SYNTHESIS AND NEUROPHARMACOLOGICAL ACTIVITY OF 1,2,4-TRIAZINE-3-THIONE DERIVATIVES

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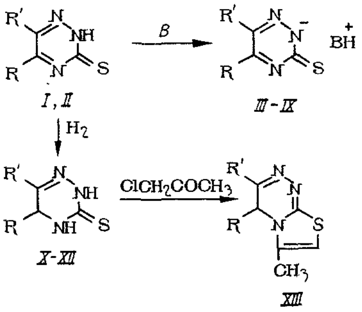
The results of an investigation into the pharmacological properties of 1,2,4-triazine-3-thiones have led to interest in this system. The derivatives of 2,3-dihydroindolo[2,3-e]-1,2,4-triazine-3-thione have been found to include antiviral and antibacterial substances [1]; 2,3-dihydro-4,7-phenanthrolino[5,6-e]-1,2,4-triazine-3-thione derivatives are active against intestinal parasites [2]; and 5-substituted 2,3-dihydro-1,2,4-triazine-3-thiones act on the spinal-cord centers, have a spasmodic action [3, 4], and have antihistaminic properties [5].

We have synthesized a new group of 1,2,4,-triazine-3-thione derivatives and have developed comparatively simple methods for synthesizing them. The starting materials used were 2,3-dihydro-1,2,4-triazine-3-thione derivatives (I, II) prepared by condensation of α-dicarbonyl compounds with thiosemicarbazide in acetic acid [6, 7].

The 2,3-dihydro-1,2,4-triazine-3-thione derivatives are weakly-acid compounds which readily dissolve in alkali to form the thiol form [3]. They might also be expected to react with organic bases. In the course of our experiments it was found that they react readily with primary, secondary, and tertiary amines, and with ammonia, to form the corresponding ammonium salts (III-IX).



When we investigated the reduction of the 2,3-dihydro-1,2,4-triazine-3-thione derivatives with zinc dust or sodium hydrosulfite in alkaline media, we found that the reaction proceeds with addition of only one hydrogen molecule, forming the corresponding tetrahydro derivatives (X-XII). We used H^1 NMR to solve the problem of which of the double bonds is involved in the reaction. The H^1-NMR spectrum of XII consists of three groups with different chemical shifts: The singlet at 7.19 ppm belongs to the protons of the phenyl group; and the two doublets centered at 4.85 and 6.87 ppm belong to the protons in the 4- and 5-positions of the ring. Thus, in the number and position of its signals, the H^1-NMR spectrum of XII corresponds to the 2,3,4,5-tetrahydro structure, i.e., the 4,5 double bond is involved in the reduction of the 2,3-dihydro derivatives of 1,2,4-triazine-3-thione.

The 2,3,4,5-tetrahydro-1,2,4-triazine-3-thione derivatives react with α-halo carbonyl compounds to form a new heterocyclic system, namely, the thiazole[3,2-c]-1,2,4-triazine system [8], as a result of

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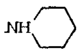
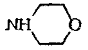
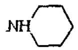
TABLE 1. 1,2,4-Triazine-3-thione Derivatives and Their Acute Toxicity

