spectra[6] of all products (Tables I and II), with absorptions at 5.60–5.65 and 5.80–5.85 μ assigned to the carbonyl groups.

The compounds in Table II were screened for antibacterial activity by reported methods.[7] Most of the hydantoins I showed slight *in vitro* activity against gram-positive and gram-negative organisms. Very limited activity was observed for these compounds (I) when tested against *Salmonella typhosa* and *Staphylococcus aureus* infections in mice.

The 5-nitrofurfurylidenehydrazides II showed slight to fair *in vitro* antibacterial activity, with limited activity against *Salmonella typhosa* and *Staphylococcus aureus* infections in mice. These compounds II exhibited parasiticidal activity[8] in chicks against *Eimeria tenella* and *Histomonas meleagridis* when mixed in feed at 0.001 and 0.002% of the ration, by the method of Johnson.[9]

In conclusion, the antibacterial properties of both the 3-[(5-nitrofurfurylidene)amino]hydantoins (I) and the 5-nitrofurfurylidenehydrazides (II) were inferior to those of the 1-[(5-nitrofurfurylidene)amino]hydantoins.[10]

Experimental Section

3-Aminohydantoin (Method A).—A mixture of N-carboxyglycine dihydrazide (70 g, 0.48 mole) and DMF (2290 ml) was heated to boiling in 0.5 hr, with mechanical stirring. The reaction solution was refluxed for 2.8 hr. The solution was evaporated under reduced pressure, and the solid residue was washed with EtOH (50 ml). Recrystallization from 25% EtOH (185 ml) gave white crystals.

3-[(5-Nitrofurfurylidene)amino]hydantoin.—A solution of 5-nitro-2-furaldehyde (47.0 g, 0.33 mole) in EtOH (350 ml) was added gradually to a solution of 3-aminohydantoin (38.4 g, 0.33 mole) in H_2O (500 ml) at 25°, with mechanical stirring. The mixture was stirred for 1.3 hr, then cooled in an ice bath. The resultant pale yellow, crystalline solid was collected and washed (H_2O), mp 217–222°, yield 33.8 g. A second crop (34.9 g, mp 223–225°) was isolated. The combined product was recrystallized from $MeNO_2$ (1800 ml).

5-Ethyl-5-methyl-3-aminohydantoin (Method C).—A solution of ethyl N-ethoxycarbonyl-DL-isovalinate (250 g, 1.15 moles), in hydrazine hydrate (570 ml, 11.4 moles) and EtOH (3300 ml), was refluxed for 82 hr. The solution was evaporated under reduced pressure, and the semicrystalline residue was triturated with Et_2O (400 ml). The filtered product, mp 120–140°, was washed with Et_2O. Recrystallization from a mixture of H_2O (4 ml) and EtOH (120 ml) gave a white, crystalline solid.

5-Ethyl-5-methyl-3-[(5-nitrofurfurylidene)amino]hydantoin was prepared by the same procedure as described for 3-[(5-nitrofurfurylidene)amino]hydantoin.

N-Ethoxycarbonylglycine 5-Nitrofurfurylidenehydrazide.—To a solution of N-ethoxycarbonylglycine hydrazide (40.5 g, 0.25 mole) in 50% EtOH (100 ml) was gradually added a solution of 5-nitro-2-furaldehyde (35.0 g, 0.25 mole) in EtOH (100 ml). The product was collected and washed with 70% EtOH; mp 174–176°, yield 66.2 g (93%). Recrystallization from EtOH (3600 ml) gave yellow crystals.

Acknowledgments.—The authors wish to thank Mr. Grant Gustin and Mr. Marvin Tefft for the elemental analyses, and Mr. Raymond Freedman for the microbiologic testing data.

(6) L. J. Bellamy, "The Infrared Spectra of Complex Molecules," 2nd ed, John Wiley and Sons, Inc., New York, N. Y., 1958, p 221.
(7) F. F. Ebetino, W. F. Carey, and B. F. Stevenson, *J. Med. Chem.*, **6**, 633 (1963).
(8) G. C. Wright, U. S. Patent 3,096,347 (1963); *Chem. Abstr.*, **60**, 660*h* (1964).
(9) C. A. Johnson, *Poultry Sci.*, **39**, 1076 (1960).
(10) S. Mintzer, E. R. Kadison, W. H. Shlaes, and O. Felsenfeld, *Antibiot. Chemotherapy*, **3**, 151 (1953).

