Biologically Oriented Organic Sulfur Chemistry. 21. Hydrodisulfide of a Penicillamine Derivative and Related Compounds[1]

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Received October 23, 1980

For study of the chemistry of an amino acid derived hydrodisulfide (**10**, $Me_2C(SR)CH(NHAc)CO_2Me$, R = SH), a synthetic route to a hydrodisulfide derivative of *N*-acetyl-DL-penicillamine was developed. Conversion of *N*-acetyl-DL-penicillamine (**3**) to the ester (**4**, R = H) was accomplished with diazomethane. With acetylsulfenyl chloride, **4** gave the acetyl sulfide (**5**, R = Ac) instead of the disulfide (**7**, R = SAc). With (methoxycarbonyl)sulfenyl chloride, **4** gave the corresponding unsymmetrical disulfide (**8**, R = SCO_2Me), which could not be converted to the hydrodisulfide. Conversion of **4** to the sulfenyl iodide (**9**, R = I) and treatment with thioacetic acid regenerated **4**. With acetyl methoxycarbonyl disulfide, however, **4** gave the acetyl disulfide **7**. Methanolysis of **7** then gave the hydrodisulfide **10**. Solutions of **10** in chloroform were unchanged after 6 days at room temperature and 1 day at 55 °C and then consumed the expected amount of iodine. Washing with bicarbonate led to immediate decomposition; neat **10** completely decomposed during storage at −10 °C for 36 h, and the half-life of **10** in HCl–methanol was about 12 h. Treatment of **10** with cyanide ion gave thiocyanate ion, and treatment with 2,4-dinitrochlorobenzene gave a dinitrophenyl derivative [**13**, R = 2,4-$(NO_2)_2C_6H_3S$] identical with **13** prepared from **4** by using 2,4-dinitrobenzenesulfenyl chloride.

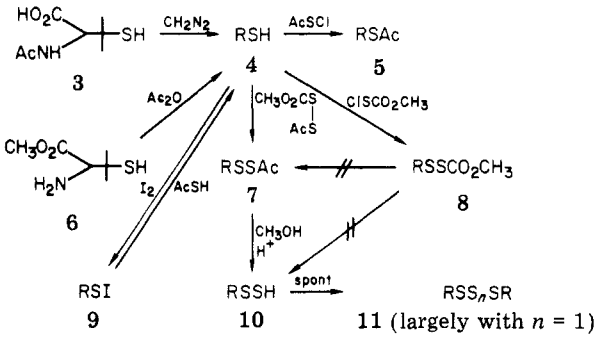
Hydrodisulfides, RSSH, have been proposed as intermediates in several important biological systems as products from the reaction of cysteine or cystine. In one of these, cytochrome P-450 is believed to oxidize phosphorothionates to phosphates [e.g., parathion, *p*-$O_2NC_6H_4OPS(OEt)_2$, to paraoxon, *p*-$O_2NC_6H_4OPO(OEt)_2$] with transfer of the sulfur to an SH group of the P-450.[2] In other systems, illustratively, cystathionase is believed to cleave cystine to give the hydrodisulfide of cysteine.[3] In order to study the chemistry of such compounds, it was desirable to develop a synthesis from a mercaptoamino acid derivative of a hydrodisulfide that might be long-lived enough to permit study. Because of the unprecedented stability of the sulfenyl iodide (**1**)[4] and thionitrite (**2**)[5] derivatives, a penicillamine derivative seemed likely to provide a stable hydrodisulfide.

$(CH_3)_2C(SI)CH[NHOCOCH_2C_6H_5]CONHC_6H_4$-*p*-Cl
1

$(CH_3)_2C(SNO)CH(NHAc)CO_2H$
2

The usual route to hydrodisulfides involves conversion of acetyl disulfide derivatives to hydrodisulfides by HCl-catalyzed methanolysis.[6] In order to minimize the number of reactive functional groups but still to maintain the features characteristic of an amino acid, we chose the methyl ester of *N*-acetyl-DL-penicillamine (**4**) as the thiol for conversion to the corresponding hydrodisulfide. The *N*-acetyl ester **4** has been reported, but no details of synthesis or characterization were given.[7] The required key intermediate was determined to be the acetyl disulfide **7** (Scheme I).

