

Our initial studies on phosphate photochemistry[2d] provided the first example of a new α-keto phototrigger as an alternative to 2-nitrobenzyl. Desyl phosphates[2] (eq 2) and other α-keto phosphates derivatives[18] have subsequently been shown to release the phosphate ligand efficiently with rate constants of 10^5 s^{-1} or higher as a consequence of the direct cleavage of the covalent bond to the caged substrate from the excited state of the phototrigger.[c,18,19] Furthermore, the byproducts of the α-keto cage ligands appeared to be biochemically benign.



(2)

Others, particularly the group of Corrie and Trentham,[18] have also reported the development of caged phosphates, *e.g.*, 3′,5′-dimethoxydesyl ATP **12**, as alternatives to the 2NP and 2NPE. Their work parallels our own investigations of desyl phosphate and desyl cAMP, further substantiating the potential of desyl derivatives. Subsequent studies by Baldwin,[19] Pirrung,[20] and Iwamura[21] have also tested the efficacy of α-keto phosphates as phototriggers. These studies demonstrated, however, that desyl derivatives are limited by low solubility in aqueous buffer and by hydrolytic instability. These limitations have prompted us to further explore other candidates for phototriggers, and

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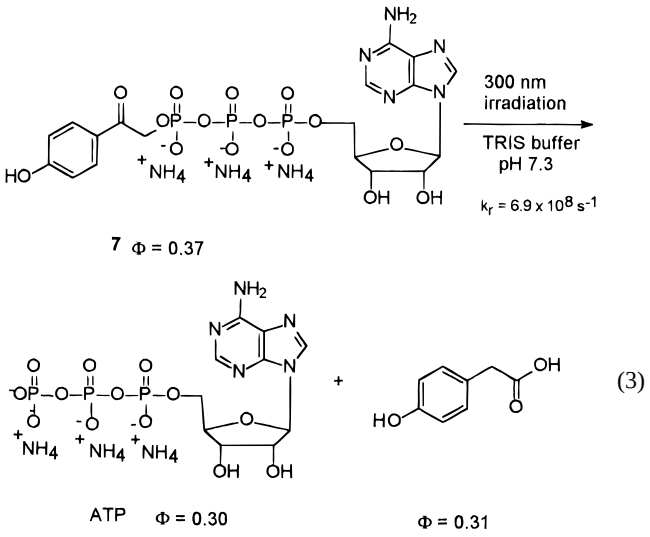
accordingly, we report here our recent studies on the synthesis, exploratory photochemistry, and quantitative mechanistic studies for substrate release from *p*-substituted phenacyl phototriggers.[22]

The photochemical release of phosphates and carboxylic acids from the *p*-methoxyphenacyl ester precursors had been reported previously by Sheehan,[23a,b] Epstein,[23c] and Baldwin.[19] However, for each of these examples, the fate of the phenacyl moiety was found to be the photoreduced form, *i.e.*, the corresponding acetophenone. These results were puzzling for two reasons: (1) Anderson and Reese[24] had reported as early as 1962 that *o*- and *p*-methoxyphenacyl as well as *p*-hydroxyphenacyl chloride gave a substantial amount of the rearranged substituted phenyl acetate ester when irradiated in methanol. (2) The results of our[2] study and that of Corrie and Trentham[15,18] on desyl phosphates would suggest a carbocation reaction intermediate which cyclizes in the desyl series to a furan, but in the *p*-hydroxyphenacyl analogues to a spiro diene dione proceeding through a carbonyl migration to the phenylacetic acid which was reported by Anderson and Reese.[24] For these reasons, we have reinvestigated and extended the study of some of these reactions and report here their application to the release of ATP (eq 3).[22]



(3)

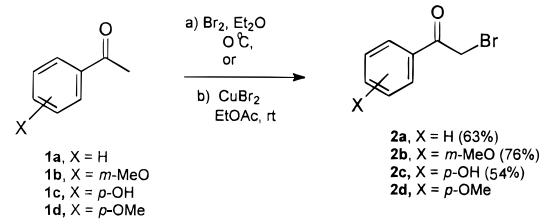
Results

A. Synthesis of *p*-Substituted Phenacyl Phosphates. In order to explore the effect of aryl substituents on the photochemistry, a series of phenacyl phosphates **3a**−**c** and **6c**−**h** were synthesized by the procedures shown in Scheme 1. *p*-Methoxyphenacyl phosphate **3d**, which had been reported previously, is included here for comparison.

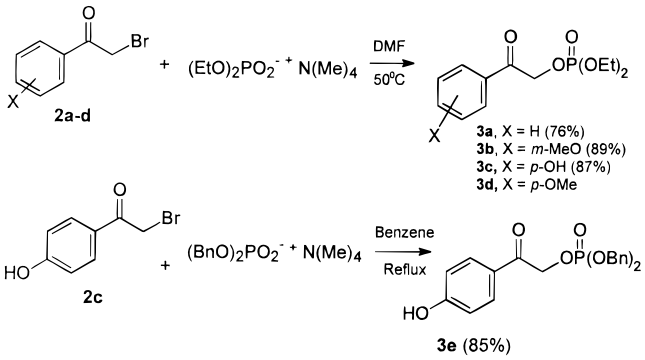
Unsubstituted **1a** and 3-methoxyacetophenone (**1b**) were converted to the α-bromoacetophenones (**2a**, 63% and **2b**, 76%, respectively) by acid-catalyzed bromination.[25] However, α-bromo-4-hydroxyacetophenone (**2c**) was synthesized by bromination of 4-hydroxyacetophenone (**1c**, 71%) with cupric bromide[26] to avoid dibromination. The α-bromoacetophenones **2a**−**d** were

Scheme 1

a. Bromination of acetophenones [24,25]



b. Conversion of α-Bromoacetophenones to Diethyl and Dibenzyl Phosphates [26]



next converted to the diethyl phosphates (**3a**−**d**) with tetramethylammonium diethyl phosphate in 76−89% yields by direct displacement of bromide,[27] taking advantage of the enhanced nucleofugacity of the leaving bromide when located α to a carbonyl. Similarly, 4-hydroxyphenacyl bromide (**2c**) was converted to 4-hydroxyphenacyl dibenzyl phosphate (**3c**, 85%) by reaction with tetramethylammonium dibenzyl phosphate in benzene under anhydrous conditions. Strictly anhydrous conditions were important for the displacement reactions with tetramethylammonium phosphates and the α-keto bromide because the presence of even small amounts of H_2O resulted in much lower yields.

Protection of ketone **3e** as its ketal **4e** was necessary to remove both benzyl protecting groups in the hydrogenolysis step and was accomplished with ethylene glycol in the presence of a catalytic amount of *p*-toluenesulfonic acid (*p*-TsOH) in benzene (Scheme 2).[28] Interestingly, without the protection of ketone **3e**, only one of the two benzyl groups was removed even after extended hydrogenation. Hydrogenolysis[29] of the benzyl groups on **4e** with H_2 on Pd/C followed by the treatment with 1% HCl gave 4-hydroxyphenacyl dihydrogen phosphate, which was further purified on an anion-exchange DEAE Sephadex column, producing 4-hydroxyphenacyl diammonium phosphate (**6c**) in a yield of 96%.

The synthesis of the *p*-hydroxyphenacyl-caged ATP (**7**, Scheme 2) required the coupling[18,30] of the monophosphate **6c** with activated ADP under rigorously anhydrous reaction conditions. Thus, the monophosphate **6c** and ADP were dried by sequential cycles of dissolution and evaporation in scrupulously dried solvents,[31] *i.e.*, pyridine or DMF, and then transformed

(22) This work has been communicated: Givens, R. S.; Park, C.-H. *Tetrahedron Lett.* **1996**, *37*, 6259−6262.

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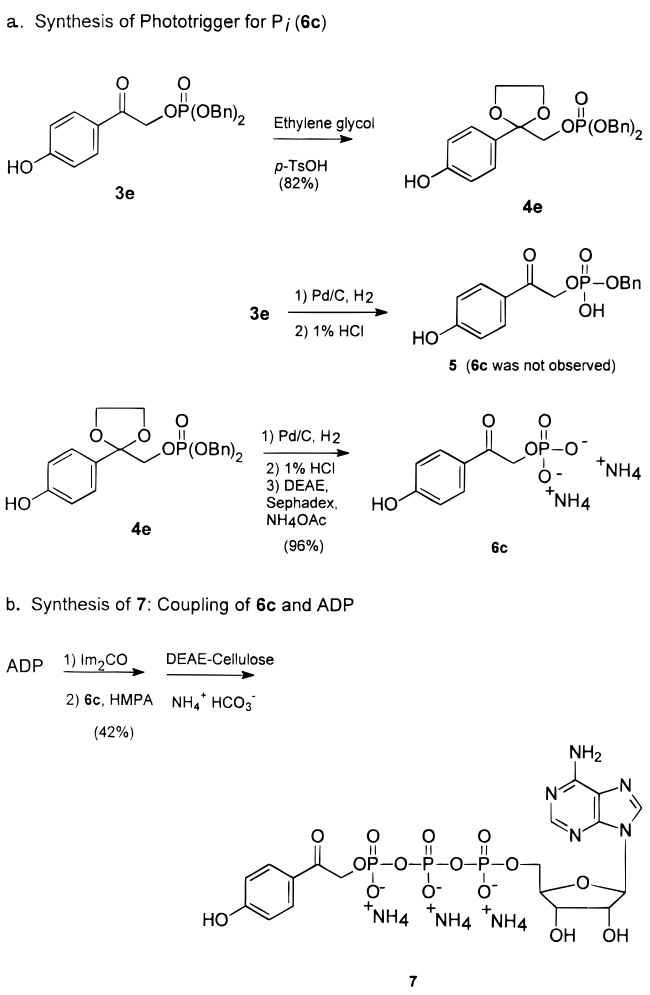
Scheme 2



to the tri-*n*-octylammonium salt. ADP was activated by first treating it with pyridine and tri-*n*-butylamine and then converting the salt to imidazolyl ADP by reaction with carbonyl diimidazole. The triphosphate was obtained from reaction of the two substrates for 72 h in HMPA (hexamethylphosphoramide) at room temperature with 20 min of sonication every 12 h, due to the poor solubility of the reactants. The product was purified by DEAE cellulose chromatography, eluting with a step gradient of aqueous ammonium bicarbonate. Purification by DEAE Sephadex chromatography with 10% methanol/ammonium acetate gave product of slightly higher purity but was a more tedious process. The yield of 4-hydroxyphenacyl ATP (**7**) from **6c** and ADP was 42%.

