N-(2-Nitrobenzenesulphenyl)-saccharin: A Useful Reagent in Peptide Synthesis

S. Romani, G. Bovermann, L. Moroder, E. Wünsch*

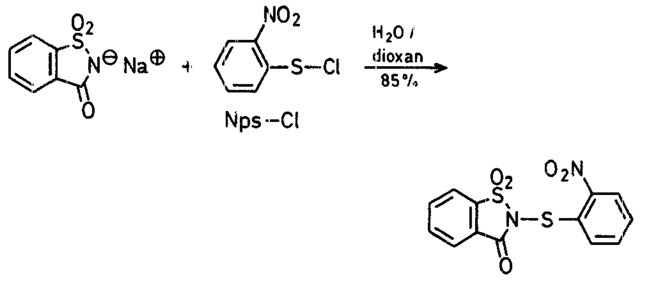
Max-Planck-Institut für Biochemie, Abteilung Peptidchemie, D-8033 Martinsried, Federal Republic of Germany

2-(2-Nitrobenzenesulphenyl)-1,2-benzoisothiazoline-3-one 1,1-dioxide (**1**; Nps-saccharin) is shown to be a useful new reagent for the synthesis of N-(2-nitrobenzenesulphenyl)-amino acids. The reagent **1** is readily prepared from 2-nitrobenzenesulphenyl chloride (Nps-Cl) and saccharin and is a crystalline compound which is stable on storage.

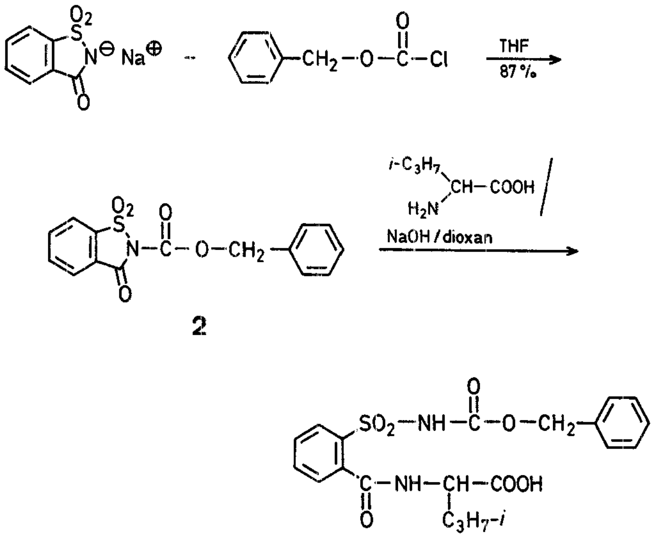
The 2-nitrobenzenesulphenyl (2-O_2N—C_6H_4—S; Nps) group[1] has found widespread application for the temporary protection of amino functions in peptide synthesis. The related amino acid derivatives have been obtained so far using 2-nitrobenzenesulphenyl chloride (Nps—Cl) as acylating agent. This reagent, however, is rather unstable; substantial amounts of the corresponding disulphide are formed on storage and recrystallisation prior to its use is often necessary.

The search for alternative Nps-donors led us to 2-(2-nitrobenzenesulphenyl)-1,2-benzoisothiazoline-2-one 1,1-dioxide (**1**; Nps-saccharin). This compound (**1**) is readily obtained in good yields as a crystalline material which is stable on storage over longer periods of time (Scheme A).

N-Acyl derivatives of saccharin have been reported to be acylating agents[2,3]. It is, however, also known that reactions of N-benzoylsaccharin with amines proceed via nucleophilic attack by the amines on the endocyclic carbonyl group of the saccharin moiety with concomitant ring opening[4]. Similarly, N-benzyloxycarbonylsaccharin (**2**), which was prepared by us as a potential benzyloxycarbonyl-donor, undergoes ring opening by amino acids under Schotten-Baumann conditions, as well as in organic solvents with tertiary amines as auxiliary bases, with quantitative formation of 2-(N-benzyloxycarbonylaminosulphonyl)-benzoylamino acids (Scheme **B**).

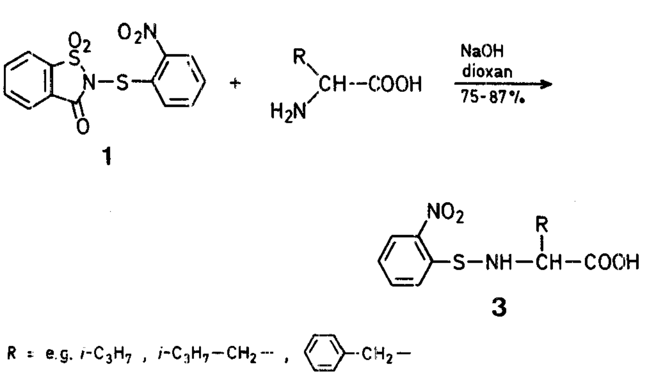


Scheme A



Scheme B

Conversely, Nps-saccharin (**1**) proved to be an efficient Nps-donor: (2-nitrobenzenesulphenyl)-amino acids **3** are obtained in yields and analytical purity comparable to those resulting from the known procedure[1] (Scheme **C**). Thus, this Nps-derivative may represent an useful new reagent in peptide chemistry, particularly because of its high stability on storage.