Scheme I[a]



[a] R = $CH_3O_2CCH(NHAc)C(CH_3)_2$

With *N*-acetyl-DL-penicillamine (**3**) as the starting material, three routes to the methyl ester **4** were explored, as well as one route from DL-penicillamine. Attempts to methylate **3** by treatment with either CH_3OH/HCl or $(CH_3O)_2SO/CH_3OH/HCl$[8] gave oily yellow products with no NMR absorption for CH_3CO at δ 2. Both products were soluble in acid and probably were largely the thiazoline formed by cyclization involving the SH group and the

(1) (a) Paper 20: Field, L.; Gallo, A. A. *J. Fluorine Chem.* **1980**, *15*, 29. (b) Presented at the Southeast-Southwest Regional Meeting of the American Chemical Society, New Orleans, LA, Dec 10–13, 1980.

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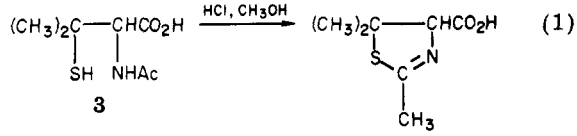
amide carbonyl (eq 1). Methylation of **3** in the THF with



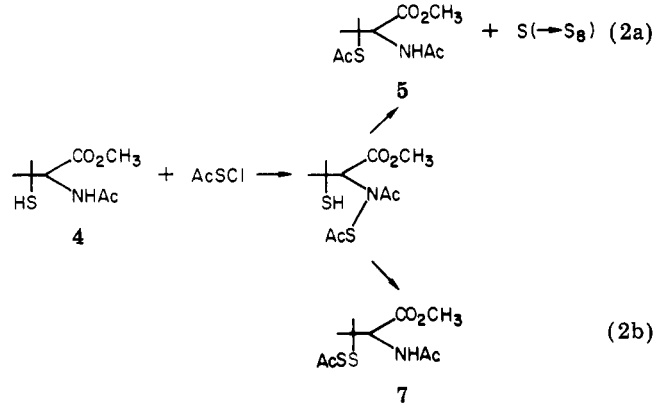
(1)

CH_2N_2 in ether gave **4** in good yield; however, the **4** was contaminated by a small amount of the *S*-methyl derivative of **4** (7–12% by NMR). Although excess CH_2N_2 increased the yield of the *S*-methyl derivative, less than the stoichiometric amount still led to dimethylated product in about 7% yield. DL-Penicillamine also could be converted to **4** by the standard method of esterification with $CH_3OH/(CH_3O)_2SO/HCl$, followed by acetylation; however, since the yield in the methylation step was only about 40%, the preferred route from **3** by treatment with CH_2N_2 proved more economical in time and materials. The *N*-acetyl ester **4** was readily freed of S-methylated product by recrystallization from ether.

The second step, conversion of **4** to the acetyl disulfide **7**, typically would be accomplished by reaction of the thiol **4** with acetylsulfenyl chloride, AcSCl.[6,9] In the present instance, the product obtained had NMR spectra consistent with the acetyl disulfide **7**, but the sulfur analysis was too low and showed the product to be the *S*-acetyl derivative **5**. Reaction of 2-acetamidoethanethiol with acetylsulfenyl chloride reportedly also failed to give the acetyl disulfide derivative, but the product was not characterized.[10] On the assumption that the reaction failed because the carbonyl group of acetylsulfenyl chloride was too reactive, (methoxycarbonyl)sulfenyl chloride[11] was used in the hope that the methoxycarbonyl disulfide **8** could be formed and converted to **7**. (Methoxycarbonyl)sulfenyl chloride condensed smoothly with the mercaptoamide **4** to give the unsymmetrical disulfide **8**, which did indeed show the expected IR and NMR spectra and which gave an analysis consistent with the presence of two sulfur atoms in the molecule. Unfortunately, **8** could not be cleaved to the hydrodisulfide **10** either by HCl in methanol or by *p*-bromoaniline in methanol, nor could it be converted to **7** by thioacetic acid in ether.

