

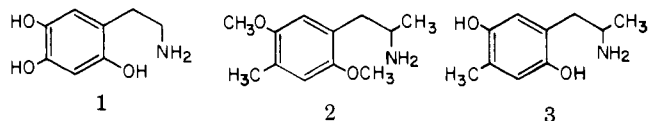
## Monomethylthio Analogues of 1-(2,4,5-Trimethoxyphenyl)-2-aminopropane

Peyton Jacob, III, George Anderson, III, Charles K. Meshul, Alexander T. Shulgin, and Neal Castagnoli, Jr.\*

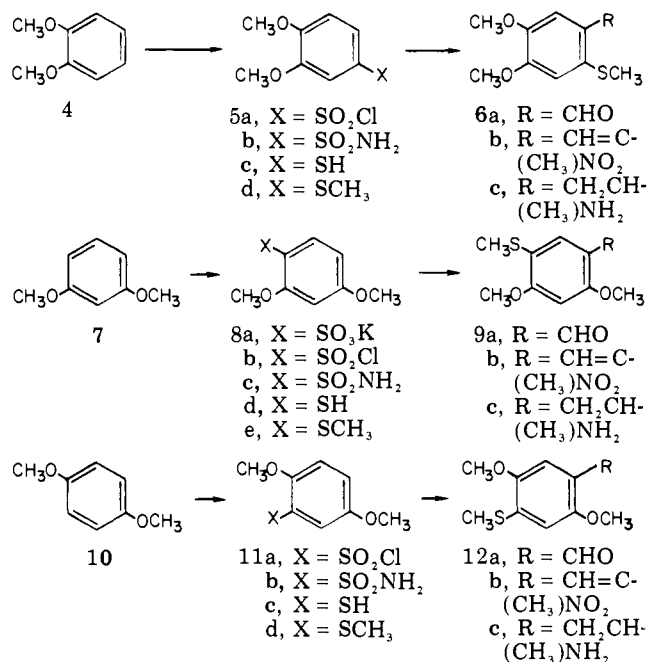
Department of Pharmaceutical Chemistry, School of Pharmacy, University of California, San Francisco, California 94143.  
Received February 9, 1977

Regiospecific syntheses of the three monomethylthio analogues of 1-(2,4,5-trimethoxyphenyl)-2-aminopropane are described. The three isomeric amines were evaluated for potential psychotomimetic potency using the rabbit hyperthermia assay. Enantiomeric compositions and time-concentration curves in rat brains were determined following intraperitoneal administration of each compound. The biological data are contrasted with the corresponding results obtained with the potent human psychotogen 1-(2,5-dimethoxy-4-methylphenyl)-2-aminopropane (DOM).

Extensive structure-activity relationship studies among the ring-substituted 1-phenyl-2-aminopropanes have established that compounds bearing methoxy groups at the 2 and 5 positions and an alkyl, alkoxy, or halo substituent at the 4 position are potent psychotomimetic agents in man.<sup>1</sup> This ring-substitution pattern also is found in the sympatholytic agent 6-hydroxydopamine (1).<sup>2</sup> We<sup>3</sup> and other workers<sup>4</sup> have examined the metabolic fate of the well-known psychotomimetic agent 1-(2,5-dimethoxy-4-methylphenyl)-2-aminopropane (2, DOM). Our studies have established that bis-O-demethylation of DOM leads to the 6-hydroxydopamine analogue 3. As anticipated, this hydroquinone metabolite possesses 6-hydroxydopamine-like properties both in terms of its chemical behavior<sup>5</sup> and its ability to cause neuronal damage in rat brains.<sup>6</sup> In order to further evaluate the structural significance of the 2,5-dimethoxy substitution pattern with regard to psychopharmacological activity, we have synthesized the three methylthio analogues of 1-(2,4,5-trimethoxyphenyl)-2-aminopropane, compounds 6c, 9c, and 12c. The present paper describes the chemical characterization of these isomers together with a preliminary evaluation of their potential psychotomimetic activity as measured by the rabbit hyperthermia model.<sup>7</sup> In an effort to evaluate the influence of structure on the partitioning of these compounds into the central nervous system, we have determined the time-concentration curves of DOM and the methylthio analogues in rat brains following intraperitoneal administration. Finally, we have examined the enantiomeric composition of these compounds in rat brains since in those cases examined<sup>7,8</sup> the *R* enantiomers of the one-ring psychotomimetic drugs have been shown to be the psychopharmacologically active isomers. Studies in our laboratory have shown that the *in vivo*<sup>9</sup> and *in vitro*<sup>10</sup> disappearance of DOM and the formation of DOM<sup>11</sup> *in vitro* metabolites are under stereochemical control.