Compound	R	R'	B	Yield (%)	Melting point (deg)*	Found (%)				Empirical formula	Calculated (%)				LD_{50}, (mg/kg)
						C	H	N	S		C	H	N	S	
I	C_6H_5	C_6H_5	—	45—50	225—226	67,8	4,4	16,0	11,8	$C_{15}H_{11}N_3S$	67,95	4,15	15,83	12,07	1200
II	C_6H_5	H	—	54,0	205—206	57,1	3,6	22,2	16,9	$C_9H_7N_3S$	57,14	3,70	22,23	16,93	520
III	C_6H_5	C_6H_5	NH_3	67,4	>100 (decomp)	64,1	5,3	19,6	11,1	$C_{15}H_{14}N_4S$	63,83	4,97	19,85	11,35	650
IV	C_6H_5	C_6H_5	$C_2H_5NH_2$	80,0	>100 (decomp)	65,8	5,7	17,9	10,5	$C_{17}H_{18}N_4S$	65,81	5,81	18,06	10,32	670
V	C_6H_5	C_6H_5	$(C_2H_5)_2NH$	83,0	>100 (decomp)	67,5	6,6	16,9	9,5	$C_{19}H_{22}N_4S$	67,46	6,51	16,56	9,47	420
VI	C_6H_5	C_6H_5	$(C_2H_5)_3N$	80,5	>100 (decomp)	68,6	6,9	15,0	—	$C_{21}H_{26}N_4S$	68,85	7,10	15,31	—	1200
VII	C_6H_5	C_6H_5		80,0	>100 (decomp)	68,3	6,0	15,6	—	$C_{20}H_{22}N_4S$	68,58	6,28	16,0	—	450
VIII	C_6H_5	C_6H_5		72,8	>100 (decomp)	64,5	5,9	15,6	—	$C_{19}H_{20}N_4OS$	64,77	5,68	15,91	—	637
IX	C_6H_5	H		62,0	>100 (decomp)	61,0	6,7	20,2	11,3	$C_{14}H_{18}N_4S$	61,31	6,57	20,45	11,67	650
X	CH_3	CH_3	—	72,3	160—161	42,0	6,3	29,1	22,6	$C_5H_9N_3S$	41,93	6,33	29,34	22,39	530
XI	C_6H_5	C_6H_5	—	91,8	222—223	67,5	4,7	15,3	11,8	$C_{15}H_{13}N_3S$	67,41	4,87	15,74	11,98	850
XII	C_6H_5	H	—	86,4	142—143	56,6	4,6	21,7	16,5	$C_9H_9N_3S$	56,54	4,71	22,00	16,75	400
XIII	C_6H_5	C_6H_5	—	—	136—137	70,5	5,0	14,0	—	$C_{18}H_{15}N_3S$	70,81	4,92	13,78	—	1250
XIV	C_6H_5	C_6H_5	—	76,1	>200 (decomp)	62,9	4,5	12,0	—	$C_{18}H_{15}N_37S\cdot HCl$	63,26	4,68	12,28	—	1180
XV	C_6H_5	C_6H_5	—	—	106—107	66,1	4,3	13,3	—	$C_{17}H_{13}N_3OS$	66,42	4,26	13,67	—	1160
XVI	C_6H_5	C_6H_5	—	78,8	>150 (decomp)	59,5	4,0	12,5	—	$C_{17}H_{13}N_3OS\cdot HCl$	59,38	4,10	12,22	—	1280

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*Compound I was crystallized from toluene; compounds II, XII, XIII, XV, and XVI, from alcohol; compound III, from a mixture of alcohol and hexane; compound IV, from ethyl acetate; compound V, from a mixture of alcohol and ethyl acetate; compound VI, VII, and VIII, from a mixture of alcohol and benzene; compound X, from water; compound IX, from benzene; and compounds XI and XIV, from n-butyl alcohol.

TABLE 2. Some Pharmacological Effects of 1,2,4-Triazine-3-thiones Administered at a Dose of 20% LD_{50}

Compound	No. of movements in 10 min (orientating activity)	Duration of hexenal-induced sleep (min)	Reserpine action		Duration of apomorphine-induced stereotype (min)	Duration of arecoline-induced hyperkinesia (min)	Corazol test (% of protected animals)
			ptosis (marks)	hypothermia (Δt, deg)			
II	418±28	52±2	2,5±0,4	1,8±0,3	59±3	15±2,8	0
VI	398±12	39±3,2	3,8±0,4	1,5±0,2	62±2,4	16±1,4	0
VII	188±22	185±12	1,5±0,3	1,6±0,3	68±4,2	18±2,6	0
VIII	205±18	72±11	2,9±0,4	2,0±0,3	62±4,3	16,2±4,2	0
IX	420±19	62±8	1,9±0,3	1,6±0,4	67±4,6	15,9±4,8	0
X	162±18	72±3	4	3,6±0,4	18±2,1	10,5±1,6	30
XI	428±28	54±2	2,2±0,2	2,0±0,2	44±2	15,8±1,6	0
XII	446±26	56±3	2,1±0,1	1,6±0,3	52±2,2	16,8±1,7	0
XIII	98±11	96±11	4	3,6±0,3	20±2	13,4±1,3	10
XIV	219±10	82±6	3,8±0,2	2,8±0,2	39,4±2,1	9,8±1,9	10
Control	393±28	42±5	4	2,5±0,6	48±6	16±2,4	—

condensation and subsequent cyclization. 5,6-Diphenyl-2,3,4,5-tetrahydro-1,2,4-triazine-3-thione (XI) reacts with chloroacetone to form 3,4-diphenyl-6-methylthiazolo[3,2-c]-1,2,4-triazine (XIII) and with chloroacetic acid to form 3,4-diphenyl-6-ketothiazolo[3,2-c]-1,2,4-triazine (XV). The presence of the keto form in XV is confirmed by the IR spectrum, in which there is an intense carbonyl-stretching band with a frequency of 1750 cm^{-1}.