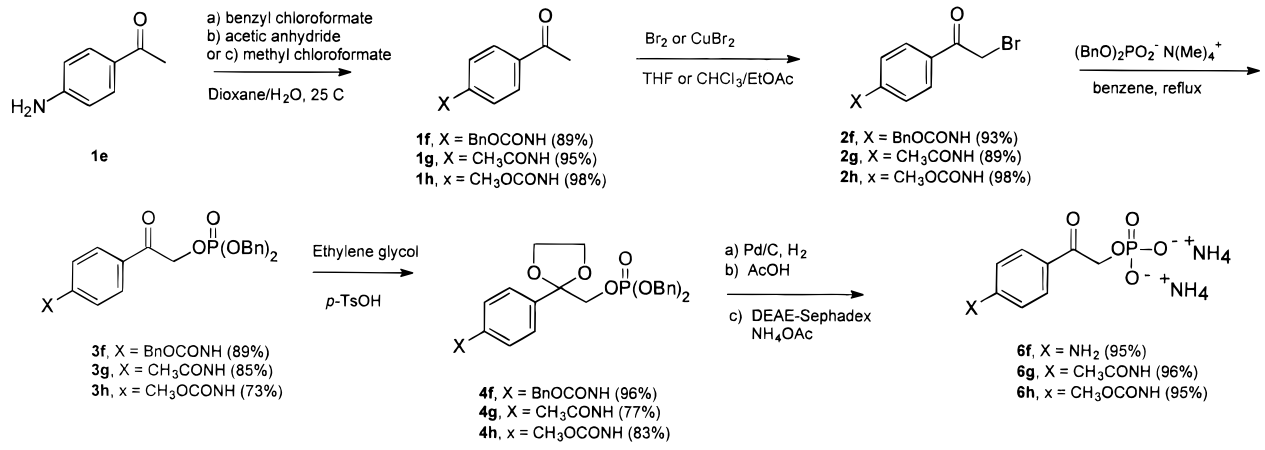
The syntheses of 4-aminophenacyl diammonium phosphate (**6f**), 4-acetamidophenacyl diammonium phosphate (**6g**), and 4-(carbomethoxyamino)phenacyl diammonium phosphate (**6h**) were accomplished by the same general strategy employed for 4-hydroxyphenacyl diammonium phosphate (Scheme 3). 4-Aminoacetophenone (**1e**) was converted to 4-((carbobenzyloxy)-amino)acetophenone (**1h**, 89%) before the bromination step to prevent formation of the undesired dibrominated products. This strategy also permitted the removal of the carbobenzyloxy group along with benzyl phosphate protecting groups under the same hydrogenolysis conditions as the phosphate group deprotection. Bromination of **1f**−**h** either by bromine[25] or by cupric bromide [26] gave the corresponding α-bromo derivatives **2f**−**h** (89−98%) followed by conversion[27] to the phenacyl dibenzyl phosphates **3f**−**h** (73−89%). Reaction of **3f**−**h** with ethylene glycol and catalytic amount of *p*-TsOH gave the ketals **4f**−**h**[28] in yields of 77−96%.

Hydrogenolysis[29] removed the two benzyl groups from the phosphate ligand along with the one in the carbobenzyloxy group of **4f**. Treatment with small amounts of 1% HCl easily removed the ketal group. Interestingly, the removal of the ketal from **4h** was much slower, requiring 48 h, in contrast to the 12 h needed for **4f** and **4g**. Conversion of the phenacyl dihydrogen phosphates to the diammonium phosphates was performed on the weakly anionic DEAE cellulose column by elution with ammonium bicarbonate. The ammonium salts were recrystallized from H_2O/MeOH solution to give the 4-amino- (**6f**, 95%), 4-acetamido- (**6g**, 96%), and 4-(carbomethoxyamino)phenacyl diammonium phosphate (**6h**, 96%), respectively.

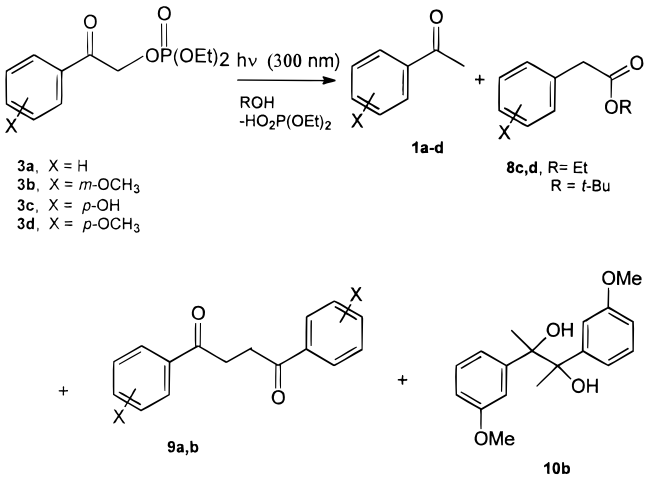
The stability of 4-hydroxyphenacyl ATP (**7**) was examined in buffered and unbuffered aqueous media. Solutions of **7** in Tris buffer (50 mM, pH 7.3), three different Ringer's solutions (pH 6.5), D_2O, and H_2O were stored in the dark at 25 °C and were periodically monitored by HPLC. The samples showed no loss of **7** over a 24 h period. In fact, for the D_2O solution of *p*-hydroxyphenacyl ATP, the NMR sample of the phototrigger was stable for over 100 days.

B. Photochemical Studies. The photochemistry of this limited collection of phenacyl phosphates is shown in Scheme 4 and eqs 4 and 5. Irradiation of all four diethyl esters **3a**−**d** in MeOH as well as **3c**,**d** in *t*-BuOH at 300 nm produced the corresponding reduction products, acetophenones **1a**−**d,** as primary products (Scheme 4). *m*-Methoxyphenacyl phosphate (**3b**) also formed radical coupling products **9b** and **10b**; whereas, the *p*-hydroxy (**3c**) and *p*-methoxy esters (**3d**) gave rearrangement products **8c**,**d** as their methyl or *tert*-butyl esters, respectively. Phosphates **3c**,**d** reacted with higher efficiencies and

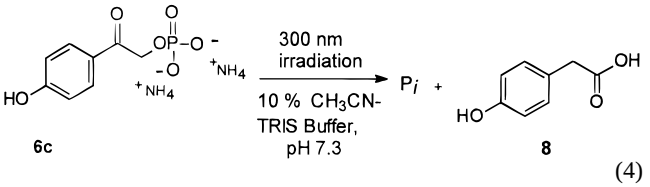
Scheme 3



Scheme 4



gave much better mass balances than the other two phenacyl diethyl phosphate derivatives.



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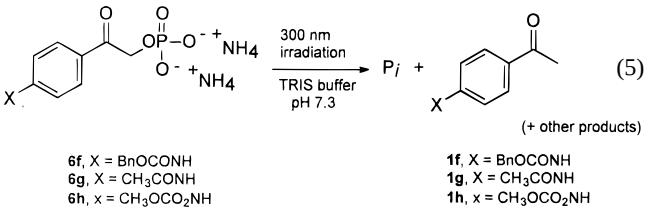


(5)

In contrast to the complex photochemistry of **3a**−**d**, *p*-hydroxyphenacyl phosphate (**6c**) and the caged ATP **7** were each converted at 300 nm exclusively to *p*-hydroxyphenylacetic acid and the released inorganic phosphate (P_i) or ATP, respectively. Essentially identical results were obtained whether the irradiations were conducted in Tris buffer or in the unbuffered aqueous media (*e.g.*, H_2O or D_2O, eq 4). No evidence of reduction or coupling products was obtained under these conditions.

Quantum efficiencies for the disappearance of *p*-hydroxyphenacyl derivatives and diethyl *p*-methoxyphenacyl phosphate[2b,c,22] are given in Table 1. Appearance efficiencies were also measured for the rearrangement, for some of the reduction products, and, in one case, for the substrate appearance, *i.e.*, ATP.

Sodium 2-naphthalenesulfonate successfully quenched the photorelease of ATP from *p*-hydroxyphenacyl ATP (**7**) and gave good Stern−Volmer (SV) kinetics (Table 2). The rate constant for the decay of the triplet, which is equated with the release rate for ATP, was 5.5×10^8 s^{-1}; whereas, the rate constant for disappearance of **7** was 6.9×10^8 s^{-1}, obtained from the SV slopes assuming a rate constant for diffusion in Tris buffer of 6.6×10^9 M^{-1} s^{-1}.

Photolyses of 4-amino- (**6f**), 4-acetamido- (**6g**), and 4-(carbomethoxyamino)phenacyl diammonium phosphate (**6h**) are shown in eq 5. In contrast to the 4-hydroxy derivatives, photoreactions for 4-aminophenacyl diammonium phosphate (**6f**) in buffered media at pH 7.3 had a low quantum efficiency for disappearance (<5%). Small amounts of the reduction product, 4-aminoacetophenone (**1e**), were also detected by HPLC of the photolysis mixture. Likewise, the reduction product **1g** from 4-acetamidophenacyl diammonium phosphate (**6g**) was obtained along with the corresponding dimers. In contrast, 4-(carbomethoxyamino)phenacyl diammonium phosphate (**6h**) did not produce the expected reduction products, instead giving a complex mixture of unidentified products (by HPLC). Quantum efficiencies for the disappearance of **6g**,**h** and the reduction products are shown in Table 3. ^{31}P NMR spectroscopy confirmed that all three aminoacetophenones triggered the release of inorganic phosphate (P_i) upon photolysis. However, the rearranged substituted phenyl acetates were not detected. Interestingly, it appears that these phototriggers release phosphate through a photoreduction process. We have not pursued the mechanism of this photorelease process.

