# A New Class of Amino Protecting Group Removable by Reductive Acidolysis: the 4-Methylsulphinylbenzyloxycarbonyl (Msz) Group†

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A safety-catch type of amino protecting group, the 4-methylsulphinylbenzyloxycarbonyl (Msz) group is stable under both acidic and basic conditions, but can be smoothly removed by a one-pot reaction involving reductive acidolysis using tetrachlorosilane–trifluoroacetic acid–scavengers; this new *N*-Msz group was successfully applied to the synthesis of a tachykinin peptide, scyliorhinin I, by a new orthogonal protection methodology.

Protection of the amino function is important in synthetic chemistry, especially in peptide synthesis.[1] We now report a safety-catch type of amino protecting group, the 4-methylsulphinylbenzyloxycarbonyl (Msz) group which is stable under both acidic and basic conditions but can be smoothly removed by a one-pot reaction involving reductive acidolysis using tetrachlorosilane ($SiCl_4$)–trifluoroacetic acid (TFA)–scavengers.

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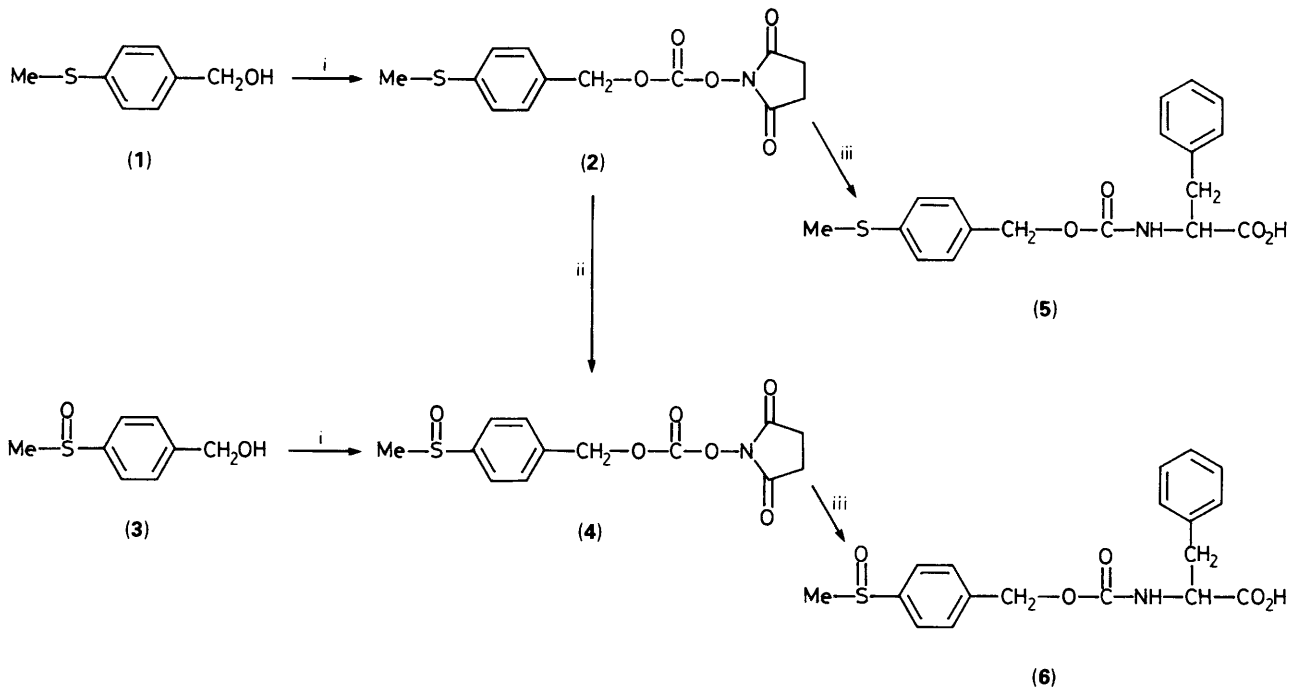
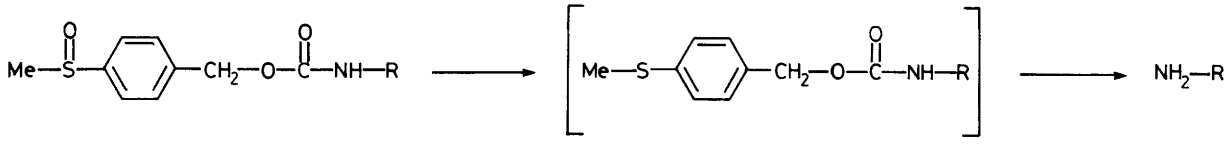
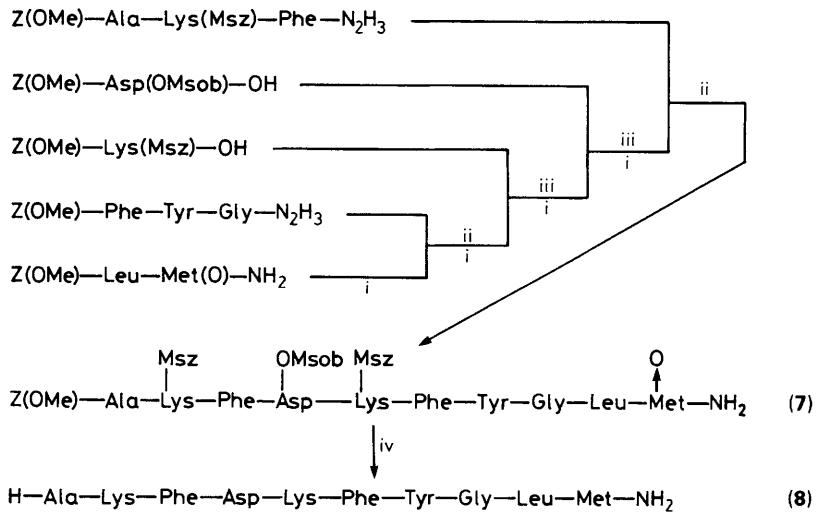
The Msz group-introducing reagent, 4-methylsulphinylbenzyl succinimidyl carbonate (Msz-OSu) (**4**) was prepared through two synthetic routes as shown in Scheme 1. The 4-methylthiobenzyl succinimidyl carbonate (Mtz-OSu) (**2**), m.p. 169—170 °C, was prepared in 95% yield from 4-methylthiobenzyl alcohol (**1**) and disuccinimidyl carbonate (DSC)[2] in MeCN in the presence of triethylamine at room temp. for 1 h. Oxidation of (**2**) by $NaBrO_2 \cdot 3H_2O$[3] in AcOEt–$H_2O$ gave Msz-OSu (**4**), m.p. 83—85 °C, in 62% yield. Alternatively, (**4**) was prepared in 40.3% yield from 4-methylsulphinylbenzyl alcohol (**3**) and DSC. Msz-Phe-OH (**6**), m.p. 139—142 °C, $[\alpha]_D^{20}$ −47.6° [*c* 0.6, dimethylformamide (DMF)], was prepared in 45% yield from (**4**) and L-phenylalanine in MeCN–$H_2O$ in the presence of triethylamine. Also, Mtz-Phe-OH (Mtz = 4-methylthiobenzyloxycarbonyl) (**5**), m.p. 87—89 °C,



