Chloroperoxidase-catalyzed oxidation of methionine derivatives

Herbert L. Holland, Frances M. Brown, Damian Lozada, Benjamin Mayne, W. Rick Szerminski, and Aaron J. van Vliet

Abstract: Treatment of *N*-methoxycarbonyl *C*-carboxylate ester derivatives of L- and D-methionine and L-ethionine by chloroperoxidase–hydrogen peroxide resulted in oxidation at sulfur to produce the (R_S) sulfoxide in moderate to high diastereomeric excess. The (R_S) sulfoxide of methionine was also obtained in moderate to high diastereomeric excess from $(\pm)_{SO}$-*N*-methoxycarbonyl-L-methionine methyl ester sulfoxide by ester hydrolysis using α-chymotrypsin, *Aspergillus* sp. protease or subtilisin Carlsberg.

Key words: amino acid oxidation, biocatalysis, biotransformation, chloroperoxidase, enzyme catalysis, lipase, sulfoxidation.

Résumé : Le traitement de dérivés *N*-méthoxycarbonyle *C*-carboxylates des L- et D-méthionines et de la L-éthionine par de la chloroperoxydase et du peroxyde d'hydrogène conduit à l'oxydation au niveau du soufre et à la formation de sulfoxydes (R_S), avec des excès diastéréomériques qui vont de modérés à élevés. Le (R_S) sulfoxyde de la méthionine a aussi été obtenu avec un excès diastéréomérique élevé à partir de l'ester méthylique du de sulfoxyde $(\pm)_{SO}$-*N*-méthoxycarbonyl-L-méthionine, par hydrolyse de l'ester à l'aide d'α-chymotrypsine, de protéase d'*Aspergillus* sp. ou de subtilisine Carlsberg.

Mots clés : oxydation d'acide aminé, biocatalyse, biotransformation, chloroperoxydase, catalyse enzymatique, lipase, sulfoxydation.

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Introduction

Sulfoxides of the sulfur-containing amino acids possess various biological properties. Oxidation of methionine residues in proteins is the target of continued interest, and can result in structural and functional changes in protein (1) and (or) cellular oxidative-stress properties (2). The reversible oxidation of methionine (**1**) can also be involved in other biological functions such as the modulation of potassium-channel activity (3) and the oxidation of high- density lipoproteins (4, 5). It is also implicated in ageing processes by the effect of methionine sulfoxidation on the activity of the human keratinocyte growth factor (6) and on the immunopeptide thymosin β-4 (7); the in vivo oxidation of methionine to the sulfoxide is an age-dependent process in humans (8). The carcinogenic properties of ethionine (**2**) have been linked to its in vivo oxidation to sulfoxide (9), while *S*-methylcysteine sulfoxide (**3**-sulfoxide) is responsible for the antibacterial action of cabbage (10) and possesses the ability to lower serum-cholesterol levels (11, 12).

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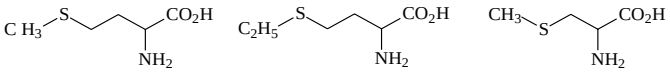
This paper is dedicated to Professor J. Bryan Jones, whose introductory work, continued research, and present-day creations are all fundamental areas of biocatalysis.

H.L. Holland,[1] F.M. Brown, D. Lozada, B. Mayne, W.R. Szeminski, and A.J. van Vliet. Department of Chemistry, Brock University, St. Catharines, ON L2S 3A1, Canada.

[1]Corresponding author (e-mail holland@chemiris.labs.brocku.ca).



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With the exception of L-methionine sulfoxide isolated from the blowfly *Phormia regina* (13), and *S*-methyl-L-cysteine sulfoxide from cabbage (14), both of which have the (S_S) configuration, no data are available on the configuration at sulfur in naturally occurring amino acid sulfoxides.

The stereoselective chemical oxidation at sulfur of a thia-amino acid has not been reported, and only recently have other chemical routes for the preparation of diastereomerically defined amino acid sulfoxides been systematically investigated (14). The value of biocatalytic oxidation for the stereoselective oxidation of sulfides to sulfoxides, however, is well established (15, 16), and was used for the oxidation of methionine by *Penicillium citreoviride*, but no stereochemical details were reported (17). Fungal biocatalysts oxidize various prochiral sulfides to chiral sulfoxides. The use of *Beauveria bassiana* ATCC 7159 and *B. caledonica* ATCC 64970 for the stereoselective oxidation of *N*-phthaloyl-methionines and -ethionines to the (S_S) methionine and ethionine sulfoxides has recently been reported (18, 19). The application of isolated enzymes for chiral sulfoxidation is less common, but chloroperoxidase (CPO) from *Caldariomyces fumago* has been systematically used for the oxidation of various sulfides (16, 20–22). Cyclohexanone monooxygenase (CMO) from *Acinetobacter* species has also been employed for moderate diastereomeric sulfoxidation of *S*-allylcysteine (alliin) (23).

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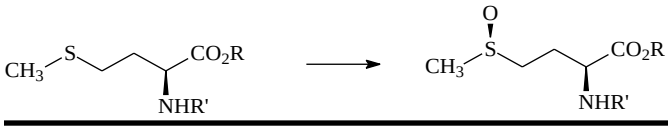
Table 1. CPO-catalyzed oxidations of L-methionine derivatives.



R	R′	Conversion (%)	de (%)	Sulfoxide stereochemistry
H	Ac	0	—	—
	MOC	2	—	—
	Phthaloyl	0	—	—
CH_3	Ac	20	72	*R*
	MOC	84	82	*R*
	ClAc	85	12	*R*
	CHO	25	30	*R*
	t-Boc	10	72	*R*
C_2H_5	MOC	60	72	*R*
n-C_3H_7	MOC	72	84	*R*
n-C_4H_9	MOC	60	48	*R*
n-C_5H_{11}	MOC	50	60	*R*

With the latter exception, the present paper describes the first use of isolated enzymes, particularly the oxidizing species chloroperoxidase–hydrogen peroxide, for the stereoselective oxidation at sulfur in methionine and ethionine derivatives.

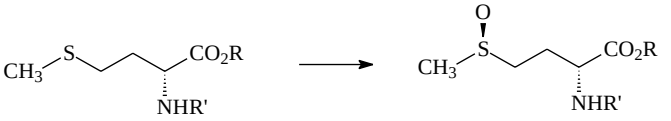
Results and discussion

Table 1 summarizes the optimum use of substrate-protecting groups for the chloroperoxidase-catalyzed oxidation of L-methionine derivatives, where maximum yield and diastereomeric excesses were obtained with the *N*-methoxycarbonyl (*N*-MOC) *C*-carboxylate methyl ester derivatives.

