designations for 13 result from arbitrarily focusing upon the stereochemical disposition of the phosphoryl oxygen in this bicyclic structure. For the sake of simplicity in this paper, only one enantiomer of 11 and 13 is shown, and in other chiral molecules no attempt is made to specify relative and/or absolute stereochemical relationships.
(18) P. Savignac, J. Chenault, and M. Dreux, *J. Organomet. Chem.*, **66**, 63 (1974); J. F. Brault and P. Savignac, *J. Organomet. Chem.*, **66**, 71 (1974). See also, B. Corbel, J. P. Paugam, M. Dreux, and P. Savignac, *Tetrahedron Lett.*, 835 (1976).
(19) Either diasteriomer of 23 has seven anisochronous carbon positions; however, the ^{13}C nucleus directly bonded to phosphorus was not observed under the employed NMR conditions due to its relatively long spin–lattice relaxation time.
(20) W. Egan and G. Zon, *Tetrahedron Lett.*, 813 (1975).
(21) W. Egan, G. Govil, and G. Zon, unpublished conformational energy calculations for 1 using a classical potential function.
(22) G. Zon, S. M. Ludeman, and W. Egan, *J. Am. Chem. Soc.*, **99**, 5785 (1977).
(23) J. E. Baldwin, *J. Chem. Soc., Chem. Commun.*, 734 (1976); J. E. Baldwin, R. C. Thomas, L. I. Kruse, and L. Silberman, *J. Org. Chem.*, **42**, 3846 (1977).
(24) P. Laszlo and P. Stang, "*Organic Spectroscopy: Principles and Applications*", Harper and Row, New York, N.Y., 1971.
(25) W. G. Bentrude and H.-W. Tan, *J. Am. Chem. Soc.*, **95**, 4666 (1973), and pertinent references cited therein.
(26) If **30** → **13** proceeds via an S_N2 mechanism, it follows that *cis*- and *trans*-11 will give *endo*- and *exo*-**13**, respectively. Cis/trans assignments for faster and slower eluting 11 can, therefore, be made by identifying the NaH product of either pure **11a** or **11b** as pure *endo*- or *exo*-**13**. Diminishing supplies of 11, however, precluded pursuit of this experiment.
(27) G. Zon, W. Egan, and J. B. Stokes, *Biochem. Pharmacol.*, **25**, 989 (1976).
(28) R. W. Brockman, S. C. Shaddix, M. Williams, and R. F. Struck, *Cancer Treat. Rep.*, **60**, 1317 (1976). For the analogous cyclization of 3-chloro-1-ethylpiperidine, see C. F. Hammer, M. McCarthy Ali, and J. D. Weber, *Tetrahedron*, **29**, 1767 (1973).
(29) For example, see P. L. Levins and Z. B. Papanastassiou, *J. Am. Chem. Soc.*, **87**, 826 (1965).
(30) Unpublished ^{31}P NMR studies of phosphoramidic mustard by G. Zon and W. Egan have indicated a 33-min half-life at pH 7.9 and ca. 37 °C. See also, P. L. Levins and I. W. Rogers, *Cancer Chemother. Rep.*, **44**, 15 (1965).
(31) R. I. Geran, N. H. Greenberg, M. M. Macdonald, A. M. Schumacher, and B. J. Abbott, *Cancer Chemother. Rep., Part 3*, **3(2)**, 1 ff. (1972).
(32) The value of T/C = 252 for 1 is the average of three randomly selected T/C values which were obtained at doses of 250 mg/kg.
(33) We thank Dr. Peter B. Farmer of the Chester Beatty Research Institute for providing us with these results. See Farmer and Cox[16] for a lead reference to details regarding this test.
(34) In contrast to the identical LD_{50} values for 1 and 11 in Table II, the L1210 tests with a dosage of 500 mg/kg for 1 gave ≤50% survivors vs. the 100% survivor rate listed in Table I for **11a** at this dose.
(35) Human epidermoid carcinoma of the nasopharynx is the tumor utilized in the KB cell culture tube assay.[31] Synthetic compounds with an ED_{50} value ≤4.0 μg/mL are considered active in this test system.
(36) Aside from the obvious steric "bulk" of an added bromine substituent, solubility perturbations of a more subtle nature may also be a factor in controlling the relative rate of substrate turnover by the microsomal enzyme system. For the role of substrate lipophilicity in determining type I microsomal P-450 binding characteristics, see K. A. S. Al-Gailany, J. B. Houston, and J. W. Bridges, *Biochem. Pharmacol.*, **27**, 783 (1978).
(37) O. M. Friedman and co-workers, *Adv. Cancer Chemother.*, in press.
(38) All chemical shifts in aqueous solutions are internally referenced to the sodium salt of 3-(trimethylsilyl)propionic acid (TSP), unless specified otherwise.
(39) We thank Dr. S. Sojka (Naval Research Laboratory) for obtaining these spectral data and for performing ^1H spin-decoupling experiments to allow for signal assignments.
(40) D. H. Ball, J. M. Williams, and L. Long, Jr., *J. Org. Chem.*, **28**, 1589 (1963).
(41) O. M. Friedman, E. Bogner, V. Grubliauskas, and H. Sommer, *J. Med. Chem.*, **6**, 50 (1963).
(42) Instruction 14, "Screening Data Summary Interpretation and Outline of Current Screen", Drug Evaluation Branch, Drug Research and Development, Division of Cancer Treatment, National Cancer Institute, Bethesda, Md., 1973. Consult all subsequent insert pages for most recent updating.

Aryloxyalkyloxy- and Aralkyloxy-4-hydroxy-3-nitrocoumarins Which Inhibit Histamine Release in the Rat and Also Antagonize the Effects of a Slow Reacting Substance of Anaphylaxis

Derek R. Buckle,* D. James Outred, Janet W. Ross, Harry Smith, Raymond J. Smith, Barbara A. Spicer, and Brian C. Gasson

Beecham Pharmaceuticals-Research Division, Brockham Park, Betchworth Surrey RH3 7AJ, England. *Received July 26, 1978*

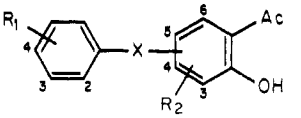
The syntheses and structure–activity relationships of a number of 4-hydroxy-3-nitrocoumarins, which are both antagonists of a slow reacting substance of anaphylaxis and potent inhibitors of antigen-induced histamine release in the rat, are described. Most active among these are 7-[3-(4-acetyl-3-hydroxy-2-*n*-propylphenoxy)-2-hydroxypropoxy] derivatives having hydrogen or lower alkyl substituents at the C-8 position of the coumarin ring, **168**, **171**, **173**, and **174**.

Disodium cromoglycate (DSCG) has been shown to inhibit the antigen-induced release of the mediators of allergic reaction[1] but it is not equally effective at inhibiting the release of all mediators. In particular, it is poor at inhibiting the release of a slow reacting substance of anaphylaxis (SRS-A) when antibodies other than IgE are involved.[2] It has been suggested that the failure of DSCG to benefit some patients with bronchial asthma might be due to the involvement of IgG antibodies, which might release SRS-A from sources other than the mast cell by a mechanism resistant to treatment with DSCG.[3] If this is so, then a compound with the ability to antagonize the effects of SRS-A, in addition to the stabilization of mast cells, might be expected to be of greater therapeutic value, than DSCG, in the treatment of bronchial asthma.

We have reported previously that some compounds containing the 2-nitro-1,3-dicarbonyl moiety showed potent antiallergic activity, as determined by the inhibition of rat

