Nucleophilic and acid catalysis in phosphoramidite alcoholysis

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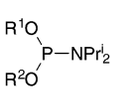
Numerous triflates, mesylates,† chlorides, trifluoroacetates and tetrazolides of trialkylammonium, pyridinium and azolium ions have been studied as activators for the reaction of *N*,*N*,*O*,*O*′-tetraisopropylphosphoramidite with propan-2-ol in acetonitrile. The progress of the reactions was followed by ^{31}P NMR spectroscopy, and the pK_a values of the activators were determined by a ^{13}C NMR spectroscopic method based on competing simultaneous protonation of two bases. The salts promoted the reaction both as acids and nucleophiles, the acidity playing a more important role than the nucleophilicity. The Brønsted α value for the general acid catalysis was observed to range from 0.6 to 0.9 and the β_{nucl} value to be 0.2 (pK_a of the conjugate acid used as the measure of nucleophilicity). Mixtures of neutral azoles or pyridines and weakly acidic ammonium salts were also shown to be useful activators that allow the acidity and nucleophilicity to be tuned independently of each other.

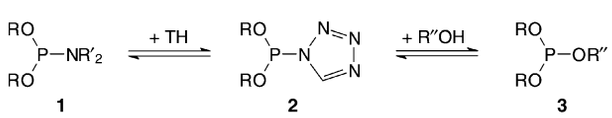
Introduction

Phosphoramidites (**1**) are phosphitylating agents utilised in automated solid-support DNA synthesis; a nucleoside 3′-phosphoramidite (**1b**) is repeatedly reacted with the 5′-hydroxy function of the growing support-bound oligonucleotide chain.[1,2] The reaction requires an acidic activator to promote the displacement of the amino group by the entering nucleophile. 1*H*-tetrazole[3] (TH) has become the most widely used activator in small-scale oligonucleotide synthesis. Although good coupling yields have been achieved with this activator, it also suffers from several disadvantages. Tetrazole is rather expensive, explosive when heated, hygroscopic and sparingly soluble in acetonitrile, the solvent commonly used in the coupling reaction. These problems have become more pronounced on attempting to scale-up the process for the synthesis of modified oligonucleotides necessitated by antisense drug research.[4] Consequently, many alternative catalysts have been screened to find an optimal activator for the phosphoramidite alcoholysis. These include azoles, such as 5-(4-nitrophenyl)-1*H*-tetrazole,[5] 5-ethylthio-1*H*-tetrazole[6] and 4,5-dicyanoimidazole,[7] azolium salts, such as benzimidazolium,[8] imidazolium[9] and *N*-methylimidazolium triflates,[10] and pyridinium salts, such as pyridinium hydrochloride,[11,12] hydrobromide,[12] sulfonates,[12] tetrafluoroborate[13] and trifluoroacetate.[14] Pyridinium salts have also been applied together with imidazole.[11,14] However, a systematic search for alternative catalysts has been hampered by insufficient understanding of the contributions of protolytic and nucleophilic processes to the activation.



1a: $R^1 = R^2 = Pr^i$
1b: R^1 = nucleosid-3′-yl, $R^2 = -CH_2CH_2CN$

Phosphoramidite alcoholysis is known to be catalysed by acids. No reaction takes place in the absence of acid.[15] For ammonium hydrohalide promoted reactions with *tert*-butyl alcohol and methanol, α values of 0.13 and 0.65 indicative of general acid catalysis have been determined.[16] In acetonitrile, the most frequently used solvent, not much is known of the effect of acid strength on catalytic efficiency. The dissociation constants for only a limited number of catalysts are known in acetonitrile, and the values referring to aqueous solution do not allow firm conclusions even at semiquantitative level. In addition to being an acid catalyst, the activator also participates as a nucleophile, although not necessarily before or during the rate-limiting step. In the absence of alcohol, tetrazole has been shown to react readily with phosphoramidites giving tetrazolylphosphonites (**2**) that then easily phosphitylate alcohols.[17,18]



1 **2** **3**

Intermediary accumulation of **2** has been observed during the tetrazole-promoted alcoholysis of phosphoramidites, and kinetic evidence for **2** lying on the reaction pathway has been obtained.[15,19] Accordingly, nucleophilic catalysis cannot be ignored as a factor that possibly affects the applicability of the activator. To elucidate this nucleophilic contribution in more detail we now report on studies with salts that potentially may participate as both acids and nucleophiles in the activation process. Many of these salts are actually viable alternative catalysts for the phosphoramidite alcoholysis, and a systematic study on their reactivity facilitates the search for an optimal catalyst. For this purpose, the reactions of a simple model compound, *O*,*O*′-diisopropyl *N*,*N*-diisopropylphosphoramidite (**1a**), with several ammonium, pyridinium and azolium salts have been studied both in the absence and presence of propan-2-ol in acetonitrile, and the observed catalytic efficiency has been compared to the nucleophilicity and acidity of the salt.

