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Two Sulfur Containing Protecting Groups for Alcoholic Hydroxyl Function

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Abstract

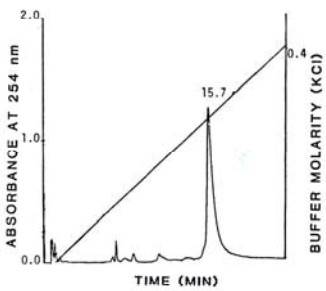
Two sulfur containing protecting groups for hydroxyl functions are described: the S-pixyl group, as in (**2**), is almost as labile as the pixyl group (as in (**1**)) whereas the chloro-S-pixyl group, as in (**3**), is much less labile to acidic hydrolysis. The application of these two new protecting groups is demonstrated by two independent synthesis of octathymidylic acids.

Acid labile protecting groups have played a very important role in developing the chemical synthesis of oligopeptides, oligosaccharides, oligoribo- and deoxyribo-nucleotides (RNA and DNA respectively) and in the synthesis of several other categories of natural products [1] on which the life process is completely dependent. The regioselective protection of the primary hydroxyl function, in presence of other secondary hydroxyl groups, by the triphenylmethyl (trityl) and its derivatives like 4-methoxy (MMTr) and 4,4′-dimthodxytrityl group (DMTr), especially the latter one, has been used as a key step in the synthesis of oligodeoxyribonucleotides (DNA) of defined sequences by both phosphodiester [2] and the triester approach [3]. Very recently, 9-phenylxanthen-9-yl (Pixyl) [4] (**1**) has been proposed as an alternative to the DMTr-group for the protection of the primary hydroxyl group of the pentose sugar moiety of appropriately *N*-protected 2′-deoxyribonucleoside blocks. Later on, the pixyl group has been successfully applied in the synthesis of DNA segments of defined sequences as a 5′-protecting group [5] or as a 3′-protecting group [6] in two different strategies. It has also been successfully employed as a 5′-protecting group in the ribo series for the synthesis of an effective protein synthesis inhibitor, pppA2′p5′A2′p5′A [7]. The main advantage of the pixyl group over the DMTr group was that the former gave crystalline derivatives and this property could be conveniently used as an additional purification step [4]. It is quite clear that the acid lability of a substituted trityl protecting group entirely depends upon the ability of the substituent on the phenyl ring(s) to delocalize the positive charge on the transition state [2]. Thus, the DMTr-group is more labile than MMTr-group which is more labile than the trityl group in acidic hydrolytic condition. Since, the lability of the pixyl group is comparable to the DMTr group, it was interesting to examine the effect of the change of heteroatom at the 10-position of the xanthene ring of the pixyl group from oxygen to sulfur so far as its ability to delocalize the positive charge on the transient carbonium ion was concerned. The lesser reactivities of furan and phenol, toward electrophilies, than the pyrrole and aniline respectively are classically explained [9] by the poorer ability of oxygen to accomodate a positive charge than nitrogen. Similarly the poorer reactivity of thiophen compared to furan, toward electrophiles, is explained [9] by the smaller positive electromeric effect of the sulfur lone pair than that of oxygen because of the lesser degree of overlap of *p*-orbitals of carbon and sulfur than in case of carbon and oxygen. We, therefore, rationalized that the replacement of oxygen at 10-position of the 9-phenylxanthene ring with nitrogen would make the latter more acid labile, which is undesirable so far as its application in a multistep oligodeoxyribonucleotide synthesis was concerned and replacement with sulfur would make it slightly less acid labile than the pixyl group.

Table I. *80% aqueous acetic acid hydrolysis of S-pixyl and chloro-S-pixyl groups at 20° C*

5′-*O*-S-pixyl derivatives		$t_{1/2}$ (min)	t_∞ (min)	5′-*O*-Cl-S-pixyl derivatives		$t_{1/2}$ (min)	t_∞ (min)
	% yields				*% yields*		
(**6**)	70.1	1	13	(**10**)	76.0	13	120
(**7**)	63.0	1	8	(**11**)	66.0	15	75
(**8**)	68.0	2	11	(**12**)	77.0	20	115
(**9**)	64.0	2	15	(**13**)	60.0	20	170

