Peptide Synthesis with Benzo- and Naphthosultones

Francine Acher and Michel Wakselman*

C.E.R.C.O.A.-C.N.R.S., Thiais, France

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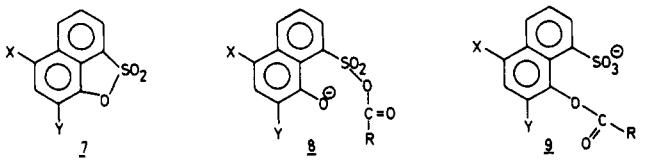
6-Nitro- and 6,8-dinitronaphth[1,8-*cd*]-1,2-oxathiole *S*,*S*-dioxides (**7b** and **7c**) have been prepared from the parent naphthosultone **7a** and compared with 5-nitrobenz[1,6-*d*]-3*H*-1,2-oxathiole *S*,*S*-dioxide (**1b**) as coupling reagents for peptide synthesis. Nucleophilic attack of a carboxylate salt on these strained five-membered sultones leads to activated esters **3** and **9** which rapidly react with amines (except in the case of **9c**). The rate constant for the formation of ester **9b** is higher than that of **3b**. Amides or peptides are formed in slightly better yields with the naphthosultone **7b** than with the benzosultone **1b**. The naphthosultones are also preferred over the benzosultones from the point of view of amount of 5(4*H*)-oxazolone formation from *N*-benzoyl amino acids and the degree of racemization. However the rate of alkaline hydrolysis of **7b** is slower than that of **1b**. All these results may be rationalized by a better intramolecular acyl transfer reaction in the more rigid mixed anhydride intermediate **8b**. There is no dependence on [amine]^2 in the rate equation for aminolysis of esters **3b** and **9b** by benzylamine in THF, acetonitrile, or DMF and the aminolysis is probably anchimerically assisted by a neighbouring S=O group. Esters **3b** are more selective acylating agents for primary amino groups in the presence of secondary ones than esters **9b** and this observation is exploited in a synthesis of maytenine by selective acylation of spermidine.

Introduction

Recently, we proposed 5-nitrobenz[1,6-*d*]-3*H*-1,2-oxathiole *S*,*S*-dioxide (**1b**) as a new stable reagent for coupling in peptide synthesis.[1] Nucleophilic attack by carboxylate ion on this strained sultone[2] leads to a mixed anhydride **2b** and then to an activated ester **3b** (Scheme I).

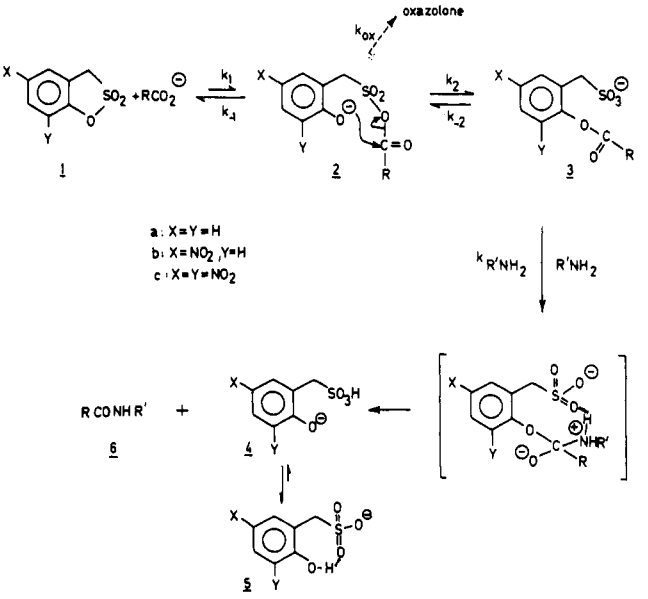
Aminolysis of **3b** is then a fast process probably due to the intramolecular base catalysis of a neighbouring S=O group. Other advantages of reagent **1b** are the water solubilities of ester **3b** and of byproduct **5b**. Consequently, acylation may be run in aqueous media and isolation of peptide **6** is easy. However, yields of ester **3b** and amide **6** are not quantitative. This may be due to an incomplete intramolecular seven-membered O → O acyl transfer reaction in the mixed anhydride intermediate **2b** in which the side chain can easily rotate out of the plane of the aromatic ring. Therefore, we looked for a sultone which could lead to a *more rigid mixed anhydride* with fewer degrees of rotational freedom.[3]

The preparation of 6-nitronaphth[1,8-*cd*]-1,2-oxathiole *S*,*S*-dioxide (**7b**) and 6,8-dinitronaphth[1,8-*cd*]-1,2-oxathiole *S*,*S*-dioxide (**7c**) and the use of **7b** as a coupling reagent in peptide synthesis are reported here. Comparisons of the reactivities of the oxy anions OH^- and RCO_2^- with sultones **1b** and **7b** and the aminolysis of esters **3b** and **9b** in different solvents have also been done. These kinetic studies and the racemization assays support the initial hypothesis.



7 **8** **9**

Scheme I



Results and Discussion

Synthesis. Generally, electrophilic substitutions[4] occur para to the oxygen atom of naphthosultone **7a**. The nitration of this sultone with 1 or 2 equiv of nitric acid in sulfuric acid gives mixtures of mono- and dinitro derivatives. In acetic anhydride at 55 °C, nitration of **7a** leads to a 52% yield of the 4-nitro derivative **7b**. The pure dinitro derivative **7c** being prepared in 58% yield from the mononitro **7b**.

