converted directly to the quinone.

A solution of 170 mg (0.47 mmol) of the aminoaldehyde in 32.5 ml of acetone was treated with a solution of 612 mg (2.3 mmol) of potassium nitrosodisulfonate in 16 ml of 0.167 M potassium dihydrogen phosphate and 32.5 ml of acetone. After 20 h the resulting solution was diluted with water and extracted with dichloromethane. This extract was dried (Na_2SO_4) and concentrated and the residue was chromatographed on silica gel with chloroform and then ethyl acetate as eluents. Concentration of the main yellow band gave 50 mg (28%) of quinone **13**: glistening orange crystals from ethanol; mp 247 °C dec; ir 5.68 (ester), 5.9–6.2 μ (quinone, amide, and aldehyde carbonyls); NMR δ 10.30 (s, 1, CHO), 8.35 (m, 1, NH), 5.80 ppm (broad, 2, CHNAc and CHOAc).

cis-**1-Acetamido-2-acetoxy-2,3-dihydro-9-hydroxymethyl-7-methoxy-6-methyl-1*H*-pyrrolo[1,2-*a*]indole-5,8-dione Methyl Carbamate (1-Acetamido-2-acetoxy-7-methoxy-*N*-methylmitosene, 15).** A stirred solution of 60 mg of **13** in 10.5 ml of ethanol and 10.5 ml of tetrahydrofuran, at 0 °C under nitrogen, was treated with 120 mg (excess) of sodium borohydride. After 30 min, acetone (3 ml) was added. The mixture was stirred 10 min, treated with 0.40 ml of 1 M ferric chloride in 0.1 M hydrochloric acid, stirred another 5 min, and diluted with water. The resulting mixture was extracted with dichloromethane and this extract was dried and concentrated to give 60 mg of crude hydroxymethylquinone **14**. This product, which showed only one spot on TLC (R_f 0.08 in 3:1 chloroform–acetone), was used directly to prepare the methyl carbamate.

A mixture of 30 mg of crude **14**, 5 ml (excess) of methyl isocyanate, 5 ml of tetrahydrofuran, and 0.1 ml of triethylamine was kept at room temperature for 4 h and then concentrated under reduced pressure. The residual solid was chromatographed on a silica gel column (15 mm × 30 cm) with 1:1 chloroform–acetone as solvent. Following a blue impurity and a small yellowish band, the main yellow band eluted. Concentration of this main eluate gave a yellow solid which was recrystallized from ethanol. Golden crystals (10 mg) were obtained: mp 226 °C dec; ir 3.1 (NH), 5.65–5.8 (carbamate and ester), 6.0–6.2 μ (amide and quinone carbonyls).

Acknowledgment. This investigation was supported by Grant CA 11686 from the National Institutes of Health. We wish to express our appreciation to Dr. M. Misiek and Dr. W. T. Bradner of Bristol Laboratories for determining the antibacterial and antitumor activities, respectively, of the compounds reported in this article.

References and Notes

(1) J. S. Webb, D. B. Cosulich, J. H. Mowat, J. B. Patrick, R. W. Broschard, W. E. Meyer, R. P. Williams, C. F. Wolf, W. Fulmor, C. Pidacks, and J. E. Lancaster, *J. Am. Chem. Soc.*, **84**, 3185 (1962).
(2) J. B. Patrick, R. P. Williams, W. E. Meyer, W. Fulmor, D. B. Cosulich, R. W. Broschard, J. S. Webb, *J. Am. Chem. Soc.*, **86**, 1889 (1964).
(3) W. G. Taylor and W. A. Remers, *J. Med. Chem.*, **18**, 307 (1975).
(4) G. Leadbetter, D. L. Fost, N. N. Ekwuribe, and W. A. Remers, *J. Org. Chem.*, **39**, 3580 (1974).
(5) S. Kinoshita, K. Uzu, K. Nakano, M. Shimizu, T. Takahashi, S. Wakaki, and M. Matsui, *Prog. Antimicrob. Anticancer Chemother.*, **2**, 1058 (1970).
(6) M. Tomasz, C. M. Mercado, J. Olson, and N. Chatterjie, *Biochemistry*, **13**, 4878 (1974).
(7) W. A. Remers and C. S. Schepman, *J. Med. Chem.*, **17**, 729 (1974).
(8) I. Usubuchi, T. Sobajima, T. Hongo, T. Kawaguchi, M. Sugawara, M. Matsui, S. Wakaki, and K. Uzu, *Gann*, **58**, 307 (1967).
(9) S. Oboshi, M. Matsui, S. Ishii, N. Masago, S. Wakaki, and K. Uzu, *Gann*, **58**, 315 (1967).
(10) R. Kojima, J. Driscoll, N. Mantel, and A. Goldin, *Cancer Chemother. Rep.*, **3**, 121 (1972).
(11) W. A. Remers, R. H. Roth, and M. J. Weiss, *J. Org. Chem.*, **30**, 2910 (1965).
(12) G. R. Allen, Jr., and M. J. Weiss, *J. Am. Chem. Soc.*, **86**, 3877 (1964).
(13) G. R. Allen, Jr., and M. J. Weiss, *J. Org. Chem.*, **30**, 2904 (1965).
(14) J. F. Poletto, G. R. Allen, Jr., and M. J. Weiss, *J. Med. Chem.*, **11**, 882 (1968).
(15) *Cancer Chemother. Rep., Part 3*, **3**, 9 (1972).

