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Studies on the Syntheses of N-Heterocyclic Compounds. III.[1] Hypocholesterolemic 1,2,4-Oxadiazole Derivatives (1)

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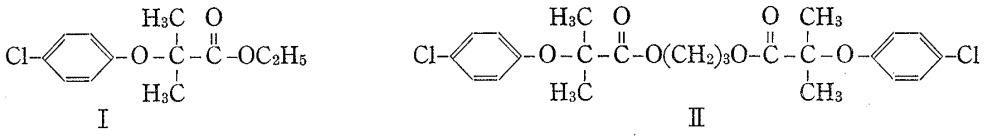
3,5-Disubstituted-1,2,4-oxadiazole derivatives containing aryl and heteryl substituent at 3 position and amino, mercapto, aryl and heteryl substituent at 5 position have been synthesized. Among these compounds, 3-[4-(1-ethoxycarbonyl-1-methylethoxy)phenyl]-3-(3-pyridyl)-1,2,4-oxadiazole exhibited a considerable hypocholesterolemic activity.

There has been a great demand for the development of a potent agent which reduces serum cholesterol as well as β-lipoprotein, since most of the cardiovascular diseases have been proved to be closely related with arteriosclerosis.[3] For the purpose of reducing endogeneous cholesterol, which has been supposed to be the predominant cause of hypercholesterolemia, a number of drugs which inhibits the cholesterol biosynthesis has been reported,[3,4-7] among them are ethyl *p*-chlorophenoxyisobutylate (clofibrate) (I)[8] and 1,3-propyl-bis-(2-*p*-chlorophenoxy-2-methylpropanoate) (simfibrate) (II).[9] On the other hand, it has been reported



I II

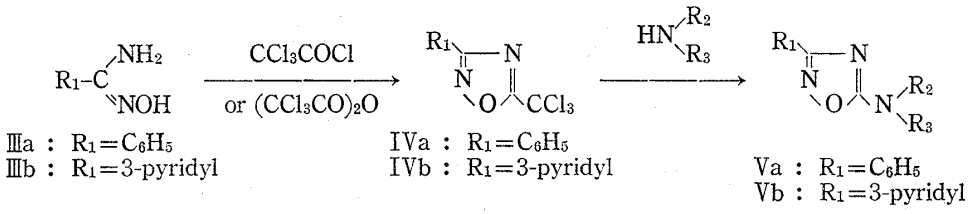
Chart 1

that some of the isoxazole and pyrazole derivatives[10] show favorable effects to lipid metabolism, in addition to significant hypoglycemic activity.

This result led us to an investigation of 1,2,4-oxadiazole derivatives, which bears structural resemblance to isoxazole and pyrazole, anticipating hypoglycemic or hypocholesterolemic activity. The present paper is concerned with the chemical modification of the 3 and 5 positions of 1,2,4-oxadiazole leading to a clofibrate-like compound.

1) Part II: S. Yurugi, M. Hieda, T. Fushimi, and M. Tomimoto, *Chem. Pharm. Bull.* (Tokyo), **19**, 2354 (1971).
2) Location: *Juso, Higashiyodogawa-ku, Osaka.*
3) D. Kritchevsky, *Federation Proceeding*, **30**, 835 (1971).
4) G.R. Griot, S. African Patent 67-04612 (1969) [*C.A.*, **71**, 101723 (1969)].
5) *a*) M.M. Best and C.H. Duncan, *J. Atherosclerosis, Res.*, **10**, 103 (1969); *b*) G. Hartmann and G. Forster, *ibid.*, **10**, 253 (1969).
6) Sandoz-Wander, U.S. Patent 3558626, 3558645 (1971).
7) K. Nakamura, Japan. Patent 41540 (1971).
8) *a*) J.M. Throp and W.S. Waring, *Nature*, **194**, 948 (1962); *b*) D.R. Avog, E.A. Swyryd, and R.G. Gould, *J. Lipid Res.*, **6**, 369 (1965); *c*) D.L. Azarvoff, D.R. Twker, and G.A. Barr, *Metabolism*, **14**, 959 (1965).
9) M. Nakanishi, T. Kobayakawa, T. Okada, and T. Tsumagari, *Yakugaku Zasshi*, **90**, 921 (1970).
10) *a*) W.E. Dulin and G.C. Gerritsen, *Proc. Soc. Exptl. Biol. Med.*, **113**, 683 (1963); *b*) G.C. Derritsen and W.E. Dulin, *Diabetes*, **14**, 507 (1965); *c*) W.E. Dulin, G.H. Lund, and G.C. Garritsen, *Proc. Soc. Exptl. Biol. Med.*, **118**, 449 (1965).

Employing the method of Elog,[11] several new 3-phenyl-5-hydroxyalkylamino-1,2,4-oxadiazoles (Va) were synthesized by the reaction of 3-phenyl-5-trichloromethyl-1,2,4-oxadiazole (IVa), derived from benzamidoxime and trichloroacetyl chloride, with alkylamines. This procedure was further applied to the synthesis of 3-(3-pyridyl)analogue (Vb) starting from 3-pyridylamidoxime (IIIb)[12] by way of 3-(3-pyridyl)-5-trichloromethyl-1,2,4-oxadiazole (IVb). The yield and analytical data of the obtained compounds (Va, b) are summarized in Table I.



