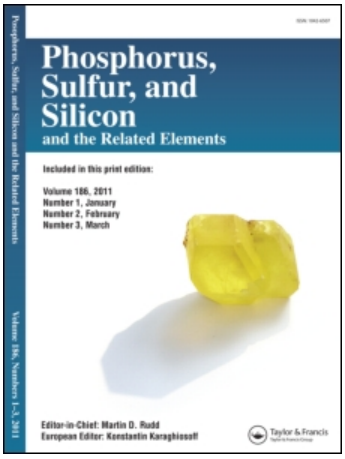


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# Synthesis and Reactivity of Five-Membered Cyclic Phosphorylating Reagents and Other Auxiliaries for the Synthesis of Oligonucleotides

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# SYNTHESIS AND REACTIVITY OF FIVE-MEMBERED CYCLIC PHOSPHORYLATING REAGENTS AND OTHER AUXILIARIES FOR THE SYNTHESIS OF OLIGONUCLEOTIDES.

IVAR UGI, NORBERT BACHMEIER, RUDOLPF HERRMANN, PETER JACOB, ROSMARIE KARL, MANUELA KLEIN, BERND LANDGRAF, PETER LEMMEN, WOLFGANG RICHTER and UWE VERFÜRTH
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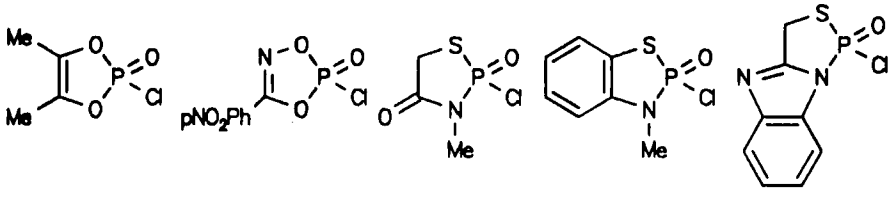
Abstract Reagents and preparative methods for the synthesis of oligonucleotides are the topic of this paper. The preparation of some highly reactive five-membered cyclic phosphorylating reagents, the 2-chloro-2-oxo-phospholes **3a-e**, is described, as well as their behaviour as phosphorylating reagents. In addition, the 1,1-dianisyl-2,2,2-trichloroethyl (DATE) protective group, and the oxidation of P(III)-compounds by oxaziridines, including the destructively stereoselective oxidation of stereoisomeric P(III) compounds are presented.



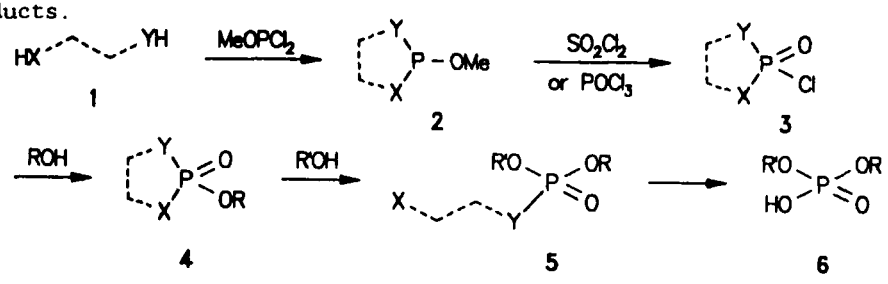
Although the P(III)-methods for the synthesis of oligonucleotides include an extra step, the oxidation of P(III) intermediates, the phosphite amidites are reagents of choice in the automated solid-phase syntheses of oligonucleotides[1], because the formation of phosphite esters from suitable P(III)-reagents proceeds generally much faster than phosphorylation by P(V)-reagents. With the exception of the highly reactive five-membered cyclic phosphorylating reagents[2], the P(V)-reagents react too sluggishly for the given purpose. In this article we wish to present some five-membered cyclic phosphorylating reagents, **3a-e**, that react fast with alcohols. One of these, **3c**, is endowed with all properties that a phosphorylating reagent for the synthesis of oligonucleotides must have.[3,4]




**3a (CEP–Cl)** **3b** **3c** **3d** **3e**

A direct cyclization according to **1** → **3** with $POCl_3$ would be the simplest conceivable way to synthesize **3a-e**. However, none of the compounds **3a-e** is thus formed, but only a mixture of unidentified compounds, presumably phosphorane derivatives and their decomposition

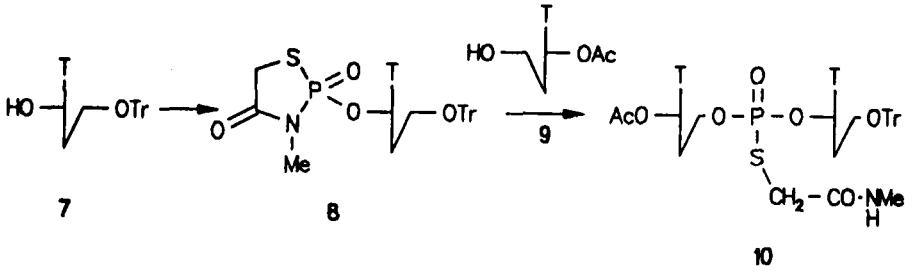
products.



