On the Use of Five-Membered Heterocycles in Peptide Chemistry*

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N-Acyl derivatives of 2-benzoxazolethiol, 2-benzothiazolethiol and 2-benzimidazolethiol are proposed for use both as efficient acylating agents for the synthesis of *N*-benzyloxycarbonyl and *N*-fluorenylmethyloxycarbonyl amino acid derivatives and as activated amides for the synthesis of peptides.

N- and *S*-acyl derivatives of five-membered heterocycles have found widespread application in the last years as acylating agents in synthetic chemistry[1-6]; we have recently explored their potential usefulness in various aspects of peptide synthesis***.

1. Amine-Protecting Reagents

1.1. New Benzyloxycarbonyl Donors

Since the discovery of the benzyloxycarbonyl group for the temporary protection of amine functions[7], the related amino acid derivatives are mainly prepared under Schotten-Baumann conditions using benzyloxycarbonyl chloride as acylating agent. The main disadvantages of this reagent are as follows.

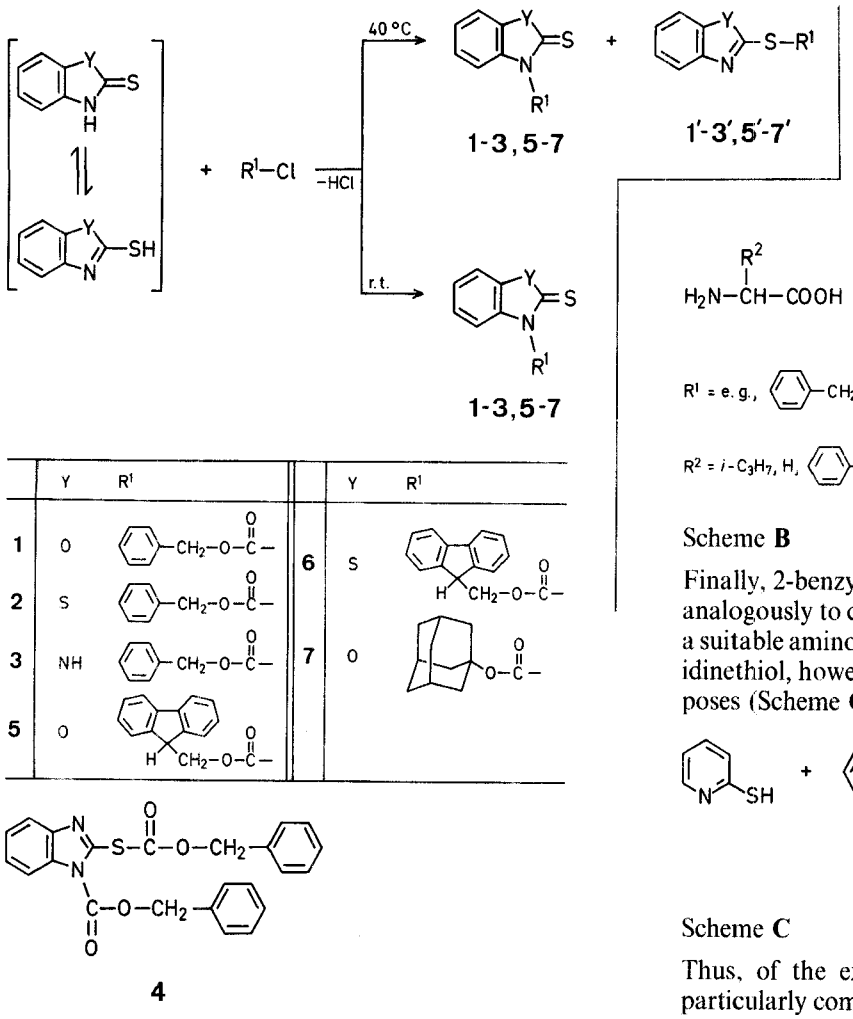
(1) It is not readily obtained in a high degree of purity and usually it contains benzyl chloride to various extents.
(2) Traces of metal ions as well as of chloride ions catalyze further decomposition to benzyl chloride on storage over longer periods of time even in the cold. Consequently, the yields of benzyloxycarbonyl amino acids are lowered and side reactions may occur particularly in the case of polyfunctional amino acids.
(3) The synthesis of benzyloxycarbonyl amino acids using benzyl carbonochloridate may be accompanied by the formation of oligopeptides via the mixed anhydride mechanism. Depending on the reaction conditions (pH, concentration, temperature) up to 20% *N*-benzyloxycarbonyl-dipeptides were recently detected in crude benzyloxycarbonyl-L-threonine and benzyloxycarbonyl-L-serine preparations[8].

