drogenase but somewhat less strongly than does T_4.[27] These observations suggest that 2,6-DIHQ merits further attention as a tool for elucidating molecular mechanisms underlying thyroid hormone action. Furthermore, the possibility should not be overlooked that 2,6-DIHQ may be a physiologically important thyroactive compound. It has been isolated from the thyroid gland[28] and has been produced from iodinated thyronines[29] and from 3,5-diiodotyrosine[28b,30] under conditions that could prevail *in vivo*.

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5,6-Dihydro-4H-1,3,4-thiadiazines. III. Chemistry and Pharmacology of a Series of Basic Derivatives[1]

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A series of basically substituted 5,6-dihydro-4H-1,3,4-thiadiazines was synthesized and tested for central nervous system depressant activity in mice. None of the compounds showed significant activity in the maximal electroshock, hydrochloric acid writhing, strychnine lethality, or the metrazol seizure threshold tests. All the compounds in the series showed some activity in the hexobarbital sleep time test.

Prior to the start of this work a series of substituted 5,6-dihydro-4H-1,3,4-thiadiazines had been synthesized and tested for pharmacologic activities.[3,4] These compounds were, in general, devoid of pharmacological activity in the mouse.[5] Certain members of this previous series of compounds inhibited monoamine oxidase activity of the brain tissue of the rat and possessed antimicrobial activity against certain types of bacteria and fungi.[4] Because of the novelty of the 5,6-dihydro-4H-1,3,4-thiadiazine heterocycle and because previously we had observed that certain pyridyl-substitued 5,6-dihydro-4H-1,3,4-oxadiazines exhibited central nervous system depressant activity in mice,[6] we decided to synthesize and test pharmacologically a series of basically substituted 5,6-dihydro-4H-1,3,4-thiadiazines. This paper reports the results of this study.

Chemistry.—The two synthetic methods used to prepare the members of the previously reported[3,4] series of thiadiazines are the treatment of a 2-(β-hydroxyalkyl)carboxylic acid hydrazide with P_2S_5 and the concentrated H_2SO_4 cyclodehydration of a 2-(β-hydroxyalkyl)thiocarboxylic acid hydrazide. Because the treatment of 2-methyl-2-(β-hydroxypropyl)isonicotinic acid hydrazide with P_2S_5 gave 5,6-dihydro-4,6-dimethyl-2-(4-pyridyl)-4H-1,3,4-thiadiazine (isolated as the dihydrochloride) in only 6% yield, and because previously[6] we had encountered difficulty in preparing the variously substituted 2-(β-hydroxyalkyl)nicotinic, -isonicotinic, and -picolinic acid hydrazides, we used four different synthetic methods to prepare the basically substituted thiadiazines listed in this paper. These methods are condensation of a β-hydrazinoalkylthiol with either (A) a nitrile, (B, C) an imino ester, (D) an aldehyde, or (E) cyanogen bromide (see Chart I).

Method A is straightforward and of wide scope as evidenced by the fact that it yielded a thiadiazine from aromatic nitrile,[1] aliphatic nitrile,[1] tertiary aminoalkylnitrile, and pyridylnitrile. This method, which involves heating at the reflux temperature for 18 hr a mixture of nitrile, β-hydrazinoalkylthiol, and ethanol and then distilling the mixture *in vacuo*, yielded an oil composed of the desired basically substituted thiadiazine plus impurities that exhibit infrared absorption at 2.9, 3.0, 3.1 (m, broad), and 6.16 μ (m). The oil was easily purified by treating it with alumina (Merck No. 71695) in benzene. Methods B and C demonstrate the ability of the imino ester derivative of a cyanopyridine to condense with a β-hydrazinoalkylthiol. The condensation of 1-(1-methylhydrazino)-2-propanethiol with 3-pyridinecarboxaldehyde (method D) in refluxing ethanol in the presence of pyridine gave tetrahydro-4,6-dimethyl-2-(3-pyridyl)-2H-1,3,4-thiadiazine (III). In contrast, we previously[1] had observed that condensation of 1-(1-methylhydrazino)-2-propanethiol with benzaldehyde in refluxing ethanol in the presence of pyridine gave the (2-mercaptopropyl)methylhydrazone of benzaldehyde. Method E illustrates condensation of a cyanogen halide with a β-hydrazinoalkylthiol to give an aminothiadiazine directly.

