mM DTT), 0.03–0.1 mL of enzyme solution, 0.1 mL of substrate solution, and enough 1.15% KCl to bring the final volume to 2.5 mL. A 2-min temperature equilibration period was initiated by addition of the enzyme solution to 25-mL Erlenmeyer flasks, containing KCl and buffer, in a 37 °C shaker bath. Reactions were started by the addition of the substrate solution and were carried out at 37 °C in air. Reactions were terminated by the addition of 2.5 mL of 20% (w/v) trichloroacetic acid in 50% (v/v) $EtOH/H_2O$, and the resulting mixture was centrifuged for 20 min in a desk-top centrfuge. The supernatants were analyzed spectrophotometrically at 497 nm.[11] *N*-Arylhydroxamic acids were omitted for the incubation mixtures that were used as reference standards. Incubations were performed in triplicate.

The substrate solutions consisted of the *N*-arylhydroxamic acid and AAB dissolved in either 95% EtOH (compounds 1 and 2) or $Me_2SO/MeOCH_2CH_2OH$ (1:1). The final protein concentration was 1.0 mg/mL except for assays with **5** (0.5 mg/mL) and **7** (0.6 mg/mL). The final *N*-arylhydroxamic acid concentration was 1.0 mM except for **5** (0.5 mM) and **7** (0.6 mM). The final AAB concentration was 0.15 mM except for assays with **5** and **7** in which it was 0.075 and 0.10 mM, respectively. The incubation time for assays with **5** was 3.5 min; for all other compounds it was 2.0 min.

Kinetics of AHAT Inactivation. The preincubation mixtures contained 1 mL of sodium pyrophosphate buffer (pH 7, 1 mM DTT), *N*-arylhydroxamic acid (0.005–1.0 mM final concentration) dissolved in 0.05 mL of 95% EtOH or 0.05 mL of $Me_2SO/MeOCH_2CH_2OH$ (1:1), 0.03–0.1 mL of hamster hepatic AHAT preparation (protein concentration as described under transacetylation assay), and sufficient 1.15% KCl to give a final volume of 2.45 mL. Control flasks contained 0.05 mL of the appropriate solvent instead of the *N*-arylhydroxamic acid solution. The preincubations were started by the addition of enzyme and were carried out at 37 °C in air. At the end of the preincubation periods (1–15 min), 0.05 mL of a substrate solution consisting of the appropriate *N*-arylhydroxamic acid and AAB were added as described under the description of the transacetylation assay in order to measure the amount of remaining enzyme activity. The mixtures were then incubated in air at 37 °C for either 2 or 3.5 min (compound **5**) and the reactions were terminated and the mixtures were assayed as described above. The calculation of the kinetic constants was accomplished as reported previously, and is summarized under Biological Results.[7a,15]

Inactivation of AHAT in the Presence of Nucleophiles. Incubations were conducted in the same manner as in the kinetic studies except each flask contained 10 mM of a low-molecular-weight nucleophile (L-methionine, L-cysteine, glutathione, guanosine phosphate) or 1 mg/mL of tRNA. A saturating concentration of inactivator (*N*-arylhydroxamic acid) was used and preincubation times (2–20 min) were adjusted such that maximum inactivation of the enzyme occurred. Control flasks contained nucleophiles, but 0.05 mL of the appropriate solvent was added instead of the inhibitor solution. The presence of the nucleophiles did not significantly affect the AAB transacetylion rates in the control flasks. Preincubations with inhibitors were initiated by addition of enzyme. At the end of the preincubation period, 0.05 mL of substrate solution (*N*-arylhydroxamic acid and AAB) was added in order to measure the remaining enzyme activity. The incubations were carried out and terminated as described above. The method of determination of the amount of protection of AHAT from inactivation by the *N*-arylhydroxamic acids was described previously and is summarized under Biological Results.[7a]

