Synthesis and Antineoplastic Evaluation of α-Substituted Alkanesulfonates: Analogues of Clomesone

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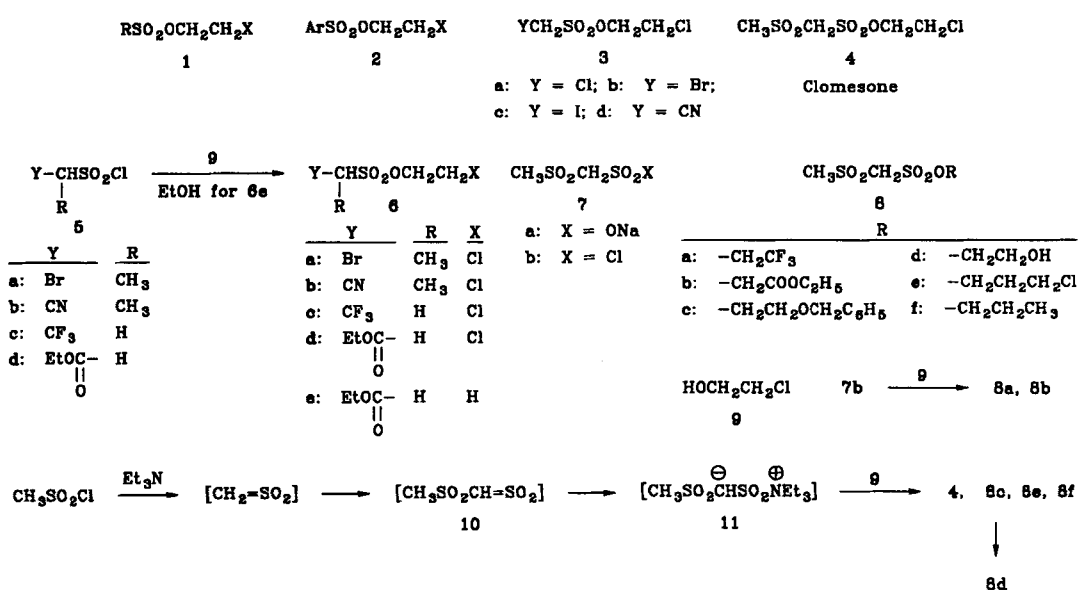
Abstract □ 2-Chloroethyl (methylsulfonyl)methanesulfonate (clomesone) is highly effective against certain experimental neoplasma and is now undergoing initial clinical trials. Two groups of analogues have been prepared to explore further the anticancer activity of this type of sulfonates. The first group is comprised of several 2-chloroethyl sulfonates that have electron-attracting groups α to the sulfonate group; among these, the α-chloroethanesulfonate and the (trifluoromethyl)-methanesulfonate caused increases in lifespan of 45 and 72%, respectively, in tests against P388 leukemia in mice. The second group is comprised of several (methylsulfonyl)methanesulfonates that possess alkylating groups other than the 2-haloethyl groups. 2-Hydroxyethyl (methylsulfonyl)methanesulfonate was active against P388 leukemia (increases in lifespan, 66 and 94%), but was less effective than clomesone, which effects cures. The 3-chloropropyl and the propyl derivatives caused modest increases in lifespan. Therefore, several 2-chloroethyl α-substituted methanesulfonates are less effective against P388 leukemia than is the α-(methylsulfonyl) derivative (clomesone), and several substituted alkyl (methylsulfonyl)methanesulfonates are also less effective than is the 2-chloroethyl derivative (clomesone). The synthesis of clomesone was simplified to one operational step from methanesulfonyl chloride.

Because of the superior effectiveness of (2-chloroethyl)triazenes and 2-chloroethyl-*N*-nitrosoureas against experimental neoplasma, 2-haloethyl sulfonates (**1**, **2**) that represent a spectrum of expected haloethylating activities were synthesized and tested against P388 leukemia in mice.[1] Arenesulfonates (**2**), regardless of the degree of expected reactivity, were ineffective. The most effective sulfonates were 2-chloroethyl methanesulfonates (**3**) that have an electron-attracting group at the α-position. Thus, 2-chloroethyl chloromethanesulfonate (**3a**) was significantly more effective than the long-known chloroethyl methanesulfonate (**1**; R = CH_3, X = Cl), which showed only minimal activity.[1] The (methylsulfonyl)methanesulfonate structure was then chosen for investigation, and 2-chloroethyl (methylsulfonyl)methanesulfonate (**4**) was shown to be vastly superior to the previously synthesized 2-haloethyl sulfonates in tests against P388 leukemia.[2] Furthermore, a group of (methylsulfonyl)methanesulfonates, including **4**, were more effective than the corresponding methanesulfonates and chloromethanesulfonates.[2] The 2-chloroethyl derivative (**4**) has been designated clomesone by the National Cancer Institute. It has been shown to be highly effective, also, against lymphoid leukemia L1210, melanotic melanoma B16, Lewis lung carcinoma, and M5076 sarcoma in mice[3,4] and is undergoing initial clinical trials.