A determination of the change in the pH (ΔpH) for a solution of each of the three phenacyl diammonium phosphates **6c**, **6f**, and **6g** in H_2O was measured as a function of photolysis time (Table 4). The ΔpH for solutions of 4-hydroxyphenacyl diammonium phosphate (**6c**) and 4-acetamidophenacyl diammonium phosphate (**6g**) were 0.7 and 1.1, respectively. However, the ΔpH of solutions of 4-aminophenacyl diammonium phosphate (**6f**) was practically 0, a result of the low quantum efficiency for the release of inorganic phosphate (P_i) from this phototrigger.

Discussion

From among the limited selection of substituted phenacyl phosphates examined in our study, the most versatile phototrigger appears to be *p*-hydroxyphenacyl. The *meta*-substituted and unsubstituted phosphates yield complex mixtures of products in MeOH including acetophenone products from photoreduction and dimeric products resulting from radical coupling processes. In contrast, both *p*-methoxy- and *p*-hydroxyphenacyl phosphate rearranged to the *p*-methoxyphenyl- and *p*-hydroxyphenylacetates when photolyzed in hydroxylic solvents, MeOH or H_2O, respectively.

This predominance of the photorearrangement is even more remarkable since apparently only these two oxygen-containing electron donors are capable of promoting the phenacyl to phenylacetate rearrangement. The three *p*-amino derivatives **6f**−**h** gave only photoreduction products of the trigger.

For the *p*-hydroxyphenacyl derivatives which possess both a requisite *para* electron-donating group and a good nucleofuge, the rearrangements are efficient and virtually free of byproducts, as shown in Table 1. The disappearance efficiencies for the caged phosphates **6c** and **7** are 0.37 ± 0.04 and product yields are essentially quantitative by NMR. The measured appearance quantum efficiencies were $0.30(\pm 0.03)$ for the formation of the two photoproducts from **7**.

The results for the phosphate series are in excellent agreement with the results reported earlier by Anderson and Reese.[24] In that study, only strong electron-donating substituents, *i.e.*, OH or OCH_3, located either *ortho* or *para* to the carbonyl underwent aryl group migration to the substituted phenylacetic acids. Additional examples of aryl migrations bearing strongly electron-donating *para* substituents are found in ground state chemistry as well, for example, the well-established neighboring phenyl group participation[32] observed in the solvolysis of β-anisylethyl tosylates, brosylates, and bromides. Our results with the *p*-hydroxyphenacyl derivatives are congruent with the earlier suggestion that arylmethyl phosphates yield a photoproduct distribution consistent with a carbocation intermediates[2] and give a Hammett substituent correlation with a negative slope of 0.9

(32) Cram, D. J. *J. Am. Chem. Soc.* **1952,** *74*, 2129−2137 and references therein.

Table 1. Quantum Efficiencies for the Photorearrangements of 4-Hydroxy- (**3c**, **6c**, and **7**) and 4-Methoxyphenacyl (**3d**)[2b,c,19,23c] Phosphates[a,b]

aryl substituent	R_1	R_2	solvent	Φ_{dis}	Φ_{red}	$\Phi_{rearrange}$	$\Phi_{phosphate}$	ref
p-MeO[b] (**3d**)	Et	Et	CH_3OH	0.42	0.07	0.20		2b,c, 23c
(**3d**)	Et	Et	CD_3OD		0.013	0.14		2b
(**3d**)	Et	Et	CH_3OD		0.053	0.11		2b
p-OH[c] (**3c**)	Et	Et	CH_3OH	0.77	0.05	0.33		*e*
p-OH[c,d] (**6c**)	NH_4+	NH_4+	aq CH_3CN	0.38	0.00	0.12		*e*
(**7**)	ATP	NH_4+	Tris	0.37	0.00	0.31	0.30	*e*

[a] Error limits for the quantum efficiencies are estimated to be ±10%. [b] Irradiated at 300 nm. [c] Irradiated at 350 nm. [d] 10%CH_3CN/90% Tris buffer. [e] This work.

Table 2. Stern−Volmer Quenching Data for Photolysis of **7** in Tris Buffer at 350 nm[a]

sodium naphthalenesulfonate [mM]	Φ_{dis} (**7**)	Φ_{rearr} (**8**)	Φ_{ATP}
0	0.37 (0.04)	0.31 (0.03)	0.30 (0.03)
50	0.31	0.25	0.25
100	0.27	0.20	0.23
150	0.24	0.16	0.19
slope (K_{sv}), M^{-1}	3.56	6.38	3.61
τ, ns	0.54 (0.09)	0.97 (0.10)	0.55 (0.06)
10^8 k, s^{-1}	6.90 (1.15)	3.20 (0.33)	5.50 (1.00)

[a] Error shown in parenthesis.

Table 3. Quantum Efficiencies for *p*-Substituted Aminophenacyl Diammonium Phosphates **6f**−**h** in Tris Buffer at 300 nm[a]

p-substituent	concn (mM)	Φ_{dis}	Φ_{red}	others
4-NH_2 (**6f**)	19	<0.05	<0.05	
4-CH_3CONH (**6g**)	6.5	0.38	0.11	dimers
4-CH_3OCONH (**6h**)	4.2	0.34		2 unknowns

[a] Error limits for the quantum efficiencies are estimated to be 10%.

Table 4. pH Changes for the Nonbuffered Photolysis Solution of Phenacyl Diammonium Phosphates **6c**, **6f**, and **6g** in H_2O[a]

		pH		
p-substituent	concn (mM)	before	after	ΔpH
4-OH (**6c**)	7.5	7.4	6.7	0.7
4-NH_2 (**6f**)	7.5	6.8	6.7	0.1
4-CH_3CONH (**6g**)	7.5	4.7	3.6	1.1

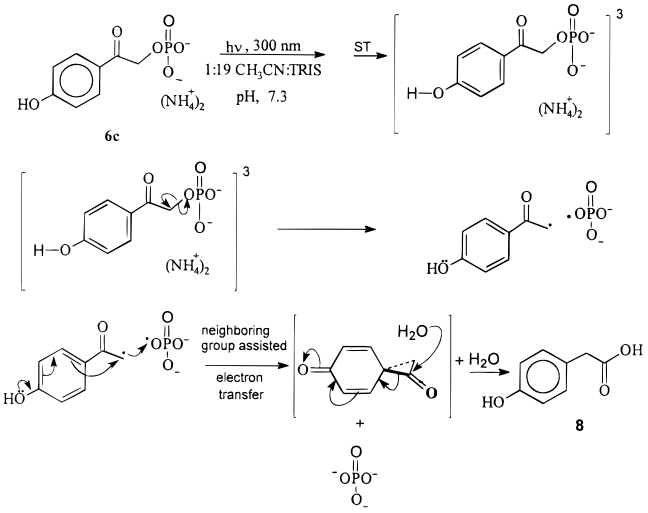
[a] Irradiated at 300 nm for 10 min.

versus Hammett σ reflecting positive charge character in the product-forming step.[33]

Quenching studies of **7** with sodium 2-naphthalenesulfonate (Table 2) confirmed the triplet as the reactive excited state for the photorelease and the rearrangement reactions. The linear dependence of the Stern−Volmer quenching by 2-naphthalenesulfonic acid to greater than 90% of the phosphate photorelease accords with an exclusively triplet state pathway for the reaction and parallels our earlier results for the desyl phosphates[2b,c] and desyl amino acids.[1] The rate constants calculated for release of phosphates derived from the SV quenching were 6.9×10^8 s^{-1} for the disappearance of **7** and 5.5×10^8 s^{-1} for the appearance of ATP and the rearranged *p*-hydroxyphenylacetic acid (**8**). These rate constants compare very favorably with the value of 2.9×10^8 s^{-1} for desyl-caged cAMP[2b,c] and 1.2×10^7 s^{-1} obtained for caged γ-*O*-desyl glutamate.[1]

Other than the determination of the multiplicity, the overall mechanistic picture for the photoreaction remains to be established. A reasonable suggestion for the triplet is homolysis to

Scheme 5



a radical pair that undergoes subsequent electron transfer to an ion pair, as outlined in Scheme 5 for release of phosphate form **6c**.[34] Pincock[34a,b] has proposed this mechanistic pathway for the photochemical release of carboxylates from benzyl and naphthyl esters via the excited singlet state during irradiation of the esters in methanol. Recently, Peters[34c] provided additional evidence for this pathway for the photosolvolysis of benzhydryl chlorides in polar media such as acetonitrile. Peters suggested that as much as 30% of the contact ion pairs generated from the singlet state of *p*-methoxybenzhydryl chloride originates from a geminate radical pair via an electron transfer (ET) process that competes with diffusion of the radical pair from the cage. The effect of substituents on the partitioning of the ET−diffusion competition is particularly significant in the benzhydryl series. The ion pair formation must be negligible for unsubstituted and *p*-methylbenzhydryl chlorides but dramatically increases to 20% or higher with methoxy substitution on one or both of the aryl rings.

For our studies, either water or buffer (highly polar, ionic media) is the solvent, the substituent is a *p*-hydroxy group (a better electron donor), and the radical pair is initially a triplet. While this combination of factors has not been present in any of the photochemical substrates to date that have been studied mechanistically, a sequence of steps parallel to those reported by Peters[34c] may very well obtain here. This proposed mechanism is shown in Scheme 5.

Structural features of this new α-keto phototrigger and its efficient photorearrangement to *p*-hydroxyphenylacetic acid invite some additional comments. First, this new caging group lacks a chiral center, allaying any concerns about generating a

(33) (a) Givens, R. S.; Matuszewski, B.; Athey, P. S.; Stoner, R. M. *J. Am. Chem. Soc.* **1990**, *112*, 6016−6023. (b) Givens, R. S.; Singh, R. *Tetrahedron Lett.* **1991**, 7013−7017.