The structures assigned to the compounds in Table I were substantiated by elemental, infrared, and nmr analyses.

Pharmacology.—These thiadiazines were evaluated for central nervous system depressant activity using a battery of screening methods; the data are summarized in Table I. None showed significant activity in the maximal electroshock, strychnine lethality, hydrochloric acid writhing, or the metrazol seizure threshold tests.

(1) Paper II: *J. Heterocyclic Chem.*, **4**, 254 (1967).

(2) To whom inquires should be addressed.

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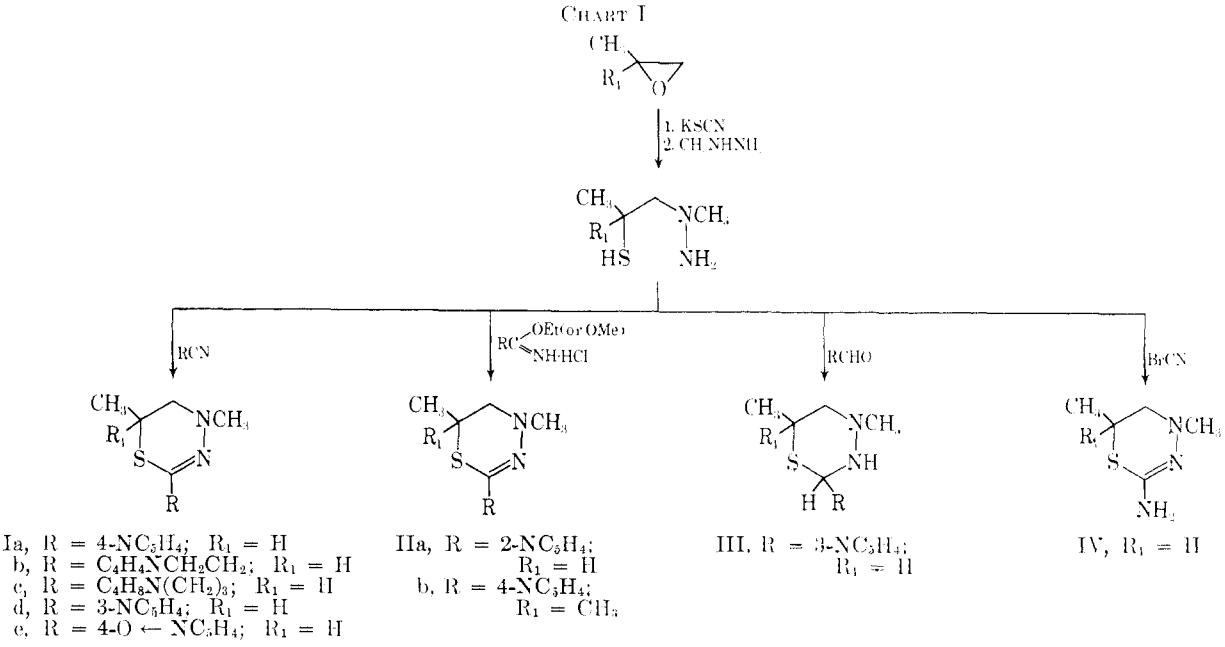
(5) In mice, these compounds did not show significant activity in the following standard screening tests: hexobarbital sleep time, HCl writhing, tremorine antagonism, maximal electroshock, strychnine protection, *d*-amphetamine aggregation, and monoamine oxidase inhibiting.

(6) D. L. Trepanier, P. E. Krieger, and J. N. Eble, *J. Med. Chem.*, **8**, 802 (1965).

CHART I



Ia, R = 4-NC_5H_4; R_1 = H
b, R = $C_4H_4NCH_2CH_2$; R_1 = H
c, R = $C_4H_8N(CH_2)_3$; R_1 = H
d, R = 3-NC_5H_4; R_1 = H
e, R = 4-O ← NC_5H_4; R_1 = H

IIa, R = 2-NC_5H_4; R_1 = H
b, R = 4-NC_5H_4; R_1 = CH_3

III, R = 3-NC_5H_4; R_1 = H

IV, R_1 = H

All compounds in this series showed some activity in the hexobarbital sleep time test. The more active compounds were selected for testing for reinduction of sleep in hexobarbital-treated mice. They were also active in this test which suggests a central nervous system rather than a metabolic mechanism of action.

