Notes

[Chem. Pharm. Bull. 17(9)1955—1958(1969)]

UDC 547.487.51.07

Studies on Synthesis of Coumarin Derivatives. XXII.[1] On the Preparation of N-Pyridyl-2-alkyl-8-oxo-8*H*-pyrano[3,2-*g*]benzoxazole-7-carboxamide Derivatives

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(Received January 13, 1969)

In the previous paper[3] of this series, it was reported that N-(2-pyridyl)-2-methyl-8-oxo-8*H*-pyrano[3,2-*g*]benzoxazole-7-carboxamide(V) showed the strongest tuberculostatic activity (minimum inhibitory concentration: 3.2 μg/ml) in the derivatives of three isomeric pyranobenzoxazoles and that the cleavage of the oxazole ring extremely decreased such activity in this field.

In connection with the above mentioned, we have further studied the structural features for tuberculostatic activity, related to the derivatives of 8-oxo-8*H*-pyrano[3,2-*g*]benzoxazole-7-carboxamide.

This paper describes synthetic study of ethyl 2-alkyl-8-oxo-8*H*-pyrano[3,2-*g*]benzoxazole-7-carboxylates and tuberculostatic activity of 2-alkyl-8-oxo-8*H*-pyrano[3,2-*g*]benzoxazole-7-carboxamide series derived from them.

In that report,[4] we have studied on the oxazole ring formation of ethyl *o*-amino-hydroxy-3-coumarincarboxylate with dehydrocyclization through between the amino group and the hydroxy group and found that it is so difficult that these oxazole rings are barely formed by

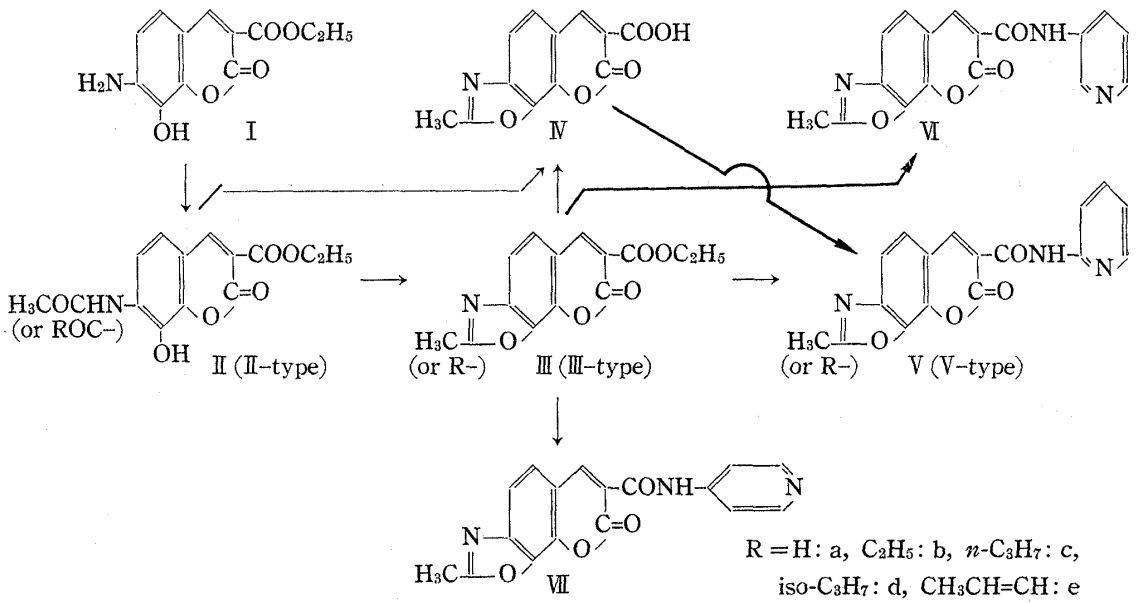

Chart 1

1) Part XXI: H. Ichibagase, M. Ichikawa and N. Shimojo, *Yakugaku Zasshi*, **86**, 755 (1968).
2) Location: *Oehon-machi, Kumamoto.*
3) M. Ichikawa and H. Ichibagase, *Chem. Pharm. Bull.* (Tokyo), **16**, 2093 (1968).
4) H. Saikachi and M. Ichikawa, *Chem. Pharm. Bull.* (Tokyo), **14**, 1162 (1966).

fusing ethyl *o*-acetamidohydroxy-3-coumarincarboxylate in the presence of phosphorus pentoxide.