(34) (a) Hilborn, J. W.; MacKnight, E.; Pincock, J. A.; Wedge, P. J. *J. Am. Chem. Soc.* **1994**, *116*, 3337−3346. (b) Pincock, J. A. *Acc. Chem. Res.* **1997**, *30*, 43−49. (c) Lipson, M.; Deniz, A. A.; Peters, K. S. *J. Am. Chem. Soc.* **1996**, *118*, 2992−2997.

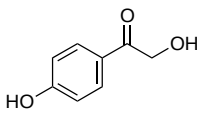
Table 5. Photoefficiencies of 4-Hydroxyphenacyl Adenosine 5′-Triphosphate (**7**) in Buffered Solution

phototriggers	conditions[a]	pH	$\Phi_{4(dis)}$	$\Phi_{ATP(app)}$	$\Phi_{8(app)}$
7	tris Buffer[b]	7.3	0.37	0.30	0.31
6c	45% Tris/55% CH_3CN	6.5	0.38	*c*	0.12

[a] Irradiated at 300 nm at room temperature. [b] Stability was demonstrated for this buffer, H_2O, D_2O and for Ringer's solutions I, II, and III for greater than 24 h. For D_2O, the phototrigger was stable for over 100 h by HPLC. [c] Not determined

mixture of diastereomers when employed with a chiral substrate. This feature will significantly simplify the purification and characterization of the synthetic caged substrates for phenacyl cages. Second, unlike 2-NB, 2-NPE, and even the desyl cages, the chromophore of the rearranged photoproduct, *p*-hydroxyphenylacetic acid, does not compete for the incident radiation in the 300−400 nm region, since it is blue-shifted relative to the phenacyl cage. This eliminates product interference of the incident light absorption by the phototrigger which would reduce the yields and prevent complete conversion in the release of the substrate. Third, the attachment of the substrate to a primary methylene, α to a carbonyl, should stabilize the derivative to any "S_N1-like" ground state release of the substrate. Direct nucleophilic substitution, however, remains a potential debilitating reaction.

As a test of the solvolytic stability, *p*-hydroxyphenacyl ATP was subjected to various buffers and to aqueous media employed in typical biological studies. Six different media were explored in this study: H_2O, D_2O, Tris, and Ringers solutions I, II, and III. Solutions of the two ester in the series of solvents and buffers were sampled periodically over a 24 h period and the aliquots examined by HPLC. As shown in Table 5, the *p*-hydroxyphenacyl ATP phototrigger was stable for at least 24 h under all conditions examined. In fact, **7** remained unchanged in D_2O for over 100 h. Additional studies with other *p*-hydroxyphenacyl carboxylic esters have also been stable to solvolytic conditions.[35] In addition, HPLC analysis confirmed that neither of these two esters were measurably degraded or hydrolyzed to α,*p*-dihydroxyacetophenone (**11**) when stored in the dark.



11

In contrast, Corrie and Trentham *et al.*[7] found that 3′,5′-dimethoxybenzoin-caged ATP (**12**) undergoes slow hydrolysis to ADP and 3′,5′-dimethoxybenzoin phosphate both in phosphate and carbonate buffers. The instability of **12** was attributed by the authors to anchimeric assistance of the carbonyl of the hydrated benzoyl group, as shown in Scheme 6.[7,8] That this was not observed for the *p*-hydroxyphenacyl series may be due in part to the greater electron donating ability of the hydroxy group to the carbonyl, rendering it less electrophilic and thus less reactive toward hydration. It should be noted that Iwamura *et al.*[21,36] reported that desyl cAMP hydrolyzed spontaneously in the absence of light in Ringer's solution containing 1% DMSO to yield benzoin and cAMP. This hydrolysis must be occurring by some other mechanism, *e.g.*, S_N2 displacement of cAMP. In our series, we have not encountered the equivalent hydrolyses of *p*-hydroxyphenacyl-caged phosphates **6c** or **7** in Ringer's solutions I, II, and III or Tris or in H_2O or D_2O at temperatures between 25 and 40 °C over a 24 h period to yield P_i and ATP, which would have been accompanied by α,*p*-dihydroxyacetophenone (**11**). No **11** was detected in any of these control experiments. Thus, caged phosphates **6c** and **7** are stable to most common solvents, yet trigger the release of the phosphate ligand upon 300−350 nm radiation.

(35) C.-H. Park, W. Bartlett, A. Jung, J. Weber, and R. Givens, work in progress.

(36) For additional details, see: ref 21. Futura, T.; Torigai, H.; Iwamura, M. *Chem Lett.* **1993**, 1179−1182. Futura, T.; Torigai, H.; Osawa, T.; Iwamura, M. *J. Chem. Soc., Perkin Trans. 1* **1993**, 3139−3142.

Conclusions

The *p*-hydroxyphenacyl cage fulfills several of the criteria advanced by Lester,[6c] especially the requirements of a high efficiency, a benign photobyproduct, and good stability of the caged derivative in ionic media. Most importantly, the p-hydroxyphenacyl phototrigger has no chiral center but does possess a much improved aqueous buffer solubility when compared with that of the desyl analogues. The photorelease is a primary process, occurring with rate constants of 10^7-10^8 s^{-1}, several orders of magnitude faster that the 2NB and 2NPE derivatives and approximately the same as the desyl analogues. These very desirable features of the new generation of phototriggers provide a promising future for their application to mechanistic biochemistry, physiology, and related fields.

Experimental Section

General Method. All nonaqueous reactions were performed under an atmosphere of argon with a slight positive pressure and continuous magnetic stirring. All reagents were used as received without further purification unless otherwise stated. All solvents used for chromatographic purposes were of reagent grade or better or were distilled prior to used. Acetophenone was distilled under vacuum. Triethylamine was dried by refluxing over potassium hydroxide and distilling under argon, collecting the middle fraction, and stored in the dark. Pyridine and *N*,*N*-dimethylformamide were dried over phosphorus pentoxide and distilled under reduced pressure, collecting the middle fraction, and stored over sodium hydroxide and molecular sieves, respectively. Benzene was distilled from sodium/benzophenone ketyl prior to use. Technical grade diethyl ether, methanol, methylene chloride, and tetrahydrofuran were distilled from calcium hydride. Flash column chromatography was performed with ethyl acetate/methylene chloride to purify the phosphate triesters. All NMR spectra are reported in ppm (δ) with either tetramethylsilane (^1H and ^{13}C) or 85% phosphoric acid (^{31}P) as the internal standards. Melting points are uncorrected. Mass spectra were provided by the Mass Spectrometry Laboratory at the University of Kansas.

2-Bromoacetophenone (**2a**) was prepared by the method of Cowper and Davidson.[25]

2-Bromo-3′-methoxyacetophenone (**2b**)[25] was prepared by the same method as above to yield 11.6 g (76%) of 2-bromo-3′-methoxyacetophenone (**2b**): mp 55−57 °C; ^1H NMR ($CDCl_3$) δ 3.82 (s, 3H), 4.51 (s, 2H), 7.13−7.60 (m, 4H); IR ($CHCl_3$) 3100, 2954, 1684, 1598, 1583, 1037 cm^{-1}; UV-vis (CH_3CN) λ_{max} (ϵ) 312 (2881), 253 (8413), 218 (21 900).

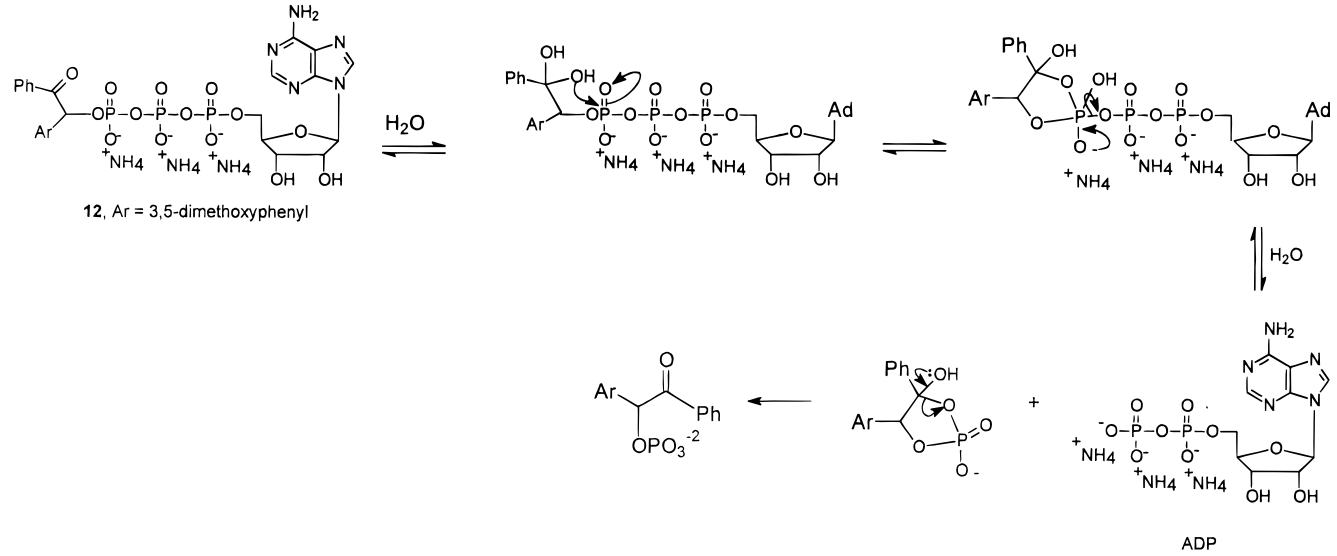
2-Bromo-4′-hydroxyacetophenone (**2c**) was prepared by the method of Buu-Hoi and Lavit[26] to give 2.53 g (11.8 mmol, 53.9%) of 2-bromo-4′-hydroxyacetophenone (**2c**): mp 129−131 °C; ^1H NMR (acetone-d_6) δ 4.64 (s, 2H), 6.97 (d, $J = 8.7$ Hz, 2H), 7.97 (d, $J = 8.7$ Hz, 2H); ^{13}C NMR (acetone-d_6) δ 38.38, 122.22, 132.83, 138.29, 169.31, 196.35.