Several alternative benzyloxycarbonyl donors have been already proposed in the past, e.g. mixed carbonates with various phenols[9,10], 1-hydroxypiperidine[11] and *N*-hydroxysuccinimide[12,13], or benzyloxycarbonyl-thiosulfate sodium salt[14], 1-benzyloxycarbonyl-3-methylimidazolium chloride[15], 3-benzyloxycarbonyl-2-oxazolone[5]. These reagents, however, have not found so far much application, thus prompting us to search for further, inexpensive, efficient benzyloxycarbonyl donors with a good stability on storage.

For such purposes, freshly prepared benzyl carbonochloridate was reacted with a few selected heterocycles such as 2-benzoxazolethiol, 2-benzothiazolethiol, and 2-benzimidazolethiol. The reaction was found to proceed smoothly at room temperature with production of the corresponding benzyloxycarbonyl derivatives in good yields as crystalline compounds which are stable on storage over longer periods of time. Acylation of the above mentioned heterocycles is expected to yield, depending on the reaction conditions, *N*- and *S*-derivatives or a mixture of both isomers, whereby the *N*-derivatives should be the thermodynamically more stable isomers[6]. In fact, benzyloxycarbonylation of 2-benzoxazolethiol at room temperature under the conditions reported in the experimental part led to 3-benzyloxycarbonyl-benzoxazoline-2-thione **(1)**, as unequivocally determined by an X-ray structure analysis of the reaction product: the acyl substituent is located at the nitrogen in position 3[16] (Scheme **A**).

On performing the acylation reaction at 40 °C, a mixture of the kinetically controlled *S*-isomer and the thermodynamically preferred *N*-isomer is formed as determined by N.M.R. spectrometric analysis of the signals for the benzyl CH_2 group [^{1}H-N.M.R. ($CDCl_3$): δ = 5.53 ppm (s, 2H) for the *N*-isomer and 5.30 ppm (s, 2H) for the *S*-isomer; ^{13}C-N.M.R. ($CDCl_3$): δ = 70.4 ppm for the *N*-isomer and 71.0 ppm for the *S*-isomer].

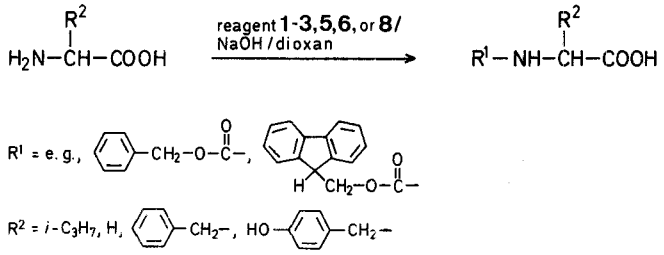
Using the ^{1}H- and ^{13}C-N.M.R. spectra of compound **1** as a reference, it has been possible to assign the *N*-acyl structure to the benzyloxycarbonyl derivatives of 2-benzothiazolethiol **(2)** and 2-benzimidazolethiol **(3)**. Benzyloxycarbonylation of 2-benzimidazolethiol in absence of base leads to 1-benzyloxycarbonylbenzimidazoline-2-thione hydrochloride (**3**·HCl) whereas in the presence of a base, e.g. triethylamine or sodium carbonate, besides the monosubstituted derivative



Scheme A

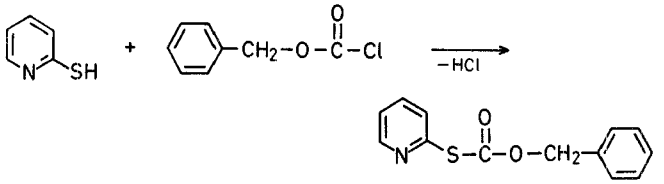
1-benzyloxycarbonyl-2-benzyloxycarbonylthio-benzimidazole (**4**) is also formed to some extent.

