toires Delagrange, France), apomorphine hydrochloride (Laboratoires Aguettant, France), α-methyl-*p*-tyrosine methyl ester hydrochloride (Sigma U.S.A.), and 6-hydroxydopamine hydrobromide (Sigma, U.S.A.) are expressed for the salts. The reserpine dose indicated (E. Merck, Darmstadt, West Germany) refers to the free base.

All controls were given acacia gum aqueous supension.

Animals. The animals used were male Swiss C.F. mice (25 ± 5 g) and male Wistar C.F. rats (200 ± 20 g), which were kept in a quiet room where the behavioral tests were performed. Room temperature was kept at 21 ± 1 °C, and artificial lighting was used (lights on between 7 a.m. and 7 p.m.). Food and water were given ad libitum.

Motility of Naive Mice. Motility experiments were performed between 9 a.m. and 1 p.m. when locomotor activity was relatively high. Animals were placed in activity cages (Boissier photoactimeter, Apelex, Bagneux, France). Each cage (L = 25.5 cm, W = 20 cm, H = 9 cm) was fitted with two photoelectric units with infrared lights located 1 cm above the floor of the cage in the middle of each side. The activity cages were placed in a closed compartment, which reduced the effects of external stimuli such as light or noise, and connected to a counter. Motility was expressed as the number of interruptions of photocell beams.

Mice were placed for 60 min in the photoactimeter, with one animal in each box. Drugs were administered 1 h before testing. Preliminary experiments were carried out on 6–12 mice and successive ones on 9–25.

Motility of Reserpine-Treated Mice. Tested drugs were administered 20 h after reserpine (10 mg/kg ip) was given to mice and 1 h before the animals (three/box) were placed in the photoactimeter. Motility was recorded for 60 min. Experiments were carried out on at least six mice.

Motility of Reserpine- and α-Methyl-*p*-tyrosine-Treated Mice. Mice were treated in the manner described in the above paragraph, with an additional injection of α-methyl-*p*-tyrosine methyl ester (250 mg/kg ip) 16 h after reserpine.

Motility of Reserpine- and Apomorphine-Treated Mice. Apomorphine (2 mg/kg ip) was injected 20 h after reserpine administration (10 mg/kg ip), as indicated by Elliott et al.,[13] benzamides having been administered 1 h before apomorphine. The motility of each mouse was recorded during 60 min following apomorphine injection.

Circling Behavior. Circling behavior was studied in unilaterally striatal 6-hydroxydopamine-lesioned mice.[21] Six days after lesioning, hypersensitivity to direct dopaminergic agonists was monitored by injection of apomorphine (2 mg/kg ip), which caused contraversive circling. This behavior, recorded for 30 min following acacia gum aqueous suspension or compound 11 administration, is expressed as the number of contraversive turns (m ± SEM). Animals (n = 7) served as their own controls.

HVA Levels in Rat Striatum and Limbic System. The effects of molecules on dopamine turnover in the striatum and limbic system of the rat were studied by determination of HVA concentrations. Brain samples were removed (5 h after drug administration) as described by Bartholini.[22] Extraction from tissue was done according to the method of Murphy et al.,[23] as modified by Pearson and Sharman,[24] and HVA was estimated fluorimetrically.[25]

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Registry No. **1**, 94843-50-2; **2**, 94843-51-3; **3**, 36845-09-7; **4**, 94843-52-4; **5**, 36845-10-0; **6**, 85367-13-1; **7**, 94843-53-5; **8**, 94843-54-6; **9**, 94843-55-7; **10**, 94843-56-8; **11**, 94843-57-9; **12**, 94843-58-0; **13**, 94843-59-1; **14**, 94843-60-4; **15**, 94843-61-5; **16**, 94843-62-6; **17**, 86439-15-8; **18**, 94843-63-7; **19**, 86425-35-6; **20**, 94843-64-8; **21**, 94843-65-9; **22**, 94843-66-0; **23**, 94843-67-1; **24**, 94843-68-2; **25**, 5407-88-5; **26**, 94843-69-3; **27**, 94843-70-6; **28**, 94843-71-7; **29**, 94843-72-8; **30**, 94843-73-9; **31**, 94843-74-0; **32**, 94843-75-1; **33**, 94843-76-2; 2-amino-4,6-dimethylpyridine, 5407-87-4; *m*-nitrobenzoyl chloride, 121-90-4; 4-fluorobenzoyl chloride, 403-43-0.

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Studies on Antianaphylactic Agents. 7.[1] Synthesis of Antiallergic 5-Oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridines[2]

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5-Oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carboxylic acids **23** and their tetrazole analogues **24** were synthesized from 4-oxo-4*H*-1-benzopyran-3-carbonitriles **3** or 2-amino-4-oxo-4*H*-1-benzopyran-3-carboxaldehydes **4**. When administered intravenously, they exhibited antiallergic activity in a reaginic PCA test in rats. In the carboxylic acid series, the activity was influenced by the substituents at the 2-position and increased substantially in the following order: Me, OMe < NH_2 < OH, H < NHOMe. On the other hand, in the tetrazole series, 2-unsubstituted derivatives showed the highest activity. Regardless of the kinds of substituents at positions 2 and 3, compounds bearing an alkyl group, especially an isopropyl group at the 7-position, were superior in activity to the corresponding unsubstituted compounds. Among these alkyl derivatives, 3-carboxylic acid derivatives, i.e., **23c** (7-ethyl), **23g** (2-amino-7-isopropyl), **23r** [2-(methoxyamino)-7-isopropyl], and a 3-tetrazole derivative **24c** (7-isopropyl), were 41–184 times as potent as disodium cromoglycate. They also exhibited remarkable activity when administered orally; clinical studies on **23g** (AA-673) are in progress.

The introduction of disodium cromoglycate (DSCG) (**1**) as the first prophylactic agent for treating bronchial asthma[3] prompted many laboratories, including our own, to search for orally active compounds.[4] Recently, we published a report on 3-(1*H*-tetrazol-5-yl)chromones **2**[5] that showed significant oral activity in the rat IgE-mediated

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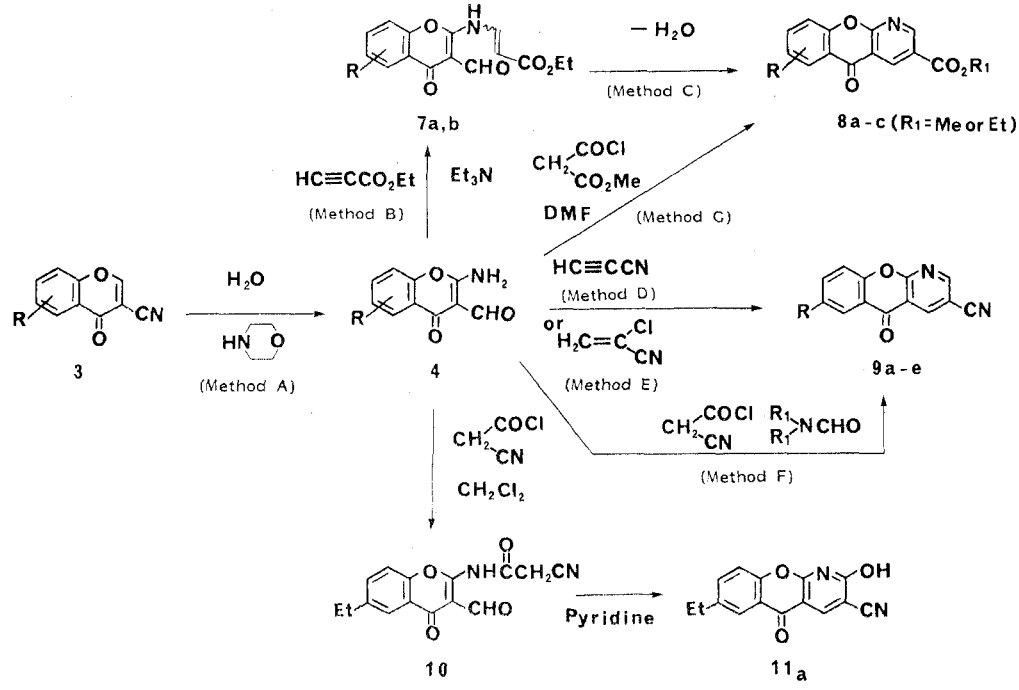
(1) Part 6: Nohara, A.; Kuriki, H.; Ishiguro, T.; Saijo, T.; Ukawa, K.; Maki, Y.; Sanno, Y. *J. Med. Chem.* **1979**, *22*, 290.
(2) Some parts of this paper have been published as a preliminary communication: Ishiguro, T; Ukawa, K.; Sugihara, H.; Nohara, A. *Heterocycles* **1981**, *16*, 733.