The reagents **3** are best synthesized by the route **1** → **2** → **3**. The 2-methoxy-phospholes **2** are readily available from suitable bifunctional acyclic starting materials and $MeOPCl_2$. The conversion of the 2-methoxy-phospholes **2** into **3** is either achieved with $SO_2Cl_2$ (**3a**[5]: 27%, $^{31}$P-NMR ($CDCl_3$, $H_3PO_4$, Bruker HX-90 36.42 MHz): $\delta$ = 20.79 ppm; **3b**[6]: 81%, $\delta$ = 17.90 ppm; **3c**[3]: 88%, $\delta$ = 39.06 ppm), or with $PCl_5$ (**3d**[3,7]; 83%, $\delta$ = 50.34 ppm; **3e**[3]: ca.20%, chemically instable; $\delta$ = 31.62 ppm). It is noteworthy, that **3a** is readily available from the corresponding 2-chlorophosphole[8] by oxidation with $N_2O_4$ (56%)[9], while **3b - e** cannot be prepared by oxidation of the respective 2-chlorophospholes by the standard oxidizing reagents.[10]

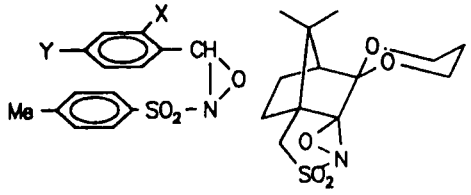
An ideal five-membered cyclic phosphorylating reagent **3** reacts rapidly with an alcohol ROH to yield **4**. The latter should not react any further with alcohols, unless deliberately activated. In the presence of a suitable catalyst, however, **4** must attack an alcohol R'OH fast to yield **5**.

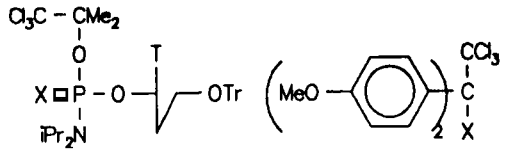
As we found in model experiments with EtOH and iPrOH, **3c** reacts precisely in the aforementioned way. With DMAP or N-methyl-imidazole as the catalyst, **5c** quickly is formed from **4c**. The selective cleavage of **5c** yields the phosphodiester **6** and N-methyl mercaptoacetamide **1c**[3]. As a first test, a synthesis of the thymidine dinucleotide derivative **10** (65% chromatographically purified product, one single $^{31}$P-NMR signal; $\delta$ = 28.06 ppm) confirmed the validity of the concept.



The cyclic chlorophosphates **3a**, **b**, **d** and **e** are less suitable for oligonucleotide syntheses. With **3a**, **b** and **e** both phosphorylating steps

**3** → **4** and **4** → **5** are fast, and can only be executed separately in solid-phase syntheses. While **3d** reacts fast according to **3d** → **4d**, **4d** is very sluggish to form **5d**, even in the presence of catalysts.[7]





**11**, a: X = Y = Cl; b: X = H; Y = CN

**12**

**13**, X = :

**14**, X = O

**15**, X = OH

**16**, X = $OSO_2CF_3$

The oxaziridines are generally capable of oxidizing P(III) to P(V) under anhydrous conditions[11]. The oxaziridines **11a** and **b** are well soluble in organic solvents, and react fast enough to be useful for the oxidation step of automated solid-phase syntheses of oligonucleotides by the phosphite amidite method.[1] Chiral oxaziridines[12], e.g. **12**, oxidize with destructive selectivity[13] one of the enantiomers of racemic phosphites, - kinetic resolution is a special case of destructive selectivity -, or one of the diastereoisomers in mixtures of diastereomeric phosphites[13], e.g. **13**.

The diastereomers **13a** ($\delta$ = 141.54 ppm) and **13b** ($\delta$ = 142.09 ppm) differ by their configuration at the P-atom. The oxidation of **13a** by **12** yields **14a** ($\delta$ = 2.72 ppm) and proceeds faster than the corresponding conversion of **13b** into **14b** ($\delta$ = 2.32 ppm). The stereoselectivity of this reaction is enhanced in the presence of suitably chosen Lewis acids. The strongly non-linear nature[13] of the destructively stereoselective oxidation of 0.01 m **13a** + **13b** (1 + 1) in ether at -60°C in the presence of 0.01 m $Ti(OiPr)_4$ is illustrated by the following observations:

When half of the required **12** is used to oxidize **13a** + **b**, a 47/53 mixture of **13a** and **13b** is obtained. With 80% of **12**, a concentration ratio **13a**/**13b** = 10/90 is observed at the end of the reaction, and with 95% of the **12** needed, only **13b** is left over; no **13a** can be detected ($^{31}$P-NMR) in the remaining **13**.