In addition to the above result, the oxazole ring formation of ethyl 7-acetamido-8-hydroxy-3-coumarincarboxylate (II) was examined under using polyphosphoric acid (PPA). Thereby, II in PPA forms the oxazole ring in good yield after heating at between 100° and 110° for 5 hours. However, in the process of separating the product from the reaction solution, which is poured into an ice water after reaction, 2-methyl-8-oxo-8*H*-pyrano[3,2-*g*]benzoxazole-7-carboxylic acid(IV) is yielded by hydrolysis of ethyl 2-methyl-8-oxo-8*H*-pyrano[3,2-*g*]-benzoxazole-7-carboxylate (III) formed. Therefore, the reaction solution is poured into an ice water after reaction and then has to be neutralized with sodium carbonate at below 5°, in order to obtain only III. Both III and IV were identical with ethyl 2-methyl-8-oxo-8*H*-pyrano[3,2-*g*]benzoxazole-7-carboxylate[4] and 2-methyl-8-oxo-8*H*-pyrano[3,2-*g*]benzoxazole-7-carboxylic acid[5] on the admixed melting point test and comparison of the infrared absorption spectrum, separately.

In order to obtain 2-alkyl-8-oxo-8*H*-pyrano[3,2-*g*]benzoxazole series, additionally, ethyl 7-amino-8-hydroxy-3-coumarincarboxylate (I) was heated with acid anhydride at a low temperature for a short time to be led to ethyl 7-acylamino-8-hydroxy-3-coumarincarboxylate(II-type), which was lightly dissolved in diluted caustic alkalis and recovered from the alkaline solution by acidification, and ethyl 7-acylamino-8-hydroxy-3-coumarincarboxylate was fused with a small amount of phosphorus pentoxide. These obtained 2-alkyl-8-oxo-8*H*-pyrano[3,2-*g*]benzoxazole series(III-type) are insoluble in diluted caustic alkalis and show a characteristic peak at 1640 cm^{-1}(C=N) and no absorption band owing to N-H, O-H streching vibration.

Finally, ethyl 2-alkyl-8-oxo-8*H*-pyrano[3,2-*g*]benzoxazole-7-carboxylate was fused with 2-aminopyridine to be derived to N-(2-pyridyl)-2-alkyl-8-oxo-8*H*-pyrano[3,2-*g*]benzoxazole-7-carboxamide (V-type).

However, any prepared carboxamide series showed no tuberculostatic activity against *Mycobacterium tuberculosis* $H_{37}Rv$.

Experimental

Most of all product are listed in every items.

Ethyl 7-Formamido-8-hydroxy-3-coumarincarboxylate (II_a)——Ten grams of formic acid was refluxed with 2 g of I for 2 hr and then poured into an ice water. The resulting solid was collected by suction, washed with H_2O, dried and recrystallized from EtOH, giving II_a in 90% yield (Table I).

Ethyl 7-Acylamino-8-hydroxy-3-coumarincarboxylates (II_b, II_c, II_d and II_e)——A suspension of 1 g of I in 5 ml of acid anhydride was heated at 80—90° for 1 hr and then poured into an ice water. The resulting solid was collected by suction, washed with H_2O, dried and recrystallized, giving products (II_b, II_c, II_d and II_e) in about 80% yield (Table I).

Table I

Compound No.	mp (°C)	Appearance ( ): cryst. solvent	Formula	Analysis (%) Calcd. C	Calcd. H	Calcd. N	Found C	Found H	Found N
IIa	232 (decomp.)	yellow prisms (EtOH)	$C_{13}H_{11}O_6N$	56,31	3.97	5.05	55.98	4.03	5.12
IIb	230	light yellow needles (EtOH)	$C_{15}H_{15}O_6N$	59.01	4.91	4.59	59.12	5.18	4.53
IIc	206	light yellow needles (EtOH)	$C_{16}H_{17}O_6N$	60.18	5.33	4.38	60.21	5.46	4.07
IId	193	light yellow needles (EtOH)	$C_{16}H_{17}O_6N$	60.18	5.33	4.38	59.82	5.26	4.53
IIe	229	yellow plates (EtOH)	$C_{16}H_{15}O_6N$	60.56	4.73	4.41	60.48	4.87	4.07

5) H. Saikachi and M. Ichikawa, *Chem. Pharm. Bull.* (Tokyo), **14**, 1350 (1966).