Dialysis Experiments. The incubations were run on a scale 8–16 times that of a standard incubation. The 16× standard incubation mixture contained 16.0 mL of 0.05 M sodium pyrophosphate buffer (pH 7, 1 mM DTT), 0.1 mM *N*-arylhydroxamic acid (dissolved in 0.8 mL of $Me_2SO/MeOCH_2CH_2OH$, 1:1), 0.48–1.6 mL of enzyme solution (1 mg/mL final protein concentration) and sufficient 1.15% KCl to give a 40-mL final incubation volume. The control flasks contained 0.8 mL of solvent instead of the *N*-arylhydroxamic acid solution. The incubation period was initiated by the addition of *N*-arylhydroxamic acid solution. The mixture was incubated in air at 37 °C. The incubation times (5–40 min) were adjusted to obtain maximum inactivation of the enzyme. At the end of the incubation period, samples (2.45 mL) were removed for AHAT activity assay. The remaining incubation mixture was dialyzed against four 800-mL portions of cold 0.05 M sodium pyrophosphate buffer (pH 7, 1 mM DTT) containing 2.04% $Me_2SO/MeOCH_2CH_2OH$, 1:1). The dialysis buffer solutions were changed four times during the 4-h dialysis period. Nitrogen was bubbled through the buffer before and during the dialysis. At the end of the dialysis period, samples (2.45 mL) were removed and assayed for activity. The substrate solution used to initiate the transacetylation assay contained AAB and the same *N*-arylhydroxamic acid used used in the preincubation. The concentrations of substrates and the solvents used are given under the description of the transacetylation assay.

Acknowledgment. This investigation was supported in part by NIH Grant CA-24427.

Registry No. 1, 53-95-2; 2, 2508-18-1; 3, 92901-04-7; 4, 92901-05-8; 5, 70952-93-1; 6, 92901-06-9; 7, 92901-07-0; 8, 92901-08-1; 9, 92901-09-2; 10, 92901-10-5; 12, 6939-05-5; 13, 23055-47-2; 14, 34172-49-1; 15, 92901-11-6; 16, 6633-40-5; 17, 54961-21-6; 18, 92901-12-7; 19, 92901-13-8; 20, 39150-37-3; AHAT, 52660-15-8; 2-nitrofluorene, 607-57-8; ethylene glycol, 107-21-1; 3-chloroperbenzoic acid, 937-14-4; glutathione, 70-18-8; L-cysteine, 52-90-4; L-methionine, 63-68-3; guanosine 2′-phosphate, 130-50-7; guanosine 3′-phosphate, 117-68-0.

[1,4]Benzoxazine-2,3-diones as Antiallergic Agents

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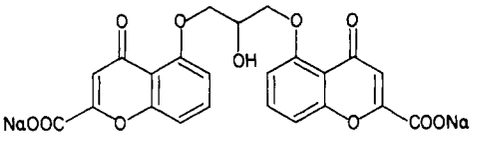
The synthesis of a series of [1,4]benzoxazine-2,3-diones and a new class of compounds, benzobisoxazinetetrones, is described. These compounds were evaluated for their effect in the rat mast cell (RMC) test passively sensitized in vitro with rat antiovalbumin serum and for their effect in inhibitory passive cutaneous anaphylaxis (PCA) in the rat. Some of these compounds are of the same potency level as disodium cromglycate in the RMC test and some are effective orally in PCA.

