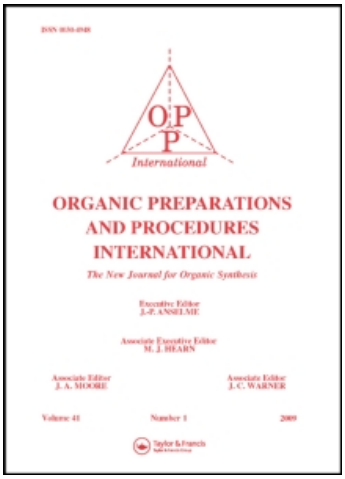

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LARGE-SCALE PREPARATION OF THE SULFUR-TRANSFER REAGENT 3H-1,2-BENZODITHIOL-3-ONE 1,1-DIOXIDE

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LARGE-SCALE PREPARATION OF THE SULFUR-TRANSFER REAGENT 3H-1,2-BENZODITHIOL-3-ONE 1,1-DIOXIDE

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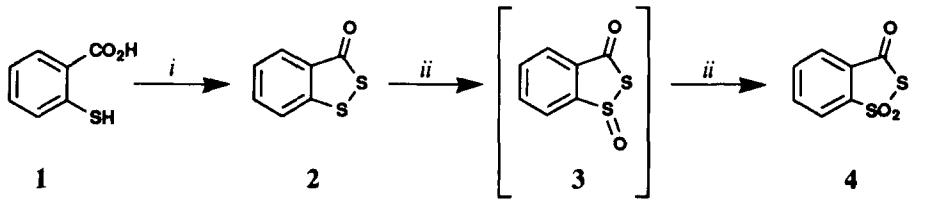
The thiosulfonate 3H-1,2-benzodithiol-3-one 1,1-dioxide (**4**) is a powerful sulfur-transfer reagent for the synthesis of oligodeoxyribonucleoside phosphorothioates *via* the phosphoramidite approach.[1] These oligonucleotide analogues exhibited antiviral activity in cell cultures[2] and have been effective in the control of gene expression by inhibiting the translation of specific messenger RNAs.[3]

A detailed concentration study revealed that the sulfurization of oligonucleotidic phosphite triesters can be effected consistently to near quantitative yields, within 30 s, with a 0.05 M solution of

3H-1,2-benzodithiol-3-one 1,1-dioxide (**4**) in acetonitrile.[4] The sulfurizing efficiency of **4** is comparable to that of elemental sulfur (S_8) which has also been used as a sulfur-transfer reagent in the synthesis of oligodeoxyribonucleoside phosphorothioates[5] and phosphorodithioates[6] in addition to 1,2-diacyl-*sn*-glycero-3-thiophosphate[7] and *myo*-inositol phosphorothioate derivatives.[8] In contrast to S_8, **4** is soluble in various organic solvents and provides rapid sulfurization kinetics. It is, therefore, anticipated that like S_8, **4** may find broad application toward the synthesis of biologically important phosphorothioated molecules. The synthetic protocol pertaining to the preparation of 3H-1,2-benzodithiol-3-one 1,1-dioxide reported earlier[1b] is unsuitable for the large-scale synthesis of the reagent.[1c] The methodology has been modified to improve the availability of **4** and the details of this procedure are reported herein.