The route involving acetylsulfenyl chloride probably failed because of formation of an intermediate by reaction of the sulfenyl chloride initially at nitrogen rather than at the sulfur of the thiol, the sulfur being essentially in a neopentyl position. If the initial site of sulfenylation were the nitrogen, the adjacent thiol could react either by carbonyl substitution [losing sulfur and forming the S-acetyl derivative **5** (eq 2a)] or by displacement on sulfur as with sulfenamides to give the disulfide (eq 2b). In the sulfenylation by (methoxycarbonyl)sulfenyl chloride, the carbonyl group of the product would be expected to be less reactive than that of the corresponding carbonyl from acetylsulfenyl chloride, so that formation of the disulfide would be favored relative to acyl transfer.

Another possible route to **7** was conversion of **4** to the sulfenyl iodide, **9**, followed by reaction of **9** with thioacetic acid to give **7**. The conversion of **4** to **9** was accomplished easily by the procedure used earlier for the formation of **1** from the thiol;[4] it was complete in less than 3 min at room temperature (NMR). The sulfenyl iodide (**9**) was stable for several hours at room temperature when care



(2a)

(2b)

was taken to wash out excess iodine and iodide ion (60% decomposition in 15 h); otherwise, extensive decomposition occurred in 2 h (80%). Attempts to crystallize **9** from $CHCl_3$ at 0 °C by addition of CCl_4 led to rapid precipitation of a solid, but NMR analysis of the redissolved solid showed it to be identical with the product from decomposition in $CHCl_3$ solution, the corresponding disulfide **11** ($n = 0$). Addition of thioacetic acid to the freshly prepared sulfenyl iodide **9** in $CHCl_3$ did not give the expected acetyl disulfide, **7**, but instead reduced **9** to the original thiol (**4**), as evidenced by reappearance of the spectrum of **4**. Attempts to prepare **7** by conversion of **4** to the sulfenyl chloride through treatment with SO_2Cl_2, followed by the addition of thioacetic acid, gave a mixture of products from which **7** could not be isolated.

The final and successful route to the unsymmetrical disulfide **7** was through reaction of **4** with acetyl methoxycarbonyl disulfide. Methoxycarbonyl disulfides have been used as intermediates in preparing unsymmetrical disulfides because of their facile fragmentation upon treatment with a thiol and a catalytic amount of an amine to give methanol, carbonyl sulfide, and the disulfide.[11] Reaction of (methoxycarbonyl)sulfenyl chloride[11] with thioacetic acid proceeded readily, and the unsymmetrical disulfide formed (**12**) is a conveniently handled liquid (eq 3). Purification of the disulfide **12** is easier if a slight

$$AcSH + ClSCO_2CH_3 \rightarrow \underset{12}{AcSSCO_2CH_3} \xrightarrow{4,\ Et_3N} 7 + COS + CH_3OH \quad (3)$$

excess of the sulfenyl chloride is used, since the boiling point of the sulfenyl chloride is such that it is easily removed by distillation; use of excess thioacetic acid results in the formation of diacetyl disulfide, which is less readily separated from **12**. The unsymmetrical disulfide **12** could be stored for at least 1 month in a sealed glass ampule under normal laboratory conditions without detectable decomposition.