0022-2623/79/1822-0158$01.00/0 © 1979 American Chemical Society

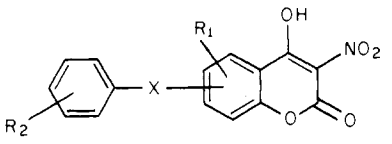
Table I. 2'-Hydroxyacetophenones



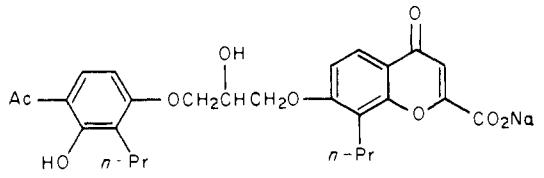
compd	posit of side chain attach.	R_1	R_2	X	mp, °C[a]	formula	anal.	yield,[b] %
1	6	H	H	$O(CH_2)_3O$	89-90	$C_{17}H_{18}O_4$	C, H	79
2	6	H	H	$O(CH_2)_4O$	81	$C_{18}H_{20}O_4$	C, H	91
3	5	H	H	$O(CH_2)_2O$	113-114	$C_{16}H_{16}O_4$	C, H	43
4	5	H	H	$O(CH_2)_3O$	87-88	$C_{17}H_{18}O_4$	C, H	70
5	5	H	H	$O(CH_2)_5O$	45	$C_{19}H_{22}O_4$	C, H	78
6	5	H	H	$O(CH_2)_6O$	74-77	$C_{20}H_{24}O_4$	H, C[c]	43
7	4	H	H	$O(CH_2)_2O$	132-133	$C_{16}H_{16}O_4$	C, H	62
8	4	H	H	$O(CH_2)_3O$	82-82.5	$C_{17}H_{18}O_4$	C, H	77
9	4	H	H	$O(CH_2)_4O$	95-96	$C_{18}H_{20}O_4$	C, H	87
10	4	H	H	$O(CH_2)_5O$	51-52	$C_{19}H_{22}O_4$	C, H	63
11	4	H	H	$O(CH_2)_6O$	71-74	$C_{20}H_{24}O_4$	C, H	69
12	3	H	H	$O(CH_2)_3O$	56-57	$C_{17}H_{18}O_4$	C, H	32
13	6	H	H	$(CH_2)_3O$	95-96	$C_{17}H_{18}O_3$	C, H	75
14	5	H	H	$(CH_2)_2O$	36	$C_{16}H_{16}O_3$	C, H	21
15	5	H	H	$(CH_2)_3O$	34-35	$C_{17}H_{18}O_3$	C, H	54
16	4	H	H	CH_2O	104-105[d]	$C_{15}H_{14}O_3$		85
17	4	H	H	$(CH_2)_2O$	69	$C_{16}H_{16}O_3$	C, H	45
18	4	H	H	$(CH_2)_3O$	75-77	$C_{17}H_{18}O_3$	C, H	83
19	4	H	H	$(CH_2)_4O$	55	$C_{18}H_{20}O_3$	C, H	59
20	4	4-Cl	H	$(CH_2)_4O$	73-75	$C_{18}H_{19}ClO_3$	C, H, Cl	71
21	4	4-Me	H	$(CH_2)_4O$	69-70	$C_{19}H_{22}O_3$	C, H	72
22	4	H	5-Et	$O(CH_2)_3O$	60.5-61.5	$C_{19}H_{22}O_4$	C, H	74
23	4	H	3-Pr[n]	$O(CH_2)_3O$	67-68	$C_{20}H_{24}O_4$	C, H	84
24	4	4-F	H	$O(CH_2)_3O$	70-72	$C_{17}H_{17}FO_4$	C, H	59
25	4	4-Cl	H	$O(CH_2)_3O$	82-83	$C_{17}H_{17}ClO_4$	C, H, Cl	80
26	4	4-OMe	H	$O(CH_2)_3O$	75-76	$C_{18}H_{20}O_5$	C, H	59
27	4	4-CN	H	$O(CH_2)_3O$	131-133	$C_{18}H_{17}NO_4$	C, H, N	65
28	4	4-Ph	H	$O(CH_2)_3O$	118-121	$C_{23}H_{22}O_4$	C, H	72
29	4	4-Me	H	$O(CH_2)_3O$	85-87	$C_{18}H_{20}O_4$	C, H	67
30	4	3-Me	H	$O(CH_2)_3O$	64	$C_{18}H_{20}O_4$	C, H	80
31	4	2-Me	H	$O(CH_2)_3O$	70	$C_{18}H_{20}O_4$	C, H	65
32	4	3,4-tetramethylene	H	$O(CH_2)_3O$	80-82	$C_{21}H_{22}O_4$	C, H	59

[a] Recrystallized from EtOH. [b] Prepared according to method A; see text and Experimental Section. [c] C: calcd, 71.80; found, 73.15. [d] L. Jurd and L. A. Polle, *J. Am. Chem. Soc.*, 80, 5527 (1958), quote mp as 104-104.5 °C.

passive cutaneous anaphylaxis (PCA),[4] and the compounds seemed to owe this activity to their ability to stabilize mast cells.[5] By the attachment of selected side chains to some of these nuclei, as in I, we have attempted to confer SRS-A



I



II

antagonist activity on these compounds while retaining a high level of mast cell stabilizing activity.

Specific SRS-A antagonist activity has been reported for a series of chromones,[6] of which FPL 55712 (II)[6,7] was the most active.

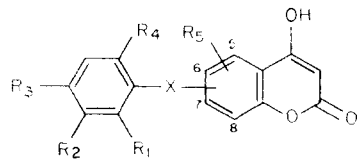
In this paper, we report the synthesis and some biological activities of a selection of 4-hydroxy-3-nitrocoumarins of formula I in which the substitutions leading to the antagonism of SRS-A have been established.

Chemistry. The synthesis of aryloxyalkyloxy- and aralkyloxy-4-hydroxy-3-nitrocoumarins, I, is outlined in Scheme I. Alkylation of the dihydroxyacetophenones III with the appropriate halide IV using potassium carbonate in acetone or butanone (method A) led to the monoalkylated 2'-hydroxyacetophenones **1-32** (Table I) which readily cyclized with diethyl carbonate as previously described[4b] (method B) to give the 4-hydroxycoumarins **33-53, 55-59, 62, 63,** and **65-68** (Table II).

For compounds with base-sensitive side chains and those derived from the halohydrins of Table III, it was found to be advantageous to use a preformed coumarin nucleus for the synthesis of the precursor 4-hydroxycoumarins. Our first choice was to use coumarins of type V in which the hydroxylactone function was protected as its enamine. Alkylation of the sodium salt of V with the halides VI in *N,N*-dimethylformamide (DMF, method C) afforded good yields of the aminocoumarins **98-103** (Table IV), which on acid hydrolysis (method D) gave the corresponding hydroxycoumarins **54, 60, 64, 69, 71,** and **72** (Table II) in moderate yield. Concomitant hydrolysis of ester functions also occurred during this step. The low solubility of these aminocoumarins in the hydrolysis medium caused difficulties with several derivatives, especially where R''' = H, which led to a search for an alternative protecting group.

Direct alkylation of 4,7-dihydroxycoumarin was difficult due to the low solubility of its alkali metal salts in common