Scheme I



**Chemistry.** The synthetic route to the three required methylthio analogues 6c, 9c, and 12c in each case started with the appropriately substituted dimethoxybenzene and involved two critical regiospecific aromatic substitution reactions (Scheme I). For example, 1,2-dimethoxybenzene (4) was converted to 3,4-dimethoxybenzenesulfonyl chloride (5a) by chlorosulfonation with chlorosulfonic acid. The product was further characterized by conversion to the corresponding sulfonamide 5b. Reduction of the sulfonyl chloride moiety with zinc and sulfuric acid provided the thiophenol derivative 5c which was readily methylated with methyl iodide to give 5d. Vilsmeier formylation of 5d gave 4,5-dimethoxy-2-methylthiobenzaldehyde (6a) in good yield. The expected 1,2,4,5-substitution pattern was confirmed by NMR spectroscopy

Table I. Hyperthermic Effects of the Three Isomeric Methylthio Compounds and of DOM in Rabbit

Compd	Dose, $\mu\text{mol/kg}$	Period of peak effect, min	Mean max temp rise (method A)	Integration of time-temp curve (0-300 min) (method B)	Approx dose for 1 °C temp rise, $\mu\text{mol/kg}$	Potency rel to DOM	
						Method A	Method B
6c	14.2	75-105	1.01 $\pm$ 0.13	0.545	~14	0.017	0.022
	7.1	105-135	0.82 $\pm$ 0.06	0.448			
9c	21.3	180-210	1.62 $\pm$ 0.06	1.07	9.77	0.028	0.039
	10.6	135-165	1.09 $\pm$ 0.07	0.683			
12c	5.3	120-165	0.60 $\pm$ 0.07	0.451	0.50	0.54	0.64
	2.0	195-225	2.31 $\pm$ 0.51	1.384			
	0.67	165-195	1.15 $\pm$ 0.22	0.731			
2 (DOM)	0.22	120-135	0.60 $\pm$ 0.16	0.343	0.27	1.0	1.0
	1.00	165-210	2.85 $\pm$ 0.35	1.631			
Mescaline	0.50	195-225	1.98 $\pm$ 0.14	1.121			
	0.25	135-165	0.80 $\pm$ 0.11	0.489			
LSD						0.0006 <sup>a</sup>	33 <sup>a</sup>

<sup>a</sup> From ref 7; normalized to DOM = 1.

which revealed two singlets in the aromatic region consistent with a para orientation of the aromatic protons. Condensation of 6a with nitroethane gave the 1-phenyl-2-nitropropene 6b, which was smoothly reduced with  $\text{LiAlH}_4$  to the desired 1-phenyl-2-aminopropane 6c. The two positional isomers 9c and 12c were prepared in an analogous manner (Scheme I), except that direct chlorosulfonation of 7 failed. The desired intermediate 8b was obtained by treatment of 7 with sulfuric acid followed by phosphorus oxychloride. The 4-methylthio isomer 12c has been previously prepared by a different procedure.<sup>12</sup> Satisfactory microanalyses and spectra data were obtained for all new compounds.

**Pharmacology and Discussion.** In a detailed paper by Aldous et al.<sup>7</sup> variously substituted 1-phenyl-2-aminopropanes were examined for their abilities to increase the rectal temperatures of trained rabbits. An exceptionally good correlation was observed between reported human psychotomimetic potency and the hyperthermia activity of the compounds examined. Recently, additional studies<sup>13</sup> have confirmed the value of this animal model. Consequently, we determined the hyperthermia properties of the amines 6c, 9c, 12c, and DOM (2) according to this procedure.

The hyperthermia data are summarized in Table I, which includes the potencies of mescaline and LSD<sup>7</sup> for comparison. The results clearly establish that only the 4-methylthio isomer 12c possesses significant hyperthermia activity. Compounds 6c and 9c, while somewhat more potent than mescaline, are approximately  $1/50$ th as active as racemic DOM and  $1/20$ th as active as 12c.

Although a number of studies have attempted to correlate physical properties, such as partition coefficients,<sup>14</sup> with the pharmacological properties of psychotomimetic drugs, relationships between potency and brain concentration have not been reported. We have determined the enantiomeric ratios (Table II) and concentrations (Figure 1) of DOM (2) and the three methylthio analogues (6c, 9c, and 12c) in the whole rat brain at various times following intraperitoneal administration of racemic drugs. At 15 mg/kg (61.2  $\mu\text{mol/kg}$ ) DOM reaches a peak concentration in the brain at approximately 15-30 min following intraperitoneal administration. The percentage of the administered dose appearing in the brain is significantly higher for DOM than for the methylthio analogues which were studied at 30 mg/kg (114.9  $\mu\text{mol/kg}$ ). Additionally, it is apparent that peak brain concentrations are achieved more rapidly with DOM than for the methylthio analogues. Interestingly, significantly higher brain concentrations were

Table II. Enantiomeric Ratios (*R/S*) of the Three Isomeric Methylthio Compounds and of DOM in Rat Brain Following Administration of the Racemates

Compd	Amt injected, ip	Enantiomeric ratio ( <i>R/S</i> ) at time <i>t</i> (min)				
		5	15	30	60	120
6c	30 mg/kg (114.9 $\mu\text{mol/kg}$ )		1.14	1.09	1.43	
9c	30 mg/kg (114.9 $\mu\text{mol/kg}$ )		1.10	1.11	1.35	
12c	30 mg/kg (114.9 $\mu\text{mol/kg}$ )		1.54	1.14	1.23	1.62
2	15 mg/kg (61.2 $\mu\text{mol/kg}$ )	0.95	1.06	1.46	1.55	1.34

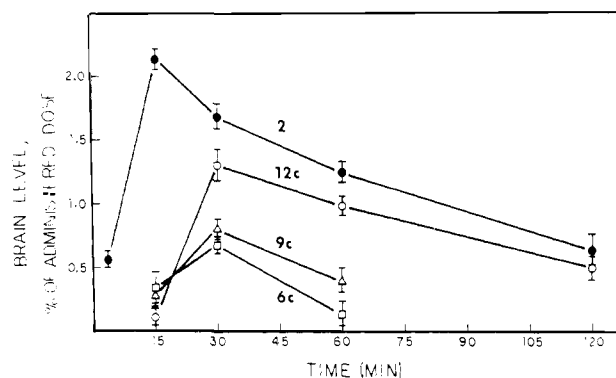


Figure 1. Time-concentration curves for the three isomeric methylthio compounds and DOM in rat brain. Each point represents an average obtained from at least three rats.

reached with the 4-methylthio derivative 12c than with the other two isomers. The time course of events appears to be approximately the same for all three methylthio compounds with peak brain concentrations being achieved between 30 and 60 min, slightly longer than observed for DOM.