Because of the high anticancer activity of clomesone, the effect on activity of both parts of the clomesone structure (namely, the α-substituted sulfonate part and the alkyl group) has been explored further. The synthesis and testing of some additional α-substituted 2-chloroethyl sulfonates (**6**) and some (methylsulfonyl)methanesulfonates (**8**) with alkylating groups other than 2-haloethyl groups are described in this report (Scheme I). The rationales for these two types of structure follow. On the basis of studies of the mechanism of action of 2-chloroethyl-*N*-nitrosoureas by Kohn and by others[5–8], it was expected[2] that clomesone and other active 2-chloroethyl sulfonates would chloroethylate DNA and that interstrand or intrastrand cross-linking of DNA would then occur. Unlike the chloroethylnitrosoureas, the chloroethyl



Scheme I

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sulfonates cannot cause carbamoylation,[2] which is evidently undesirable, and little, or no, 2-hydroxyethylation would be expected a priori. Investigations by Kohn, Gibson, and coworkers have shown that clomesone does indeed chloroethylate DNA.[9-11] DNA cross-links then form in DNA of cells that are deficient in DNA-guanine-O^6-alkyltransferase[9,12,13] or in isolated DNA treated with clomesone.[14,15] There are differences, however, between the chloroethylnitrosoureas and clomesone in alkylation and in cross-linking of DNA.[10-17] The new 2-chloroethyl derivatives represented by structure **6** have electron-attracting groups at the α-position of the sulfonate moiety and permit further investigations of the antineoplastic activity of this type of structure. It has been suggested that interstrand DNA–DNA cross-linking might not be the primary mechanism of cytotoxicity[9,17]; the (methylsulfonyl)methanesulfonates (**8**) are derivatives that can alkylate but cannot cross-link.

Results and Discussion

Chemistry—The original synthesis of clomesone (**4**) began with methanesulfonyl chloride and proceeded via the isolated intermediates sodium (methylsulfonyl)methanesulfonate (**7a**) and (methylsulfonyl)methanesulfonyl chloride (**7b**). Clomesone was obtained in yields of 45–50% by treating **7b** in ethyl acetate with 1.1 equivalents of 2-chloroethanol (**9**) and 1.2–1.3 equivalents of triethylamine. It appeared likely that clomesone could be formed directly from (methylsulfonyl)-sulfene (**10**) or its triethylamine adduct (**11**), which are intermediates in situ.[2,18,19] This possibility proved to be practicable; thus, clomesone was obtained in one operational step in yields of 50–60% by treating a cold solution of methanesulfonyl chloride, triethylamine, and acetonitrile with 2-chloroethanol.

The 2-chloroethyl α-substituted-sulfonates (**6a–d**) were prepared from the appropriate α-substituted sulfonyl chloride (**5**) and 2-chloroethanol, and the ethyl sulfonate **6e** was prepared from **5d** and ethanol. (Methylsulfonyl)methanesulfonates **8a** and **8b** were prepared similarly from **7b**. The (benzyloxy)ethyl (**8c**), 3-chloropropyl (**8e**), and propyl (**8f**) (methylsulfonyl)methanesulfonates were prepared from methanesulfonyl chloride by the one-step procedure described for the improved synthesis of clomesone. The (benzyloxy)ethyl derivative (**8c**) was prepared to serve as a precursor of 2-hydroxyethyl (methylsulfonyl)sulfonate (**8d**), which was obtained by hydrogenolysis of **8c**.

Biological Evaluation—Because the bromomethanesulfonate (**3b**) and the cyanomethanesulfonate (**3d**) increased the lifespan of mice inoculated intraperitoneally with P388 leukemia,[1] the analogous 2-chloroethyl ethanesulfonates (**6a** and **6b**) were prepared for testing. Similar chloroethyl sulfonates with the trifluoromethyl (**6c**) or the ethoxycarbonyl (**6d**) groups as the electron-attracting centers at the α-position were also prepared. In tests against P388 leukemia performed in accordance with standard protocols, the cyanoethanesulfonate (**6b**) was active at 50 mg/kg/day [ratio of treated to control (T/C), 145 and 172%], and the 2,2,2-trifluoroethanesulfonate (**6c**) increased lifespan in two tests at 100 mg/kg/day (T/C, 145 and 138%). For comparison, results of tests of clomesone at two doses against P388 leukemia, according to the same protocols, are summarized in Table I.

Further variations of the alkylating moiety of the (methylsulfonyl)methanesulfonate structure are represented by structure **8**. Compounds **8a** and **8b** possess potential two-carbon alkylating moieties; they were not active in routine tests against P388 leukemia. The clomesone homologue (the chloropropyl derivative **8e**) displayed only minimal activity in tests against P388 leukemia (T/C, 131 and 135% at 200 mg/kg/day), whereas similar tests with clomesone had resulted in survival of some of the treated animals until the tests were terminated at 60 days.[2] The T/C ratios resulting from tests of **8e** were not significantly different from those from tests of the propyl derivative (**8f**).

Interactions of (2-chloroethyl)nitrosoureas and their transformation products with DNA result in 2-hydroxyethylation as well as 2-chloroethylation. 2-Hydroxyethylation is a major result of these interactions,[11,20,21] but 2-hydroxyethylation evidently is not responsible for antitumor activity.[10] Clomesone cannot generate the intermediates that form from chloroethylnitrosoureas and that evidently are responsible for 2-hydroxyethylation of DNA.[22-24] 2-Hydroxyethyl (methylsulfonyl)methanesulfonate (**8d**) was prepared to determine whether the clomesone analogue would have activity. Compound **8d** increased lifespan in tests against P388 leukemia at doses of 50, 100, and 200 mg/kg/day, the values of T/C being 166 and 194% in two tests at the latter dose (Table I).