The reaction of carboxylate salts with **7b** is analogous to that of sultone **1b** (Scheme I, **8** corresponding to **2**, **9** to **3**, **10** to **4**, and **11** to **5**). Aminolysis of activated esters **9b**, formed by intramolecular rearrangements of the intermediate mixed anhydrides **8b**, gave amides or peptides (Table I). The literature references for melting points and $[\alpha]_D$ of amides and peptides **6** are reported on Table I of the supplementary material. Yields obtained when using equimolecular amounts of carboxylate salt, amine, and coupling reagent **7b** have not been optimized but are

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(2) The reaction is probably of the $SN_2(S)$ type with a pentacoordinated transition state (Suttle, N. A.; Williams, A. *J. Chem. Soc., Perkin Trans. 2* **1983**, 1563).

(3) A representative value of ca 4.5 eu has been estimated for the entropy contribution of the internal rotation around a C–C single bond (Mandolini, L. *J. Am. Chem. Soc.* **1978**, *100*, 550. Graafland, T.; Wagenaar, A.; Kirby, A. J.; Engberts, J. B. F. N. *J. Am. Chem. Soc.* **1979**, *101*, 6981).

(4) (a) Schetty, G. *Helv. Chim. Acta* **1947**, *30*, 1650. (b) Mustafa, A. *Chem. Rev.* **1954**, *54*, 195.

0022-3263/84/1949-4133$01.50/0

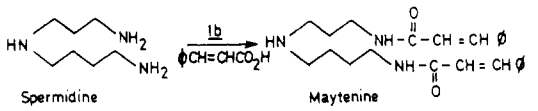
Table I. Synthesis of Di- and Tripeptides with Sultone 7b and Comparison with Sultone 1b[1]

RCO_2H	$R'NH_2$	solvent	time of first step, h	yield,[p] %	mp °C	$[\alpha]_D$ deg (concn, solvent, temp, °C)
CH_3CO_2H	$C_6H_5CH_2NH_2$	THF	8	83	60–61	
a		DMF	2	75	60–61	
ZGly	GlyOEt	THF	8	82	79–80	
a		DMF	1	78.5 (55)	79–80	
ZGly	$NHEt_2$	THF	8	80.5 (63)	oil[e]	
a		DMF	4	83	oil	
ZGly	ProOMe	THF	8	78 (76)	45	−82.2 (*c* 1, MeOH, 20)
a				53[o]	49–50	−84.0 (*c* 1, MeOH, 20)
ZGly	ValOMe	THF	15	82 (70.5)	oil[f]	−18.6 (*c* 1.5, MeOH, 28)
a						
ZGly	SerOMe	THF	15	70 (60)	96	+25.5 (*c* 1.02, $CHCl_3$, 27)
a						
ZGly (*a*)	TyrOMe	THF	15	72.5 (74.5)	oil[g]	+18.3 (*c* 1, MeOH, 20)
BocGly	GlyOEt	THF	15	76 (55.5)	oil	
a		CH_2Cl_2[d]				
N-Z-*S*-BzlCys	ValOMe	THF	15	63.2 (71.5)[j]	78	−32.1 (*c* 2, THF, 22)
a		DMF	3	71.5	78	−30.5
ZMet	GlyOEt	THF	15	85 (65)	93–94	−18.1 (*c* 4.5, EtOH, 27)
a						
ZVal	ProOMe	THF	15	80	oil	−90.6 (*c* 0.5, MeOH, 20)
a						
ZVal	ValOMe	THF	15	76 (68.5)	107–108	−29.0 (*c* 1, EtOH, 22)
a		DMF	2.5	84	110	−34.5
ZSer	GlyOEt	THF	15	60[h] (49)	104	−5.7 (*c* 1, EtOH, 20)
a						
BzLeu	GlyOEt	THF	15	72.0 (56.7)[i]	152–153	−32.6 (*c* 3.1, EtOH, 20)[k]
a		DMF	2.5	78.0 (66.0)[n]	151–152	−33.2[m]
ZGlyPhe	GlyOEt	THF	15	75[i] (77)	118–119	−12.5 (*c* 2, EtOH, 25)
a		DMF	2	75[i] (81)	118–119	−12.5
ZGlyPhe	*p*-$H_2NC_6H_4CH_2OH$	THF	15	77	167	
a						
BocGly	ValOBzl	THF	15	74.5	oil	−21.9 (*c* 1, MeOH, 20)
a		DMF/H_2O[d] (3/7)				
ZGly	PheONa	THF	15	40 (65)	124–125	+34.6 (*c* 2, EtOH, 26)
a		THF/H_2O[d] (3/7) pH 8.5–9.0				
ZGly	GlyOEt	THF	15	80 (53.2)	79	
b						
ZGly	GlyOEt	DMF	3	69 (65)	79	
c						

[a] Triethylammonium salt. [b] Tetrabutylammonium salt. [c] Dicyclohexylammonium salt. [d] Solvent for the second step. [e] Anal. Calcd for $C_{14}H_{20}O_3N_2$: C, 63.62; H, 7.63. Found C, 63.53; H, 7.56. [f] In our hands the mixed anhydride method[28] led also to an oil having the same R_f on TLC (R_f 0.49, SiO_2, AcOEt). [g] Anal. Calcd for $C_{20}H_{22}N_2O_6$: C, 62.12; H, 5.48. Found: C, 61.86; H, 5.64. [h] A small amount of ZΔAlaGlyOEt[29] was identified. [i] No D,L form was isolated in the Anderson's test. [j] $[\alpha]^{22}_D$ −32.1° (*c* 2, THF). [k] 95.8% L. [l] $[\alpha]^{20}_D$ −6.8 (20.0% L). [m] 97.5% L. [n] $[\alpha]^{20}_D$ −28.1° (82.7% L). [o] After recrystallization. [p] Yields in parentheses are using **1b**.