1 **2** **3** **4**

i: $CH_3C(O)SH/H_2SO_4$; *ii:* $H_2O_2/CF_3CO_2H/ \leq 40°$

EXPERIMENTAL SECTION

Thiosalicylic acid and thiolacetic acid were purchased from Aldrich Chemical Company Inc. Sulfuric acid and trifluoroacetic acid (Peptide synthesis grade) were obtained from Mallinckrodt and Applied Biosystems Inc., respectively. All reagents were used without further purification. Teflon membrane filters (47 mm, 0.45 μm) were purchased from Millipore Corp. Thin-layer chromatography was performed on plastic plates coated with a 0.2 mm thick layer of silica gel 60 F_{254} (EM Science). Melting points were determined on a Büchi 510 melting point apparatus. Elemental analyses were done by Atlantic Microlab Inc. (Norcross, GA). NMR spectra were recorded with a General Electric Model GN 300 spectrometer operating in the presence of broad-band decoupling at 7.05 Tesla (300 MHz for ^1H). ^{13}C NMR spectra were recorded in $CDCl_3$ using the middle line of resonances obtained from the solvent (δ = 77.0 ppm) as internal reference. Electron-ionization mass spectra were recorded with a Hewlett-Packard 5995 gas chromatograph/mass spectrometer equipped with an HP 59970C MS Chem Station data system. The ionization potential was 70 eV and the ionizing current was 220 μA.

3H-1,2-Benzodithiol-3-one (2).- In a well-ventilated fume hood, a 7.5 L cylindrical Pyrex reservoir[9] was placed in a water bath kept at ambient temperature. The reservoir was charged with thiosalicylic acid (300 g, 1.92 mol) and conc. sulfuric acid (3 L). Thiolacetic acid (282 mL, 3.94 mol) was then added dropwise over 1.5 hr to the mechanically stirred suspension. Upon completion of the addition, the reaction mixture was rapidly warmed to an internal temperature of 44-46° and was stirred at that temperature for 2 hrs.[10] The dark brown mixture was then gradually added to crushed ice (30 L). The precipitate was left in the ice for 15 min before being collected on a coarse glass-sintered funnel. To facilitate and expedite the filtration, excess ice in the funnel was melted by adding warm water (*ca.* 50°) and the drained precipitate was periodically transferred to a large beaker (4 L). The crude solid

was partitioned between chloroform (1.5 L) and a saturated solution of sodium bicarbonate (500 mL) which was slowly added to avoid excessive release of carbon dioxide. The suspension was then filtered by suction and the aqueous portion of the filtrate (*ca.* 500 mL) was discarded while the organic layer was extracted once more with a saturated solution of sodium bicarbonate (500 mL). The organic phase was dried over anhydrous magnesium sulfate, filtered and evaporated to dryness under reduced pressure affording 270.1 g (82%) of 3H-1,2-benzodithiol-3-one as a dark yellow solid.

The crude product was recrystallized in a batchwise manner according to the following protocol. The compound (50 g) was dissolved in hexane (825 mL) upon stirring and *moderate* heating on a hot plate. The boiling yellow solution was carefully decanted from a black oil to an Erlenmeyer flask (1 L), allowed to cool at ambient temperature and then at -10° overnight. The crystalline material was collected, washed with hexane (200 mL) and dried under vacuum. The filtrate obtained from each recrystallization batch were pooled and evaporated to dryness under reduced pressure. The residue was recrystallized from hexane as above.

The total amount of recrystallized product (249.6 g) was dissolved in acetone (*ca.* 2.3 L) and filtered from insoluble material.[11] The filtrate was evaporated to dryness under reduced pressure affording 241.8 g (75%) of 3H-1,2-benzodithiol-3-one,[12] mp. 73-74°, lit.[12b] mp. 77°.

^{13}C NMR ($CDCl_3$): δ 193.5, 148.2, 133.5, 129.0, 127.2, 125.6, 124.6. MS (70 eV): *m/z* 170 (M+2) (9), 169 (M+1) (9), 168 (M) (100, base peak), 140 (36), 104 (49), 96 (60), 76 (46), 69 (52), 50 (60).