Conclusion—In tests versus P388 leukemia in mice, none of the sulfonate moieties reported here or previously[1] are superior to the (methylsulfonyl)methanesulfonate group as the carrier group of the chloroethyl group, and none of the alkylating groups (ref 2 and structure **8**) attached to the (methylsulfonyl)methanesulfonate group are superior to the chloroethyl group.

Experimental Section

General Methods—Melting points (mp) were determined in capillary tubes heated in a Mel-Temp apparatus. MS data were taken from low-resolution, electron-impact spectra determined at 70 eV with a Varian/MAT 311A spectrometer; M = molecular ion ($M^{+\cdot}$). Other MS peaks were assigned to fragments [(M minus a fragment)$^+$ or (fragment)$^{+\cdot}$] that are represented for simplicity as uncharged species. IR absorption data were taken from spectra recorded with a Nicolet 10 MXE spectrometer; s = strong, vs = very strong, br = broad, sh = shoulder. The IR spectra of solids were recorded from specimens in pressed KBr disks; spectra of liquids or low-melting solids were recorded from capillary films on KBr windows. NMR spectra were determined at 300.64 MHz with a Nicolet 300 NB NMR spectrometer or at 100.1 MHz with a Varian model XL-100-15 NMR spectrometer. Tetramethylsilane was the internal standard, and the solvent was $CDCl_3$ or deuterated dimethylsulfoxide. The apparent multiplicity of the signals and the assigned positions of the protons are listed parenthetically with the chemical shifts; s = singlet, t = triplet, m = multiplet, qt = quartet, qn = quintet. Specimens of the sulfonate esters were protected from atmospheric moisture and were stored in a freezer.

2-Chloroethyl (Methylsulfonyl)methanesulfonate (Clomesone, 4): Synthesis in One Operational Step from Methanesulfonyl Chloride—A solution of 133 g (1.32 mol) of triethylamine in 400 mL of anhydrous acetonitrile was cooled to −30–−40 °C, and a solution of 100 g (0.88 mol) of methanesulfonyl chloride in anhydrous acetonitrile (67 mL) was added in a dropwise manner at a rate that prevented the temperature from rising above −30 °C. The mixture was stirred at −30–−40 °C for 1 h, and a solution of 35.4 g (0.44 mol) of anhydrous 2-chloroethanol in anhydrous acetonitrile (30 mL) was added in a dropwise manner. The resulting mixture was stirred for 2 h at −30–−40 °C and then was filtered to separate triethylamine hydrochloride. The latter material was washed with ethyl acetate, the washings were combined with the filtrate, and the organic solution was concentrated under reduced pressure to an oil. A solution of the residual oil in ethyl acetate (1200 mL) was washed quickly with two portions (2 × 200 mL) of dilute NaCl solution and then with two portions (2 × 200 mL) of saturated NaCl solution, dried ($MgSO_4$), filtered, and concentrated under reduced pressure. The residual solid (70 g) was dissolved in methylene chloride (450 mL), and the solution was filtered and diluted slowly with cyclohexane (450 mL). The mixture, protected from atmospheric moisture, was allowed to stand at room temperature for ~1 h and was then stored at low temperatures (~5 °C) overnight. The precipitate was collected by filtration, washed with cyclohexane, and dried under reduced pressure over phosphorus pentoxide: yield, 58.7 g (56%); mp, 60–62 °C; ^1H NMR (300.64 MHz, $CDCl_3$): δ 3.26 (s, CH_3), 3.80 (t, CH_2Cl), 4.65 (t, OCH_2), 4.67 (s, SCH_2S). The IR and mass spectra were identical with those of **4** prepared by the route reported previously.[2]

Table I—Chloroethyl α-Substituted-Sulfonates and (Methylsulfonyl)methanesulfonic Esters versus P388 Leukemia In Vivo[a,b]