At all times studied and with all four compounds (except for the 5-min observation with 2) it was observed that the *R* enantiomer was present in the brain at a higher concentration than was the *S* isomer. The ratio (*R/S*) observed for the three methylthio isomers was essentially constant at 1.10 at the time of maximum brain concentration (30 min following injection) and in general showed an increased enrichment in the *R* isomer (or a concomitant decrease in *S* isomer) at both earlier and later times, when the total concentration was lower.

The most striking fact to emerge from the above data is the clear-cut pharmacological distinction of the 4-methylthio compound **12c** from the positional isomers **6c** and **9c**. It seems unlikely that the differences in biological properties could be due to lipophilicity differences, since these three isomeric compounds would be expected to have nearly identical partition coefficients. Furthermore, all three methylthio compounds should have nearly identical steric features, which would seem to argue against a sterically sensitive interaction of the parent compound with a receptor.

Our previous observation that DOM (**2**) is metabolically O-demethylated to the hydroquinone derivative **3** has led us to propose that the psychotomimetic properties of DOM may be mediated by this "6-hydroxydopamine-like" metabolite.<sup>3</sup> Of the three methylthio analogues **6c**, **9c**, and **12c**, only the 4-methylthio compound **12c** has the potential for direct hydroquinone formation by O-demethylation.<sup>25</sup> It is of interest therefore that compound **12c** proved to be the most potent of the three isomers in the rabbit hyperthermia assay. The differences in brain concentrations of the methylthio isomers alone would probably not account for this difference in biological potency. Further studies on specifically substituted 1-phenyl-2-aminopropanes as well as studies concerning the formation of 6-hydroxydopamine-like metabolites are currently in progress.

## Experimental Section

**Chemistry.** Melting points were taken on a Thomas-Hoover apparatus and are uncorrected. Infrared spectra were obtained from mineral oil mulls using a Perkin-Elmer Model 337 spectrophotometer. NMR spectra were recorded on a Varian A-60 or Perkin-Elmer R-12B instrument. Chemical shifts are reported in parts per million relative to Me<sub>4</sub>Si(CDCl<sub>3</sub>) or DSS (D<sub>2</sub>O). Microanalyses were performed by the Microanalytical Laboratory, University of California, Berkeley, Calif. The syntheses and properties of the nitropropene **12b** and amine **12c** have been described previously.<sup>12</sup>

**3,4-Dimethoxybenzenesulfonyl Chloride (5a).** The procedure was based on the method of Huntress and Carten.<sup>16</sup> Chlorosulfonic acid (50 g, 0.43 mol) was added portionwise, with stirring, to neat veratrole (**4**, 27.6 g, 0.20 mol) over a 20-min period. The reaction was exothermic and HCl was evolved. The viscous blue-green mixture was poured into 400 mL of crushed ice and after standing 1 h the mixture was extracted with methylene chloride (2 × 150 mL). The extract was concentrated in vacuo to give a colorless oil which solidified on standing. The yield of crude product was 37.1 g (79%): mp 63–66 °C (lit.<sup>15</sup> mp 72–73 °C); NMR (CDCl<sub>3</sub>) δ 4.00 (s, 3 H, OCH<sub>3</sub>), 4.02 (s, 3 H, OCH<sub>3</sub>), 7.07 (d, 1 H, *J* = 9 Hz, ArH), 7.50 (d, 1 H, *J* = 2.5 Hz, ArH), 7.74 (d of d, 1 H, ArH).

The sulfonamide **5b** was recrystallized from ethanol as colorless needles: mp 132–133 °C (lit.<sup>16</sup> mp 135–136 °C).

**2,5-Dimethoxybenzenesulfonyl Chloride (11a).** This isomer was prepared from **10** as described for **5a**, with the exception that the chlorosulfonation was carried out with methylene chloride as solvent. The yield of **11a** was 42%: mp 109–112 °C; NMR (CDCl<sub>3</sub>) δ 3.84 (s, 3 H, OCH<sub>3</sub>), 4.02 (s, 3 H, OCH<sub>3</sub>), 7.14–7.56 (m, 3 H, ArH).

The sulfonamide **11b**, from ethanol, had mp 147.5–148.5 °C (lit.<sup>16</sup> mp 148 °C).