Compounds **1**–**3** were found to represent efficient benzyloxycarbonyl donors (Table 1) although their acylating potency is lower than that of benzyl carbonochloridate, as clearly demonstrated by the fact that *N*-benzyloxycarbonyl-L-tyrosine is obtained in good yields (75%) directly from L-tyrosine with reagent **1** without concomitant acylation of the phenolic function (Scheme **B**).



Scheme **B**

Finally, 2-benzyloxycarbonylthio-pyridine (**8**) was prepared analogously to compound **1** and was also found to represent a suitable amino-protecting reagent. The high costs of 2-pyridinethiol, however, reduce its significance for practical purposes (Scheme **C**).



Scheme **C**

Thus, of the examined *N*-benzyloxycarbonyl derivatives, particularly compounds **1** and **2** may represent suitable benzyloxycarbonyl donors because of their low production costs, high stability, and sufficient acylating potency.

1.2. New 9-Fluorenylmethyloxycarbonyl Donors

9-Fluorenylmethyloxycarbonyl amino acid derivatives[17] have recently become commonly used intermediates for peptide synthesis. Their preparation by the standard procedures using 9-fluorenylmethyl carbonochloridate as acylating agent is accompanied by a side reaction similar to that discussed above for benzyloxycarbonyl amino acids; as recently reported[8,18–21], the resulting amino acid derivatives are contaminated by substantial amounts of 9-fluorenylmethyloxycarbonyl oligopeptides.

Table 1. Synthesis of *N*-Protected Amino Acid Derivatives (Scheme **B**)

Amino Acid	Reagent	Yield [%]	m.p. [°C]	Optical Rotations		
				$[\alpha]_D^{20}$	$[\alpha]_{546}^{20}$	*c* 1, solvent
Z-Val-OH	**1**	81	59–60°	+15.7°	+18.6°	$CHCl_3$
Z-Val-OH	**2**	90	60°	+15.7°	+18.5°	$CHCl_3$
Z-Val-OH	**3**	91	62°	+15.4°	+18.4°	$CHCl_3$
Z-Val-OH	**8**	82	62°	+15.4°	+18.6°	$CHCl_3$
Z-Gly-OH	**1**	79	120°	–	–	–
Z-Phe-OH	**1**	70	88–89°	– 4.7°	– 6.1°	CH_3COOH
Z-Tyr-OH · DCHA[a]	**1**	75	91–93°	+25°	+30°	CH_3OH
Fmoc-Val-OH	**6**	91	147–147.5[a]	–17.4°	–21.4°	DMF
Fmoc-Gly-OH	**6**	88	176–177°	–	–	–
Fmoc-Phe-OH	**5**	70	185–186.5°	–39.5°	–47.2°	DMF

[a] Dicyclohexylammonium salt.

To bypass this side reaction various other fluorenylmethyl carbonates have been proposed, among which the mixed carbonate with *N*-hydroxysuccinimide was found to be the most efficient amino-protecting reagent[20,21].

The successful use of the five-membered heterocycles for the preparation of benzyloxycarbonyl donors prompted us to investigate their potential application for the present problem. Both 2-benzoxazolethiol and 2-benzothiazolethiol react with 9-fluorenylmethyl carbonochloridate to give the corresponding *N*-acyl derivatives **5** and **6** (Scheme **A**). These compounds are stable, crystalline solids which react with amino acids (Scheme **B**, Table 1) to produce related 9-fluorenylmethyloxycarbonyl amino acids. Formation of oligopeptides was not observed to occur by this procedure.