0022-2623/85/1828-0559$01.50/0

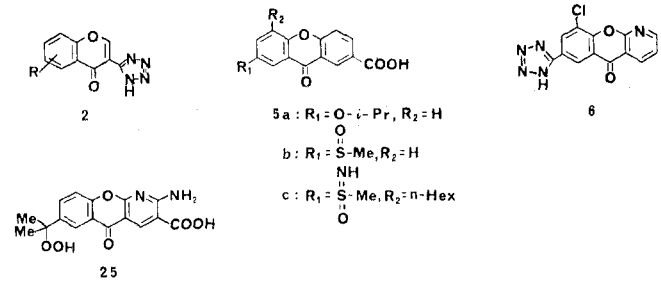
Scheme I



passive cutaneous anaphylaxis test (PCA), a possible model for the mode of action of DSCG. In the course of a synthetic study of **2**, we found, independently of Petersen et al.,[6] that 4-oxo-4*H*-1-benzopyran-3-carbonitriles **3** could be converted into 2-amino-4-oxo-4*H*-1-benzopyran-3-carboxaldehydes **4**.[2,7] Since the compounds **4** were thought to be valuable starting materials for heterocycles, we attempted to synthesize antiallergic agents from **4**.

Because xanthone-2-carboxylic acids (**5a–c**)[8–10] and compound **6**[11,12] carrying a tetrazole group on the benzene ring of the 5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine skelton possessed oral antiallergic activity, we attempted to synthesize compounds carrying a carboxyl or a tetrazole group on the pyridine ring of the skelton to ascertain whether or not they also possessed such activity. In this paper, we describe the synthesis and antiallergic activity of these compounds.

Reactions of carboxaldehydes **4** or carbonitriles **3** with some reactive methylene compounds afford 2,3-disubstituted-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine derivatives.[6,13] However, there is no report on the synthesis of 3-substituted-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridines that carry no substituent at the 2-position. Furthermore, in view of the structure–activity relations of 3-(1*H*-tetrazol-5-yl)chromones **2**,[5] 3-(4-oxo-4*H*-1-benzopyran-3-yl)-acrylic acids,[14] and xanthone-2-carboxylic acids **5**,[8] it seemed that 5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carboxylic acids and 3-tetrazole derivatives without a substituent at the 2-position would be the most desirable for high antiallergic activity. For these reasons, we initially investigated the synthesis of these compounds and then of their 2-substituted derivatives.



Chemistry. I. Synthesis of 2-Unsubstituted-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridines Carrying Alkoxycarbonyl, Cyano, and Formyl Groups at the 3-Position. The starting materials, 2-amino-4-oxo-4*H*-1-benzopyran-3-carboxaldehydes **4**, were synthesized from 4-oxo-4*H*-1-benzopyran-3-carbonitriles **3**[5] by a method modified from one in ref 6, i.e., by heating **3** in the presence of morpholine in DMF–H_2O at 60 °C for 2 h (method A, Scheme I) (Table I).

The reaction of **4** with ethyl propiolate in the presence of triethylamine in DMF initially afforded aminoacrylates **7** (method B), which were converted to ethyl 5-oxo-5*H*-

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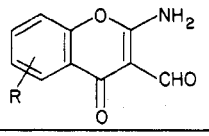
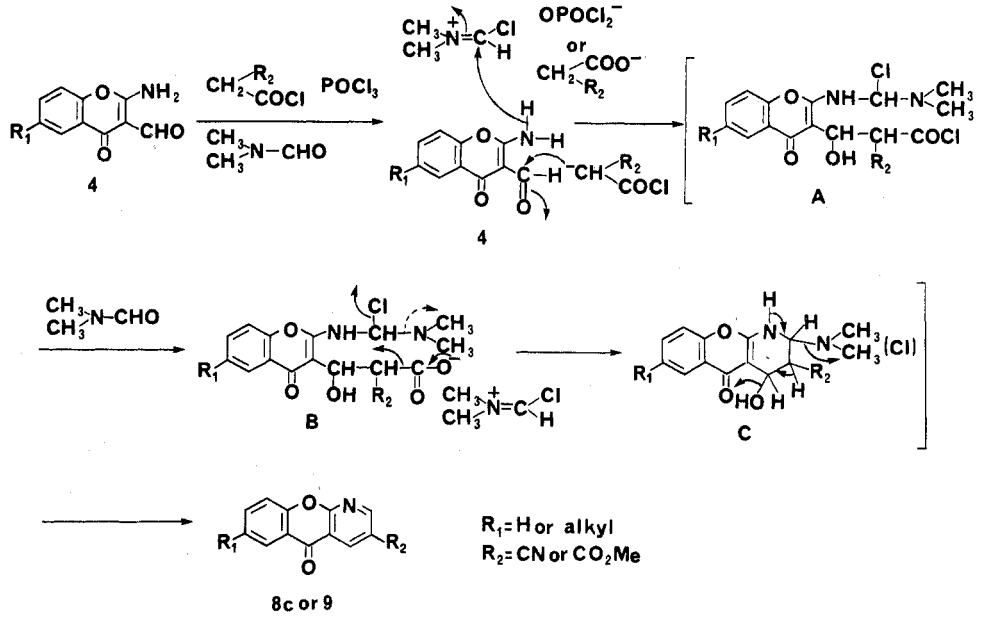
Table I. 2-Amino-4-oxo-4*H*-1-benzopyran-3-carboxaldehydes **4**



compd	R	mp, °C	recrystn solvent	yield, %	formula[a]
4a[b]	H	252–255 dec	AcOH	70	$C_{10}H_7NO_3$
4b	6-Me	282–284 dec	AcOH	60	$C_{11}H_9NO_3$
4c	6-Et	246–249 dec	AcOH	74	$C_{12}H_{11}NO_3$
4d	6-*i*-Pr	206–208	AcOH	65	$C_{13}H_{13}NO_3$
4e	6-*t*-Bu	240–242	AcOH	64	$C_{14}H_{15}NO_3$
4f	6,7-Me_2	259–263 dec	AcOH	61	$C_{12}H_{11}NO_3$
4g	6-MeO	251–254 dec	$CHCl_3$	68	$C_{11}H_9NO_4$
4h	8-MeO	235–238	$CHCl_3$	68	$C_{11}H_9NO_4$
4i	5,6-benzo	258–260	AcOH	62	$C_{13}H_9NO_3$

[a] All compounds were analyzed for C, H, and N; the analytical results obtained for these elements were within ±0.4% of the theoretical value. [b] Lit.[6] mp 248–250 °C dec (EtOH).

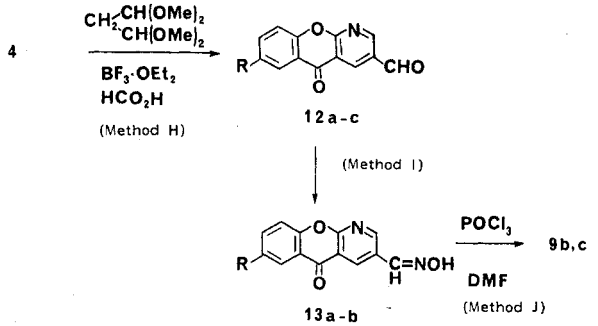
Scheme II



[1]benzopyrano[2,3-*b*]pyridine-3-carboxylates **8** by further heating (method C). Naturally, it was possible to obtain **8** directly from **4** without isolating and purifying the intermediate **7**. In the reaction of **4** with cyanoacetylene, a basic catalyst, such as triethylamine, was not necessary, and 3-cyano derivatives **9** were prepared in one step (method D). However, because cyanoacetylene has several undesirable properties, i.e., sublimation at low temperature, instability, pungent odor, etc., a substitute was needed. α-Chloroacrylonitrile could react with **4c** (R = 6-Et) in the presence of triethylamine to afford the desired **9b** (method E).

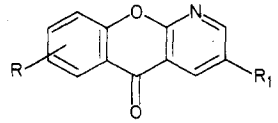
As mentioned previously, 2,3-disubstituted-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine derivatives have been synthesized from **4** and reactive methylene compounds.[6] However, when **4c** was treated in DMF with cyanoacetyl chloride generated in situ by the reaction of cyanoacetic acid with PCl_5 in dichloromethane, the expected cyanoacetamide intermediate **10** or 2-hydroxy-3-cyanobenzopyranopyridine derivative **11a** was not isolated, but 2-unsubstituted-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carbonitrile (**9b**) was (method F). Similarly, the reaction of **4c** in DMF with methyl malonyl chloride generated in situ, as noted above, gave 2-unsubstituted 3-carboxylate **8c** (method G). On the other hand, the reaction of **4c** in dichloromethane instead of DMF with cyanoacetyl chloride afforded only the amide **10**, which was converted to **11a** by heating in pyridine. Also, when the reaction was carried out in formamide, **9b** was not obtained. The fact that 2-unsubstituted-benzopyranopyridine derivatives could be formed only in DMF suggested the participation of Vilsmeier reagent arising from acid chloride and DMF. When *N*,*N*-diethylformamide or *N*-formylmorpholine was used instead of DMF, **9b** was obtained from **4c** likewise. A plausible mechanism to account for the above results is shown in Scheme II.