Table II. 4-Hydroxycoumarins

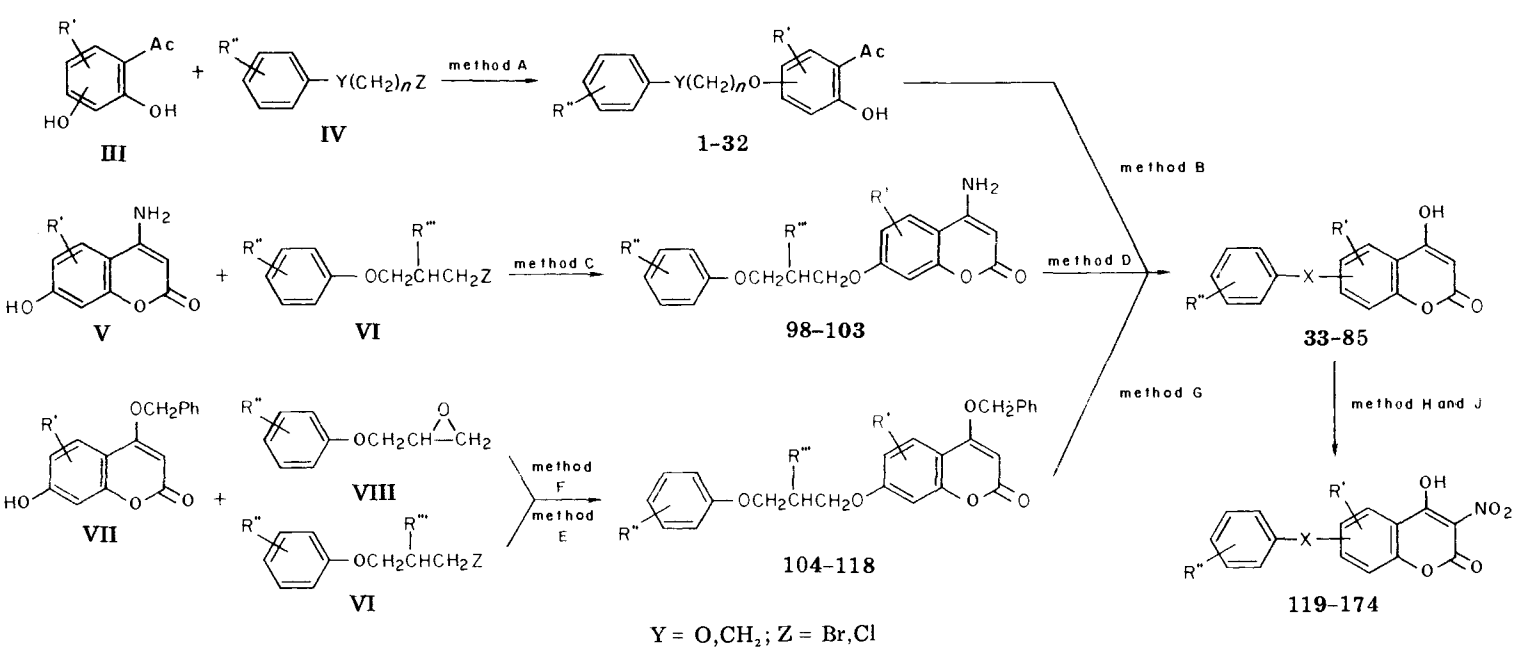


compd	posit of side-chain attach.	R_1	R_2	R_3	R_4	R_5	X	mp, °C[a]	formula	anal.	yield, %	meth of prep[b]
33	5	H	H	H	H	H	$O(CH_2)_3O$	179-180	$C_{18}H_{16}O_5$	C, H	71	B
34	5	H	H	H	H	H	$O(CH_2)_4O$	101-102	$C_{19}H_{18}O_5$	C, H	38	B
35	6	H	H	H	H	H	$O(CH_2)_2O$	211	$C_{17}H_{14}O_5$	C, H	39	B
36	6	H	H	H	H	H	$O(CH_2)_3O$	204-205	$C_{18}H_{16}O_5$	C, H	71	B
37	6	H	H	H	H	H	$O(CH_2)_5O$	163-164	$C_{20}H_{20}O_5 \cdot H_2O$	C, H	40	B
38	6	H	H	H	H	H	$O(CH_2)_6O$	162-163	$C_{21}H_{22}O_5$	H, C[c]	58	B
39	7	H	H	H	H	H	$O(CH_2)_2O$	244	$C_{17}H_{14}O_5$	C, H	39	B
40	7	H	H	H	H	H	$O(CH_2)_3O$	190-192	$C_{18}H_{16}O_5$	C, H	65	B
41	7	H	H	H	H	H	$O(CH_2)_4O$	194	$C_{19}H_{18}O_5$	C, H	70	B
42	7	H	H	H	H	H	$O(CH_2)_5O$	180	$C_{20}H_{20}O_5$	C, H	49	B
43	7	H	H	H	H	H	$O(CH_2)_6O$	170	$C_{21}H_{22}O_5 \cdot 0.5H_2O$	C, H	22	B
44	8	H	H	H	H	H	$O(CH_2)_3O$	187-188	$C_{18}H_{16}O_5 \cdot H_2O$	C, H	48	B
45	5	H	H	H	H	H	$(CH_2)_3O$	80-81	$C_{18}H_{16}O_4$	C, H	27	B
46	6	H	H	H	H	H	$(CH_2)_2O$	189	$C_{17}H_{14}O_4$	C, H	71	B
47	6	H	H	H	H	H	$(CH_2)_3O$	212	$C_{18}H_{16}O_4$	C, H	65	B
48	7	H	H	H	H	H	CH_2O	278[d]	$C_{16}H_{12}O_4$		58	B
49	7	H	H	H	H	H	$(CH_2)_2O$	203	$C_{17}H_{14}O_4$	C, H	49	B
50	7	H	H	H	H	H	$(CH_2)_3O$	255	$C_{18}H_{16}O_4$	C, H	56	B
51	7	H	H	H	H	H	$(CH_2)_4O$	192	$C_{19}H_{18}O_4$	C, H	42	B
52	7	H	H	Cl	H	H	$(CH_2)_4O$	209-210.5	$C_{19}H_{17}ClO_4$	C, H, Cl	63	B
53	7	H	H	Me	H	H	$(CH_2)_4O$	198-201	$C_{20}H_{20}O_4$	C, H	57	B
54	7	H	H	H	H	5-Me	$O(CH_2)_3O$	218-219	$C_{19}H_{18}O_5$	C, H	65	D
55	7	H	H	H	H	6-Et	$O(CH_2)_3O$	233-234	$C_{20}H_{20}O_5$	C, H	73	B
56	7	H	H	H	H	8-Pr^n	$O(CH_2)_3O$	149-150	$C_{21}H_{22}O_5$	C, H	95	B
57	7	H	H	F	H	H	$O(CH_2)_3O$	203-206	$C_{18}H_{15}NO_5$	C, H	58	B
58	7	H	H	Cl	H	H	$O(CH_2)_3O$	230-234	$C_{18}H_{15}ClO_5$	C, H, Cl	71	B
59	7	H	H	OMe	H	H	$O(CH_2)_3O$	193-196	$C_{19}H_{18}O_6$	C, H	73	B
60	7	H	H	Ac	H	H	$O(CH_2)_3O$	202-204	$C_{20}H_{18}O_6$	C, H	55	D
61	7	H	H	CO_2Me	H	H	$O(CH_2)_3O$	218	$C_{20}H_{18}O_7$	C, H	83	G
62	7	H	H	CN	H	H	$O(CH_2)_3O$	180-184	$C_{19}H_{15}NO_5$	C, N, H[e]	50	B
63	7	H	H	Ph	H	H	$O(CH_2)_3O$	240-242	$C_{24}H_{20}O_5$	C, H	76	B
64	7	H	H	CO_2H	H	H	$O(CH_2)_3O$	275-276[f]	$C_{19}H_{16}O_7$	crude	80	D[g]
65	7	H	H	Me	H	H	$O(CH_2)_3O$	209-212	$C_{19}H_{18}O_5$	C, H	59	B
66	7	H	Me	H	H	H	$O(CH_2)_3O$	192	$C_{19}H_{18}O_5$	C, H	26	B
67	7	Me	H	H	H	H	$O(CH_2)_3O$	203	$C_{19}H_{18}O_5$	C, H	24	B
68	7	H	tetramethylene		H	H	$O(CH_2)_3O$	199-201	$C_{22}H_{22}O_5$	C, H	42	B
69	7	H	OH	Ac	H	H	$O(CH_2)_3O$	220-222	$C_{20}H_{18}O_7$	C, H	71	D
70	7	Pr^n	OH	Ac	H	H	$O(CH_2)_3O$	166	$C_{23}H_{24}O_7$	C, H	70	G

71	7	H	H	H	H	H	$OCH_2CH(OH)CH_2O$	181–183	$C_{18}H_{16}O_6$	C, H	54	D
72	7	H	H	CO_2H	H	H	$OCH_2CH(OH)CH_2O$	169–172 (foams)	$C_{19}H_{16}O_8$	crude	55	D
73	7	Et	H	H	H	H	$OCH_2CH(OH)CH_2O$	177	$C_{20}H_{20}O_6$	C, H	71	G
74	7	Pr^n	H	H	H	H	$OCH_2CH(OH)CH_2O$	197	$C_{21}H_{22}O_6 \cdot 0.5H_2O$	C, H	89	G
75	7	Pr^n	H	F	H	H	$OCH_2CH(OH)CH_2O$	195	$C_{21}H_{21}NO_6$	C, H	95	G
76	7	Pr^i	H	H	H	H	$OCH_2CH(OH)CH_2O$	172	$C_{21}H_{22}O_6 \cdot 0.5H_2O$	C, H	97	G
77	7	Pr^n	H	Ac	H	H	$OCH_2CH(OH)CH_2O$	oil	$C_{23}H_{23}NO_9$	crude	87	G
78	7	H	OH	Ac	H	H	$OCH_2CH(OH)CH_2O$	~205	$C_{20}H_{18}O_8$	crude	60	G
79	7	Me	OH	Ac	H	H	$OCH_2CH(OH)CH_2O$	230	$C_{21}H_{20}O_8$	C, H	64	G
80	7	Pr^n	OH	Ac	H	H	$OCH_2CH(OH)CH_2O$	foam	$C_{23}H_{24}O_8$	M^+ 428	100	G
81	7	Pr^n	OH	EtCO	H	H	$OCH_2CH(OH)CH_2O$	89	$C_{24}H_{26}O_8$	C, H	61	G
82	7	H	OH	Ac	Et	H	$OCH_2CH(OH)CH_2O$	240	$C_{22}H_{22}O_8$	C, H	87	G
83	7	Pr^n	OH	Ac	H	Me	$OCH_2CH(OH)CH_2O$	118–120	$C_{24}H_{26}O_8 \cdot H_2O$	C, H	93	G
84	7	Pr^n	OH	Ac	H	Et	$OCH_2CH(OH)CH_2O$	170	$C_{25}H_{28}O_8$	C, H	88	G
85	7	Pr^n	OH	Ac	H	Pr^n	$OCH_2CH(OH)CH_2O$	foam	$C_{26}H_{30}O_8$		100	G

[a] Recrystallized from EtOH with addition of water, if necessary. [b] See text and Experimental Section. [c] C: calcd, 71.17; found, 70.37. [d] M. A. Hermodson, W. M. Barker and K. P. Link, *J. Med. Chem.*, 14, 167 (1971), quote mp 272–273 °C. [e] H: calcd, 4.45; found, 5.03. [f] Recrystallized from aqueous DMF. [g] Hydrolysis assisted by the addition of AcOH.

Scheme I



solvents, although its methylation has been previously reported.[8] However, reaction of the monosodium salt in DMF proceeded preferentially at the C-4 hydroxyl, as shown by IR and NMR spectroscopy (the C-3 proton no longer being exchanged with D_2O), with no product alkylated solely at the C-7 hydroxyl being isolated under these conditions. With benzyl chloride, a 28% yield of 4-benzyloxy-7-hydroxycoumarin VII (R′ = H) was isolated. Similar yields of VII in which R′ = alkyl were obtained from alkyl-substituted 4,7-dihydroxycoumarins, but, in general, better yields of C-4 benzylated products were accomplished under esterification conditions. Further alkylation of VII gave moderate to good yields of the coupled products **104–118** (Table V), whether the alkylating agent was a simple halide, a halohydrin (VI, R‴ = H or OH, respectively; method E), or an epoxide VIII (method F). Hydrogenolysis of the benzyl group proceeded rapidly at atmospheric pressure (method G) to give high yields of the deprotected coumarins **61**, **70**, and **73–85**. 4,7-(Dibenzyloxy)coumarin could be similarly debenzylated to 7-(benzyloxy)-4-hydroxycoumarin (**48**) in near quantitative yield, which clearly established the selectivity with which the 4-benzyl group could be removed.

Direct nitration of the compounds of Table II was simply accomplished in the majority of cases using fuming nitric acid in chloroform at 0 °C[4b] (method H). At higher temperatures and in cases where the terminal aromatic ring was activated by two or more electron donating substituents, secondary nitration occurred. Thus, nitration of **65** resulted in the simultaneous nitration of the terminal aromatic ring to give **157**, the position of insertion of the second nitro group being unambiguously assigned as C-2′ by NMR spectroscopy. Similarly, nitration of **83** at room temperature gave the dinitro derivative **172** in which the position of the second nitro group was again unambiguously assigned from its NMR spectrum.

In these cases of more highly activated nuclei, the nitro group at C-3 was introduced in a stepwise fashion by nitrosation and oxidation (method J). Compounds **119**, **120**, **144**, **146**, **149**, **150**, **152–155**, **161–165**, and **173** were synthesized in this way. Generally, nitration by the direct route is to be preferred over nitrosation and oxidation whenever possible on account of the greater yields obtainable.

