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Pharmacology of Ethyl Thioncarbamate

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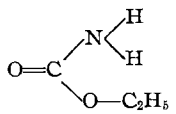
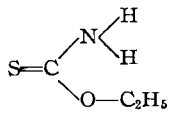
The introduction of sulfur into the molecule of certain of the barbiturates produces more rapid onset of the hypnotic effect. This suggests the possibility of introducing sulfur into the molecule of other hypnotics and this report deals with the preparation and pharmacological properties of ethyl thioncarbamate which differs from ethyl carbamate (urethane) by having a sulfur atom replacing the oxygen of the carbonyl group. The following formulas show this relation:



Ethyl carbamate
(Ethyl urethane)



Ethyl thioncarbamate
(Ethyl oxythiocarbamate)
(Ethyl thionurethane)

The ethyl thioncarbamate was prepared by the method of Salomon (1). This method depends upon the formation of potassium ethyl xanthogenate by the reaction of carbon disulfide, potassium hydroxide and ethyl alcohol. The potassium is then replaced by an ethyl group as a result of reaction with ethyl bromide. After washing and drying, the resulting sulfocarbonyl-oxyethylsulfethyl is mixed with alcoholic ammonia, placed in an ice bath, and ammonia gas passed through it. This yields ethyl mercaptan and ethyl thioncarbamate which is secured in crystalline form by evaporation of the reacting media. The compound was recrystallized from ether and washed with petroleum ether. The yield was 35 per cent; monoclinic crystals, melting point, 38° C. (uncorr.); nitrogen determinations by the Kjeldahl method gave 13.80, 13.93 per cent (theoretical 13.33, Wheeler and Barnes (2) reported 13.02). Ethyl thioncarbamate is soluble in water to the extent of 2.3 per cent at 22° C. Upon standing in open air, it volatilizes. A 25 per cent solution in alcohol is stable and was, therefore, used as a stock solution. For pharmacological experiments, this was diluted with water to make a 2.5 per cent solution which contained 10 per cent alcohol.

This compound was prepared by Salomon in 1874 and a paper published by Smith (3) in 1893 reports the pharmacological action of thioncarbamate and thiolcarbamate. A dog after a total dose of 0.5 Gm. of the thioncarbamate by mouth became very depressed and restless. He ate a little but vomited. The next day albumin appeared in the urine. The dog became weaker,

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showed a black tongue and died during the following night. Sections of the stomach mucosa were red and swollen in the cardiac section as were the intestines. The tubules and urinary canals of the kidney were partly filled with red corpuscles and the entire substance of the brain showed vascular destruction, the white matter being sprinkled with red. The urine, however, did not have the characteristic odor of ethyl thioncarbamate. No thiocyanate was found in the urine or kidneys.

Imase (4) has recently reported that phenyl thiourethane depresses the central nervous system, slows respiration, lowers blood pressure of rabbits and mice and depresses the activity of isolated heart and smooth muscle of the intestine and uterus. Blood vessels of perfused toad legs and the rabbit's ear dilated with small doses and constricted with large amounts.

EXPERIMENTAL

Determination of Lethal and Effective Doses.—These determinations were made on mixed adult albino rats. Injections were all made intraperitoneally. There were complications caused by delayed death which occurred even after several days. able I shows results of administration of various doses of this compound. Deaths are tabulated in two groups: those dying within 48 hours after administration and those dying later. The loss of righting reflex was taken as the time of onset of the hypnotic effect and its reappearance was taken as the time of recovery. On this basis an effective dose is 275 mg. per Kg. It produces loss of righting reflex in 1.5 minutes and recovery in 7.5 minutes. A few similar determinations were made with ethyl urethane. These showed loss of righting reflex with 800 mg. per Kg. to take place in 7 minutes and recovery in 16 minutes. The lethal dose of ethyl urethane was found to be about 2 Gm. per Kg.

The ethyl thioncarbamate, therefore, is effective in much smaller doses than the ethyl urethane. The striking thing, however, in these experiments was the death which occurred several days after the administration of the ethyl thioncarbamate.

General Effects of Ethyl Thioncarbamate.—Observations were now made on cats and rabbits after the intravenous injection of various doses of the ethyl thioncarbamate. The general effects on cats with various doses were as follows: An intravenous dose of 100 mg. per Kg. produced immediate loss of the righting reflex and disappearance of the pain response. Recovery was very rapid. In two or three minutes the righting reflex and cortical responses as measured by placement reactions had returned. For ten to twelve hours the cat was apparently normal although quite restless. Gradual depression then set in and death occurred sixty hours after the administration of the drug.

With larger doses of 150 mg. per Kg. there was immediate loss of the righting reflex after injection. There was also imminent danger of respiratory failure, but artificial respiration usually restored this function. The righting reflex was lost for about 18 minutes. Recovery, however, was not complete for some time later, but at the end of about 45 minutes cortical placement reactions were present and the animal would eat. From this time on there was gradually increasing depression. At 12 hours the righting reflex was very sluggish and at 18 hours it was absent. Respiration became very slow and rectal temperature fell to 24° C. Death occurred at the end of 60 to 70 hours. Congestion of the vital organs was observed and the bladder contained bloody urine.