Here we describe the use of two new sulfur-containing protecting group, 5′-*O*-(9-phenylthioxanthen-9-yl)- (S-pixyl) and 5′-*O*-(7-chloro-9-phenylthioxanthen-9-yl)- and (chloro-S-pixyl), as in (**2**) and (**3**) respectively. The relative rates of acid hydrolysis of 5′-S-pixyl and 5′-chloro-S-pixyl-groups from the appropriately *N*-acyl protected deoxyribonucleoside derivatives [8] (Table I) make it clear that the S-pixyl group has a comparable acid stability to that of the pixyl group [4] while the chloro-S-pixyl group is considerably less acid labile. Thus, what seems to be apparent, in the 9-phenylxanthene and 9-phenylthioxanthene systems, (**1**) and (**6**)–(**9**) respectively, is that the sulfur and the oxygen, as heteratoms, have a comparable ability to delocalize the transient positive charge at the 9-position. This can presumably be attributed to a nearly identical *p*-orbital overlap of carbon and sulfur in S-pixyl, and carbon and oxygen in the pixyl group. The poorer acid labile properties of 7-chloro-9-phenylthioxanthen-9-yl protecting group, (as in (**10**) to



Elution profile: 1; S-pixyl has been used as the 5′-protecting group.

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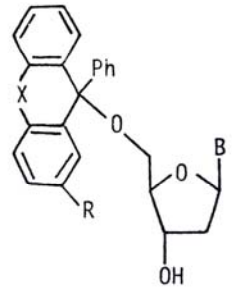
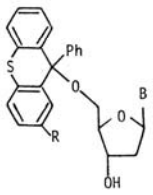


(1) X=O , R=H
(2) X=S , R=H
(3) X=S , R=Cl
(4) X=SO_2 , R=H
(5) X=SO_2 , R=Cl

(13)), can be explained by the inductive effect of chlorine which, as expected, destabilizes the carbonium ion. However, it is interesting to note that the deblocking of the S-pixyl and chloro-S-pixyl groups from (6)–(13) were complete within 90 seconds at 20° C with a 1.1 equivalent of 4-toluenesulfonic acid. H_2O in 2% ethanol-$CHCl_3$ solution and, thus, thymidine and other *N*-acyl protected deoxyribonucleosides were isolated in quantitative yields. It should be noted here that the corresponding sulfones (4) and (5) were completely resistant to acidic hydrolysis both in 80% aqueous acetic acid and in 4-tolunesulfonic acid. H_2O condition. Like the pixyl group, the S-pixyl and chloro-S-pixyl groups, as in (2) and (3) respectively, showed fluorescence under 366 nm UV light after the spots on the TLC plates were sprayed with 10% ethanolic H_2SO_4 and this way the S-pixyl and chloro-S-pixyl containing derivatives could be detected upto 2×10^{-9} *M* level.

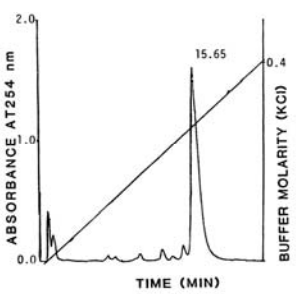
Finally, the application of the S-pixyl and chloro-S-pixyl



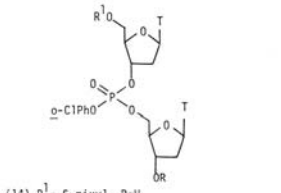
(6) B=T , R=H, (7) B=A , R=H , (8) B=C , R=H,
(9) B=G , R=H, (10)B=T , R=Cl, (11)B=A , R=Cl,
(12)B=C , R=Cl,(13)B=G , R=Cl,
T=Thymin-1-yl
A=6-N-m-clorobenzoyladenin-9-yl
C=4-N-benzoylcytosin-1-yl
G=2-N-t-butylbenzoylguanin-9-yl

groups were demonstrated by the independent synthesis of two octathymidylic acid fragments following a method adopted by us previously [5, 10].