The most effective compound in the hexobarbital sleep time test was the 4-pyridyldimethylthiadiazine (Ia). Substitution in the 2 position of the thiadiazine ring with a 3-pyridyl (Id) or a 2-pyridyl (IIa) moiety, instead of the 4-pyridyl, resulted in reduction of activity. The conversion of Ia to its N-oxide (Ie) resulted in only a slight loss of activity as did substitution of a methyl group (IIb) for the proton in the 6 position. Replacement of the 4-pyridyl moiety of Ia by a pyrrolylethyl (Ib), pyrrolidinopropyl (Ic), or amino (IV) moiety resulted in reduction of activity. The 2-pyridyl derivative of the saturated dimethyl thiadiazine (III) was also less active.

Compound Ia was selected for more extensive evaluation. It was found to be as active orally as parenterally in prolonging hexobarbital sleep times in mice. Intraperitoneal doses of 25 and 50 mg/kg resulted in average sleep times of 109 ± 11 and 204 ± 14 min; oral doses of 25 and 50 mg/kg resulted in average sleep times of 95 ± 11 and 175 ± 13 min (controls 26 min). At doses as high as 40 mg/kg, Ia did not protect mice in the *d*-amphetamine aggregate toxicity test nor did it antagonize the hyperthermic response of rats to brewers yeast. Ia exhibited no significant effect on spontaneous motor activity (measured in Woodard circular activity cages) in mice at either 10 or 25 mg/kg. At a dose of 50 mg/kg at 60 min after dosing, the motor activity was depressed more than 50% from the saline control. The intravenous administration of 20 mg/kg of Ia in an aqueous solution to anesthetized dogs resulted in only a transient hypotension. No autonomic effects, adrenergic blockade, or antihistaminic, or anticholinergic effects were observed. The pharmacological activity found for this thiadiazine series is quite similar to that reported previously for a series of analogous oxadiazines.[6]

Experimental Section

The melting points were obtained in a capillary tube with the Thomas–Hoover Uni-Melt and are corrected. The elemental analyses were done by Midwest Microlabs., Inc., Indianapolis, Ind. The nmr spectra were obtained at 60 Mc, with a Varian A-60 spectrometer, for 10% $CDCl_3$ solutions containing tetramethylsilane (TMS) as an internal standard. Chemical shifts are measured as shielding (cps) relative to the shielding of the TMS protons. Infrared spectra were obtained with a Perkin-Elmer 337 grating spectrophotometer.

Propylene sulfide (bp 72–73°, 73% yield) and **isobutylene sulfide** (bp 83–85°, 40% yield) were prepared by the reaction of the oxides with potassium thiocyanate in aqueous solution.[7]

1-(1-Methylhydrazino)-2-propanethiol[1] and **2-methyl-1-(1-methylhydrazino)-2-propanethiol** [bp 70–75° (13 mm), 46%] were prepared by the reaction of propylene sulfide and isobutylene sulfide with methylhydrazine. **Isonicotinonitrile 1-oxide** (mp 225–227°, yield 90%) was prepared by H_2O_2–AcOH oxidation of isonicotinonitrile.[8]

Basically Substituted Thiadiazines (Table I). Method A.—A mixture of 120 g (1.0 mole) of 1-(1-methylhydrazino)-2-propanethiol, 104 g (1.0 mole) of isonicotinonitrile, and 350 ml of ethanol was refluxed for 18 hr and distilled *in vacuo* to give 137 g (66%) of light yellow oil, bp 169–175° (3.0 mm); glpc (5-ft column, 4% SE30 Chromosorb W, acid-washed, 180°, He flow rate 300 ml/min), 9 sec (18%), 12 sec (8%), and 117 sec (74%); λ_{max}^{film} 2.91, 3.02 and 3.14 (m, broad), 6.16 (m), and 6.29 (s) μ. The oil (137 g) was dissolved in 1500 ml of benzene, 900 g of alumina (Merck No. 71695) was added, and the mixture was stirred for 20 min and filtered. The alumina was washed with 200 ml of benzene, and the benzene was evaporated *in vacuo* to give 97 g (47%) of a faintly straw-colored oil: it exhibited one peak (glpc), t = 110 sec; infrared, absence of 2.91, 3.02, 3.14, and 6.16-μ bands and presence of SC=N band at 6.29 μ (s); nmr, 78 (CH_3, doublet, J = 7 cps), 150 (quartet, 1 proton), 179 (NCH_3, singlet), 182 (quartet, 1 proton), 213 (multiplet, 1 proton), 451 (multiplet, 2-pyridyl protons), and 511 (multiplet, 2 pyridyl protons) cps.