Results

Table 1 lists the dissociation constants determined in acetonitrile for the protolytes employed. Values not obtained from the literature were measured by a ^{13}C NMR spectroscopic method based on competing simultaneous protonation of two different bases (for details see ref. 19). It is worth noting that in some cases the relative acidities differ considerably from those

† The IUPAC name for triflate is trifluoromethanesulfonate and that for mesylate is methanesulfonate.

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Table 1 The dissociation constants (pK_{BH+} or pK_{HA}) in acetonitrile for the protolytes employed

Protolyte	pK_{BH+} or pK_{HA} [h]
Triethylammonium ion [a]	18.5
N,*N*-Diisopropylammonium ion	16.5
N-Methylmorpholinium ion [b]	15.6
Triallylammonium ion	15.2
N,*N*-Diisopropylanilinium ion	14.5
N,*N*-Dimethylanilinium ion	8.6
2,6-Lutidinium ion [c]	14.4
Pyridinium ion [c]	12.6
3-Chloropyridinium ion [c]	10.0
2-Chloropyridinium ion [c]	6.8
Imidazolium ion	17.1
N-Methylimidazolium ion	17.1
Benzimidazolium ion	14.3
N-Methylbenzimidazolium ion	14.1
1,2,4-Triazolium ion	10.0
Tetrazole	14.5
Trifluoroacetic acid [d]	12.7
Methanesulfonic acid [e]	8.4
HCl [f]	8.9
HBr [f]	5.5
Trifluoromethanesulfonic acid [g]	2.6

[a] J. F. Coetzee and G. R. Padmanabhan, *J. Am. Chem. Soc.*, 1965, **87**, 5005. [b] P. Beltrame, G. Gelli and A. Loi, *Gazz. Chim. Ital.*, 1980, **110**, 491. [c] D. Augustin-Nowacka and L. Chmurzyñski, *Anal. Chim. Acta*, 1999, **381**, 215. [d] T. Jasinski, A. A. El-Harakany, F. G. Halaka and H. Sadek, *Croat. Chem. Acta*, 1978, **51**, 1. [e] T. Fujinaga and I. Sakamoto, *J. Electroanal. Chem.*, 1977, **85**, 185. [f] I. M. Kolthoff, S. Bruckenstein and M. K. Chantooni Jr., *J. Am. Chem. Soc.*, 1961, **83**, 3927. [g] T. Fujinaga and I. Sakamoto, *J. Electroanal. Chem.*, 1977, **85**, 185. [h] All values measured by us have statistical errors less than ±0.1. *Notation*: Many of the values are included in the compendium: K. Izutsu, *Acid-base Dissociation Constants in Dipolar Aprotic Solvents*, Blackwell scientific, Oxford, 1990.

observed in aqueous medium. For instance, tetrazole is only 2 pK_{HA} units weaker an acid than trifluoroacetic acid, and the latter is barely capable of protonating pyridine. Methanesulfonic acid, in turn, is barely sufficiently strong to protonate *N*,*N*-dimethylaniline.

The kinetic runs were performed by mixing equal amounts of **1a** and the activator salt to obtain an initial concentration of 0.1 mol dm^{-3} for each, and monitoring the progress of the reaction by ^{31}P NMR spectroscopy. Similar measurements were carried out in excess (usually 10 equiv.) of propan-2-ol (Pr^iOH). In the absence of alcohol, displacement of the amino group by the anion or deprotonated cation of the salt usually took place. Ammonium trifluoroacetates, for example, gave trifuoroacetylphosphonite, and all azolium salts gave the corresponding azolylphosphonites. In an equimolar mixture of **1a** and the activator, complete displacement of the amino function is possible only if the entering nucleophile donates a proton upon nucleophilic attack (*cf*. reaction I in Scheme 1). Otherwise, the departing diisopropylamine abstracts the proton from the acidic activator, decreasing the concentration of the acid catalyst (II and III in Scheme 1). Hence, *N*-methylazolium salts expectedly resulted in only a 50% substitution, while the non-methylated azolium cations yielded a 100% conversion (see Scheme 1).