**Scheme 1.** *Reagents*: i, DSC; ii, $NaBrO_2 \cdot 3H_2O$; iii, L-phenylalanine.



**Scheme 2.** Reductive acidolysis of the *N*-Msz group.



**Scheme 3.** Synthesis of scyliorhinin I by a new orthogonal protection methodology. *Reagents and conditions*: i, TFA–anisole; ii, azide method; iii, DCC–HOBt; iv, $SiCl_4$–TFA–anisole.

$[\alpha]_D^{20}$ −38.9° (*c* 0.6, DMF), was similarly prepared using (**2**).

Msz-Phe-OH (**6**) was stable to acidic conditions [TFA–anisole (25 °C, 24 h)] and basic conditions [1 M aq. NaOH (25 °C, 24 h), $NH_2NH_2$ in MeOH (25 °C, 24 h)], while it was quantitatively deblocked by $SiCl_4$ (10 equiv.)–TFA–anisole (10 equiv.) (0 °C, 60 min)[4—7] which had strong reductivity.[4,5] In this reaction, $SiCl_4$–TFA exhibited strong reductivity with any species of scavengers tested, such as anisole, phenols, sulphides, selenides and thiols, but it had no reductivity in the absence of scavengers.[4,5] Mtz-Phe-OH (**5**) was deprotected with TFA–anisole (0 °C, 60 min). In this deprotection reaction using $SiCl_4$–TFA–scavengers, the Msz group is first reduced to the TFA-labile Mtz group and then cleaved by acidolysis; *i.e.*, the Msz group is deprotected by the reduction of the sulphoxide moiety and subsequent acidolysis (reductive acidolysis)[4—6] (Scheme 2).

Using this acid-stable Msz group as 'semi-permanent' side-chain protecting group in combination with the acid-labile 'temporary' $N^{\alpha}$-protecting group [Boc = t-butoxycarbonyl or Z(OMe) = 4-methoxybenzyloxycarbonyl], we have developed a novel two-dimensional protection methodology in peptide synthesis. This method is based on a final deprotection strategy using 'reductive acidolysis'. The Msz group was introduced into the $N^{\varepsilon}$-function of lysine; *i.e.*, Z(OMe)-Lys(Msz)-OH, m.p. 84—86 °C, $[\alpha]_D^{21}$ −9.4° (*c* 0.4, DMF), was prepared from Lys·1/2$Cu^{2+}$ and Msz-OSu (**4**) (1.1 equiv.) followed by the reaction of Z(OMe)-$N_3$. Boc-Lys(Msz)-OH, $[\alpha]_D^{30}$ −6.7° (*c* 0.45, DMF), was prepared similarly. Lys(Msz) was stable to TFA–anisole (25 °C, 24 h), and it was readily deprotected by $SiCl_4$–TFA–anisole (0 °C, 60 min).