Phenacyl diethyl phosphate (**3a**), **3-methoxyphenacyl diethyl phosphate** (**3b**), and **4-hydroxyphenacyl diethyl phosphate** (**3c**) were prepared by the general method of Zwiezak and Kluba.[27,37]

4-Hydroxyphenacyl Dibenzyl Phosphate (**3e**).[27] A mixture of tetramethylammonium dibenzyl phosphate (1.60 g, 4.70 mmol), 4-hydroxyphenacyl bromide (**2c**, 1.00 g, 4.70 mmol), and 20 mL of benzene was placed in a 50 mL round-bottom flask and refluxed with efficient

(37) See Supporting Information for details including experimental procedures and spectral data.

Scheme 6



stirring for 2 h. The solution was then cooled to room temperature and extracted with H_2O/EtOAc solution. After the solvent was removed, a pale yellow solid product was purified by silica gel flash column chromatography (CH_2Cl_2/EtOAc 80:20) to yield 1.63 g (85%) of 4-hydroxyphenacyl dibenzyl phosphate (**3e**): mp 116.5−118 °C; ^{1}H NMR (DMSO-d_6) δ 5.13 (d, J = 7.8 Hz, 4H), 5.36 (d, J = 10.7 Hz, 2H), 6.87 (d, J = 7.6 Hz, 2H), 7.38 (m, 10H), 7.82 (d, J = 7.6 Hz, 2H), 10.51 (s, 1H); ^{13}C NMR (DMSO-d_6) δ 68.65 (d, J = 22.3 Hz), 68.73 (d, J = 25.8 Hz), 115.39, 125.14, 127.81, 128.30, 128.41, 130.34, 136.01 (d, J = 29.8 Hz), 162.65, 190.80 (d, J = 16.5 Hz); ^{31}P NMR (DMSO-d_6) δ 0.56; IR ($CHCl_3$) 3212, 1684, 1600, 1456, 1373, 1281, 1115, 984 cm^{-1}; exact mass calcd for $C_{22}H_{21}O_6P$ 412.1154, found 412.1163. Anal. Calcd for $C_{22}H_{21}O_6P$: C, 64.08; H, 5.13. Found: C, 64.46; H, 5.18.

4-Hydroxyphenacyl Dibenzyl Phosphate (Ethylene Ketal) (4e).[28] To the solution of 4-hydroxyphenacyl dibenzyl phosphate (**3e**, 3.45 g, 8.4 mmol) in 100 mL of benzene, a catalytic amount of *p*-TsOH (79.9 mg, 0.42 mmol) was added followed by the addition of excess amount of ethylene glycol (8.86 g, 145.1 mmol). The solution was refluxed for 12 h with a Dean−Stark apparatus to remove the water that was generated. When reaction was complete, $NaHCO_3$ (0.71 g, 8.4 mmol) was added to neutralize the mixture. After benzene was removed *in vacuo*, ethylene glycol was removed by extraction with H_2O/EtOAc. Further purification was done by recrystallization from Et_2O to give 3.19 g (82.1%) of 4-hydroxyphenacyl dibenzyl phosphate (ethylene ketal) (**4e**): mp 97−98 °C; ^{1}H NMR (DMSO-d_6) δ 3.80 (m, 2H), 4.00 (m, 2H), 4.03 (d, J = 6.7 Hz, 2H), 4.95 (d, J = 7.7 Hz, 4H), 6.74 (d, J = 8.6 Hz, 2H), 7.27 (d, J = 8.6 Hz, 2H), 7.33−7.38 (m, 10 H), 9.53 (s, 1H); ^{13}C NMR (DMSO-d_6) δ 64.91, 68.34 (d, J = 22.7 Hz), 69.03 (d, J = 24.9 Hz), 107.06 (d, J =33.4 Hz), 114.71, 127.21, 127.71, 128.28, 128.40, 129.11, 135.96 (d, J = 28.3 Hz), 157.60; ^{31}P NMR (DMSO-d_6) δ −0.02; IR ($CHCl_3$) 3280, 1610, 1512, 1455, 1273, 1168, 1002 cm^{-1}; exact mass calcd for $C_{24}H_{25}O_7P$ 456.1416, found 456.1400. Anal. Calcd for $C_{24}H_{25}O_7P$: C, 63.16; H, 5.52. Found: C, 63.00; H, 5.49.

Exploratory Hydrogenation of 4-Hydroxyphenacyl Dibenzyl Phosphate (3e).[29] To 4-hydroxyphenacyl dibenzyl phosphate (**3e**, 0.35 g, 0.84 mmol) dissolved in 10 mL of MeOH was added 35 mg of 10% Pd/C. The same hydrogenation procedure (see below) was used as that employed for 4-hydroxyphenacyl dibenzyl phosphate (ethylene ketal) (**4e**). Only one benzyl group was removed even with extended hydrogenation to give 4-hydroxyphenacyl benzyl phosphate (**5c**).

4-Hydroxyphenacyl Diammonium Phosphate (6c).[29] To 4-hydroxyphenacyl dibenzyl phosphate (ethylene ketal) (**4e**, 1.86 g, 4.1 mmol) dissolved in 30 mL of MeOH was added 186 mg of 10% Pd/C. The solution was hydrogenated under 10 psi of H_2 with 45 min of stirring followed by the addition of 1% HCl (1 mL). After filtration, the filtrate was concentrated *in vacuo* to yield a viscous liquid which was loaded on 5 g of DEAE Sephadex column pretreated with 10 mM of ammonium acetate. The products were eluted with stepping gradient of ammonium acetate solution (200 mL each of 0.0−0.2 M solution with 0.05 M increments). Diammonium salt **6c** was eluted at 0.2 M ammonium acetate. Excess ammonium acetate and water were lyophilized from the solution to yield a solid which was recrystallized from H_2O/MeOH to give 1.05 g (96.2%) of 4-hydroxyphenacyl diammonium phosphate (**6c**): mp 177−192 °C (dec); ^{1}H NMR (D_2O) δ 5.08 (d, J = 5.6 Hz, 2H), 6.92 (d, J = 8.8 Hz, 2H), 7.89 (d, J = 8.8 Hz, 2H); ^{13}C NMR (D_2O) δ 69.37 (d, J = 14.2 Hz), 118.46, 128.59, 133.42, 165.03, 200.50 (d, J = 33.3 Hz); ^{31}P NMR (D_2O) δ 1.18; IR (KBr) 3350, 3088, 2950, 1679, 1594, 1570, 1440, 1268, 1090 cm^{-1}; UV-vis (CH_3CN/H_2O) λ_{max} (ϵ) 220 (8400), 282 (14 000). Anal. Calcd for $C_8H_{15}N_2O_6P$: C, 36.10; H, 5.68; N, 10.52. Found: C, 35.82; H, 5.38; N, 10.33.

4-Hydroxyphenacyl Adenosine 5′-Triphosphate, Triammonium Salt (7).[30] Dowex 50W resin (30 g) in a sintered glass funnel was treated with 5% hydrochloric acid (60 mL × 2) and washed with water until the filtrate was neutral. The resin was then treated with a 20% aqueous pyridine solution and washed with water until the filtrate was neutral. A solution of adenosine 5′-diphosphate (ADP, potassium salt, 1.105 g, 2.33 mmol) in 20 mL of water was stirred with the resin for 5 min, the resin was filtered and washed with water (20 mL × 4). Into the filtrate was added tributylamine (0.87 g, 4.7 mmol) followed by 30 min of stirring. The solution was lyophilized to yield 1.95 g of a white powder which was further dried by sequential cycles of dissolution and evaporation of dry pyridine (10 mL × 2) and dry DMF (10 mL × 2). The dried residue was redissolved in 10 mL of dry DMF, carbonyl diimidazole (1.65 g, 10 mmol) was added, and the resulting solution was stirred under argon for 24 h at ambient temperature. The reaction was quenched by adding methanol (0.3 mL, 8 mmol); the resulting solution was stirred for 1 h, and the solvent was removed *in vacuo*. Meanwhile, a solution of 4-hydroxyphenacyl diammonium phosphate (**6e**, 1.17 g, 4.44 mmol) in 40 mL of water was stirred for 30 min with the activated Dowex 50W resin (30 g, pyridinium form) as described above. The resin was filtered and washed with water (20 mL × 3). Tri-*n*-octylamine (1.65 g, 4.44 mmol) was added to the filtrate, and the mixture was lyophilized to yield 3.21 g of a white powder which was dissolved in dry pyridine (15 mL × 2) followed by evaporation and then dissolved in dry DMF (15 mL × 2) and evaporated. The gummy residue was redissolved in 10 mL of dry DMF and added to the above ADP imidazole solution. The DMF was evaporated *in vacuo* at room temperature and hexamethylphosphoramide (HMPA, 30 mL) added to the pale yellow residue. The solution was stirred under argon for 3 days during which time the solution was sonicated for two 20 min periods each day. The reaction was quenched by adding 120 mL of water and washed with chloroform (100 mL × 4) and hexane (100 mL). The aqueous layer was lyophilized and purified by DEAE-cellulose (HCO_3^- form, 500 mL dry volume) column eluted with a stepping gradient of ammonium bicarbonate solution with

the concentrations from 0.0 to 0.3 M in 0.05 M increments. Fractions with only one component were combined and lyophilized to yield 0.55 g (42.3%) of 4-hydroxyphenacyl adenosine 5′-triphosphate, triammonium salt (**7**): mp 137 °C (dec); ^{1}H NMR (D_2O) δ 4.21 (m, 1H), 4.27 (s, 2H), 4.37 (t, J = 4.3 Hz, 1H), 4.44 (t, J = 21.5 Hz, 1H), 5.10 (m, 2H), 5.77 (d, J = 5.1 Hz, 1H), 6.52 (d, J = 8.5, 2H), 7.46 (d, J = 8.8 Hz, 2H), 8.02 (s, 1H), 8.29 (s, 1H); ^{13}C NMR (D_2O) δ 68.12 (d, J = 20.7 Hz), 70.87 (d, J = 19.4 Hz), 72.92, 77.77, 86.51 (d, J = 37.1 Hz), 90.20, 117.93, 121.14, 128.20, 133.00, 142.91, 151.02, 152.90, 156.33, 164.12, 197.95 (d, J = 33.2 Hz); ^{31}P NMR (D_2O) δ −25.42 (t, J = 48.3 Hz), −14.09 (d, J = 46.1 Hz), −13.74 (d, J = 49.4 Hz); IR (KBr) 3320, 3150, 2991, 2821, 1710, 1679, 1594, 1570, 1453, 1372, 1248, 1112, 1041 cm^{-1}; UV-vis (CH_3CN/H_2O) λ_{max} (ϵ) 286 (14 600); exact mass calcd for $C_{18}H_{22}O_{15}P_3$ (free acid) 641.0482, found 641.0460.

