# Methylase Models: Intramolecular Alkylation of a Phenol by an Adenosyl Sulphonium Salt

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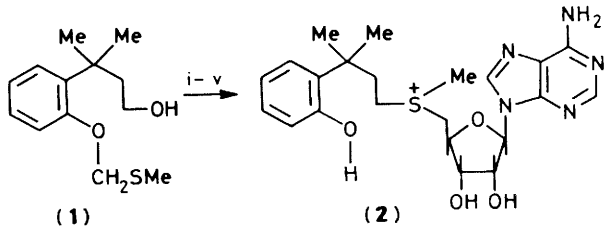
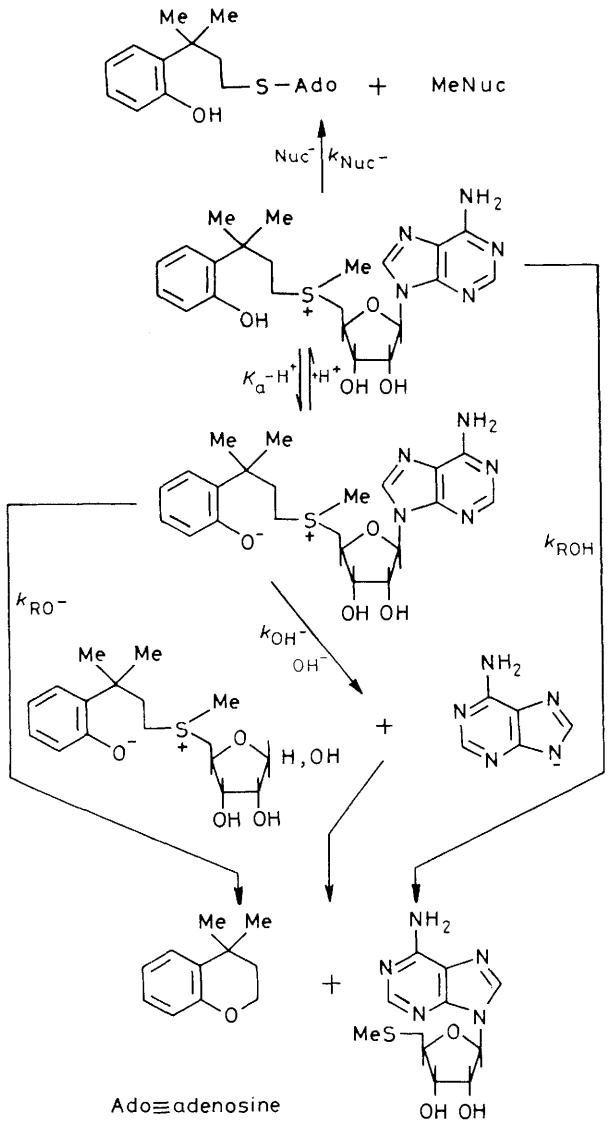
The newly synthesized adenosylsulphonium ion, (**2**), undergoes a facile ring closure reaction in aqueous oxyanion buffers; in contrast, amine buffers effect an intermolecular dealkylation in competition with the lyate reaction.

We have previously shown that the enzyme-catalysed methylation of pyro-catecholamines proceeds with inversion of configuration at the methyl carbon atom,[1] in agreement with a random, sequential kinetic mechanism.[2] In related model reactions, we have demonstrated that intramolecular alcohol[3] and phenol[4] alkylations by alkylmethyl-*p*-nitrophenylsulphonium salts are catalysed by general bases. We now report the extension of these model reaction studies to include alkylmethyl-5′-adenosylsulphonium salts [*e.g.* (**2**)]. In this case, the leaving group 5′-deoxy-5′-methylthioadenosine (MTA), is the same as for enzyme-catalysed aminopropyl transfer (*e.g.* spermidine synthase and spermine synthase).[5] It is also electronically similar to *S*-adenosylhomocysteine, the leaving group in enzyme-catalysed methyl transfer.[5] The present work represents the first example of a model reaction for enzyme-catalysed alkyl transfer, amenable to detailed kinetic studies, in which the leaving group is the biochemically relevant 5′-thioadenosine, MTA.