Regardless of the nucleophilicity of the activator salt anion or the deprotonated cation, an additional substitution product was repeatedly observed: a ^{31}P NMR signal at 131.0 ppm (detected before,[15] but then unexplained) refers to diisopropylphosphite anhydride, $(Pr^iO)_2P–O–P(OPr^i)_2$ (**13**).[20] This compound may be formed by a nucleophilic attack of diisopropyl phosphonate, $(Pr^iO)_2P(O)H$, on **1**, as depicted in Scheme 2. The phosphonate is, in turn, produced by the reaction of **1a** with traces of water present. Hence, both the nature of the activator and the moisture content of the solution affect the extent of formation of **13**. The anhydride also reacts with alcohol but not quite as fast as most of the other initial displacement products.

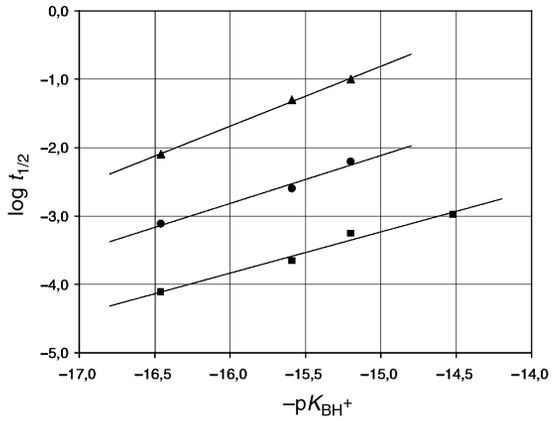


Fig. 1 The reaction of **1a** with Pr^iOH in the presence of various trialkylammonium triflates (■), mesylates (•) and trifluoroacetates (▲) in acetonitrile at 20 °C: Brønsted dependence of log $t_{1/2}$ on $-pK_{BH+}$ of ammonium ions.

The ^{31}P NMR spectroscopic data for the substitution products are given in Table 2. It is worth noting that the chemical shifts of azolylphosphonites, especially those of the positively charged *N*-methylazolylphosphonites, seem to depend on the identity of the salt anion, probably due to electrostatic ion–ion or ion–dipole interactions. The products of the initial nucleophilic displacement (**2**–**12**) were observed to be highly reactive, undergoing a rapid reaction upon addition of Pr^iOH, consistent with the fact that no sign of their intermediary accumulation could be detected in the presence of alcohol. The rate of alcoholysis of **1a** depended on the nature of the activator salt, as discussed below in more detail, but not on the concentration of the alcohol; no deceleration took place on using less than 10 equiv. of Pr^iOH. The results are summarized in Tables 3–5.

Ammonium salts as activators

Ammonium salts were the least efficient of the activators studied. The reactions were so slow that the differences in rate could be quantified by ^{31}P NMR spectroscopy. Accordingly, the key feature of the catalysis, *viz*. the possible involvement of both an acidic and a nucleophilic contribution to the catalysis, could best be examined with these activators (Table 3). On using ammonium triflates, mesylates or trifluoroacetates as activators, the catalytic efficiency was increased with the acidity of the ammonium ion, the Brønsted α value for the alcoholysis being 0.6 (triflates), 0.7 (mesylates) and 0.9 (trifluoroacetates) (all values ± 0.1), as illustrated in Fig. 1. If the cation was too weakly acidic, no reaction took place, as was the case with the triethylammonium salts.

Measurements in the absence of alcohol showed that ammonium triflates did not result in detectable accumulation of a covalent intermediate. The only reaction observed was slow formation of **13**. By contrast, the mesylates and trifluoroacetates of all ammonium ions gave a covalent displacement product as an intermediate. Among triallylammonium salts, the mesylate and bromide ions resulted in partial substitution, while chloride, trifluoroacetate and tetrazolide ions instantly exhibited the expected 50% displacement. Interestingly, the level of displacement by the anion also to some extent depended on the acidity of the cation. Triallylammonium trifluoroacetate resulted in a 43% displacement, while only 30% and 16% displacements were achieved with the corresponding *N*-methylmorpholinium and *N*,*N*-dimethylbenzylammonium salts, respectively. Even these partial substitutions reached their equilibrium almost instantly. The tetrazolides of all the cations studied caused a complete (50%) displacement.