Ethyl 8-Oxo-8*H*-pyrano[3,2-*g*]benzoxazole-7-carboxylate (III_a)——Two grams of II_a was fused till beginning to occur a yellow sublimating substance. After cooling, resulting solid was extracted with boiling benzene and then concentrated. Separated crystals were collected by suction and recrystallized from EtOH, giving III_a in 70% yield (Table II).

Ethyl 2-Alkyl-8-oxo-8*H*-pyrano[3,2-*g*]benzoxazole-7-carboxylates (III_b, III_c, III_d and III_e)——One gram of ethyl 7-acylamino-8-hydroxy-3-coumarincarboxylate was fused with a small amount of P_2O_5 at 190—200° for about 5 min.

After cooling, resulting solid was extracted with boiling benzene and concentrated. Separated crystals were collected by suction and recrystallized, giving products (III_b, III_c, III_d, and III_e) in 70% yield(Table II).

Table II

Compound No.	mp (°C)	Appearance ( ): cryst. solvent	IR ν_{max}^{KBr} cm^{-1}: $\nu_{C=N}$	Formula	Analysis (%) Calcd. C	H	N	Found C	H	N
IIIa	211	light yellow prisms (EtOH)	1640	$C_{13}H_9O_5N$	60.23	3.47	5.40	60.37	3.16	4.95
IIIb	173	light yellow needles (EtOH)	1640	$C_{15}H_{13}O_5N$	62.72	4.53	4.88	62.72	4.83	4.69
IIIc	136	light yellow needles (EtOH)	1635	$C_{16}H_{15}O_5N$	63.78	4.98	4.65	63.81	5.05	4.50
IIId	136	light yellow needles (EtOH)	1635	$C_{16}H_{15}O_5N$	63.78	4.98	4.65	63.70	5.03	4.72
IIIe	209	orange yellow plates (EtOH)	1635	$C_{16}H_{13}O_5N$	64.21	4.34	4.68	64.26	4.51	4.66

N-(2-Pyridyl)-2-alkyl-8-oxo-8*H*-pyrano[3,2-*g*]benzoxazole-7-carboxamides (V_a, V_b, V_c, V_d, and V_e)——One gram of ethyl 2-alkyl-8-oxo-8*H*-pyrano[3,2-*g*]benzoxazole-7-carboxylate obtained was generally fused with 1 g of 2-aminopyridine at 170—180° for 10 min. After cooling, resulting solid was treated with a small amount of EtOH. The insoluble substance was collected by suction and recrystallized, giving products (V_a, V_b, V_c, V_d, and V_e) in 70% yield (Table III).

N-(3-Pyridyl)-2-methyl-8-oxo-8*H*-pyrano[3,2-*g*]benzoxazole-7-carboxamide (VI) and N-(4-Pyridyl)-2-methyl-8-oxo-8*H*-pyrano[3,2-*g*]benzoxazole-7-carboxamide (VII)——One gram of III was fused with 3-aminopyridine at 190°—200° for 10 min. After cooling,resulting solid was treated with EtOH. The insoluble substance was collected by suction and recrystallized from AcOH, giving VI 80% yield. VII was yielded by fusing III with 4-aminopyridine in the same manner as the above synthetic method in 80% yield (Table III).