The configurations at sulfur of the sulfoxides listed in Table 1 were obtained by analysis of their ^{13}C NMR spectra. Although the ^{1}H NMR spectra of the two diastereomers (S_SS_C) and (R_SS_C) were essentially identical, the signals in ^{13}C NMR spectra from the α and β carbons to the sulfoxide sulfur were separable in methanol. Comparison with ^{13}C NMR spectra from diastereomerically pure L-methionine sulfoxide standards with similar protection groups (19), were then used to assign the configurations obtained by the CPO-catalyzed oxidations. The sulfoxides listed in Table 1 were predominantly (R_SS_C) configurations, and illustrate a consistent excess of the upfield-signal intensity in the stereochemically dependent pairs of signals at ca. δ 24 (C-3), 37 (S(O)-CH_3), and 50 (C-4). Confirmation that the diastereomeric pairs of all the sulfoxide products (possessing both (R_SS_C) and (S_SS_C) configurations) exhibited base-line peak separation in the ^{13}C NMR spectra was obtained from the ^{13}C NMR spectra of such products obtained using hydrogen peroxide as the oxidation reagent for the *N*-MOC ester derivatives.

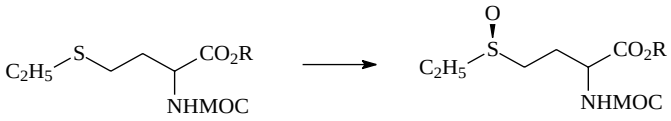
The results obtained from CPO-catalyzed oxidation of D-methionine derivatives are listed in Table 2. The configuration at sulfur was again determined to be (R_S) by ^{13}C NMR spectral analysis of the α and β carbons to the sulfoxide sulfur (ca. δ 24 (C-3), 37 (S(O)-CH_3), and 50 (C-4)). The predominant (R_SR_C) configurations of the products illustrate a consistent excess of the downfield ^{13}C NMR signals in the stereochemically dependent pairs.

Table 2. CPO-catalyzed oxidations of D-methionine derivatives.



R	R′	Conversion (%)	de (%)	Sulfoxide stereochemistry
CH_3	MOC	42	50	*R*
	t-Boc	6	—	—
C_2H_5	MOC	22	50	*R*
n-C_3H_7	MOC	28	46	*R*
n-C_4H_9	MOC	19	24	*R*
n-C_5H_{11}	MOC	10	0	—

Table 3. CPO-catalyzed oxidations of L-and D-ethionine derivatives.



C-2 configuration	R	Conversion (%)	de (%)	Sulfoxide stereochemistry
L (*S*)	CH_3	30	91	*R*
L (*S*)	C_2H_5	40	80	*R*
D (*R*)	CH_3	8	0	—

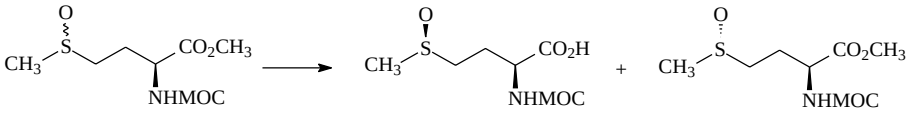
Table 3 illustrates that only the L-ethionine derivatives were effective substrates for CPO-catalyzed oxidation at sulfur. The configurations at sulfur were determined as described above, i.e., using ^{13}C NMR spectral analysis of α carbons to the sulfoxide sulfur (ca. δ 24 (C-3) and 51 (SCH_2CH_3)), and by comparison with the ^{13}C NMR spectra of stereochemically known samples (19). The predominant signal from the (R_SS_C) configurations of both diastereomerically enriched sulfoxides occurs as the upfield component of the stereochemically dependent pair of signals.

The formation of diastereomerically enriched sulfoxides by stereoselective hydrolysis of the methyl ester from diastereomeric *N*-MOC-L-methionine sulfoxides (1:1) is summarized in Table 4.

Of the hydrolytic enzymes that were successful in the stereoselective hydrolysis of this diastereomeric pair, subtilisin Carlsberg was the most efficient and gave both the acid and the unreacted ester products in moderate yield and high diastereomeric excess. Again, configuration at sulfur of both products was determined by ^{13}C NMR spectral analysis of the α and β carbons to the sulfoxide sulfur (ca. δ 24 (C-3), 37 (S(O)-CH_3), and 50 (C-4)), with (R_SS_C) configurations of diastereomerically enriched sulfoxides occurring as the upfield component of the stereochemically dependent signal pair and (S_SS_C) configurations with the predominant peak as the downfield component.

The CPO-catalyzed oxidations of a series of methionine-containing dipeptides are summarized in Table 5. Although the enzyme was capable of the oxidation of several of these substrates in acceptable yields, only one sulfoxide, from the

Table 4. Stereoselective hydrolysis of *N*-MOC-L-methionine methyl ester sulfoxides.



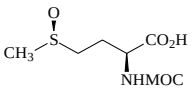
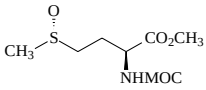
Enzyme	Time (min)	 Yield (%)	de (%)	Sulfoxide stereochemistry	 Yield (%)	de (%)	Sulfoxide stereochemistry
α-Chymotrypsin	10	42	82	*R*	35	80	*S*
Aspergillus sp. protease	5	39	40	*R*	46	28	*S*
Subtilisin Carlsberg	15	41	90	*R*	38	86	*S*
Aspergillus lipase	10	25	0	—	58	0	—

Table 5. CPO-catalyzed oxidations of L-methionine-containing protected dipeptides

N-MOC-Me-ester dipeptide	Yield (%)	de (%)	Sulfoxide stereochemistry
MET-GLY	90	0	—
MET-ALA	80	0	—
MET-LEU	90	0	—
MET-PRO	17	0	—
MET-PHE	<5	0	—
GLY-MET	95	0	—
ALA-MET	90	98	*R*
LEU-MET	16	0	—
PRO-MET	33	5	—
PHE-MET	<5	0	—

methyl ester of *N*-MOC, ALA-MET, was produced in a stereochemically acceptable form.

In the absence of a stereochemical standard for this product, the assignment of the configuration at sulfur was again based on ^{13}C NMR spectral analysis of the α and β carbons to the sulfoxide sulfur (ca. δ 24 (C-3), 37 (S(O)-CH_3), and 50 (C-4)), with the assumption that the (R_SS_C) configuration of *N*-MOC-ALA-MET methyl ester sulfoxide presumably resulted in signal intensities consistent with the standard L-methionine products. Nevertheless, the description of the methionine sulfoxide configuration of the dipeptide product as (R_SS_C) must be considered tentative.

The removal of both *N*-MOC- and methyl ester protecting groups from the diastereomerically enriched L-methionine, D-methionine, and L-ethionine sulfoxide enzyme-catalyzed products was performed in moderate yield by routine hydrolysis with barium hydroxide, generating the appropriate free amino-acid sulfoxides.