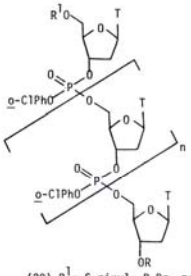
The dimer blocks (14) and (15) were obtained in 77.0 and 67.0% yields respectively using a "one pot" procedure described by us previously which we have subsequently used to prepare two sets of the fully protected dimer blocks (16), (17), (18) and (19) using a standard procedure [5, 10]. We noticed at this stage of the preparation of partially protected 5′-hydroxy block that the S-pixyl from (16) and (18) could be removed easily under a standard condition of the removal of the pixyl group (treatment of 4-toluenesulfonic acid H_2O (1.1 equiv.) in 2% EtOH–$CHCl_3$ for 90 s at 20°C) but the chloro-S-pixyl group from (17) and (19) required a prolonged treatment for 180 seconds for complete removal under the above condition. We then prepared the fully protected tetramer blocks (20) to



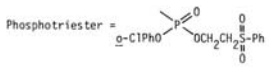
Elution profile: 2; Cl-S-pixyl has been used as the 5′-protecting group.



(14) R^1= S-pixyl, R=H
(15) R^1= chloro-S-pixyl, R=H
(16) R^1= S-pixyl, R= Phosphotriester
(17) R^1= chloro-S-pixyl, R=Phosphotriester
(18) R^1= S-pixyl, R= Bz
(19) R^1= chloro-S-pixyl, R=Bz



(20) R^1= S-pixyl, R=Bz, n=2
(21) R^1= S-pixyl, R=Phosphotriester, n=2
(22) R^1= chloro-S-pixyl, R=Bz, n=2
(23) R^1= chloro-S-pixyl, R=Phosphotriester, n=2
(24) R^1= S-pixyl, R=Bz, n=6
(25) R^1= chloro-S-pixyl, R=Bz, n=6

Phosphotriester = 

(23) by the block condensation of the 3′-phosphodiester salts of (14) and (15) with the respective 5′-hydroxy blocks again using a standard condition [5, 10]. The preparation of the 5′-hydroxy tetramer components by the acid hydrolysis of S-pixyl and chloro-S-pixyl groups again gave the same results in that the S-pixyl could be easily removed within 90 seconds under the normal condition while chloro-S-pixyl needed at least 180 seconds. After the preparation of the 5′-protected tetramer phosphodiester component, the two octamer blocks were assmebled, (24) and (25), following an usual condition in 73.1 and 81.0% yields (powders). They were deprotected in an usual fashion and chromatographed through HPLC under a standard condition [5, 10] (elution profile 1 and 2). The material under the main peaks were collected in 91.2 and 87% yield. An aliquot of each of these two preparations were ^{32}P-labelled and characterized [5, 10] unambiguously by their electrophoretic mobilities, partial digestion with snake venom phosphodiesterase (eight differently charge species on electrophoresis) and a complete digestion to establish the 3′ → 5′ linked phosphodiester linkages.

Acknowledgements

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References

1. *Protecting Groups in Organic Chemistry* (Edited by McOmie, J. F. W., Plenum, New York (1973).
2. Schaller, H., Weimann, G., Lerch, B. and Khorana, H. G., *J. Am. Chem. Soc.* **85**, 3821 (1963); for a recent review see Amarnath, V. and Broom, A. D., *Chem Rev.* **77**, 183 (1977).
3. Reese, C. B., *Tetrahedron* **34**, 3143 (1978).
4. Chattopadhyaya, J. B.. and Reese, C. B., *J. C. S. Chem. Comm.* 639 (1978).
5. Balgobin, N., Josephson, S. and Chattopadhyaya, J. B., *Acta Chem. Scand.* **B35**, 201 (1981).
6. Chattopadhyaya, J. B. and Reese, C. B., *Nucleic Acid Res.* **8**, 2039 (1980).
7. Chattopadhyaya, J. B., *Tetrahedron Lett.* **21**, 4113 (1980).
8. The S-pixyl- and Chloro-S-Pixyl-derivatives have been prepared basically following the procedure for the preparation of the pixyl derivatives (see [4]). All new monomeric compounds have been characterized by ^{1}H–NMR, I. R., mass spectrometry and element analysis. The S-pixyl and Chloro-S-Pixyl derivatives lacked the crystalline property of the pixyl derivatives.
9. March, J., *Advanced Organic Chemistry* McGraw Hill, New York (1977).
10. Balgobin, N., Josephson, S. and Chattopadhyaya, J. B., *Tetrahedron Lett.* **22**, 1915 (1981); Josephson, S. and Chattopadhyaya, J. B., *Chemica Scripta* **18**, 184 (1981).