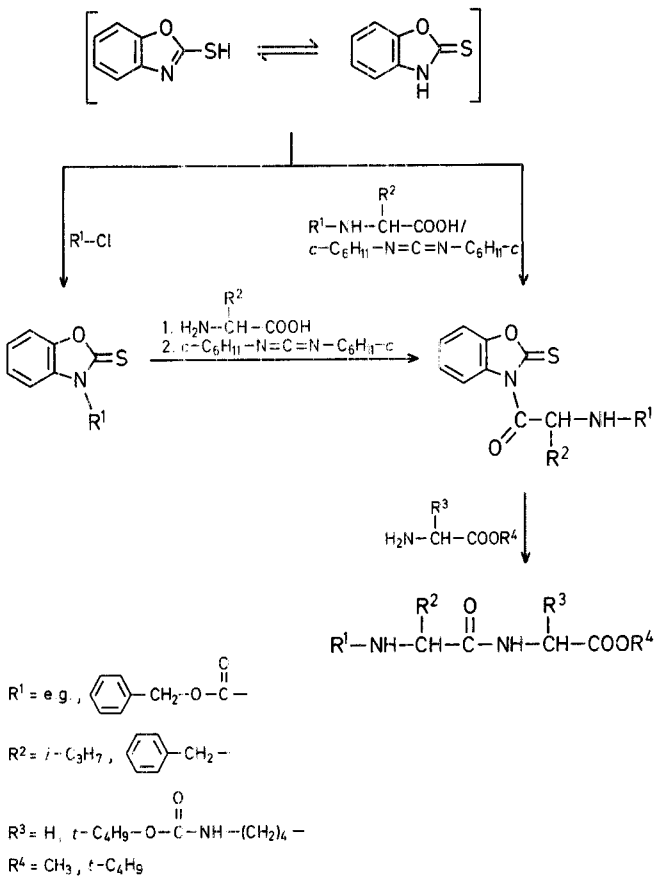
1.3. New Adamantyloxycarbonyl Donors

We have recently proposed adamantyl carbonofluoridate[22] as reagent for the preparation of adamantyloxycarbonyl amino acids, since it was found to be remarkably more stable than the corresponding carbonochloridate[23]. To avoid possible oligopeptide formation, as discussed above for other carbonochloridates, we have now explored the use of heterocyclic adamantyloxycarbonyl derivatives as amino-protecting reagents.

Adamantyl carbonofluoridate reacts with 2-benzoxazolethiol to yield the corresponding 3-adamantyloxycarbonyl-benzoxazoline-2-thione (**7**, Scheme **A**), whereas reaction of the carbonofluoridate with benzothiazole-2-thiol leads to quantitative auto-oxidation of the heterocyclic compound catalyzed by the fluoride ions. Although reagent **7** proved to be an efficient adamantyloxycarbonyl donor, it is not of practical usefulness for the preparation of simple amino acid derivatives; for solubility reasons a quantitative removal of the resulting 2-benzoxazolethiol is hardly possible.

2. New Methods for Peptide Bond Formation

The successful application of this series of heterocycles as leaving groups in amino-protecting reagents led us to investigate their usefulness for peptide bond formation (Scheme **D**).



Scheme **D**

N-Protected amino acids were reacted e.g. with 2-benzoxazolethiol using dicyclohexylcarbodiimide as condensing agent. The thermodynamically more stable *N*-acyl derivatives are formed in good yields as stable "activated" amino acid derivatives (Table 2). These were found to react smoothly with amino acid esters to give dipeptides in yields comparable to those obtained by using known active esters, e.g. by *N*-hydroxysuccinimide esters. Racemisation was not found to occur to detectable degrees as determined by gas chromatographic chiral analysis according to Ref.[24].

Table 2. 3-(*N*-Benzyloxycarbonylaminoacyl)-benzoxazoline-2-thiones[a]

Amino Acid Derivative	Yield [%] (Method)	m.p. [°C] (solvent)	Optical Rotations (c 1, dioxan) $[\alpha]_D^{20}$	$[\alpha]_{546}^{20}$	Molecular Formula[b]
Z-Val	73 (A) 56 (B)	113–116° (*c*-C_3H_7OH)	−18.9°	−21.2°	$C_{13}H_{17}NO_4$ (251.3)
Z-Trp	72 (A)	155–159° (C_2H_5OAc/pE)	+16.5°	+21.3°	$C_{19}H_{18}N_2O_4$ (338.4)
Z-Phe	84 (A)	108–111° (C_2H_5OAc/pE)	−23.9°	−27.1°	$C_{17}H_{17}NO_4$ (299.3)
Z-Gly	65 (A)	140–141° (C_2H_5OAc/ether)	—	—	$C_{10}H_{11}NO_4$ (209.2)