Scheme III



The condensation of **4** with malonaldehyde was examined next. The reaction of **4d** (R = 6-*i*-Pr) with malonaldehyde bis(dimethyl acetal) in the presence of formic acid in boron trifluoride etherate gave the desired 7-iso-

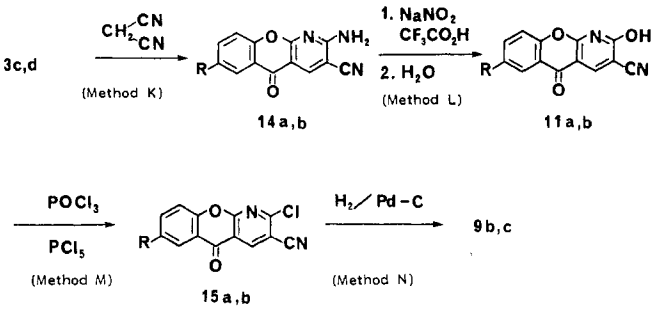
Table II. 3-Substituted-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridines **8**, **9**, **12**, and **13**



compd	R	R_1	mp, °C	recrystn solvent[a]	yield, %	method[b]	formula[c]
8a	H	CO_2Et	139–140	A	49	C	$C_{15}H_{11}NO_4$
8b	7-Et	CO_2Et	140–142	B	62	C	$C_{17}H_{15}NO_4$
8c	7-Et	CO_2Me	156–157	A	36	G	$C_{16}H_{13}NO_4$
9a	H	CN	220–226	B	32	D	$C_{13}H_6N_2O_2$
					39	F	
9b	7-Et	CN	183–185	C	28	D	$C_{15}H_{10}N_2O_2$
					33	E	
					49	F	
					84[d]	J	
					76	N	
9c	7-*i*-Pr	CN	230–231	D	36	D	$C_{16}H_{12}N_2O_2$
					35	E	
					52	F	
					87	J	
					72	N	
9d	7-*t*-Bu	CN	247–249	C	38	D	$C_{17}H_{14}N_2O_2$
9e	7,9-Me_2	CN	254–257	C	47	D	$C_{15}H_{10}N_2O_2$
12a	H	CHO	220–222	C	10	H	$C_{13}H_7NO_3$
12b	7-Et	CHO	175–178	C	31	H	$C_{15}H_{11}NO_3$
12c	7-*i*-Pr	CHO	211–213	E	61	H	$C_{16}H_{13}NO_3$
13a	7-Et	CH=NOH	250–252	B	93	I	$C_{15}H_{12}N_2O_3$
13b	7-*i*-Pr	CH=NOH	247–249	D	95	I	$C_{16}H_{14}N_2O_3$

[a] A = MeOH, B = EtOH, C = MeCN, D = $CHCl_3$–EtOH, E = AcOEt. [b] Method C: cyclization of **7**. Method D: reaction of **4** with cyanoacetylene. Method E: reaction of **4** with α-chloroacrylonitrile. Method F: reaction of **4** with cyanoacetyl chloride. Method G: reaction of **4** with methyl malonyl chloride. Method H: reaction of **4** with malonaldehyde bis(dimethyl acetal). Method I: reaction of **12** with hydroxylamine. Method J: reaction of **13** with $POCl_3$. Method N: catalytic hydrogenation of **15**. [c] All compounds were analyzed for C, H, and N; analytica results obtained for these elements were within ±0.4% of the theoretical value. [d] Overall yield from **12b**.

Scheme IV



propyl-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carboxaldehyde (**12c**) (method H, Scheme III). The oxime **13** prepared from **12** was treated with $POCl_3$ in DMF at room temperature to give the nitrile **9** (method J) (Table II).

Another synthetic route to **9** was also investigated. Compound **3** was converted to 2-amino-5-oxo-5*H*-[1]-benzopyrano[2,3-*b*]pyridine-3-carbonitriles **14** (method K,[13] Scheme IV) which upon deamination by sodium nitrite in trifluoroacetic acid afforded **11** (method L). Chlorination of **11** with $POCl_3$–PCl_5 (method M) followed by catalytic hydrogenation over 5% Pd–C in the presence of K_2CO_3 in DMF gave **9** (method N).

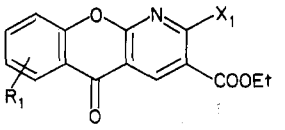
II. Synthesis of 2-Substituted-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridines Carrying Alkoxycarbonyl or Cyano Groups at the 3-Position. 2-Substituted-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carboxylates (**16–18**, **21**; Table III) were synthesized by the route shown in Scheme V. The esters **16a** (X_1 = Me) and **16b–f** (X_1 = NH_2) were prepared from **3** (method K) or **4** (method O) by the method of ref 13 or 6. The ester **16g** (X_1 = COOEt, R_1 = 7-Et) was synthesized by the reaction of **4c** (R_1 = 6-Et) with diethyl acetylenedicarboxylate (method P). Although the esters **17** were obtained by the reaction of **4** with diethyl malonate in low yield,[6] they could also be prepared by deamination of **16** (X_1 = NH_2) with sodium nitrite in aqueous acetic acid in good yield (method L). The 2-methoxy derivative **18** was prepared by methylation of **17c** (R_1 = 7-Et) with diazomethane; the *N*-methyl derivative **19** was also obtained as a minor product (method Q). The 2-alkoxyamino derivatives **21** were prepared by chlorination of **17** with $POCl_3$–PCl_5, followed by alkoxyamination of the resulting chloro derivatives **20** (method R).

III. Synthesis of 5-Oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carboxylic Acids and Their Tetrazole Derivatives. The route for synthesizing 5-oxo-5*H*-[1]-benzopyano[2,3-*b*]pyridine-3-carboxylic acids **23** and their tetrazole analogues **24** is shown in Scheme VI. Generally, the esters **16–18** and **21** were hydrolyzed to **23** under acidic conditions (50% H_2SO_4–AcOH) (method T) to avoid the ring opening (e.g., formation of a pyridone derivative (**22**) that occurred under basic conditions). The acids **23b–d** (X_1 = H) were prepared either by hydrolysis of **8** (X_1 = H) or by decarboxylation of the diacid **23v** (X_1 = COOH), which was synthesized from the diester **16g** (X_1 = COOEt). Though the acids **23f,g** (X_1 = NH_2) could be prepared in one step by the condensation–ring closure reaction of carboxaldehyde **4** with cyano acetic acid in pyridine (method S), the route from the esters **16** was preferred. Compounds **24** were synthesized by reaction of nitriles **9** with sodium azide in the presence of ammonium chloride in DMF (method U), but with aluminum chloride–sodium azide in tetrahydrofuran no reaction occurred.

Biological Results and Discussion

The biological activities were measured by the rat PCA test and the results are shown in Tables IV and V. The 5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine ring was found to be essential for activity; when the ring structure of **23a** was destroyed, the activity disappeared, as shown by the result with the pyridone derivative **22**. Initially, the effect

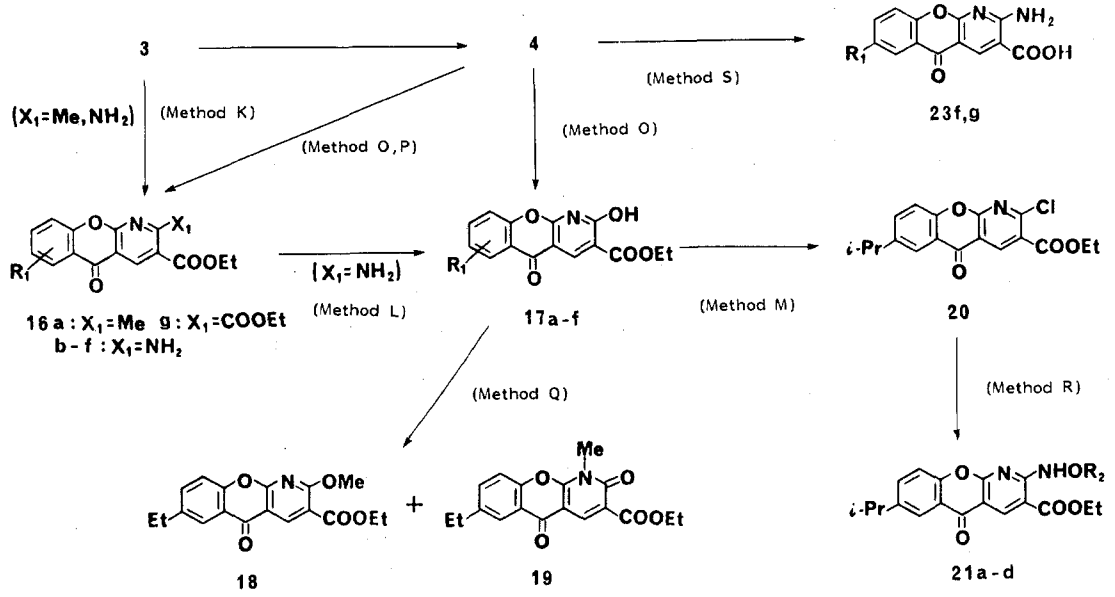
Table III. Ethyl 5-Oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carboxylates