higher than with the earlier reported sultone **1b** (Table I). Coupling may be achieved with triethylammonium, tetrabutylammonium,[5] or dicyclohexylammonium salts and only needed 1 equiv of base per molecule of acid (and amine) used. Presumably the hydrogen-bonded *p*-nitrophenoxy grouping of **11b** (vide infra) is too weakly acidic in aprotic medium to protonate the amine. Generally, the activation with **7b** and the coupling to provide **6** were carried out in "one pot" in THF or DMF [Abreviations used are those recommended by the IUPAC-IUB Joint commission on Biochemical Nomenclature (*Eur. J. Biochem.* **1984**, *138*, 9). Other abreviations used are PNPA (*p*-nitrophenyl acetate), DMF (dimethylformamide), THF (tetrahydrofuran), AcOEt (ethyl acetate), Z (benzyloxycarbonyl), Bzl (benzyl), Bz (benzoyl), and DCC (dicyclohexylcarbodiimide).] without isolation of the activated ester **9b**. However ester **9b** could be isolated and aminolysis run in aqueous media. Sterically demanding dipeptide derivatives such as ZValValOMe were formed with good yields. A free hydroxyl function in the amino component did not require protection prior to the reaction. Acylation of secondary amines (Et_2NH, ProOMe) occurred

Scheme II



but was a much slower process than with primary amines (see kinetic studies) and was less selective than in the case of **1b**. Aminolysis of ester **9b** (RCO = ZGly) with an equimolar mixture of ethylamine and diethylamine gave a mixture of ZGlyNHEt (76.7%) and $ZGlyNEt_2$ (13.6%) in THF. Under the same conditions aminolysis of the analogous ester **3b** gave only ZGlyNHEt (60%). As an application of this good selectivity, maytenine[6] has been synthetized in 71% yield with **1b** (1 equiv) *trans*-cinnamic acid (1 equiv), and spermidine (0.5 equiv) (Scheme II). In this reaction a small amount (5%) of the triacylated de-

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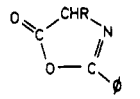
rivative of spermidine was easily separated. Reaction of **7b** with a mixture of *N*-benzoylglycine (hippuric acid) and triethylamine (1 equiv of each) in THF gave solely ester **9b** (80%), whereas **1b** under the same conditions gave a mixture of ester **3b** and 2-phenyl-5(4*H*)-oxazolone **12a**.[7]



12 a: R=H
b: R=i Bu

In the same way, activation of *N*-benzoyl-L-leucine with **1b** or **7b** in THF showed the formation of 2-phenyl-4-isobutyl-5(4*H*)-oxazolone (**12b**) only in the case of **1b**. Coupling of Bz-L-Leu with ethyl glycinate in THF gave the dipeptide BzLeuGlyOEt (Young's test[8]) containing 80.0% racemate when using **1b** and 4.2% when using **7b**. Activation and coupling in DMF gave 17.3% racemate when using **1b** and only 2.5% when using **7b** (Table I). In the Anderson's test (preparation of ZGlyPheGlyOEt;[9] which is about 10 times less sensitive than the Young's test[10,11a]) no racemate could be detected by partial crystallization when using **7b** in THF or DMF (Table I). Under controlled conditions (absence of chloride ion) aminolysis of *p*-nitrophenyl esters does not lead to racemization but considerable racemization may occur during the preparation of *p*-nitrophenyl peptide esters (86% D,L with DCC/*p*-nitrophenol in the Anderson test[12]).

Thus the degree of racemization is very weak compared to DCC and is lower with **7b** than with **1b**. The risk of racemization will decrease if the acyl transfer from highly activated species such as the mixed anhydride **2b** and **8b** to esters **3b** and **9b** is highly efficient and allowed no leakage to 5(4*H*)-oxazolone.[11a,11c] The solvent effect on racemization is much larger in the case of the benzosultone **1b** than in the case of naphthosultone **7b**. In DMF ion pairs are more dissociated than in THF. Therefore the phenolate anion is more nucleophilic, the rates of formation of the esters **3b** and **9b** will be higher, and the ratios of the rate constants k_2/k_{ox} (Scheme I) will increase.[11] However in the case of naphthosultone **7b** the acyl-transfer reaction is already very efficient in THF and the amount of racemate only slightly decreases from 4.2 to 2.5% (Table I).

Racemization may also occur through direct α-hydrogen abstraction as in the case of active esters of protected L-cystein.[13] When 1 equiv of Et_3N was used, *N*-Z-*S*-Bzl-L-Cys-L-ValOMe[13a] was obtained with a good optical purity (Table I).

After separation of peptide **6**, the byproduct **11b** may be extracted from the aqueous phase as a diethylanilinium salt[14] and recyclized with $POCl_3$ to give back reagent **7b** with a good yield. In the same manner, reagent **1b** may be recovered from **5b** but with a poorer yield.