Studies on Antianaphylactic Agents. 5.[1] Synthesis of 3-(1*H*-Tetrazol-5-yl)chromones, a New Series of Antiallergic Substances

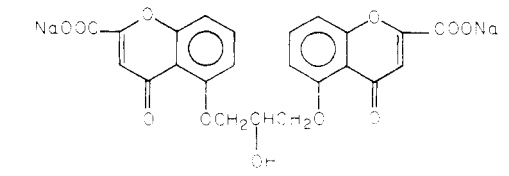
Akira Nohara,* Hisashi Kuriki, Taketoshi Saijo, Hirosada Sugihara, Morio Kanno, and Yasushi Sanno

Medicinal Research Laboratories, Central Research Division, Takeda Chemical Industries, Ltd., Yodogawa-ku, Osaka 532, Japan. *Received May 17, 1976*

A number of 3-(1*H*-tetrazol-5-yl)chromones were synthesized and found to have antiallergic activity in the rat passive cutaneous anaphylaxis (PCA) test. These compounds are active when administered orally in rats and of possible value for the treatment of asthma.

Disodium cromoglycate (DSCG, **1**) has been established as being of use in the treatment of some types of bronchial asthma.[2] It has been shown to inhibit the liberation of mediators of immediate type allergic reactions initiated by reaginic antibody–antigen interactions.[3] It inhibits homologous passive cutaneous anaphylaxis (PCA) reactions in the rat induced by reaginic antibody and this reaction has been used as a routine screen for compounds with similar biological activities.[4] Although many kinds of compounds possessing a similar activity have been reported, much attention is being devoted to the antiallergic agents which can be administered orally.

Following the observation that introduction of a carbonyl group at the 3 position enhanced the antiallergic activity of chromones,[5] we began a program of investigation of 3-substituted chromone derivatives. As part of this program we have reported the activity of 3-(4-oxo-4*H*-1-benzopyran-3)acrylic acids (**2**).[1] We have also found, in sharp contrast to DSCG-type 4-oxo-4*H*-1-benzopyran-2-carboxylic acids (**3**), that some 4-oxo-4*H*-1-benzopyran-3-carboxylic acids (**4**) are inactive in inhibiting PCA in rats.[6] Inactivity of **4** was attributed to its weak acidity (**4**, R = H; $pK_a' = 8.85$[6]) due to intramolecular hydrogen bond formation.



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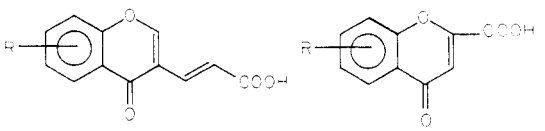

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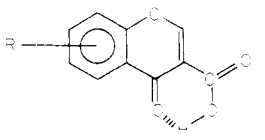


4

Recently, attempts to replace the carboxylic acid group with a tetrazole ring have been made with chromones,[7] xanthones,[8] and a thioxanthone derivative,[9] because 1*H*-tetrazoles generally show acidity comparable with carboxylic acids.[10] Furthermore, these examples indicate that biologically active tetrazoles can be obtained from the corresponding active carboxylic acids but not from the inactive acids, suggesting that the substitution of the tetrazole group for the carboxylic acid group in **4** would give rise to inactive compounds. Contrary to previous findings, however, 3-(1*H*-tetrazol-5-yl)chromone derivatives (**7**),[11] which proved to be stronger acids than **4** (e.g., **6**, R = H; $pK_a' = 5.85$), were found to be active not only when administered intravenously but also orally. This is particularly significant because DSCG (**1**) is orally inactive and is usually administered by inhalation. This paper describes the synthesis and some of the biological properties of these new antiallergic agents.