The dihydroxyacetophenones III and halides IV also required in this work were either available commercially or prepared by standard procedures. In general, compounds IV were prepared from α,ω-dibromoalkanes,[9] the products being distilled under vacuum but used without further purification. The halohydrins VI of Table III were prepared from the appropriate phenol using the method of Stephenson.[10]

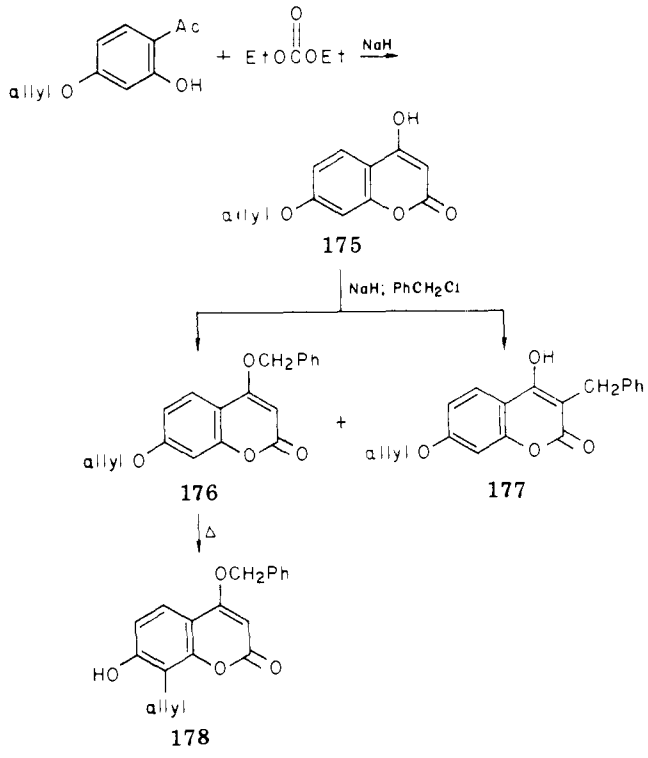
Synthesis of the aminocoumarin intermediates V was accomplished from the corresponding resorcinol by condensation with ethyl cyanoacetate.[11-13]

The necessary 8-allyl derivative **178** was prepared as shown in Scheme II. Condensation of 4′-(allyloxy)-2′-hydroxyacetophenone[14] with diethyl carbonate gave the coumarin **175**, which on alkylation with benzyl chloride gave a mixture of the *O*-benzyl-**176** and *C*-benzyl-**177** derivatives in 38 and 4% yields, respectively. Pyrolysis of **176** in decalin then gave the rearrangement product **178** in good yield.

Results and Discussion

Following the discovery of the potent SRS-A antagonist II[5] we were prompted to look for analogous activity in a series of compounds containing the 4-hydroxy-3-nitrocoumarin nucleus. Members of this series carrying simple

Scheme II



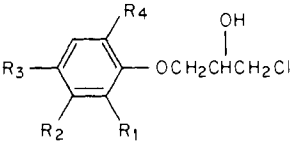
alkyl substituents showed little SRS-A antagonist activity, and it seemed to us that it might be possible to introduce this activity into the series, by the incorporation of an appropriate side chain, while still retaining the high antiallergy activity shown by some compounds containing this nucleus.[4b]

As a starting point, we selected the unsubstituted phenoxypropoxy compound **126** (Table VI), since it was known[4b] that nitrocoumarins substituted at C-7 with alkyloxy groups were among the most potent antiallergic compounds of this series. On finding that this compound retained a high level of activity as an inhibitor of histamine release when given intraperitoneally to rats prior to passive peritoneal anaphylaxis (PPA) (Table X) and yet displayed antagonism of SRS-A, we were encouraged to look further at other derivatives of this general class.

Simple manipulation of the alkyl side chain and the position of its attachment to the coumarin ring, compounds **119–130** (Table VI), indicated that SRS-A antagonist activity tended to fall off with increasing chain length in the C-7 series, with the trimethylene chain, **126**, being optimal. The effect of chain length was less critical in the C-6 series where the tri- and pentamethylene compounds, **122** and **123**, respectively, were equipotent to **126**, and the hexamethylene analogue **124** was marginally more potent. Substitution at either positions C-5 or C-8 as in compounds **119**, **120**, and **130**, resulted in a marked reduction of antagonistic activity. Without exception substitution at C-7 gave compounds of greatest activity as inhibitors of histamine release during rat PPA, with **126** being the most potent of this group. The C-6 hexamethylene compound **124** had low activity in this test despite its good antagonism of SRS-A, and further elaboration was therefore centered on the C-7 derivatives.

Replacing the terminal ether function of this series by the methylene group to give the compounds of Table VII showed a similar trend with regard to positional effects of the side chain. In this instance, variation of the chain length from one to four carbon atoms, **135–138**, had little influence on the level of SRS-A antagonism. A possible

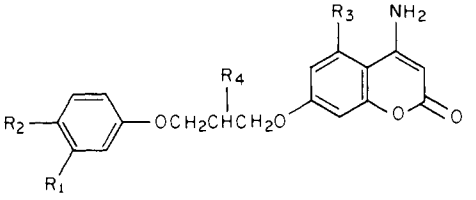
Table III. 1-(Aryloxy)-3-chloropropan-2-ols



compd	R_1	R_2	R_3	R_4	mp or bp (mm), °C	formula	anal.	yield,[a] %
86	H	H	H	H	112 (0.6)[b]	$C_9H_{11}ClO_2$		71
87	H	H	CO_2Me	H	170-172 (0.5)	$C_{11}H_{13}ClO_4$	C, H, Cl	75
88	Et	H	H	H	104 (0.5)	$C_{11}H_{15}ClO_2$		70
89	Pr^n	H	H	H	136-140 (2.0)	$C_{12}H_7ClO_2$		44
90	Pr^n	H	F	H	127 (0.6)	$C_{12}H_{16}ClFO_2$	C, H, Cl	73
91	Pr^i	H	H	H	170 (2.0)	$C_{12}H_{17}ClO_2$		72
92	Pr^n	H	Ac	H	oil	$C_{14}H_{19}ClO_3$		100
93	H	OH	Ac	H	oil	$C_{11}H_{13}ClO_4$		60
94	Me	OH	Ac	H	112-116	$C_{12}H_{15}ClO_4$	C, H, Cl	63
95	Pr^n	OH	Ac	H	oil	$C_{14}H_{19}ClO_4$		89
96	Pr^n	OH	EtCO	H	oily solid	$C_{15}H_{19}ClO_4$	C, H, Cl	100
97	H	OH	Ac	Et	68	$C_{13}H_{17}ClO_4$	C, H, Cl	74

[a] Prepared according to the method of O. Stephenson, *J. Chem. Soc.*, 1571 (1954). [b] W. Bradley, J. Forrest, and O. Stephenson, *J. Chem. Soc.*, 1589 (1951), quote bp as 120-135 °C (3.0 mm).

Table IV. 4-Aminocoumarins



compd	R_1	R_2	R_3	R_4	mp, °C	formula	anal.	yield,[a] %
98	H	H	H	OH	213-214 (EtOAc)	$C_{18}H_{17}NO_5 \cdot 0.5H_2O$	C, H, N	92
99	H	H	Me	H	164-167 (EtOH)	$C_{19}H_{19}NO_4$	C, H, N	91
100	H	Ac	H	H	165-168 (AcOH-H_2O)	$C_{20}H_{19}NO_5$	C, H, N	94
101	OH	Ac	H	H	158-159 (EtOH-H_2O)	$C_{20}H_{19}NO_6$	C, H, N	60
102	H	CO_2Me	H	H	155 (AcOH-H_2O)	$C_{20}H_{19}NO_6 \cdot H_2O$	C, H, N	100
103	H	CO_2Me	H	OH	212-215 (MeOH)	$C_{20}H_{19}NO_7 \cdot 0.5H_2O$	C, H, N	94

[a] Prepared by method C.

advantage with this modification lay in the increased aqueous solubility of their sodium salts.

The terminal aromatic ring was elaborated with a range of substituents (Table VIII, compounds **144–158**). Using compound **126** as a reference, these substitutions generally resulted in a reduction in the ability to antagonize SRS-A, with the exception of the two halo compounds **144** and **145** and the acetyl and methyl compounds **147** and **152**, respectively, which showed a similar SRS-A antagonist activity to that shown by **126**. From these four compounds, the methyl derivative **152** was selected in order to study the influence of the substituent position on SRS-A antagonist activity. Very little difference was noticed in the meta and para isomers **152** and **153**, respectively, but an enhancement of SRS-A antagonism occurred in the ortho isomer **154**.

The reduction in SRS-A antagonism found with the carboxy analogue **151**, which was one of the more water-soluble derivatives of the group, was of interest. Indeed, in our attempts to confer greater aqueous solubility to the group by incorporation of the glyceryl side chain (Table IX), a noticeable drop in SRS-A antagonism relative to the nonhydroxylated analogue generally occurred. Compounds **159** and **126** are typical examples of this.

In view of the structure of II, hydroxyl and acetyl functions were incorporated into the terminal nucleus of I to give the two derivatives **156** and **166**. These compounds were no more active as antagonists than their unsubstituted counterparts **126** and **159**, respectively. The addition of an *n*-propyl group at the ortho position, as in compounds **158** and **168**, resulted in increased activity. Furthermore, the presence of any lower alkyl group in the ortho position, as in compounds **154**, **158**, **161–165**, and **167–174**, resulted in the most active members of the series.

Replacing the acetyl function of **168** by proprionyl, **169**, had little effect on activity, as did branching of the *o*-alkyl group (compound **164**).

The effect of alkyl groups in the coumarin ring of I was shown in the basic system using compounds **141–143** and in the more complex case using compounds **171**, **173**, and **174**. It was clearly established that alkylation at either C-5 or C-6 reduced the ability to antagonize SRS-A, while little difference could be demonstrated between simple alkyl substitution at C-8, compounds **171**, **173**, and **174**, and the dealkyl derivative **168**.