4-((Carbobenzyloxy)amino)acetophenone (**1f**). To a solution of 4-aminoacetophenone (**1e**, 5 g, 37 mmol) and NaOH (1.49 g) in 50 mL of dioxane/H_2O (70:30) was added dropwise benzyl chloroformate (6.3 g) for 30 min at 0 °C followed by 2 h of vigorous stirring at room temperature. After the reaction was complete, the solvent was removed *in vacuo*. The resulting solid was extracted with EtOAc/H_2O. The organic layer was concentrated and purified by silica gel column chromatography (CH_2Cl_2/hexane 50:50). Further purification was accomplished by recrystallization from CH_2Cl_2/hexane giving 8.84 g (89%) of yellowish crystalline 4-((carbobenzyloxy)amino)acetophenone (**1f**): mp 123.5−124.5 °C; ^{1}H NMR ($CDCl_3$) δ 2.56 (s, 3H), 5.22 (s, 2H), 7.39−7.41 (m, 5H), 7.49 (d, J = 8.7 Hz, 2H), 7.93 (d, J = 8.7 Hz, 2H); ^{13}C NMR (DMSO-d_6) δ 26.28, 66.27, 117.22, 128.08, 128.12, 128.40, 129.49, 130.99, 136.21, 143.55, 153.11, 196.43; IR ($CHCl_3$) 3426, 3050, 2935, 1739, 1676, 1600, 1524, 1500, 1409, 1359, 1313, 1272, 1178 cm^{-1}; FABMS *m/z* (rel intensity) 185 (72), 270 (M + 1, 100); exact mass calcd for $C_{16}H_{15}NO_3$ (M + H) 270.1130, found 270.1113.

4-((Carbobenzyloxy)amino)phenacyl Bromide (**2f**).[25] To a solution of 4-((carbobenzyloxy)amino)acetophenone (**1f**, 0.92 g, 3.7 mmol) in 30 mL of THF containing a catalytic amount of $AlCl_3$ (10 mg) was added 0.55 g of Br_2 dropwise for 30 min at 0 °C. After completion of the addition, the solution turned to pale yellow within 10 min. The solvent was removed *in vacuo* followed by the extraction of the resulting solid with H_2O/EtOAc. The organic layer was concentrated and purified by silica gel column chromatography (EtOAc/hexane 30:70). Further purification was accomplished by recrystallization from EtOAc/hexane to give 1.1 g (93%) of 4-((carbobenzyloxy)amino)phenacyl bromide (**2f**): mp 165 °C (dec); ^{1}H NMR (acetone-d_6) δ 4.70 (s, 2H), 5.22 (s, 2H), 7.38−7.46 (m, 5H), 7.75 (d, J = 8.8 Hz, 2H), 8.04 (d, J = 8.8 Hz, 2H), 9.23 (s, 1H); ^{13}C NMR (DMSO-d_6) δ 33.68, 66.13, 117.31, 127.92, 128.13, 128.20, 128.43, 130.23, 136.16, 144.22, 153.08, 190.16; IR (mineral oil) 3355, 3190, 3100, 2929, 2854, 1732, 1682, 1592, 1533, 1460, 1415, 1284, 1219, 1181, 1045, 972 cm^{-1}.

4-((Carbobenzyloxy)amino)phenacyl Dibenzyl Phosphate (**3f**).[27] To a solution of tetramethylammonium dibenzyl phosphate (0.6 g, 1.7 mmol) in hot benzene was added 0.5 g (1.44 mmol) of 4-((carbobenzyloxy)amino)phenacyl bromide (**2f**). After 2 h at reflux, the resulting solution was cooled to ambient temperature and the solvent was removed *in vacuo*. The resulting solid was purified by silica gel column chromatography (EtOAc/CH_2Cl_2 20:80) to give 0.70 g (89%) of 4-((carbobenzyloxy)amino)phenacyl dibenzyl phosphate (**3f**): mp 112−113 °C; ^{1}H NMR (acetone-d_6) δ 5.17 (s, 2H), 5.21 (d, J = 5.2 Hz, 4H), 5.38 (d, J = 11.1 Hz, 2H), 7.36−7.47 (m, 15H), 7.45 (d, J = 8.9 Hz, 2H), 7.95 (d, J = 8.9 Hz, 2H), 9.23 (s, 1H); ^{13}C NMR (acetone-d_6) δ 73.29, 75.56 (d, J = 22.2 Hz), 75.83 (d, J = 24.6 Hz), 124.35 (d, J = 30.0 Hz), 134.79, 134.95, 135.01, 135.09, 135.28, 135.38, 136.04, 137.52, 143.41, 151.15, 151.25, 159.97, 197.79 (d, J = 16.2 Hz); ^{31}P NMR (acetone-d_6) δ −2.07; IR ($CHCl_3$) 3430, 3048, 2928, 2879, 1737, 1698, 1598, 1525, 1500, 1438, 1410, 1209, 1178 cm^{-1}; UV-vis (CH_3CN) λ_{max} (ϵ) 289 (22 500); FABMS *m/z* (rel intensity) 105 (23), 117 (84), 185 (100), 277 (30), 456 (17), 546 (M + 1, 83); exact mass calcd for $C_{30}H_{29}NO_7P$ (M + H) 546.1682, found 546.1703.

4-((Carbobenzyloxy)amino)phenacyl Dibenzyl Phosphate (Ethylene Ketal) (**4f**).[28] To a solution of 4-((carbobenzyloxy)amino)-phenacyl dibenzyl phosphate (**3f**, 3.5 g, 6.4 mmol) in 50 mL of benzene was added a catalytic amount of *p*-TsOH (60.9 mg) and an excess of ethylene glycol (6.75 g). The solution was refluxed and stirred for 24 h using a Dean−Stark apparatus to remove the water. After the reaction was complete, the mixture was cooled to ambient temperature and neutralized by addition of $NaHCO_3$ (0.54 g) followed by removal of the solvent *in vacuo*. Extraction of the crude product with H_2O/EtOAc and purification by silica gel column chromatography (EtOAc/hexane 50:50) gave 3.62 g (96%) of 4-(((carbobenzyloxy)amino))phenacyl dibenzyl phosphate (ethylene ketal) (**4f**): ^{1}H NMR (acetone-d_6) δ 2.89 (m, 2H), 4.10 (m, 2H), 4.12 (d, J = 6.8 Hz, 2H), 4.99 (d, J = 7.8 Hz, 4H), 5.18 (s, 2H), 7.34−7.39 (m, 15H), 7.44 (d, J = 8.7 Hz, 2H), 7.59 (d, J = 8.7 Hz, 2H), 8.85 (s, 1H); ^{13}C NMR (acetone-d_6) 64.38, 72.28, 75.32 (d, J = 20.2 Hz), 75.79 (d, J = 22.6 Hz), 125.35 (d, J = 30.0 Hz), 134.79, 134.95, 135.01, 135.09, 135.28, 135.38, 136.04, 137.52, 137.79 (d, J = 16.2 Hz), 143.41, 152.15, 152.38, 160.38; ^{31}P NMR (acetone-d_6) δ −2.63; IR ($CHCl_3$) 3428, 3050, 2931, 2881, 1687, 1598, 1525, 1502, 1435, 1410, 1212, 1182 cm^{-1}.

4-Aminophenacyl Diammonium Phosphate (**6f**).[29] To 4-((carbobenzyloxy)amino) phenacyl dibenzyl phosphate (ethylene ketal) (**4f**, 3.62 g, 6.14 mmol) dissolved in 30 mL of MeOH was added 362 mg of 10% Pd/C. The solution was hydrogenated at 10 psi of H_2 for 25 min with stirring followed by the addition of 1% HCl (1 mL). After filtration, the filtrate was evaporated *in vacuo*. The resulting viscous liquid was loaded on 5 g of a DEAE Sephadex (A-50-120, Sigma) column pretreated with 10 M of ammonium acetate. The diammonium salt of was eluted at 100 M ammonium acetate. Excess ammonium acetate was lyophilized, and the resulting solid was recrystallized from H_2O/MeOH to give 1.55 g (95%) of 4-aminophenacyl diammonium phosphate (**6f**): mp 160 °C (dec); ^{1}H NMR (D_2O) δ 5.08 (d, J = 5.6 Hz, 2H), 6.92 (d, J = 8.8 Hz, 2H), 7.89 (d, J = 8.8 Hz, 2H); ^{13}C NMR (D_2O) δ 69.26 (d, J = 14.2 Hz), 117.16, 126.30, 133.25, 156.14, 199.80 (d, J = 31.6 Hz); ^{31}P NMR (D_2O) δ −1.71; IR (mineral oil) 3239, 3205, 3043, 2923, 1659, 1603, 1567, 1419, 1321, 1111 cm^{-1}; UV-vis (CH_3CN/H_2O) λ_{max} (ϵ) 230 (5960), 316 (19 100); FABMS (free acid) *m/z* (rel intensity) 110 (18), 185 (100), 202 (32), 232 (M + 1, 22); exact mass calcd for $C_8H_{11}NO_5P$ (free acid, M + H) 232.0375, found 232.0350.