Disodium cromoglycate (DSCG, **1**[1]) is the only compound on the market that is indicated for prophylactic treatment of certain types of extrinsic asthma. It is claimed to act by inhibiting the release of mediators stimulated by antibody–antigen interactions.[2] Because



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DSCG is effective in asthma only when inhaled,[3] a search

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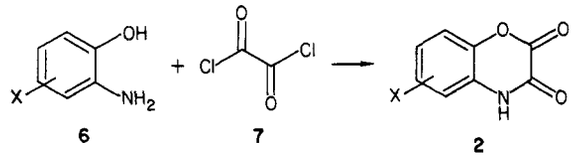
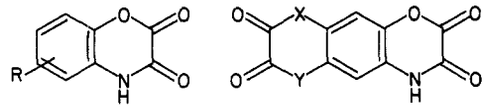
Scheme I



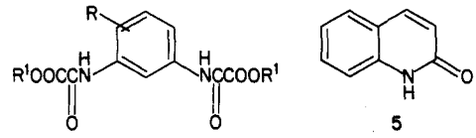
for orally effective prophylactic agents has continued since its introduction. Recently there have been numerous reports of compounds that possess antiallergic properties similar to cromolyn in animal test systems, but many of them have failed in humans—either due to lack of activity or due to toxicity. As a result, the search for new compounds for use in oral treatment of asthma continues.

We report herein, two new classes of compounds, the benzoxazinediones **2** and benzobisoxazinetetrones **3**, found to be potent inhibitors of antigen-induced histamine release from rat mast cells (RMC) and which are active orally and intraperitoneally in the rat passive cutaneous (PCA) test.



3a, X=O; Y=NH
b, X=NH; Y=O

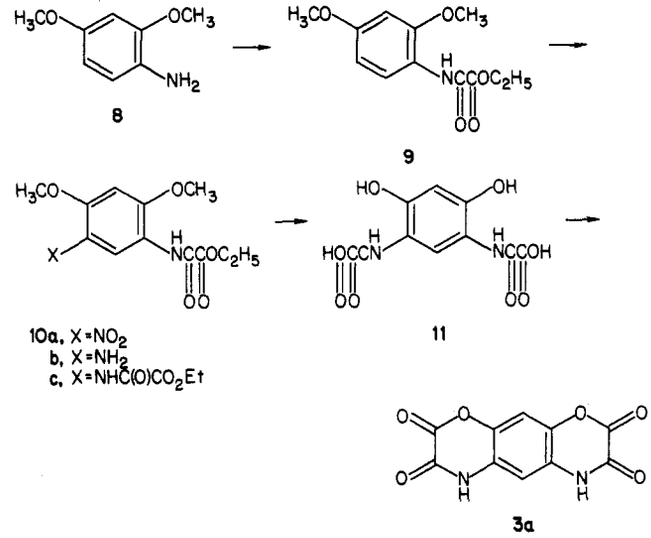
Among recently discovered, orally active (in animals) antiasthma agents are the oxanilic acids and esters (**4**) reported by Wright and Jonnson.[4] Similar structures were independently discovered by Sellstedt and co-workers.[5] These molecules contain both the aniline and oxalic acid moieties; because both anilides and oxalic acids possess toxic potential, we considered ways in which this potential might be negated while retaining the structural feature required for activity.



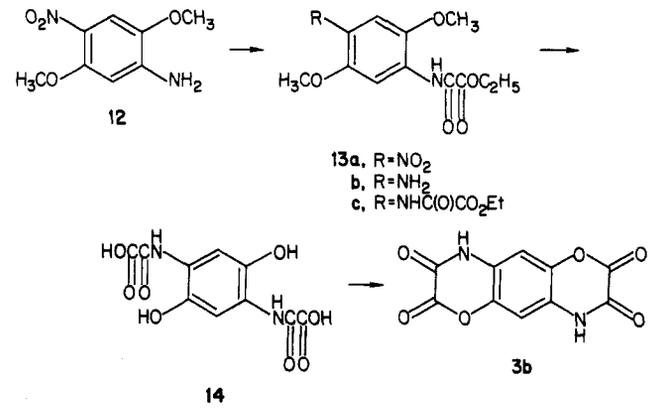
It is well known that anilides, e.g., acetanilide, can be rendered less toxic by introducing an oxygen function, e.g., as in *p*-hydroxyacetanilide or acetophenetidine. We were also aware from our previous studies in the carbostyril series (**5**) that cyclized anilides are stable and relatively nontoxic. Theoretically, combining these two into one structure, one arrives at a benzobisoxazinetetrone type structure, **3**. Such compounds have not been described previously.