compd	R_1	X_1	mp, °C	recrystn solvent[a]	yield, %	method[b]	formula[c]
16a	7-Et	Me	149–151	A	51	O	$C_{18}N_{17}NO_4$
16b	7-Et	NH_2	279–280	C	66	O	$C_{17}H_{16}N_2O_4$
16c	7-*i*-Pr	NH_2	243–244	A	69	O	$C_{18}H_{18}N_2O_4$
					89	K	
16d	7-*n*-Bu	NH_2	234.5–235	A	74	O	$C_{19}H_{20}N_2O_4$
16e	9-OMe	NH_2	>300	A	74	O	$C_{16}H_{14}N_2O_5$
16f	7-OMe	NH_2	285–286	B	73	O	$C_{16}H_{14}N_2O_5$
16g	7-Et	COOEt	112–113	A	91	P	$C_{20}H_{19}NO_6$
17a[d]	H	OH	245–248	C	92	L	$C_{15}H_{11}NO_5$
					26	O	
17b	7-Me	OH	221–222	A	92	L	$C_{16}H_{13}NO_5$
17c	7-Et	OH	204–206	A	96	L	$C_{17}H_{15}NO_5$
					27	O	
17d	7-*i*-Pr	OH	158–159	A	79	L	$C_{18}H_{17}NO_5$
17e	9-OMe	OH	252–254	C	90	L	$C_{16}H_{13}NO_6$
17f	7-OMe	OH	269–270	B	75	L	$C_{16}H_{13}NO_6$
18	7-Et	OMe	145–146	D	59	Q	$C_{18}H_{17}NO_3$
20	7-*i*-Pr	Cl	111–112	E	58	M	$C_{18}H_{16}ClNO_4$
21a	7-*i*-Pr	NHOMe	208–209	F	79	R	$C_{19}H_{20}N_2O_5$
21b	7-*i*-Pr	NHOEt	182–184	G	24	R	$C_{20}H_{22}N_2O_5$
21c	7-*i*-Pr	NHO-*n*-Bu	134–135	G	36	R	$C_{22}H_{26}N_2O_5$
21d	7-*i*-Pr	NHOH	254–255	H	31	R	$C_{18}H_{18}N_2O_5$

[a] A = EtOH, B = $CHCl_3$–EtOH, C = DMF, D = Me_2CO, E = *i*-Pr_2O, F = $CHCl_3$–MeCN, G = MeCN, H = EtOH–H_2O. [b] Method K: reaction of **3d** with ethyl cyanoacetate. Method L: deamination of **16b–f**. Method M: reaction of **17** with $POCl_3$–PCl_5. Method O: reaction of **4** with ethyl acetoacetate, ethyl cyanoacetate, or diethyl malonate. Method P: reaction of **4c** with diethyl acetylenedicarboxylate. Method Q: reaction of **17c** with diazomethane. Method R: reaction of **22** with hydroxylamine or *O*-alkylhydroxylamine. [c] All compounds were analyzed for C, H, and N; the analytical results obtained for these elements were within ±0.4% of the theoretical value. [d] Lit.[6] mp 239–242 °C (DMF).

Scheme V



on the antiallergic activity of the substituents at position 2 of the 5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridinecarboxylic acids and their tetrazole analogues was examined. On the basis of the analogy of structure–activity studies of **2**[5] and 3-(4-oxo-4*H*-1-benzopyran-3-yl)acrylic acid,[14] 5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carboxylic acid derivatives **23b–d** carrying no substituent at the 2-position were expected to be the most desirable compounds for high activity. This presumption was found to be true: the 2-unsubstituted derivative **23c** was 21, 4, and 1.2 times as potent as the corresponding 2-substituted derivatives **23a** (Me), **23f** (NH_2), and **23l** (OH), respectively. The respective substitution of the hydrogen, hydroxyl, and amino group at the 2-position with an alkyl, alkyloxy, and alkylamino group resulted in a decrease in activity as shown by the comparison of **23a** to **23c**, **23l** to **23p**, and **23g** to **23q**. Unexpectedly, however, when the 2-amino group was replaced with an alkoxyamino or hydroxyamino group, the activity was enhanced remarkably. The methoxyamino and hydroxyamino derivatives **23r** and **23u**, respectively, were 4.2 and 1.8 times as potent as the amino derivative **23g**, respectively. Elongation of the alkoxy chain in the methoxyamino group also resulted in a decrease of activity as shown by the comparison of **23r** to **23s** or **23t**. It fol-

Scheme VI

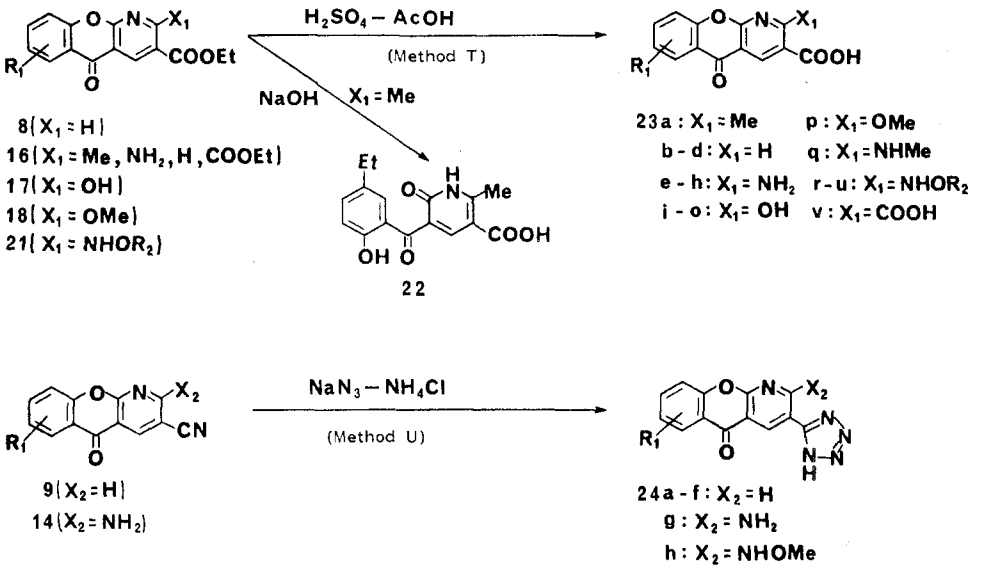
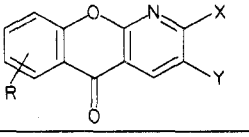


Table IV. Anti-PCA Activity of 5-Oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridines



compd	R	X	Y	mp, °C	recrystn solvent[a]	formula[b]	rat PCA:[c] ID_{50}, mg/kg, iv
23a	7-Et	Me	COOH	243–245	A	$C_{16}H_{13}NO_4$	0.73 (0.52–1.2)
23b	H	H	COOH	270	B	$C_{13}H_7NO_4$	0.63 (0.52–0.76)
23c	7-Et	H	COOH	238–239	B	$C_{15}H_{11}NO_4$	0.034 (0.029–0.040)
23d	7-*i*-Pr	H	COOH	259–261	C	$C_{16}H_{13}NO_4$	0.059 (0.016–0.13)
23e	9-OMe	NH_2	COOH	>300	D	$C_{14}H_{10}N_2O_5$	0.45 (0.25–0.80)
23f	7-Et	NH_2	COOH	>300	E	$C_{15}H_{12}N_2O_4$	0.14 (0.056–0.24)
23g	7-*i*-Pr	NH_2	COOH	>300	D	$C_{16}H_{14}N_2O_4$	0.032 (0.028–0.036)
23h	7-*n*-Bu	NH_2	COOH	303–304	D	$C_{17}H_{16}N_2O_4$	1.1 (0.68–10)
23i	7,9-Me_2	OH	COOH	>300	D	$C_{15}H_{11}NO_5$	0.12 (0.079–0.64)
23j	H	OH	COOH	>300	D	$C_{13}H_7NO_5$	0.62 (0.42–0.89)
23k	7-Me	OH	COOH	>300	D	$C_{14}H_9NO_5$	0.10 (0.079–0.12)
23l	7-Et	OH	COOH	303–305 dec	B	$C_{15}H_{11}NO_5$	0.041 (0.0054–0.086)
23m	7-*i*-Pr	OH	COOH	295–300 dec	C	$C_{16}H_{13}NO_5$	0.020 (0.015–0.026)
23n	9-OMe	OH	COOH	>300	D	$C_{14}H_9NO_6$	0.10 (0.077–0.17)
23o	7-OMe	OH	COOH	>300	D	$C_{14}H_9NO_6$	0.042 (0.021–0.067)
23p	7-Et	OMe	COOH	226–228	F	$C_{16}H_{13}NO_5$	0.50 (0.23–0.78)
23q	7-*i*-Pr	NHMe	COOH	>300	B	$C_{17}H_{16}N_2O_4$	0.39 (0.33–0.45)
23r	7-*i*-Pr	NHOMe	COOH	270–274 dec	H	$C_{17}H_{16}N_2O_5$	0.0076 (0.0064–0.0093)
23s	7-*i*-Pr	NHOEt	COOH	267–271 dec	H	$C_{18}H_{18}N_2O_5$	0.014 (0.012–0.015)
23t	7-*i*-Pr	NHOBu	COOH	244–247 dec	H	$C_{20}H_{22}N_2O_5$	0.028 (0.023–0.034)
23u	7-*i*-Pr	NHOH	COOH	237–239 dec	I	$C_{16}H_{14}N_2O_5$	0.018 (0.013–0.024)
24a	H	H	Te[d]	>300	D	$C_{13}H_7N_5O_2$	0.44 (0.35–0.57)
24b	7-Et	H	Te	262–265 dec	D	$C_{15}H_{11}N_5O_2$	0.11 (0.087–0.16)
24c	7-*i*-Pr	H	Te	275–277 dec	D	$C_{16}H_{13}N_5O_2$	0.0096 (0.0086–0.011)
24d	7-*t*-Bu	H	Te	273–275 dec	F	$C_{17}H_{15}N_5O_2$	0.047 (0.036–0.069)
24e	7,9-Me_2	H	Te	294–298 dec	D	$C_{15}H_{11}N_5O_2$	0.31 (0.056–0.54)
24f	6,7-benzo	H	Te	291–293 dec	D	$C_{17}H_9N_5O_2$ C_3H_7NO	0.84 (0.58–1.67)
24g	7-*i*-Pr	NH_2	Te	>300	D	$C_{16}H_{14}N_6O_2$	0.064 (0.0096–0.42)
24h	7-*i*-Pr	NHOMe	Te	295–298 dec	H	$C_{17}H_{16}N_6O_3$	0.19 (0.17–0.23)
22				236–238 dec	B	$C_{16}H_{15}NO_5$	>20
1							1.4 (1.3–1.6)