Compound	Dose, mg/kg/day (q.d. 1–5)	Deaths by Day 5/Total[c]	Wt. Change Diff. By Day 5[d] (T − C), g	Survival Time[e]	
				Days, T/C	T/C Ratio, %
6a	200	6/6	—[f]	t[g]	—
	100	0/6	−0.6	12.3/12.0	102
	50	0/6	0.0	12.3/12.0	102
	25	0/6	0.23	11.8/12.0	98
6b	100	6/6	—	t	—
	50	0/6	−2.2	16.3/11.2	145
	50	0/6	−1.4	19.8/11.5	172
	25	0/6	−1.1	14.8/11.5	128
	12.5	0/6	0.3	13.9/11.5	120
6c	200	6/6	—	t	—
	100	0/6	−3.6	15.3/10.5	145
	50	0/6	−1.1	14.0/10.5	133
	25	0/6	−0.3	12.3/10.5	117
	100	0/6	−3.3	18.0/13.0	138
	50	0/6	−1.1	13.3/13.0	102
	25	0/5	−0.2	13.5/13.0	103
6d	200	0/6	−1.9	12.3/11.7	105
	100	1/6	−0.1	12.7/11.7	108
	50	0/6	−1.1	12.0/11.7	102
6e	200	0/6	−0.7	12.0/11.0	109
	100	0/6	0.0	11.0/11.0	100
	50	0/6	−0.2	11.8/11.0	107
8a	200	4/6	—	t	—
	100	0/6	0.2	10.8/10.3	104
	50	0/6	−0.8	11.0/10.3	106
8b	200	6/6	—	t	—
	100	2/6	−5.2t	12.3/11.2	109
	50	0/6	−3.6	12.4/11.2	110
	50	0/6	−2.7	11.7/11.2	104
	25	0/6	−1.7	11.0/11.2	98
	12.5	0/6	−0.8	12.1/11.2	108
8c	200	1/6	−3.4	12.9/11.9	108
	100	0/6	−2.4	12.1/11.9	101
	50	0/6	0.0	11.8/11.9	99
	25	0/6	−0.2	13.0/11.9	109
8d	200	2/6	−2.9	18.8/11.3	166
	100	0/6	−0.6	17.0/11.3	150
	50	0/6	−3.0	14.8/11.3	130
	25	0/6	−1.1	13.7/11.3	121
	200	0/6	−3.7	21.0/10.8	194
	100	0/6	−1.6	17.7/10.8	163
	50	0/6	−0.9	14.8/10.8	137
	25	0/6	−0.7	12.3/10.8	113
8e	400	5/6	—	t	—
	200	0/6	−1.3	13.9/10.6	131
	100	0/6	−0.5	12.3/10.6	116
	50	0/6	0.3	11.4/10.6	107
	200	0/6	−2.5	14.8/10.9	135
	100	0/6	0.1	11.4/10.9	104
	50	0/6	−0.8	12.3/10.9	112
	25	0/6	0.1	10.8/10.9	99
8f	200	0/6	−4.9t	14.0/11.0	127
	100	0/6	−1.7	12.8/11.0	116
	50	0/6	−0.3	11.3/11.0	102
	25	0/6	−0.2	11.9/11.0	108
	400	6/6	—	t	—
	200	0/6	−2.0	13.0/11.3	115
	100	0/6	−0.9	11.3/11.3	100
	50	0/6	−0.5	12.0/11.3	106
Clomesome[2,3]					
6 Experiments (5 or 6 mice/expt.)	33	0/34	−1.5–−2.8	—	179–209
5 Experiments (6 mice/expt.)	50	0/30	−1.6–−3.4	2/6–6/6[h]	225–335[i]

[a] Mice were inoculated with 10^6 P388 leukemia cells on Day 0; C = control group of animals; T = treated animals. [b] Compounds were administered *intraperitoneally within ~5 min* of the time of preparation of solutions or suspensions (5-min unstable compound); **6a**, **6b**, **6d**, **6e**, **8a**, **8c**, and **8e** were administered in saline plus polysorbate 80; **6e**, **8b**, and **8d** were administered in saline; and **8f** was administered in aqueous hydroxypropyl cellulose. [c] Number of treated mice dead on or before day 5/number of treated mice. [d] Average change in weight by day 5 of treated mice minus average change in weight of control mice. [e] A compound is considered[30] to be toxic (t) if mortality by day 5 ≥ 3 of 6, T/C <85%, or the weight change difference (T − C) is greater in magnitude than −4g. [f] —, Not applicable. [g] t, Compound is toxic. [h] 60-Day survivors. [i] Percent T/C for non-survivors.

2-Chloroethyl α-Bromoethanesulfonate (6a)—A solution of 2.13 g (26.5 mmol) of 2-chloroethanol, 2.92 g (28.9 mmol) of dry triethylamine, and 5 mL of ethyl acetate was added in a dropwise manner during 20 min to a solution, chilled in an ice bath, of 5.0 g (24.1 mmol) of α-bromoethanesulfonyl chloride[25] (**5a**) in 50 mL of ethyl acetate. The mixture was stirred during 4 h in the ice bath, and the triethylamine hydrochloride was separated by filtration and washed with ethyl acetate. The filtrate (plus the washings) was washed with saturated aqueous NaCl solution (3 × 20 mL), dried ($MgSO_4$), filtered, and concentrated under reduced pressure to an oil; 5.4 g (89% yield). The oil was distilled: yield, 3.5 g (58%); bp, 79–80.5 °C at 0.01 mm Hg; MS (direct-probe temperature 20 °C): *m/z* 251 (M + H), 201 and 203 (M − CH_2Cl), 186 (M − CH_2CH_2Cl + H), 171 (M − Br), 127 ($ClCH_2CH_2OSO$), 107 and 109 (CH_3CHBr), 79 (CH_2CH_2Cl), 63 ($ClCH_2CH_2$); IR (strong and medium bands): 2975, 1445, 1385 sh, 1363 vs, 1305, 1189, 1167 vs, 1151 s, 1069, 1052, 1000 s, 955 s, 905 s, 778, 715, 670, 635, and 570 cm^{-1}; ^1H NMR (100.1 MHz, $CDCl_3$): δ 2.09 (d, CH_3), 3.78 (t, CH_2Cl), 4.60 (t, OCH_2), 5.00 (qt, CH).

Anal.—Calcd for $C_4H_8BrClO_3S$: C, 19.10; H, 3.21. Found: C, 19.44; H, 3.20.