Table III

Compound No.	mp (°C)	Appearance ( ): cryst. solvent	Formula	Analysis (%) Calcd. C	H	N	Found C	H	N
Va	>300	yellow prisms (EtOH)	$C_{16}H_9O_4N_3$	62.54	2.93	13.68	62.60	2.91	13.92
Vb	245—246	light yellow needles (EtOH)	$C_{18}H_{13}O_4N_3$	64.47	3.88	12.53	64.53	4.17	12.44
Vc	258	light yellow needles (EtOH)	$C_{19}H_{15}O_4N_3$	65.32	4.29	12.03	65.13	4.55	12.32
Vd	268	yellow needles (EtOH)	$C_{19}H_{15}O_4N_3$	65.32	4.29	12.03	65.33	4.46	11.73
Ve	>300	light yellow prisms ($CHCl_3$)	$C_{19}H_{13}O_4N_3 \cdot \frac{1}{2}H_2O$	64.04	3.93	11.79	63.97	3.81	11.54
VI	>300	light yellow needles (AcOH)	$C_{17}H_{11}O_4N_3 \cdot \frac{1}{2}H_2O$	61.82	3.64	12.73	61.72	3.83	12.95
VII	>300	light yellow needles (AcOH)	$C_{17}H_{11}O_4N_3 \cdot \frac{1}{2}H_2O$	61.82	3.64	12.73	61.77	3.74	12.74

Ethyl 2-Methyl-8-oxo-8*H*-pyrano[3,2-*g*]benzoxazole-7-carboxylate (III)——After a mixture of 1 g of II and 10 ml of tetraphosphoric acid had been heated at 100°—110° for 5 hr, the sticky solution was poured into about 200 ml of an ice water. As soon as the product was separated, the pH of the solution was quickly adjusted to about 7 with Na_2CO_3 at below 5°. Separating crystals were collected by suction, washed with H_2O, dried and recrystallized from benzene to give the product (III) as pale yellow needles (0.7 g), mp 211°, which was identical with ethyl 2-methyl-8-oxo-8*H*-pyrano[3,2-*g*]benzoxazole-7-carboxylate on the admixed melting point test and comparison of the infrared spectrum.

2-Methyl-8-oxo-8*H*-pyrano[3,2-*g*]benzoxazole-7-carboxylic Acid (IV)——After a mixture of 1 g of II and 10 ml of tetraphosphoric acid had been heated at 100°—110° for 5 hr, the sticky solution was poured into about 100 ml of an ice water. After standing at between 5° and 10° for 24 hr, separating crystals were collected by suction, washed with H_2O, dried and recrystallized from EtOH to give the product (IV) as light yellow needles (0.7 g), mp 248° (decomp.), which was identical with 2-methyl-8-oxo-8*H*-pyrano[3,2-*g*]-benzoxazole-7-carboxylic acid on the admixed melting point test.

N-(2-Pyridyl)-2-methyl-8-oxo-8*H*-pyrano[3,2-*g*]benzoxazole-7-carboxamide (V)——After a mixture of 1 g of IV and 20 ml of $SOCl_2$ had been heated under reflux for 1 hr, the excess of $SOCl_2$ was removed *in vacuo*. The residue was suspended in 30 ml dried benzene and then 20 ml of 5% 2-aminopyridine benzene solution was gradually added to the suepension at below 10°. After standing at room temperature for 24 hr, benzene was evaporated *in vacuo* and the residue was treated with 2% aq. AcOH solution in an ice bath. The resulting solid was collected by suction, washed with H_2O,dried and recrystallized from AcOH to give the product (V) as light yellow needles (1 g), mp>300°. *Anal.* Calcd. for $C_{17}H_{16}O_4N_3$: C, 63.55; H, 3.42; N, 13.08. Found: C, 63.24; H, 3.51; N, 12.87.

Acknowledgement The authors wish to express their gratituder to Dr. M. Shimizu, Dr. Y. Oshima and Mr. S. Nagasaki Daiichi Seiyaku Co., Ltd. for collaboration in microbiological test. The microanalyses were performed by Miss Ogata and the measurement of infrared spectra were carried out by Miss Sato of the Central Analysis Room of this Faculty to whom we are also grateful.

[Chem. Pharm. Bull. 17(9)1958—1962(1969)]

UDC 547.964.4.07

Studies on Peptides. XXIV.[1,2] Some Observation on the Urethan Formation during the Mixed Anhydride Procedure in Peptide Synthesis

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(Received March 12, 1969)

Control of racemization during the mixed anhydride procedure[4–6] in peptide synthesis has been studied by Applewhite, *et al.*[7] and more recently by Anderson, *et al.*,[8,9] especially in the case of acylpeptide anhydrides. Usefulness of this rapid peptide-forming reaction was thus further evaluated. However, the urethane formation, a possible side reaction of this procedure seems to be a limitation in some instances for the use of this method.

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