The reaction of **4** with **12** proceeds readily in the presence of triethylamine in methanol, with noticeable production of a gaseous product during the first few minutes (eq 3). Formation of the unsymmetrical disulfide **7** was observed always to be accompanied by formation of small to moderate amounts of polysulfides (**11**) comprised mostly of the trisulfide, perhaps because of base-catalyzed decomposition of **7** (vide infra); **7** was purified by chromatography over silica gel in ethyl acetate. A solution of **7** in pure methanol showed no decomposition over a 3-day period. Addition of *p*-bromoaniline, in an effort to generate the hydrodisulfide **10**, resulted in greater than 50% decomposition in 1 day with no evidence by NMR for acetylation of *p*-bromoaniline or for presence of the hydrodisulfide **10**; the base-induced decomposition products

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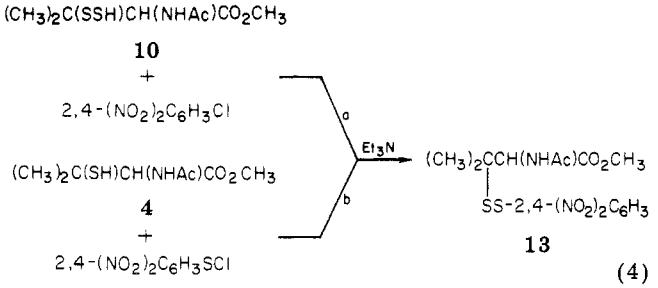
appeared to be the polysulfide (11), identified by TLC and NMR, and H_2S which was identified by its odor.

As expected, acid-catalyzed methanolysis of 7 led to 10, the hydrodisulfide, the reaction taking place in about 2 h when the solution was 0.5 N in HCl. The hydrodisulfide was not stable in acidic methanol; after about 8 h the odor of H_2S could be detected over the solution, and NMR peaks corresponding to the polysulfide 11 could be observed. After 1 day in this solution, 10 was more than 50% decomposed. Even washing a solution of the hydrodisulfide in $CHCl_3$ with aqueous bicarbonate, and in one instance merely with water, resulted in complete decomposition. A sample of 10 from which the solvent had been removed when stored as a glass at −10 °C showed complete decomposition in less than 36 h. The best procedure for formation of 10 seemed to be to perform the methanolysis, checking the progress of the reaction by the easily observed changes in the NMR spectrum in the δ 4.5–5.0 region for the methine doublet, and then, after evaporation of the reaction mixture to near dryness, to dissolve the residue immediately in $CDCl_3$ and wash the $CDCl_3$ solution carefully with a minimal amount of water.

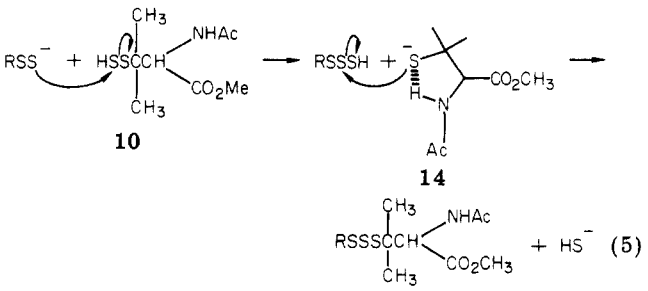
Evidence for the formation of 10, which to the best of our knowledge is the first amidohydrodisulfide to be characterized (although it was too unstable for elemental analysis), can be summarized as follows. (1) During the methanolysis, NMR peaks at δ 2.48 (CH_3COS) and δ 4.57 (CH doublet) characteristic of 7 disappear and are replaced by peaks characteristic of the formation of 10 at δ 2.04 [$CH_3CO_2CH_3$] and δ 4.76 (CH doublet). This second set of peaks also disappears with time, coincident with the evolution of H_2S as determined by its odor and the blackening of lead acetate paper. (2) The isolated product of the decomposition reaction is the polysulfide 11, as shown by comparison of the NMR spectrum and TLC behavior with those of the product 11 obtained in the preparation of 7. (3) The peak at δ 3.27, consistent with the expectation of "δ ~3" for RSSH,[10] disappeared when 10 in chloroform was shaken with D_2O. (4) Addition of the methanolysis reaction mixture to a solution of 1-chloro-2,4-dinitrobenzene and triethylamine in methanol resulted in the formation of disulfide 13 in 69% yield (eq 4a). This material was identical with 13 prepared by the