The **maleate** was prepared using 1 molar equiv of maleic acid in ethanol–ether. The hydrochlorides in Table I were prepared by treating an ether solution of the base with ethereal HCl.

Method B.—To a stirred mixture of 14.1 g (0.12 mole) of 1-(1-methylhydraziuo)-2-propanethiol, 4.3 g of gaseous HCl, and 75 ml of methanol was added, dropwise, over a period of 0.5 hr, a solution of 16 g (0.12 mole) of methyl picolinimidate in 20 ml of methanol. The mixture was stirred at ambient temperature

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Table I: Basically Substituted 5,6-Dihydro-4H-1,3,4-thiadiazines

No.	R	R_1	Bp (mm) or mp, °C	Yield, %	Method[a]	Calcd, % C	Calcd, % H	Calcd, % N	Calcd, % S	Found, % C	Found, % H	Found, % N	Found, % S	Toxicity LD_{50}, mg/kg ip	Screening dose, mg/kg ip	Hexobarbital sleep time, ratio T/C	Hexobarbital resleep test[b]
Ia	4-NC_5H_4	H	116–117[c]	42	A	52.00	5.30	13.00	9.91	52.19	5.63	13.08	10.20	88	20	5.5	10/10
b	$C_4H_4NCH_2CH_2$	H	132–139 (0.5)	37	A	59.16	7.67	18.82		59.23	7.87	18.76		681	50	2.3	10/10
c	$C_4H_8NCH_2CH_2CH_2$	H	111–116 (0.2)	28	A	59.71	9.60	17.41		59.91	9.80	17.62		68	50	1.9	
d	3-NC_5H_4	H	168–170[d]	21	A	49.27	5.78	17.24	14.55[e]	49.27	6.05	16.86	14.78[e]	464	50	2.0	
e	4-O←NC_5H_4	H	196–198[d]	16	A	46.24	5.43	16.18	12.34	46.68	5.62	16.52	12.38	147	25	4.3	9/10
IIa	2-NC_5H_4	H	103–104	26	B												
b	4-NC_5H_4	CH_3	227–229[d] dec	38	C	51.25	6.25	16.30	13.75[e]	50.74	6.39	16.01	13.81[e]	83	50	2.5	
															25	4.9	10/10
III			137–138	26	D	57.38	7.22	20.07		57.55	7.20	20.42		681	200	6.4	
															25	2.7	
IV	H_2N	H	78–80 (0.1)	26	E	41.35	7.63	28.94		40.90	8.28	28.40		316	50	5.4	10/10
															25	2.8	

[a] See Experimental Section. [b] The ratio is the number of mice in which sleep was reinduced/number of mice tested. [c] Maleate. [d] Hydrochloride. [e] Chlorine.

for 2 hr, concentrated *in vacuo*, treated with water, basified with NaOH solution, and extracted with ether. The washed (H_2O) and dried ($MgSO_4$) ether extract was distilled *in vacuo* to give 6.5 g (26%) of yellow oil, bp 151–163° (0.5–2.5 mm). The oil was dissolved in ether and treated with ethereal HCl until precipitation of the hydrochloride was complete.