[a] A = Me_2CO, B = DMF–H_2O, C = EtOH, D = DMF, E = AcOH–H_2O, F = DMF–EtOH, G = MeOH–H_2O, H = AcOH, I = EtOH–H_2O, J = DMF–AcOEt. [b] All compounds were analyzed for C, H, and N; the analytical results obtained for these elements were within ±0.4% of the theoretical value. [c] ID_{50} = 50% inhibition dose. Numerals in parentheses are 95% confidence limits. [d] Te = 1*H*-tetrazol-5-yl.

lowed, as a consequence, that when the members of carboxylic acid series were administered intravenously, their activity increased substantially in the following order: Me, OMe < NH_2 < OH, H < NHOMe, and the methoxyamino derivative **23r** was the most potent compound.

On the other hand, when the members of the tetrazole series were administered intravenously, 2-unsubstituted derivatives **24a–f** showed, as expected, high antiallergic activity. However, in contrast with the result of the carboxylic acid series, the derivative carrying an NHOMe group at the 2-position (**24h**) showed low activity. In the tetrazole series, the activity increased in the following order: NHOMe < NH_2 < H, and 2-unsubstituted derivative **24c** was the most potent compound.

Subsequently, the effect on the activity of the substituents on the benzene ring of 5-oxo-5*H*-[1]benzopyrano-

Table V. Oral Anti-PCA Activity of 5-Oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridines

compd	ID_{50},[a,b] mg/kg	compd	ID_{50},[a,b] mg/kg
23c	0.95 (0.65–1.4)	**23s**	0.58 (0.42–0.82)[c]
	0.18 (0.13–0.26)[c]	**23t**	3.5 (2.5–4.6)
23d	1.2 (0.83–1.55)	**23u**	1.0 (0.64–1.7)
23f	5.4 (3.2–10.0)	**24b**	6.4 (2.7–21.4)
23g	6.1 (4.8–7.8)	**24c**	0.46 (0.29–0.77)
	0.73 (0.38–1.15)[c]		0.034 (0.021–0.051)[c]
23r	4.6 (4.1–5.2)	**24d**	6.0 (3.1–39)
	0.59 (0.44–0.79)[c]		

[a] See Table IV, footnote *c*. [b] Nonfasted rats were used except where indicated by superscript *c*. [c] Fasted rats were used.

[2,3-*b*]pyridine skelton was examined. The kinds and positions of substituent(s) were selected on the basis of the structure–activity relations study of chromone derivatives **2**[5] and from the synthetic viewpoint. Regardless of the kinds of substituent at positions 2 and 3, compounds bearing an alkyl group, especially the isopropyl group at the 7-position, were superior to the corresponding unsubstituted compounds in activity as shown by the comparison of **23b** to **23d**, **23j** to **23m**, **24a** to **24c**; the isopropyl derivatives were 11, 31, and 46 times as potent as the corresponding unsubstituted compounds. Whereas an alkyl or an ether group at the 7-position enhanced activity, a 6,7-benzo group lowered the activity, in contrast with the result from the chromone derivatives. All compounds synthesized herein were highly potent inhibitors of the PCA reaction; in particular, alkyl derivatives **23c**, **23g**, **23r**, and **24c** were 41–184 times as potent as DSCG and 10–46 times as potent as **2** (R = 6-Et).

The ID_{50} values of the representative compounds administered orally are shown in Table V. In all cases, the tested compounds were absorbed better in fasted than in nonfasted rats; 5, 8, 8, and 14 times better absorptions were obtained in the fasted rats for **23c**, **23g**, **23r**, and **24c**, respectively.

In the course of the study, it was found that **23g** prevented the contraction of guinea pig ileum induced by the slow-reacting substance of anaphylaxis (SRS-A) (IC_{50} = 4.6×10^{-5} M). This result was in a marked contrast to the recent report[15] that the structurally related 2-isopropylpyrido[2,1-*b*]quinazoline-8-carboxylic acid was inactive, but the corresponding 8-(diethylamino)ethyl ester showed antagonistic activity against SRS-A. In addition to this antagonistic activity, **23g** inhibited SRS-A generation (IC_{50} = 1.7×10^{-5} M) from the lung fragments of the actively sensitized guinea pigs. When an alkaline solution of **23g** was aerated under irradiation with a mercury lamp, a 7-(1-hydroperoxy-1-methylethyl) derivative (**25**)[16] was detected. This fact suggests that the 7-isopropyl group may play a role in scavenging peroxy radicals and contribute to inhibiting generation of SRS-A.

After the pharmacological and toxicological properties of **23c**, **23g**, **23r**, and **24c** were examined, **23g** (AA-673) was selected as the most promising compound, and clinical studies on it are in progress.

Experimental Section

All melting points were determined on a micro melting point apparatus (Yanagimoto) and are uncorrected. The following instruments were used to obtain physical data: NMR spectra, a Varian T-60 spectrometer; IR spectra, a Hitachi 215 grating infrared spectrophotometer; mass spectra, a Hitachi RMU-6D mass spectrometer and a JMS-01SC mass spectrometer (Japan Electron Optics Co.). Spectral data were in accord with the assigned structure. Analytical results obtained for elements were within ±0.4% of theoretical values except where otherwise indicated.

2-Amino-6-isopropyl-4-oxo-4*H*-1-benzopyran-3-carboxaldehyde (4d) (Method A). 6-Isopropyl-4-oxo-4*H*-1-benzopyran-3-carbonitrile[5] (**3d**; 533 g, 2.5 mol) was added to a mixture of morpholine (540 mL), DMF (850 mL), and water (2.8 L) over a period of 15 min with stirring at 60 °C, and the mixture was stirred for 2 h at the same temperature. After the reaction mixture had been cooled with ice, the separated crystals were collected by filtration, washed with water, and dried in vacuo. The crystals were recrystallized from DMF and washed with EtOH to give 391 g (67.6%) of yellow crystals: mp 198–204 °C. The analytical sample was obtained by recrystallization from AcOH to afford pale yellow crystals: mp 206–208 °C. Anal. ($C_{13}H_{13}NO_3$) C, H, N.

Ethyl 3-[(3-Formyl-4-oxo-4*H*-1-benzopyran-2-yl)amino]acrylate (7a) (Method B). A mixture of 2-amino-4-oxo-4*H*-1-benzopyran-3-carboxaldehyde (**4a**; 5.5 g, 30 mmol), ethyl propiolate (5 g, 52 mmol), and Et_3N (one drop) in DMF (25 mL) was stirred for 1 h at 90 °C and concentrated in vacuo. MeOH was added to the residue, and an insoluble substance was collected by filtration, washed with MeOH, and recrystallized from MeOH to afford 3.5 g (39%) of colorless needles: mp 201–203 °C. Anal. ($C_{15}H_{13}NO_5$) C, H, N.

Ethyl 3-[(6-Ethyl-3-formyl-4-oxo-4*H*-1-benzopyran-2-yl)amino]acrylate (7b). Compounds **7b** was prepared by method B: mp 201–203 °C (MeOH), 52% yield. Anal. ($C_{17}H_{17}NO_5$) C, H, N.