Chemistry. Synthesis of 3-(1*H*-tetrazol-5-yl)chromones was carried out by the synthetic route shown in Scheme I. Reaction of 4-oxo-4*H*-1-benzopyran-3-carboxaldehyde (**5**)[12,13] with hydroxylamine hydrochloride in 95% ethanol in the presence of hydrochloric acid[14] afforded in one step[15] the 3-carbonitrile derivative **6a**. In a similar manner, **6b–l** and naphtho[2,1-*b*]pyran-2-carbonitrile (**8b**) were synthesized from **5b–l**[13] and **8a**.[16] Hydroxycarbonitrile derivatives (**6q,r**) were prepared from the appropriate acetoxy starting materials (**5m**,[13] **5n**[13]). In the case of **5o** and naphtho[1,2-*b*]pyran-3-carboxaldehyde (**9a**),[16] the isolated oximes were converted to the nitrile derivatives (**6o, 9b**) by the reaction with acetic anhydride. The 6-nitrocarbonitrile derivative **6p** was prepared in good yield by the nitration of **6a** with fuming nitric acid in 95% sulfuric acid. The 7-butoxy derivative **6s** was prepared from **6q** by alkylation with butyl iodide (Table I).

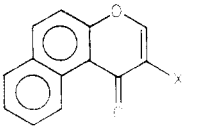
Conversion of the cyano group to the tetrazole ring was achieved with sodium azide in the presence of anhydrous aluminum chloride in tetrahydrofuran.[17] By this method, 3-(1*H*-tetrazol-5-yl)chromones **7a–l, 7o–s, 8c**, and **9c** were prepared from **6a–l, 6o–s, 8b**, and **9b**. The 6-methoxytetrazole derivative **7j** was demethylated by hydriodic acid to give the 6-hydroxy derivative **7t**. The 6-nitrotetrazole derivative **7p** was also prepared by the nitration of **7a**.

In order to investigate the importance of acidity of the tetrazole ring for biological activity, 3-(2-*tert*-butyl-2*H*-tetrazol-5-yl)chromone (**10**) was prepared by the reaction of **7a** with *tert*-butyl alcohol in trifluoroacetic acid in the presence of 95% sulfuric acid, and conversion of tetrazole **7a** to the corresponding oxadiazole derivative **11** was conducted with acetic anhydride.[18] Hydrolytic degradation of **7a** in sodium hydroxide afforded the acetophenone derivative **12**. The ring closure of **12** was effected by acetic anhydride and pyridine to give 2-methylchromone derivative **13**.

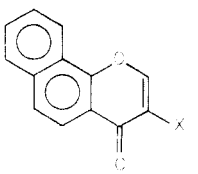
Table I. 4-Oxo-4*H*-1-benzopyran-3-carbonitriles

Compd	Mp, °C	Crystn solvent[a]	Yield, %	Formula[b]
6a	177–178	A	76	$C_{10}H_5NO_2$
6b	152.5–153.5	A	58	$C_{11}H_7NO_2$
6c	123–124	A	69	$C_{12}H_9NO_2$
6d	102–104	A	52	$C_{13}H_{11}NO_2$
6e	118–120	A	62	$C_{13}H_{11}NO_2$
6f	94–95	A	63	$C_{14}H_{13}NO_2$
6g	55	B	25	$C_{16}H_{17}NO_2$
6h	164–165	C	41	$C_{16}H_{15}NO_2$
6i	210–213	C	51	$C_{10}H_4ClNO_2$
6j	194–195	A	49	$C_{11}H_7NO_3$
6k	167–168	D	36	$C_{12}H_{10}N_2O_2$
6l	196–198	E	51	$C_{12}H_9NO_2$
6o[c]	164–165	F	37	$C_{13}H_9NO_3$
6p[c]	211–213	G	95	$C_{10}H_4N_2O_4$
6q	278–280	A	48	$C_{10}H_5NO_3$
6r	>300	H	28	$C_{10}H_5NO_4$
6s	120–121	I	68	$C_{14}H_{13}NO_3$
8b	194.5–195.5	A	67	$C_{12}H_7NO_2$
9b[c]	229–230	E	69	$C_{13}H_7NO_2$

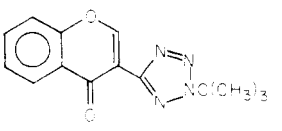
[a] A = EtOH, B = EtOH-hexane, C = MeOH, D = $CHCl_3$-EtOAc, E = Me_2CO, F = benzene, G = MeOH-$CHCl_3$, H = DMF-H_2O, I = benzene-hexane. [b] All compounds were analyzed for C, H, and N. [c] See Experimental Section.