[a] The *N*-acyl structure was assigned on the basis of the ^1H- and ^{13}C-N.M.R. data as shown by the example of 3-(*N*-benzyloxycarbonyl-L-valyl)-benzoxazoline-2-thione:
^1H-N.M.R. ($CDCl_3$): δ = 0.86 (d, 3H, J = 7.0 Hz, Val-$H_{\gamma b}$); 1.14 (d, 3H, J = 6.7 Hz, Val-$H_{\gamma a}$); 2.42 (m, 1H, Val-H_β); 5.14 (s, 2H $CH_2C_6H_5$); 5.52 (br. d, 1H, Val-H_α); 6.60–8.06 ppm (m, 10H, aromatic H + NH).
^{13}C-N.M.R. ($CDCl_3$): δ = 15.60, 19.63 (Val, 2C$_\gamma$); 30.09 (Val, C$_\beta$); 58.69 (Val, C$_\alpha$); 67.26 ($CH_2C_6H_5$); 109.77 (C-7); 116.27 (C-4); 125.63–146.55 (other aromatic C); 173.5 ppm (C-5).

[b] Satisfactory microanalyses obtained: C ±0.26, H ±0.15, N ±0.12, S ±0.42.

The main shortcomings of the use of sulfur-containing heterocycles in peptide chemistry are related to their poisoning effect on the palladium catalyst routinely used for hydrogenolytic debenzyloxycarbonylation of synthetic peptide intermediates. However, the high solubility of the thiol compounds in most organic solvents and the easy removal of trace contaminations by short treatments with charcoal, offer an easy bypass to such expected difficulties. The model dipeptide Z-Phe-Gly-OCH_3 was quantitatively debenzyloxycarbonylated by hydrogenolysis in the usual reaction time.

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, Darmstadt) using the solvent systems: (a) cyclohexane/chloroform/acetic acid 45 : 45 : 10, (b) methanol/ethyl acetate/dichloromethane 1 : 2 : 3; (c) *n*-heptane/t-butanol/acetic acid 3 : 2 : 1; (d) *n*-butanol/acetic acid/water 3 : 1 : 1; compounds were visualized by chlorine test. The microanalyses were carried out on elemental analyzer Perkin Elmer model 240. The N.M.R. spectra were recorded on a 90 MHz spectrometer (model HX—90 R, Bruker-Physik, Karlsruhe-Forchheim); chemical shifts are given in ppm vs. TMS as internal standard.

3-Benzyloxycarbonylbenzoxazoline-2-thione (1):

Method A: To a solution of 2-benzoxazolethiol (7.5 g, 50 mmol) and triethylamine (8.3 ml, 60 mmol) in tetrahydrofuran (100 ml), benzyl carbonochloridate (9.5 ml, 60 mmol) is added dropwise under vigorous stirring at room temperature. After 1 h the bulk of the solvent is evaporated and the residue distributed between ethyl acetate (100 ml) and water (50 ml). The organic phase is dried with sodium sulfate and concentrated to a small volume. On addition of petroleum ether the crystalline product separates and is collected and dried; yield: 11 g (77%); m.p. 89–92 °C; homogeneous on T.L.C., solvent systems (a), (b), and (c).

$C_{15}H_{11}NO_3S$	calc.	C 63.14	H 3.89	N 4.91	S 11.24
(285.3)	found	63.24	3.86	4.63	11.27

^{1}H-N.M.R. ($CDCl_3$): δ = 5.53 (s, 2 H; $CH_2C_6H_5$); 7.17–7.74 ppm (m, 9 H_{arom}).

^{13}C-N.M.R. ($CDCl_3$): δ = 70.4 ($CH_2C_6H_5$); 109.9 (C-7); 115.1 (C-4); 125.2, 125.9 (C-5, C-6); 128.8–133.1 (C-3a, C-1′–C-6′); 146.1 (C-7a); 149.6 (O—CO—); 176.6 ppm (C-2).

Method B: To a solution of 2-benzoxazolethiol (22.7 g; 150 mmol) in ethyl acetate (250 ml), sodium carbonate (9.30 g, 88 mmol) is added followed by benzyl carbonochloridate (23.5 ml, 176 mmol) in portions. After 1 h, insoluble salts are removed by filtration, the filtrate is washed with water, dried with sodium sulfate, and concentrated to a small volume. The product **1** crystallizes on addition of petroleum ether; yield: 38.3 g (89%); m.p. 90–91 °C.