2-Chloroethyl α-Cyanoethanesulfonate (6b)—Compound **6b** was prepared from α-cyanoethanesulfonyl chloride[26] by a procedure similar to that described for the preparation of **6a**: yield after distillation, 41%; bp, 79–80 °C at 0.02 mm Hg; MS (direct-probe temperature 20 °C): *m/z* 198 (M + 1), 162 (M − Cl), 157, 148 (M − CH_2Cl), 127 ($ClCH_2CH_2OSO$), 117, 93, 63 ($ClCH_2CH_2$), 54 (CH_3CHCN); IR (strong and medium bands), 2940, 1455, 1391 s, 1372 vs, 1307, 1197, 1180 vs, 1166 s, 1069, 995 vs, 956 vs, 908 s, 777, 715, and 601 cm^{-1}; weak IR band, 2255 cm^{-1} (CN); ^1H NMR (100.1 MHz, $CDCl_3$): δ 1.85 (d, CH_3), 3.81 (t, CH_2Cl), 4.22 (qt, CH), 4.68 (t, OCH_2).

Anal.—Calcd for $C_5H_8ClNO_3S$: C, 30.38; H, 4.08; N, 7.09. Found: C, 30.25; H, 4.06; N, 6.86.

2-Chloroethyl 2,2,2-Trifluoroethanesulfonate (6c)—Compound **6c** was prepared from 2,2,2-trifluoroethanesulfonyl chloride[27] by a procedure similar to that described for the preparation of **6a**: yield after distillation, 37%; bp, 80–81 °C at 0.5 mm Hg; IR (strong and medium bands): 3015, 2965, 1393 vs, 1379 vs, 1332 vs, 1273 s, 1257 vs, 1183 vs, 1143 vs, 1092 s, 1000 s, 959 s, 912, 674, and 592 cm^{-1}; ^1H NMR (100.1 MHz, $CDCl_3$): δ 3.77 (t, CH_2Cl), 3.98 (m, CF_3CH_2), 4.56 (t, OCH_2).

Anal.—Calcd for $C_4H_6ClF_3O_3S$: C, 21.20; H, 2.67. Found: C, 21.34; H, 2.94.

Ethyl [(2-Chloroethyloxy)sulfonyl]acetate (6d)—Compound **6d** was prepared from ethyl (chlorosulfonyl)acetate[28] by a procedure similar to that described for the preparation of **6a**: yield after distillation, 65%; bp, 107–107.5 at 0.01 mm Hg; MS (direct-probe temperature 20 °C): *m/z* 231 (M + H), 185 (M − OEt), 181 (M − CH_2Cl), 167 ($EtOCOCH_2SO_3$), 151 ($EtOCOCH_2SO_2$), 63 ($ClCH_2CH_2$); IR (strong and medium bands): 2985, 2960 br, 1745 vs, 1371 vs, 1306 s, 1176 s, 1128, 1070, 1024, 1002 s, 960, and 914 br cm^{-1}; ^1H NMR (100.1 MHz, $CDCl_3$): δ 1.34 (t, CH_3CH_2OCO), 3.78 (t, CH_2Cl), 4.18 (s, $OCCH_2S$), 4.30 (qt, CH_2CH_3), 4.57 (t, OCH_2CH_2Cl).

Anal.—Calcd for $C_6H_{11}ClO_5S$: C, 31.24; H, 4.81. Found: C, 31.32; H, 4.91.

Ethyl [(Ethyloxy)sulfonyl]acetate (6e)—Compound **6e** was prepared by a procedure similar to that described for the preparation of **6a**: yield after distillation, 82%; bp, 74–74.5 °C at 0.005–0.001 mm Hg; MS (~150 °C): *m/z* 196 (M), 181 (M − CH_3), 151 ($EtOCOCH_2SO_2$), 123 (M − COOEt); IR (strong and medium bands): 2990, 2960, 2945, 1746 vs, 1390 sh, 1373 vs, 1362 s, 1291 s, 1177 s, 1126, 1026, 1001 s, 930 s, 915 sh, 805, 625, and 525 cm^{-1}; ^1H NMR (100.1 MHz, $CDCl_3$): δ 1.33 (t, CH_3CH_2OCO), 1.44 (t, $SO_2OCH_2CH_3$), 4.10 (s, $OCCH_2S$), 4.28 (t, CH_3CH_2OCO), 4.44 (t, $SO_2OCH_2CH_3$).

Anal.—Calcd for $C_6H_{12}O_5S$: C, 36.72; H, 6.16. Found: C, 36.74; H, 6.32.

