Scientific Edition

JOURNAL OF THE AMERICAN PHARMACEUTICAL ASSOCIATION

JUSTIN L. POWERS, EDITOR, WASHINGTON, D. C.

VOLUME XXXI | *DECEMBER, 1942* | NUMBER 12 CONSECUTIVE NO. 23

The Action of Calycanthine*

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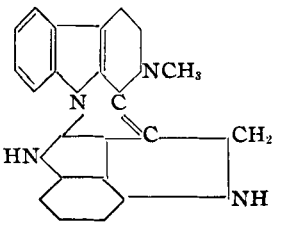
The seeds of *Calycanthus glaucus*, or colloquially "bubby," have been a source of poisoning to cattle and sheep in the Tennessee Valley. Eccles (1) was the first to isolate from the seeds an alkaloid named calycanthine although he did not give the melting point or any analytical data. Wiley (2) made a chemical examination, but added very little information to the subject. Characterization of calycanthine was achieved by Gordin (3) who proposed the empirical formula $C_{11}H_{14}N_2$. Interestingly, the same author obtained isocalycanthine from a second lot of the seeds of *Calycanthus glaucus* (4, 5). Späth and Stroh (6) revised Gordin's empirical formula to $C_{22}H_{28}N_4$ by doubling.

Manske, in a series of brilliant investigations (7, 8, 9, 10), isolated calycanthine from *Calycanthus floridus* and *Meratia praecox*, arrived at the correct empirical formula $C_{22}H_{26}N_4$, and advocated the following structural formula:



It should be noted that the molecule of calycanthine has two tryptamine nuclei, one being reduced. Manske has also isolated calycanthine from *Calycanthus glaucus* and *C. occidentalis*, and observed that Gordin's isocalycanthine is a physical isomer of calycanthine because recrystallization or inoculation gives rise to calycanthine at once (private communication).

An attempt to solve the structure of calycanthine was undertaken by Barger and his associates prior to his death (11). From the seeds of *C. floridus*, Barger, Jacob and Madinaveitia (12) succeeded in isolating a new alkaloid called calycanthidine, $C_{13}H_{16}N_2$. Recently, Manske (13) obtained cocositol, $C_6H_{12}O_6$, an optical isomer of inositol, from the leaves of *C. floridus* and *C. glaucus*.

The plant *Meratia praecox*, from which Manske (7) isolated calycanthine, has been

* Received July 9, 1942, from the Lilly Research Laboratories, Eli Lilly and Company, Indianapolis, Ind.

† The authors are indebted to Messrs. Charles L. Rose, Robert C. Anderson, E. Brown Robbins and Francis Henderson for their valuable assistance in several of the experiments.

widely cultivated in China. The native name is "La Mei." It is so closely related to the *Calycanthus* species that it has been identified by some authorities as *Calycanthus praecox* (14). It blossoms in midwinter, namely, January. Its fragrant flowers, strung on a thin wire, are often worn by women as hair ornaments. They are also used in medicine as a cooling and sialagogue remedy. Apparently the Chinese have not recognized the toxic properties of its seeds since no mention of them has been made in literature.

Cushny (15) conducted a pharmacologic study of calycanthine, chiefly on its acute toxicity and the cardiac depressant action. McGuigan and von Hess (16) concluded that isocalycanthine and calycanthine had an identical effect in animals.

Dr. Richard H. F. Manske, National Research Laboratories, Ottawa, Canada, provided us with a generous supply of calycanthine in the form of hydrochloride. It was proposed to extend the pharmacologic studies with this alkaloid, which had been so admirably initiated by Cushny (15).

EXPERIMENTAL

Calycanthine hydrochloride is easily soluble in water. A 1–5% solution foams readily upon shaking, but it does not hemolyze the rabbit's erythrocytes in various concentrations from 1:100,000 to 1:100.

1. Acute Toxicity.—By intravenous injection, the median lethal doses were determined in mice and rats as shown in Table I. The alkaloid is more than twice as toxic to rats as to mice. Lethal doses were followed, immediately or within 10–15 min., by a series of clonic convulsions, then succeeded by tonic convulsions. Death usually occurred in from $^1/_2$ to 3 hrs.

Calycanthine hydrochloride appears to be even more toxic to larger laboratory animals such as rabbits. Doses varying from 10 to 40 mg./Kg. were invariably fatal. The maximal tolerated dose was 7.5 mg./Kg. A total of 16 rabbits was employed. Restlessness, tonic convulsions, cyanosis and twitching of the legs preceded death. The animal was often in an opisthotonos position during convulsions.

TABLE I.—ACUTE TOXICITY OF CALYCANTHINE HCl

Animal	Dose, Mg./Kg.	Number Died / Number Injected	Median Lethal Dose ± Standard Error, Mg./Kg.
Mice	30	0/10	43.79 ± 1.89
	34	0/8	
	40	4/10	
	50	7/8	
	60	9/10	
	70	5/5	
Rats	13	1/10	17.16 ± 0.82
	16	2/10	
	20	8/9	

"Sodium Amytal" (Sodium Iso-amyl Ethyl Barbiturate, Lilly) intraperitoneally injected prior to calycanthine afforded a very slight protection. For example, calycanthine alone in a separate test killed all of 5 mice in the amount of 40 mg./Kg. The same dose following the barbiturate killed 6 out of 8 mice. The antidotal action of "Sodium Amytal" is therefore negligible if it is compared with that against strychnine (17), coriamyrtin and picrotoxin (18) and tutin (19). Cushny (15) believed that calycanthine affected a higher part of the central axis primarily. In the light of present findings, the alkaloid probably acts on a center higher than the medulla since "Sodium Amytal" is most effective in detoxifying poisons that act on the medulla and spinal cord.