Experimental

Materials and methods

Melting points were determined on a Kofler hot stage and are uncorrected. The ^{1}H NMR spectra were recorded on a Bruker Advance series 300 spectrometer in $CDCl_3$ using residual $CHCl_3$ as the internal standard or CD_3OD using CH_3OH as the internal standard unless otherwise stated; chemical shifts are reported in ppm (δ) and the signals quoted as s (singlet), d (doublet), t (triplet), q (quartet), or m (multiplet). The ^{13}C NMR spectra were recorded at 75 MHz on the same spectrometer in $CDCl_3$, CD_3OD, or D_2O. Diastereomeric excess (de) was determined by ^{13}C NMR analysis of signals α and β to sulfur. Mass spectra were obtained using a Kratos 1S spectrometer. Optical rotations were recorded at ambient temperature in the stated solvent using a Rudolph Autopol 3 polarimeter. TLC was performed on Merck silica gel F_{254} plates, 0.2 mm, and column chromatography used Merck silica gel 9385, 230–400 mesh.

Chloroperoxidase from *Caldariomyces fumago* was obtained from the Sigma Chemical Co. (catalogue no. C-0278). α-Chymotrypsin, *Aspergillus* sp. protease, subtilisin Carlsberg, and *Aspergillus* lipase were obtained from Altus Biologics Inc.

Preparation of substrates

N-*Protection*

N-*t*-Boc-L- and D-methionines, *N*-acetyl-L-methionine, and *N*-formyl-L-methionine are commercial samples. *N*-chloroacetyl-L-methionine (24) and *N*-phthaloyl-L-methionine (19) were prepared from the amino acids as previously described.

N-MOC-L- and D-methionines, *N*-MOC-L- and -D- ethionines, and *N*-MOC-dipeptides were prepared by the following standard procedure. A stirred solution of L-methionine (2.08 g) in aqueous sodium hydroxide (40 mL, 2 M) at 0°C was treated with methyl chloroformate (10.4 mL) in 1 mL increments over 1 h. The pH was maintained above 10.5 by addition of 2 M sodium hydroxide as required. After the final aliquot of methyl chloroformate (0.4 mL) was added, the reaction mixture was allowed to reach room temperature and was then washed with an equal volume of ether. The aqueous layer was then cooled to 0°C, acidified to pH 2–3 with 10% HCl, and extracted with ethyl acetate (3 × 100 mL). The extract was dried ($MgSO_4$) and evaporated to yield *N*-MOC-L-methionine (2.34 g, 84%) as an oil. NMR data are listed below. Unless stated otherwise all products gave *m*/*z* MH^+ ions using FAB-NBA spectra.

N-*MOC-L- and D-methionines*

^{1}H NMR ($CDCl_3$) δ: 1.95–2.26 (m, 2H), 2.10 (s, 3H, SCH_3), 2.59 (t, 2H), 3.70 (s, 3H, OCH_3), 4.50 (q, 1H), 5.39

(d, 1H), 6.47 (d, 1H). ^{13}C NMR ($CDCl_3$) δ: 15.6 (SCH_3), 30.2 (SCH_2), 31.9 (CH_2), 53.0 (OCH_3), 53.3 (CH), 157.5 (CO), 176.2 (CO). EI-MS *m/z* (%): 207 (26), 175 (40), 133 (47), 115 (58), 75 (66), 61 (100).

N-*MOC-L- and D- ethionines*

^{1}H NMR ($CDCl_3$) δ: 1.21 (t, 3H), 1.81–2.17 (m, 4H, $2CH_2$) 3.71 (s, 3H, OCH_3), 4.34 (q, 1H), 4.84 (s, 1H), 7.35 (d, 1H). ^{13}C NMR (CD_3OD) δ: 14.1 (CH_3), 25.4 (SCH_2), 27.3 (SCH_2), 31.6 (CH_2), 52.6 (CH), 53.3 (OCH_3), 163.5 (CO), 173.4 (CO).

N-*MOC-MET-GLY*

^{1}H NMR (CD_3OD) δ: 1.8–2.1 (m, 2H, CH_2), 2.08 (s, 3H, SCH_3), 2.55 (m, 2H), 3.34 (s, 3H, OCH_3), 3.67 (br s, 2H), 4.28 (m, 1H), 5.6 (br s, 1H), 6.8 (br s, 1H). ^{13}C NMR (CD_3OD) δ: 14.1 (SCH_3), 30.0, 31.8, 40.9, 54.4, 56.4, 158.1, 171.9, 173.8.

N-*MOC-MET-ALA*

^{1}H NMR (D_2O) δ: 1.22 (d, 3H), 1.88 (m, 2H, CHC*H*$_3$), 2.0 (s, 3H, SCH_3), 2.51 (m, 2H), 3.57 (s, 3H, OCH_3), 4.05 (q, 1H, C*H*CH_3), 4.16 (m, 1H). ^{13}C NMR (D_2O) δ: 14.5, 17.8, 29.7, 31.0, 51.3, 53.0, 54.3, 159.3, 172.6, 173.2.

N-*MOC-MET-LEU*

^{1}H NMR (CD_3OD) δ: 0.75 (d, 3H, CHC*H*$_3$), 0.81 (d, 3H, CHC*H*$_3$), 1.55 (m, 2H), 1.95–2.2 (m, 2H), 2.08 (s, 3H, SCH_3), 2.85 (m, 1H), 3.20 (m, 2H), 3.56 (s, 3H, OCH_3), 4.17 (m, 1H), 4.34 (t, 1H). ^{13}C NMR (D_2O) δ: 13.8, 20.9 (2), 22.6, 24.5, 30.5, 39.9, 52.6, 52.9, 158.5, 173.8, 175.1.

N-*MOC-MET-PRO*

^{1}H NMR (CD_3OD) δ: 1.88–2.15 (m, 6H), 2.09 (s, 3H, SCH_3), 2.25 (m, 2H), 2.60 (t, 2H), 3.62 (s, 3H, OCH_3), 4.49 (m, 1H), 4.62 (m, 1H). ^{13}C NMR (CD_3OD) δ: 14.3, 24.9, 29.0, 29.8, 31.1, 51.7, 59.4, 78.5, 156.8, 172.1, 173.0.

N-*MOC-MET-PHE*

^{1}H NMR ($CDCl_3$) δ: 1.8–2.0 (m, 2H), 2.08 (s, 3H, SCH_3), 2.50 (m, 2H), 3.03 (m, 2H), 3.64 (s, 3H, OCH_3), 4.22 (m, 1H), 4.72 (m, 1H), 7.23 (m, 5H). ^{13}C NMR (CD_3OD) δ: 14.2, 29.9, 31.9, 37.2, 51.7, 54.1, 54.3, 126.9, 128.5, 129.3, 137.0, 155.9, 172.2, 173.2.

N-*MOC-GLY-MET*

^{1}H NMR (CD_3OD) δ: 1.9–2.2 (m, 2H), 2.07 (s, 3H, SCH_3), 2.5–2.7 (m, 4H), 3.68 (s, 3H, OCH_3), 4.57 (q, 1H). ^{13}C NMR (CD_3OD) δ: 14.2, 30.1, 31.1, 43.8, 51.5, 51.8, 157.7, 171.5, 171.8.