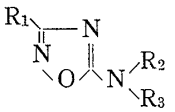
IIIa : $R_1=C_6H_5$
IIIb : R_1=3-pyridyl

IVa : $R_1=C_6H_5$
IVb : R_1=3-pyridyl

Va : $R_1=C_6H_5$
Vb : R_1=3-pyridyl

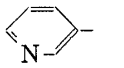
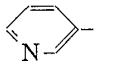
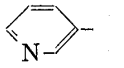
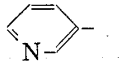
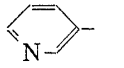
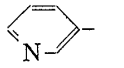
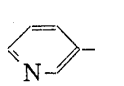
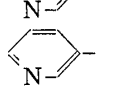
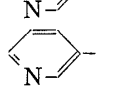
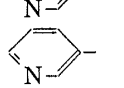
Chart 2

TABLE I. 3-Phenyl- and 3-(3-Pyridyl)-5-substituted Amino-1,2,4-oxadiazoles (V)

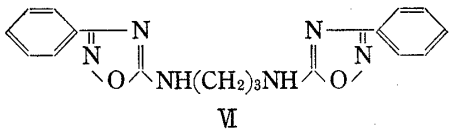


V	R_1	$N<^{R_2}_{R_3}$	Yield (%)	mp (°C)	Formula	Analysis (%) Calcd. C	H	N	Found C	H	N
1	C_6H_5	$NH(CH_2)_3OH$	44	78—79	$C_{11}H_{13}O_2N_3$	60.26	5.98	19.17	60.02	6.00	19.05
2	C_6H_5	$NHCH_2\underset{CH_3}{\underset{\vert}{C}}HOH$	30	93—95	$C_{11}H_{13}O_2N_3$	60.26	5.98	19.17	60.26	6.05	19.10
3	C_6H_5	$NH\underset{CH_3}{\underset{\vert}{C}}HCH_2OH$	23	67—70	$C_{11}H_{13}O_2N_3$	60.26	5.98	19.17	60.43	5.96	18.98
4		NH_2	94	261—263	$C_7H_6ON_4$	51.85	3.73	34.55	51.47	3.62	33.50
5		$NHC<^{NH}_{NH_2}$	56	259—261	$C_8H_8ON_6$	47.05	3.95	41.16	47.10	3.89	41.36
6		$NHNHC<^{NH}_{NH_2}$	79	190—193	$C_8H_9ON_7$	40.51	4.67	41.17	40.46	4.44	39.89
7		$NHC<^{NH_2}_{N—C<^{NH_2}_{NH}}$	33	236—238	$C_9H_{10}ON_8$	41.22	3.84	42.34	42.72	4.22	42.32
8		$NHC<^{NH_2}_{NCONH_2}$	35	200	$C_9H_9O_2N_7$	38.16	3.92	34.62	38.45	3.88	36.84
9		$NH(CH_2)_2OH$	30	128—129	$C_9H_{10}O_2N_4$	52.41	4.89	27.17	52.60	4.83	26.96
10		$N(CH_2CH_2OH)_2$	40	99—101	$C_{11}H_{14}O_3N_4$	49.25	6.00	20.70	49.02	5.88	20.75
11		$NHCH_2\underset{CH_3}{\underset{\vert}{C}}HOH$	53	115—118	$C_{10}H_{12}O_2N_4$	54.53	5.49	25.44	54.56	5.36	25.91
12		$NH(CH_2)_3OH$	35	121—122	$C_{10}H_{12}O_2N_4$	54.35	5.49	25.44	54.68	5.25	25.24

11) F. Elog and R. Lenaers, *Helv. Chim. Acta*, **49**, 1430 (1966).
12) L. Michaelis, *Chem. Ber.*, **24**, 3441 (1891).

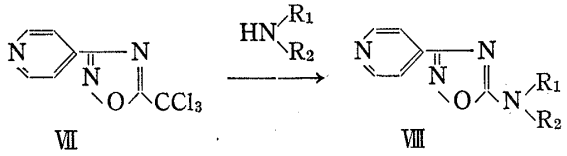
V	R_1	$N\langle^{R_2}_{R_3}$	Yield (%)	mp (°C)	Formula	Analysis (%) Calcd. C	H	N	Found C	H	N
13		$N(CH_3)(CH_2)_2OH$	26	101—102	$C_{10}H_{12}O_2N_4$	54.53	5.49	25.44	54.30	5.38	25.00
14			81	80—81	$C_{11}H_{12}ON_4$	61.09	5.60	25.91	61.38	5.50	25.78
15			51	75—76	$C_{12}H_{14}ON_4$	62.59	6.17	24.33	61.74	6.06	24.39
16			10	146—147	$C_{11}H_{12}O_2N_4$	56.89	5.21	24.13	56.56	5.17	24.12
17		$NH(CH_2)_2NHCOCH_3$	27	181—183	$C_{11}H_{13}O_2N_5$	53.43	5.30	28.33	53.68	5.29	28.33
18		$NH(CH_2)_3NHCOCH_3$	36	128—130	$C_{12}H_{15}O_2N_5$	55.17	5.75	26.82	55.03	5.64	26.67
19		$NHCH_2CH(CH_3)NHCOCH_3$	49	180—182	$C_{12}H_{15}O_2N_5$	55.17	5.75	26.82	55.25	5.71	26.75
20		$NH(CH_2)_2SCH_3$	75	85—87	$C_{10}H_{12}ON_4S$	50.83	5.12	23.71	50.71	5.23	24.00
21		$NH(CH_2)_2SOCH_3$	75	120—122	$C_{10}H_{12}O_2N_4S$	47.60	4.80	22.21	47.79	4.76	22.52
22		$NH(CH_2)_2SO_2CH_3$	59	137—139	$C_{19}H_{12}O_3N_4S$	44.76	4.51	20.88	44.42	4.38	20.81