Table 2 ^{31}P NMR spectroscopic data (δ given in ppm and J in Hz) of the substitution products

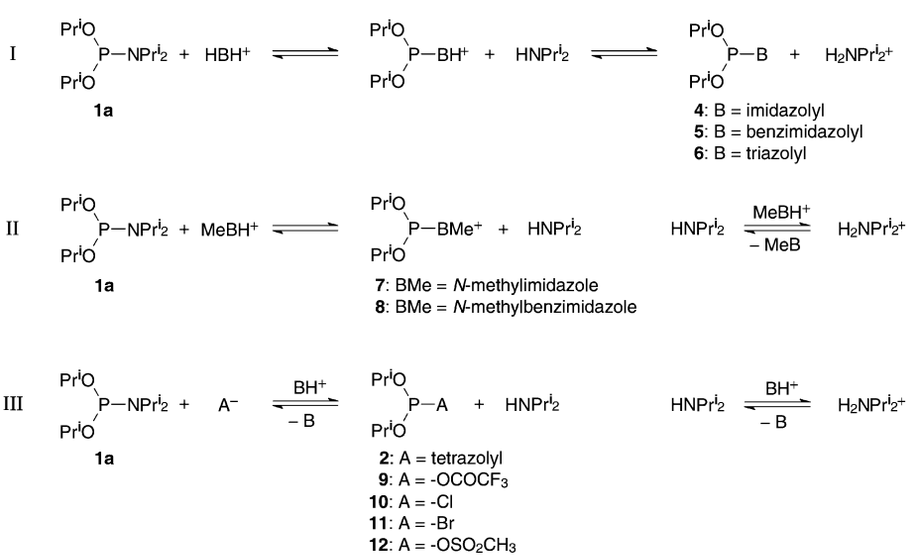
Compound	δ	Multiplicity	$^3J_{PH}$
Diisopropyl tetrazolylphosphonite (**2**)	128.1	t	8.6
Diisopropyl imidazolylphosphonite (**4**)	126.3/126.9	br s	
Diisopropyl benzimidazolylphosphonite (**5**)	128.6/128.5	br s	
Diisopropyl triazolylphosphonite (**6**)	129.7/124.6	br s	
Diisopropyl *N*-methylimidazolylphosphonite (**7**)	129.4/136.0	br s	
Diisopropyl *N*-methylbenzimidazolylphosphonite (**8**)	129.4/138.1	br s	
Diisopropyl trifluoroacetylphosphonite (**9**)	138.5	t	8.6
Diisopropyl phosphorochloridite (**10**)	168.4	t	9.5
Diisopropyl phosphorobromidite (**11**)	186.2	br s	
Diisopropyl methanesulfonylphosphonite (**12**)	137.1	t	9.1
Diisopropyl phosphite anhydride (**13**)	131.0	tt	6.3 5.4 [a]

[a] Value for $^5J_{PH}$.

Table 3 Half-lives for the reaction of **1a** (0.1 mol dm^{-3}) with propan-2-ol (1.0 mol dm^{-3}) in acetonitrile at 20 °C on using ammonium salts (0.1 mol dm^{-3}) as activators

Activator	Intermediate [a]	$t_{1/2}$/s [b]
N,N-Dimethylbenzylammonium triflate	—	13000
N-Methylmorpholinium triflate	—	4500
Triallylammonium triflate	—	1800
N,N-Diisopropylanilinium triflate	—	950
N,N-Dimethylanilinium triflate	100% (*P*-protonated **1a**)	20
N,N-Dimethylbenzylammonium mesylate	—	1300
N-Methylmorpholinium mesylate	—	400
Triallylammonium mesylate	12% (**12**)	120
Triallylammonium bromide	12% (**11**)	200
Triallylammonium chloride	50% (**10**)	20
Triethylammonium trifluoroacetate	—	∞ [c]
N,N-Dimethylbenzylammonium trifluoroacetate	16% (**9**)	125
N-Methylmorpholinium trifluoroacetate	30% (**9**)	20
Triallylammonium trifluoroacetate	43% (**9**)	10
Triethylammonium tetrazolide	—	∞ [c]
N,N-Dimethylbenzylammonium tetrazolide	50% (**2**)	30
N-Methylmorpholinium tetrazolide	50% (**2**)	15
Triallylammonium tetrazolide	50% (**2**)	15

[a] The level of substitution in absence of alcohol. The identity of the substitution product is given in parentheses. [b] Half-lives for the formation of **3**. The half-lives for the formation of the intermediate in the absence of alcohol were always less than 5 s (the lower limit for the detection). [c] No reaction detected.



Scheme 1 Displacement of the diisopropylamino group of **1a** with a protonated azole (reaction I), protonated *N*-methylazole (II) and an acid anion (III). The overall stoichiometry is determined by both the actual displacement and the subsequent protolytic equilibrium.

In the presence of alcohol, the catalytic efficiency of the tertiary ammonium salts depended on the nucleophilicity of the anion (see Fig. 1): the reaction rate was increased in the order triflates < mesylates < trifluoroacetates < tetrazolides. If the basicity of the anion is taken as a measure of its nucleophilicity, a β_{nucl} value of 0.2 ± 0.1 is obtained for the triallylammonium, *N*-methylmorpholinium and *N,N*-dimethylbenzylammonium salts.