(4)

reaction of 2,4-dinitrobenzenesulfenyl chloride with 4 (eq 4b). (5) A sample of 10 in chloroform, which had proved stable under ambient conditions for 6 days and then at ~55 °C for an additional day, consumed 100% of the theoretical amount of iodine, presumably giving a tetrasulfide corresponding to 11 (with $n = 2$). (6) Treatment of 10 with cyanide ion gave thiocyanate ion in a reaction characteristic of most hydrodisulfides,[12] including the hydrodisulfide of cytochrome P-450,[2] although the test for SCN^- (with Fe^{3+}) was rather weak.

The weak test for SCN^- may find its explanation in the instability of the hydrodisulfide 10 to bases (e.g., CN^-). This instability undoubtedly also is the basis for formation of polysulfides 11 encountered in the preparation of the precursor 7 and in the decomposition of 10. We suggest that the facile reaction of 10 with bases stems from nucleophilic displacement of RS^- from RSSH by RSS^-. In the present instance the stability of RS^- might be enhanced by a hydrogen bonding interaction with the adjacent amide function (eq 5). The thiolate anion 14 then could displace



(5)

SH^- from the hydrotrisulfide to yield a trisulfide, the major component of 11, and ultimately H_2S. Extension of the mechanism of eq 5 to $RSSS^-$, etc., would lead to higher polysulfides, which probably are present but could not be separated from the trisulfide.

Experimental Section

Melting points were determined by using a Thomas-Hoover stirred-liquid apparatus and are corrected. NMR spectra were recorded in $CDCl_3$ with either a JEOL JNM-MH-100 spectrometer or a JEOL FX-60Q spectrometer using Me_4Si as an internal standard and are reported as parts per million (δ). IR spectra were obtained by using KBr pellets with Perkin-Elmer Model 521 and 727 spectrometers. Elemental analyses were performed by Galbraith Laboratories. Solvents were removed under reduced pressure by using a rotary-flask evaporator. TLC was done by using Eastman Chromagram sheets of 13181 silica gel. *N*-Acetyl-DL-penicillamine and 2,4-dinitrobenzenesulfenyl chloride were purchased from Aldrich Chemical Co.

Methyl 2-Acetamido-3-mercapto-3-methylbutanoate (4). To a solution of 8.0 g (42 mmol) of DL-*N*-acetylpenicillamine (3) in THF was added an ethereal solution of diazomethane which had been generated from nitrosomethylurea.[13] Addition was continued until the THF solution remained yellow. Excess diazomethane was decomposed by adding a small amount of acetic acid, and the solvent was removed under reduced pressure, yielding 10 g of crude 4. Crystallization from Et_2O at ~−10 °C (yields are poor at higher temperatures) gave 5.8 g (67%) of 4, mp 71–76 °C. One more crystallization from Et_2O gave 4: mp 78–79 °C; mass spectrum, m/e 205 (calcd for $C_8H_{15}NO_3S$, 205); NMR ($CDCl_3$) δ 6.74 (d, 1 NH), 4.68 (d, 1, J = 9 Hz, CHN), 3.78 (s, 3, CH_3O), 2.08 (s, 3, CH_3CO), 2.04 (s, 1, SH), 1.48 (s, 3, CH_3C), 1.38 (s, 3, CH_3C).

Anal. Calcd for $C_8H_{15}NO_3S$: C, 46.81; H, 7.37; N, 6.82; S, 15.62. Found: C, 46.92; H, 7.46; N, 6.73; S, 15.58.