Our search for "orthogonal" protection[14] of hydroxyl groups for nucleotide syntheses led to the DATE (1,1-dianisyl-2,2,2-trichloroethyl) group[4] that survives the deprotection conditions of customary protecting groups, and can be removed by reductive fragmentation under conditions that do not affect other blocking groups.[14] Just like the Tr and DMTr groups, the DATE group can be introduced by alkylating with DATE

triflate[16] in pyridine. DATE triflate is obtained from **15** which, in turn, is formed from dianisyl ketone and trichloromethyl lithium[15] in ether/petroleum ether/THF at -110°C in 18% yield. Although many potential alternatives have been checked[16], no better synthesis for **15** has been found.

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REFERENCES

1. M.J. Gait, H.W.D. Matthes, M. Sing, B.S. Sproat and R.C. Titmus, in **Chemical and Enzymatic Synthesis of Gene Fragments**, edited by H.G. Gassen and A. Lang (Verlag Chemie, Weinheim, 1982), p.1.
2. F. Westheimer, Acc. Chem. Res. **11**, 70 (1968); D. Marquarding, F. Ramirez, I. Ugi and P.D. Gillespie, Angew. Chem. **85**, 99 (1973); Angew. Chem. Int. Ed. Engl. **12**, 91 (1973); F. Ramirez and I. Ugi, Phosphorus & Sulfur **1**, 231 (1976).
3. I. Ugi, J. Bauer, E. Fontain, J. Götz, G. Hering, P. Jacob, B. Landgraf, R. Karl, P. Lemmen, R. Schneiderwind-Stöcklein, R. Schwarz, P. Sluka, N. Balgobin, J. Chattopadhyaya, T. Pathak and X.-X. Zhou, Chem. Scripta **26**, 205 (1986).
4. I. Ugi, J. Achatz, M. Baumgartner-Rupnik, B. Danzer, C. Fleck, G. Glahsl, R. Herrmann, P. Jacob, C. Kambach, R. Karl, M. Klein, B. Landgraf, P. Lemmen, H. Martin, G. Neyer, R. Obrecht and B. Westinger, in Natural Products Chemistry III, edited by A. ur Rahman, P. Le Quesne (Springer Verlag, Heidelberg, 1988), p. 107-133.
5. see also: I.P. Gozman, Izv. Akad. Nauk. SSR **1968**, 2362; F. Ramirez, H. Okazoki and J.F. Marceck, Synthesis **1975**, 637; **1976**, 819.
6. see also: E. Fluck and M. Vargas, Z. Anorg. Allgem. Chem. **437**, 53 (1977); E.v. Hinrichs and I. Ugi, J. Chem. Research (S) **1978**, 338; J. Chem. Research (M) **1978**, 3973.
7. P. Jacob, Doctoral Thesis, Techn. Univ. München, 1989.
8. T.N. Kudriavtseva, N.B. Karlstedt, M.V. Proskurnina, N.V. Boganova, T.G. Shestakova and I.F. Lutsenko, Zh. Obshch. Khim. **52**, 1049 (1982).
9. I. Ugi and J. Götz, Heterocycles **21**, 265 (1984).
10. H.A. Kellner and I. Ugi, Z. Naturforschung **34b**, 1303 (1979).
11. I. Ugi, P. Jacob, B. Landgraf, C. Rupp, P. Lemmen and U. Verfürth, Nucleosides and Nucleotides **7**, 605 (1988).
12. G. Glahsl and R. Herrmann, J. Chem. Soc. Perkin I **1988**, 1753.
13. J. Brandt, C. Jochum, I. Ugi and P. Jochum, Tetrahedron **33**, 1353 (1977); I. Ugi, Chimia **40**, 340 (1986).
14. H. Eckert, W. Breuer, J. Geller I. Lagerlund, M. Listl, D. Marquarding, S. Stüber, I. Ugi, S. Zahr and H.v. Zychlinski, Pure & Appl. Chem. **51**, 1219 (1979); I. Ugi and R.G.K. Schneiderwind, Tetrahedron **30**, 2207 (1983).
15. G.Köbrich, K. Flory and R.H. Fischer, Chem. Ber. **99**, 1793 (1966)
16. R. Karl, Doctoral Thesis, Techn. Univ. München, 1989.