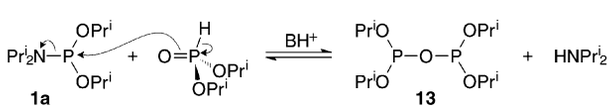
Table 4 Half-lives for the reaction of **1a** (0.1 mol dm^{-3}) with propan-2-ol (1.0 mol dm^{-3}) in acetonitrile at 20 °C on using pyridinium and azolium salts (0.1 mol dm^{-3}) as activators

Activator	Intermediate[b]	$t_{1/2}$/s[a]
2,6-Lutidinium triflate	—	100
2,6-Lutidinium mesylate	50% (**12**)	10
2,6-Lutidinium bromide	35% (**11**)	10
2,6-Lutidinium trifluoroacetate	50% (**9**)	<5
Pyridinium triflate	—	<5
3-Chloropyridinium triflate	—	<5
2-Chloropyridinium triflate	100% (*P*-protonated **1a**)	25
N-Methylbenzimidazolium triflate	50% (**8**)	<5
N-Methylimidazolium triflate	50% (**7**)	<5
1,2,4-Triazolium triflate	100% (**6**)	<5
Benzimidazolium triflate	100% (**5**)	<5
Imidazolium triflate	100% (**4**)	<5
N-Methylbenzylimidazolium trifluoroacetate	50% (**8**)	<5
N-Methylimidazolium trifluoroacetate	50% (**7**)	<5
Benzimidazolium trifluoroacetate	100% (**5**)	<5
Imidazolium trifluoroacetate	100% (**4**)	<5

[a,b] See the corresponding footnotes in Table 3. The half-lives for the formation of the intermediate in the absence of alcohol were less than 5 s, except for 2,6-lutidinium bromide, where a value of 50 s was observed.

Table 5 Half-lives for the reactions of **1a** (0.1 mol dm^{-3}) in the presence and absence of propan-2-ol (1.0 mol dm^{-3}) in acetonitrile at 20 °C on using mixtures of triethylammonium triflate ($TEAH^+$ triflate, 0.1 mol dm^{-3}) and pyridine or azole as activators

Activator	$t_{1/2}$/s
$TEAH^+$ triflate + imidazole	3000
$TEAH^+$ triflate + imidazole + propan-2-ol	3000
$TEAH^+$ triflate + benzimidazole	7200
$TEAH^+$ triflate + triazole	>18000
$TEAH^+$ triflate + *N*-methylimidazole + propan-2-ol	<60
$TEAH^+$ triflate + *N*-methylbenzimidazole + propan-2-ol	3600
$TEAH^+$ triflate + 2,6-lutidine + propan-2-ol	180
$TEAH^+$ triflate + pyridine + propan-2-ol	10800



Scheme 2 The formation of diisopropylphosphite anhydride (**13**).

Pyridinium and azolium salts as activators

The results obtained with the pyridinium and azolium salts are summarized in Table 4. Generally, pyridinium salts were more effective catalysts of the alcoholysis than the ammonium salts. Among them, 2,6-lutidinium salts were least reactive. Prolonged treatment of **1a** with lutidinium triflate resulted in formation of unidentified by-products exhibiting ^{13}C NMR signals at 164.5 ppm and 43.6 ppm (doublets with J = 303.5 Hz). The final product of alcoholysis, however, proved to be the expected phosphite **3**. With the strongly acidic 2-chloropyridinium ion, *P*-protonation took place in the absence of alcohol.

As with ammonium salts, all anions except the triflate ion served as nucleophiles in the absence of alcohol: mesylate, bromide and trifluoroacetate ions all resulted in displacement of the diisopropylamino ligand on using 2,6-lutidinium ion as an acid. No direct evidence of intermediates having a covalent phosphorus–pyridine was obtained. Two observations are, however, noteworthy. While 2,6-lutidinium and *N*,*N*-diisopropylanilinium cations are approximately as acidic, their mesylates behave in a rather different manner. Firstly, 2,6-lutidinium mesylate converted **1a** quantitatively to mesyl phosphonate, whereas only a 10% displacement took place on using *N*,*N*-diisopropylanilinium mesylate. Secondly, the alcoholysis was considerably faster in the presence of 2,6-lutidinium triflate than in the presence of *N*,*N*-diisopropylanilinium triflate.