N-*MOC-ALA-MET*

^{1}H NMR (CD_3OD) δ: 1.32 (d, 3H), 1.9–2.2 (m, 2H), 2.09 (s, 3H, SCH_3), 2.50 (m, 2H), 3.66 (s, 3H, OCH_3), 4.12 (m, 1H), 4.58 (m, 1H). ^{13}C NMR (CD_3OD) δ: 14.2, 17.3, 30.1, 31.3, 50.9, 51.1, 51.6, 157.0, 173.8, 174.7.

N-*MOC-LEU-MET*

^{1}H NMR (CD_3OD) δ: 0.96 (d, 3H), 0.98 (d, 3H), 1.58 (m, 2H), 1.70 (m, 1H), 1.9–2.2 (m, 2H), 2.05 (s, 3H, SCH_3), 2.55 (m, 2H), 3.66 (s, 3H, OCH_3), 4.16 (m, 1H), 4.57 (m, 1H). ^{13}C NMR (CD_3OD) δ: 14.2, 20.9, 22.3, 24.7, 30.1, 31.2, 41.0, 51.5, 51.6, 53.8, 156.8, 173.2, 174.6.

N-*MOC-PRO-MET*

^{1}H NMR (CD_3OD) δ: 1.8–2.3 (m, 8H), 2.09 (s, 3H, SCH_3), 2.60 (m, 2H), 3.64 (s, 3H, OCH_3), 4.28 (m, 1H), 4.58 (m, 1H). ^{13}C NMR (CD_3OD) δ: 14.3, 24.4, 30.1, 30.4, 31.1, 51.7, 51.8, 60.2, 60.5, 156.3, 172.6, 174.3.

N-*MOC-PHE-MET*

^{1}H NMR (CD_3OD) δ: 1.82–2.12 (m, 2H), 2.04 (s, 3H, SCH_3), 2.52 (m, 2H), 2.88 and 3.14 (m, total 2H), 3.58 (s, 3H, OCH_3), 4.35 (m, 1H), 4.60 (m, 1H), 7.07 (d, 1H, NH), 7.24 (m, 5H), 8.40 (d, 1H, NH). ^{13}C NMR (CD_3OD) δ: 14.2, 30.0, 31.0, 38.0, 51.6, 51.7, 56.6, 126.7, 128.4 (2), 129.3 (2), 137.5, 157.3, 172.5, 173.4.

Esterification

The esters of *N*-protected amino acids and dipeptides were all obtained by the following standard procedure using the appropriate alcohol. *N*-MOC-L-methionine (2.34 g) was dissolved in methanol (50 mL) and a catalytic amount of hydrochloric acid added to adjust the solution to pH 3.0. The solution was heated under reflux for 12 h, then evaporated. Water (50 mL) was added and the product extracted with ethyl acetate. The solution was washed with saturated $NaHCO_3$, dried, and evaporated to give *N*-MOC-L-methionine methyl ester as an oil (2.03 g, 81%). NMR data are listed below. Unless listed otherwise, all products gave *m/z* MH^+ ions using FAB-NBA spectra.

N-*Acetyl-L-methionine methyl ester*

^{1}H NMR (CD_3OD) δ: 1.9–2.2 (m, 2H), 1.94 (s, 3H), 2.08 (s, 3H, SCH_3), 2.42 (m, 2H), 3.60 (s, 3H, OCH_3), 4.58 (m, 1H), 6.72 (d, 1H, NH). ^{13}C NMR (CD_3OD) δ: 14.3, 21.4, 30.1, 31.0, 51.8, 51.9, 172.4, 173.0.

N-*Chloroacetyl-L-methionine methyl ester*

^{1}H NMR ($CDCl_3$) δ: 1.98–2.25 (m, 2H), 2.1 (s, 3H, SCH_3), 2.50 (t, 2H), 3.78 (s, 3H, OCH_3), 4.08 (2H, s), 4.70 (m, 1H). ^{13}C NMR ($CDCl_3$) δ: 15.9, 30.2, 31.7, 42.8, 52.2, 53.1, 166.3, 172.1.

N-*Formyl-L-methionine methyl ester*

^{1}H NMR ($CDCl_3$) δ: 1.9–2.2 (m, 2H), 2.05 (s, 3H, SCH_3), 2.52 (t, 2H), 3.73 (s, 3H, OCH_3), 4.70 (m, 1H), 4.80 (s, 1H, NH), 8.10 (s, 1H). ^{13}C NMR ($CDCl_3$) δ: 15.9, 30.2, 32.0, 50.6, 53.1, 161.1, 172.4.

N-*MOC-L- and D-methionine methyl esters*

^{1}H NMR ($CDCl_3$) δ: 1.92–2.24 (m, 2H), 2.10 (s, 3H, SCH_3), 2.54 (t, 2H), 3.70 (s, 3H, OCH_3), 3.79 (s, 3H, OCH_3), 4.51 (q, 1H), 5.35 (d, 1H, NH). ^{13}C NMR ($CDCl_3$) δ: 15.0, 29.6, 31.4, 52.0, 52.1, 52.8, 156.4, 172.4.

N-*MOC-L- and D-methionine ethyl esters*

^{1}H NMR ($CDCl_3$) δ: 1.26 (t, 3H), 1.90–2.2 (m, 2H), 2.11 (s, 3H, SCH_3), 2.52 (m, 2H), 3.67 (s, 3H, OCH_3), 4.20 (2H, q), 4.45 (m, 1H), 5.36 (m, 1H, NH). ^{13}C NMR ($CDCl_3$) δ: 14.5, 15.8, 30.2, 32.4, 52.7, 53.6, 62.0, 156.9, 172.4.

N-*MOC-L- and D-methionine n-propyl esters*

^{1}H NMR ($CDCl_3$) δ: 0.92 (t, 3H), 1.67 (sextet, 2H), 1.90–2.15 (m, 2H), 2.08 (s, 3H, SCH_3), 2.51 (t, 2H), 3.68 (s, 3H, OCH_3), 4.08 (q, 2H), 4.42 (m, 1H), 5.39 (m, 1H, NH).

^{13}C NMR ($CDCl_3$) δ: 10.7, 15.8, 22.3, 30.3, 32.5, 52.7, 53.6, 67.6, 156.9, 172.5.

N-*MOC-L- and D-methionine n-butyl esters*

^{1}H NMR ($CDCl_3$) δ: 0.92 (t, 3H), 1.33 (sextet, 2H), 1.60 (sextet, 2H), 1.87–2.15 (m, 2H), 2.07 (s, 3H, SCH_3), 2.50 (t, 2H), 3.66 (s, 3H, OCH_3), 4.10 (q, 2H), 4.41 (m, 1H), 5.40 (m, 1H, NH). ^{13}C NMR ($CDCl_3$) δ: 14.0, 15.8, 19.4, 30.3, 30.9, 32.5, 52.7, 53.6, 65.8, 156.9, 172.5.

N-*MOC-L- and D-methionine n-pentyl esters*

^{1}H NMR ($CDCl_3$) δ: 0.87 (t, 3H), 1.30 (m, 6H), 1.62 (m, 2H), 1.85–2.18 (m, 2H), 2.07 (s, 3H, SCH_3), 2.49 (m, 2H), 3.64 (s, 3H, OCH_3), 4.08 (q, 2H), 4.40 (m, 1H), 5.38 (m, 1H, NH). ^{13}C NMR ($CDCl_3$) δ: 14.3, 15.8, 22.6, 28.3, 28.6, 30.3, 32.5, 52.7, 53.6, 66.1, 156.9, 172.5.