In the reaction of IV with ammonia and guanidine derivatives, methanol was used as the solvent, while in other cases the reactions were carried out without solvent. When IVa was reacted with 1,3-diaminopropane, bis-substituted compound (VI) was afforded along with Va. The reaction of IVb with ethylene diamine and 1,2-diaminopropane, however,



VI

Chart 3



VII VIII

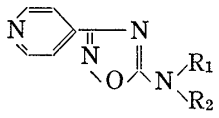
Chart 4

afforded only monosubstituted compound (Vb). The structure of the latter product was proved to be 3-(3-pyridyl)-5-(2-aminopropylamino)-1,2,4-oxadiazole by the fact that the diazoation of the product gave 3-(3-pyridyl)-5-(2-hydroxypropylamino)-1,2,4-oxadiazole (**12**), which was identified with an authentic sample prepared by the reaction of IVb with isopropanolamine. The obtained 5-aminoalkylamino-1,2,4-oxadiazoles were acetylated to give corresponding acetylamino compound (Table I, **17**, **18**, **19**). The methylsulfinyl (**21**) and methylsulfonyl (**22**) derivatives were prepared by the oxydation of the methylthio derivative (**20**). Three 3-(4-pyridyl) derivatives (VIII) were further synthesized by the same procedure with the 3-pyridyl derivative, which are listed in Table II.

Though, none of the synthesized compounds showed hypoglycemic activity, some of the 3-pyridyl derivatives were found to possess hypocholesterolemic activity (A-action)[13]

13) A-action: Hypocholesterolemic activity in hypercholesterolemic rats.

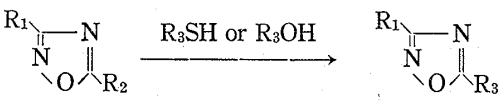
Table II. 3-(4-Pyridyl)-5-substituted Amino-1,2,4-oxaliazoles (VIII)



VIII	$N\langle^{R_1}_{R_2}$	Yield (%)	mp (°C)	Formula	Analysis (%) Calcd. C	H	N	Found C	H	N
23	$NH(CH_2)_2N(C_2H_5)_2$	54	198—199	$C_{13}H_{19}ON_5 \cdot 2(COOH)_2$	46.26	5.22	15.87	46.14	5.14	16.06
24	$NH(CH_2)_3N(C_2H_5)_2$	58	140—143	$C_{14}H_{21}ON_5 \cdot 2(COOH)_2$	47.47	5.49	15.38	47.40	5.47	15.43
25	$NH(CH_2)_3N(CH_3)_2$	93	76—79	$C_{12}H_{17}ON_5$	58.30	6.88	28.34	58.56	6.88	28.89

as shown in Table V. With respect to B-action[14] however, those compounds were negative, showing somewhat hypercholesterolemic tendency.

Subsequently introduction of functional groups containing oxygen or sulfur atom instead of NH-group to the 5-position was undertaken, fixing the 3-position to phenyl and 3-pyridyl. As for the phenyl derivatives five compounds were obtained starting from 3-phenyl-5-chloro-1,2,4-oxadiazole (IX).[15] The reaction of IX with sodium ethoxide and sodium benzyloxide gave **27** and **28**, respectively. The reaction with ethyl α-mercapto-α-substituted acetates in the presence of sodium ethoxide afforded **28**, **29** and **30**, whereas the reaction of IX with ethyl α-hydroxy-α-substituted acetate which are less nucleophilie than the mercapto compound failed to react under the same condition. The synthesis of the 3-(3-pyridyl) derivatives were carried out by the reaction of IVa with the above alcohols and mercaptans in the presence of sodium ethoxide, but most of the reactions failed to give the desired compound, yielding only 3-(3-pyridyl)-5-ethoxy-1,2,4-oxadiazole (**31**). Another four compounds **32**, **33**, **34** and **35** were prepared by the reactions of 5-chloromethyl derivatives (X)[16] and (XI),[17] with ethyl α-mercapto-α-substituted acetates. In this case also, ethyl α-hydroxy-α-substituted acetates failed to react with X and XI. The obtained compounds and their biological activities were listed in Table III and V.



IX: $R_1=C_6H_5$, $R_2=Cl$
X: $R_1=C_6H_5$, $R_2=CH_2Cl$
XI: R_1=3-pyridyl, $R_2=CH_2Cl$

XII

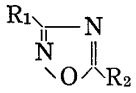
Chart 5

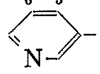
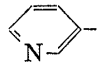
From the above result it became clear that the compounds **29** and **30** possessed considerable hypocholesterolemic activities which were positive in both the A and B effects in rat. However those compounds showed anorectic effect to rat possibly because of the smell of the mercaptan. The result also indicated that 3-(3-pyridyl) derivatives possess almost the same activity with 3-phenyl derivatives.