Azolium salts proved to be most efficient among the activators studied in the present work: all alcoholyses and displacement reactions were instant. In the absence of alcohol, azolium salts always resulted in quantitative displacement, the azole, not the anion, becoming always bonded to the phosphorus atom. According to ^{31}P NMR spectroscopy, even the *N*-methylazoles gave covalent azole–phosphorus adducts at the expected 50% conversion level. While this manifests the superior nucleophilicity of the azoles, it is as well to bear in mind that among the azoles used in this work, only tetrazole is able to react with phosphoramidites when introduced in neutral form.

Cooperativity of activators

Since the high catalytic efficiency of azolium and pyridinium salts prevented the quantification of their relative reactivities, a series of experiments with the respective nucleophiles, neutral azoles and pyridines, were carried out in the presence of a very weak acid, triethylammonium ion. When triethylammonium triflate was used as an acid, reactions both in the presence and in the absence of alcohol could be monitored by ^{31}P NMR spectroscopy. The results are presented in Table 5.

The catalytic efficiency clearly correlates with the basicity of the nucleophile, and the tertiary *N*-methylated azoles are more reactive than their unsubstituted counterparts. The reactivity of *N*-methylimidazole is striking even when used together with such a weak acid as triethylammonium ion. It is worth noting that the reaction was enabled even by pyridines, though no covalent adducts between them and phosphorus could be detected. Finally, the reaction between **1a** and imidazole was followed in the presence of triethylammonium triflate using imidazole in excess. The presence of imidazole was indispensable for the alcoholysis to take place, but the use of imidazole in excess did not further accelerate the imidazolysis reaction. Within experimental error, use of up to 5 equiv. of imidazole had no effect on the reaction rate.

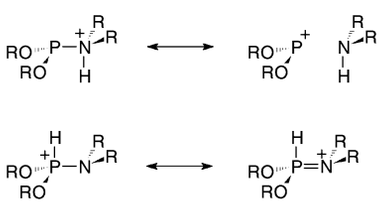
Discussion

Acid catalysis

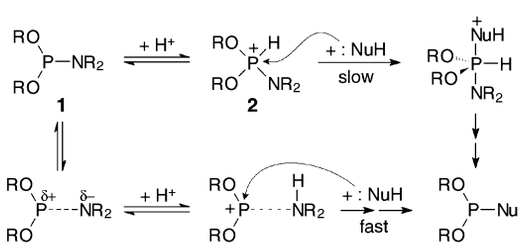
The alcoholysis of **1a** in MeCN is subject to general acid catalysis, proceeding only in the presence of a sufficiently strong proton donor. On using tertiary ammonium ions as an acid, an ion having a pK_a value less than 18.5 was required to obtain alcoholysis; no triethylammonium salt was able to catalyse the process, not even triethylammonium tetrazolide, in striking

contrast to tetrazolides of more acidic ammonium ions. The Brønsted α values observed for the general acid catalysis by ammonium, pyridinium and azolium ions range from 0.6 to 0.9, suggesting a rather advanced proton transfer in the transition state.

It has been shown previously[21] that phosphoramidites protonated on phosphorus are rather unreactive, and observable for instance by ^{31}P NMR spectroscopy. According to the present results, *P*-protonated phosphoramidites are formed only by acids having a pK_a value less than 10 (methanesulfonic acid, and *N*,*N*-dimethylanilinium and 2-chloropyridinium ions as triflates). Therefore, the acid catalysis by proton donors weaker than this in all likelihood involves protonation of the nitrogen atom, which cannot be observed directly. According to molecular modelling,[22] *N*-protonation lengthens and weakens the P–N bond and gives a highly unstable structure resembling electronically that of a phosphenium–amine complex. Protonation of phosphorus, in turn, strengthens and shortens the P–N bond.



As discussed previously,[21] the phosphorus atom is protonated by strong acids (pK_a < 10), since in the ground state of the molecule it is more basic than the nitrogen. The resulting phosphonium structure can then react with nucleophiles *via* an associative reaction path. Acids that are too weak for *P*-protonation may protonate the nitrogen atom of the exited state of **1**: on stretching the P–N bond the electron density on nitrogen increases until it becomes more basic than the phosphorus and basic enough even for acids with pK_a = 17. *N*-Protonation can take place only in the presence of a suitable nucleophile that in a more or less concerted manner traps the developing phosphorus cation resulting in substitution by a dissociative mechanism (Scheme 3).



Scheme 3 Discussed associative (higher) and dissociative (lower) reaction pathways triggered by strong and weak acids, respectively.