Scheme C

Melting points were determined on a Tottoli capillary melting point apparatus and are uncorrected. Optical rotations were measured in a jacketed 1 dm cell on a Perkin Elmer model 141 polarimeter. T. L. C. was performed on precoated silica gel 60 plates (Merck AG) using the solvent system: (a) cyclohexane/chloroform/acetic acid,

Table. (2-Nitrobenzenesulphenyl)-amino Acids **3** prepared using Reagent **1** (A) or Nps-Cl (B)

Amino Acid[a]	Yield [%]	m.p. [°C] A	B	$[\alpha]_D^{20}$ (c1, $CHCl_3$) A	B	$[\alpha]_{546}^{20}$ (c1, $CHCl_3$)
Nps-Val-OH · DCHA	87	193–193.5°	197–198°	−91.2°	− 92.6°	−106.7°
Nps-Phe-OH · DCHA	82	186.5°	186–187°	− 5.3°	− 4.0°	+ 14.3°
Nps-Leu-OH · DCHA	75	187.5–188°	186–187°	−70.3°	− 70.5°	− 82.0°

[a] Dicyclohexylammonium salts.

45 : 45 : 10; (b) methanol/ethyl acetate/dichloromethane, 1 : 2 : 3; compounds were visualized by chlorine test. Microanalyses were carried out on an elemental analyzer Perkin Elmer model 240. The ^1H- and ^{13}C-N.M.R. spectra were recorded on a 90 MHz spectrometer (model HS-90 R, Bruker-Physik, Karlsruhe–Forchheim); chemical shifts (δ) are given in ppm vs. with TMS as internal standard.

2-(2-Nitrobenzenesulphenyl)-1,2-benzoisothiazoline-3-one 1,1-Dioxide (1; Nps-saccharin):

To a solution of saccharin sodium salt (5.1 g, 25 mmol) in 3/1 dioxan/water (40 ml), 2-nitrobenzenesulphenyl chloride (4.7 g, 25 mmol) in dioxan (10 ml) is added dropwise at room temperature under vigorous stirring. After 2 h, water (100 ml) is added to complete the precipitation. The precipitate is collected by filtration, washed with water and ethyl acetate, and dried; yield: 7.1 g (85 %); m.p. 185–186 °C (dec.); homogeneous on T. L. C. in solvent systems (a) and (b).

$C_{13}H_8N_2O_5S_2$	C 46.42	H 2.40	N 8.33	S 19.07
(336.3)	46.57	2.39	8.36	18.82

(2-Nitrobenzenesulphenyl)-amino Acids 3; General Procedure:

To a chilled solution of the amino acid or amino acid derivative (10 mmol) in 1 normal aqueous sodium hydroxide (10 ml) and dioxan (10 ml), Nps-saccharin (**1**; 3.36 g, 10 mmol) in tetrahydrofuran (40 ml) is added dropwise under vigorous stirring. The mixture is kept at room temperature for 12 h, then the organic solvents are evaporated, the aqueous solution diluted with water (50 ml), acidified with 2 % aqueous potassium hydrogen sulphate to pH 3, and extracted with ethyl acetate (3 × 50 ml). The amino acid derivatives are isolated by known procedures (Table).

2-Benzyloxycarbonyl-1,2-benzoisothiazoline-3-one 1,1-Dioxide (2; *N*-Benzyloxycarbonyl-saccharin):

To a suspension of saccharin sodium salt (20.5 g, 0.1 mol) in ice-cold tetrahydrofuran (200 ml), benzyloxycarbonyl chloride (16 ml, 0.1 mol) is added dropwise under stirring. The mixture is then kept at 50 °C for 6 h and at room temperature for additional 12 h. Insoluble material is filtered off and the filtrate evaporated. The oily residue crystallises from ether; yield: 27.6 g (87 %); m.p. 104 °C; homogeneous on T. L. C. in the solvent systems (a) and (b).

$C_{15}H_{11}NO_5S$	calc.	C 56.78	H 3.49	N 4.41	S 10.10
(317.3)	found	56.58	3.48	4.34	10.02

^{1}N.M.R.($CDCl_3$/TMS): δ = 5.49 (s,2H, CH_2); 7.25–8.20 ppm (m, 9 H_{arom}).

^{13}C-N.M.R. ($CDCl_3$/TMS); δ = 155.9 (C-3); 147.0 (—O—CO—); 137.4 (C-1'); 136.3 (C-7a); 134.9, 134.0 (C-5, C-6); 128.8, 128.7 (C-3', C-5', C-3a); 128.2 (C-2', C-6'); 126.2 (C-6); 125.7 (C-4'); 121.3 (C-7); 69.8 ppm (CH_2).

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* Address for correspondence.

[1] Zervas, L., Borovas, D., Gazis, E. *J. Am. Chem. Soc.* **1963**, *85*, 3660.

[2] Ramegowda, N.S., Narang, C.K., Bachawat, J.M., Mathur, N.K. *Indian J. Chem.* **1972**, *10*, 1194.

[3] Michel, F., Lorenz, M. *Liebigs Ann. Chem.* **1966**, *698*, 242.

[4] Imai, Y., Okunoyama, H., Ohkashi, M. *Nippon Kagaku Kaishi* **1975**, 1, 123; *C.A.* **1976**, *84*, 17204.