The structural requirements for in vitro SRS-A antagonist activity shown by compounds containing the 4-hydroxy-3-nitrocoumarin nucleus were very similar to

Table V. 4-(Benzyloxy)coumarins

compd	R_1	R_2	R_3	R_4	R_5	X	mp, °C[a]	formula	anal.	yield,[b] %
104	H	H	CO_2Me	H	H	$O(CH_2)_3O$	153–155	$C_{27}H_{24}O_7$	C, H	69
105	Et	H	H	H	H	$OCH_2CH(OH)CH_2O$	82–83	$C_{27}H_{26}O_6$	C, H	45
106	Pr[n]	H	H	H	H	$OCH_2CH(OH)CH_2O$	109–110	$C_{28}H_{28}O_6$	C, H	55[c]
107	Pr[i]	H	F	H	H	$OCH_2CH(OH)CH_2O$	152	$C_{28}H_{28}O_6$	C, H	30
108	Pr[n]	H	F	H	H	$OCH_2CH(OH)CH_2O$	124–126	$C_{28}H_{27}FO_6$	C, H	41
109	Pr[n]	H	H	H	H	$OCH_2CH(OH)CH_2O$	154	$C_{30}H_{30}O_7$	C, H	18
110	H	OH	Ac	H	H	$OCH_2CH(OH)CH_2O$	192–195	$C_{27}H_{24}O_8$	crude	33
111	H	OH	Ac	Et	H	$OCH_2CH(OH)CH_2O$	195	$C_{29}H_{28}O_8$	C, H	37
112	Me	OH	Ac	H	H	$OCH_2CH(OH)CH_2O$	157	$C_{28}H_{26}O_8 \cdot 0.5H_2O$	C, H	30
113	Pr[n]	OH	Ac	H	H	$OCH_2CH(OH)CH_2O$	172–173	$C_{30}H_{30}O_8$	C, H	42
114	allyl	OH	EtCO	H	H	$OCH_2CH(OH)CH_2O$	117	$C_{31}H_{30}O_8$	C, H	25
115	Pr[n]	OH	Ac	H	Me	$OCH_2CH(OH)CH_2O$	201–203	$C_{31}H_{33}O_8 \cdot 0.5H_2O$	C, H	37
116	Pr[n]	OH	Ac	H	Et	$OCH_2CH(OH)CH_2O$	195[d]	$C_{29}H_{28}O_8$	C, H	36
117	Pr[n]	OH	Ac	H	allyl	$OCH_2CH(OH)CH_2O$	foam	$C_{33}H_{34}O_8$	R_f ($CHCl_3/SiO_2$) 0.5	41
118	Pr[n]	OH	Ac	H	H	$O(CH_2)_3O$	190	$C_{30}H_{30}O_7$	C, H	32

[a] Recrystallized from EtOH. [b] Prepared by method E. [c] Prepared using method F. [d] Recrystallized from AcOH.

those previously reported for a series of 2-carboxychromones.[6] Compounds containing the former nucleus showed, in addition, a high level of antiallergy activity, as demonstrated by their ability to inhibit histamine release when given to rats, intraperitoneally, prior to PPA. Table X gives the PPA results of seven of the preferred compounds of this class, together with Ti and disodium cromoglycate (DSCG) as reference markers. Compounds **168** and **174** were the most potent inhibitors of histamine release, and these two compounds also were among the most potent of the nitrocoumarins at antagonizing the effects of SRS-A.

Experimental Section

Melting points were determined using a Büchi melting-point apparatus and are uncorrected. The structures of all compounds were consistent with their IR and NMR spectra, the latter of which were determined as solutions in either $CDCl_3$ or Me_2SO-d_6. Where represented by elemental symbols, the analyses for these elements fall within ±0.4% of the calculated values.

Aryloxyalkyl Halides (IV, Y = O). Aryloxyalkyl bromides were either commercially available or synthesized from the corresponding phenols by the procedure of Marvel and Tanenbaum.[9] Those synthesized had boiling points in agreement with literature values. The following chlorides were prepared by alkylation of the appropriate phenols with 1-bromo-3-chloropropane.

4′-(3-Chloropropoxy)-2′-hydroxyacetophenone. A mixture of 2′,4′-dihydroxyacetophenone (7.6 g, 0.05 mol), anhydrous K_2CO_3 (10.4 g, 0.075 mol), and 1-bromo-3-chloropropane (11.8 g, 0.075 mol) in dry Me_2CO (40 mL) was stirred at reflux for 5 h, cooled, and filtered. Evaporation of the filtrate gave a dark solid, which was recrystallized from EtOH to give 9.5 g (84%) of material of mp 73 °C. Anal. ($C_{11}H_{13}ClO_3$) C, H.

4′-(3-Chloropropoxy)-2′-hydroxy-3′-*n*-propylacetophenone. Alkylation of 2′,4′-dihydroxy-3′-*n*-propylacetophenone[14] with 1-bromo-3-chloropropane as described above gave 80% of the title compound of mp 37–38 °C. Anal. ($C_{14}H_{19}ClO_3$) C, H, Cl.

Aralkyl Bromides (IV, Y = CH_2). These compounds were either commercially available or prepared from the corresponding alcohols using hydrobromic acid following the procedure of v. Braun.[15]

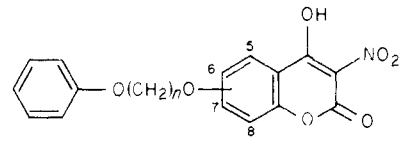
1-(Aryloxy)-3-chloropropan-2-ols (Table III). These were prepared by the procedure of Stephenson[10] by reaction of the appropriate phenol with 1-chloro-2,3-epoxypropane.

4-(Benzyloxy)-7-hydroxycoumarins VII. 4-(Benzyloxy)-7-hydroxycoumarin. (a) By Alkylation with Benzyl Chloride. A solution of $PhCH_2Cl$ (12.7 g, 0.1 mol) in dry DMF (10 mL) was added dropwise at 100 °C to a stirred solution of 4,7-dihydroxycoumarin[16] (17.8 g, 0.1 mol) and NaH (2.4 g, 0.1 mol) in dry DMF (75 mL), and the mixture was kept at this temperature for a further 4 h. After cooling, the solvent was removed in vacuo, and water was added to the residue. The oily solid which separated was filtered off, washed with water, and recrystallized from EtOH to give 7.5 g (28%) of the 4-(benzyloxy) derivative: mp 234–236 °C; IR ν_{max} (mull) 3160, 1708, 1625 cm^{-1}; NMR δ (Me_2SO-d_6) 5.32 (2 H, s, benzyl CH_2), 5.80 (1 H, s, nonexchangeable, vinyl CH), 6.80 (2 H, m), 7.33–7.73 (6 H, m), 10.6 (1 H, br, exchangeable, OH). Anal. ($C_{16}H_{12}O_4$) C, H.

(b) By Alkylation with Benzyl Alcohol. Thoroughly dried 4,7-dihydroxycoumarin[16] (50 g) was dissolved in $PhCH_2OH$ (400 mL), and dry HCl was passed through the solution for 1 h. The mixture was warmed to 50 °C for 2 h, and the dark solution was decolorized and concentrated in vacuo. On cooling, 25.6 g of the 4-(benzyloxy) derivative, mp 232–234 °C, separated, which was identical with that prepared above. Dilution of the mother liquor with water gave 23 g of 4,7-dihydroxycoumarin, giving an overall yield of 63% on the basis of unreclaimed diol.

4-(Benzyloxy)-7-hydroxy-8-methylcoumarin. Dry HCl was passed through a stirred suspension of 4,7-dihydroxy-8-methylcoumarin[18] (15 g) in $PhCH_2OH$ (150 mL) at 0 °C for 30 min, and the mixture was stirred at 100 °C for 2 h. After cooling, the mixture was concentrated in vacuo and the residue diluted with EtOH (50 mL). The benzyl derivative which separated was filtered off and recrystallized from EtOH to give 6.0 g (27%) of

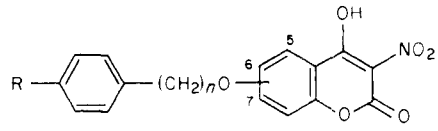
Table VI. 4-Hydroxy-3-nitrocoumarins



compd	posit of side-chain attach.	*n*	mp, °C[a]	formula	anal.	yield,[b] %	antag of SRS-A concn, μM, for 50% inhib[c]
119	5	3	138-139	$C_{18}H_{15}NO_7$	C, H, N	65[d]	18
120	5	4	120	$C_{19}H_{17}NO_7 \cdot 0.5H_2O$	C, H, N	56[d]	>10
121	6	2	164-165	$C_{17}H_{13}NO_7$	C, H, N	45	17
122	6	3	138-140	$C_{18}H_{15}NO_7$	C, H, N	48	2.3
123	6	5	110	$C_{20}H_{19}NO_7$		20	2.0
124	6	6	90-91	$C_{21}H_{21}NO_7$	H, N, C[e]	37	0.78 (0.17-3.5)
125	7	2	188	$C_{17}H_{13}NO_7$	C, H, N	69	>10
126	7	3	148-150	$C_{18}H_{15}NO_7$	C, H, N	82	1.6 (0.26-9.0)
127	7	4	152-154	$C_{19}H_{17}NO_7$	C, H, N	96	3.6
128	7	5	143-145	$C_{20}H_{19}NO_7$	C, H, N	77	8.4
129	7	6	110	$C_{21}H_{21}NO_7$	C, H, N	66	>20
130	8	3	115-116	$C_{18}H_{15}NO_7$	C, H, N	31	>20

[a] Recrystallized from EtOH. Melting usually accompanied by decomposition. [b] Prepared by method H, unless otherwise noted. [c] Figures in parentheses are 95% confidence limits. The value for the standard compound, FPL 55712 (II), under these conditions was 0.17 μM (0.006-4.8). [d] Prepared by method J. [e] C: calcd, 63.15; found, 62.61.