3H-1,2-Benzodithiol-3-one 1,1-Dioxide (4).- In a 3 L three-necked flask was added 3H-1,2-benzodithiol-3-one (235.6 g, 1.4 mol) and trifluoroacetic acid (1.5 L). The solution was magnetically stirred for 5 min at ambient temperature and then cooled to 5° in a wet ice/acetone bath. Aqueous hydrogen peroxide (30%, 240 mL) was added dropwise over 30 min. The reaction mixture was then removed from the cold bath and stirred at room temperature until the solution exothermically reached an internal temperature of 40° which was maintained for 30 min through repeated cooling sessions. A second portion of hydrogen peroxide (240 mL) was added, as above, to the cooled (33°) reaction mixture. Upon completion of the addition, the internal temperature of the solution was similarly maintained at 37-41° for 30 min. A third and final addition of peroxide (240 mL) was carried out and the reaction went to completion, at 37-41°, within the next 2.5 hrs.[13]

The reaction mixture was then poured onto 20 L of crushed ice and allowed to stand 15 min before the solid was collected. As above, the excess ice in the coarse glass-sintered funnel was melted by adding warm water and the white crystals were thoroughly washed with water until the filtrate was neutral and low in peroxides (*ca.* 1 mg/L).[14] The white solid was dissolved in methylene chloride (600 mL) and the solution was extracted once with 1% aqueous sodium bisulfite (250 mL) and then twice with water (250 mL). The organic phase was dried over anhydrous sodium sulfate, filtered, and evaporated to dryness under reduced pressure. The crude solid (136.0 g, 0.68 mol) was dissolved in methylene chloride (500 mL) with *moderate* heating. The solution was filtered through a teflon membrane, brought to a volume of 900 mL with hexane, heated to boiling, allowed to cool to room temperature and then placed in the freezer (-10°) overnight. The white crystals (104.2 g) were collected, washed

with hexane (100 mL) and dried under vacuum. The filtrate was taken to dryness and the material was recrystallized as above affording a second crop of colorless crystals (24.0 g) to a total yield of 128.2 g (46%) of 3H-1,2-benzodithiol-3-one 1,1-dioxide,[15] mp. 102.5-103°, lit.[16] mp. 98-99°.
^{13}C NMR ($CDCl_3$): δ 182.9, 148.3, 136.5, 134.5, 130.0, 125.6, 121.9. MS (70 eV): *m/z* 202 (M+2) (4), 201 (M+1) (4), 200 (M) (45), 136 (100, base peak), 108 (41), 104 (37), 76 (92), 69 (31).
Anal. Calcd. for $C_7H_4O_3S_2$: C, 41.99; H, 2.01; S, 32.02. Found: C, 42.00; H, 1.97; S, 31.95.

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9. The cylindrical reservoir is a 22x25 cm chromatography developing chamber purchased from Fisher Scientific.

10. The bath temperature was 52°.

11. Analysis of the insoluble material by mass spectrometry (70 eV) indicated that the compound (*m/z*= 256) has a fragmentation pattern identical to that observed with elemental sulfur (S_8).

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13. The reaction rates were monitored by thin-layer chromatography ($CHCl_3$) until the presence of the slow-moving thiosulfinate **3** (R_f= 0.27) was negligible (less than 5%).

14. Peroxides were qualitatively measured with Quantofix test sticks purchased from Aldrich Chemical Co.

15. The solid reagent can be stored indefinitely, at ambient temperature, in amber glass containers. Alternatively, a 0.05 M solution of **4** in acetonitrile can be kept for at least one month at 20° in a properly cleaned and "siliconized" bottle.[1b]

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SYNTHESIS OF 4-AROYL-[(5-PHENYL-2(2H)-TETRAZOLYL)-1-OXOETHYL]-THIOSEMICARBAZIDES AND 4-AROYL-5-[(5-PHENYL-2(2H)-TETRAZOLYL)METHYL)-1,2,4-(4H)-TRIAZOLE-3-THIONE

Submitted by (09/13/91)

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Acylthiosemicarbazides have been reported to possess physiological activity.[1-3] We had previously prepared acylthiosemicarbazides and related heterocyclic derivatives by reaction of cyanoacetylhydrazine[4] and α-phenylcyanoacetylhydrazine[5] with aroylisothiocyanates and observed that they promote growth of wheat plumules at low concentrations. A number of 5-aryl-2H-tetra-