N-*MOC-L- and D- ethionine methyl esters*

^{1}H NMR (CD_3OD) δ: 1.21 (t, 3H), 1.80–2.15 (m, 2H), 2.52 (m, 4H), 3.62 (s, 3H, OCH_3), 3.72 (s, 3H, OCH_3), 4.33 (m, 1H), 7.35 (m, 1H, NH). ^{13}C NMR (CD_3OD) δ: 14.1, 25.1, 27.5, 31.6, 51.7, 51.8, 53.3, 158.3, 173.4.

N-*MOC-L-ethionine ethyl ester*

^{1}H NMR (CD_3OD) δ: 1.22 (t, 3H), 1.30 (t, 3H), 1.90–2.16 (m, 2H), 2.55 (m, 4H), 3.68 (s, 3H, OCH_3), 4.21 (q, 2H), 4.35 (m, 1H). ^{13}C NMR (CD_3OD) δ: 13.5, 14.1, 25.5, 27.5, 31.6, 51.8, 71.6, 158.3, 173.0.

N-*MOC-MET-GLY methyl ester*

^{1}H NMR ($CDCl_3$) δ: 1.86–2.18 (m, 2H), 2.07 (s, 3H, SCH_3), 2.56 (m, 2H), 3.64 (s, 3H, OCH_3), 3.76 (s, 3H, OCH_3), 4.02 (m, 2H), 4.42 (m, 1H), 5.76 (m, 1H, NH), 6.98 (m, 1H, NH). ^{13}C NMR (CD_3OD) δ: 15.1, 31.0, 32.8, 41.8, 52.6, 52.8, 55.4, 159.1, 171.6, 175.1. EI-MS *m/z* (%): 278 (8), 246 (11), 204 (60), 172 (22), 90 (64), 61 (100),

N-*MOC-MET-ALA methyl ester*

^{1}H NMR (D_2O) δ: 1.38 (d, 3H), 1.82–2.04 (m, 2H), 2.01 (s, 3H, SCH_3), 2.50 (m, 2H), 3.57 (s, 3H, OCH_3), 3.68 (s, 3H, OCH_3), 4.12 (q, 1H), 4.33 (m, 1H). ^{13}C NMR (D_2O) δ: 14.5, 16.3, 29.5, 31.0, 49.1, 53.0, 53.3, 54.2, 158.9, 174.6, 175.2.

N-*MOC-MET-LEU methyl ester*

^{1}H NMR (CD_3OD) δ: 0.74 (d, 3H, CHC*H*$_3$), 0.81 (d, 3H, CHC*H*$_3$), 1.55 (m, 2H), 1.95–2.2 (m, 2H), 2.10 (s, 3H, SCH_3), 2.85 (m, 1H), 3.24 (m, 2H), 3.55 (s, 3H, OCH_3), 3.60 (s, 3H, OCH_3), 4.17 (m, 1H), 4.35 (t, 1H). ^{13}C NMR (D_2O) δ: 13.6, 20.9 (2), 22.6, 24.5, 30.5, 39.9, 52.6, 52.9, 53.3, 158.8, 173.8, 175.1.

N-*MOC-MET-PRO methyl ester*

^{1}H NMR (CD_3OD) δ: 1.88–2.15 (m, 6H), 2.10 (s, 3H, SCH_3), 2.25 (m, 2H), 2.60 (t, 2H), 3.62 (s, 3H, OCH_3), 3.80 (s, 3H, OCH_3), 4.49 (m, 1H), 4.62 (m, 1H). ^{13}C NMR (CD_3OD) δ: 14.3, 24.9, 29.0, 29.8, 30.1, 31.1, 51.7 (2), 59.4, 78.5, 158.1, 172.1, 173.0.

N-*MOC-MET-PHE methyl ester*

^{1}H NMR (CD_3OD) δ: 1.7–2.1 (m, 2H), 2.02 (s, 3H, SCH_3), 2.48 (m, 2H), 3.05 (m, 2H), 3.64 (s, 3H, OCH_3), 3.70 (s, 3H, OCH_3), 4.22 (m, 1H), 4.72 (m, 1H), 7.25 (m, 5H). ^{13}C NMR (CD_3OD) δ: 14.2, 29.9, 31.9, 37.2, 51.7 (2), 54.1, 54.3, 126.9, 128.5, 129.3, 137.0, 157.9, 172.2, 173.2.

N-*MOC-GLY-MET methyl ester*

^{1}H NMR (CD_3OD) δ: 1.94–2.2 (m, 2H), 2.07 (s, 3H, SCH_3), 2.56 (m, 2H), 3.67 (s, 3H, OCH_3), 3.72 (s, 3H, OCH_3), 4.61 (m, 1H). ^{13}C NMR (CD_3OD) δ: 14.2, 30.0, 30.9, 43.7, 51.6, 51.8, 158.7, 171.3, 172.7.

N-*MOC-ALA-MET methyl ester*

^{1}H NMR (CD_3OD) δ: 1.33 (d, 3H), 1.9–2.18 (m, 2H), 2.07 (s, 3H, SCH_3), 3.64 (s, 3H, OCH_3), 3.73 (s, 3H, OCH_3), 4.16 (m, 1H), 4.59 (m, 1H). ^{13}C NMR (CD_3OD) δ: 14.2, 17.3, 30.1, 31.3, 50.9, 51.1, 51.6 (2), 157.9, 173.8, 174.7.

N-*MOC-LEU-MET methyl ester*

^{1}H NMR (CD_3OD) δ: 0.92 (d, 3H), 0.94 (d, 3H), 1.54 (m, 2H), 1.70 (m, 1H), 1.9–2.18 (m, 2H), 2.05 (s, 3H, SCH_3), 2.52 (m, 2H), 3.60 (s, 3H, OCH_3), 3.71 (s, 3H, OCH_3), 4.12 (m, 1H), 4.58 (m, 1H), 7.0 (d, 1H, NH), 8.35 (d, 1H, NH). ^{13}C NMR (CD_3OD) δ: 14.2, 21.0, 22.3, 24.8, 30.0, 30.9, 41.0, 51.6, 51.7, 53.7, 158.1, 172.6, 174.7.

N-*MOC-PRO-MET methyl ester*

^{1}H NMR (CD_3OD) δ: 1.82–2.3 (m, 8H), 2.1 (s, 3H, SCH_3), 2.60 (m, 2H), 3.64 (s, 3H, OCH_3), 3.73 (s, 3H, OCH_3), 4.28 (m, 1H), 4.58 (m, 1H). ^{13}C NMR (CD_3OD) δ: 14.3, 24.4, 30.1, 30.4, 31.1, 51.7, 51.8, 52.2, 60.2, 60.5, 156.0, 172.6, 174.3.