**2,4-Dimethoxybenzenesulfonyl Chloride 8b.** The chlorosulfonic acid procedure described above for the sulfonation of the *o*- and *p*-dimethoxybenzenes **4** and **10**, respectively, was not effective for the sulfonation of **7**. The procedure employed was essentially that of Suter and Hansen.<sup>17</sup> Concentrated sulfuric acid (29 g, 0.30 mol) was added to neat 1,3-dimethoxybenzene (**7**, 27 g, 0.20 mol), with stirring, over a 15-min period. The mixture was stirred 1 h at ambient temperature and then poured slowly into 250 mL of saturated aqueous potassium carbonate. The resulting precipitate was collected by filtration and dried (air oven, 125 °C) to give 59.6 g of crude potassium 2,4-dimethoxybenzene-

sulfonate (**8a**). Phosphorus oxychloride (35 g) was added to the pulverized salt **8a** (30 g) and the mixture was heated on a steam bath with occasional swirling (2 h). After cooling to room temperature the mixture was poured onto 300 mL of crushed ice, stirred until the ice had melted, and extracted with ether (2 × 150 mL). The combined extracts were washed with saturated aqueous NaCl (50 mL) and evaporated under reduced pressure to give 14.2 g (62% based on **7**) of a white solid: mp 69–72 °C (lit.<sup>17</sup> mp 70.5 °C); NMR (CDCl<sub>3</sub>) δ 3.92 (s, 3 H, OCH<sub>3</sub>), 4.05 (s, 3 H, OCH<sub>3</sub>), 6.50–6.75 (m, 2 H, ArH), 7.90 (d, 1 H, *J* = 9 Hz, ArH).

A small portion was converted to the amide **8c** by heating with concentrated ammonium hydroxide. Recrystallization from ethanol provided white needles, mp 165.5–166.5 °C (lit.<sup>16</sup> mp 166–167 °C).

**2,4-Dimethoxythiophenol (8d).** The procedure was based on the method of Field and Grunwald.<sup>18</sup> A solution of **8b** (13.2 g, 0.056 mol) in ether was added dropwise to a stirred, refluxing suspension of 11.0 g of lithium aluminum hydride in 750 mL of ether. The mixture was heated under reflux with stirring for 48 h, cooled externally with ice, and treated dropwise with 600 mL of 10% H<sub>2</sub>SO<sub>4</sub> (w/v). The resulting two-phase system was separated, and the aqueous phase was extracted with ether (2 × 200 mL). The ether extracts were pooled, washed once with water (200 mL), and flash evaporated to yield a pale amber oil with a slight sulfide smell. This was dried by the azeotropic removal of added methylene chloride and distilled through a short-path still. The final product was a colorless oil: bp 89–92 °C (0.5 mm) (8.0 g, 84% yield); NMR (CDCl<sub>3</sub>) δ 3.54 (s, 1 H, SH), 3.71 (s, 3 H, OCH<sub>3</sub>), 3.80 (s, 3 H, OCH<sub>3</sub>), 6.26–6.55 (m, 2 H, ArH), 7.15 (d of d, 1 H, ArH).

**3,4-Dimethoxythiophenol (5c).** The method of Adams and Marvel<sup>19</sup> was employed. The sulfonyl chloride **5a** (33 g, 0.14 mol) was pulverized in a mortar and added to a mixture of 55 mL of concentrated H<sub>2</sub>SO<sub>4</sub> and 900 cm<sup>3</sup> of crushed ice contained in a 2-L flask. Zinc dust (50 g, 0.77 mol) was added portionwise with vigorous stirring over a 10-min period. A condenser was attached, and the mixture was held at reflux for 1.5 h with vigorous stirring. The mixture was cooled to room temperature, decanted from unreacted zinc, and extracted with ether (3 × 150 mL). The combined extracts were washed with 40 mL of saturated aqueous NaCl, concentrated under reduced pressure, and distilled through a short-path distillation head to give 20.8 g (87%) of a colorless liquid: bp 86–88 °C (0.4 mm) [lit.<sup>20</sup> bp 138 °C (14 mm)]; NMR (CDCl<sub>3</sub>) δ 3.46 (s, 1 H, SH), 3.87 (s, 6 H, OCH<sub>3</sub>), 6.60–7.03 (m, 3 H, ArH).

**2,5-Dimethoxythiophenol (11c).** This isomer was prepared from **11a** as described for **5c**. It was obtained as a colorless liquid in 59% yield: bp 95–97 °C (0.6 mm); *n*<sub>D</sub><sup>20</sup> 1.5829 [lit.<sup>21</sup> bp 138–140 °C (14 mm); *n*<sub>D</sub><sup>20</sup> 1.5848].

**3,4-Dimethoxythioanisole (5d).**<sup>22</sup> To a nitrogen-flushed 250-mL flask was added 3,4-dimethoxythiophenol (**5c**, 10 g, 59 mmol), absolute ethanol (50 mL), and KOH (5 g, 85%, ~75 mmol) in 80 mL of ethanol. A reflux condenser was attached to the flask and methyl iodide (6 mL, ~90 mmol) was added. The reaction was exothermic, and a white precipitate formed. The solution was heated under reflux for 30 min, cooled, and poured into 200 mL of water. The mixture was extracted with ether (3 × 50 mL) and the combined extracts were washed with saturated aqueous sodium hydrosulfite. The ether was removed under reduced pressure and the residue distilled to provide 10.3 g (90%) of a colorless liquid, bp 94–95 °C (0.4 mm), that solidified on standing: mp 31–32 °C; NMR (CDCl<sub>3</sub>) δ 2.45 (s, 3 H, SCH<sub>3</sub>), 3.85 (s, 3 H, OCH<sub>3</sub>), 3.87 (s, 3 H, OCH<sub>3</sub>), 6.80–7.00 (m, 3 H, ArH).