Table VII. 4-Hydroxy-3-nitrocoumarins



compd	R	posit of side-chain attach.	*n*	mp, °C[a]	formula	anal.	yield,[b] %	antag of SRS-A, concn in μM, for 50% inhib[c]
131	H	5	3	134-137	$C_{18}H_{15}NO_6$	C, H, N	55	>20
132	H	6	1	194-195	$C_{16}H_{11}NO_6$	C, H, N	99	22
133	H	6	2	166	$C_{17}H_{13}NO_6$	C, H, N	73	18
134	H	6	3	137-138	$C_{18}H_{15}NO_6$	C, H, N	79	5.5
135	H	7	1	205-206	$C_{16}H_{11}NO_6$	C, H, N	78	1.5
136	H	7	2	154	$C_{17}H_{13}NO_6$	C, H, N	83	4.4
137	H	7	3	192-195	$C_{18}H_{15}NO_6$	C, H, N	73	7.8
138	H	7	4	132	$C_{19}H_{17}NO_6$	C, H, N	88	4.5
139	Cl	7	4	157-160	$C_{19}H_{16}ClNO_6$	C, H, Cl, N	62	3.8
140	Me	7	4	145-147	$C_{20}H_{19}NO_6$	C, H, N	88	2.5

[a] See footnote *a*, Table VI. [b] Prepared by method H. [c] See footnote *c*, Table VI.

material, mp 274-276 °C. Anal. ($C_{17}H_{14}O_4$) C, H.

4-Amino-8-ethyl-7-hydroxycoumarin. Ethyl cyanoacetate (28.7 g, 0.254 mol), 2-ethylresorcinol[17] (35 g, 0.25 mol), and anhydrous $ZnCl_2$ (16 g) in dry Et_2O (100 mL) were stirred at 0 °C for 2 h, during which time dry HCl was passed through the solution. The mixture was allowed to stand for 3 days, and the ethereal phase was separated. Dilution of the inorganic phase with water gave the title compound, which on recrystallization from aqueous EtOH gave 18.8 g (36%) of product, mp 240-243 °C, which was used without further purification. Other 4-aminocoumarins of structure V were similarly prepared.

4,7-Dihydroxy-8-ethylcoumarin. 4-Amino-8-ethyl-7-hydroxycoumarin (18.5 g) was hydrolyzed according to method D, to give 11.9 g (64%) of product, mp 235-237 °C (EtOH). Anal. ($C_{11}H_{10}O_4 \cdot EtOH$) C, H.

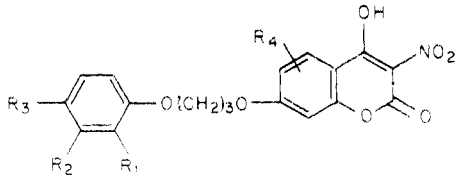
4-(Benzyloxy)-8-ethyl-7-hydroxycoumarin. Benzylation of 4,7-dihydroxy-8-ethylcoumarin as described above for the 8-methyl homologue gave 31% of the 4-benzyl derivative, mp 233-237 °C (MeOH). Anal. ($C_{18}H_{16}O_4$) C, H.

7-(Allyloxy)-4-hydroxycoumarin (175). A solution of 4′-(allyloxy)-2′-hydroxyacetophenone[14] (35.8 g, 0.186 mol) in dry PhH (400 mL) was added with stirring to NaH (10.4 g, 0.43 mol) in dry PhH (400 mL) over 30 min at reflux. After a further 10 min, a solution of diethyl carbonate (46.4 g, 0.38 mol) in dry PhH (400 mL) was added over 1 h, and the mixture was stirred at reflux overnight. After cooling, the solution was poured onto iced 2 N HCl (2 L), and the phases were separated. The dried organic phase was evaporated to dryness to give an oil, which solidified on heating to 100 °C in vacuo. Recrystallization from EtOH gave 30.5 g (75%) of **175**, mp 235-236 °C (dec). Anal. ($C_{12}H_{10}O_4$) C, H.

7-(Allyloxy)-4-(benzyloxy)coumarin (176). A suspension of **175** (21.8 g, 0.1 mol) in dry DMF (80 mL) was treated with NaH (2.4 g, 0.11 mol) and stirred at 100-110 °C during the dropwise addition of excess $PhCH_2Cl$ (12.7 mL) in dry DMF (10 mL). After a further 5 h at 100 °C, the mixture was stirred at room temperature overnight and the solvent removed in vacuo. The residue was partitioned between aqueous 1 N NaOH and $CHCl_3$, and the phases were separated. Evaporation of the dried organic phase gave an oily solid, which after recrystallization from EtOH gave 11.8 g (38%) of **176**: mp 133-134 °C; IR ν_{max} (mull) 1715, 1620 cm^{-1}. Anal. ($C_{19}H_{16}O_4$) C, H.

Acidification of the alkaline phase gave ca. 13 g of a mixture of **175** and the *C*-benzyl derivative **177**, from which 1.2 g (4%) of **177** was isolated by extraction into Et_2O and recrystallization from EtOH: mp 168-169 °C, IR ν_{max} (mull) 2700 (br), 1650, 1620, 1603 cm^{-1}. Anal. ($C_{19}H_{16}O_4$) C, H. From the insoluble material, 3 g of unreacted **175** was reclaimed.

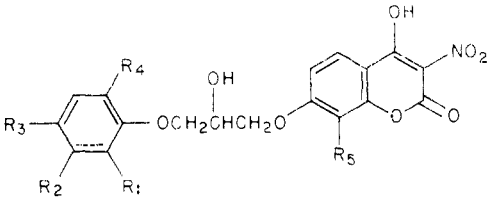
Table VIII. 4-Hydroxy-3-nitrocoumarins



compd	R_1	R_2	R_3	R_4	mp, °C[a]	formula	anal.	yield, %	meth of prep[b]	antag of SRS-A concn, in μM, for 50% inhib[c]
141	H	H	H	5-Me	134-137	$C_{19}H_{17}NO_7$	C, H, N	67	H	5.2
142	H	H	H	6-Et	154-155	$C_{20}H_{19}NO_7$	C, H, N	88	H	>10
143	H	H	H	8-Pr^n	136-137	$C_{21}H_{21}NO_7$	C, H, N	84	H	0.54-3.5
144	H	H	F	H	173-176	$C_{18}H_{14}FNO_7$	C, H, N	24	J	1
145	H	H	Cl	H	139-145	$C_{18}H_{14}ClNO_7$	C, H, Cl, N	72	H	4
146	H	H	OMe	H	175	$C_{19}H_{17}NO_8$	C, H, N	52	J	4 to >10
147	H	H	Ac	H	159-161	$C_{20}H_{17}NO_8$	C, H, N	52	H	6.1
148	H	H	CO_2Me	H	164-168	$C_{20}H_{17}NO_9$	C, H, N	87	H	>10
149	H	H	CN	H	204	$C_{19}H_{14}N_2O_7$	C, H, N	40	J	>10
150	H	H	Ph	H	180-182	$C_{24}H_{19}NO_7$	C, H, N	76	J	>20
151	H	H	CO_2H	H	239-240	$C_{19}H_{15}NO_9$	C, H, N	55	H	>20
152	H	H	Me	H	170-172	$C_{19}H_{17}NO_7$	C, H, N	21	J	4.5
153	H	Me	H	H	142	$C_{19}H_{17}NO_7$	C, H, N	64	J	4.4
154	Me	H	H	H	146	$C_{19}H_{17}NO_7$	C, H, N	70	J	0.59
155	H	tetramethylene		H	127-134	$C_{22}H_{21}NO_7$	C, H, N	40	J	>2
156	H	OH	Ac	H	199-200	$C_{20}H_{17}NO_9$	C, H, N	75	H	2.8
157	NO_2	H	Me	H	183-185	$C_{19}H_{16}N_2O_9$	C, H, N	20	H	>10
158	Pr^n	OH	Ac	H	148	$C_{23}H_{23}NO_9$	C, H, N	74	H	0.1

[a] See footnote *a*, Table VI. [b] See text and Experimental Section. [c] See footnote *c*, Table VI.

Table IX. 4-Hydroxy-3-nitrocoumarins



compd	R_1	R_2	R_3	R_4	R_5	mp, °C[a]	formula	anal.	yield, %	meth of prep[b]	antag of SRS-A concn, in μM, for 50% inhib[c]
159	H	H	H	H	H	179-180	$C_{18}H_{15}NO_8$	C, H, N	50	H	12
160	H	H	CO_2H	H	H	220-221	$C_{19}H_{15}NO_{10}$	C, H, N	49	H	>10
161	Et	H	H	H	H	140	$C_{20}H_{19}NO_8 \cdot 0.5H_2O$	C, H, N	52	J	2.7
162	Pr^n	H	H	H	H	125-126	$C_{21}H_{21}NO_8$	C, H, N	87	J	0.11
163	Pr^n	H	F	H	H	136-138	$C_{21}H_{20}FNO_8$	C, H, N	92	J	0.14
164	Pr^i	H	H	H	H	83	$C_{23}H_{23}NO_9$	C, H, N	65	J	0.16 (0.27-1.6)
165	Pr^n	H	Ac	H	H	145-148	$C_{23}H_{22}NO_9$	C, H, N	21	J	0.7
166	H	OH	Ac	H	H	120	$C_{20}H_{17}NO_{10} \cdot 0.5H_2O$	C, H, N	49	H	>20
167	Me	OH	Ac	H	H	204	$C_{21}H_{19}NO_{10} \cdot 0.5H_2O$	C, H, N	67	H	0.5
168	Pr^n	OH	Ac	H	H	201-203	$C_{23}H_{23}NO_{10}$	C, H, N	78	H	0.08 (0.03-0.23)
169	Pr^n	OH	EtCO	H	H	121	$C_{24}H_{25}NO_{10}$	C, H, N	83	H	0.15
170	H	OH	Ac	Et	H	150	$C_{22}H_{21}NO_{10} \cdot 0.5H_2O$	C, H, N	30	H	3.5
171	Pr^n	OH	Ac	H	Me	208	$C_{24}H_{25}NO_{10} \cdot H_2O$	C, H, N	12	H	0.025
172	Pr^n	OH	Ac	NO_2	Me	127	$C_{24}H_{24}N_2O_{12} \cdot 0.5H_2O$	C, H, N	58	H	1.60
173	Pr^n	OH	Ac	H	Et	168	$C_{25}H_{27}NO_{10}$	C, H, N	40	J	0.05-0.6
174	Pr^n	OH	Ac	H	Pr^n	foam	$C_{26}H_{29}NO_{10} \cdot 0.75H_2O$	C, H, N	98	H	0.20 (0.006-6.9)

[a] See footnote *a*, Table VI. [b] See text and Experimental Section. [c] See footnote *c*, Table VI.