Doses of 50 mg. per Kg. both intravenously and intraperitoneally resulted in a short period of depression followed by recovery but subsequently death occurred in about 70 hours. With smaller doses there was no depression observed and the animals generally survived. Rabbits receiving comparable doses showed effects similar to those just described for cats.

Table I.—Effect of Various Doses of Ethyl Thioncarbamate on Albino Rats after Intraperitoneal Administration

Dose, mg. per Kg.	No. of Rats	No. Losing Righting Reflex	Righting Reflex Lost, Average Min.	Righting Reflex Absent, Average Min.	Number Dying in Less than 48 Hours	Number Dying in More than 48 Hours
200	10	1	1.5	3	..	..
250	10	5	1.5	7.5	..	1
275	10	9	1.4	24.5	..	1
300	16	13	1.4	23.0	..	1
325	18	16	1.3	274.1	1	2
350	13	11	1.2	144.0	1	1
375	10	10	1.05	517.0	2	3[a]
400	13	13	1.1	555.0	2	8
425	13	13	1.0	Not regained	10	3
450	6	6	1.0	Not regained	6	0
475	3	3	1.0	Not regained	3	0

[a] One died after 5 days. One died after 7 days.

Respiration and Blood Pressure.—Kymographic tracings of respiration and blood pressure on cats and rabbits anesthetized with ether showed that the intravenous administration of 75 mg. per Kg. produced stoppage of respiration. Smaller doses slowed respiration. This respiratory depression was apparently central because after cessation of respiration it was possible to stimulate the sectioned phrenic nerve and observe contraction of the diaphragm. The heart continued to beat for some time after respiratory failure and it was possible to restore the animal by artificial respiration or by the adminstration of picrotoxin in adequate doses.

Except with very small doses, it was found that blood pressure fell after intravenous administration of the compound. With progressively larger doses the fall in blood pressure became greater. For example, in doses of 25 mg. per Kg. the blood pressure dropped 15 mm. of mercury, with 50 mg. the fall was 31 mm., while with 75 mg. the fall was 55 mm. The blood pressure returned to its original level in about three minutes in all of these cases.

Stimulation of the vagus and cervical sympathetic nerves produced characteristic effects on the heart and pupil under all conditions of dosage and therefore ethyl thioncarbamate, like urethane, is apparently without autonomic action.

Smooth and Cardiac Muscle Experiments.—Application of various concentrations of ethyl thioncarbamate to the smooth muscle of the intestine in an isolated tissue bath showed that a concentration of 1:800 stopped contractions. Recovery took place when washed with Ringer's solution. A 1:1600 solution stopped the contractions for 1.5 minutes after which they resumed their normal rate. A solution of 1:3200 produced relaxation of the muscle for about one-quarter of a minute after which the rythum was resumed. Rat uterus was relaxed by a concentration of 1:800.

The rate of isolated frog heart was depressed by concentrations of 1:200 and stopped in concentrations of 1:150. In all cases, control experiments with equivalent amounts of alcohol showed that the alcohol contained in the original stock solution was without noticeable effect.

Pathology.—The delayed death described above seems most likely due to damage of the various tissues. We therefore ran liver function tests (bromsulfthalein) and kidney function tests (phenolsulfonphthalein) at various times after the administration of the drug. The liver function test made on rabbits showed maximum retention of the dye in the blood after oral doses of 350, 375 and 400 mg. per Kg. 24 hours after administration. With a dose of 300 mg. per Kg., the dye was only partially removed from the blood at the end of 24 hours and not completely at the end of 48 hours. The phenolsulfonphthalein test also made on rabbits showed a decrease in the excretion of the dye 24 hours after the administration of 300 mg. per Kg. All of the animals receiving the above doses eventually died.

Microscopic sections were prepared of liver, kidney and spleen of a rabbit which was killed 3 days after it has received 350 mg. per Kg. by mouth. These showed marked destruction of the cells lining the tubules of the kidney, the cell walls being ruptured. Liver and spleen sections also showed degenerative changes to have taken place.

DISCUSSION AND CONCLUSIONS

Investigation of the pharmacology of ethyl thioncarbamate indicates that the replacement of oxygen in the carbonyl group of ethyl carbamate (urethane) by sulfur produces marked changes in the action of this compound. The depression produced is more rapid in its onset and shorter in its duration. This is similar to the changes occurring in the action of the barbiturates when the carbonyl oxygen is replaced with sulfur to form the thiobarbiturates.

The secondary effects produced by the ethyl thioncarbamate are such that the compound is definitely contraindicated for clinical use. These secondary effects are mainly damage to liver and kidney tissue. It is possible that these undesirable side actions are due to a breakdown product of the ethyl thioncarbamate. From the chemistry of the compound, the breakdown products may be ethyl mercaptan and cyanuric acid. However, ethyl mercaptan cannot be detected on the exhaled breath of animals given the original compound. Another possible breakdown product may be thiocyanic acid but we were able to verify Smith in not being able to detect this in blood or urine. However, the presence of a very characteristic odor in the breath and also in the tissues and urine of animals leads to the belief that the original compound does break down in the body. Because of the probability that the undesirable effects are due to a break down product and the possibility that higher homologues of this series might not break down in the same way, they will be studied.

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