N-*MOC-PHE-MET methyl ester*

^{1}H NMR (CD_3OD) δ: 1.82–2.1 (m, 2H), 2.06 (s, 3H, SCH_3), 2.52 (m, 2H), 2.87 (m, 1H), 3.14 (m, 1H), 3.58 (s, 3H, OCH_3), 3.70 (s, 3H, OCH_3), 4.35 (m, 1H), 4.60 (m, 1H), 7.25 (m, 5H), 8.39 (d, 1H, NH). ^{13}C NMR (CD_3OD) δ: 14.2, 30.0, 31.0, 38.0, 51.6, 51.7, 51.8, 56.6, 126.7, 128.4, 129.3, 137.4, 157.9, 172.5, 173.4.

Preparation of standard protected sulfoxides

Standard diastereomeric sulfoxide mixtures of esters of *N*-protected amino acids and dipeptides were prepared using the following representative procedure: *N*-MOC-L-methionine methyl ester (2.03 g) was dissolved in methanol (40 mL) in an Erlenmeyer flask (100 mL). In a separate flask, hydrogen peroxide (1.13 g of 30% aqueous solution, 1.1 molar equivalents) was dissolved in methanol (15 mL). Both solutions were cooled to –20°C, then mixed and allowed to stand at –20°C overnight. The solvent was then removed by vacuum to give a quantitative yield of a 1:1 diastereomeric mixture of *N*-MOC-L-methionine methyl ester sulfoxides (oil).

N-*Acetyl-L-methionine methyl ester, sulfoxide*

^{1}H NMR (CD_3OD) δ: 1.94 (s, 3H, $COCH_3$), 2.2–2.4 (m, 2H), 2.52 (s, 3H, $SOCH_3$), 2.8–3.0 (m, 2H), 3.60 (s, 3H, OCH_3), 4.62 (m, 1H). ^{13}C NMR (CD_3OD) δ: 21.4, 24.8, 37.2/37.3, 49.9, 51.6, 52.0, 172.0, 173.0.

N-*Chloroacetyl-L-methionine methyl ester, sulfoxide*

^{1}H NMR (CD_3OD) δ: 2.15 (m, 1H), 2.35 (m, 1H), 2.76 (s, 3H, $SOCH_3$), 2.88 (m, 2H), 3.78 (s, 3H, OCH_3), 4.10 (s, 2H), 4.60 (m, 1H). ^{13}C NMR (CD_3OD) δ: 24.7, 37.1/37.2, 41.9, 49.9, 52.0, 52.6, 166.8, 171.4.

N-*Formyl-L-methionine methyl ester, sulfoxide*
^{1}H NMR (CD_3OD) δ: 2.0–2.3 (m, 2H), 2.62 (s, 3H, $SOCH_3$), 2.80 (m, 2H), 3.80 (s, 3H, OCH_3), 4.80 (m, 1H), 8.22 (s, 1H). ^{13}C NMR (CD_3OD) δ: 25.8, 38.1/38.2, 50.71/50.75, 51.0, 53.1, 163.7, 172.4.

N-*MOC-L- and D-methionine methyl esters, sulfoxides*
^{1}H NMR (CD_3OD) δ: 2.2–2.6 (m, 2H), 2.66 (s, 3H, $SOCH_3$), 2.90 (m, 2H), 3.62 (s, 3H, OCH_3), 3.73 (s, 3H, OCH_3), 4.30 (m, 1H). ^{13}C NMR (CD_3OD) δ: 24.82/24.94, 37.1/37.25, 49.96/50.06, 52.0, 52.1, 53.2, 158.2, 172.4.

N-*MOC-L- and D-methionine ethyl esters, sulfoxides*
^{1}H NMR (CD_3OD) δ: 1.28 (t, 3H), 2.2–2.58 (m, 2H), 2.60 (s, 3H, $SOCH_3$), 2.94 (m, 2H), 3.68 (s, 3H, OCH_3), 4.18 (q, 2H), 4.30 (m, 1H). ^{13}C NMR (CD_3OD) δ: 13.4, 24.72/24.95, 37.12/37.27, 48.8, 50.1, 51.73/51.83, 61.7, 158.3, 172.0.

N-*MOC-L- and D-methionine n-propyl esters, sulfoxides*
^{1}H NMR (CD_3OD) δ: 0.94 (t, 3H), 1.67 (m, 2H), 2.2–2.5 (m, 2H), 2.68 (s, 3H, $SOCH_3$), 2.80 (m, 2H), 3.62 (s, 3H, OCH_3), 4.08 (q, 2H), 4.44 (m, 1H), 7.32 (m, 1H, NH). ^{13}C NMR (CD_3OD) δ: 9.8, 22.3, 24.70/24.94, 37.11/37.27, 51.71/51.80, 52.7, 53.6, 67.1, 158.3, 172.0.

N-*MOC-L- and D-methionine n-butyl esters, sulfoxides*
^{1}H NMR (CD_3OD) δ: 0.95 (t, 3H), 1.40 (sextet, 2H), 1.60 (sextet, 2H), 2.2–2.6 (m, 2H), 2.68 (s, 3H, $SOCH_3$), 2.90 (t, 2H), 3.70 (s, 3H, OCH_3), 4.13 (q, 2H), 4.32 (m, 1H). ^{13}C NMR (CD_3OD) δ: 13.0, 19.4, 24.68/24.92, 32.5, 37.12/37.27, 50.2, 51.7, 53.4, 65.4, 158.3, 172.0.

N-*MOC-L- and D-methionine n-pentyl esters, sulfoxides*
^{1}H NMR (CD_3OD) δ: 0.87 (t, 3H), 1.30 (m, 6H), 1.62 (m, 2H), 2.22–2.5 (m, 2H), 2.63 (s, 3H, $SOCH_3$), 2.65 (m, 2H), 3.62 (s, 3H, OCH_3), 4.15 (q, 2H), 4.33 (m, 1H). ^{13}C NMR (CD_3OD) δ: 14.2, 22.3, 24.69/24.93, 28.4, 31.0, 37.13/37.29, 50.10/50.16, 51.7, 53.4, 65.7, 157.6, 172.0.

N-*MOC-L- and D- ethionine methyl esters, sulfoxides*
^{1}H NMR (CD_3OD) δ: 1.33 (t, 3H), 2.11 (m, 1H), 2.33 (m, 1H), 2.72–2.97 (m, 4H), 3.64 (s, 3H, OCH_3), 3.77 (s, 3H, OCH_3), 4.34 (m, 1H). ^{13}C NMR (CD_3OD) δ: 6.2, 24.89/25.12, 45.3, 51.93/52.11, 52.3, 53.29, 53.62, 158.2, 172.5.

N-*MOC-L-ethionine ethyl ester, sulfoxides*
^{1}H NMR (CD_3OD) δ: 1.22 (t, 3H), 1.30 (t, 3H), 2.15 (m, 1H), 2.33 (m, 1H), 2.7–2.95 (m, 4H), 3.68 (s, 3H, OCH_3), 4.20 (q, 2H), 4.36 (m, 1H). ^{13}C NMR (CD_3OD) δ: 6.1, 13.5, 24.88/25.11, 45.26/45.35, 51.71/51.81, 53.4, 53.5, 71.6, 158.3, 172.0.