8-Allyl-4-(benzyloxy)-7-hydroxycoumarin (178). A solution of **176** (11.3 g) in decalin (500 mL) was stirred at reflux under N_2 for 12 h, and the mixture was then cooled. After dilution with $CHCl_3$, the product was extracted into 1 N NaOH solution, from which 7.03 g (62%) of **178**, mp 205–207 °C, was isolated. Recrystallization from *i*-PrOH raised the mp to 224–226 °C. Anal. ($C_{19}H_{16}O_4 \cdot 0.25H_2O$) C, H. From the organic phase, 2.8 g of **176** was reclaimed.

5′-Ethyl-2′-hydroxy-4′-(3-phenoxypropoxy)acetophenone (22, Method A). Anhydrous K_2CO_3 (10.4 g, 0.075 mol) was added to a solution of 2′,4′-dihydroxy-5′-ethylacetophenone[18] (9.0 g, 0.05 mol) and 1-bromo-3-phenoxypropane (10.8 g, 0.05 mol) in dry Me_2CO (200 mL), and the mixture was stirred at reflux for 24 h. The mixture was cooled and filtered, and the filtrate was evaporated. Recrystallization of the residue from EtOH gave compound **22** (11.6 g, 74%), mp 60.5–61.5 °C.

6-Ethyl-4-hydroxy-7-(3-phenoxypropoxy)coumarin (54, Method B). A solution of **22** (11.4 g, 0.036 mol) in dry PhH (90 mL) was added over 30 min to a stirred suspension of NaH (1.97 g, 0.082 mol) in dry PhH (90 mL) at reflux. After a further 10 min, a solution of diethyl carbonate (8.8 g, 0.072 mol) in dry PhH (90 mL) was added over 1 h, and the mixture was stirred at reflux overnight. The cooled mixture was poured onto ice (100 g) and 2 N HCl (400 mL), and the aqueous phase was separated and

Table X. Rat Passive Peritoneal Anaphylaxis Data

compd	inhib of histamine release in rat PPA, ED_{50} mol/rat[a]
FPL55712 (II)	3.2×10^{-8} (1.2×10^{-7} to 7.9×10^{-9}, -62.8, 11)
DSCG[b]	5.2×10^{-9} (1.6×10^{-8} to 1.6×10^{-9}, -37.4, 16)
126	2.1×10^{-9} (7.4×10^{-9} to 5.6×10^{-10}, -43.6, 21)
158	4.8×10^{-9} (6.06×10^{-8} to 4.4×10^{-10}, -70.5, 16)
162	1.1×10^{-9} (4.8×10^{-9} to 2.1×10^{-10}, -33.4, 15)
163	1.7×10^{-9} (9.8×10^{-9} to 2.0×10^{-10}, -39.1, 13)
168	6.5×10^{-10} (4.1×10^{-9} to 1.1×10^{-10}, -77.6, 19)
171	2.4×10^{-9} (7.1×10^{-8} to 4.1×10^{-11}, -34.4, 14)
174	6.2×10^{-10} (1.7×10^{-9} to 2.3×10^{-10}, -76.2, 19)

[a] Dose required to reduce the concentration of histamine in the peritoneal fluid to 50% of that in controls (95% confidence limits, slope of histamine concentration/log dose line, number of rats). [b] Disodium cromoglycate.

extracted with Et_2O. The combined organic phases were extracted with 2 N NaOH, from which 9.1 g (73%) of coumarin was isolated by acidification and filtration. Recrystallization from EtOH gave material of mp 233–234 °C (dec).

4-Amino-5-methyl-7-(3-phenoxypropoxy)coumarin (99, Method C). NaH (1.40 g, 10% excess) was added to a stirred suspension of 4-amino-7-hydroxy-5-methylcoumarin[12] (9.67 g, 0.051 mol) in dry DMF (40 mL), and the mixture was refluxed for 90 min to complete formation of the sodium salt. A solution of 1-bromo-3-phenoxypropane (10.9 g, 0.051 mol) in dry DMF (10 mL) was added dropwise over 1 h to the hot solution, and refluxing was maintained for a further 4 h. After cooling, the solvent was removed in vacuo, and the residue was triturated with water, filtered, and dried. Trituration with anhydrous Et_2O gave 15.13 g (91%) of material, mp 154–155 °C. Recrystallization from EtOH after decolorizing with charcoal gave the following: mp 164–167 °C, IR ν_{max} (mull) 3500, 3300, 3150, 3070, 1695, 1645, 1605 cm^{-1}; NMR δ (Me_2SO-d_6) 2.18 (2 H, quintet, J = 6 Hz, CCH_2C) 2.68 (3 H, s, CH_3), 4.11 (2 H, t, J = 6 Hz, OCH_2), 4.20 (2 H, t, J = 6 Hz, OCH_2), 5.18 (1 H, s, vinyl CH), 6.70–7.42 (9 H, complex m, collapses to 7 H on D_2O exchange, aromatic CH + NH_2).

4-Hydroxy-5-methyl-7-(3-phenoxypropoxy)coumarin (54, Method D). A mixture of **99** (6.8 g) and 50% (v/v) H_2SO_4 (160 g) was heated with stirring at 100 °C for 8 h, and the suspension was cooled and poured into water. Filtration and recrystallization of the residue from EtOH afforded 4.4 g (65%) of **54**, mp 217–219 °C (dec).

In general, a hydrolysis time of 2–8 h was sufficient, depending on the solubility of the aminocoumarin.

4-(Benzyloxy)-7-[3-(4-carbomethoxyphenoxy)propoxy]coumarin (104, Method E). A solution of 4-(benzyloxy)-7-hydroxycoumarin (2.68 g, 0.01 mol) in dry DMF (7.5 mL) was treated with 100% NaH (0.265 g) and stirred at 100 °C for 1 h. Methyl 4-(3-chloropropoxy)benzoate[19] (2.285 g, 0.01 mol) in dry DMF (1 mL) was added portionwise over 1 h to the hot solution, and the mixture was stirred at 100 °C for an additional 4 h. After cooling, the solvent was removed in vacuo, water was added, and the precipitated solid was separated. Recrystallization from MeOH (1.5 L) gave 3.16 g (69%) of **104**, mp 153–155 °C. Anal. ($C_{27}H_{24}O_7$) C, H.

4-(Benzyloxy)-7-[2-hydroxy-3-(2-*n*-propylphenoxy)propoxy]coumarin (106, Method F). To a suspension of 4-(benzyloxy)-7-hydroxycoumarin (5.38 g, 0.02 mol) in EtOH (50 mL) was added a solution of NaOH (0.4 g) in water (10 mL) followed by 1,2-epoxy-3-(2-*n*-propylphenoxy)propane[6] (3.84 g, 0.02 mol) in EtOH (20 mL). The mixture was refluxed for 4 h and cooled, and the solvent was evaporated under reduced pressure. The residue was diluted with water, and the product was extracted into EtOAc. Evaporation of the dried extract gave an oily solid, which recrystallized from EtOH to give 4.52 g (55%) of **106**, mp 109–110 °C. Anal. ($C_{28}H_{28}O_6$) C, H.

7-[3-(4-Carbomethoxyphenoxy)propoxy]-4-hydroxycoumarin (61, Method G). A solution of **104** (2.87 g) in dry DMF (100 mL) was added to 10% palladinized charcoal (0.15 g), and the mixture was hydrogenated at atmospheric pressure until 1 equiv of H_2 was absorbed (15 min). The filtered solution was concentrated in vacuo, water was added, and the white solid was filtered off. Recrystallization from MeOH gave 1.91 g (83%) of **61**, mp 218 °C. Anal. ($C_{20}H_{18}O_7$) C, H.

Partial Debenzylation of 4,7-(Dibenzyloxy)coumarin to 7-(Benzyloxy)-4-hydroxycoumarin (48). Hydrogenation of 4,7-(dibenzyloxy)coumarin (0.358 g, mp 138–140 °C, prepared by method E) over 10% palladinized charcoal in dry DMF (15 mL) resulted in a rapid uptake of 1 equiv of H_2 to give 0.227 g (85%) of benzyloxycoumarin **48**, identical in all respects with that prepared from **16** using method B.

4-Hydroxy-3-nitro-7-(3-phenoxypropoxy)coumarin (126 method H). Fuming HNO_3 (6 mL, d 1.52) was added over 1 h to a stirred suspension of **40** (2.0 g) in $CHCl_3$ (200 mL) at 0 °C, and the dark solution was stirred at this temperature for a further 15 min. Cold 5 N HCl (100 mL) was added, and the $CHCl_3$ was removed in vacuo at 0 °C. The yellow solid which separated was filtered off, washed well with water, and recrystallized from EtOH to give 1.90 g (82%) of **126**: mp 148–150 °C (dec); IR ν_{max} (mull) 1750, 1605, 1520 cm^{-1}. Anal. ($C_{18}H_{15}NO_7$) C, H, N.