4-Acetamidoacetophenone (**1g**). To a solution of 4-aminoacetophenone (**1e**, 10.0 g, 74.0 mmol) in 50 mL of dioxane/H_2O (50:50) was added acetic anhydride (6.3 g, 111.0 mg) over a 30 min period at 0 °C followed by 2 h of vigorous stirring at room temperature. The solvent was removed *in vacuo*, and the resulting solid was filtered and rinsed with dioxane. The crude product was dried and recrystallized from EtOAc or EtOAc/benzene to give 12.5 g (95%) of 4-acetamidoacetophenone (**1g**): mp 165.5−166.5 °C; ^{1}H NMR (DMSO-d_6) δ 2.06 (s, 3H), 2.52 (s, 3H), 7.71 (d, J = 8.8 Hz, 2H), 7.91 (d, J = 8.7 Hz, 2H), 10.29 (s, 1H); ^{13}C NMR (DMSO-d_6) δ 24.10, 26.30, 118.04, 129.38, 131.40, 143.56, 168.85, 196.35; IR ($CHCl_3$) 3431, 3048, 2939, 1695, 1684, 1599, 1515, 1403, 1362, 1312, 1270, 1178 cm^{-1}; FABMS *m/z* (rel intensity) 120.1 (11), 136.1 (27), 178.1 (M + 1, 100); exact mass calcd for $C_{10}H_{12}NO_2$ (M + H) 178.0868, found 178.0874.

4-Acetamidophenacyl Bromide (**2g**),[26] **4-Acetamidophenacyl Dibenzyl Phosphate** (**3g**).[27] **4-Acetamidophenacyl Dibenzyl Phosphate (Ethylene Ketal)** (**4g**),[28] and **4-Acetamidophenacyl Diammonium Phosphate** (**6g**).[29] These were prepared according to the same procedure used for **2f**, **3f**, **4f**, and **6f**, respectively.[37]

4-(Carbomethoxyamino)acetophenone (**1h**). To a solution of 4-aminoacetophenone (**1e**, 5.0 g, 37.0 mmol) and NaOH (1.5 g, 37.0 mmol) in 50 mL of dioxane/H_2O (50:50) was added dropwise methyl chloroformate (3.5 g, 37.0 mmol) for 30 min at 0 °C followed by 2 h of vigorous stirring at room temperature. After the reaction was complete, the solvent was removed *in vacuo*. The resulting solid was extracted with EtOAc/H_2O. The organic layer was concentrated and purified by silica gel column chromatography (EtOAc/CH_2Cl_2/hexane 30:50:20). Further purification was accomplished by recrystallization from CH_2Cl_2/hexane to give 7.1 g (98%) of white crystals of 4-(carbomethoxyamino)acetophenone (**1h**): mp 160−162 °C; ^{1}H NMR (DMSO-d_6) δ 2.51 (s, 3H), 3.70 (s, 3H), 7.59 (d, J = 8.7 Hz, 2H), 7.90 (d, J = 8.8 Hz, 2H), 10.07 (s, 1H); ^{13}C NMR (DMSO-d_6) δ 26.28, 51.83, 117.15, 129.49, 130.93, 143.63, 153.72, 196.33; IR ($CHCl_3$) 3428, 3050, 2948, 1739, 1675, 1603, 1525, 1408, 1359, 1314, 1272, 1179 cm^{-1}; FABMS *m/z* (rel intensity) 185.1 (100), 194.0 (M + 1, 45); exact mass calcd for $C_{10}H_{12}NO_3$ (M + H) 194.0817, found 194.0800.

4-(Carbomethoxyamino)phenacyl Bromide (**2h**),[26] **4-(Carbomethoxyamino)phenacyl Dibenzyl Phosphate** (**3h**),[27] **4-(Carbomethoxyamino)phenacyl Dibenzyl Phosphate (Ethylene Ketal)** (**4h**),[28] and **4-(Carbomethoxyamino)phenacyl Diammonium Phosphate** (**6h**).[29] These compounds were prepared according to the same procedure used for **2f**, **3f**, **4f**, and **6f**, respectively.[37]

2,4′-Dihydroxyacetophenone (**11**).[38] To the solution of 2-bromo-4′-hydroxyacetophenone (**2c**, 0.5 g, 2.3 mmol) and formic acid (0.13 g, 2.8 mmol) in 30 mL of benzene was added DBU (0.43 g, 2.8 mmol) at 0 °C. The reaction was monitored by TLC. After the reaction was complete, the solvent was removed *in vacuo*, and the residue was purified by silica gel column chromatography (90% CH_2Cl_2/10% EtOAc). The intermediate **11** was identified by ^{1}H NMR [(DMSO-d_6) δ 5.49 (s, 2H), 6.88 (d, J = 8.7 Hz, 2H), 7.86 (d, J = 8.7 Hz, 2H), 8.40 (s, 1H), 10.53 (s, 1H)]. To a solution of the intermediate **11** dissolved in 20 mL of methanol was added sodium hydroxide (0.2 g, 4.0 mmol) at room temperature. The solution was stirred for 1 h and neutralized by hydrochloric acid. After the solvent was removed *in vacuo*, the mixture was extracted with H_2O/EtOAc. The crude product was recrystallized from EtOAc/hexane to give 0.35 g (99%) of 2,4′-dihydroxyacetophenone (**11**): mp 165−167 °C; ^{1}H NMR (CD_3CN) δ 3.4 (br, 1H), 4.72 (s, 2H), 6.87 (d, J = 8.7 Hz, 2H), 7.81 (d, J = 8.7 Hz, 3H); ^{13}C NMR (CD_3CN) δ 64.55, 115.09, 129.92, 161.72, 196.81; IR (mineral oil) 3362, 3124, 2928, 1677, 1596, 1537, 1411, 1280, 1228, 1207, 1169, 1076 cm^{-1}; HRMS (EI, CH_3OH) 153* (M^+ + 1), 152 (M^+), 136 (M^+ − 16 [O]), 121 (M^+ − CH_3O), 93 (M^+ − C_2H_3O), 65.

General Procedure for Photolysis. All photochemical starting materials were synthesized and used as described below. Acetonitrile (HPLC grade) was used without further purification. All water was distilled and passed through a Nanopure deionizing system. Phosphate buffer for analytical HPLC was prepared using 85% phosphoric acid, tetrabutylammonium phosphate (TBAP, 0.5 mM), and potassium hydroxide to afford a solution of 0.04 M buffer at pH 6. The HPLC system employed consisted of two pumps, a controller, an auto injector fitted with a 10 μL loop, an UV-vis spectrophotometric detector set at 254 or 280 nm, a recorder and a C18 5 μm 250 mm × 4.6 mm column. All analytical HPLC analyses employed either a solvent gradient or an isocratic elution with a flow rate of 1.0−1.5 mL/min. Photolyses were performed in a Southern New England photoreactor fitted with a merry-go-round apparatus using 16 × 300 or 4 × 350 nm lamps. The light output for the determination of quantum yields was measured using the potassium ferrioxalate method.[39]

Samples for irradiation were placed in 20 × 180 mm Pyrex tubes and either 5 or 10 mL of an aqueous or alcoholic solution containing the phosphate ester was introduced into the tubes along with the appropriate internal standard. The concentration of the phosphate was adjusted to assure complete absorption of the incident radiation, *i.e.*, greater than 3 absorbance units at the excitation wavelength. The tube was sealed with a septum, deaerated with argon for at least 20 min at 0 °C, and photolyzed. Aliquots (100 μL) were removed periodically, stored in the dark, and analyzed by HPLC.[40]

Photolysis of Phenacyl Diethyl Phosphate (**3a**). Into three Pyrex tubes containing 10 mL of MeOH or *tert*-butyl alcohol were introduced 217 mg (0.86 mmol) of phenacyl diethyl phosphate (**3a**) and 0.2 g (1.88 mmol) of anisole as an internal standard. The solution was deaerated and irradiated at 300 nm for 3 h for MeOH and 48 h for *tert*-butyl alcohol solutions. Aliquots were taken at 30 min intervals and prepared for HPLC analysis. For both solvents, the photoproducts were isolated by silica gel column chromatography and identified as acetophenone and 1,4-diphenylbutane-1,4-dione (**9a**) [^{1}H NMR ($CDCl_3$) δ 3.48 (s, 4H), 7.56−8.05 (m, 10H); IR ($CHCl_3$) 3050, 1682, 1600, 1050 cm^{-1}; mass spectrum *m/e* (rel intensity) 239 (18.1, M + 1), 238 (78.7, M^+), 149 (20.1), 133 (38.7), 105 (100.0), 77 (53.5), 51 (22.0)], and acetophenone. Further characterization of the products was not attempted.

Photolysis of 3-Methoxyphenacyl Diethyl Phosphate (**3b**). Into three Pyrex tubes containing 10 mL of MeOH or *tert*-butyl alcohol were introduced 302 mg (0.90 mmol) of 3-methoxyphenacyl diethyl phosphate (**3b**) and 0.2 g (1.88 mmol) of anisole as an internal standard. The solution was deaerated and irradiated at 300 nm for 105 min for MeOH and 600 min for *tert*-butyl alcohol solutions. Aliquots were taken at 30 min intervals and prepared for HPLC analysis. For both solvents, the photoproducts were isolated by silica gel column chromatography and identified as 3-methoxyacetophenone, 1,4-bis-[3-methoxyphenyl]-butane-1,4-dione (**9b**) [^{1}H NMR ($CDCl_3$) δ 3.45 (s, 4H), 3.86 (s, 6H), 7.69−8.20 (m, 8H); IR ($CHCl_3$) 3050, 2897, 1684, 1600, 1029 cm^{-1}; mass spectrum *m/e* (rel intensity) 299 (28.9, M + 1), 298 (59.1, M^+), 163 (32.6), 135 (100.0), 107 (73.1), 92 (54.1), 77 (59.9)], and 2,3-bis-[4-methoxyphenyl]-butan-2,3-diol (**10b**) [^{1}H NMR ($CDCl_3$) δ 1.61 (s, 6H), 2.60 (s, 2H), 3.85 (s, 6H), 7.11−7.68 (m, 8H); IR ($CHCl_3$) 3300, 3050, 1606, 909 cm^{-1}]. Further characterization of products was not attempted.