N-*MOC-MET-GLY methyl ester, sulfoxides*
^{1}H NMR (CD_3OD) δ: 2.0–2.32 (m, 2H), 2.66 (s, 3H, $SOCH_3$), 2.92 (t, 2H), 3.67 (s, 3H, OCH_3), 3.73 (s, 3H, OCH_3), 3.95 (q, 2H), 4.30 (m, 1H). ^{13}C NMR (CD_3OD) δ: 25.5, 37.08/37.14, 40.8, 49.63/49.77, 51.6, 51.8, 53.9, 157.9, 171.8, 173.0.

N-*MOC-MET-ALA methyl ester, sulfoxides*
^{1}H NMR (CD_3OD) δ: 1.30 (d, 3H), 1.98 (m, 1H), 2.10 (m, 1H), 2.58 (s, 3H, $SOCH_3$), 2.75–2.90 (m, 2H), 3.55 (s, 3H, OCH_3), 3.60 (s, 3H, OCH_3), 4.16 (d, 1H), 4.30 (q, 1H). ^{13}C NMR (CD_3OD) δ: 16.0, 24.71/24.79, 36.69/36.78, 49.65/49.74, 52.90, 53.07, 53.64/53.86, 158.2, 172.3, 173.5.

N-*MOC-MET-LEU methyl ester, sulfoxides*
^{1}H NMR (CD_3OD) δ: 0.75 (d, 3H), 0.81 (d, 3H), 1.50–1.60 (m, 3H), 2.58 (s, 3H, $SOCH_3$), 2.75–2.90 (m, 2H), 3.53 (s, 3H, OCH_3), 3.62 (s, 3H, OCH_3), 4.16 (d, 1H), 4.34 (q, 1H). ^{13}C NMR (CD_3OD) δ: 20.6, 22.3 (2), 24.52/24.68, 37.06/37.13, 40.0, 50.6, 51.3, 51.8, 53.4, 157.9, 172.4, 173.5.

N-*MOC-MET-PRO methyl ester, sulfoxides*
^{1}H NMR (CD_3OD) δ: 1.88–2.15 (m, 4H), 2.40–2.60 (m, 2H), 2.60 (t, 2H), 2.70 (s, 3H, $SOCH_3$), 2.90 (m, 2H), 3.62 (s, 3H, OCH_3), 3.80 (s, 3H, OCH_3), 4.50 (m, 1H), 4.62 (m, 1H). ^{13}C NMR (CD_3OD) δ: 24.8, 24.9, 29.0, 31.1, 37.23/37.34, 51.7 (2), 51.8, 59.4, 78.4, 158.1, 171.0, 172.9.

N-*MOC-MET-PHE methyl ester, sulfoxides*
^{1}H NMR (CD_3OD) δ: 2.2–2.6 (m, 2H), 2.62 (s, 3H, $SOCH_3$), 2.88 (m, 2H), 3.06 (m, 2H), 3.65 (s, 3H, OCH_3), 3.72 (s, 3H, OCH_3), 4.22 (m, 1H), 4.72 (m, 1H), 7.25 (m, 5H). ^{13}C NMR (CD_3OD) δ: 25.56/25.61, 37.2, 37.31/37.39, 49.87/49.92, 51.7 (2), 54.1, 54.3, 126.9, 128.5, 129.3, 136.8, 157.9, 172.2, 173.3.

N-*MOC-GLY-MET methyl ester, sulfoxides*
^{1}H NMR (CD_3OD) δ: 2.16 (m, 1H), 2.36 (m, 1H), 2.68 (s, 3H, $SOCH_3$), 2.77–2.97 (m, 2H), 3.68 (s, 3H, OCH_3), 3.78 (s, 3H, OCH_3), 3.80 (2H, s), 4.22 (m, 1H), 4.60 (m, 1H). ^{13}C NMR (CD_3OD) δ: 24.8, 37.16/37.23, 43.8, 49.9, 51.3, 51.7/51.8, 52.0, 158.7, 171.7, 171.8.

N-*MOC-ALA-MET methyl ester, sulfoxides*
^{1}H NMR (CD_3OD) δ: 1.35 (d, 3H), 2.10 (m, 1H), 2.36 (m, 1H), 2.68 (s, 3H, $SOCH_3$), 2.87 (m, 2H), 3.65, (s, 3H, OCH_3), 3.74 (s, 3H, OCH_3), 4.10 (m, 1H), 4.60 (m, 1H). ^{13}C NMR (CD_3OD) δ: 17.1, 24.67/24.90, 37.18/37.27, 49.9, 50.8, 51.57/51.69, 52.0, 157.9, 171.6, 174.9.

N-*MOC-LEU-MET methyl ester, sulfoxides*
^{1}H NMR (CD_3OD) δ: 0.92 (d, 3H), 0.94 (d, 3H), 1.54 (m, 2H), 1.70 (m, 1H), 2.2–2.45 (m, 2H), 2.66 (s, 3H, $SOCH_3$), 2.82 (m, 2H), 3.63 (s, 3H, OCH_3), 3.73 (s, 3H, OCH_3), 4.15 (m, 1H), 4.58 (m, 1H). ^{13}C NMR (CD_3OD) δ: 21.6, 22.0, 25.5, 25.7, 38.14/38.20, 42.0, 50.9, 51.6, 51.7, 54.6, 159.1, 173.6, 175.6.

N-*MOC-PRO-MET methyl ester, sulfoxides*
^{1}H NMR (CD_3OD) δ: 1.82–2.3 (m, 6H), 2.68 (s, 3H, $SOCH_3$), 2.80 (m, 2H), 3.62 (s, 3H, OCH_3), 3.73 (s, 3H, OCH_3), 4.30 (m, 1H), 4.58 (m, 1H). ^{13}C NMR (CD_3OD) δ: 23.5, 24.37/24.46, 30.4, 37.19/37.23, 49.97, 51.4, 51.8, 52.2, 60.2, 60.5, 156.3, 172.6, 174.4.

N-*MOC-PHE-MET methyl ester, sulfoxides*
^{1}H NMR (CD_3OD) δ: 2.25–2.60 (m, 2H), 2.66 (s, 3H, $SOCH_3$), 2.88 (m, 2H), 2.87 (m, 1H), 3.14 (m, 1H), 3.63 (s, 3H, OCH_3), 3.75 (s, 3H, OCH_3), 4.45 (m, 1H), 4.60 (m, 1H), 7.25 (m, 5H). ^{13}C NMR (CD_3OD) δ: 24.62/24.76, 37.51/37.60, 38.0, 50.08/50.16, 51.6 (2), 51.8, 56.8, 126.6, 128.4, 129.5, 137.6, 158.3, 172.3, 173.6.