The unsubstituted sultone **7a** only reacts with carboxylate ions in THF at reflux temperature whereas the dinitro derivative **7c** rapidly reacts at room temperature. However the rate of aminolysis of esters **9c** is slow due to the steric hindrance of the *o*-nitro substituent.

Kinetic Studies. (1) Alkaline Hydrolysis of Sultones 1b and 7b. Calculation of the second-order rate constants at various DMF concentrations from the pseudo-first-order rate constants measured at different hydroxide concentrations gave the values tabulated in Table II (available as suplementary material). For instance $k^{1b} = 4.4 \times 10^3$ and $k^{7b} = 1.2 \times 10^3$ M^{-1} s^{-1} at 20% DMF concentration and the ratio of the rate constants for the two sultones (k^{7b}/k^{1b}) is about 3.70.

For the analogous unsubstituted benzo- and naphthosultones **1a** and **7a** the ratio of the rate constants for alkaline hydrolysis is 1.5.[15] This difference of reactivity may be due to a variation of the electronic effect of a nitro substituent in the phenol and naphthol leaving groups in the transition state. In this particular case the direct comparison of the leaving groups' abilities with the p*K*s of the phenol leaving groups (p*K* **5b** 7.08[16], 6.8[17], 7.05; pK **11b** 7.20, 7.30 (see Experimental Section)) is probably misleading for the strength of the intramolecular hydrogen bond[18] in **5** and **11** (Scheme I) has an influence on the p*K* value and the real leaving groups are the phenolates **4** or **10**.

(2) Reaction of 1b and 6b with Carboxylate Anions. When, IR spectroscopy is used,[13a] it is possible to follow the kinetics of reaction of both sultones with *p*-toluate ion in THF. The ratio of the rate constants for the formation of esters is 0.67 ($k^{3b} = 0.64$, $k^{9b} = 0.95$ M^{-1} min^{-1}). A simplified calculation (available as a supplementary material) shows that the intramolecular acyl-transfer reaction should be more than five times faster in the case of the naphthosultone **7b** than in the case of its benzo analogue **1b**.

(3) Aminolysis of Triethylammonium Salts of Acetates 3b and 9b and of *p*-Nitrophenyl Acetate. Generally, aminolysis of aryl esters takes place according to a two-step process involving a tetrahedral intermediate ($IT^{\pm}$). In aprotic solvents the decomposition of the tetrahedral intermediate is rate determining.[19] The base catalyst for ammonium proton removal from $IT^{\pm}$ can be the solvent, a neighboring group, another molecule of amine, or an external catalyst.[20] A low degree of racemization is observed when ester aminolysis occurs by intramolecular anchimeric assistance.[11a]

For the *p*-nitrophenyl acetate our results are in good agreement with D. S. Kemp's observations with *p*-nitrophenyl esters of N-protected amino acids ($k_{R'NH2} = k'_2$[amine] + k'_3[amine]^2).[21] On plotting $k_{R'NH2}$ versus benzylamine concentration we observe a linear variation ($k'_3 = 0$) in THF and acetonitrile[22] for amine concentrations below 0.15 M but for higher concentrations the k'_3(amine)^2 term could not be neglected (Table III). Compared to

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Table III. Second-Order (k'_2) and Third-Order (k'_3) Rate Constants for the Aminolysis of Esters 3b, 9b (R = CH_3), and PNPA by Benzylamine and Dibutylamine

		benzylamine			di-*n*-butylamine		
ester	solvent	amine max concn, M	k'_2,[a] M^{-1} s^{-1}	k'_3,[a] M^{-2} s^{-1}	amine max. concn, M	$10^3 k'_2$,[a] M^{-1} s^{-1}	$k'_{2PhCH_2NH_2}/k'_{2Bu_2NH}$
PNPA	CH_3CN	0.44	0.0276	0.047			
3b		0.15	0.532				
9b		0.15	0.280				
PNPA	THF	0.44	0.084	0.088	0.540	3.0	28
3b		0.021	3.15		0.371	57.5	55
9b		0.091	0.630		0.371	37.3	17
PNPA	DMF	0.046	1.26				
3b		0.021	3.98				
9b		0.091	0.702				
PNPA	borate	0.030	0.490	33.0			
3b	buffer pH 8.85	0.030	0.430[b]	23.0			
9b	μ 0.49	0.15	0.113[b]	0.843			

[a] $k_{R'NH_2} = k'_2[\text{amine}] + k'_3[\text{amine}]^2$. [b] The first-order rate constants of hydrolysis of **3b** and **9b** in the same buffer are respectively 1.06×10^{-4} s^{-1} and 1.06×10^{-5} s^{-1} and the smallest recorded values for $k_{R'NH_2}$ 5.83×10^{-3} s^{-1} and 4.14×10^{-3} s^{-1} for **3b** and **9b**, therefore the hydrolysis of esters could be neglected compared to aminolysis.

those of PNPA, the rates of aminolysis of acetate **3b** and **9b** show an enhancement and no second-order dependance on amine concentration on the whole scale of concentrations $k'_3 = 0$; Table III). This behavior is expected for esters having a neighboring S=O group which assists the aminolysis by intramolecular general base catalysis and the probable mechanism is shown on Scheme I (formula in brackets). In DMF the intermolecular assistance by the solvent is very efficient, a linear variation is observed for any amine concentration investigated and the rate of aminolysis of PNPA is higher than that of **9b** but lower than that of **3b**. In the borate buffer (pH 8.85) no linear variation was noted for any of the three esters (Table III). The difference of rates observed between **3b** and **9b** in favor of **3b** in the four investigated solvents (Table III) presumably originates from steric hindrance[23] in the case of **9b**. The largest rate variation between **3b** and PNPA is noted in THF, assistance by the solvent is weak,[11b] and the effect of the neighboring sulfonate is very large. In the aqueous borate buffer the sequence of rates is reversed. In this protic and polar solvent, the sulfonate group is highly solvated and the intramolecular catalysis is suppressed. This solvatation increases the steric hindrance of the sulfonate group and the rates of aminolysis of **3b** and **9b** are lower than that of PNPA.