Although the *P*-protonation overwhelmingly predominates in the presence of strong acids, a minor amount of *N*-protonation may also take place, and hence the dissociative pathway may compete with the associative reaction of the predominant ionic form. The alcoholysis initiated by *P*-protonation with *N*,*N*-dimethylanilinium (pK_a = 8.6) and 2-chloropyridinium (pK_a = 6.8) ions is, for example, considerably faster than that triggered by triflic acid, which may well refer to competition between the associative and dissociative mechanisms. Although *N*,*N*-dimethylaniline and 2-chloropyridine present in solution after the protonation are very weak bases, they still are considerably more efficient proton acceptors than the triflate anion. On addition of a nucleophile, the bases can mediate the proton transfer from phosphorus to the nitrogen atom of the departing amine, thus facilitating decomposition by a dissociative pathway.

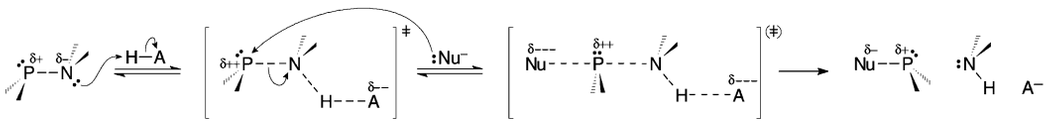
The acidity of the catalyst may also affect the level of substitution in the absence of alcohol, as in the case of ammonium trifluoroacetates. This could be attributed to the protolytic equilibrium between the acid catalyst and the amine replaced by the nucleophile (see Scheme 1). If the equilibrium constant of the substitution is close to unity, a strong acid that effectively traps the amine may drive the reaction to completion. A weak acid capable only of partial protonation results in an equilibrium depending also on the pK_a of the catalyst. Ammonium tetrazolides, in turn, constantly give complete replacement because the tetrazolylphosphonite is more stable, and due to the high equilibrium constant of the substitution reaction it is drawn to completion regardless of the position of the protolytic equilibrium.

Nucleophilic catalysis

In addition to acid catalysis, the alcoholysis of **1a** also seems to be susceptible to nucleophilic catalysis, as indicated by the following observations. 1) Most activators displace the diisopropylamino ligand from **1a** in the absence of alcohol. 2) These displacement reactions are usually quantitative and clearly faster than the alcoholysis in the presence of a poorly nucleophilic triflate salt of comparable acidity. 3) The products of these reactions react rapidly with alcohols. 4) The alcoholysis in the presence of a nucleophilic salt is considerably faster than that in the presence of a non-nucleophilic salt of equal acidity. For instance, the alcoholysis of **1a** in the presence of triallylammonium triflate exhibited a half-life of 1800 s while triallylammonium trifluoroacetate and tetrazolide gave the phosphite **3** instantly, as also does imidazolium triflate in spite of its lower acidity. Assistance by a good nucleophile also appears to enable alcoholysis under conditions that are not otherwise sufficiently acidic for the reaction: triethylammonium triflate alone does not catalyse the alcoholysis, but addition of imidazole promotes the reaction with alcohol, and in the absence of alcohol a covalent substitution product is formed.

Measurements with ammonium salts suggest that the nucleophilicity of the anions studied increases in the order $CF_3SO_3^-$ < $CH_3SO_3^-$ ≈ Br^- < Cl^- ≈ CF_3COO^- < tetrazolide ion. Whether triflate anion is capable of acting as a nucleophile towards **1a** at all is uncertain as no displacement products have been detected, in contrast to the other anions. Nevertheless, this possibility cannot be ruled out, since covalent and ionic triflate adducts with other trivalent phosphorus compounds have been detected by ^{31}P NMR.[23] In the absence of a better nucleophile, reaction proceeding *via* dialkylphosphite anhydride after nucleophilic assistance of hydrogen phosphonate traces must also be considered as an alternative. Interestingly, all neutral nitrogen bases seem to be better nucleophiles than the anions. With the weakly acidic triethylammonium ion as a proton donor, the tetrazolide anion is unable to promote the alcoholysis, whereas all neutral azoles and pyridines, when used together with triethylammonium triflate, enabled the reaction. Although pyridines have not been observed to yield displacement products with **1a**, they catalyse the alcoholysis in a way that would be best explained by a nucleophilic contribution. The high catalytic efficiency of pyridinium triflate compared to that of almost equally acidic *N*,*N*-diisopropylanilinium triflate is a good indication of this.