Methyl 2-Acetamido-3-(acetylthio)-3-methylbutanoate (5). A solution of 3.15 g (15.3 mmol) of methyl 2-acetamido-3-mercapto-3-methylbutanoate (4) in 50 mL of CCl_4 was treated with acetylsulfenyl chloride[9] until the yellow color of the sulfenyl halide persisted; the resulting yellow solution was allowed to stand for 1 day. The CCl_4 was removed under reduced pressure; the resulting oil solidified upon standing. The solid was dissolved in Et_2O, and the solution was filtered and kept at ~−10 °C for 2 days. Filtration yielded 2.2 g (58%) of 5 as colorless solid, mp 93–96 °C. Two recrystallizations from Et_2O gave 5: mp 95.5–96.5 °C; IR (KBr) 3280, 1735, 1685, 1650 cm^{-1}; NMR ($CDCl_3$) δ 6.7 (d, 1, J = 9 Hz, NH), 4.84 (d, 1, J = 9 Hz, CHN), 3.74 (s, 3, CH_3O), 2.28 (s, 3, CH_3COS), 2.08 (s, 3, CH_3CON), 1.84 (s, 6, $(CH_3)_2C$).

Anal. Calcd for $C_{10}H_{17}NO_4S$: C, 48.56; H, 6.93; N, 5.66; S, 12.96. Found: C, 48.38; H, 7.14; N, 5.64; S, 13.09.

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Methyl 2-Acetamido-3-(acetyldithio)-3-methylbutanoate (7). To a solution of 1.02 g (5.0 mmol) of **4** in 15 mL of CH_3OH was added 0.83 g (5.0 mmol) of methoxycarbonyl acetyl disulfide. A few drops of Et_3N were added, and the reaction mixture was allowed to stand overnight. The CH_3OH was removed under reduced pressure and the resulting oil chromatographed over 20 g of silica gel in EtOAc. The first portion, 0.7 g, was further purified by preparative TLC to give 0.7 g (50%) of oily **7** that crystallized upon standing; mp 80–87 °C. Two recrystallizations from Et_2O gave material with a constant melting point of 89–90 °C: IR (KBr) 3325, 1742, 1732, 1646, 1531 cm^{-1}; NMR ($CDCl_3$) δ 6.75 (d, 1, J = 9 Hz, NH), 4.60 (d, 1, J = 9 Hz, CHN), 3.75 (s, 3, CH_3O), 2.46 (s, 3, CH_3COS), 2.09 (s, 3, CH_3CON), 1.43 (s, 3, CH_3C), 1.34 (s, 3, CH_3C).

Anal. Calcd for $C_{10}H_{17}NO_4S_2$: C, 42.99; H, 6.13; S, 22.95. Found: C, 43.23; H, 6.35; S, 22.72.

The second fraction, 0.4 g, solidified after being allowed to stand; mp 127–153 °C. This material, **11**, yielded H_2S, as detected by lead acetate paper and by its odor, upon treatment with Zn and HCl in CH_3OH; **11** is best characterized by CCH_3 signals at δ 1.46 and 1.44 and (less clearly) by two sets of CH methine signals at δ 4.84 and 4.81.

Anal. Calcd for $C_{16}H_{28}N_2O_6S_3$: S, 21.83. Found: S, 20.97 (23.70 after recrystallization from EtOAc).