The IR spectra of I, II, and X-XII do not exhibit the characteristic absorption band of the mercapto group in the 2600-2550-cm^{-1} region, and the clear signal in the 3500-3300-cm^{-1} region indicates the presence of an NH group. The absorption spectra of the ammonium salts III-IX show similarities with the spectra of I and II, which indicates that they have a thione structure, i.e., the salt-forming reaction takes place not through the sulfur atom, but throught the atom in the 2 position of the triazine ring.

The main properties of the compounds synthesized, and their acute toxicity on intraperitoneal injection into mice (LD_{50}) are given in Table 1.

The compounds synthesized were tested for neuropharmacological activity on hyperaggressive mice of both sexes weighing 20-22 g. We determined the acute toxicity on intraperitoneal injection, and the effect of the substances on spontaneous behavior, motor activity (from the number of movements in 10 min in the motor-activity recorder in the experimental-industrial workshops of the USSR Academy of Medical Sciences), and induced aggression in the mice [9]. We studied the effect of the substances on animal body temperature, on spasms induced by maximum electric shock (50 mA, 0.2 sec) and by Corazol (150 mg/kg), on the duration of hexenal-induced (75 mg/kg) sleep, of apomorphine-induced (30 mg/kg) stereotypy, of arecoline-induced (25 mg/kg) hyperkinesia, and of reserpine-induced (2.5 mg/kg) ptosis, and on hypothermia. The antihistaminic activity was determined on a section of the small intestine of a rat and on isolated rat cervix on the basis of the ability of the substances to affect contracture induced by histamine ($2\cdot10^{-7}$) [sic]. The test compounds were injected intraperitoneally 45 min before administering the hexenal, apomorphine, arecoline, reserpine, and Corazol, which were also injected intraperitoneally. Psychotropic activity was noted in compounds II, VI, VII, IX, X, XI, XII, and XIV.

In the test for prolongation of hexenal-induced sleep, the highest activity was observed for compounds VII, VIII, X, XIII, and XIV. These same compounds depressed orientating motor activity.

Compounds VI and VII raised the body temperature of the mice somewhat (by 1-2°), while X and XIV lowered it (by 2-3°).

In subsequent tests, it was found that compounds VII and VIII potentiate the effect of apomorphine and weaken the action of reserpine, whereas X, XIII, and XIV acted as weak apomorphine antagonists and, in addition, potentiated the effect of reserpine (Table 2).

In the induced-agression test, only compounds X and XIV prevented combat between the male mice when they were placed in pairs in cages with floors made of plates onto which an electric current was applied.

The substances studied had no significant effect on spasms induced by Corazol and maximum electric shock; only compound X prevented Corazol-induced spasms in 30% of the animals.

Compounds II, VI, VII, VIII, and XII somewhat increased the "group toxicity" of amphetamine [10], but this effect was pronounced only in the case of compound VII. Amphetamine at a dose of 8 mg/kg caused the loss of 40% of the grouped animals, while compound VII administered in a dose equal to 20% of its LD_{50} together with the amphetamine increased the mortality rate of the mice to 60%. Only compound VII diminished etaperazine-induced catalepsy, which was evaluated by a known method [11].

Substances II, VII, and XII increased the number of fast waves in the electroencephalographs of rats with permanently-implanted electrodes, especially in the cortex and hypothalamus; substance X increased the proportion of slow activity; and the other substances had no significant effect on the bioelectric activity of the rat brain.

Central m-cholinolytic activity could not be detected in the majority of the compounds. Only compounds X and XIV, which have weak sedative properties, shorten slightly the duration of arecoline-induced hyperkinesia (see Table 2). Antihistaminic properties were observed in none of the substances studied.