1,2,4-Oxadiazolylpyridinium Salts. Oral Hypoglycemic Agents

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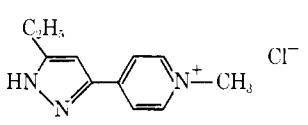
A series of 1,2,4-oxadiazolylpyridinium quaternary salts has been synthesized. These compounds display interesting hypoglycemic activity in mice.

4-[3(5)-Pyrazolyl]pyridinium salts (**1**, for instance) have been found to display interesting oral hypoglycemic activity in alloxan-diabetic mice.[1] As an initial development of this lead, the pyrazole ring was replaced by an isoxazole ring to obtain some novel isoxazolylpyridinium salts[2] which also exhibited interesting hypoglycemic activity in laboratory animals.[3] 1-Methyl-4-(3-methyl-5-isoxazolyl)pyridinium chloride (**2**) has been chosen for extensive evaluation as a potential antidiabetic agent.[4] As a further development of the lead, we now describe the synthesis and hypoglycemic activity of a number of new 1,2,4-oxadiazolylpyridinium salts, **5**, for instance.



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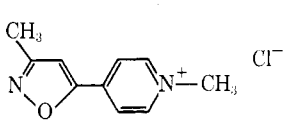
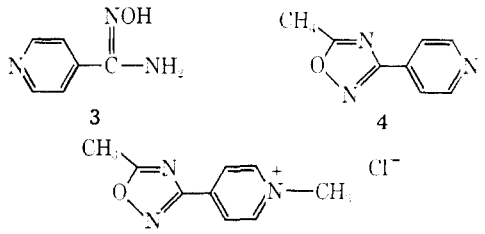


2

The synthesis of unsymmetrically substituted 1,2,4-oxadiazoles by the condensation of an amidoxime with Ac_2O has been described.[5,6] Thus the reaction of Ac_2O with isonicotinamidoxime (**3**)[7] provided 4-(5-methyl-

(1) V. J. Bauer, H. P. Dalalian, W. J. Fanshawe, S. R. Safir, E. C. Tocus, and C. R. Boshart, *J. Med. Chem.*, **11**, 981 (1968).
(2) V. J. Bauer, W. J. Fanshawe, H. P. Dalalian, and S. R. Safir, *ibid.*, **11**, 984 (1968).
(3) D. A. Blickens and S. J. Riggi, *Toxicol. Appl. Pharmacol.*, in press.
(4) S. J. Riggi, D. A. Blickens, and C. R. Boshart, *Diabetes*, **17**, 646 (1968).
(5) F. Tiemann, *Ber.*, **17**, 126 (1884).
(6) K. Clarke, *J. Chem. Soc.*, 4251 (1954).
(7) E. Bernasek, *J. Org. Chem.*, **22**, 1263 (1957).

1,2,4-oxadiazol-3-yl)pyridine (**4**). Quaternization of the pyridine base **4** with methyl chloride afforded 1-methyl-4-(5-methyl-1,2,4-oxadiazol-3-yl)pyridinium chloride (**5**).



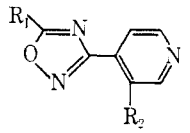
3 **4**

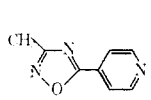
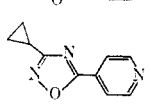
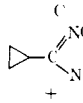
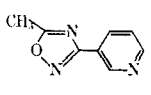
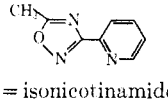
5

When it was observed that **5** displayed interesting hypoglycemic activity in mice (Table II), the preparation of a series of analogs was undertaken. The choice of substituents was influenced by the structure–activity correlation already developed for the pyrazolylpyridinium salts.[1] Reaction of the appropriate amidoxime with various anhydrides gave the desired 1,2,4-oxadiazolylpyridine bases **6–14** (Table I). 4-(1,2,4-Oxadiazol-3-yl)pyridine (**15**) was prepared by the condensation of **3** with triethyl orthoformate. Quaternization of these bases with a variety of halides produced the 1,2,4-oxadiazolylpyridinium salts **16–28** (Table II).