**2,4-Dimethoxythioanisole (8e).** This isomer was prepared from **8d** as described for **5d**: yield 83%; bp 100–103 °C (0.6 mm); mp 35–37 °C [lit.<sup>17</sup> bp 115–118 °C (4 mm); mp 38–39 °C]; NMR (CDCl<sub>3</sub>) δ 2.36 (s, 3 H, SCH<sub>3</sub>), 3.77 (s, 3 H, OCH<sub>3</sub>), 3.86 (s, 3 H, OCH<sub>3</sub>), 6.37–6.61 (m, 2 H, ArH), 7.20 (d, 1 H, *J* = 9 Hz, ArH).

**2,5-Dimethoxythioanisole (11d).** This isomer was prepared from **11c** as described for **5d**. The product was a colorless oil, obtained in 83% yield by bulb-to-bulb distillation: bp 150 °C (0.6 mm) [lit.<sup>20</sup> bp 86–88 °C (0.4 mm)].

**4,5-Dimethoxy-2-methylthiobenzaldehyde (6a).** Phosphorus oxychloride (15 g, 0.10 mol) was mixed with *N*-methylformanilide (14 g, 0.10 mol) and warmed gently on the steam bath. 3,4-Dimethoxythioanisole (**5d**, 8.2 g, 42 mmol) was added portionwise

over 5 min. The reaction was exothermic, and the color changed from orange to dark red. The mixture was heated on the steam bath for 20 min and then poured into 200 mL of water. After stirring 15 min the precipitated product was collected by filtration and recrystallized from ethanol (100 mL) to give 8.05 g (85%) of off-white needles: mp 112–113 °C; NMR (CDCl<sub>3</sub>) δ 2.50 (s, 3 H, SCH<sub>3</sub>), 3.93 (s, 3 H, OCH<sub>3</sub>), 3.99 (s, 3 H, OCH<sub>3</sub>), 6.94 (s, 1 H, ArH), 7.40 (s, 1 H, ArH), 10.43 (s, 1 H, CHO). Anal. Calcd for C<sub>10</sub>H<sub>12</sub>O<sub>3</sub>S: C, 56.58; H, 5.70. Found: C, 56.85; H, 5.78.

**2,4-Dimethoxy-5-methylthiobenzaldehyde (9a).** This isomer was prepared from **8e** as described for **6a**. Recrystallization from methanol provided a 96% yield of fine white crystals, mp 124.5–125.5 °C. This compound exhibits dimorphism. In one experiment, the product melted at 109–110 °C and displayed an infrared spectrum (Nujol mull) with major peaks at 691, 734, 819, and 994 cm<sup>-1</sup>. These were lost and an infrared spectrum of the more stable higher melting form (694, 731, 839, and 897 cm<sup>-1</sup>) was obtained following intense grinding prior to mull preparation or by recrystallization from a more concentrated methanolic solution: NMR (CDCl<sub>3</sub>) δ 2.42 (s, 3 H, SCH<sub>3</sub>), 3.95 (s, 3 H, OCH<sub>3</sub>), 3.99 (s, 3 H, OCH<sub>3</sub>), 6.47 (s, 1 H, ArH), 7.68 (s, 1 H, ArH), 10.32 (s, 1 H, CHO). Anal. Calcd for C<sub>10</sub>H<sub>12</sub>O<sub>3</sub>S: C, 56.58; H, 5.70. Found: C, 56.55; H, 5.71.

**2,5-Dimethoxy-4-methylthiobenzaldehyde (12a).** This isomer was prepared in 53% yield from **11d** as described for **6a**. Off-white crystals from methanol were obtained: mp 97.5–98.5 °C. The mixture melting point with an authentic sample (lit.<sup>12</sup> mp 95–96.5 °C) was 96–96.5 °C: NMR (CDCl<sub>3</sub>) δ 2.49 (s, 3 H, SCH<sub>3</sub>), 3.90 (s, 3 H, OCH<sub>3</sub>), 3.95 (s, 3 H, OCH<sub>3</sub>), 6.75 (s, 1 H, ArH), 7.26 (s, 1 H, ArH), 10.42 (s, 1 H, CHO).

**1-(4,5-Dimethoxy-2-methylthiophenyl)-2-nitropropene (6b).** A mixture of aldehyde **6a** (2.0 g, 9.4 mmol) and ammonium acetate (0.45 g) in 8 mL of nitroethane was heated on a steam bath for 4.5 h. Removal of the excess nitroethane in vacuo gave a red oil, which upon addition of methanol (5 mL) spontaneously crystallized. The crude product (mp 98–101 °C) was recrystallized from 25 mL of boiling methanol and provided 1.85 g (73%) of bright orange crystals: mp 104–105 °C; NMR (CDCl<sub>3</sub>) δ 2.42 (br s, 3 H, CH<sub>3</sub>), 2.47 (s, 3 H, SCH<sub>3</sub>), 3.90 (s, 3 H, OCH<sub>3</sub>), 3.97 (s, 3 H, OCH<sub>3</sub>), 6.83 (s, 1 H, ArH), 7.02 (s, 1 H, ArH), 8.40 (br s, 1 H, ArCH). Anal. Calcd for C<sub>12</sub>H<sub>13</sub>NO<sub>4</sub>S: C, 53.51; H, 5.61; N, 5.20. Found: C, 53.39; H, 5.59; N, 5.19.