Aminolysis of esters **3b** or **9b** by the di-*n*-butylamine in THF at 25.0 °C shows no second-order term in amine ($k'_3 = 0$) on the range of the investigated concentrations and is also accelerated by an intramolecular assistance (Table III), yet the observed rates were much slower than with the benzylamine especially in the case of ester **3b**. As a result, esters **3b** show a very good selectivity for the acylation of primary amino groups in the presence of secondary amines. (This result has been exploited in the preparation of maytenine (vide supra).)

Conclusion

The comparison of the reactivities of the benzo- and naphthosultones (**1b** and **7b**) supports our initial hypothesis. In spite of a lower intrisic reactivity (the rate of alkaline hydrolysis is lower) the stable and easily prepared naphthosultone **7b** is a better coupling reagent for peptide synthesis than **1b** because of a more efficient intramolecular acyl-transfer reaction in the more rigid mixed anhydride **8b**. The rates of reaction with carboxylate anions and the yields of activated esters **9b** and peptides **6** are higher. The ratio of the rate constants for the formation of the activated ester and the competitive formation of the 5(4*H*)-oxazolone increase and the degree of racemization decreases (2.5% of racemate in the supersensitive Young's test in DMF, a very useful solvent for segment coupling in peptide synthesis). However aminolysis of **9b** is less selective than that of **3b**.

Experimental Section

General Methods. The rates of saponification of **1b** and **7b** as well as the rates of aminolysis of **3b**, **9b**, and PNPA were followed by means of a Cary 210 spectrophotometer equipped with a thermostated cell compartment maintained at 25.0 ± 0.2 °C. The rates of esterification of **1b** and **7b** were measured by following the increase of the absorbance at the ester absorption frequency with a Perkin-Elmer 167 spectrophotometer. NMR spectra were recorded at 90 MHz on a Brucker WH 90DS spectrometer. $[\alpha]_D$ measurements were carried out on a Perkin-Elmer 241 polarimeter and pH measures on a radiometer PHM 52 pH meter.

Materials. 5-Nitrobenz[1,6-*d*]-3*H*-1,2-oxathiole *S*,*S*-dioxide (1b) was prepared by the described method.[15a]

Naphth[1,8-*cd*]-1,2-oxathiole *S*,*S*-Dioxide. The commercial sultone **7a** (Fluka A.G.) was purified by filtration of a dichloromethane solution on a short silica column.

6-Nitronaphth[1,8-*cd*]-1,2-oxathiole *S*,*S*-Dioxide (7b). A solution of 5.3 g (25.7 mmoles) of sultone **7a** in 20 mL of acetic anhydride was stirred and heated to 55 °C. Then 3.2 mL of fuming nitric acid ($d = 1.52$) was added dropwise (about 1 drop per minute so as to keep the internal temperature between 55 and 70 °C). After stirring for a further 1 h at 55 °C the mixture was cooled to 40 °C and filtered. The collected solid was washed with dry hexane, recrystallized from acetic anhydride, washed again with hexane, and dried in a vacuum dessicator: yield 3.38 g (52.3%); mp 224–225 °C. Anal. Calcd for $C_{10}H_5NO_5S$: C, 47.81; H, 2.01; N, 5.58. Found: C, 47.64; H, 2.04; N, 5.37. MS, *m/e* (relative intensity) 251 (M^+, 100), 235 (4), 221 (17), 206 (29), 157 (40), 142 (29), 141 (26), 129 (29), 114 (54), 113 (55), 101 (32); ^1H NMR (CD_3COCD_3) δ 9.3 (dd, 1 H, H_5, $J_{5-7} = 0.6$, $J_{5-6} = 8.5$), 9.0 (d, 1 H, H_3, $J_{2-3} = 8.5$), 8.7 (dd, 1 H, H_7, $J_{5-7} = 0.6$, $J_{6-7} = 7.4$), 8.5 (dd, 1 H, H_6, $J_{5-6} = 8.5$, $J_{6-7} = 7.4$), 7.7 (d, 1 H, H_2, $J_{2-3} = 8.5$); IR (CH_2Cl_2) 1635, 1580, 1525, 1480, 1380, 1335, 1200, 1190, 1155 cm^{-1}.