Ethyl 5-Oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carboxylate (8a) (Method C). A mixture of **7a** (3.2 g, 11 mmol), toluene (20 mL), and Et_3N (5 mL) was refluxed for 24 h. The reaction mixture was concentrated in vacuo, and the residue was recrystallized from MeOH to afford 1.42 g (49%) of pale yellow crystals: mp 139–140 °C. Anal. ($C_{15}H_{11}NO_4$) C, H, N.

7-Isopropyl-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carbonitrile (9c) (Method D). A mixture of **4d** (2.31 g) and cyanoacetylene (2.04 g) in DMF (60 mL) was stirred for 1 h at 100 °C and then for 10 h at 140 °C. After the solvent was removed in vacuo, the residue was chromatographed on a column of silica gel (150 g) and eluted with $CHCl_3$. The desired fraction was concentrated and recrystallized from EtOH to afford 0.955 g (36%) of colorless needles: mp 228–229 °C. Anal. ($C_{16}H_{12}N_2O_2$) C, H, N.

7-Ethyl-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carbonitrile (9b) (Method E). α-Chloroacrylonitrile (1.31 g) and Et_3N (1.6 g) were added to a solution of **4c** (0.98 g) in DMF (50 mL), and the reaction mixture was heated at 120 °C for 14 h. After the solvent was removed in vacuo, the residue was chromatographed on a column of silica gel (60 g) and eluted with $CHCl_3$. The eluate was concentrated in vacuo and the residue was recrystallized from CH_3CN to give colorless needles (0.41 g, 33%) of **9b**: mp 183–185 °C. Anal. ($C_{15}H_{10}N_2O_2$) C, H, N.

7-Ethyl-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carbonitrile (9b) (Method F). A mixture of **4c** (1.82 g) and cyanoacetyl chloride (3.5 g) in DMF (40 mL) was stirred for 3 h at 60 °C. After the solvent was evaporated, the residue was chromatographed on a column of silica gel (100 g) and eluted with $CHCl_3$. The eluate was concentrated in vacuo and the residue was recrystallized from CH_3CN to give 1.03 g (49%) of **9b**. Its NMR spectrum was identical with that of **9b** described above.

Methyl 7-Ethyl-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carboxylate (8c) (Method G). PCl_5 (0.312 g) was added to a solution of malonic acid monomethyl ester (0.177 g) in CH_2Cl_2 (7 mL) and the mixture was refluxed for 0.5 h and then cooled. The reaction mixture was added to a solution of **4c** (0.217 g) in DMF (10 mL) with stirring at room temperature. After the mixture had been stirred for 0.5 h at room temperature, it was heated for 1.5 h at 65 °C and the solvent was evaporated in vacuo. The residue was chromatographed on a column of silica gel (20 g) and eluted with $CHCl_3$. The eluate was concentrated and the residue was recrystallized from MeOH to afford 0.103 g (36%) of **8c**: mp 156–157 °C. Anal. ($C_{16}H_{13}NO_4$) C, H, N.

2-[(Cyanoacetyl)amino]-6-ethyl-4-oxo-4*H*-1-benzopyran-3-carboxaldehyde (10). A mixture of cyanoacetic acid (1.27 g)

(15) Tilley, J. W.; Levitan, P.; Welton, A. F.; Crowley, H. J. *J. Med. Chem.* **1983**, *26*, 1638.
(16) Unpublished data.

and PCl_5 (3.12 g) in CH_2Cl_2 (25 mL) was refluxed for 0.5 h, and **4c** (2.12 g) was added to the solution. The mixture was refluxed for 2 h. The separated crystals were collected by filtration and recrystallized from AcOH to give 1.05 g (40%) of **10**: mp 170–175 °C dec; mass spectrum, *m/e* 284 (M^+). Anal. ($C_{15}H_{12}N_2O_4$) C, H, N.

7-Ethyl-2-hydroxy-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carbonitrile (11a). A solution of **10** (0.10 g) in pyridine (3 mL) was heated at 80 °C for 2 h and pyridine was evaporated in vacuo. The residue was recrystallized from AcOH to give 51 mg (55%) of **11a**: mp >300 °C. Anal. ($C_{15}H_{10}N_2O_3$) C, H, N.

7-Isopropyl-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carboxaldehyde (12c) (Method H). A mixture of **4d** (500 mg, 2.16 mmol), HCO_2H (0.3 mL), malonaldehyde bis(dimethyl acetal) (0.5 mL), and $BF_3{\cdot}OEt_2$ (2 mL) was stirred at 60 °C for 2 h. The reaction mixture was poured into H_2O (100 mL) and the precipitate was collected by filtration and recrystallized from AcOEt to afford 350 mg (60.5%) of **12c**: mp 211–213 °C. Anal. ($C_{16}H_{13}NO_3$) C, H, N.

7-Isopropyl-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carboxaldehyde Oxime (13b) (Method I). A mixture of **12c** (367 mg, 1 mmol) and $NH_2OH{\cdot}HCl$ (80 mg, 1.12 mmol) in EtOH (4 mL) was refluxed for 0.5 h. After the reaction mixture had been cooled, the separated crystals were collected by filtration to give 267 mg (94.7%) of colorless scales: mp 241–243 °C. A part of these crystals (68 mg) was dissolved in $CHCl_3$, chromatographed on a column of silica gel (10 g), and eluted with $CHCl_3$–Me_2CO–HCO_2H (20:1:0.1). The eluate was concentrated and the residue was recrystallized from $CHCl_3$–EtOH to afford 25 mg of colorless crystals: mp 247–249 °C. Anal. ($C_{16}H_{14}N_2O_3$) C, H, N.

7-Isopropyl-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carbonitrile (9c) (Method J). $POCl_3$ (0.5 mL) was added to a solution of **13b** (150 mg, 0.53 mmol) in DMF (5 mL) and the mixture was stirred for 0.5 h at room temperature. The reaction mixture was poured into H_2O, and the precipitated crystals were collected by filtration and recrystallized from $CHCl_3$–EtOH to afford 121 mg (86.5%) of colorless scales: mp 228–230 °C. Some of the material was chromatographed on a column of silica gel, eluted with $CHCl_3$–Me_2CO–HCO_2H (20:1:0.1), and recrystallized from $CHCl_3$–EtOH to give colorless scales: mp 230–231 °C. Anal. ($C_{16}H_{12}N_2O_2$) C, H, N.

2-Amino-7-isopropyl-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carbonitrile (14b) (Method K). A mixture of **3d** (50 g, 0.235 mol), malononitrile (31 g, 0.47 mol), piperidine (25 mL, 0.253 mol), and EtOH (1 L) was refluxed for 3 h and cooled. The separated solid was collected by filtration, washed with EtOH, and recrystallized from DMF to afford 57.2 g (89%) of pale yellow needles: mp >300 °C. Anal. ($C_{16}H_{13}N_3O_2$) C, H, N. **14a** (R = Et) was also prepared by method K: mp >300 °C (DMF), 92% yield. Anal. ($C_{15}H_{11}N_3O_2$) C, H, N.

2-Hydroxy-7-isopropyl-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carbonitrile (11b) (Method L). $NaNO_2$ (17.1 g) was added to an ice-cooled solution of **14b** (57.2 g) in CF_3CO_2H (333 mL) over a period of 1.5 h with stirring. After an additional 0.5 h of stirring, the reaction mixture was poured into ice–water (3 L). A precipitate was collected by filtration and washed with H_2O. A 5% aqueous Na_2CO_3 solution (1 L), triethanolamine (33 g), H_2O (3.5 L), and AcOEt (1 L) were added to a suspension of the precipitate in H_2O (0.5 L), and the mixture was stirred. An insoluble material was filtered off and the separated aqueous layer was extracted with two 1-L portions of AcOEt. The aqueous layer was acidified with 10% HCl and allowed to stand for 3 days at room temperature. The precipitate was collected by filtration, washed with H_2O, and dried in vacuo to give 47.1 g (82%) of **11b**, mp >300 °C, which was recrystallized from DMF–EtOH to afford colorless prisms: mp >300 °C. Anal. ($C_{16}H_{12}N_2O_3$) C, H, N. **11a** (R = Et) was prepared by method L: mp >300 °C (DMF–H_2O), 60% yield.

2-Chloro-7-isopropyl-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carbonitrile (15b) (Method M). A mixture of **11b** (41 g), $POCl_3$ (300 mL), and PCl_5 (45 g) was stirred for 2 h at 120 °C. Additional PCl_5 (5 g) was added and the mixture was stirred for a further 3-h period. After the reaction mixture had been cooled, the separated crystals were collected by filtration, washed with isopropyl ether, and dried in vacuo to give 32.8 g (75%) of crystals: mp 242–243 °C. Anal. ($C_{16}H_{11}ClN_2O_2$) C, H, N. **15a** (R = Et) was prepared by method M: mp 233–234 °C (AcOEt), 76% yield. Anal. ($C_{15}H_9ClN_2O_2$) C, H, N.