A benzoxazinedione prototype molecule (**2**, R = H) was synthesized and surprisingly found to possess interesting activity, initiating the program described in this paper. Benzoxazinedione was first reported in 1932;[6] no biological activity of these compounds has previously been reported.

Scheme II



Scheme III



Chemistry. The benzoxazinediones (Table I) were prepared readily by reaction of the appropriate *o*-aminophenol with oxalyl chloride[7] (Scheme I) in boiling toluene. The two isomeric benzobisoxazinetetrone heterocyclic systems have never been described previously. The synthesis of benzo[1,2-*b*:5,1-*b*]bis[1,4]oxazinetetrone (**3a**) is shown in Scheme II. Nitration of the oxanilic ester **9** gave **10a** as the only product as indicated by NMR data. Hydrogenation of **10a** followed by reaction with ethyloxalyl chloride gave the bisoxanilate **10c**. Demethylation and hydrolysis of the diester was accomplished in a single step by treatment of **10c** with boron tribromide. The resulting acid **11** was cyclized to **3a** in refluxing acetic anhydride/acetic acid.

Isomeric benzo[1,2-*b*:4,5-*b*]bis[1,4]oxazinetetrone (**3**:) was prepared in a similar fashion (Scheme III). Thus reaction of 2,5-dimethoxy-4-nitroaniline (**12**) with ethyloxalyl chloride gave **13a**. The nitro group was reduced by catalytic hydrogenation to give **13b**, which was reacted with ethyloxalyl chloride to give the bisoxaniliate **13c**. Demethylation and deesterification by boron tribromide followed by ring closure in acetic acid–acetic anhydride gave the benzobisoxazinetetrone **3b**.

Results and Discussion

As a class, benzoxazine-2,3-diones are potent inhibitors of antigen-induced release of histamine from passively

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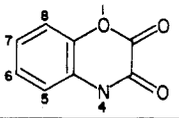
Table I. Inhibition of RMC and Rat PCA by Benzoxazinediones



compd	substit	formula	mp, °C	RMC:[a] I_{50}, μM	PCA % inhib 50 mg/kg (ip)	PCA 25 mg/kg (po)	PCA ED_{50} (po), mg/kg
2a	H	$C_8H_5NO_3$	264–266[b]	42	70		
2b	6-Cl	$C_8H_4ClNO_3$	289–292[b]			14	
2c	8-OCH_3	$C_9H_7NO_4$	251–254	7 (2)	89[c]	88[c]	14
2d	6-OCH_3	$C_9H_7NO_4$	249–253	15		21[d]	
2e	7-OCH_3	$C_9H_7NO_4$	248–252	28		33[c]	
2f	7-OH	$C_8H_5NO_4$	184–186	170			
2g	6-CF_3	$C_9H_4F_3NO_3$	208–213	100		20[d]	
2h	7-CH_3	$C_9H_7NO_3$	148–149	32	97[c]		
2i	6-NO_2	$C_8H_4N_2O_5$	268–274[b]	0.8	91[c]	24[e,f]	
2j	7-CN	$C_9H_4N_2O_3$	298–302	15		14[f]	23 (ip)
2k	6-CO_2Et	$C_{11}H_9NO_5$	232–234	10		0	
2l	7-CO_2H	$C_9H_5NO_5$	>300	16	83[c]	7[e]	
2m	6-$NHCOCO_2Et$	$C_{12}H_{10}N_2O_6$	266–268	60			
2n	6,7-$(OCH_3)_2$	$C_{10}H_9NO_5$	268–271	30	100[c]	45[c]	20
2o	6-CO_2CH_3, 8-OCH_3	$C_{11}H_9NO_6$	285–287	3.4 (2)		35	
2p	8-OCH_3, 6-CH_2-CH=CH_2	$C_{12}H_{11}NO_4$	204–206[g]	10		77[c]	7
2q	6,7-$CH_2CH_2CH_2CH$	$C_{12}H_{11}NO_3$	262–266	10		15	96
2r	6,7-CH=CHCH=CH	$C_{12}H_7NO_3$	6	45[e]	45[e]	11	
2s	5,6-CH=CHCH=CH	$C_{12}H_7NO_3$	>300	<100	18		
2t	4-CH_3	$C_9H_7NO_3$	175–178	*h*			
3a	6,7-NHC(O)COO	$C_{10}H_4N_2O_6$	>300	3 (2)		13	>125
3b	6,7-OCOCONH	$C_{10}H_4N_2O_6$	>300	1.2 (2)		0	>125
DSCG				4.5 (96)			6 (ip)