Subsequently chemical modifications of the phenyl group were undertaken by introducing various substituents at the *para* position. Thus, starting from 4-hydroxybenzonitrile (XIII),[18] 3-(4-alkoxyphenyl)-1,2,4-oxadiazole derivatives (XVI) were synthesized *via* 4-alkoxybenzonitrile (XIV) and amidoximes (XV). Among the obtained derivatives, compounds **37**, **38**, **39** and **42** showed considerable hypocholesterolemic activities (Table IV, V). It has been

14) B-action: Hypocholesterolemic activity in normocholesterolemic rats.
15) K. Fujita, T. Fujie, and A. Ide, *Yakugaku Zasshi*, **84**, 1061 (1964).
16) C. Palazzo, M. Tanella, G. Strani, and B. Silvestrine, *J. Med. Chem.*, **4**, 351 (1961).
17) Lab. Toraude, Dutch. Patent 6611571 (1967) [*C.A.*, **67**, 64402 (1967)].
18) *a*) J.N. Ashley, H.J. Burber, A.J. Ewis, G. Newberg, and A.D.H. Shelf, *J. Chem. Soc.*, **1942**, 103; *b*) A. Findlay and C.S. Tang, *Can. J. Chem.*, **45**, 1014 (1967).

TABLE III. 1,2,4-Oxadiazole Derivatives (XII)



XII	R_1	R_2	Yield (%)	mp (°C)	Formula	Analysis (%) Calcd. C	H	N	Found C	H	N
26	C_6H_5	OC_2H_5	84	oil	$C_{10}H_{10}O_2N_2$	63.14	5.30	14.73	63.10	4.91	14.64
27	C_6H_5	$OCH_2C_6H_5$	52	59	$C_{15}H_{12}O_2N_2$	71.41	4.80	11.11	71.69	4.96	11.06
28	C_6H_5	$SCH_2CO_2C_2H_5$	53	59	$C_{12}H_{12}O_3N_2S$	53.53	4.58	10.60	54.26	4.41	10.48
29	C_6H_5	$SCHCO_2C_2H_5$ (CH_3)	83	oil	$C_{13}H_{14}O_3N_2S$	56.10	5.07	10.07	56.26	4.95	9.82
30	C_6H_5	$SC(CH_3)_2CO_2C_2H_5$	51	oil	$C_{14}H_{16}O_3N_2S$	57.51	5.52	9.58	57.78	5.57	9.63
31		OC_2H_5	75	40—42	$C_9H_9O_2N_3$	56.54	4.74	21.98	56.10	4.82	21.63
32	C_6H_5	$CH_2SCH_2CO_2C_2H_5$	84	oil	$C_{13}H_{14}O_3N_2S$	56.10	5.07	10.07	55.88	5.11	9.83
33	C_6H_5	$CH_2SC(CH_3)_2CO_2C_2H_5$	81	oil	$C_{15}H_{18}O_3N_2S$	58.81	5.92	9.15	58.52	5.98	9.21
34		$CH_2SCH_2CO_2C_2H_5$	70	97—98[a]	$C_{12}H_{13}O_3N_2S$	45.53	4.09	11.37	45.48	4.08	11.44
35		$CH_2SC(CH_3)_2CO_2C_2H_5$	50	57—60[a]	$C_{14}H_{17}O_3N_3S$	48.36	4.82	10.58	48.11	4.75	10.84

a) mono oxalate

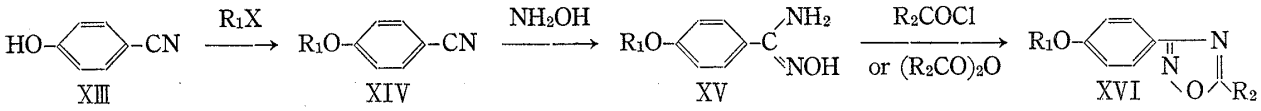
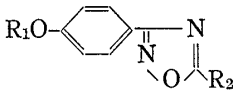
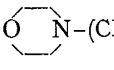


Chart 6

TABLE IV. 3-(4-Substituted alkoxy)-phenyl-5-substituted-1,2,4-oxadiazoles (XVI)



XVI	R_1	R_2	Yield (%)	mp (°C)	Formula	Analysis (%) Calcd. C	H	N	Found C	H	N
36	$(C_2H_5)_2N(CH_2)_2$	CH_3	51	175—176	$C_{15}H_{21}O_2N_3 \cdot HCl$	57.78	7.11	13.33	57.57	7.11	13.48
37	$(C_2H_5)_2N(CH_2)_2$	C_6H_5	56	156—158	$C_{20}H_{23}O_2N_3 \cdot (COOH)_2$	61.81	5.90	9.83	61.97	5.72	9.80
38	$(C_2H_5)_2N(CH_2)_2$		66	42—43	$C_{19}H_{22}O_2N_4 \cdot H_2O$	64.02	6.79	15.72	63.82	6.37	15.72
39	$(C_2H_5)_2N(CH_2)_2$		39	86—87	$C_{19}H_{22}O_2N_4 \cdot H_2O$	64.02	6.79	15.72	64.26	6.15	15.50
40			36[a]	119—120	$C_{19}H_{20}O_3N_4$	64.76	5.72	15.90	64.55	5.73	16.18
41	CH_3CH_2		53	134—135	$C_{15}H_{13}O_2N_3$	67.40	4.90	15.72	67.15	5.14	15.55
42	$C_2H_5OCOC(CH_3)_2$		70	75—77	$C_{19}H_{19}O_4N_3$	64.58	5.42	11.89	64.43	4.92	11.95

a) Yield was based on 4-(2-morpholino)ethoxy benzonitrile.