7-Isopropyl-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carbonitrile (9c) (Method N). A mixture of **15b** (1.72 g), anhydrous K_2CO_3 (0.795 g), and 5% Pd–C (200 mg) in DMF (50 mL) was hydrogenated under atmospheric pressure at room temperature over a 2.5-h period. After the reaction was terminated, DMF was evaporated. $CHCl_3$ (100 mL) and H_2O (50 mL) were added to the residue, and the insoluble material was filtered off. The $CHCl_3$ layer was separated, washed with 5% aqueous Na_2CO_3 (40 mL × 3) and twice with H_2O (40 mL × 2), dried over anhydrous Na_2SO_4, and evaporated in vacuo. The residue was recrystallized from DMF–EtOH to afford 1.09 g (72%) of **9c**: mp 228–229 °C.

Ethyl 7-Ethyl-2-methyl-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carboxylate (16a) (Method K). A mixture of **4c** (2.17 g), ethyl acetoacetate (4.0 mL), EtOH (50 mL), and piperidine (5 mL) was refluxed for 2 h with stirring. After the reaction mixture had been cooled, the precipitate was collected by filtration, washed with EtOH, and recrystallized from EtOH (charcoal) to give 1.60 g (51%) of yellow neeldes: mp 149–151 °C. Anal. ($C_{18}H_{17}NO_4$) C, H, N.

Ethyl 2-Amino-7-isopropyl-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carboxylate (16c) (Method K). A mixture of **3d** (250 g), EtOH (3.5 L), piperidine (117 mL), and ethyl cyanoacetate (173 g) was refluxed for 5 h. After the reaction mixture was allowed to stand overnight at room temperature, the separated crystals were collected by filtration, washed with EtOH (800 mL), and recrystallized from EtOH to afford 363.8 g (95%) of colorless needles: mp 243–244 °C. Anal. ($C_{18}H_{18}N_2O_4$) C, H, N.

Ethyl 2-Amino-7-ethyl-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carboxylate (16b) (Method O). A mixture of **4c** (217 mg), ethyl cyanoacetate (0.6 mL), piperidine (0.3 mL), and EtOH (10 mL) was refluxed for 3 h. After the reaction mixture had been cooled to room temperature, the separated crystals were collected by filtration and recrystallized from DMF to give 206 mg (66%) or colorless needles: mp 278–279 °C. Anal. ($C_{17}H_{16}N_2O_4$) C, H, N.

Diethyl 7-Ethyl-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-2,3-dicarboxylate (16g) (Method P). A mixture of **4c** (2.17 g), diethyl acetylenedicarboxylate (2.55 g), EtOH (50 mL), and Et_3N (0.5 mL) was refluxed for 2 h. While the reaction mixture was hot, an insoluble material was filtered off and the filtrate was cooled. The separated crystals were collected by filtration and washed with EtOH to afford 3.35 g (91%) of colorless needles: mp 113–114 °C. Anal. ($C_{20}H_{19}NO_6$) C, H, N.

Ethyl 7-Ethyl-2-hydroxy-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carboxylate (17c). A. Method O. A mixture of **4c** (3.17 g), EtOH (50 mL), piperidine (15 mL), diethyl malonate (8 g), and 1,5-diazabicyclo[3.4.0]non-5-ene (1 mL) was refluxed for 15 h. The reaction mixture was concentrated and the residue was acidified with HCl. The precipitated material was collected by filtration, washed with H_2O, and recrystallized from EtOH to afford 1.23 g (27%) of pale yellow needles: mp 200–204 °C. Anal. ($C_{17}H_{15}NO_5$) C, H, N.

B. Method L. $NaNO_2$ (4.0 g) was added to a suspension of **16b** (2.5 g, 8.0 mmol) in 90% AcOH (250 mL) over a period of 30 min at 75 °C with stirring. After an additional 2-h period of stirring at 75 °C, the reaction mixture was concentrated. H_2O (150 mL) was added to the residue and the mixture was stirred for 30 min at 80 °C and cooled with ice–water. The separated crystals were collected by filtration, washed with H_2O, and recrystallized from EtOH to afford 2.4 g (96%) of colorless needles: mp 204–206 °C.

Ethyl 7-Ethyl-2-methoxy-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carboxylate (18) and Ethyl 7-Ethyl-1-methyl-2,5-dioxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carboxylate (19) (Method Q). An excess of diazomethane ethereal solution was added dropwise to a solution of **17c** (1.0 g, 3.2 mmol) in $CHCl_3$ (20 mL) over a period of 15 min at room temperature and stirred for 45 min. A small amount of AcOH was added to the reaction mixture in order to destroy diazomethane and the mixture was concentrated in vacuo. The residue was chromatographed on a

column of silica gel (100 g) and eluted with $CHCl_3$–Me_2CO–HCO_2H (20:1:0.1). The compound eluted first was recrystallized from Me_2CO to afford 615 mg (59%) of **18** as colorless needles: mp 145–146 °C. Anal. ($C_{18}H_{17}NO_3$) C, H, N. The compound eluted second was recrystallized from Me_2CO to afford 255 mg (24%) of **19** as colorless needles: mp 240–241 °C. Anal. ($C_{18}H_{17}NO_3$) C, H, N.

Ethyl 2-Chloro-7-isopropyl-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carboxylate (20) (Method M). A mixture of **17d** (R_1 = 7-*i*-Pr, 1.09 g), PCl_5 (1.38 g), and $POCl_3$ (14 mL) was stirred for 7 h at 120 °C. The reaction mixture was concentrated in vacuo, and the residue was dissolved in EtOH (10 mL). This reaction mixture was concentrated in vacuo, and the residue was chromatographed on a column of silica gel (25 g) and eluted with benzene. The eluate was evaporated to dryness and the residue was recrystallized from isopropyl ether to afford 0.67 g (58%) of colorless needles: mp 160–161 °C. Anal. ($C_{18}H_{16}ClNO_4$) C, H, N.

Ethyl 7-Isopropyl-2-(methoxyamino)-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carboxylate (21a) (Method R). A mixture of **20** (285 g), *O*-methylhydroxylamine hydrochloride (100 g), and triethylamine (240 g) in $CHCl_3$ (4.2 L) was refluxed for 24 h and then cooled. The reaction mixture was washed with H_2O (1.5 L) and dried over anhydrous Na_2SO_4. The solvent was evaporated in vacuo and the residue was recrystallized from benzene to afford 231.5 g (79%) of colorless crystals: mp 208–209 °C. Anal. ($C_{19}H_{20}N_2O_5$) C, H, N.

3-(2-Hydroxy-5-ethylbenzoyl)-6-methyl-2(1*H*)-pyridone-5-carboxylic Acid (22). The ester **16a** (106 mg) was added to 1 N NaOH (5 mL), and the mixture was stirred for 3 h at 100 °C. After the reaction mixture was cooled, an insoluble material was filtered off and the filtrate was acidified with 1 N HCl. The precipitate was collected by filtration and recrystallized from DMF–H_2O to afford 70 mg (68%) of yellow fine crystals: mp 236–238 °C dec. Anal. ($C_{16}H_{15}NO_5$) C, H, N.

2-Amino-7-isopropyl-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carboxylic Acid (23g) (Method T). A suspension of **16c** (1.5 g) in 50% H_2SO_4–AcOH (1:1) (36 mL) was stirred for 3 h at 130 °C. After the reaction was terminated, the reaction mixture was poured into water. The precipitate was collected by filtration, washed with water, and recrystallized from DMF to give 1.20 g (88%) of colorless crystals: mp >300 °C. Anal. ($C_{16}H_{14}N_2O_4$) C, H, N.

7-Ethyl-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-2,3-dicarboxylic Acid (23v). Diethyl ester **16g** (3.69 g, 10 mmol) was dissolved in 80% H_2SO_4 (20 mL) and AcOH (3.5 mL) at 70 °C, and H_2O (7 mL) was added to this solution. The mixture was stirred for 1 h at 70 °C, then H_2O (100 mL) was added, and the mixture was cooled. The separated solid was collected by filtration and dissolved in 1 N NaOH (60 mL). After the solution was stirred for 1 h at room temperature, it was acidified with concentrated HCl. The precipitate was collected by filtration, washed with water, and recrystallized from MeOH–H_2O to afford 2.64 g (84%) of colorless needles: mp 206–207 °C dec. Anal. ($C_{16}H_{11}NO_6$) C, H, N.

7-Ethyl-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carboxylic Acid (23c) (Method T). A suspension of **8b** (3.27 g) in 50% H_2SO_4–AcOH (1:2) (36 mL) was stirred for 3 h at 130 °C. After the reaction was terminated, the reaction mixture was poured into water. The precipitate was collected by filtration, washed with water, and recrystallized from DMF-H_2O to give 2.93 g (90%) of colorless crystals: mp 238–239 °C. Anal. ($C_{15}H_{11}NO_4$) C, H, N.

7-Ethyl-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carboxylic Acid (23c). Dicarboxylic acid **23v** (313 mg, 1.0 mmol) was molten by gentle heating on a hot plate. After effervescence had ceased, the product was recrystallized twice from acetone (charcoal) to give 126 mg (46%) of colorless needles: mp 238–239 °C.