**1-(2,4-Dimethoxy-5-methylthiophenyl)-2-nitropropene (9b).** This isomer was prepared in 80% yield from **9a** with nitroethane as described for **6b**: mp 112–113 °C (ethanol); NMR (CDCl<sub>3</sub>) δ 2.42 (br s, 6 H, SCH<sub>3</sub>, CCH<sub>3</sub>), 3.96 (s, 3 H, OCH<sub>3</sub>), 4.00 (s, 3 H, OCH<sub>3</sub>), 6.56 (s, 1 H, ArH), 7.30 (s, 1 H, ArH), 8.30 (s, 1 H, ArCH). Anal. Calcd for C<sub>12</sub>H<sub>13</sub>NO<sub>4</sub>S: C, 53.51; H, 5.61; N, 5.20. Found: C, 53.45; H, 5.62; N, 5.21.

**1-(4,5-Dimethoxy-2-methylthiophenyl)-2-aminopropane Hydrochloride (6c).** A 200-mL three-neck flask was flushed with nitrogen and then charged with THF (50 mL) and lithium aluminum hydride (1.3 g, 34 mmol). The mixture was heated to reflux with stirring, and a solution of **6b** (1.65 g, 6.1 mmol) in THF (20 mL) was added dropwise over a 30-min period. Heating at reflux was continued overnight, while maintaining a static pressure of nitrogen. The mixture was cooled externally with ice water and the excess hydride destroyed by careful addition of a solution of 1.3 mL of water in 10 mL of THF. Aqueous NaOH (1.3 mL of 4 N) was added, followed by 3.9 mL of water. The mixture was filtered to remove the precipitated salts, and the filter cake was washed with 20 mL of THF. Concentration of the filtrate in vacuo gave a light yellow oil which was taken up in 20 mL of 2-propanol and converted to the hydrochloride by the addition of 0.9 mL of concentrated HCl. The solution was diluted with 200 mL of anhydrous ether, which resulted in the formation of pale yellow crystals (1.20 g, 71%): mp 218–219.5 °C. An analytical sample was obtained from methanol: mp 222–223 °C; NMR (D<sub>2</sub>O) δ 1.42 (d, *J* = 7 Hz, 3 H, CCH<sub>3</sub>), 2.56 (s, 3 H, SCH<sub>3</sub>), 3.12 (d, *J* = 7 Hz, CH<sub>2</sub>), 3.6–3.9 (m, 1 H, CH), 3.96 (s, 6 H, OCH<sub>3</sub>), 7.03 (s, 1 H, ArH), 7.07 (s, 1 H, ArH). Anal. Calcd for C<sub>12</sub>H<sub>20</sub>ClNO<sub>2</sub>S: C, 51.87; H, 7.25; N, 5.04. Found: C, 51.71; H, 7.13; N, 4.99.

**1-(2,4-Dimethoxy-5-methylthiophenyl)-2-aminopropane Hydrochloride (9c).** This isomer was prepared from **9b** as described for **6c**. The free base was isolated (71% yield) by short-path distillation as a colorless oil, bp 125–128 °C (0.1 mm),

that solidified on standing, mp 47–48.5 °C. The hydrochloride salt was prepared as described for **6c**: mp 140.5–142 °C; NMR (D<sub>2</sub>O) δ 1.37 (d, *J* = 7 Hz, 3 H, CCH<sub>3</sub>), 2.45 (s, 3 H, SCH<sub>3</sub>), 2.96 (d, 2 H, *J* = 7 Hz, CH<sub>2</sub>), 3.56–3.83 (m, 1 H, CH), 3.99 (s, 6 H, OCH<sub>3</sub>), 6.78 (s, 1 H, ArH), 7.23 (s, 1 H, ArH). Anal. Calcd for C<sub>12</sub>H<sub>20</sub>ClNO<sub>2</sub>S: C, 51.87; H, 7.25; N, 5.04. Found: C, 51.65; H, 7.14; N, 4.95.

**Pharmacology. Rabbit Hyperthermia.** New Zealand rabbits weighing 2.9–3.5 kg were used. Rectal temperatures were measured using a Yellow Springs thermistor and a Honeywell-Brown 12 channel recorder. The animals were housed at a constant temperature of 22 °C. During experimental sessions they were lightly restrained in metal stocks and the rectal probes were inserted about 8 cm. At the end of a 2–3 week training period during which time the rabbits were submitted to the experimental procedure without drug, only modest temperature variations (<0.2 °C) were recorded when vehicle only was administered. Groups of four rabbits were used for each dose which was administered in the marginal ear vein as a solution of the amine hydrochloride in nonpyrogenic saline. A period of at least 1 week intervened between experiments to allow the animals to fully recover from the preceding dose. Each drug was studied at various doses which, depending on the potency of the particular compound, ranged from 0.25 to 21.2 μmol/kg. The data obtained (Table I) are expressed in terms of the period of peak effect (the mean maximum temperature rise, method A; and integration of the time-temperature curve, method B). The values are also expressed in terms of potencies relative to DOM.