[a] Inhibition of AIR was concentration dependent. DSCG was routinely used as positive control in each experiment and has an I_{50} values of 4.5 ± 0.5 μM (average ± SE, N = 96). Number of experiments indicated in parentheses. [b] Reference 7. [c] $p < 0.001$. [d] $p < 0.05$. [e] $p < 0.01$. [f] Inhibition at 50 mg/kg. [g] Reference 6. [h] No activity at 100 μM.

sensitized rat mast cells (RMC) (Table I). In the RMC assay some compounds showed activity similar to that of DSCG.

In the in vitro screen, substitution on the aromatic ring resulted, in general, in increased activity relative to the unsubstituted compounds. There was no consistent pattern relating the effect of introducing electron-donating or electron-withdrawing groups. Disubstitution generally enhanced potency. The benzobisoxazinetetrones **3a** and **3b** were among the more potent compounds in this series.

There is poor SAR correlation between the in vitro and in vivo screens. The 6-nitrobenzoxazinedione **2i** with an I_{50} value of 0.8 μM in RMC is not active orally in PCA. Two compounds, **2c** and **2p**, with a methoxy group at the 8-position showed good PCA activity when given orally. Although the PCA activity found for compounds in this series generally seems to be less active than that of the oxanilic acids reported in the literature,[4,5] the most active compound, **2p**, had an ED_{50} value of 7 mg/kg (po), which is as active as DSCG (ED_{50} = 6 mg/kg) when given intraperitoneally.

We have described the synthesis and antiallergic activities of a series of benzoxazinediones and a new class of compounds, benzobisoxazinetetrones (**3a** and **3b**). Several of these compounds showed the same level of potency as DSCG in the tests used. As further modification of this series of compounds led to the discovery of other new classes of antiallergic agents[10,11] that displayed even more potent antiallergic activities, we did not further investigate the possibility that benzoxazine-2,3-diones hydrolyzed in vivo to the corresponding oxanilic acids.

Experimental Section

Melting points were determined on a Thomas-Hoover apparatus and are uncorrected. Spectra were recorded for all compounds and were consistent with the assigned structure. NMR were recorded on a Varian EM-390 spectrometer. IR spectra were recorded on a Perkin-Elmer Model 298 spectrophotometer. All compounds had elemental analyses for C, H, and N within ±0.4% of theoretical values unless otherwise indicated.

General Procedure for Preparation of Benzoxazinediones.[7] To oxalyl dichloride (0.6 mol) solution in toluene at 100 °C was added an appropriate amount of *o*-aminophenol (0.5 mol) portionwise over a period of 1 h. The reaction mixture was heated to reflux for 2 h and then cooled to room temperature. The desired product was isolated by filtration followed by recrystallization.

Compounds **2a–s** were prepared according to the above procedure.