4-Hydroxy-7-[3-(2-methylphenoxy)propoxy]-3-nitrocoumarin (154, Method J). $NaNO_2$ (0.24 g, 0.003 mol) was added in one portion to a vigorously stirred suspension of **67** (1 g, 0.003 mol) in glacial AcOH (30 mL). After 2 h at ambient temperature, the reddish solution was poured into water (150 mL), and the precipitated yellow solid was filtered off. Recrystallization from aqueous EtOH gave 0.82 g (70%) of **154**, mp 146 °C. Anal. ($C_{19}H_{17}NO_7$) C, H, N.

SRS-A Antagonism. The material used as an SRS-A standard was the heated supernatant of combined peritoneal fluids collected, 5 min after antigen challenge, from rats subjected to passive peritoneal anaphylaxis.[20] It was assayed on the isolated terminal strip of a guinea pig ileum preparation suspended in 4 mL of Tyrode's solution containing atropine (5×10^{-7} M) and mepyramine maleate (10^{-6} M), as described previously.[20] The antagonist, in 0.1 mL of water, was added to the Tyrode's solution 30 s before a volume of the SRS-A solution (max 0.2 mL) that in the absence of the antagonist produced a substantial but less than maximal response. At least three different inhibitory concentrations of the antagonist were used, and the activity of each antagonist was determined on more than one sample of guinea pig ileum. The concentration of the antagonist, in the 4 mL of solution in the bath, required for 50% inhibition of the SRS-A response was determined by eye from the log concentration–inhibition curve or by calculation of the line of best fit and 95% confidence limits as described in ref 21.

Passive Peritoneal Anaphylaxis. Passive peritoneal anaphylaxis (PPA) was carried out in the rat as described previously.[20] Briefly, antiserum to ovalbumin (Sigma grade III) was raised in rats using Bordetella pertussis vaccine (Burroughs Wellcome, London) as adjuvant. Rats were given intraperitoneal injections of 2 mL of a 1:5 dilution of the rat antiserum in isotonic saline. Two hours later, 0.3 mL of a 5% solution of Pontamine Sky Blue (Raymond A. Lamb, London) in isotonic saline was injected intravenously, followed immediately by an intraperitoneal injection with 1 mL of a solution of a test compound in saline or of saline alone for control rats. Each dose of a compound or of saline was given to groups of five to seven rats, and treatments were randomized. After 30 s, the rats were given an intraperitoneal injection of 5 mL of a Tyrode solution containing 50 μg/mL heparin and 0.4 mg/mL ovalbumin. Exactly 5 min after challenge, the rats were stunned and bled, and the peritoneal fluids were collected and assayed for histamine as described.[20] The line of best fit for the log dose–response curve and the 95% confidence limits were calculated as described.[21]

References and Notes

(1) J. S. G. Cox, J. E. Beach, A. M. J. N. Blair, A. J. Clarke, J. King, T. B. Lee, D. E. E. Loveday, G. F. Moss, T. S. C. Orr, J. T. Ritchie, and P. Sheard, *Adv. Drug Res.*, **5**, 115 (1970).

(2) R. P. Orange and K. F. Austen, *Int. Arch. Allergy Appl. Immunol.*, **41**, 79 (1971).
(3) D. H. Bryant, M. W. Burns, and L. Lazarus, *J. Allergy Clin. Immunol.*, **56**, 417 (1975).
(4) (a) D. R. Buckle, N. J. Morgan, J. W. Ross, H. Smith, and B. A. Spicer, *J. Med. Chem.*, **16**, 1334 (1973); (b) D. R. Buckle, B. C. C. Cantello, H. Smith, and B. A. Spicer, *J. Med. Chem.*, **18**, 391 (1975); (c) *ibid.*, **18**, 726 (1975); (d) D. R. Buckle, B. C. C. Cantello, H. Smith, R. J. Smith, and B. A. Spicer, *J. Med. Chem.*, **20**, 1059 (1977).
(5) B. A. Spicer, J. W. Ross, and H. Smith, *Clin. Exp. Immunol.*, **21**, 419 (1975).
(6) R. A. Appleton, J. R. Bantick, T. R. Chamberlain, D. N. Harden, T. B. Lee, and A. D. Pratt, *J. Med. Chem.*, **20**, 371 (1977).
(7) J. Augstein, J. B. Farmer, T. B. Lee, P. Sheard, and M. L. Tattersall, *Nature (London), New Biol.*, **245**, 215 (1973).
(8) I. M. Heilbron and D. W. Hill, *J. Chem. Soc.*, 1705 (1927).
(9) C. S. Marvel and A. L. Tanenbaum, *J. Am. Chem. Soc.*, **44**, 2645 (1922).
(10) O. Stephenson, *J. Chem. Soc.*, 1571 (1954).
(11) S. Iguchi, *Yakugaku Zasshi*, **72**, 122 (1952); *Chem. Abstr.*, **46**, 11187a (1952).
(12) S. Iguchi and N. Utsugi, *Yakugaku Zasshi*, **73**, 1290 (1953); *Chem. Abstr.*, **49**, 304g (1955).
(13) C. F. Spencer, C. H. Stammer, J. O. Rodin, E. Walton, F. W. Holly, and K. Folkers, *J. Am. Chem. Soc.*, **78**, 2655 (1956).
(14) W. Baker and O. M. Lothian, *J. Chem. Soc.*, 628 (1935).
(15) J. V. Braun, *Chem. Ber.*, **43**, 2837 (1910).
(16) A. A. Shamshurin and L. L. Simonova, *Chem. Abstr.*, **65**, 10572h (1966).
(17) A. Russell, J. R. Frye, and W. L. Mauldin, *J. Am. Chem. Soc.*, **62**, 1441 (1940).
(18) K. W. Rosenmund, R. Buchwald, and Th. Deligiannis, *Arch. Pharm. (Weinheim, Ger.)*, **271**, 342 (1933).
(19) J. D. Genzer, C. P. Huttrer, and G. C. van Wessem, *J. Am. Chem. Soc.*, **73**, 3159 (1951).
(20) J. W. Ross, H. Smith, and B. A. Spicer, *Int. Arch. Allergy Appl. Immunol.*, **51**, 226 (1976).
(21) K. A. Brownlee, "Statistical Theory and Methodology in Science and Engineering", Wiley, New York, N.Y., 1965.

Synthesis and Pharmacologic Characterization of an Alkylating Analogue (Chlornaltrexamine) of Naltrexone with Ultralong-Lasting Narcotic Antagonist Properties

P. S. Portoghese,* D. L. Larson, J. B. Jiang,

Department of Medicinal Chemistry, College of Pharmacy

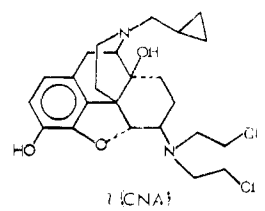
T. P. Caruso, and A. E. Takemori

Department of Pharmacology, School of Medicine, University of Minnesota, Minneapolis, Minnesota 55455.
Received July 19, 1978

Chlornaltrexamine (CNA) produces ultralong-lasting (3–6 days) narcotic antagonism in mice and persistent stereospecific binding to rat-brain homogenate. Protection studies in mice suggest that CNA mediates its narcotic antagonist effects by interacting with the same receptors that are occupied by naloxone. A single icv dose of CNA also has been found to inhibit the development of physical dependence in mice for at least 3 days. These studies suggest that CNA exerts its sustained effects by selective covalent association with opioid receptors.

Narcotic antagonists are used extensively as pharmacologic tools for the investigation of opioid receptors.[1] Indeed, the recent research literature attests to the impact that such antagonists have made in this active research area.[2] However, the reversible nature of conventional narcotic antagonists (e.g., naloxone and naltrexone) is an inherent limitation to their utility, particularly with regard to the use of such compounds in the isolation and purification of opioid receptors. Ligands that specifically form covalent bonds with opioid receptors, therefore, would represent a major addition to the armamentarium of agents employed as investigational tools.

For this reason, considerable effort has been devoted to the design and synthesis of agents having this potential.[3-9] In this publication we describe our detailed studies concerning the first example of an alkylating agent which covalently associates with receptors which mediate narcotic antagonist activity in vivo and in vitro. We have named this ultralong-acting antagonist, chlornaltrexamine (CNA) **1**.[10]



1 (CNA)

Design Considerations and Chemistry. Three factors will affect the efficiency of receptor alkylation once an affinity labeling agent[11] reaches the biophase. These are (1) the affinity of the ligand for the receptor, (2) the intrinsic chemical reactivity of the alkylating moiety, and (3) the proximity of the reactive moiety of the ligand to a receptor nucleophile in the drug-receptor complex. While criteria 1 and 2 can be met without much difficulty, criterion 3 is not easily attained because there is no information on the location of nucleophiles on or adjacent to the receptor.

Since previous studies involving the attachment of reactive moieties to the aromatic ring of the *N*-phenethyl group of anileridine[4,6] and *N*-phenethyl-3-hydroxymorphinan[9] gave inconclusive results, we decided to modify our approach by attaching the reactive moiety to a narcotic antagonist rather than an agonist. Also, the position of attachment was changed so as to explore a different receptor locus for the nucleophile.

Inasmuch as naltrexone[12] (**2**) is a relatively "pure" narcotic antagonist with high receptor affinity,[13] we selected it for modification. The C-6 center was chosen as the point of attachment for the bis(α-chloroethyl)amino group because a variety of substituents in this position do not destroy receptor affinity.[13-15]

CNA (**1**) was synthesized from naltrexone (**2**) by two different routes (see Scheme I). In the first route, reductive amination of **2** with $NaCNBH_3$[16] and diethanol-

0022-2623/79/1822-0168$01.00/0 © 1979 American Chemical Society