**Rat Brain Kinetics and Enantiomeric Composition Studies.** Male Sprague-Dawley rats, weighing 200–250 g, provided Purina Rat Chow and water ad libitum, were housed in air-conditioned quarters with automatic light regulation. All experiments were done at approximately the same time of the day. Compounds were administered intraperitoneally as their racemates: 15 mg (61.2 μmol)/kg for DOM-HCl and 30 mg (114.9 μmol)/kg for the three methylthio compounds **6c**-HCl, **9c**-HCl, and **12c**-HCl. At appropriate times after injection, the rats were decapitated in a cold room. The whole brain was isolated, washed in cold isotonic saline, blotted dry, weighted, and homogenized in 4.5 mL of 0.2 N HCl for 1 min. The homogenate was transferred to a disposable culture tube and internal standard was added [30 nmol of (S)-(+)-1-(2,5-dimethoxy-4-bromophenyl)-2-aminopropane hydrochloride<sup>23</sup> for the DOM studies and 5 nmol of (R)-(-)-DOM<sup>9</sup> for the methylthio analogue studies]. After the homogenate was made strongly basic with 10% NaOH and extracted with benzene, the isolated amines were derivatized with (S)-(-)-*N*-pentafluorobenzoylpropyl-1-imidazole (PFBPI).<sup>10,24</sup> Electron-capture gas chromatography employed a Hewlett-Packard 5713A equipped with a <sup>63</sup>Ni detector. Argon-methane (95:5) was the carrier gas with a flow rate of 10 mL/min. The column packing used was 3% SP-2250 (1 m × 2 mm) on Supelcoport. The column temperature was 250 °C for the DOM analysis and 260 °C for the three methylthio compounds. In all cases the diastereomeric pair resulting from PFBPI derivatization showed near baseline resolution. Based on past experience<sup>9,24</sup> we have tentatively assigned the fast-eluting peak to the PFBP derivative of the *R* enantiomer.

**Acknowledgment.** This research was supported by U.S. Public Health Service Research Grant MH 21219, a National Institutes of Health postdoctoral research fellowship to P.J., and PHS Training Grant 3920 to G.A.

## References and Notes

- (1) A. T. Shulgin, T. Sargent, and C. Naranjo, *Nature (London)*, 221, 537 (1969); A. T. Shulgin and D. C. Dyer, *J. Med. Chem.*, 18, 1201 (1976); A. T. Shulgin, T. Sargent, and C. Naranjo, *Pharmacology*, 5, 103 (1971).
- (2) R. Kostrzewa and D. Jacobowitz, *Pharmacol. Rev.*, 26, 199 (1974).
- (3) J. S. Zweig and N. Castagnoli, Jr., *Psychopharmacol. Commun.*, 1, 359 (1975); J. S. Zweig and N. Castagnoli, Jr., *J. Med. Chem.*, 20, 414 (1977).
- (4) B. T. Ho, V. Estevez, L. W. Tansey, L. F. Englert, P. J. Creaven, and W. M. McIsaac, *J. Med. Chem.*, 14, 158 (1971); B. T. Ho, V. Estevez, and G. E. Fritchie, *Brain Res.*, 29, 166 (1971).

- (5) J. S. Zweig and N. Castagnoli, Jr., *J. Med. Chem.*, **17**, 747 (1974); P. Jacob, III, and N. Castagnoli, Jr., unpublished data.
- (6) L. L. Butcher, *J. Neural. Transm.*, **37**, 189 (1975).
- (7) F. A. B. Aldous, B. C. Barrass, K. Brewster, D. A. Buxton, D. M. Green, R. M. Pinder, P. Rich, M. Skeels, and K. J. Tutt, *J. Med. Chem.*, **17**, 1100 (1974).
- (8) A. T. Shulgin, *J. Pharm. Pharmacol.*, **25**, 271 (1973); F. Benington, R. D. Morin, J. Beaton, J. R. Smythies, and R. J. Bradley, *Nature (London), New Biol.*, **242**, 185 (1975).
- (9) S. B. Matin, P. S. Callery, J. S. Zweig, A. O'Brien, R. Rapoport, and N. Castagnoli, Jr., *J. Med. Chem.*, **17**, 877 (1974).
- (10) N. P. McGraw, P. S. Callery, and N. Castagnoli, Jr., *J. Med. Chem.*, **20**, 185 (1977).
- (11) J. Gal, L. Gruenke, and N. Castagnoli, Jr., *J. Med. Chem.*, **18**, 683 (1975); R. Weinkam, J. Gal, P. Callery, and N. Castagnoli, Jr., *Anal. Chem.*, **48**, 203 (1976).
- (12) D. E. Nichols and A. T. Shulgin, *J. Pharm. Sci.*, **65**, 1554 (1976).
- (13) R. T. Standridge, H. G. Howell, J. A. Gyls, R. A. Partyka, and A. T. Shulgin, *J. Med. Chem.*, **19**, 1400 (1976).
- (14) C. F. Barfknecht, D. E. Nichols, and W. J. Dunn, *J. Med. Chem.*, **18**, 208 (1975).
- (15) A. Quilico, *Gazz. Chim. Ital.*, **57**, 793 (1927).
- (16) E. H. Huntress and F. H. Carten, *J. Am. Chem. Soc.*, **62**, 603 (1940).
- (17) C. M. Suter and H. L. Hansen, *J. Am. Chem. Soc.*, **55**, 2080 (1933).
- (18) L. Field and F. A. Grunwald, *J. Org. Chem.*, **16**, 946 (1951).
- (19) R. Adams and C. S. Marvel, "Organic Syntheses", Collect. Vol. I, Wiley, New York, N.Y., 1941, p 504.
- (20) K. Fries, H. Koch, and H. Stukenbrock, *Justus Liebigs Ann. Chem.*, **468**, 162 (1929).
- (21) H. Burton and E. Hoggarth, *J. Chem. Soc.*, **14** (1945).
- (22) A. Kiss, E. Vinkler, and E. Csetneky, *Acta Phys. Chem.*, **2**, 192 (1949); *Chem. Abstr.*, **44**, 5706g (1966).
- (23) D. E. Nichols, C. F. Barfknecht, and D. B. Rusterholz, *J. Med. Chem.*, **16**, 480 (1973).
- (24) S. B. Matin, M. Rowland, and N. Castagnoli, Jr., *J. Pharm. Sci.*, **62**, 821 (1973).
- (25) Although O- and S-demethylations of **6c** and **9c** would lead to thiohydroquinones, subsequent two-electron oxidations of these species to quinoid species are unlikely in view of the high energy of the carbon-sulfur  $\pi$  bond: J. B. Hendrickson, D. J. Cram, and G. S. Hammond, "Organic Chemistry", 3rd ed, McGraw-Hill, New York, N.Y., 1970, pp 790-792.