One might expect that since the reaction is subject to nucleophilic catalysis its rate would be dependent on the nucleophile concentration. In order to study this, one should vary the concentration of the nucleophile while keeping the acid concentration constant, which cannot be accomplished using salts alone. A possible approach is to use imidazole, one of the best nucleophiles for this work, together with a weak acid such as triethylammonium triflate. Use of one equiv. of imidazole (0.1 mol dm^{-3}) as a nucleophile resulted in a slow imidazolysis, but additions up to 5 equiv. (0.5 mol dm^{-3}) did not further



Scheme 4 Proposed reaction mechanism. Attack of the nucleophile takes place only after the proton transfer. Magnitude of the partial charges developing on nitrogen and phosphorus atoms depends on the strength of the acid and reactivity of the nucleophile, respectively. The rate limiting step is either the proton transfer alone or together with nucleophilic attack, depending on the identity of the nucleophile.

enhance the reaction. In fact, the nucleophilic substitutions of phosphoramidites have rarely been detected to depend on the nucleophile concentration.[24] Kinetic studies on alcoholysis have been performed in pseudo-first order conditions[16] and the transaminolysis of phosphoramidites has been reported to be independent of the amine concentration.[25] Reaction of phosphoramidites with tetrazole[19] is dependent on tetrazole concentration because of the acidic role of the reactant.

The finding that the reaction rate depends on the identity of the nucleophile suggests that the nucleophile is present in the transition state. The observed zero-order dependence of the imidazolysis rate on the nucleophile concentration is in at least apparent contradiction to this. One may presume that the nucleophile attacks the phosphorus atom as soon as the protonation-induced partial positive charge at this site is sufficiently high. The proton transfer leads to a stable intermediate only when the electron deficient phosphorus centre is trapped by the nucleophile, otherwise the amine and phosphenium ion rapidly reassociate to the starting material. Previously, a preassociation type mechanism was discussed,[21] but this would explain the observed kinetics only if the association equilibrium was quantitatively on the side of the associate and hence every protonation would lead to nucleophilic attack and displacement reaction. A better way to rationalise our observations is simply to think that at excess concentration of a very good nucleophile the trapping of the developing phosphorus cation is so fast that the *N*-protonation facilitated P–N bond cleavage alone becomes rate-limiting. Attack of a poor nucleophile involves an energy barrier comparable to that of the proton donation, resulting in an energy profile of two slightly separated peaks of almost similar height. The lower activation energy of a good nucleophile, in turn, is only a shoulder after the protonation peak. Hence, if the trapping is quantitative, *i.e.* if the departed amine is no longer able to compete for the phosphenium ion with the nucleophilic activator, the activator is not present in the transition state but participates after that (Scheme 4). Accordingly, at high concentrations of the nucleophilic activator the reaction rate becomes independent of the activator concentration.

Conclusion

Triflates, mesylates, chlorides, bromides, trifluoroacetates and tetrazolides of trialkylammonium, pyridinium and azolium ions serve as activators for the phosphoramidite alcoholysis promoting the reaction both as acids and nucleophiles. In all likelihood, the reaction proceeds by rate-limiting proton transfer to the departing amide ion, followed by trapping of the developing, only marginally stable phosphenium ion with a preassociated nucleophile. The Brønsted α value for the general acid catalysis falls in the range of 0.6 to 0.9, while the β_{nucl} value is only 0.2 (pK_a of the conjugate acid used as the measure of nucleophilicity).

Mixtures of neutral azoles or pyridines and weakly acidic ammonium salts may also be used as activators. Since in this case the proton donor (ammonium ion) and nucleophile (azole, pyridine) are independent of each other, both of them may be tuned in the desired manner to optimise the activation process. The advantage is that undesired cleavage of acid labile protecting groups may be avoided by using a weakly acidic activator of high nucleophilicity, such as imidazolium triflate, or a combination of an ammonium ion of similar acidity and *N*-methylimidazole.

Experimental

The synthesis of **1a** has been published previously.[15] The activator salts were generated *in situ* in an NMR tube by mixing the bases with tetrazole, trifluoroacetic acid, methanesulfonic acid and trifluoromethanesulfonic acid. Hydrochlorides and hydrobromides were prepared separately from desired bases and PCl_3 or PBr_3, respectively. Alcohols and amines were distilled and stored over molecular sieves (4 Å) and KOH pellets, respectively. Liquid acids were stored under airtight septa. Acetonitrile was dried with CaH_2 and stored over it.

In kinetic runs, **1a**, triflic acid, methanesulfonic acid and trifluoroacetic acid were introduced neat, other reagents were used as solutions that were dried with molecular sieves (4 Å). All reactions were performed in oven-dried, septum-sealed NMR tubes, into which the reagents were introduced by syringes. The ^{31}P NMR spectroscopic method for following the reactions[15] and the ^{13}C NMR spectroscopic method for determining the dissociation constants[19] have been published previously. The NMR spectra were recorded on a 500 MHz spectrometer magnet (202.35 MHz for ^{31}P, 125.65 MHz for ^{13}C).

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