TABLE I

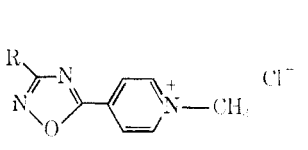
1,2,4-OXADIAZOLYLPYRIDINES



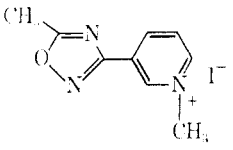
Compd	R_1	R_2	Reactants[a]	Mp, °C	Recrystn solvent	Formula[b]
15	H	H	$HC(OEt)_3$ + A	147–148	H_2O	$C_7H_5N_3O$
4	CH_3	H	Ac_2O + A	92–93	EtOH	$C_8H_7N_3O$
6	C_2H_5	H	$(EtCO)_2O$ + A	42–44	Petr ether (30–60)	$C_9H_9N_3O$
7	*c*-C_3H_5	H	$(c\text{-}C_3H_5CO)_2O$[c] + A	79–81	MeOH–H_2O	$C_{10}H_9N_3O$
8	*c*-C_4H_7	H	$(c\text{-}C_4H_7CO)_2O$ + A	Liquid[d]		$C_{11}H_{11}N_3O$
9	C_6H_5	H	$(C_6H_5CO)_2O$ + A	146–147.5	EtOH	$C_{13}H_9N_3O$
10	CH_3	CH_3	Ac_2O + B	Oil[e]		$C_9H_9N_3O$
11			MeC(=NOH)–NH_2 + C	88–89	Hexane	$C_9H_7N_3O$
12			 + C	92–93	Hexane	$C_{10}H_9N_3O$
13			Ac_2O + D	107–111[f]		$C_9H_7N_3O$
14			Ac_2O + E	89–90	*i*-PrOH	$C_9H_7N_3O$

[a] A = isonicotinamidoxime,[7] B = 3-methylisonicotinamidoxime, C = isonicotinic acid anhydride [A. W. Shrecker and P. B. Maury, *J. Amer. Chem. Soc.*, **76**, 5803 (1954)], D = nicotinamidoxime,[6,7] E = picolinamidoxime.[7] [b] Compounds have been analyzed for C, H, N. [c] F. Fichter and H. Reeb, *Helv. Chim. Acta*, **6**, 457 (1923). [d] Bp 105–108° (0.07 mm). [e] Characterized as the methiodide **23** (Table II). [f] Lit.[6] mp 113°.

The ability of **5** to lower blood glucose levels in mice when contrasted with the failure of the pyridine base **4** to exhibit this property, demonstrates that the presence of the quaternary pyridinium salt moiety is necessary for hypoglycemic activity in this series. This requirement has been previously observed for the analogous pyrazolyl and isoxazolyl systems.[1,2] In the pyrazolylpyridinium salt series,[1] activity is confined to those compounds in which the 3(5)-pyrazolyl position is bonded to the 4-pyridine position. However, the location of the 1,2,4-oxadiazole–pyridinium ring attachment is not specific for maintaining biological activity. Thus, 1-methyl-4-(3-methyl-1,2,4-oxadiazol-5-yl)pyridinium chloride (**25**), as well as the 3-pyridinium (**27**) and 2-pyridinium (**28**) analogs of **5**, possesses hypoglycemic activity on oral administration to mice (Table II).



25, R = CH_3
26, R = 



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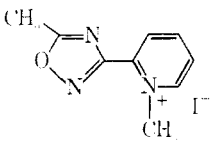


28

Activity is retained when the 5 position of the (1,2,4-oxadiazol-3-yl)pyridinium system is substituted by H (**16**), with ethyl (**20**), cycloalkyl (**21**, **22**), or phenyl (**24**). The isomeric (1,2,4-oxadiazol-5-yl)pyridinium system is also active with either a methyl (**25**) or cyclopropyl group (**26**) in the 3 position. Replacement of the N-methyl substituent of **5** with an ethyl (**17**), allyl (**18**), or 2-ethoxyethyl group (**19**) also results in compounds with hypoglycemic activity.