8a, X = CHO
b, X = CN
c, X = 



9a, X = CHO
b, X = CN
c, X = 



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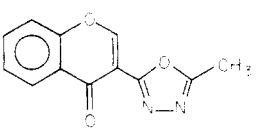

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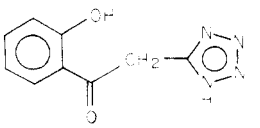

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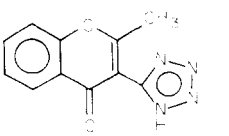


13

In the preceding paper,[1] *trans*-3-(4-oxo-4*H*-1-benzopyran-3)acrylic acids (**2**) were reported as antiallergic agents. To determine the effect of the substitution of the carboxy group by a tetrazole in this series, the acrylonitrile derivatives **14a,b** prepared from **5a,f**[13] were converted to **15a,b** by aluminum azide as shown above. The propionitrile derivative **16**, obtained by the catalytic reduction of **14a**, was also converted to tetrazole **17** by reaction with ammonium azide in dimethylformamide.[19] Also, **18b** was

Table II. 3-(1*H*-Tetrazol-5-yl)chromones

Compd	Mp, °C	Crystn solvent[a]	Yield, %	Formula[b]	PCA assay, rel potency
1					1.0
19					1.4
7a	297-299 dec	A	80	$C_{10}H_6N_4O_2$	3.5
7b	258-259	B	44	$C_{11}H_8N_4O_2$	2.4
7c	220-222	C	81	$C_{12}H_{10}N_4O_2$	4.0
7d	214-215	B	40	$C_{13}H_{12}N_4O_2$	3.4
7e	222-223	B	48	$C_{13}H_{12}N_4O_2$	3.4
7f	206-209	B	32	$C_{14}H_{14}N_4O_2$	1.8
7g	207-210	B	85	$C_{16}H_{18}N_4O_2$	0.6
7h	252-253	D	72	$C_{16}H_{16}N_4O_2$	0.5
7i	275-277 dec	B	43	$C_{10}H_5ClN_4O_2$	4.0
7j	292-293 dec	A	59	$C_{11}H_8N_4O_3$	2.2
7k	303-305 dec	E	57	$C_{12}H_{11}N_5O_2$	2.3
7l	274-275 dec	A	58	$C_{12}H_{10}N_4O_2$	11.6
7o	271-273 dec	E	49	$C_{13}H_{10}N_4O_4$	2.9
7p	285-286 dec	A	78	$C_{10}H_5N_5O_4$	4.0
7q	>300	F	70	$C_{10}H_6N_4O_3$	1.1
7r	>300	F	79	$C_{10}H_6N_4O_4 \cdot 0.5H_2O$[c]	1.8
7s	236-238 dec	E	78	$C_{14}H_{14}N_4O_3$	0.9
7t	>300	F	69	$C_{10}H_6N_4O_3$	4.5
8c	282-285 dec	F	38	$C_{14}H_8N_4O_2$	5.0
9c	303-305 dec	F	43	$C_{14}H_8N_4O_2$	8.3

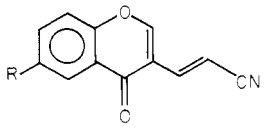
[a] A = DMF, B = DMF-MeOH, C = EtOH, D = THF-MeOH, E = DMF-Me_2CO, F = DMF-H_2O. [b] All compounds were analyzed for C, H, and N. [c] C: calcd, 47.07; found, 47.82.

Table III. Miscellaneous Compounds

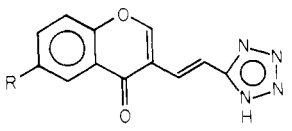
Compd	Mp, °C	Crystn solvent[a]	Yield, %	Formula[b]	PCA assay, rel potency
10	120-121	A	77	$C_{14}H_{14}N_4O_2$	*c*
11	165-166	B	72	$C_{12}H_8N_2O_3$	*c*
12	200-201	B	70	$C_9H_8N_4O_2$	0.35
13	280-282 dec	C	64	$C_{11}H_8N_4O_2$	0.1
15a	254.5-255 dec	D	18	$C_{12}H_8N_4O_2$	0.3
15b	247-250	C	11	$C_{16}H_{16}N_4O_2$	0.07
17	179-181 dec	A	20	$C_{12}H_{10}N_2O_2$	0.07
18b	252-253.5	E	50	$C_{12}H_{10}N_4O_2$	*c*

[a] A = *n*-heptane, B = EtOH, C = DMF-Me_2CO, D = MeOH, E = DMF-MeOH. [b] All compounds were analyzed for C, H, and N. [c] These compounds have no significant effect up to 20 mg/kg.