2,2,2-Trifluoroethyl (Methylsulfonyl)methanesulfonate (8a)—A solution of 1.71 g (17.1 mmol) of 2,2,2-trifluoroethanol, 1.88 g (18.7 mmol) of triethylamine, and 5 mL of ethyl acetate was added in a dropwise manner for 20 min to a solution, chilled in an ice bath, of 3.0 g (15.6 mmol) of (methylsulfonyl)methanesulfonyl chloride[2] (**7b**) in 20 mL of ethyl acetate. The mixture was stirred in the ice bath for 3 h and then stored in a refrigerator overnight, and the triethylamine hydrochloride was separated by filtration and washed with ethyl acetate. The filtrate (plus washings) was washed successively with cold water (2 × 20 mL) and saturated NaCl solution (2 × 20 mL), dried ($MgSO_4$), filtered, and concentrated under reduced pressure to a solid residue: yield of crude product, 3.0 g (75%); mp, 65–68 °C. A solution of the residue in ethyl acetate was filtered, diluted with hexane, and stored in a refrigerator overnight. The crystalline product was collected by filtration and washed with hexane: yield, 1.4 g (35%); mp, 74–76 °C; MS (direct-probe temperature, 100 °C): *m/z* 257 (M + H), 256 (M), 241 (M − CH_3), 237 (M − F), 187 ($CH_3SO_2CH_2SO_2OCH_2$), 178, 162 ($SO_2OCH_2CF_3$ − H), 157 ($CH_3SO_2CH_2SO_2$), 147, 94 (CH_2SO_3), 83 (CH_2CF_3), 79 (CH_3SO_2), 69 (CF_3), 63 (CH_3SO); IR (medium and strong bands): 2990, 2925, 1375 vs, 1317 vs, 1307 vs, 1275, 1240, 1186 vs, 1148 s, 1130, 1042 s, 960 s, 873, 862 s, 835, 775, 768, 587, 525, 503, and 465 cm^{-1}; ^1H NMR (100.1 MHz, Me_2SO-d_6): δ 3.25 (s, CH_3SO_2), 5.04 (m, CH_2CF_3), 5.89 (s, SCH_2S).

Anal.—Calcd for $C_4H_7F_3O_5S_2$: C, 18.75; H, 2.75. Found: C, 19.01; H, 2.81.

Ethyl [(Methylsulfonyl)methanesulfonyl]oxyacetate (8b)—Compound **8b** was prepared from ethyl hydroxyacetate and **7b** by the procedure described for the preparation of **8a**. The crude product (65% yield) was dissolved in ethyl acetate:hexane and purified by chromatography on silica gel, with ethyl acetate:hexane (2:1) being used for development and elution: yield, 35%; mp, 58–60 °C; MS (direct-probe temperature, 220 °C): *m/z* 260 (M), 215 (M − OEt), 187 ($CH_3SO_2CH_2SO_2OCH_2$), 157 ($CH_3SO_2CH_2SO_2$), 94 (CH_2SO_3), 79 (CH_3SO_2), 63 (CH_3SO); IR (strong and medium bands): 2977, 2912, 1750 vs, 1420, 1375 vs, 1324, 1302 s, 1297 s, 1253 s, 1247 s, 1179 vs, 1127, 1120, 1043 s, 1012, 975, 878 vs, 805 s, 515 s, and 465 cm^{-1}; ^1H NMR (100.1 MHz, Me_2SO-d_6): δ 1.23 (t, CH_2CH_3), 3.23 (s, CH_3SO_2), 4.22 (qt, CH_2CH_3), 5.01 (s, SO_3CH_2), 5.72 (s, SCH_2S).

Anal.—Calcd for $C_6H_{12}O_7S_2$: C, 27.68; H, 4.65. Found: C, 27.62; H, 4.58.

2-[(Phenylmethyl)oxy]ethyl(Methylsulfonyl)methanesulfonate (8c)—Compound **8c** was prepared from 125 mmol of methanesulfonyl chloride, 188 mmol of triethylamine, and 65.7 mmol of 2-[(phenylmethyl)oxy]ethanol by the one-step procedure described for the preparation of clomesone (4). The light yellow oil obtained by concentrating the acetonitrile:ethyl acetate solution under reduced pressure was dissolved in ethyl acetate, and the solution was washed quickly with water and with saturated aqueous NaCl, filtered, dried ($MgSO_4$), and concentrated under reduced pressure to a light yellow oil (14.0 g). The crude product was dissolved in ethyl acetate:hexane (3:1) and subjected to flash chromatography on silica gel. The fractions that contained only **8c**, determined by TLC, were combined and concentrated under reduced pressure. The residual oil was stored at low temperatures under argon, to deter discoloration, and then crystallized from ethyl acetate:cyclohexane: yield of white needles, 7.0 g (34%); mp, 60–62 °C; MS (direct-probe temperature, 20 °C): *m/z* 308 (M), 201 (M − $OCH_2C_6H_5$), 187 ($CH_3SO_2CH_2SO_2OCH_2$), 175 ($CH_3SO_2CH_2SO_3$ + 2H), 157 ($CH_3SO_2CH_2SO_2$), 107 ($C_6H_5CH_2O$), 91 ($C_6H_5CH_2$), 79 (CH_3SO_2), 63 (CH_3SO); IR (strong and medium bands): 2995, 2932, 1384 s, 1374 s, 1369 s, 1360, 1315 vs, 1184 vs, 1165 vs, 1128, 1120, 1002, 932 s, 923 s, 870, 804, and 742 cm^{-1}; ^1H NMR (300.64 MHz, $CDCl_3$): δ 3.13 (s, CH_3SO_2), 3.77 (m, $SO_3CH_2CH_2O$), 4.59 (m, $SO_3CH_2CH_2O$ + $OCH_2C_6H_5$), 4.73 (s, SCH_2S), 7.45 (m, C_6H_5).

Anal.—Calcd for $C_{11}H_{16}O_2S_2$: C, 42.84; H, 5.23. Found: C, 42.61; H, 5.26.