2-Amino-7-ethyl-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine-3-carboxylic Acid (23f) (Method S). A mixture of **4c** (10.9 g), cyanoacetic acid (10 g), and pyridine (25 mL) was stirred at 120 °C. After 15 min and another 45 min, two additional portions of cyanoacetic acid (10 g) were added, and the mixture was stirred at the same temperature for 1.5 h. The reaction mixture was cooled, and the precipitate was collected by filtration. Recrystallization from DMF afforded 7.80 g (55%) of pale yellow crystals: mp >300 °C. Anal. ($C_{15}H_{12}N_2O_4$) C, H, N.

7-Isopropyl-3-(1*H*-tetrazol-5-yl)-5-oxo-5*H*-[1]benzopyrano[2,3-*b*]pyridine (24c) (Method U). A mixture of **9c** (161.8 g), DMF (3.5 L), NaN_3 (119.6 g), and NH_4Cl (98.4 g) was stirred for 2.5 h at 120 °C. After the reaction mixture had been cooled, an insoluble material was filtered off. The filtrate was acidified with concentrated HCl (20 mL) and concentrated in vacuo. Water was added to the residue and the precipitate was collected by filtration, washed with water, and dissolved in hot DMF (900 mL). EtOH (1.2 L) was added to the solution, and the mixture was allowed to stand overnight in a refrigerator. The precipitate was collected by filtration. Triethanolamine (113 g), H_2O (2.6 L), and AcOEt (800 mL) were added to the precipitate, and the mixture was stirred. The aqueous layer was separated from the organic layer and filtered to remove an insoluble material. The filtrate was acidified with concentrated HCl (90 mL) at 60 °C. After the reaction mixture was stirred for 30 min at 60 °C, the separated crystals were collected by filtration, washed with H_2O, and dried in vacuo to give 184 g (97%) of **24c**, mp 272–275 °C dec, which was recrystallized from DMF to afford colorless prisms: mp 275–277 °C dec. Anal. ($C_{16}H_{13}N_5O_2$) C, H, N.

Biological Assay. Male Sprague–Dawley rats, 8 weeks old and weighing about 280 g, were used. Rat antiserum containing homocytotropic antibody was prepared according to the method of Mota.[17] In brief, the animals were sensitized by intramuscular injections of 1 mg of egg albumin in 1 mL of saline solution concomitantly with an intraperitoneal injection of 2×10^{10} of killed *Bordetella pertussis*. Serum collected from each animal 12 days after sensitization was pooled and frozen until use. The biological properties of the skin sensitizing antibody contained in these sera satisfy the requirements for a homocytotropic antibody; i.e., it fixes homologous skin tissue for a long time and is heat labile. The antisera showed passive cutaneous anaphylaxis (PCA, 72–h latent period) titers of 1:32 to 1:64. The homologous rat PCA response was elicited as follows. Four 0.05-mL aliquots of serum diluted fourfold with physiological saline solutions were injected intradermally into the shaved dorsal skin of the rat. After 72 h, the rat was challenged with an intravenous injection of 1 mL of saline solution containing 5 mg of egg albumin and 10 mg of Evans blue. The drugs to be tested or vehicles (saline or polyethylene glycol 400) were administered intravenously immediately before antigen challenge. In the case of oral administration, the drugs suspended in 5% gum arabicum were administered 5 min before antigen challenge. Rats were killed by bleeding 30 min later, and the area of the dye leakage was measured in square millimeters. The ID_{50} value, i.e., the dose required to cause 50% inhibition of the PCA reaction, was calculated from the relation between the logarithmic dose and the area of dye leakage by the method of least squares. Fiducial limits of the ID_{50} values were calculated according to Fieller's theorem.[18] The anti-SRS-A activity in guinea pig ileum and the inhibitory effect on SRS-A generation in guinea pig lung were assayed according to the method described previously.[19]

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Registry No. **3a**, 50743-17-4; **3b**, 50743-18-5; **3c**, 50743-19-6; **3d**, 50743-32-3; **3e**, 68301-74-6; **3f**, 94978-86-6; **3g**, 50743-21-0; **3h**, 53428-23-2; **3i**, 50743-28-7; **4a**, 61424-76-8; **4b**, 68301-75-7; **4c**, 68301-76-8; **4d**, 68301-82-6; **4e**, 68301-87-1; **4f**, 94978-87-7; **4g**, 68301-78-0; **4h**, 68301-84-8; **4i**, 68301-85-9; **7a**, 68302-19-2; **7b**, 68302-39-6; **8a**, 68302-20-5; **8b**, 68302-41-0; **8c**, 78921-06-9; **9a**, 68302-31-8; **9b**, 68302-29-4; **9c**, 68302-32-9; **9d**, 68302-35-2; **9e**, 68302-34-1; **10**, 78893-28-4; **11a**, 78893-29-5; **11b**, 68302-42-1; **12a**, 78893-30-8; **12b**, 78893-31-9; **12c**, 78893-32-0; **13a**, 78893-34-2; **13b**, 78893-35-3; **14a**, 68302-09-0; **14b**, 68302-12-5; **15a**, 78893-36-4; **15b**,

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Studies on 4(1*H*)-Quinazolinones. 5.[1] Synthesis and Antiinflammatory Activity of 4(1*H*)-Quinazolinone Derivatives

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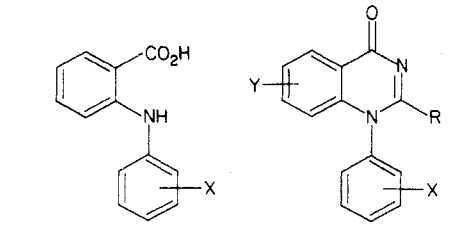
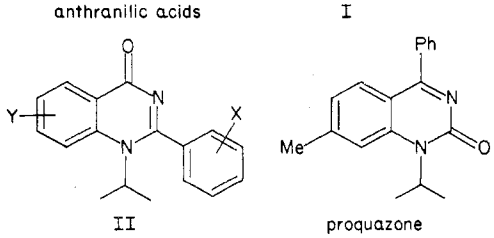
A number of new 4(1*H*)-quinazolinones were synthesized and evaluated in the carrageenin-induced paw edema test. Most of the compounds were obtained by the cyclization of the appropriately substituted anthranilamides with acid chlorides, followed by further chemical transformation. Structure–activity data suggest that 2-isopropyl-1-phenyl-, 2-cyclopropyl-1-phenyl-, and 1-isopropyl-2-phenyl-4(1*H*)-quinazolinones afford optimal potency and the presence of a halogen atom is preferred for activity. Adrenalectomy does not affect the antiinflammatory test results. The best result taking into account both efficacy and side effects was displayed by 1-isoproyl-(2-fluorophenyl)-4-(1*H*)-quinazolinone (**50**).

Various kinds of nonsteroidal antiinflammatory agents have been clinically used. However, most of these agents have gastrointestinal toxicity as a side effect. In order to search for a new drug without this characteristic side effect, we prepared 1-phenyl-4(1*H*)-quinazolinones I, because these were considered to be cyclic analogues of *N*-phenylanthranilic acids, which are known to possess potent antiinflammatory activity. In the course of this study, 2-isopropyl-1-phenyl-4(1*H*)-quinazolinones (I, R = isopropyl) were found to have marked antiinflammatory activity. We recognized the structural resemblance between I (R = isopropyl) and proquazone.[2] As a result of our continuing study, it was found that 1-isopropyl-2-phenyl-4(1*H*)-quinazolinones II also showed a good level of activity. In this paper, we report the synthesis and antiinflammatory activity of these 4(1*H*)-quinazolinones.





Chemistry. Most of the quinazolinone derivatives in Tables I–III were prepared by the methods illustrated in Schemes I–III. Scheme I shows the synthetic route to I and the dihydro derivative VII (Table II, **47**). Schemes II and III show the syntheses of 4-isopropyl-1-phenyl-2-(1*H*)-quinazolinone (IX, Table II, **44**), II, and the dihydro compounds XIII.

The starting *N*-phenylanthranilic acids were prepared by the Ullmann reaction. The anthranilic acids III were allowed to react with thionyl chloride (method A) or phosgene (method B), followed by treatment with aqueous ammonia to afford the corresponding 2-anilinobenzamides IV. Alternatively, two of the amides, 2-(2-toluidino)- and 2-anilino-5-methoxybenzamide (IVd and IVn, respectively), were prepared from the methyl *N*-phenylanthranilates VI (method C). Compounds I were generally obtained in moderate yield from IV by our reported method[1a] using an acyl chloride in chloroform (*N*,*N*-dimethylformamide was used as a solvent in those cases where solubility of IV in chloroform was low). The reaction of 2-(4-nitroanilino)benzamide (IVh) with cyclopropanecarbonyl chloride or cyclobutanecarbonyl chloride gave a mixture of intermediate *N*-(2-carbamoylphenyl)-*N*-(4-nitrophenyl)acylamide V and the cyclized 4(1*H*)-quinazolinone **27** or **29**. Without purification, each mixture was refluxed in acetic acid containing boron trifluoride etherate to afford

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