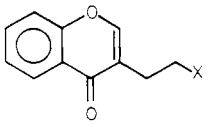
prepared from the acetonitrile derivative **18a**.[1]



14a, R = H
b, R = *n*-Bu

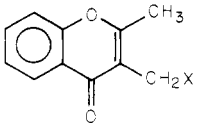


15a, R = H
b, R = *n*-Bu



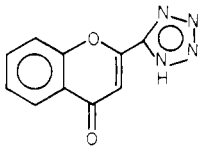
16, X = CN

17, X = 



18a, X = CN

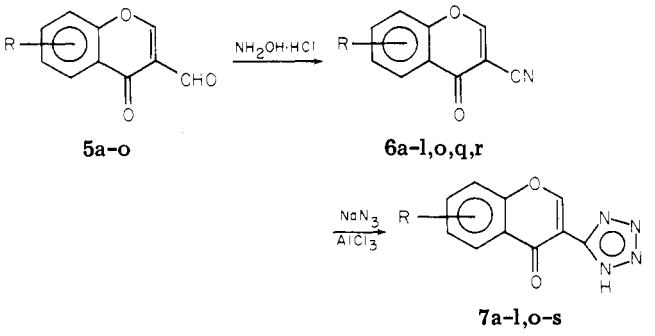
b, X = 



19

Structure–Activity Relationships. The biological activities were measured by the standard rat PCA tests as described in the Experimental Section and compared with disodium cromoglycate (**1**). The results are shown in Tables II–IV.

Scheme I



5a–o **6a–l,o,q,r** **7a–l,o–s**

a, R = H	k, R = 6-NMe_2
b, R = 6-Me	l, R = 6,8-Me_2
c, R = 6-Et	m, R = 7-OAc
d, R = 6-*n*-Pr	n, R = 6,7-$(OAc)_2$
e, R = 6-*i*-Pr	o, R = 6-COOEt
f, R = 6-*n*-Bu	p, R = 6-NO_2
g, R = 6-*n*-hexyl	q, R = 7-OH
h, R = 6-cyclohexyl	r, R = 6,7-$(OH)_2$
i, R = 6-Cl	s, R = 7-O-*n*-Bu
j, R = 6-OMe	t, R = 6-OH

The effects of varying substituents on the carbocyclic ring of the chromone nucleus are shown in Table II. All the compounds shown in Table II were highly potent inhibitors of the PCA reaction. The most noteworthy compounds were **7c,i,l,p,t**, **8c**, and **9c** which were ca. 4–10 times as active as disodium cromoglycate (**1**). When a comparison was made between the biological activity of

Table IV. Inhibitory Effects of Orally Administered 3-(1*H*-Tetrazol-5-yl)chromones on Passive Cutaneous Anaphylaxis in Rats

Compd	No. of rats used	No. of expt	ID_{50},[a] mg/kg
7a	18	2	5.3[b] (5.5, 5.0)
7b	9	1	10.0
7c	30	4	6.9 ± 1.7[c]
7e	9	1	17.0
7j	9	1	7.0
7l	9	1	1.25
8c	9	1	8.5
9c	9	1	7.4
1	9	1	>100

[a] ID_{50}: 50% inhibition dose. [b] Mean value of two experiments. [c] The value is mean ± standard error of four experiments.

analogues bearing a tetrazole ring on the 2 and 3 positions of the chromone ring, **7a** proved to be 2.5 times as active as the isomer **19**.[7] Moreover, **7** and its analogues **8c** and **9c** are active when administered orally. Introduction of methyl group at the 2 position of the chromone nucleus (i.e., **13**) reduces the biological activity. Similar results have also been obtained with the acrylic acid derivatives **2**.[1] It is interesting that the acetophenone derivative **12** which is not as rigid as **7** also shows some activity. On the other hand, **15** and **17** show the same extent of activities as the corresponding carboxy derivatives. The need for the acidic tetrazole ring is shown by the inactivity of **10** and **11**. The fact that **12**, **13**, and **18b** show weak or no activity seems to show that there exists a severe structural restriction for a compound to exert antiallergic activity.