The desired substrate, (**2**), is obtained as shown in Scheme 1, starting with the *o*-substituted phenyl ether, (**1**).[4] The general synthetic method follows procedures recently developed in our laboratory for the synthesis of complex adenosine-5′-thioethers.[6,7] The reaction of (**2**) in aqueous buffers can be shown to procede in a manner similar to that previously described with *p*-nitrothioanisole as the leaving group.[3,4] Thus, (**2**) reacts at 40 °C over the range of pH 4—10, using acetate, phosphate, and carbonate buffers, to yield MTA and 4,4-dimethylchroman. At pH > 10, adenine is observed in addition to MTA, as a result of a hydroxide-catalysed fragmentation. This reaction of adenosylsulphonium compounds and hydroxide is well known.[8] Product analysis of the reaction solutions was carried out by h.p.l.c. using reverse-phase (Whatman ODS-2) chromatography; flow rate 1.0 ml $min^{-1}$. With 35% aqueous methanol as the mobile phase, adenine ($t_r$ 10.2 min) and MTA ($t_r$ 20.1 min) were easily quantitated, and 4,4-dimethylchroman ($t_r$ 28.2 min) was determined using 70% aqueous methanol as the mobile phase. The compound [*Me*-$^{14}$C]-(**2**) was synthesized, and its reaction studied in both oxyanion (*e.g.* carbonate, phosphate, and acetate) and amine




**Scheme 1.** *Reagents and conditions:* i, *p*-$MeC_6H_4SO_2Cl$, pyridine, 0 °C, 10 h; ii, KSAc, acetone, reflux, 4 h; iii, 5′-deoxy-5′-chloroadenosine, $Me_2SO$, NaOH, ambient temp., 4 h, under $N_2$; iv, MeI, $HCO_2H$; v, ion exchange on Dowex 1, $ClO_4^-$ form.




Scheme 2

(*e.g.* *N*-methylmorpholine) buffers. The reactions were maintained at 40 °C for a period of at least $10t_{\frac{1}{2}}$, and then analysed for products by h.p.l.c. The reactions carried out in oxyanion buffers gave rise to [*Me*-$^{14}$C]-MTA; no other radioactive product was observed. In contrast, the reactions carried out in amine buffers gave rise not only to [*Me*-$^{14}$C]-MTA, but also to a radioactive material eluting very early on reverse-phase h.p.l.c.; $t_r$ 3 min. These data are consistent with the formation of both MTA and an *N*,*N*-dimethylmorpholinium salt in the presence of the amine buffer. This result is in accord with our previous, more extensive studies on oxyanion-mediated general base catalysis *vs.* amine-mediated nucleophilic dealkylation at sp$^3$ carbon. A reaction scheme consistent with these results is shown in Scheme 2. Qualitatively, these data suggest that, in spite of a poorer dialkyl thioether leaving group, model sulphonium salts such as (**2**) react similarly to sulphonium salts in which *p*-nitrothioanisole is the leaving group.

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## References

1 R. W. Woodard, M-D. Tsai, H. G. Floss, P. A. Crooks, and J. K. Coward, *J. Biol. Chem.*, 1980, **255**, 9124.
2 J. K. Coward, E. P. Slisz, and F. Y-H. Wu, *Biochemistry*, 1973, **12**, 2291.
3 J. O. Knipe and J. K. Coward, *J. Am. Chem. Soc.*, 1979, **101**, 4339.
4 J. O. Knipe, P. J. Vasquez, and J. K. Coward, *J. Am. Chem. Soc.*, 1982, **104**, in the press.
5 J. K. Coward and P. A. Crooks, in 'Transmethylation,' eds. E. Usdin, R. T. Borchardt, and C. R. Creveling, Elsevier North-Holland, New York, 1979, p. 215.
6 G. L. Anderson, D. L. Bussolotti, and J. K. Coward, *J. Med. Chem.*, 1981, **24**, 1271.
7 K-C. Tang, R. Mariuzza, and J. K. Coward, *J. Med. Chem.*, 1981, **24**, 1277.
8 L. W. Parks and F. Schlenk, *J. Biol. Chem.*, 1958, **230**, 295; J. Baddiley, W. Frank, N. A. Hughes, and J. Wieczorkowski, *J. Chem. Soc.*, 1962, 1999; R. T. Borchardt, *J. Am. Chem. Soc.*, 1979, **101**, 458.