The hypoglycemic activity of each compound was tested in Carworth Farms male mice (CF-1 or CF-1-S), 25–30 g.[8] The compounds (1.5 or 3.0 mmoles/kg) were dissolved in 0.9% saline or suspended in 0.9% aqueous carboxymethylcellulose and administered by gavage in a volume of 0.25 ml/25-g mouse. Controls received an equal volume of vehicle. Blood samples (0.05 ml) obtained from retrobulbar plexuses 3 and 5 hr after dosing were assayed[9] for blood glucose (estimated as reducing sugar content) using the method of Hoffman[9] as adapted for the Technicon AutoAnalyzer. Results are included in Table II. Further work is in progress to determine potential clinical utility of these oxadiazolylpyridinium salts.

Experimental Section[10]

3-Methylisonicotinamidoxime (29).—To a stirred solution of 5.0 g (0.042 mole) of 3-methylisonicotinonitrile[11] and 2.8 g

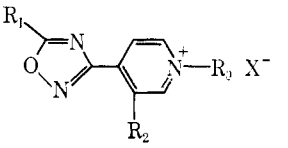
(8) Technical assistance of Miss L. Will and Miss A. Greening is greatly appreciated.

(9) W. S. Hoffman, *J. Biol. Chem.*, **120**, 51 (1937).

(10) Melting points were determined in a Hershberg apparatus and are uncorrected. Microanalyses were performed by Mr. L. M. Brancone and staff; where analyses are indicated only by symbols of the elements, analytical results obtained for those elements were within ±0.4% of the theoretical values. Uv spectra were determined in MeOH solution with a Cary 11 spectrophotometer by Mr. W. Fulmor and staff.

(11) H. Tani, *Yakugaku Zasshi*, **81**, 141 (1961).

TABLE II
1,2,4-OXADIAZOLYLPYRIDINIUM SALTS



Compd	R_1	R_2	R_3	X	Mp, °C dec	Recrystn solvent	Formula[a]	% decrease in blood glucose[b] 1.5 mmoles/kg	3.0 mmoles/kg	Control
16	H	H	CH_3	Cl	206	*i*-PrOH	$C_8H_8ClN_3O\cdot0.5H_2O$[c]	35 ± 5[d]	30 ± 4[e]	7 ± 6
5	CH_3	H	CH_3	Cl	163–165	CH_3CN	$C_9H_{10}ClN_3O\cdot H_2O$	34 ± 7[d]	68 ± 7[e]	39 ± 6
17	CH_3	H	C_2H_5	I	140–142	*i*-PrOH	$C_{10}H_{12}IN_3O$	34 ± 2	48 ± 5	−8 ± 4
18	CH_3	H	$CH_2CH{=}CH_2$	Cl	188–190	*i*-PrOH–hexane	$C_{11}H_{12}ClN_3O$	15 ± 5	41 ± 11	6 ± 4
19	CH_3	H	$CH_2CH_2OC_6H_5$	Cl	187–188	*i*-PrOH–Et$_2$O	$C_{18}H_{18}ClN_3O_2\cdot H_2O$[c]	14 ± 3	27 ± 4	9 ± 5
20	C_6H_5	H	CH_3	ClO_4[f]	128–129	EtOH	$C_{15}H_{12}ClN_3O_5$	57 ± 8	93 ± 1	−5 ± 5
21	*c*-C_3H_5	H	CH_3	Cl	211–212	CH_3CN	$C_{11}H_{12}ClN_3O\cdot0.5H_2O$	62 ± 13	87 ± 3	6 ± 4
22	*c*-C_4H_7	H	CH_3	Cl	200–203	CH_3CN	$C_{12}H_{14}ClN_3O$[c]	19 ± 6	60 ± 11	8 ± 4
23	CH_3	CH_3	CH_3	I	108–110	*i*-PrOH	$C_{10}H_{12}IN_3O$	56 ± 10	82 ± 3	4 ± 5
24	C_6H_5	H	CH_3	Cl	260–262	CH_3CN	$C_{14}H_{12}ClN_3O\cdot H_2O$	56 ± 13		31 ± 7
25		See **25** in text		Cl	173–175	*i*-PrOH–hexane	$C_9H_{10}ClN_3O\cdot H_2O$	32 ± 5	33 ± 5	15 ± 4
26		See **26** in text		Cl	226–227	CH_3CN	$C_{11}H_{12}ClN_3O\cdot0.5H_2O$	22 ± 4	29 ± 4	6 ± 3
27		See **27** in text		I	203–205	MeOH	$C_9H_{10}IN_3O$[g]	35 ± 2	38 ± 6	18 ± 8
28		See **28** in text		I	156	*i*-PrOH	$C_8H_{10}IN_3O$[c]	27 ± 7	50 ± 12	13 ± 6