demonstrated that compounds **37**, **38** and **39** exhibit hypocholesterolemic activity by blocking the enzymatic reduction of desmosterol to cholesterol just in the same manner as triparanol.[19] Thus, by determining the pattern of plasma sterols by gas chromatography, and unusual accumulation of desmosterol was observed when those compounds were administered to rat. (Fig. 1). However, compound **42**, which involves the component of clofibrate, 1-ethoxycarbonyl-1-methylethoxy group, showed no accumulation of desmosterol. It became clear by our experiment[20] that this compound inhibits cholesterol biosynthesis at the stage of transformation of acetate into mevalonate similarly to clofibrate.[8a] Since compound **41**, which has a slight hypocholesterolemic activity, also shows no accumulation of desmosterol, it is assumed that the property to accumulate desmosterol might have some connection with the tertiary amino group in the alkoxy side chain.

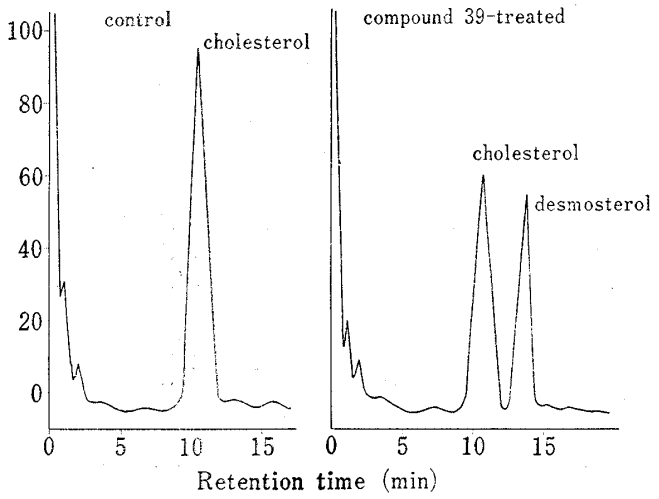
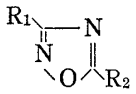
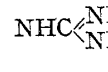
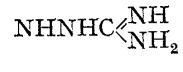
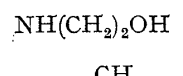
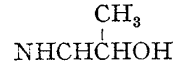
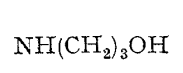
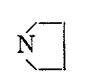
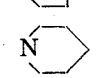
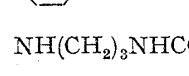


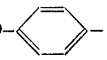
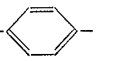
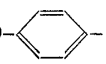
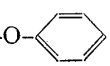
Fig. 1. Gas Chromatogram of Rat Plasma Sterol

Table V. Hypocholesterolemic Activity of 1,2,4-Oxadiazoles in Rat



No.	R_1	R_2	Efficacy[a] Plasma total cholesterol A	Plasma total cholesterol B	Liver total cholesterol A	Liver total cholesterol B
5			−		+ ↓	
6			+ ↓		+ ↓	
9			## ↓		−	
11			−		+ ↓	
12			## ↓	+ ↑	+ ↓	+ ↑
14			## ↑		−	
15			## ↓		+ ↓	
18			## ↑		+ ↑	
27	C_6H_5	$OCH_2C_6H_5$	+ ↑	−	+ ↓	−

19) H.Y.C. Wong, H.E. Vroman, and H.C. Mendez, *Life Sciences*, **5**, 629 (1966).
20) Y. Imai and K. Shimamoto, *Atherosclerosis*, **17**, 121, 131 (1973).

No.	R_1	R_2	Efficacy[a] Plasma total cholesterol A	Plasma total cholesterol B	Liver total cholesterol A	Liver total cholesterol B
28	C_6H_5	$SCH_2CO_2C_2H_5$	−	+ ↓	−	+ ↑
29	C_6H_5	$SCHCO_2C_2H_5$ (CH$_3$)	‡ ↓	‡ ↓	+ ↑	−
30	C_6H_5	$SC(CH_3)_2CO_2C_2H_5$	‡ ↓	‡ ↓	+ ↑	−
31		OC_2H_5	+ ↑	+ ↓	+ ↓	−
32	C_6H_5	$CH_2SCH_2CO_2C_2H_5$	−	+ ↓	−	+ ↑
33	C_6H_5	$CH_2SC(CH_3)_2CO_2C_2H_5$	‡ ↓	+ ↓	‡ ↑	−
34		$CH_2SCH_2CO_2C_2H_5$	−	+ ↑	+ ↑	−
35		$CH_2SC(CH_3)_2CO_2C_2H_5$	+ ↑	+ ↓	+ ↑	−
36	$(C_2H_5)_2N(CH_2)_2$–O– 	CH_3	+ ↓	‡ ↓	+ ↓	−
37	$(C_2H_5)_2N(CH_2)_2$–O– 	C_6H_5	+ ↓	‡ ↓	‡ ↓	−
38	$(C_2H_5)_2N(CH_2)_2$–O– 		‡ ↓	‡ ↓	+ ↓	‡ ↓
39	$(C_2H_5)_2N(CH_2)_2$–O– 		−	‡ ↓	‡ ↓	‡ ↓
40	 N–$(CH_2)_2$–O– 		+ ↓	+ ↓	+ ↓	+ ↓
41	C_2H_5O– 		+ ↓	+ ↓	+ ↓	+ ↓
42	$C_2H_5OCOC(CH_3)_2$–O– 		‡ ↓	‡ ↓	+ ↓	+ ↓

a) A: (−) <10%, (+) 10—30%, (++) 30—50%, (+++)> 50%
B: (−) < 5%, (+) 5—15%, (++) 15—30%, (+++)> 30%
↑: hypercholesterolemic activity
↓: hypocholesterolemic activity