6,8-Dinitronaphth[1,8-*cd*]-1,2-oxathiole *S*,*S*-Dioxide (7c). To a stirred solution of 0.502 g (2 mmol) of **7b** in 5 mL of concentrated sulfuric acid was added dropwise 1.60 mL of fuming nitric acid ($d = 1.52$, 6 mmol) at 10 °C. The internal temperature was raised to 50 °C for 90 min by means of an oil bath. After cooling to 0 °C small pieces of ice were added one by one (2 h) until the end of the precipitation. The precipitate was very rapidly filtered on sintered glass, rapidly washed with 2 mL of dry hexane, immediately recrystallized in the minimum amount of dry acetone,

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and collected by filtration: yield 0.343 g (58%); mp 244–245 °C. Anal. Calcd for $C_{10}H_4N_2O_7S$: C, 40.58; H, 1.39. Found: C, 40.34; H, 1.10. MS, m/e 296 (M^+, 100), 266 (14), 202 (19), 186 (12), 140 (12), 128 (29), 116 (14), 112 (34), 111 (26), 110 (11), 100 (41), 86 (63), 75 (30), 74 (42); ^1H NMR (CD_3COCD_3) δ 9.45 (s, 1 H, H_3), 9.40 (dd, 1 H, H_5, J_{5-7} = 0.6, J_{5-6} = 8.5), 9.00 (dd, 1 H, H_7, J_{5-7} = 0.6, J_{6-7} = 7.4), 8.70 (dd, 1 H, H_6, J_{6-7} = 7.4, J_{5-6} = 8.5); IR (nujol) 1685, 1540, 1460, 1395, 1375, 1330, 1195, 1185 cm^{-1}.

Triethylammonium Salt of Acetates (or Esters of *p*-Toluic Acid) 3b and 9b. Acetic acid (or *p*-toluic acid) (1 mmol) and 1 mmol of triethylamine in 2 mL of dry solvent (THF, acetonitrile) were added to 1 mmol of sultone **1b** or **7b** in 3 mL of solvent. The solution (0.2 M) was stirred overnight at room temperature and diluted to 50 or 100 mL to give 2×10^{-2} M and 10^{-2} M stock solutions.

***p*-Nitrophenyl Acetate (PNPA).** The commercial product was recrystallized from petroleum ether, mp 76–78 °C (lit.[20b] mp 78.4 °C).

Benzylamine, dibutylamine, and triethylamine were distilled from potassium hydroxide. Boiled distilled water was used for all aqueous solutions. "SDS puran", DMF, and acetonitrile were used. THF was distilled from sodium (with benzophenone as a colorimetric indicator of the absence of water) just before use.

The values of pK_w, water dissociation constant, and γ_{OH^-}, the activity coefficient of hydroxide ion equal to γ_{KCl}, the activity coefficient of potassium chloride in the solution, were taken from the literature.[25]

Coupling (Table I). General Procedure. To a stirred solution of 0.251 g (1 mmol) of sultone **7b** in 2 mL of dry DMF or a suspension in dry THF at room temperature was added a mixture of acid and triethylamine (1 mmol each) in 2 mL of solvent. After stirring for several hours a solution of 1 mmol of amine (or an equimolecular mixture of amine hydrochloride and triethylamine) in 1 mL of solvent was added. After 2 h, the organic solvent was removed in vacuo and 15 mL of a 7% aqueous solution of sodium bicarbonate was added to the residue. Extraction with 2 × 15 mL of ethyl acetate, washing with 4 × 5 mL of bicarbonate, and drying over sodium sulfate led to slightly yellow products which were purified on short silica (Merck 9385) columns (6 cm × 2 cm). First, dichloromethane eluted a small amount of sultone **7b** and then pure amide **6** was obtained with a mixture of diethyl ether and ethyl acetate (2:1) as eluent (Table I). TLC on silica gel Merck 5735 shows only one spot using ethyl acetate as eluent.

Maytenine. 1b (0.215 g, 1 mmol) and a mixture of *trans*-cinnamic acid (0.148 g) and triethylamine (0.101 g) (1 mmol of each) were allowed to react overnight in 4 mL of dry THF. Spermidine (0.073 g, 0.5 mol) in 1 mL of THF was then added and the solution stirred for 4 h. The crude product was extracted from a (1 M) sodium hydroxide solution (15 mL) with ethyl acetate (3 × 10 mL) (yield 0.162 g, 79%, mp 153–154 °C (lit.[6] 157 °C)) and purified on a short (6 cm × 2 cm) neutral alumina (Merck 1077) column.[6a] Upon elution with ethyl acetate (50 mL) and AcOEt–MeOH (95:5) (20 mL) the triacylated compound which was present in about 5% was eliminated. The pure maytenine was recovered with methanol (20 mL) (yield 0.144 g, 71.3%, mp 157 °C).

Recovering of the Coupling Reagent 7b. After extraction of the amide **6** the aqueous phases were concentrated to 5 mL and acidified to pH 1–2 with concentrated sulfuric acid. Then 0.149 g (1 mmol) of diethylaniline and 10 mL of dichloromethane were added. The aqueous layer was extracted with 10-mL portions of dichloromethane and the combined layers were dried over anhydrous sodium sulfate. The solvent was removed under reduced pressure. The oily residue crystallized and was precipitated with dry ether from dichloromethane: yield of the diethylanilinium salt of **11b** 0.376 g (90%); mp 151 °C. Anal. Calcd for $C_{20}H_{22}N_2O_6S$: C, 57.40; H, 5.30. Found: C, 57.45; H, 5.47. ^1H NMR ($CDCl_3$) δ 11.1 and 10.8 (2 s, 1.5 H, OH and NH), 8.85 (dd, 1 H, H_5, J_{5-7} = 1, J_{5-6} = 9), 8.5 (dd, 1 H, H_7, J_{5-7} = 1, J_{6-7} = 7), 8.3 (d, 1 H, H_3, J_{2-3} = 8.5), 7.7 (dd, 1 H, H_6, J_{6-7} = 7, J_{5-6} = 9), 7.1 (d, 1 H, H_2, J_{2-3} = 8.5), 7.5 (s, 5 H, NC_6H_5), 3.9 to 3.1 (broad m, 4 H, NCH_2CH_3), 1.1 (t, 6 H, NCH_2CH_3).