On the basis of the experimental results, we can conclude that substances II, VI, VII, VIII, IX, XI, and XII have properties characteristic of antiaggressants. They decrease animal motility, potentiate the effect of hexenal and apomorphine, and are reserpine antagonists, and compound VII is also an etaperazine antagonist.

The most active substance in this group is VII, but it is inferior to known antiaggressants (a comparison was made with melipramine). Thus, in the test for prolongation of hexenal-induced (38 mg/kg) sleep, the ED_{50} of melipramine is 16.9 (11.1-21.4) mg/kg and that of VII is 44 (38.8-49.7) mg/kg. In the test for potentiation of apomorphine-induced sterotypy in mice, the ED_{50} of melipramine is 2.9 (1.5-3.8) mg/kg, and that of VII is 98.6 (78-114.6) mg/kg.

EXPERIMENTAL

5,6-Diphenyl-2,3-dihydro-1,2,4-triazine-3-thione (I). Prepared by condensation of benzil with thiosemicarbazide in acetic acid [6].

5-Phenyl-2,3-dihydro-1,2,4-triazine-3-thione (II). Synthesized by reacting phenylglyoxal with thiosemicarbazide by the method described in [7].

Ammonium Salt of 5,6-Diphenyl-2,3-dihydro-1,2,4-triazine-3-thione (III). Concentrated ammonia solution (1.5 ml) is added to a suspension of 5.3 g I in 20 ml benzene. The color of the precipitate gradually changes. The yellow crystals are filtered off and recrystallized from a mixture of alcohol and hexane (3 : 1) to obtain 3.8 g of III.

Compounds IV-IX were prepared analogously (see Table 1).

5,6-Diphenyl-2,3,4,5-tetrahydro-1,2,4-triazine-3-thione (XI). Technical sodium hydrosulfite (40-45 g) or zinc dust (10-12 g) is added in small portions to a boiling solution of 26.5 g I in 500 ml of 10% aqueous sodium hydroxide solution while agitating vigorously over a period of 5-10 min. The boiling solution becomes colorless, and a yellowish precipitate is deposited from the reaction mixture. After cooling, the precipitate is filtered off on a glass–cloth filter, washed with water, and dried. Recrystallization from a mixture of alcohol and benzene (1 : 1) and then from n-butyl alcohol gives 24.5 g of XI.

Compounds X and XII were prepared analogously.

3,4-Diphenyl-6-methylthiazolo[3,2-c]-1,2,4-triazine (XIII). A mixture of 2.67 g XI, 0.93 g chloroacetone and 10 ml n-butyl alcohol is heated to boiling and boiled for several minutes until complete dissolution is obtained. The light yellow solution is cooled to precipitate white crystals, which are recrystallized from n-butyl alcohol. The yield of the hydrochloride of XIII (XIV) is 2.6 g. On treatment with aqueous sodium-bicarbonate solution, the white crystals of XIV are converted into yellow crystals of the free base XIII.

3,4-Diphenyl-6-ketothiazolo[3,2-c]-1,2,4-triazine (XV) and its hydrochloride (XIV) were prepared analogously.

LITERATURE CITED

1. USA Patent No. 6410823 (1965); Chem. Abstr., 63, 13,295 (1965).
2. Belgian Patent No. 615152 (1962); Chem. Abstr., 58, 12,586 (1963).
3. S. Rossi, Gazz. Chir. Ital., 83, 133 (1953).

4. P. Mantegazza, R. Tomasini, R. Fusco, et al., Arch. Int. Pharmacodyn., 95, 123 (1953).
5. S. Rossi, Atti Acc. Naz. Lincei R. C. Classe Sci. Fis. Mat. Nat., 14, 113 (1953).
6. J. Klosa, Arch. Pharm. (Weingheim), 288, 465 (1955).
7. R. Fusco, S. Rossi, F. Mantegazza, et al., Ann. Chim. (Rome), 42, 94 (1952).
8. V. E. Bogachev and M. G. Fomenko, Author's Certificate No. 175968 (1965); Byull. Izobret., No. 21, 20 (1965).
9. D. H. Tedeschi, R. E. Tedeschi, and E. J. Fellows, Arch. Int. Pharmacodyn., 132, 172 (1959).
10. M. R. Chance, J. Pharmacol. Exp. Ther., 87, 214 (1946).
11. C. Morpurgo, Arch. Int. Pharmacodyn., 137, 84 (1962).