## Synthesis and Evaluation of $\alpha$ -Hydroxythiol Esters as Antitumor Agents and Glyoxalase I Inhibitors<sup>1</sup>

Stan S. Hall,\* Lidia M. Doweiko, Arthur M. Doweiko, and Judith S. Ryan Zilenovski

Carl A. Olson Laboratories, Department of Chemistry, Rutgers University, Newark, New Jersey 07102.  
Received January 31, 1977

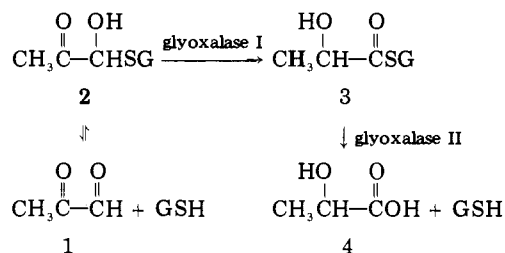
Synthesis of a series of  $\alpha$ -hydroxythiol esters made available, for the first time, product-like molecules that were evaluated as inhibitors of the enzyme glyoxalase I and as potential antitumor agents. All the  $\alpha$ -hydroxythiol esters tested were competitive inhibitors of the enzyme, albeit weak; however, the relative  $[I]_{50}$  values suggested information about the active site. Antileukemic activity in L1210 lymphoid leukemia indicated no significant activity by these  $\alpha$ -hydroxythiol esters.

The glyoxalase system, which consists of glyoxalase I [S-lactoylglutathione methylglyoxal lyase (isomerizing); E.C. 4.4.1.5], the coenzyme glutathione (GSH), and glyoxalase II (S-2-hydroxyacylglutathione hydrolase; E.C. 3.1.2.6), converts methylglyoxal (**1**) to lactic acid (**4**) (see Scheme I).

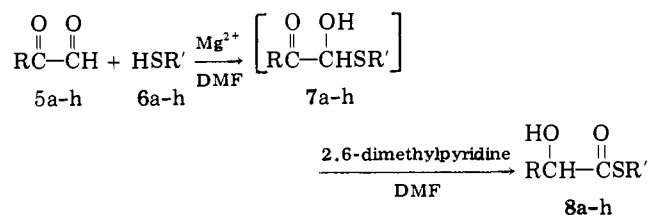
It has been suggested that the glyoxalase enzyme system, which is found widely distributed in cells of all forms of life,<sup>2,3</sup> may be involved in the regulation of cell growth by maintaining a proper concentration of one of the substrate components methylglyoxal (**1**).<sup>4,5</sup> The deficiency of this ketoaldehyde **1**<sup>6</sup> and the unusually high concentration of lactic acid<sup>7</sup> in cancer cells imply that such cells, having lost the ability to maintain a proper concentration of methylglyoxal, continue to proliferate at an unchecked rate. Selective inhibition of glyoxalase I that may result in a buildup of methylglyoxal (**1**) again in these cells might provide an effective means of chemotherapy.

Numerous studies, testing this hypothesis, have been carried out using substrate-related molecules. The carcinostatic activity of  $\alpha$ -ketoaldehydes, including methylglyoxal (**1**), is known<sup>8</sup> but not effective since these agents are rapidly metabolized to the corresponding  $\alpha$ -hydroxy acids by the glyoxalase enzyme system.<sup>4</sup> S-Alkyl derivatives and related compounds of the coenzyme glutathione (GSH) cause potent competitive inhibition of glyoxalase I<sup>4,9-11</sup> and cytotoxic activity against L1210 leukemia and KB cells in tissue culture.<sup>12</sup> However, the rapid degradation of S-substituted glutathione derivatives by glu-

Scheme I



Scheme II



tathionase and cysteinylglycinase renders many of these inhibitors ineffective when tested in vivo.<sup>13,14</sup> Consequently attempts have been made to find degradation resistant analogues.<sup>13</sup>

The discovery in this laboratory of a convenient method to prepare  $\alpha$ -hydroxythiol esters<sup>15</sup> made available a unique series of compounds that should be competitive inhibitors of the glyoxalase system and might be effective anticancer