[a] Compounds have been analyzed for C, H, N, halogen. [b] Values are means ± standard errors of four to six mice. Maximal reductions in blood glucose concentrations 3 or 5 hr after dosing are expressed as per cent decrease from predose values. Control animals were dosed orally with vehicle. An increase in blood glucose is indicated by a negative sign (−). Average predose blood glucose concentration for 88 control mice was 128 ± 2 mg %. [c] ±0.5 of the theoretical value. [d] 1.6 mmoles/kg. [e] 3.2 mmoles/kg. [f] The crude N-methyl chloride was converted to the perchlorate salt for characterization. [g] ±0.6 of the theoretical value.

(0.042 mole) of $HONH_3^+Cl^-$ in 25 ml of EtOH and 10 ml of H_2O was slowly added 2.2 g (0.021 mole) of Na_2CO_3. The solution was heated under reflux for 3 hr and then concentrated under reduced pressure to give a white solid residue. The solid was taken up in hot EtOH, filtered, and concentrated under reduced pressure to give a tacky, white solid. The solid was recrystallized (*i*-PrOH–hexane) to yield 1.32 g (18%) of colorless crystals, mp 102–109°, and an additional recrystallization afforded colorless crystals, mp 135–139°. *Anal.* ($C_7H_9N_3O\cdot0.5H_2O$) C, H, N.

Cyclopropanecarboxamidoxime Hydrochloride (30).—A MeOH solution (200 ml) of 20 g (0.37 mole) of NaOMe was slowly added to a MeOH solution (200 ml) of 25 g (0.37 mole) of cyclopropyl cyanide and 25.7 g (0.37 mole) of $HONH_3^+Cl^-$. The mixture was filtered and concentrated under reduced pressure to a viscous liquid which was taken up in EtOH. The mixture was filtered and the filtrate was concentrated under reduced pressure to 33 g (89%) of a clear, viscous liquid. A 3-g sample of the cyclopropanecarboxamidoxime was treated with an excess of ethanolic HCl. The addition of Et_2O precipitated a solid which was twice recrystallized (*i*-PrOH–hexane) to provide off-white crystals, mp 178–181°. *Anal.* ($C_4H_9ClN_2O$) C, H, Cl, N.

4-(5-Methyl-1,2,4-oxadiazol-3-yl)pyridine (4).—A mixture of 27 g (0.2 mole) of isonicotinamidoxime[7] (**3**) and 50 ml of Ac_2O was heated under reflux for 3 hr. The solvent was distilled and the residual brown liquid was treated with 100 ml of aqueous Na_2CO_3. The solid which separated was twice recrystallized (EtOH) to yield 10 g (31%) of off-white crystals: mp 92–93°; uv, 226 mμ (ε 10,200) and 273 mμ (ε 2500). *Anal.* (Table I).

1-Methyl-4-(5-methyl-1,2,4-oxadiazol-3-yl)pyridinium Chloride (5).—A mixture of 4.5 g (0.028 mole) of **4** and 10 ml of MeCl was heated for 18 hr at 120° in a glass-lined steel bomb. The excess MeCl was allowed to evaporate and the residual solid was recrystallized (MeCN) to yield 4.1 g (69%) of colorless crystals: mp 150–152°; uv, 253 mμ (ε 10,600). *Anal.* (Table II).

1,2,4-Oxadiazolylpyridines (Table I).—Compounds **6–15** were prepared by the condensation of an amidoxime with the appropriate anhydride or with triethyl orthoformate using the procedure described for **4**.

1,2,4-Oxadiazolylpyridinium Salts (Table II).—Compounds **16–28** were prepared by the reaction of the 1,2,4-oxadiazolylpyridines **6–15** with an alkyl halide in a bomb (as for **5**, above) or in an alcoholic solvent under reflux.