2-Hydroxyethyl (Methylsulfonyl)methanesulfonate (8d)—A mixture of 2.0 g of **8c**, 40 mL of ethyl acetate, and 100 mg of 5% palladium–charcoal catalyst was stirred in an atmosphere of hydrogen at atmospheric pressure. After the initial uptake of hydrogen had subsided, a second portion of the catalyst (150 mg) was added and the stirring was continued. The rate of hydrogenolysis was slow at first, then increased, and finally declined until the uptake of hydrogen essentially ceased. The catalyst was separated by filtration and washed with ethyl acetate, and the filtrate (plus washings) was concentrated to an oil that was stored at low temperatures under argon. The product crystallized to a white solid and was dried thoroughly under reduced pressure (oil pump) at room temperature: yield, 1.4 g (99%); mp, 36–39 °C; MS (direct-probe temperature, 20 °C): *m/z* 219 (M + H), 201 (M − OH), 188 (M − CH_2OH + H), 175 ($CH_3SO_2CH_2SO_3$ + 2H), 157 ($CH_3SO_2CH_2SO_2$), 124 ($SO_3CH_2CH_2OH$ − H), 94 (CH_2SO_3), 79 (CH_3SO_2), 63 (CH_3SO); IR: 3540 br, 3365 br, 2985, 2930, 1370 sh, 1362 s, 1320 vs, 1240, 1183 vs, 1150 vs, 1120, 1080, 986 s, 924 vs, 860, 796, 510, and 468 cm^{-1}; ^1H NMR (300.64 MHz, $CDCl_3$): δ 3.27 (s, CH_3SO_2), 3.95 (m, CH_2OH), 4.56 (m, $SO_3CH_2CH_2$), 4.74 (s, SCH_2S).

Anal.—Calcd for $C_4H_{10}O_6S_2$: C, 22.01; H, 4.62. Found: C, 22.33; H, 4.94.

3-Chloropropyl (Methylsulfonyl)methanesulfonate (8e)—Compound 8e was prepared by the one-step procedure described for the preparation of clomesone (4). The residual oil obtained by concentrating the ethyl acetate solution under reduced pressure was crystallized from dichloromethane:cyclohexane: yield, 53%; mp, 58–59 °C; MS (direct-probe temperature, 20 °C): *m/z* 251 (M + H), 201 (M − CH_2Cl), 187 ($CH_3SO_2CH_2SO_2OCH_2$), 175 ($CH_3SO_2CH_2SO_3$ + 2H), 157 ($CH_3SO_2CH_2SO_2$), 109 ($CH_2SO_2OCH_2$ + H), 94 (CH_2SO_3), 80 (SO_3), 79 (CH_3SO_2), 63 (CH_3SO); IR (medium and strong bands): 2996 s, 2934, 1370 vs, 1355, 1329 vs, 1285, 1181 vs, 1174 s, 1166 s, 925 vs, 858 s, 778, 760, 602, 524, and 505 cm^{-1}; ^1H NMR (300.64 MHz, $CDCl_3$): δ 2.26 (qn, $CH_2CH_2CH_2$), 3.26 (s, CH_3SO_2), 3.71 (t, CH_2Cl), 4.61 (t, OCH_2), 4.63 (s, SCH_2S).

Anal.—Calcd for $C_5H_{11}ClO_5S_2$: C, 23.95; H, 4.42. Found: C, 24.16; H, 4.46.

Propyl (Methylsulfonyl)methanesulfonate (8f)—Compound 8f was prepared by the one-step procedure described for the preparation of clomesone (4). The crude solid obtained by concentrating the ethyl acetate solution was dissolved in ethyl acetate:hexane (2:1) and subjected to flash chromatography on silica gel. Fractions that contained 8f, determined by TLC with detection of spots with iodine vapor, were combined and concentrated under reduced pressure. The residue (51% yield) was recrystallized from dichloromethane/cyclohexane: yield, 33%; mp, 41–44 °C; MS (direct-probe temperature, 20 °C): *m/z* 217 (M + H), 215 (M − H), 201 (M − CH_3), 187 ($CH_3SO_2CH_2SO_2OCH_2$), 175 ($CH_3SO_2CH_2SO_3$ + 2H), 157 ($CH_3SO_2CH_2SO_2$), 109 ($CH_2SO_2OCH_2$ + H), 94 (CH_2SO_3), 80 (SO_3), 79 (CH_3SO_2), 63 (CH_3SO); IR (medium and strong bands): 2994 s, 2931 s, 1397, 1375 vs, 1314 vs, 1245, 1186 vs, 1164 vs, 1130, 1124, 936 vs, 915, 870 s, 848, 780, 528, 502, and 460 cm^{-1}; ^1H NMR (300.64 MHz, $CDCl_3$): δ 1.03 (t, $CH_2CH_2CH_3$), 1.84 (m, $CH_2CH_2CH_3$), 3.24 (s, CH_3SO_2), 4.41 (t, $CH_2CH_2CH_3$), 4.59 (s, SCH_2S); ^{13}C NMR (75.6 MHz, $CDCl_3$): δ 9.84, 22.59, 42.13, 68.01, 75.64.

Anal.—Calcd for $C_5H_{12}O_5S_2$: C, 27.77; H, 5.59. Found: C, 27.93; H, 5.87.