Ethyl *N*-(2,4-Dimethoxyphenyl)oxanilate (9). To a mixture of 2,4-dimethoxyaniline (30.6 g, 0.199 mol) and triethylamine (50 mL, 0.36 mol) in 250 mL of CH_2Cl_2 was added ethyloxalyl chloride (27.5 mL, 0.199 mol) dropwise over a period of 30 min at room temperature. After addition was completed, the reaction was stirred for an additional 3 h. The organic solution was washed with water, dried, and evaporated to give 40 g (59%) of an off-white solid: mp 242–244 °C; NMR ($CDCl_3$) δ 1.4 (t, 3 H), 3.8 (s, 3 H), 3.9 (s, 3 H), 4.38 (q, 2 H), 6.5 (q, 2 H), 8.3 (s, 1 H).

Ethyl *N*-(2,4-Dimethoxy-5-nitrophenyl)oxanilate (10a). Compound **9** (25 g, 98.4 mmol) in 200 mL of CH_2Cl_2 and 200 mL of acetic acid was nitrated with 4 mL of fuming nitric acid, which was added in four portions over a period of 30 min. The reaction mixture was stirred at room temperature for 3 h. Filtration and

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recrystallization (ethyl acetate/hexane) gave 22 g (75%) of a pale yellow solid: mp 197–198 °C; NMR ($CDCl_3$) δ 1.4 (t, 3 H), 3.95 (s, 3 H), 4.0 (s, 3 H), 4.4 (q, 2 H), 6.6 (s, 1 H), 9 (s, 2 H). Anal. ($C_{12}H_{14}N_2O_7$) C, H, N.

Ethyl *N*-(2,4-Dimethoxy-5-aminophenyl)oxanilate (10b). The nitro compound **10a** (12 g, 40.3 mmol) and 1.5 g of 5% Pd/C in 300 mL of ethanol were hydrogenated at 40 psi overnight. Filtration followed by evaporation of solvent gave 10.5 g (96.5%) of **8b** as a yellow solid: NMR ($CDCl_3$) δ 1.42 (t, 3 H), 3.88 (s, 6 H), 4.38 (22 H), 6.48 (s, 1 H), 4.84 (s, 1 H). This compound was used without further purification.

Diethyl 4,6-Dimethoxy-1,3-phenylenediiminobis(oxoacetate) (10c). To a solution of the aniline derivative **10b** (21 g, 78.36 mmol) and triethylamine (30 mL) in 300 mL of $CHCl_3$ was added dropwise a solution of ethyloxalyl chloride (11.6 mL, 79 mmol) in 40 mL of CH_2Cl_2 over a period of 30 min. The reaction mixture was stirred for an additional 3 h at room temperature. The organic solution was washed with water, 1 N HCl, and 5% $NaHCO_3$ solution, dried, and evaporated to give 2.7 g of a light yellow solid. Recrystallization from ethanol gave 23 g (79.7%) of product: mp 183–184 °C; NMR ($CDCl_3$) δ 1.4 (t, 6 H), 3.9 (s, 6 H), 4.38 (q, 4 H), 6.5 (s, 1 H), 9.18 (s, 1 H). Anal. ($C_{16}H_{20}N_2O_8$) C, H, N.

4,6-Dihydroxy-1,3-phenylenediiminobis(oxoacetic acid) (11). A mixture of **10c** (8 g, 21.7 mmol) and 10 mL of boron tribromide in 30 mL of CH_2Cl_2 was heated to reflux overnight. Excess boron tribromide was decomposed by water. The product was filtered and dried to give 4.3 g (63.5%) of **11** as a yellow solid. This compound was used without further purification: NMR (Me_2SO-d_6) δ 6.57 (s, 1 H), 8.53 (s, 1 H).