Biotransformation procedures

Chloroperoxidase-catalyzed oxidations

The reactions were carried out using the following standard procedure: *N*-MOC-L-methionine methyl ester (33 mg, 0.15 mM) was dissolved in disodium citrate buffer (16.6 mL, 0.1 M, pH 5.0) in a 50 mL flask. The solution was stirred slowly at room temperature, and chloroperoxidase (2000 units) was added. A solution of hydrogen peroxide (56.6 μL of 30% aqueous solution) in disodium citrate buffer (1.6 mL, 0.1 M, pH 5.0) was then added via a syringe pump over 70 min, with addition occurring directly below the surface level of the reaction mixture. The reaction was stirred for a further 5 min, quenched by addition of aqueous saturated sodium sulfite (0.5 mL), and then continuously extracted with dichloromethane for 3 d. Evaporation of the extract gave a quantitative recovery of amino acid material, which was directly analyzed by ^{1}H and ^{13}C NMR. Configuration at sulfur of *N*-MOC-L-methionine ester sulfoxides was identified by ^{13}C NMR analysis.

Ester hydrolysis of N-MOC-L-methionine methyl ester sulfoxides

The diastereomeric mixture of chemically prepared *N*-MOC-L-methionine methyl ester sulfoxide (100 mg) was dissolved in phosphate buffer (0.1 M, pH 7.0, 20 mL), and then the stirred solution was treated with the appropriate Altus Chiroclec® enzyme (100 mg for each of α-chymotrypsin, *Aspergillus* sp. protease, subtilisin Carlsberg, or *Aspergillus* lipase in four separation experiments), each suspended in phosphate buffer (5 mL). The resulting reaction mixture was then stirred at room temperature for 5–60 min, monitored by TLC (silica gel, EtOAc–MeOH–AcOH, 79:20:1), and on ca. 50% completion the reaction was quenched by the addition of ethanol (5 mL) and then filtered. The filtrate was acidified to pH 3 by the addition of 10% HCl, then continuously extracted with dichloromethane for 3 d. Evaporation of the extract gave a quantitative recovery of amino acid material, which was directly analyzed by ^{1}H and ^{13}C NMR, and separated by chromatography using silica gel with 10% gradient elution (EtOAc–MeOH, 80:20 to MeOH). Spectral data are quoted above for *N*-MOC-L-methionine methyl ester sulfoxide and *N*-MOC-L-methionine sulfoxide products. Configuration at sulfur of *N*-MOC-L-methionine methyl ester sulfoxides was identified by ^{13}C NMR analysis.

Removal of *N*-MOC and ester-protecting groups

The following standard reaction procedure was used: *N*-MOC-L-methionine methyl ester sulfoxide (120 mg), prepared by CPO-catalyzed oxidation of *N*-MOC-L-methionine methyl ester, was dissolved in a mixture of distilled water (4 mL) and methanol (1 mL). Barium hydroxide (300 mg) was added, and the mixture stirred and heated at reflux for 4 h. Water (10 mL) was added and the mixture was cooled and adjusted to pH 5.75 with sulfuric acid (0.1 M), filtered (celite), and evaporated to yield a semi-solid (100 mg), which was then dissolved in distilled water (1 mL), filtered, and the filtrate diluted with acetone (7 mL). The resulting cloudy mixture was refrigerated overnight to produce crystals of L-methionine sulfoxide (62 mg, 76%), identified by ^{13}C NMR and other analytical comparisons with authentic samples (19) as (R_SS_C) material; mp230–235°C (lit. (25, 26) mp 239°C). $[\alpha]_D$ –75 (c = 0.5, H_2O) (lit. (25) $[\alpha]_D$ –77 (c = 0.5, H_2O)).

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References

1. J.O. Konz, J. King, and C.L. Cooney. Biotechnol. Prog. **14**, 393 (1998).
2. J. Moskovitz, B.S. Berlett, J.M. Poston, and E.R. Stadtman. Proc. Natl. Acad. Sci. **94**, 9585 (1997).
3. M.A. Ciorba, S.H. Heinemann, H. Weissbach, N. Brot, and T. Hoshi. Proc. Natl. Acad Sci. **94**, 9932 (1997).
4. B. Garner, P.K. Witting, A.R. Waldeck, J.K. Christinson, M. Raftery, and R. Stocker. J. Biol. Chem. **273**, 6080 (1998).
5. B. Garner, A.R. Waldeck, P.K. Witting, K.-A. Rye, and R. Stocker. J. Biol. Chem. **273**, 6088 (1998).
6. C.S. Spar, L.O. Narhi, J. Speakman, S. Hieng, and Y.-R. Hsu. Tech. Protein Chem. VIII. 10th symposium of the Protein Society. Academic Press, San Diego. 1997. p. 299.
7. T. Huff and E. Hannappel. Anal. Chim. Acta, **352,** 249 (1997).
8. V.S. Sharov and C. Schöneich. Free Rad. Biol. Med. **29**, 986 (2000).
9. Z. Brada, S. Bulba, and J. Cohen. Cancer Res. **35**, 2674 (1975).
10. K.H. Kyung, D.C. Han, and H.P. Fielding. J. Food Sci. **62**, 406 (1997).
11. W. Komatsu, Y. Miura, and K. Yagasaki. Lipids, **33**, 499 (1998).
12. H.W. Chin and R.C. Lindsay. J. Agric. Food Chem. **42**, 1529 (1994).
13. F. Lucas and L. Levenbrook. Biochem. J. **100**, 473 (1966).
14. V.G. de la Rosa, M. Ordonez, and J.M. Llera. Tetrahedron: Asymmetry, **12**, 1615 (2001).
15. H.L. Holland. Chem. Rev. **88**, 473 (1988).
16. H.L. Holland. Nat. Prod. Rep. **18**, 171 (2001).
17. M. Uyeda, S. Takenobu, and M. Hongo. Agric. Biol. Chem. **38**, 1797 (1974).
18. H.L. Holland and F. M. Brown. Tetrahedron: Asymmetry, **9**, 535 (1998).
19. H.L. Holland, P.R. Andreana, and F. M. Brown. Tetrahedron: Asymmetry, **10**, 2833 (1999).
20. R.R. Vargas, E.J.H. Bechara, L. Marzorati, and B. Wladislaw. Tetrahedron: Asymmetry, **10**, 3219 (1999).
21. F. van der Velde, M. Bakker, F. van Rantwijk, G.P. Rai, L.P. Hager, and R.A. Sheldon. J. Mol. Cat. B: Enzymat. **11**, 765 (2001).
22. G.P. Rai, S. Sakai, A.M. Flórez, L. Mogollon, and L.P. Hager. Adv. Synth. Catal. **343**, 658 (2001).
23. I. Koch and M. Keusen. Pharmazie, **53**, 668 (1998).
24. A.F. Cook and D.T. Maichuk. J. Org. Chem. **35**, 1940 (1970).
25. J.P. Greenstein and M. Winitz. Chemistry of the amino acids. Vol. 3. Wiley, New York. 1961. p. 2145.
26. F. Lucas and L. Levenbook. Biochem. J. **100**, 473 (1966).