Methyl 2-Acetamido-3-[(methoxycarbonyl)dithio]-3-methylbutanoate (8). To a solution of 1.00 g (4.9 mmol) of **4** in 20 mL of Et_2O was added 0.61 g (4.9 mmol) of (methoxycarbonyl)sulfenyl chloride[11] in 15 mL of Et_2O. The yellow color of the sulfenyl chloride faded immediately, and the reaction mixture was allowed to stand for 0.5 h at room temperature. The Et_2O was removed under reduced pressure. The resulting oil (1.5 g) crystallized after being washed with Et_2O and EtOAc; mp 80–86 °C. Recrystallization once from Et_2O and then from hexane gave pure **8** (typically in yields of ~70%): mp 86.7–87.7 °C; IR 3290, 1737, 1732, 1638, 1536 cm^{-1}; NMR ($CDCl_3$) δ 6.70 (d, 1, J = 9 Hz, NH), 4.51 (d, 1, J = 9 Hz, CHN), 3.90 (s, 3, CH_3O), 3.74 (s, 3, CH_3O), 2.07 (s, 3, CH_3CON), 1.45 (s, 3, CH_3C), 1.38 (s, 3, CH_3C).

Anal. Calcd for $C_{10}H_{17}NO_5S_2$: C, 40.66; H, 5.80; S, 21.71. Found: C, 40.93; H, 5.96; S, 21.30.

1-Acetamido-1-(methoxycarbonyl)-2-methyl-2-propanesulfenyl Iodide (9). To a solution of 20 mg of **4** (0.10 mmol) in 3 mL of $CDCl_3$ were added 100 mg (0.39 mmol) of I_2 and 1 mL of D_2O. Within less than 3 min the NMR spectrum of **4** had completely disappeared, and a new spectrum had appeared: δ 4.71 (s, 1, CHND), 3.78 (s, 3, CH_3O), 2.07 (s, 3, CH_3CON), 1.54 (s, 3, CH_3C), 1.50 (s, 3, CH_3C). The solution was washed with D_2O, the excess I_2 was removed by careful addition of aqueous sodium thiosulfate, and the solution then was washed three times with small portions of D_2O. Addition of excess thioacetic acid to the solution of **9** immediately caused the red-orange color of the sulfenyl iodide to fade to yellow. The NMR spectrum of the sulfenyl iodide was replaced by that of **4**.

1-Acetamido-1-(carbomethoxy)-2-methyl-2-propyl Hydrodisulfide (10). To 50 mg of **7** in 0.3 mL of CH_3OH were added 2 drops of 35% HCl. The progress of the reaction was followed by observing the δ 4–5 region in the NMR spectrum. After 2.5 h the CH_3OH was removed under reduced pressure at 27 °C, the oil was dissolved in $CHCl_3$ and washed with ~0.1 mL of H_2O, and the $CHCl_3$ was removed under reduced pressure. The resulting colorless oil was dissolved immediately in $CDCl_3$ and analyzed by NMR: δ 6.30 (d, 1, J = 9 Hz, NH), 4.81 (d, 1, J = 9 Hz, CHN), 3.77 (s, 3, CH_3O), 3.27 (s, 1, SSH, disappears with D_2O wash), 2.05 (s, 3, CH_3CON), 1.36 (s, 6, $(CH_3)_2C$). Similar treatment of 60 mg (0.21 mmol) of **7** for 3 h followed by addition of the acidic reaction mixture to a solution of 50 mg (0.25 mmol) of 1-chloro-2,4-dinitrobenzene and 0.3 mL of Et_3N in 1 mL of CH_3OH yielded a product mixture that showed by TLC a compound with an R_f corresponding to the authentic nitrophenyl disulfide **13**. The product mixture was purified by preparative TLC to yield **13**: 60 mg (69%); mp and mmp (with authentic **13**) 161–162 °C; the IR spectrum was identical with that of authentic **13**.

Essentially as reported,[12] a solution of **10**, from 10 mg of **7** in 0.5 N methanolic HCl, was treated with 3 mg of KCN, and the pH was adjusted to ~8 with dilute NH_4OH and aqueous bicarbonate. After 3 h, the solution was extracted with $CHCl_3$ and then made slightly acidic with aqueous HCl. The addition of 1 drop of a $Fe(NO_3)_3$ solution gave the red color characteristic of ferric thiocyanate.