Benzo[1,2-*b*:5,4-*b*]bis[1,4]oxazine-2,3,7,8-tetrone (3a). To a refluxing solution of the crude compound **11** (4.3 g, 13.8 mmol) in 150 mL of acetic acid was added dropwise a solution of 4 g of acetic anhydride in 10 mL of acetic acid over a period of 40 min. Refluxing was continued for 2 h after the addition of acetic anhydride. The insoluble product thus formed was filtered hot, and the solid product was washed well with methylene chloride to give 2.7 g (79%) of tan colored powder: mp >300 °C; NMR (Me_2SO-d_6) δ 6.8 (s, 1 H), 7.3 (s, 1 H), 11.8 (s, 2 H). Anal. ($C_{10}H_4N_2O_6$) C, H, N.

Ethyl *N*-(2,5-Dimethoxy-4-nitrophenyl)oxanilate (13a). To a mixture of 5-dimethoxy-4-nitroaniline (**12**; 25 g, 0.126 mol), 13 mL of triethylamine (0.1262 mol) in 200 mL of CH_2Cl_2, and 40 mL of DMF was added dropwise a solution of ethyloxalyl chloride (18.9 g, 0.139 mol) in 50 mL of CH_2Cl_2 at room temperature over a period of 30 min. After addition, the reaction mixture was concentrated and the residue was washed with water and filtered. Recrystallization from methylene chloride–hexane gave 3 g (95.6%) of product: mp 146–147 °C; NMR ($CDCl_3$) δ 1.43 (t, 3 H), 3.97 (s, 6 H), 4.32 (q, 2 H), 7.53 (s, 1 H), 8.04 (s, 1 H). Anal. ($C_{12}H_{14}N_2O_7$) C, H, N.

Ethyl *N*-(2,5-Dimethoxy-4-aminophenyl)oxanilate (13b). A mixture of the nitro compound **13a** (30 g, 0.1 mol) and 3.6 g of 5% Pd/C in 600 mL of EtOH was hydrogenated at 40 psi overnight. The reaction mixture was filtered and the catalyst was washed well with CH_2Cl_2. The organic solution was evaporated to dryness to give 26 g (96%) of **13b**. This was used for the next reaction without further purification.

Diethyl 2,5-Dimethoxy-1,4-phenylenediiminobis(oxoacetate) (13c). Ethyloxalyl chloride (15.5 g, 0.114 mol) in 30 mL of CH_2Cl_2 was added dropwise to a solution of the crude product **13b** obtained above (23 g, 91.3 mmol) and triethylamine (40 mL) in 300 mL of CH_2Cl_2 at room temperature over a period of 30 min. After the mixture was stirred for an additional 2 h, the solvent was evaporated and water (250 mL) was added to the residue. Filtration followed by recrystallization from ethanol gave 32.6 g (97%) of **13c** as a cream yellow solid: mp 225–226 °C; NMR ($CDCl_3$) δ 1.45 (t, 6 H), 3.9 (s, 6 H), 4.42 (q, 4 H), 8.2 (s, 2 H). Anal. ($C_{16}H_{20}N_2O_8$) C, H, N.

2,5-Dihydroxy-1,4-phenylenediiminobis(oxoacetic acid) (14). A mixture of compound **13c** (15 g, 40.7 mmol) and boron tribromide (10 mL) in 80 mL of CH_2Cl_2 was heated on an oil bath at 130 °C for 5 days. Excess boron tribromide was decomposed by water, and the reaction mixture was filtered and the solid product was dried to give 10.5 g of crude. This was used for the next reaction without further purification.

Benzo[1,2-*b*:4,5-*b*]bis[1,4]oxazine-2,3,7,8-tetrone (3b). To a refluxing acetic acid solution of the crude product **14** (9 g, 28.8 mol, in 80 mL) was added dropwise a solution of 8.5 g of acetic anhydride in 10 mL of acetic acid over a period of 2 h. The reaction mixture was heated for an additional 2 h and then filtered while hot. The product obtained (5.2 g, 66.4%) is analytically pure: mp >300 °C; NMR (Me_2SO-d_6) δ 6.83 (s).