Photolysis of 4-Hydroxyphenacyl Diethyl Phosphate (3c). Into five Pyrex tubes containing 10 mL of MeOH or *tert*-butyl alcohol were introduced 100 mg (0.46 mmol) of 4-hydroxyphenacyl diethyl phosphate (**3c**) and 0.2 g (1.88 mmol) of anisole as an internal standard. The solution was deaerated and irradiated at 300 nm for 30 min. Aliquots were taken at 5 min intervals and prepared for HPLC analysis. Methyl 4-hydroxyphenylacetate[2c] (**8c**, R = Me, in MeOH) [^{1}H NMR ($CDCl_3$) δ 3.56 (s, 2H), 3.70 (s, 3H), 6.74 (d, J = 8.5 Hz, 2H), 7.11 (d, J = 8.5 Hz, 2H); IR ($CHCl_3$) 3350, 3050, 1732, 1607, 1600, 1514, 1442, 1302, 1156 cm^{-1}; mass spectrum *m/e* (rel intensity) 167 (5.4, M + 1), 166 (8.8, M^+), 107 (22.8)] and *tert*-butyl 4-hydroxyphenylacetate (**8c**, R = *t*-Bu, in *t*-BuOH) [^{1}H NMR ($CDCl_3$) δ 1.43 (s, 9H), 3.45 (s, 2H), 6.75 (d, J = 8.4 Hz, 2H), 7.11 (d, J = 8.4, 2H), 7.26 (s, 1H); IR ($CHCl_3$) 3350, 3050, 2947, 1715, 1610, 1541, 1453, 1367, 1145, 958, 871, 837 cm^{-1}; mass spectrum *m/e* (rel intensity) 208 (81.3, M^+), 153 (13.2), 121 (18.0), 107 (99.3), 57 (100.0)] were isolated and identified as photoproducts by the injection on HPLC with authentic samples. Further characterization of products was not attempted. The quantum efficiencies for the disappearance of 4-hydroxyphenacyl diethyl phosphate (**3c**), Φ_{Dis}, the appearance of 4-hydroxyacetophenone (**1c**), Φ_{red}, and methyl 4-hydroxyphenylacetate (**8c**, R = CH_3), Φ_{rearr}, are shown in Table 1.

Photolysis of 4-Hydroxyphenacyl Diammonium Phosphate (**6c**). Into five Pyrex tubes containing 10 mL of 10% CH_3CN in buffer (pH 7.3) were introduced 100 mg (0.38 mmol) of 4-hydroxyphenacyl diammonium phosphate (**6c**) and 0.2 g (1.88 mmol) of anisole as an internal standard. The solution was deaerated and irradiated at 300 nm for 30 min. Aliquots were taken at 5 min intervals and prepared for HPLC analysis. The only photoproduct observed, 4-hydroxyphenylacetic acid (**8**), was identified by co-injection on HPLC with authentic sample. The quantum efficiencies for the disappearance of 4-hydroxyphenacyl diammonium phosphate (**6c**), Φ_{Dis}, and the appearance of 4-hydroxyphenylacetic acid (**8**), Φ_{rearr}, are shown in Table 1.

Photolysis of 4-Hydroxyphenacyl Adenosine 5′-Triphosphate, Triammonium Salt (**7**). Into three Pyrex tubes containing 5 mL of Tris buffer (0.05 M, pH 7.3) were introduced 20 mg (28.9 μmol) of 4-hydroxyphenacyl adenosine 5′-triphosphate, triammonium salt (**7**) and 15.3 mg (125 μmol) of benzoic acid as an internal standard. The solution was deaerated and irradiated at 300 nm for 12 min. Aliquots were taken at 3 min intervals and prepared for HPLC analysis. 4-Hydroxyphenylacetic acid (**8**) and ATP were identified as photoproducts by co-injection on HPLC with authentic samples. The quantum efficiencies for the disappearance of triammonium salt (**7**), Φ_{Dis}, and the appearance of 4-hydroxyphenylacetic acid (**8**), Φ_{rearr}, and ATP, Φ_{ATP}, are shown in Table 1.

Photolysis of 4-hydroxyphenacyl adenosine 5′-triphosphate, triammonium salt (**7**) in lactated Ringer's solution Into a Pyrex tube containing 5 mL of Ringer's solution (each 100 mL contains 5 g of dextrose hydrous, 600 mg of sodium chloride, 310 mg of sodium lactate, 30 mg of potassium chloride, and 20 mg of calcium chloride, pH 6.5) was introduced 20 mg (28.9 μmol) of 4-hydroxyphenacyl adenosine 5′-triphosphate, triammonium salt (**7**). The solution was deaerated and irradiated at 300 nm for 12 min. The resulting solution was prepared for HPLC analysis. 4-Hydroxyphenylacetic acid (**8c**, R = H) and ATP were identified as photoproducts by co-injection on HPLC with authentic samples. Quantitative measurements were made during the quenching studies given below.

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Competitive Quenching Study of 7 with Sodium 2-Naphthalenesulfonate. To a 10 mL volumetric flask were added 15 mg (21.7 μmol) of 4-hydroxyphenacyl adenosine 5′-triphosphate, triammonium salt (**7**), 61 mg (0.5 mmol) of benzoic acid, and Tris buffer (1 M, pH 7.3) to the fill line. To each of four Pyrex tubes was added 1 mL of the above solution. To three of these tubes was added 11.5 mg (50 μmol), 23.0 mg (100 μmol), or 34.5 mg (150 μmol) of sodium 2-naphthalene sulfonate. The tubes were deaerated for 20 min with argon at room temperature and photolyzed using 4 × RPR 350 nm lamps; the reactions were monitored by HPLC. A least-squares analysis was performed on the data obtained. The results are given in Table 2.

Photolysis of 4-Aminophenacyl Diammonium Phosphate (6f). Into each of four Pyrex tubes containing 10 mL of 10% CH_3CN/Tris buffer (50 mM, pH 7.3) was introduced 50 mg (0.19 mmol) of 4-aminophenacyl diammonium phosphate (**6f**) and 0.2 g (1.88 mmol) of anisole as an internal standard. One of the four phototubes was wrapped with aluminum foil and used as a thermal control for reference. The solutions were deaerated and irradiated at 300 nm for 30 min. Aliquots were taken at 5 min intervals. All aliquots were diluted for HPLC analysis. Only one product, 4-aminoacetophenone (**1e**), was identified by co-injection on HPLC with authentic sample. The quantum efficiencies for the disappearance of 4-aminophenacyl diammonium phosphate (**6f**) and the appearance 4-aminoacetophenone (**1e**) are shown in Table 3. The reference tube did not show any product.

Photolysis of 4-acetamidophenacyl diammonium phosphate (6g) and **4-carbomethoxyaminophenacyl diammonium phosphate** (**6h**) were performed out using the same procedures employed for **6f**.[37]

Stability Test of 4-Hydroxyphenacyl Adenosine 5′-Triphosphate, Triammonium Salt (**7**) **in Various Media.** Into each of six 10 mL of test tubes, respectively, containing 5 mL of H_2O, D_2O, Tris Buffer (50 mM, pH 7.3), Ringer's solution I (each 100 mL contained 600 mg of sodium chloride, 30 mg of potassium chloride, and 20 mg of calcium chloride, pH 6.5), Ringer's solution II (each 100 mL contained I and 310 mg of sodium lactate, pH 6.5), and Ringer's solution III (each 100 mL contained (II) and 5 g of dextrose hydrous, pH 6.5) was introduced 5 mg (7.2 μmol) of 4-hydroxyphenacyl adenosine 5′-triphosphate, triammonium salt (**7**). Solutions were maintained at ambient temperature for 24 h. Aliquots were removed periodically to determine the stability of **7** in the six different media by HPLC. In all cases, **7** was stable at least 24 h or longer.

pH Test of Three Phenacyl Diammonium Phosphates. Into each of three Pyrex tubes containing 5 mL of H_2O were introduced 10 mg (37.6 μmol) of 4-hydroxyphenacyl diammonium phosphate (**6c**), 10 mg (37.6 μmol) of 4-aminophenacyl diammonium phosphate (**6f**), and 11.5 mg (37.6 μmol) of 4-acetamidophenacyl diammonium phosphate (**6g**). The pH of each solution was determined to be 7.4, 6.8, and 4.7, respectively. The solutions were deaerated and irradiated at 300 nm for 10 min. The resulting photolysis solutions gave pH measurements of 6.7, 6.7, and 3.6, respectively (Table 4).

^{31}P Experiments of *p*-Substituted Phenacyl Diammonium Phosphates for the Release of Inorganic Phosphate (P_i). Each (2 mg) of three phosphates, 4-aminophenacyl diammonium phosphate (**6f**), 4-acetamidophenacyl diammonium phosphate (**6g**), and 4-(carbomethoxyamino)phenacyl diammonium phosphate (**6h**) was introduced to three NMR tubes, respectively. All of the tubes were filled with 1 mL of D_2O, and the solutions were deaerated and irradiated for 10 min. ^{31}P NMR analyses were conducted before and after the irradiation. Since the ^{31}P reference peak (measured by 85% phosphoric acid) was not stable, the analyzing peaks being evaluated often changed P_i chemical shifts slightly. However, all three samples showed that the phosphoric acid was released upon irradiation. All of the released P_i peaks were observed between 0.2 and 0.0 ppm in the NMR spectra.

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Supporting Information Available: Experimental and spectral details for **1f**−**h**, **2f**−**h**, **3a**−**c, f**−**h**, **4g**−**h**, and **6g**−**h** and the photochemical experimental details for **6g** and **h** (14 pages). See any current masthead page for ordering and Internet access instructions.

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