To 0.418 g (1 mmol) of this salt was added phosphorus oxychloride (5 mL) and the mixture slowly heated to 60–70 °C by means of an oil bath. Dissolution occurred and after 1 h the excess of $POCl_3$ was removed by distillation. The cooled residue was then recrystallized from acetic anhydride: yield 0.213 g (85%); mp 225 °C.

Hydrolysis of 1b and 7b. The alkaline hydrolysis of sultone **1b** and **7b** was followed by spectrophotometry. Because of the low solubilities of the sultones dimethylformamide had to be added as a cosolvent. Since the hydroxide ion concentration was kept constant by the use of a buffer, pseudo-first-order kinetic was observed in each particular case. Borate buffers of 0.49 M ionic strength were prepared following Bates.[26]

We considered that the hydroxide ion concentrations of the partial organic solutions were the same ones as those of the pure aqueous buffers (pH noted as $pH_{0\%DMF}$). Hydroxide ion concentrations were calculated from eq 1 and ranged from 0.30×10^{-5} M to 2.86×10^{-5} M.

$$\log OH^- = pH_{0\%DMF} - pK_w - \log \gamma_{OH^-}$$

$$\log OH^- = pH_{0\%DMF} - 13.81 \text{ at } 25\ ^\circ C \qquad (1)$$

The procedure for a typical kinetic run was as follows: A partial organic borate buffer (3 mL) was transferred to a quartz cell which was placed into the thermostated compartment of the spectrophotometer. A stock solution (15 μL 10^{-2} M) of sultone **1b** or **7b** in dry DMF (solution prepared just before use) was added. The stoppered cell was shaken vigorously, placed back into the thermostated compartment and allowed to equilibrate. Continuous plots of the absorbance increases at 412 nm for **1b** and at 468 nm for **7b** (the wavelengths at which the maximum change occurred) were recorded.

The pseudo-first-order rate constants at various hydroxide ion concentrations and various DMF proportions were calculated from eq 2.

$$\log (A_\infty - A_t) = k_1 t + \log (A_\infty - A_0) \qquad (2)$$

p*K* Determination. Potentiometry:[27] p*K*a **5b** 7.05, p*K*a **11b** 7.20. Spectrophotometry:[27] p*K*a **11b** 7.30.

Aminolysis of Triethylammonium Salts of Acetates 3b and 9b and PNPA. The rates of aminolysis of **3b**, **9b**, and PNPA by the benzylamine were determined in dry acetonitrile, THF, and DMF, and in a borate buffer of 0.49 M ionic strength and pH equal to 8.85, aminolysis by the dibutylamine was studied in THF. According to the relative rates, amine concentrations varied from 0.00305 M to 0.540 M. As a large excess was always used, pseudo-first-order kinetics were observed.

A typical run proceeded as follows: The solvent (3 mL) was placed in a cell maintained at 25.0 ± 0.2 °C; a stock solution (15 μL, 10^{-2} M) of esters **3b**, **9b**, or PNPA and then 1–300 μl (according to the relative rates) of pure amine were added. After the cell solution had been shaken and equilibrated in the thermostated compartment, a continuous plot of absorbance increase was recorded. The following recording wavelengths were chosen: 330 nm for **3b** and PNPA and 390 nm for **9b** in acetonitrile and THF, 330 nm for **3b**, 400 nm for **9b**, and 434 nm for PNPA in DMF,

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and 412 nm for **3b**, 468 nm for **9b**, and 402 nm for PNPA in the borate buffer.

The pseudo-first-order rate constants at various amine concentrations in the various solvents were calculated from eq 2.

Reaction of Sultones 1b and 7b with Carboxylate Ion. The rates of formation of esters **3b** and **9b** were determined from the increase of the ester IR band at 1750 cm^{-1} as a function of the time. Stock solutions (2×10^{-2} M) of the sultone and of the mixture of *p*-toluic acid (1 equiv) and triethylamine (100 equiv, large excess used to suppress the acid absorbance in the 1700–1800 cm^{-1} region) in dry THF were prepared just before use and mixed together. The absorbance was periodically recorded. The temperature could not be accuratly fixed and was about 30 ± 5 °C.

The recorded spectra at 1750 cm^{-1} show an increase which stabilizes before decreasing. In fact hydrolysis of esters **3b** and **9b** can only be neglected at the very beginning of the reaction (first 0.45 and 0.25 half-lives for the reactions with **1b** and **7b**). The first-order rate constant of the hydrolysis of a 10^{-2} M stock solution of these esters by contaminant water in analogous conditions was respectively 2.1×10^{-3} and 5.8×10^{-3} min^{-1}.

From a 2.10^{-2} M *p*-toluic acid ester **3b** or **9b** solution and a 2 M triethylamine solution, a 10^{-2} M ester solution with an excess of a 100 equiv of triethylamine was prepared and immediatly recorded. These spectra enabled us to estimate the end values (A_∞) of the ester formation reaction mentioned above. Optical densities at 1750 cm^{-1} were calculated from the measurements on the absorption spectra. Beer Lambert's coefficients were determined from the same (10^{-2} M) ester solution mentioned above.