Antigen-Induced Release of Histamine from Rat Mast Cells (RMC).[8] The effect of test compounds on antigen-induced release of histamine (AIR) from passively sensitized rat mast cells was determined according to the procedure of Khandwala et al.[8] Briefly, washed RMC were passively sensitized with rat antiovalbumin serum in vitro, washed, and challenged with ovalbumin to cause release of histamine. Test compounds were added simultaneously with the antigen. Both spontaneous histamine release in the absence of antigen and AIR are expressed as percent of total extractable histamine in the cells. The compound activity is expressed as percent inhibition of AIR or as the I_{50} value (concentration of the test compound required to inhibit AIR by 50%). Test compounds were dissolved in Me_2SO (final concentration of Me_2SO was 0.17% and did not affect AIR).

Passive Cutaneous Anaphylaxis in the Rat (PCA). The effect of compounds on IgE-mediated cutaneous wheal formation in the rat was determined by a modification of the method of Watanabe and Ovary.[9] Antiserum for these studies was prepared according to the following immunization protocol. Male Sprague–Dawley rats (approximately 250 g) were injected intramuscularly on days 0, 2, and 4 with 10 μg of ovalbumin and 20 mg of aluminum hydroxide (Amphojel) in 1 mL of saline. On day 0 each rat also was given 10^9 *Bordatella pertussia* organisms by the intraperitoneal route. Rats were exsanguinated on day 8, and serum was collected by the usual methods.

The method for passive cutaneous anaphylaxis was as follows. Naive rats were sensitized at dorsal sites by intradermal injection of the syngeneic IgE antiovalbumin antiserum (1:20 dilution). After a latency period of 48 h to allow cytophilic antibodies to bind to the cutaneous mast cells, groups of four rats were given either vehicle (1% methylcellulose, 3 mL) or graded doses of compound (1 dose/group). Rats were challenged intravenously with antigen (4 mg of ovalbumin) in 1% Evans blue dye 10 min after oral administration of test compound. Thirty minutes after antigen challenge, the rats were sacrificed by cervical dislocation, the dorsal skins reflected, and blued wheal areas measured. Mean values ±SE for wheal areas in control and drug-treated groups were determined and compared statistically by the Student's *t* test.

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Registry No. **2a**, 3597-63-5; **2b**, 27393-19-7; **2c**, 81055-28-9; **2d**, 81055-30-3; **2e**, 81066-48-0; **2f**, 81055-25-6; **2g**, 92643-66-8; **2h**, 81055-21-2; **2i**, 27393-20-0; **2j**, 81055-22-3; **2k**, 92643-67-9; **2l**, 81055-27-8; **2m**, 81055-23-4; **2n**, 81055-29-0; **2o**, 81055-32-5; **2p**, 81055-31-4; **2q**, 81066-49-1; **2r**, 27383-80-8; **2s**, 92643-68-0; **2t**, 72985-52-5; **3a**, 92643-69-1; **3b**, 92643-70-4; **6a**, 95-55-6; **6b**, 95-85-2; **6c**, 40925-71-1; **6d**, 20734-76-3; **6e**, 40925-70-0; **6f**, 13066-95-0; **6g**, 454-81-9; **6h**, 2835-98-5; **6i**, 99-57-0; **6j**, 55586-26-0; **6k**, 13052-92-1; **6l**, 2374-03-0; **6m**, 92643-71-5; **6n**, 7107-04-2; **6o**, 92643-72-6; **6p**, 92643-73-7; **6q**, 28094-04-4; **6r**, 5417-63-0; **6s**, 2834-92-6; **9**, 24451-12-5; **10a**, 92643-74-8; **10b**, 92643-75-9; **10c**, 92643-76-0; **11**, 92643-77-1; **13a**, 92643-78-2; **13b**, 92643-79-3; **13c**, 92643-80-6; **14**, 92643-81-7; oxalyl dichloride, 79-37-8; 2,4-dimethoxyaniline, 2735-04-8; ethyl oxalyl chloride, 4755-77-5.