3-Benzyloxycarbonylbenzothiazoline-2-thione (2):

Prepared from benzyl carbonochloridate (9.5 ml, 60 mmol) and 2-benzothiazolethiol (8.4g ,50 mmol) as described above for **1** (Method A); yield: 11.6 g (77%); m.p. 87 °C; homogenous on T.L.C., solvent systems (a), (b), and (c).

$C_{15}H_{11}NO_2S_2$	calc.	C 59.78	H 3.68	N 4.65	S 21.28
(301.4)	found	59.55	3.64	4.64	21.26

^{1}H-N.M.R. (DMSO-d_6): δ = 5.35 (s, 2 H, $CH_2C_6H_5$); 7.38–8.06 ppm (m, 9 H_{arom}).

^{13}C-N.M.R. (DMSO-d_6): δ = 70.8 ($CH_2C_6H_5$); 121.1–128.9 (other aromatic C); 134.3 (C-7a); 136.1 (C-3a); 152.9 (O—CO—); 165.1 ppm (C-2).

1-Benzyloxycarbonylbenzimidazoline-2-thione (3):

Method A: To a solution of 2-benzimidazolethiol (1.50 g, 10 mmol) in tetrahydrofuran (50 ml), benzyl carbonochloridate (1.9 ml, 12 mmol) is added dropwise under stirring at room temperature. The precipitate is collected by filtration and washed with ether to give 1-benzyloxycarbonyl-benzimidazoline-2-thione hydrochloride; yield: 2.3 g (70%); m.p. 223–224 °C; homogeneous on T.L.C., solvent systems (a), (b), and (c).

$C_{15}H_{12}N_2O_2S$·HCl	calc.	C 56.16	H 4.08	N 8.73	S 9.99	Cl 11.05
(320.8)	found	56.59	4.10	8.70	9.95	10.99

Method B: To 2-benzimidazolethiol (3 g, 20 mmol) and triethylamine (3.4 ml, 24 mmol) in tetrahydrofuran (50 ml), benzyl carbonochloridate (3.8 ml, 24 mmol) is added dropwise at room temperature. After 12 h the solvent is evaporated and the residue distributed between ethyl acetate (100 ml) and water (50 ml). The organic phase is dried with sodium sulfate and concentrated to a small volume. The crystalline product is collected and dried to give 1-benzyloxycarbonylbenzimidazoline-2-thione **(3)**; yield: 3.3 g (59%); m.p. 140 °C.

$C_{15}H_{12}N_2O_2S$	calc.	C 63.36	H 4.25	N 9.85	N 11.27
(284.3)	found	63.27	4.21	9.66	11.39

^{1}H-N.M.R. (DMSO-d_6): δ = 5.38 (s, 2 H, $CH_2C_6H_5$); 7.25–7.62 (m, 9 H_{arom}); 12.64 ppm (br. s, 1 H, NH).

^{13}C-N.M.R. (DMSO-d_6): δ = 70.1 ($CH_2C_6H_5$); 122.8–128.4 (other aromatic C); 134.7 (C-7a); 138.9 (C-3a); 165.9 ppm (C-2).

1-Benzyloxycarbonyl-2-benzyloxycarbonylthiobenzimidazole (4):

On concentration of the mother liquors from the preparation of **3** (Method B) and addition of ether, the title compound is isolated as a crystalline material; yield: 0.8 g (10%); m.p. 119–120 °C.

$C_{23}H_{18}N_2O_4S$	calc.	C 66.02	H 4.34	N 6.69	S 7.66
(418.5)	found	66.04	4.62	6.60	7.80

^{1}H-N.M.R. (DMSO-d_6): δ = 5.26 (s, 2 H, $CH_2C_6H_5$); 5.51 ppm (s, 2 H, $CH_2C_6H_5$).

3-(9-Fluorenylmethyloxycarbonyl)-benzoxazoline-2-thione (5):

The title compound is prepared from 9-fluorenylmethyl carbonochloridate (15.5 g, 60 mmol) and 2-benzoxazolethiol (7.5 g, 50 mmol) as described for **1** (Method B): The product crystallizes from ethyl acetate/hexane; yield: 10.8 g (58%); m.p. 181–182 °C; homogeneous on T.L.C., solvent systems (a) and (c).