Experimental Section

Melting points were determined with a micromelting point apparatus (Yanagimoto) and are uncorrected. Where analyses are indicated only by symbols of the elements, the analytical results obtained for these elements were within ±0.4% of the theoretical value. NMR spectra were recorded on a Varian Associates T-60 instrument with Me_4Si as an internal standard. Ir spectra were measured on a Hitachi infrared spectrophotometer EPI-S2. Mass spectra were recorded on Hitachi RMU-6D or Hitachi RMS-4 instruments.

4-Oxo-4*H*-1-benzopyran-3-carbonitrile (6a).[11] A mixture of 4-oxo-4*H*-1-benzopyran-3-carboxaldehyde (**5a**,[12,13] 87 g, 0.5 mol), hydroxylamine hydrochloride (36 g, 0.5 mol), 95% EtOH (500 ml), and concentrated HCl (8 ml) was refluxed for 12 h with stirring. After cooling, the precipitates were collected by filtration and recrystallized from EtOH to afford 64.6 g (76%) of colorless crystals.

6-Ethoxycarbonyl-4-oxo-4*H*-1-benzopyran-3-carbonitrile (6o). HCl gas was bubbled through the suspension of 6-cyano-4-oxo-4*H*-1-benzopyran-3-carboxaldehyde[13] (3.8 g, 19 mmol) in EtOH (200 ml) for 1 h. After the reaction temperature reached 50 °C, the solvent was evaporated in vacuo and H_2O (100 ml) was added to the residue. The mixture was heated at 80 °C for 10 min and extracted with EtOAc. The extract was washed with H_2O and dried with Na_2SO_4. The solvent was evaporated in vacuo to afford 4.2 g of 6-ethoxycarbonyl-4-oxo-4*H*-1-benzopyran-3-carboxaldehyde (**5o**). A mixture of **5o** (4.2 g, 17 mmol), 95% EtOH (70 ml), and hydroxylamine hydrochloride (1.19 g, 17 mmol) was heated at 80 °C for 5 min. The resulting solution was concentrated to two-thirds volume in vacuo to afford 2.7 g of a precipitate, which was collected by filtration and dissolved in Ac_2O (50 ml). The solution was heated at 130 °C for 3 h. After evaporation of Ac_2O, the residue was recrystallized from benzene to afford 1.7 g (37%) of colorless needles (**6o**).

6-Nitro-4-oxo-4*H*-1-benzopyran-3-carbonitrile (6p). To a suspension of **6a** (342 mg, 2.0 mmol) in 95% H_2SO_4 was added fuming nitric acid (2 ml) under ice–water cooling and the reaction mixture was kept standing at room temperature for 1.5 h. The solution was poured into ice water and a precipitate was collected by filtration. Recrystallization from $CHCl_3$–MeOH afforded 412 mg (95%) of colorless crystals.

7-Butoxy-4-oxo-4*H*-1-benzopyran-3-carbonitrile (6s). A mixture of **6q** (500 mg, 2.7 mmol), dried Me_2SO (10 ml), anhydrous K_2CO_3 (500 mg), and *n*-butyl iodide (600 mg) was stirred at room temperature for 4 h, poured into ice–water (200 ml), and acidified with 1 N HCl to pH 1. A pale yellow precipitate was collected by filtration, washed with H_2O, and dried. Recrystallization from benzene–*n*-hexane afforded 445 mg (68%) of pale yellow crystals.

Naphtho[1,2-*b*]pyran-3-carbonitrile (9b). A mixture of **9a**[16] (1.12 g, 5 mmol), hydroxylamine hydrochloride (350 mg, 5 mmol), 95% EtOH (20 ml), and concentrated HCl (0.1 ml) was refluxed with stirring. After 3 h, additional 95% EtOH (30 ml) was added to the reaction mixture. After a further 4 h, the reaction mixture was cooled and an insoluble material was collected by filtration. The precipitate was added to Ac_2O (20 ml) and the mixture was refluxed for 1 h. After the solvent was removed from the mixture, the residue was recrystallized from acetone to afford 760 mg (69%) of yellow crystals.

3-(1*H*-Tetrazol-5-yl)chromone (7a).[11] To THF (600 ml) precooled in an ice bath were added pulverized anhydrous $AlCl_3$ (58.5 g, 0.44 mol), **6a** (34.2 g, 0.2 mol), and sodium azide (57.2 g, 0.88 mol) in this order, and the ice bath was removed. The mixture was then refluxed for 4 h with stirring. The following treatment was done in a well-ventilated draft. HCl (15%, 175 ml) was added to the reaction mixture and the liquid phase was obtained by decantation. After the solvent was evaporated in vacuo, the resulting solid was collected by filtration and recrystallized from DMF to afford 34.2 g (80%) of colorless needles.