In order to demonstrate the usefulness of Lys(Msz), a tachykinin peptide, scyliorhinin I (**8**)[8] was synthesized by a solution method (Scheme 3). In combination with the TFA-labile Z(OMe) group for $N^{\alpha}$-protection, we used amino acid derivatives bearing TFA-stable protecting groups removable by reductive acidolysis, *i.e.*, Z(OMe)-Lys(Msz)-OH, Z(OMe)-Asp(OMsob)-OH[5,6] (Msob = 4-methylsulphinylbenzyl), and Z(OMe)-Met(O)-OH. It is known that the Msob ester[9] can be cleaved by $SiCl_4$–TFA–scavengers.[4—6] Also, Met(O) can be easily reduced by $SiCl_4$–TFA–scavengers.[4,5]

Using the dicyclohexylcarbodiimide (DCC),1-hydroxybenzotriazole (HOBt) and azide method, (**7**) was assembled, and prior to each coupling, TFA–anisole was used to remove the $N^{\alpha}$-Z(OMe) group selectively. The protected peptide (**7**) (100 mg) was deprotected with $SiCl_4$ (299 μl)–TFA (2 ml)–anisole (420 μl) at 0 °C for 1 h. The product was gel-filtered on Sephadex G-25 and purified by ion exchange chromatography on CM-Sepharose followed by preparative h.p.l.c. on TSK gel ODS-120T. The homogeneous peptide (**8**)‡ was obtained in 39% yield (from the deprotection and purification steps) and possessed biological activities and physicochemical properties identical with an authentic sample.[5,6]

These excellent results show the potential of the new orthogonal protection methodology for peptide synthesis.

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## References

1 R. Geiger and W. König, 'The Peptides,' eds. E. Gross and J. Meinhofer, Academic Press, New York, 1981, vol. 3, p. 1.
2 H. Ogura, T. Kobayashi, K. Shimizu, K. Kawabe, and K. Takeda, *Tetrahedron Letters*, 1979, 4745.
3 T. Kageyama, Y. Ueno, and M. Okawara, *Synthesis*, 1983, 815.
4 Y. Kiso, M. Yoshida, T. Fujisaki, T. Mimoto, T. Kimura, and M. Shimokura, 'Peptide Chemistry 1986,' ed. T. Miyazawa, Protein Res. Found., Osaka, Japan, 1987, p. 205.
5 Y. Kiso, M. Yoshida, T. Kimura, T. Mimoto, M. Shimokura, and T. Fujisaki, 'Peptides: Chemistry and Biology,' ed. G. R. Marshall, ESCOM Sci. Publishers B. V., Leiden, Netherlands, 1988, p. 229.
6 Y. Kiso, M. Shimokura, T. Kimura, T. Mimoto, M. Yoshida, and T. Fujisaki, 'Peptide Chemistry 1986,' ed. T. Miyazawa, Protein Res. Found., Osaka, Japan, 1987, p. 211.
7 Y. Kiso, T. Kimura, M. Shimokura, and T. Narukami, *J. Chem. Soc., Chem. Commun.*, 1988, 287.
8 J. M. Conlon, C. F. Deacon, L. O'Toole, and L. Thim, *FEBS Lett.*, 1986, **200**, 111.
9 J. M. Samanen and E. Brandeis, *J. Org. Chem.*, 1988, **53**, 561.

‡ $[\alpha]_D^{20}$ −30.5° (*c* 1, 1 M AcOH); t.l.c. (silica, $Bu^tOH$: AcOH: pyridine: $H_2O$ 4:1:1:2), $R_f$ 0.36; h.p.l.c. [Cosmosil $5C_{18}ST$, 4.6 × 150 mm, MeCN (10—60%, 30 min) in 0.1% aq. TFA, 0.7 ml/min], retention time 18.90 min; FD mass spectrum $(MH)^+$ 1218; satisfactory elemental analyses were obtained for $C_{59}H_{87}N_{13}O_{13}S·3CF_3CO_2H·5H_2O$. Amino acid ratios (6 M HCl hydrolysate): Asp 1.03, Gly 0.99, Ala 0.99, Met 0.70, Leu 1.00, Tyr 1.01, Phe 2.05, Lys 1.90; (aminopeptidase M hydrolysate): Asp 0.99, Gly 1.03, Ala 1.25, Met 1.12, Leu 1.00, Tyr 1.01, Phe 2.32, Lys 2.25; contractile activity of synthetic scyliorhinin I ($EC_{50}$ 1.9 × $10^{-8}$ M) was comparable to substance P ($EC_{50}$ 2.0 × $10^{-8}$ M) on the guinea pig ileum. [$EC_{50}$: the concentration required to cause 50% contraction of the maximum with carbachol (1.0 × $10^{-5}$ M).]