Experimental

3-Phenyl-5-trichloromethyl-1,2,4-oxadiazole (IVa)[11]——To a solution of benzamidoxime (IIIa) (54 g), C_6H_6 (200 ml) and pyridine (100 ml) was added trichloroacetyl chloride (75 g) cooled in an ice bath. The reaction mixture was refluxed for 1.5 hr and evaporated *in vacuo*. H_2O was added to the residue and the separated material was extracted with ether. The extract was washed with H_2O, dried over anhydrous Na_2SO_4, and evaporated to give 70 g (67%) of IVa.

3-(3-Pyridyl)-5-trichloromethyl-1,2,4-oxadiazole (IVb)——A) To a suspension of 3-pyridylamidoxime (IIIb)[12] (31.7 g) in C_6H_6 (250 ml) was added dropwise trichloroacetic anhydride (100 g). The mixture was refluxed for 3 hr and evaporated *in vacuo*. H_2O was added to the residue, and the solution was neutralized with Na_2CO_3 and extracted with ether. The extract was dried over anhydrous Na_2SO_4 and evaporated *in vacuo* to give 47 g (74%) of IVb as an orange-colored oil, which was used for the subsequent reaction without further purification.

B) To a suspension of IIIb (53 g) in C_6H_6 (300 ml) were added dropwise pyridine (50 ml) and trichloroacetyl chloride (75 g). After an exothermic reaction, the mixture was separated into two layers. After cooling, H_2O was added to mixture, the benzene layer was separated, Na_2CO_3 (20 g) was added to the aqueous layer and the separated oily product was extracted with ether. The extract was combined with the benzene layer and dried over anhydrous $MgSO_4$. After evaporation *in vacuo* the residue was purified by column chromatography on silica gel eluted with C_6H_6–acetone (10:1) to afford 56 g (50%) of IVb as yellow needles, mp 50°.

3-Phenyl-5-hydroxyalkylamino-1,2,4-oxadiazoles (Va)——General Procedure: The mixture of IVa and an excess of an amine was heated at 60—120°. The reaction mixture was poured into H_2O and extracted with ether. The extract was washed with H_2O and dried over anhydrous $MgSO_4$, evaporated *in vacuo* to give crude crystals, which were recrystallized from C_6H_6 or petroleum ether to give the pure product.

3-(3-Pyridyl)-5-substituted Amino-1,2,4-oxadiazoles (Vb)——General Procedure a): To a solution of IVb in MeOH was added an excess of an amine. The precipitate was filtered and recrystallized from an appropriate solvent to give Vb.

General Procedure b): IVb and an excess of an amine were mixed to effect an immediate exothermic reaction. The completion of the reaction was checked by thin-layer chromatography (TLC), and the excess of amine was removed *in vacuo*. The residue was washed with ether and recrystallized from EtOH to give Vb.

3-(3-Pyridyl)-5-(acetoaminoalkylamino)-1,2,4-oxadiazole (Vb)——General Procedure: IVb and an excess of diamine were mixed to effect an immediate exothermic reaction. After the excess of diamine was removed *in vacuo*, Ac_2O was added to the residue. The resulting precipitate was filtered and recrystallized from EtOH.

1,3-Diamino-bis-(3-phenyl-1,2,4-oxadiazole)-propane (VI)——1,3-Diaminopropane (2 ml) was added to 3-phenyl-5-chloro-1,2,4-oxadiazole (IX)[15] (1.0 g) at 0° and left at room temperature for 4 hr. H_2O was added to the mixture and extracted with AcOEt. The extract was evaporated *in vacuo* and the residue was purified by column chromatography on silica gel to give 0.2 g (10%) of VI, mp 158—159°. *Anal.* Calcd. for $C_{19}H_{18}O_2N_6$: C, 62.97; H, 5.01; N, 23.19. Found: C, 63.28; H, 4.91; N, 22.87.

3-(3-Pyridyl)-5-(2-hydroxypropylamino)-1,2,4-oxadiazole (11)——A) IVb (8 g) was treated with isopropanolamine (4 ml) and left at room temperature overnight. H_2O was added to the mixture and extracted with ether. The aqueous layer was evaporated *in vacuo*, and the residue was dissolved in hot EtOH. The filtration of the resulting crystals and the recrystallization gave 4 g (53%) of **11**.

B) IVb (0.66 g) was treated with 1,2-propanediamine (0.5 g) and warmed at 70° for 1 hr. The mixture was evaporated *in vacuo* to dryness. The residue was dissolved in 10 ml of H_2O. To the solution were added 10% HCl (2 ml) and a solution of $NaNO_2$ (0.5 g in H_2O 2 ml) with stirring at 0° over 2 hr. After the resulting diazonium salt was decomposed by gentle heating, the solution was made alkaline with K_2CO_3 and evaporated *in vacuo*. The residue was extracted with EtOH and the extract was evaporated. The residue was purified by column chromatography on silica gel to give crystals, which showed complete identity with an authentic sample obtained in the procedure A in mixed melting point and infrared (IR) spectrum.