6-Nitro-3-(1*H*-tetrazol-5-yl)chromone (7p). To a solution of **7a** (6.425 g, 30 mmol) in 95% H_2SO_4 (40 ml) was added fuming nitric acid (4 ml) at 30–35 °C and it was kept standing at room temperature for 10 min. The solution was poured into ice water (700 ml) and a precipitate was collected by filtration and recrystallized from DMF to afford 6.04 g (78%) of colorless needles.

6-Hydroxy-3-(1*H*-tetrazol-5-yl)chromone (7t). A mixture of 6-methoxy-3-(1*H*-tetrazol-5-yl)chromone (**7j**, 244 mg, 1 mmol) and 57% hydriodic acid (8 ml) was heated at 140 °C for 2 h with stirring. After cooling, an insoluble material was collected by filtration, washed with H_2O, and recrystallized from DMF–H_2O to afford 158 mg (69%) of colorless needles.

3-(2-*tert*-Butyl-2*H*-tetrazol-5-yl)chromone (10). A solution of **7a** (856 mg, 4 mmol), *tert*-BuOH (0.8 ml), trifluoroacetic acid (4 ml), and 95% H_2SO_4 (0.1 ml) was stirred at room temperature for 2.5 h. The solution was diluted with EtOAc, washed with aqueous 1 N Na_2CO_3 followed by H_2O, dried with Na_2SO_4, and concentrated in vacuo. The residue was crystallized twice from *n*-heptane to afford 840 mg (77%) of colorless prisms.

3-(2-Methyl-1,3,4-oxadiazol-5-yl)chromone (11). A mixture of **7a** (428 mg, 2 mmol) and Ac_2O (5 ml) was refluxed for 1.5 h and Ac_2O was evaporated in vacuo. The resulting residue was recrystallized from EtOH to afford 370 mg (72%) of colorless crystals.

2-(1*H*-Tetrazol-5-yl)-2′-hydroxyacetophenone (12). A solution of **7a** (1.07 g, 5 mmol) in 1 N NaOH (20 ml) was heated at 100 °C for 1 h and acidified with 1 N HCl. The crude product, collected by filtration, was recrystallized from EtOH to afford 670 mg (70%) of colorless needles.

3-(1*H*-Tetrazol-5-yl)-2-methylchromone (13). A mixture of **12** (500 mg, 2.45 mmol), pyridine (1.0 ml), and Ac_2O (0.5 ml) was heated by an electric heater to become solution. After heating for an additional 1-min period, the solution was cooled rapidly to room temperature and the separated crystals were collected by filtration and recrystallized from DMF–Me_2CO to afford 360 mg (64%) of colorless crystals.

***trans*-3-(4-Oxo-4*H*-1-benzopyran-3)acrylonitrile (14a).** To a mixture of **5a** (10.44 g, 60 mmol) and cyanoacetic acid (5.4 g, 66 mmol) was added pyridine (25 ml) during 30 s at 110 °C. After an additional 8-min period, the reaction mixture was cooled and the separated crystals were collected by filtration. Recrystallization from EtOH (charcoal) and then EtOAc afforded 6.6 g (56%) of colorless crystals, mp 193–195 °C. Anal. ($C_{12}H_7NO_2$) C, H, N.

***trans*-3-(6-*n*-Butyl-4-oxo-4*H*-1-benzopyran-3)acrylonitrile (14b)**: mp 124–126 °C. Anal. ($C_{16}H_{15}NO_2$) C, H, N.

3-(4-Oxo-4*H*-1-benzopyran-3)propionitrile (16). Compound **14a** (1.97 g, 10 mmol) was dissolved in diethyl carbonate (100 ml) at 80 °C. To this solution was added Pd black (100 mg) and the mixture was hydrogenated under atmospheric pressure at 80 °C. After 1.8 equiv of hydrogen was absorbed, the catalyst was removed by filtration. The filtrate was concentrated in vacuo and trituration of the residue with EtOH gave a solid. Recrystallization of the solid from EtOH afforded 805 mg (40%) of pale yellow crystals, mp 112–113 °C. Anal. ($C_{12}H_9NO_2$) C, H, N.