2-[(Phenylmethyl)oxy]ethanol[29]—2-[(Phenylmethyl)oxy]ethanol was prepared by heating a mixture of ethyleneglycol (100 g), sodium methoxide, and benzyl chloride (75 g) at 155 °C: yield, 54 g (66%); bp, 82–83 °C at 0.1 mm Hg; MS: *m/z* 152 (M), 107 ($C_6H_5CH_2O$), 91 ($C_6H_5CH_2$).

Tests Against P388 Leukemia—These tests were performed as described previously.[2,30]

References and Notes

1. Shealy, Y. F.; Krauth, C. A.; Struck, R. F.; Montgomery, J. A. *J. Med. Chem.* **1983**, *26*, 1168–1173.
2. Shealy, Y. F.; Krauth, C. A.; Laster, W. R., Jr. *J. Med. Chem.* **1984**, *27*, 664–670.
3. Shealy, Y. F.; Krauth, C. A. U.S. Patent 4 611 074, September 9, 1986.
4. Dykes, D. J.; Waud, W. R.; Harrison, S. D., Jr.; Laster, W. R., Jr.; Griswold, D. P., Jr.; Shealy, Y. F.; Montgomery, J. A. *Cancer Res.* **1989**, *49*, 1182–1186.
5. Kohn, K. W. *Cancer Res.* **1977**, *37*, 1450–1454.
6. Thomas, C. B.; Osieka, R.; Kohn, K. W. *Cancer Res.* **1978**, *38*, 2448–2454.
7. Lown, J. W.; McLaughlin, L. W.; Chang, Y.-M. *Bioorg. Chem.* **1978**, *7*, 97–110.
8. cf. Other publications cited in ref 2.
9. Gibson, N. W.; Erickson, L. C.; Kohn, K. W. *Cancer Res.* **1985**, *45*, 1674–1679.
10. Gibson, N. W.; Hartley, J. A.; Strong, J. M.; Kohn, K. W. *Cancer Res.* **1986**, *46*, 553–557.
11. Hartley, J. A.; Gibson, N. W.; Kohn, K. W.; Mattes, W. B. *Cancer Res.* **1986**, *46*, 1943–1947.
12. Hartley, J. A.; Gibson, N. W. *Cancer Res.* **1986**, *46*, 3871–3875.
13. Dolan, M. E.; Pegg, A. E.; Hora, N. K.; Erickson, L. C. *Cancer Res.* **1988**, *48*, 3603–3606.
14. Brent, T. P.; Lestrud, S. O.; Smith, D. G.; Remack, J. S. *Cancer Res.* **1987**, *47*, 3384–3387.
15. Buckley, N.; Brent, T. P. *J. Am. Chem. Soc.* **1988**, *110*, 7520–7529.
16. Dolan, M. E.; Young, G. S.; Pegg, A. S. *Cancer Res.* **1986**, *46*, 4500–4504.
17. Alexander, J. A.; Bowden, B. J.; Wheeler, G. P. *Cancer Res.* **1986**, *46*, 6024–6028.
18. Opitz, G.; Kleeman, M.; Bücher, D.; Walz, G.; Rieth, K. *Angew. Chem. Int. Ed.* **1966**, *5*, 594–595.
19. Senning, A. *Synthesis* **1973**, 211–212.
20. Ludlum, D. B.; Tong, W. P. In *Nitrosoureas in Cancer Treatment*, INSERM Symposium No. 19; Serrou, B.; Schein, P. S.; Imbach, J.-L., Eds.; Elsevier/North Holland Biomedical: Amsterdam, The Netherlands, 1981; pp 21–31.
21. Tong, W. P.; Kohn, K. W.; Ludlum, D. B. *Cancer Res.* **1982**, *42*, 4460–4464.
22. Lown, J. W.; Chauhan, S. M. S. *J. Med. Chem.* **1981**, *24*, 270–279.
23. Interaction of clomesone with DNA was not expected to produce products of hydroxyethylation;[2] subsequently, a very small amount of (2-hydroxyethylguanine was identified, in addition to the much greater amount of 7-(2-chloroethyl)guanine, after interaction of clomesone with DNA.[24] This low degree of hydroxyethylation evidently proceeds by a mechanism that is different from the process that results in much more hydroxyethylation by (2-chloroethyl)nitrosoureas.
24. Struck, R. F.; Alexander, J. A.; McCain, D. M.; Shealy, Y. F.; Rose, L. M. *Biochem. Pharmacol.* **1991**, *41*, 457–459.
25. Carpino, L. A.; McAdams, L. V., III; Rynbrandt, R. H.; Spiewak, J. W. *J. Am. Chem. Soc.* **1971**, *93*, 476–484.
26. Sammes, M. P.; Wylie, C. M.; Hoggett, J. G. *J. Chem. Soc. (C)* **1971**, 2151–2155.
27. Crossland, R. K.; Wells, W. E.; Shiner, V. J., Jr. *J. Am. Chem. Soc.* **1971**, *93*, 4217–4219.
28. Vieillefosse, R. *Bull. Soc. Chim. Fr.* **1947**, 351–358.
29. Bennett, G. M. *J. Chem. Soc.* **1925**, *127*, 1277–1282.
30. Geran, R. I.; Greenberg, N. H.; Macdonald, M. M.; Schumacher, A. M.; Abbott, B. J. *Cancer Chemother. Rept., Part 3* **1972**, *3*, 1–103.

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