3-(3-Pyridyl)-5-(2-methanesulfinylethylamino)-1,2,4-oxadiazole (21)——To a solution of 3-(3-pyridyl)-5-(2-methanethiolethylamino)-1,2,4-oxadiazole (**20**) (1.2 g) in AcOH (20 ml) was added 30% H_2O_2 (0.55 g) at 0° and the solution was allowed to stand at room temperature overnight. After the excess of H_2O_2 decomposed with Na_2SO_3, the solution was treated with charcoal and filtered. The filtrate was evaporated *in vacuo* to give 1 g (75%) of **21**, mp 120—122°.

3-(3-Pyridyl)-5-(2-methanesulfonylethylamino)-1,2,4-oxadiazole (22)——To a solution of **20** (1.2 g) in AcOH (20 ml) was added 30% H_2O_2 (1.1 g) at 0° with stirring and the solution was allowed to stand overnight at room temperature. After heating at 55—60° for 2 hr, the mixture was concentrated to 30 ml and purified by column chromatography on silica gel to give 0.8 g (59%) of **22**, mp 137—139°.

3-(4-Pyridyl)-5-trichloromethyl-1,2,4-oxadiazole (VII)——To a solution of 4-pyridylamidoxime[21] (6.9 g) in C_6H_6 (200 ml) was added dropwise trichloroacetic anhydride (18.5 g) at 0° and then refluxed for 3 hr. The precipitate was filtered, the filtrate was washed with H_2O, dried over anhydrous Na_2SO_4, and evaporated *in vacuo* to give 9 g (42%) of crude VII, which was recrystallized from C_6H_6 to give 7.2 g (34%) of VII, mp 113—114°. *Anal.* Calcd. for $C_8H_4OCl_3N_3 \cdot CCl_3COOH$: C, 28.04; H, 1.17; N, 9.81. Found: C, 28.18; H, 1.15; N, 9.72.

3-(4-Pyridyl)-5-substituted Amino-1,2,4-oxadiazoles (VIII)——General Procedure: VII was boiled in EtOH with an excess of an amine for 3 hr. The reaction mixture was evaporated *in vacuo* and the residue was purified by recrystallization or column chromatography on silica gel.

3-Phenyl-5-ethoxy-1,2,4-oxadiazole (26)——A solution of IX (0.9 g) in EtOH (3 ml) was added dropwise to EtONa solution, prepared from 0.25 g of Na and 150 ml of EtOH at 0°, and heated on water bath for 2 hr. After EtOH was evaporated, H_2O (5 ml) was added to the residue and the separated oil was extracted with $CHCl_3$. The extract was evaporated *in vacuo* to give 0.8 g (84%) of **26** as colorless oil.

3-Phenyl-5-benzyloxy-1,2,4-oxadiazole (27)——Na (0.23 g) was dissolved in benzylalcohol (10 ml) with heating. To the solution was added dropwise a mixture of IX (1.8 g) and benzylalcohol (5 ml) and heated on water bath for 1 hr. The cooled reaction mixture was poured into H_2O (450 ml), the separated oil was extracted with petroleum ether. The extract was evaporated *in vacuo* and the residue was purified by column chromatography on silica gel eluted with C_6H_6–acetone (9:1) to afford 1.3 g (52%) of **27**, mp 59°.

21) G. Leandri, L. Maioli, and L. Ruzzier, *Boll. Sci. fac. chim. Ind. Bolagna*, **15**, 57 (1957) [*C.A.*, **52**, 11051 (1958)].

3-(3-Pyridyl)-5-ethoxy-1,2,4-oxadiazole (31)——A solution of IVb (6.5 g) in EtOH (10 ml) was added dropwise to EtONa solution, prepared from 0.3 g of Na and 20 ml of EtOH, at 0° and heated on water bath for 20 min. After EtOH was evaporated, H_2O was added to the residue and the separated oil was extracted with $CHCl_3$. The extract was washed with H_2O, dried over anhydrous Na_2SO_4, evaporated *in vacuo*, and purified by column chromatography on silica gel eluted with C_6H_6–acetone (4:1) to give 3.5 g (75%) of **31**, mp 40—42.

3-Phenyl-5-ethoxycarbonylmethylthio-1,2,4-oxadiazoles (28, 29 and 30)——General Procedure: Ethoxy α-mercapto-α-substituted acetate (0.01 mole) was added dropwise to the solution of EtONa, prepared from 2.3 g of Na and 23 ml of EtOH. The solution was added dropwise to the solution of IX (1.9 g) in EtOH (5 ml) at 0° and allowed to stand at room temperature for 2—5 hr. After the removal of EtOH by evaporation, H_2O was added to the residue and separated oil was extracted with AcOEt. The extract was washed with H_2O, dried over anhydrous Na_2SO_4, and evaporated *in vacuo*. The residue was purified by column chromatography on silica gel, eluted with C_6H_6 to afford **28**, **29** or **30**.