1-(4-Oxo-4*H*-1-benzopyran-3)-2-(1*H*-tetrazol-5-yl)ethane (17). A mixture of **16** (590 mg, 2.96 mmol), sodium azide (900 mg, 13.8 mmol), NH_4Cl (250 mg, 4.16 mmol), and DMF (5 ml) was heated at 150 °C for 17 h with stirring. After cooling an insoluble material was filtered off and the filtrate was concentrated in vacuo. Dilute HCl and EtOAc were added to the residue, and the EtOAc layer was extracted with 5% $NaHCO_3$ aqueous solution. The aqueous solution was acidified with concentrated HCl and then extracted with EtOAc. The EtOAc extract was dried with Na_2SO_4 and concentrated in vacuo. The residual solid was recrystallized from EtOH to afford 143 mg (20%) of colorless needles.

Biological Assay. Male Sprague–Dawley rats, 7 weeks old and weighing about 250 g, were used. Rat antiserum containing homocytotropic antibody was prepared according to the method of Mota.[20] 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 20 billion of *Bordetella pertussis* vaccine. 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 1:32–1:64. Homologous rat PCA response was elicited as follows. Four 0.05-ml aliquots of serum diluted fourfold with physiological saline solution 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. Drugs to be tested or vehicles (saline or polyethylene glycol 400) were administered intravenously immediately before antigen challenge. In the case of oral administration, drugs suspended in 5% gum arabicum were administered 5 min before antigen challenge. Rats were sacrificed by bleeding 30 min later, and the area of the dye leakage was measured in square millimeters. The dose giving 50% inhibition (ID_{50}) for each drug was calculated graphically from the dose–inhibition relationship expressed in inhibition percent of the bluing areas against doses on a logarithmic scale. At least three doses and three animals for each dose (i.e., 12 spots) were used for obtaining the dose–inhibition relationship.

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References and Notes

(1) A. Nohara, H. Kuriki, T. Saijo, K. Ukawa, T. Murata, M. Kanno, and Y. Sanno, *J. Med. Chem.*, **18**, 34 (1975).
(2) J. S. G. Cox, *Nature (London)*, **216**, 1328 (1967).
(3) T. S. C. Orr, M. C. Pollard, J. Gwilliam, and J. S. G. Cox, *Clin. Exp. Immunol.*, **7**, 745 (1970).
(4) H. Cairns, C. Fitzmaurice, D. Hunter, P. B. Johnson, J. King, T. B. Lee, G. H. Lord, R. Minshull, and J. S. G. Cox, *J. Med. Chem.*, **15**, 583 (1972).
(5) Y. Sanno, A. Nohara, H. Kuriki, and A. Koda, *J. Takeda Res. Lab.*, **33**, 225 (1974).
(6) A. Nohara, T. Umetani, K. Ukawa, and Y. Sanno, *Chem. Pharm. Bull.*, **22**, 2959 (1974).
(7) (a) G. P. Ellis and D. Shaw, *J. Med. Chem.*, **15**, 865 (1972); (b) *J. Chem. Soc., Perkin Trans. 1*, 779 (1972).
(8) E. S. K. Assem and M. K. McAllen, *Int. Arch. Allergy Appl. Immunol.*, **45**, 697 (1973).
(9) J. F. Batchelor, M. J. Follenfant, L. G. Garland, J. H. Gorvin, A. F. Green, H. F. Hodson, D. T. D. Hughes, and J. E. Tateson, *Lancet*, 1169 (1975).
(10) F. R. Benson, *Heterocycl. Compd.*, **8**, 8 (1967).
(11) A. Nohara, *Tetrahedron Lett.*, 1187 (1974).
(12) F. Eiden and H. Haverland, *Arch. Pharm. (Weinheim, Ger.)*, **300**, 806 (1967).
(13) (a) A. Nohara, T. Umetani, and Y. Sanno, *Tetrahedron Lett.*, 1995 (1973); (b) *Tetrahedron*, **30**, 3553 (1974).
(14) J. A. Findley and C. S. Tang, *Can. J. Chem.*, **45**, 1014 (1967).
(15) Similar results were obtained by the reaction using hydroxylamine in formic acid in the presence of sodium formate: T. van Es, *J. Chem. Soc.*, 1564 (1965).
(16) G. A. Reynolds and J. A. Van Allan, *J. Heterocycl. Chem.*, **6**, 375 (1969).
(17) E. Wiberg and H. Michaud, *Z. Naturforsch., B*, **9**, 496 (1954).
(18) R. Fuisgen and H. G. Margraf, *Angew. Chem.*, **72**, 359 (1960).
(19) W. G. Finnegan, R. A. Henry, and R. L. Lofquist, *J. Am. Chem. Soc.*, **80**, 3908 (1958).
(20) I. Mota, *Life Sci.*, **2**, 917 (1963).