A sample of **10** in $CDCl_3$, from 35 mg (0.13 mmol) of **7** remained unchanged, by NMR, during 6 days at room temperature and an additional day at 50–55 °C. A 2.0-mL portion of 0.10 N I_2 (in aqueous KI) was added, the solution was shaken vigorously for 1 min, and then the excess I_2 was titrated with 0.75 mL of 0.10 N $Na_2S_2O_3$ (100% of theory, 0.13 mequiv). Extraction of the titrated sample with $CHCl_3$ and evaporation of the $CHCl_3$ under reduced pressure gave an oil, presumably the tetrasulfide corresponding to **11** with n = 2; NMR ($CDCl_3$) δ 6.35 (d, 1, NH), 4.80 (2 sets of doublets, 1, J = 10 Hz, CHN), 3.78 (s, 3, CH_3O), 2.07 (s, 3, CH_3CON), 1.53 (s, 3, CH_3C), 1.46 (s, 3, CH_3C).

Acetyl Methoxycarbonyl Disulfide (12). To a solution of 6.3 g of methoxycarbonyl sulfenyl chloride[11] in Et_2O was added a solution of 3.8 g of thioacetic acid in 35 mL of Et_2O. After the addition was complete (~5 min), the solution was stirred for 0.5 h and the Et_2O removed under reduced pressure to give a yellow oil (8.1 g). Distillation at 62 °C (0.2 torr) gave a colorless liquid, n^{27}_D = 1.5244. Redistillation gave a product, n^{27}_D = 1.5212–1.5220, in a yield exceeding 38%; NMR ($CDCl_3$) δ 3.86 (s, 3, CH_3O), 2.40 (s, 3, CH_3COS).

Anal. Calcd for $C_4H_6O_3S_2$: C, 28.90; H, 3.64. Found: C, 28.83; H, 3.47.

Authentic Methyl 2-Acetamido-3-(2,4-dinitrophenyldithio)-3-methylbutanoate (13). To a solution of 100 mg (0.49 mmol) of **4** in 20 mL of Et_2O were added 50 mg (0.50 mmol) of Et_3N and 120 mg (0.51 mmol) of 2,4-dinitrobenzenesulfenyl chloride, and the solution was allowed to stand for 3 h. The yellow solid (200 mg, 98%) was removed by filtration; mp 159–161 °C. Two recrystallizations from EtOAc gave **13** with a constant melting point of 161–163 °C; NMR ($CDCl_3$) δ 8.99 (m, 1, Ar H), 8.67 (m, 2, Ar H), 7.64 (br, 1, NH), 4.81 (d, 1, CHN), 3.73 (s, 3, CH_3O), 1.97 (s, 3, CH_3CON), 1.41 (s, 6, $(CH_3)_2C$). No evidence for disproportionation of **13** could be found by TLC following 4 h in refluxing CH_3OH.

Anal. Calcd for $C_{14}H_{17}N_3O_7S_2$: C, 41.67; H, 4.25. Found: C, 41.84; H, 4.28.

Acknowledgment. We are grateful to the National Institutes of Health for support of this research through USPHS Grant ES00267 to the Center in Environmental Toxicology in the School of Medicine at Vanderbilt University.

Registry No. DL-**3**, 59-53-0; DL-**4**, 76548-18-0; DL-**5**, 76480-05-2; DL-**7**, 76480-06-3; DL-**8**, 76480-07-4; DL-**9**, 76480-08-5; DL-**10**, 76480-09-6; **11** (n = 1), 76480-10-9; **11** (n = 2), 76480-11-0; **12**, 76480-12-1; DL-**13**, 76480-13-2; methoxycarbonyl acetyl disulfide, 76480-12-1; (methoxycarbonyl)sulfenyl chloride, 26555-40-8; thioacetic acid, 507-09-5; 2,4-dinitrobenzenesulfenyl chloride, 528-76-7.