$C_{22}H_{15}NO_3S$	calc.	C 70.76	H 4.05	N 3.75	S 8.59
(373.4)	found	70.64	4.10	3.72	8.56

^{1}H-N.M.R. (DMSO-d_6): δ = 4.56 (t, 1 H, J = 5.9 Hz, H-9); 5.13 (d, 2 H, J = 5.9 Hz, CH_2O); 6.51–7.99 ppm (m, 12 H_{arom}).

3-(9-Fluorenylmethyloxycarbonyl)-benzothiazoline-2-thione (6):

The title compound is prepared from 9-fluorenylmethyl carbonochloridate (15.5 g, 60 mmol) and 2-benzothiazolethiol (8.4 g, 50 mmol) as described for **1** (Method B); the product crystallizes from ethyl acetate/hexane; yield: 13.8 g (71%); m.p. 96–98 °C; homogeneous on T.L.C., solvent systems (a) and (c).

$C_{22}H_{15}NO_2S_2$	calc.	C 67.84	H 3.88	N 3.65	S 16.47
(389.5)	found	67.88	3.88	3.57	16.52

^{1}H-N.M.R. (DMSO-d_6): δ = 4.38 (t, 1 H, J = 5.9 Hz, H-9); 4.85 (d, 2 H, J = 5.9 Hz, OCH_2); 7.23–8.20 ppm (m, 12 H_{arom}).

3-Adamantyloxycarbonylbenzoxazoline-2-thione (7):

The title compound is prepared from adamantyl carbonofluoridate (11.8 g, 60 mmol) and 2-benzoxazolethiol (7.5 g, 50 mmol) as described for **1** (Method A); the product crystallizes from ethyl acetate/ether; yield: 11.9 g (72%); m.p. 168–170 °C; homogeneous on T.L.C., solvent systems (a), (b), and (c).

$C_{18}H_{19}NO_3S$	calc.	C 65.63	H 5.81	N 4.25	S 9.73
(329.4)	found	65.62	5.82	4.20	9.70

2-Benzyloxycarbonylthiopyridine (8):

The title compound is prepared from benzyl carbonochloridate (9.5 ml, 60 mmol) and 2-pyridinethiol (5.6 g, 50 mmol) as described for **1** (Method A) and crystallization from ether/petroleum ether; yield: 8.5 g (71%); m.p. 37 °C; homogeneous on T.L.C., solvent systems (a) and (d).

$C_{13}H_{11}NO_2S$	calc.	C 63.65	H 4.52	N 5.71	S 13.07
(245.3)	found	63.59	4.50	5.72	13.44

^{1}H-N.M.R. (DMSO-d_6): δ = 5.33 ppm (s, 2H, $CH_2C_6H_5$).

^{13}C-N.M.R. (DMSO-d_6): δ = 69.3 ppm ($CH_2C_6H_5$).

Benzyloxycarbonyl- and 9-Fluorenylmethyloxycarbonylamino Acids; General Procedure:

To a chilled solution of the amino acid (10 mmol) in 1 normal sodium hydroxide solution (10 ml) and dioxan (10 ml), reagent **1**,**2**,**3**,**5**,**6**, or **8** (11 mmol) in dioxan (20 ml) is added dropwise under vigorous stirring. After 12 h at room temperature the bulk of the dioxan is evaporated, the resulting aqueous solution diluted with 5% aqueous sodium hydrogen carbonate solution (10 ml), and extracted with ether or ethyl acetate (3 × 30 ml). The aqueous phase is acidified with 1 normal sulfuric acid to pH 1 and extracted with ethyl acetate (3 × 30 ml). The combined latter extracts are washed sulfate-free with water, dried with sodium sulfate, and evaporated. The amino acid derivatives are isolated by known procedures; see Table 1.

3-(*N*-Benzyloxycarbonylaminoacyl)-benzoxazoline-2-thiones; General Procedure:

Method A: To a chilled solution of the *N*-benzyloxycarbonylamino acid (10 mmol) and 2-benzoxazolethiol (10 mmol) in ethyl acetate (or tetrahydrofuran, 30 ml), dicyclohexylcarbodiimide (10 mmol) is added. The mixture is stirred for 2 h in the ice-bath and 12 h at room temperature. The urea is filtered off and from the filtrate the amino acid derivatives are isolated by crystallization; see Table 2.