The second-order rate constants were calculated from eq 3.

$$1/(A_\infty - A_t) = k_2 t + 1/A \quad (3)$$

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Registry No. **1b**, 14618-10-1; **3b**, 91606-06-3; **7a**, 83-31-8; **7b**, 91586-81-1; **7c**, 91586-82-2; **9b**, 91606-08-5; CH_3CO_2H·triethylammonium salt, 5204-74-0; ZGly·triethylammonium salt, 35264-84-7; BocGly·triethylammonium salt, 91586-83-3; *N*-Z-*S*-BzlCys·triethylammonium salt, 91586-84-4; ZMet·triethylammonium salt, 91586-85-5; ZVal·triethylammonium salt, 57022-41-0; ZSer·triethylammonium salt, 91586-86-6; BzLeu·triethylammonium salt, 91586-87-7; ZGlyPhe·triethylammonium salt, 91586-88-8; ZGly·tetrabutylammonium salt, 75523-03-4; ZGly·dicyclohexylammonium salt, 10073-22-0; $C_6H_5CH_2NH_2$, 100-46-9; GlyOEt, 459-73-4; $NHEt_2$, 109-89-7; ProOMe, 2577-48-2; ValOMe, 4070-48-8; SerOMe, 2788-84-3; TyrOMe, 1080-06-4; p-$H_2NC_6H_4CH_2OH$, 623-04-1; PheONa, 16480-57-2; $CH_3CONHCH_2C_6H_5$, 588-46-5; ZGlyGlyOEt, 3005-87-6; ZGlyNEt$_2$, 79990-06-0; ZGlyProOMe, 66449-90-9; ZGlyValOMe, 61058-41-1; ZGlySerOMe, 10239-27-7; ZGlyTyrOMe, 66012-26-8; BocGlyGlyOEt, 25438-03-3; *N*-Z-*S*-BzlCysValOMe, 24215-87-0; ZMetGlyOEt, 27482-82-2; ZValProOMe, 51827-14-6; ZValValOMe, 1999-88-8; ZSerGlyOEt, 4526-93-6; BzLeuGlyOEt, 2418-77-1; ZGlyPheGlyOEt, 2073-59-8; ZGlyPheNH-p-$C_6H_4CH_2OH$, 91586-89-9; BocGlyValOBzl, 66415-00-7; ZGlyPheONa, 5002-73-3; spermidine, 124-20-9; maytenine, 41590-65-2; di-*n*-butylamine, 111-92-2; *trans*-cinnamic acid, 140-10-3.

Supplementary Material Available: Table I, Table II, and calculation of the ratio of the rates of acyl transfer (3 pages). Ordering information is given on any current masthead page.

The First Skeletal Rearrangement of Aspidosperma to Melodinus Alkaloids. A Facile Conversion of (-)-Vincadifformine into *N*-Methyltetrahydromeloscine†

Giovanni Palmisano,* Bruno Danieli, Giordano Lesma, Renata Riva, and Sergio Riva

Istituto di Chimica Organica della Facoltà di Scienze, Università degli Studi di Milano, 20133 Milan, Italy

Francesco Demartin and Norberto Masciocchi

Laboratorio di Strutturistica della Facoltà di Scienze, Università degli Studi di Milano, 20133 Milan, Italy

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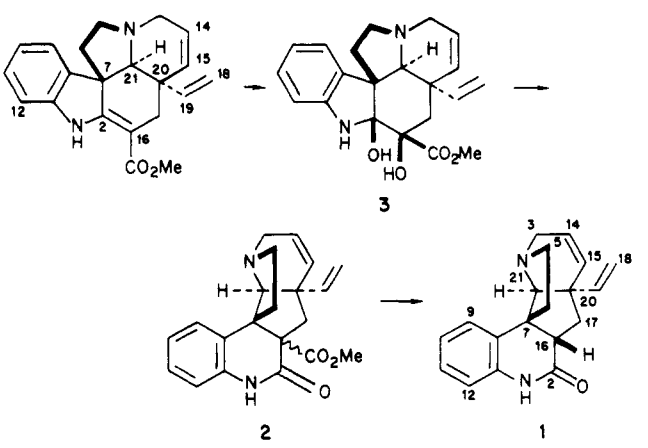
A facile approach to *N*-methyltetrahydromeloscine 18 starting from the readily available (-)-vincadifformine 11 was developed in a model study directed toward the synthesis of meloscine-type *Melodinus* alkaloids.

A characteristic of the genus *Melodinus*[1] of the Apocynaceae family is the presence of a rare type of indole alkaloids. In these, ring B has expanded to become six membered with a concomitant contraction of the ring C [abeo-7(2→16)aspidospermane],[2] meloscine 1[3] being the prototype of this group.

To date, only eight members of this family are known: meloscine, its 16-epi[3] and 16-epi *N*-oxide derivatives,[4] scandine 2,[3] meloscandonine,[5] 19-epimeloscandonine,[6] scandomeline,[6] and 19-episcandomeline.[6]

The mechanism of conversion of the *Aspidosperma* skeleton into the *Melodinus* system is under current study and there are several chemically reasonable possibilities.[3,6] The occurrence of *Melodinus* alkaloids together with 19-acetoxytabersonine in the *Melodinus* species indicated that the oxidation at C-16 followed by an α-ketol rearrangement could be the crucial steps in the biogenetic proposal (Scheme I). In an attempt to emulate this route for 1,

Scheme I



† We respectfully dedicate this work to the memory of Professor L. Canonica, untimely deceased in Aug 1984.

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