Method C.—A mixture of 18.5 g (0.083 mole) of 2-methyl-1-(1-methylhydrazino)-2-propanethiol, 11.1 g (0.083 mole) of ethyl isonicotinimidate dihydrochloride, and 250 ml of absolute EtOH was stirred and refluxed for 18 hr. The cooled reaction mixture was poured onto crushed ice, basified with NaOH solution, and extracted with $CHCl_3$. The washed (H_2O) and dried ($MgSO_4$) $CHCl_3$ solution was distilled *in vacuo* to give 7 g (38%) of yellow oil: bp 132–136° (1.0 mm); λ_{max}^{film} 2.95 (vw, broad), 5.95 (w), 6.20 (ms), and 6.31 (s) μ. It was chromatographed on 125 g of alumina (Baker No. 0537) in a 2.8 × 17 cm column using benzene as the eluent to give 5.8 g (30%) of a slightly yellow oil: λ_{max}^{film} 6.25 μ (s) (–SC=N); nmr, 81 (Me_2C, singlet), 162 (CH_2, singlet), 183 (NCH_3, singlet), 452 (2 pyridyl protons, multiplet), and 512 (2 pyridyl protons, multiplet) cps. The oil was dissolved in ether and treated with ethereal HCl.

Method D.—To a mixture of 20 g (0.17 mole) of 1-(1-methylhydrazino)-2-propanethiol, 1 ml of pyridine, and 50 ml of ethanol heated to 70° on a steam bath was added portionwise over a period of 0.5 hr, 18 g (0.17 mole) of 3-pyridinecarboxaldehyde in 50 ml of ethanol. The mixture was heated an additional 0.5 hr on the steam bath and evaporated to dryness *in vacuo*. The residual oil was kept at ambient temperature for 4 days. The solid that had formed was collected (filtration) and washed with a minimum of ether to give 13 g of white crystals, mp 120–130°. Two recrystallizations from ethyl acetate gave 9.0 g (26%) of white crystals: mp 137–138°; λ_{max}^{Nujol} 3.12 (s) (NH), 9.65 (s), 10.5 (s), 11.4 (s), 12.3 (s), 13.3 (s), and 13.5 (s) μ (no –C=N); nmr, 72 (CH_3, doublet, J = 6 cps), 113 (multiplet, one CH_2 proton), 152 (NCH_3, singlet), 189 (multiplet, one CH_2 proton), 323 (2-H doublet, J = 10 cps), 433, 465, 522, and 534 (4 pyridyl protons, 2 multiplets, a quartet, and a doublet) cps.

Method E.—To a stirred mixture of 22 g (0.20 mole) of BrCN and 200 ml of H_2O was added dropwise over a period of 1 hr, a solution of 24 g of 1-(1-methylhydrazino)-2-propanethiol in 50 ml of H_2O. After the addition was completed, the mixture was heated at reflux temperature for 1 hr, decolorized with activated charcoal, cooled, made alkaline with cold NaOH solution, and extracted with $CHCl_3$. Distillation of the dried ($MgSO_4$) extract gave 7.7 g (26%) of a slightly yellow liquid: bp 78–80° (0.1 mm); $\lambda_{max}^{CCl_4}$ 2.89, 2.97, 3.03, 3.16 (NH_2), 6.15, 6.30 (C=N), 6.92, 7.81, and 10.1 μ; nmr (CCl_4), 78 (CH_3, doublet, J = 7 cps), 152 (quintet, 1 proton), 155 (NCH_3, singlet), 171 (quartet, 1 proton), 210 (multiplet, 1 proton), and 244 cps (broad signal, NH_2); glpc (175°, 5-ft column, 4% SE30/Chromosorb W, acid-washed, He flow rate 250 ml/min), t = 360 sec.

Pharmacology.—The acute toxicity[6,9] and CNS-depressant,[6,9] analgesic,[10] and anticonvulsant[6,9,11] effects in mice were investigated by the techniques previously described. The effects on spontaneous motor activity in mice were measured in doughnut-shaped cages with a central light source impinging upon opposing photosenstive cells.[11] Antagonism to yeast-induced hyperthermia in rats was determined by the method of Hambourger and Smith.[12] Chlorpromazine was used as a positive control. Antagonism to the toxic effects of amphetamine in aggregated mice was determined as described by Mennear and Rudzik.[13]

Reinduction of sleep (loss of righting reflex) in mice was determined on an all or none basis by intraperitoneal injection of test compound immediately following recovery from hexobarbital (100 mg/kg) induced sleep.[14] Cardiovascular studies were done in dogs anesthetized with pentobarbital using methods previously described.[15]

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