Method B: To a chilled solution of the amino acid (10 mmol) and sodium carbonate (5 mmol) in 1:1 dioxan/water (20 ml), 3-benzyloxycarbonylbenzoxazoline-2-thione (11 mmol) in dioxan (40 ml) is added under stirring. After 12 h at room temperature the bulk of the dioxan is evaporated. The resulting solution is diluted with water (20 ml), acidified with 1 normal sulfuric acid to pH 1 and extracted with ethyl acetate (3 × 30 ml). The combined extracts are washed sulfate-free with water and dried with sodium sulfate. The sodium sulfate is removed by filtration, the filtrate cooled in an ice-bath, and treated with dicyclohexylcarbodiimide (11 mmol). The reaction is allowed to proceed and worked up as described above; see Table 2.

Benzyloxycarbonyl-L-valyl-glycine Methyl Ester:

To a solution of H-Gly-OCH_3 · HCl (2.5 g, 20 mmol) and triethylamine (2.78 ml; 20 mmol) in dimethylformamide (20 ml), 3-(*N*-benzyloxycarbonyl-L-valyl)-benzoxazoline-2-thione (6.9 g, 18 mmol) is added. After 24 h the solvent is evaporated and the residue distributed between ethyl acetate (50 ml) and water (20 ml). The organic layer is washed with 5% sodium hydrogen carbonate solution, 2% potassium hydrogen sulfate solution, and water, dried with sodium sulfate, and concentrated. On addition of ether the product separates; yield: 4.8 g (82%); m.p. 160–161 °C; $[\alpha]_D^{20}$: −28.32°, $[\alpha]_{546}^{20}$: −33.74° (*c* 1, methanol); chiral analysis: D-Val < 0.1%.

$C_{16}H_{22}N_2O_5$	calc.	C 59.62	H 6.88	N 8.69
(322.4)	found	59.69	6.93	8.70

Benzyloxycarbonyl-L-phenylalanyl-glycine Methyl Ester:

The title compound is prepared from H-Gly-OCH_3-HCl (2.5 g, 20 mmol) and 3-(*N*-benzyloxycarbonyl-L-phenylalanyl)-benzoxazoline-2-thione (7.8 g, 18 mmol) as described for Z-Val-Gly-OCH_3; yield: 5.6 g (84%); m.p. 120 °C; $[\alpha]_D^{20}$: −19.3°, $[\alpha]_{546}^{20}$: −22.1° (*c* 1, methanol); chiral analysis: D-Phe = 0.8%.

Preparation via Z-Phe-ONSu; yield: 73%; m.p. 117–119 °C; $[\alpha]_D^{20}$: 18.8°, $[\alpha]_{546}^{20}$: −23.0° (*c* 1, methanol); chiral analysis: D-Phe = 0.6%.

$C_{20}H_{22}N_2O_5$	calc.	C 64.85	H 5.99	N 7.56
(370.4)	found	64.77	5.91	7.50

Benzyloxycarbonyl-L-valyl-N^{ε}-*t*-butyloxycarbonyl-L-lysine *t*-Butyl Ester:

The dipeptide is obtained from H-Lys(Boc)-OC_4H_9-*t* (7.5 g, 22 mmol) and 3-(*N*-benzyloxycarbonyl-L-valyl)-benzoxazoline-2-thione (7.7 g, 20 mmol) as described above for Z-Val-Gly-OCH_3 and crystallization from ethyl acetate/petroleum ether; yield: 8.2 g (77%); m.p. 85–86 °C; $[\alpha]_D^{20}$: −25.7°, $[\alpha]_{546}^{20}$: −29.6° (*c* 1, methanol); chiral analysis: D-Val < 0.5%, D-Lys = 1.8%.

Preparation via Z-Val-ONSu; yield: 84%; m.p. 85–87 °C; $[\alpha]_D^{20}$: −25.2°, $[\alpha]_{546}^{20}$: −29.9° (*c* 1, methanol); chiral analysis: D-Val < 0.5%, D-Lys = 1.6%.

$C_{28}H_{45}N_3O_7$	calc.	C 62.78	H 8.47	N 7.84
(535.7)	found	62.81	8.39	7.82

This work is dedicated to Prof. K. Schlögl on the occasion of his 60th birthday.

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*** Standard abbreviations for amino acids and peptide derivatives are used as recommended by the IUPAC-IUB commission on biochemical nomenclature.

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