3-Phenyl- and 3-(3-Pyridyl)-5-(2-ethoxycarbonylmethylthio)-methyl-1,2,4-oxadiazoles (32, 33, 34 and 35)——General Procedure: Ethyl α-mercapto-α-substituted acetate (0.012 mole) was added dropwise to the solution of EtONa, prepared from 0.23 g of Na and 10 ml of EtOH. The solution was added to the solution of 3-phenyl- (X)[16] and 3-(3-pyridyl)-5-chloromethyl-1,2,4-oxadiazole (XI)[17] (0.01 mole) in EtOH (18 ml) and allowed to stand at room temperature for 2 hr. After the removal of EtOH by evaporation, H_2O (5 ml) was added to the residue and the separated oil was extracted with AcOEt. The extract was washed with H_2O, dried over anhydrous Na_2SO_4, and evaporated *in vacuo*. The residue was purified by column chromatography on silica gel eluted with C_6H_6 to afford **32**, **33**, **34** or **35**. Compounds 34 and 35 were crystallized as oxalate.

4-Alkoxybenzonitriles (XIV)——General Procedure: To a mixture of *p*-cyanophenol (0.1 mole) (XIII)[15] and EtONa, prepared from 0.12 mole of Na and 150 ml of EtOH, was added an alkylhalide (0.2 mole). The mixture was refluxed for 0.5—72 hr. After the removal of EtOH by evaporation *in vacuo*, the residue was poured into H_2O and extracted with ether. The extract was washed with H_2O, dried over anhydrous Na_2SO_4, and evaporated to give the crude XIV, which was purified by vacuum distillation or recrystallization. The obtained compounds are listed in Table VI. 4-Ethoxybenzonitrile[19a] and 4-(1-ethoxycarbonyl-1-methylethoxy)-benzonitrile[22] were prepared by the reported method.

Table VI. 4-Alkoxybenzonitrile (XIV)

RO-  -CN

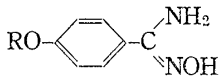
R	Yield (%)	mp/bp (°C)	Formula	Analysis % Calcd. C	H	N	Found C	H	N
$(C_2H_5)_2N(CH_2)_2$	81	144—146/0.5	$C_{13}H_{18}ON_2$	71.52	8.31	12.83	71.67	8.34	13.12
	70	88—90	$C_{13}H_{16}O_2N_2$	67.22	6.94	12.06	67.36	6.97	12.02

4-Alkoxybenzamidoximes (XV)——General Procedure: To a solution of $NH_2OH \cdot HCl$ (0.15 mole) and Na_2CO_3 (0.075 mole) in H_2O (15 ml) was added a solution of 4-alkoxybenzonitrile (0.1 mole) (XIV) in EtOH (90 ml). The mixture was refluxed for 2—8 hr. EtOH was removed *in vacuo*, the residue was poured into H_2O (50 ml) and extracted with AcOEt. The extract was washed with 50 ml of 10% HCl. The aqueous layer was adjusted to pH 9 with 20% NaOH and the separated oil was extracted with AcOEt. The extract was washed with H_2O and evaporated *in vacuo*. The residue was recrystallized from EtOH or C_6H_6 to give XV. The obtained compounds are listed in Table VII.

3-(4-Alkoxy)-phenyl-5-substituted-1,2,4-oxadiazoles (XVI)——General Procedure: XV (0.02 mole) was treated with an acid anhydride (0.02 mole) or an acyl chloride (0.025 mole) in pyridine and heated at 130—140° for 2—4 hr. After cooling, the reaction mixture was poured into ice-water and extracted with $CHCl_3$ or AcOEt. The extract was washed with H_2O, dried over anhydrous Na_2SO_4 and evaporated *in vacuo*. The residue was purified by recrystallization from MeOH–H_2O. In cases that the product failed to crystallize, the residue was purified by column chromatography on silica gel, eluted with C_6H_6 or C_6H_6–acetone (9 : 1).

22) D.T. Witiak, T. Chun-Lun Ho, and R.E. Hackney, *J. Med. Chem.*, **11**, 1086 (1968).

Table VII. 4-Alkoxybenzamidoxime (XV)



R	Yield (%)	mp (°C)	Formula	Analysis (%) Calcd. C	H	N	Found C	H	N
$(C_2H_5)_2N(CH_2)_2$	88	208	$C_{13}H_{21}O_2N_3 \cdot HCl$	48.16	7.15	12.96	48.12	7.29	13.00
$C_2H_5OCOC(CH_3)_2$  $N-(CH_2)_2$ [a]	80	119	$C_{13}H_{19}O_3N_3$	58.63	6.81	10.52	58.76	6.81	10.30

a) This compound was not purified by recrystallization and column chromatography.

Hypocholesterolemic Activity[20]——Hypocholesterolemic activity was studied in male Sprague-Dawley-JCL rat of 5 weeks-old, fed the basal CE-2 diet[23] or the same diet to which 1% cholesterol, 0.2% sodium cholate and 5% olive oil had been added. The test compound previously dissolved in EtOH or ether was mixed in diets. After rats fed diets for 7 days, plasma cholesterol levels were determined by the method of Abell, *et al.*[24] Liver lipid was extract by the method of Folch, *et al.*[25] and an aliquot of the lipid extract was analyzed for its cholesterol content, following a modification of the method of Abell, *et al.* The biological activity of the compound was estimated as follows:

$$\text{Efficacy (\%) in A-test} = \frac{C-Tc}{C-N} \times 100$$

$$\text{Efficacy (\%) in B-test} = \frac{N-Tn}{N} \times 100$$

C: Plasma cholesterol (PCh) of untreated hypercholesterolemic group.
Tc: PCh of drug-treated hypercholesterolemic group.
N: PCh of untreated normocholesterolemic group.
Tn: PCh of drug-treated normocholesterolemic group.
Values of C and N were found to be about 250 and 75 mg/100 ml, respectively.

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