In frogs calycanthine hydrochloride in doses of 0.2, 0.5 and 1 mg./Gm., injected into the lymph sac, caused gradual depression and prostration. The animal lay flat on its abdomen after a lapse of an hour or longer. When placed in a new position, its hindlegs often stretched out, toes twitched and the head retracted. At no time was there occurrence of typical spasms or convulsions. The ventricle stopped at diastole $4^1/_2$ to 6 hrs. after the administration of the largest dose, namely, 1 mg./Gm. Injection of 0.1 mg./Gm. did not appear to affect the frog.

The reaction of frogs to toxic doses of calycanthine is thus one of depression, while that of mice, rats and rabbits is certainly one of stimulation. This contrast of response between warm-blooded animals, on the one hand, and cold-blooded animals as represented by frogs, on the other, is probably comparable to what was previously observed with sodium 1:3-dimethylbutylethyl-barbiturate (20).

2. Blood Sugar and Count.—Two of the 3 rabbits were injected intravenously with 7.5 mg. of calycanthine hydrochloride per Kg. of body weight; and one, 5 mg. per Kg. At various intervals, but more frequently during the first hour, their blood sugar was determined according to the procedure of Hagedorn and Jensen (21), and their erythrocytes and leucocytes, including the differential, were counted by the usual methods. Hyperglycemia definitely occurred as shown in Fig. 1. The results in the other rabbits were confirmatory. The changes in blood counts were slight and all within normal limits of variation.

3. Circulation.—Just as Cushny (15) reported, calycanthine was found to produce a fall of arterial blood pressure in anesthetized animals. The average effective dose in cats was 10–20 mg., and that in dogs. 40–60 mg. There was a slight decrease in heart rate with an increase in amplitude of respira-

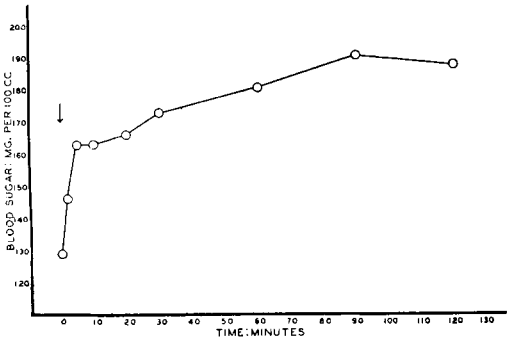


Fig. 1.—Action of calycanthine on blood sugar.

Rabbit, female, weighing 2.05 Kg., was given intravenously (at arrow) 7.5 mg. of calycanthine HCl/Kg. of body weight. Note the increase of blood sugar.

tion during the fall of carotid blood pressure. Atropinization did not nullify the depressor action. Recovery was prompt—a matter of 3–10 min.

There was a definite depression of the ventricular contraction under the influence of calycanthine. Figure 2 shows a myocardiographic tracing of a cat recorded with a dose of 10 mg. It is closely comparable to the one published by McGuigan and von Hess (16) on isocalycanthine.

4. Isolated Intestines and Uteri.—Stimulation resulted upon the addition of calycanthine hydrochloride on the isolated rabbit's intestines and uterus. Examples are given in Fig. 3. Ergotoxine ethanesulfonate did not antagonize the effect on the uterus, so that the action was not due to sympathetic

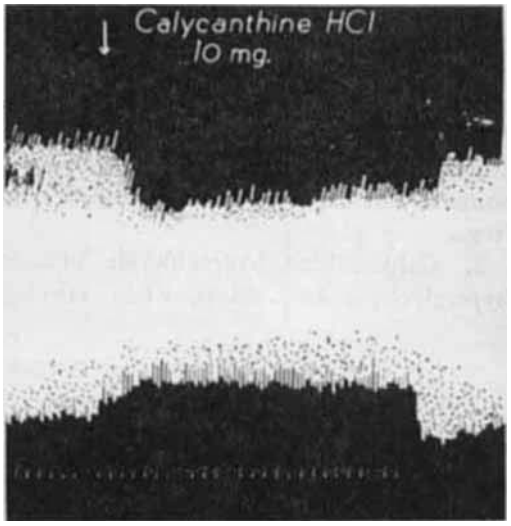


Fig. 2.—Action of calycanthine on mammalian heart.

Cat, female, weighing 2.817 Kg., was decerebrated and pithed, under artificial respiration with an open chest. The myocardiogram of the right ventricle was recorded. At arrow, a dose of 10 mg. of calycanthine HCl was injected by the femoral vein. The decrease in amplitude of contractions is obvious—with rapid recovery.

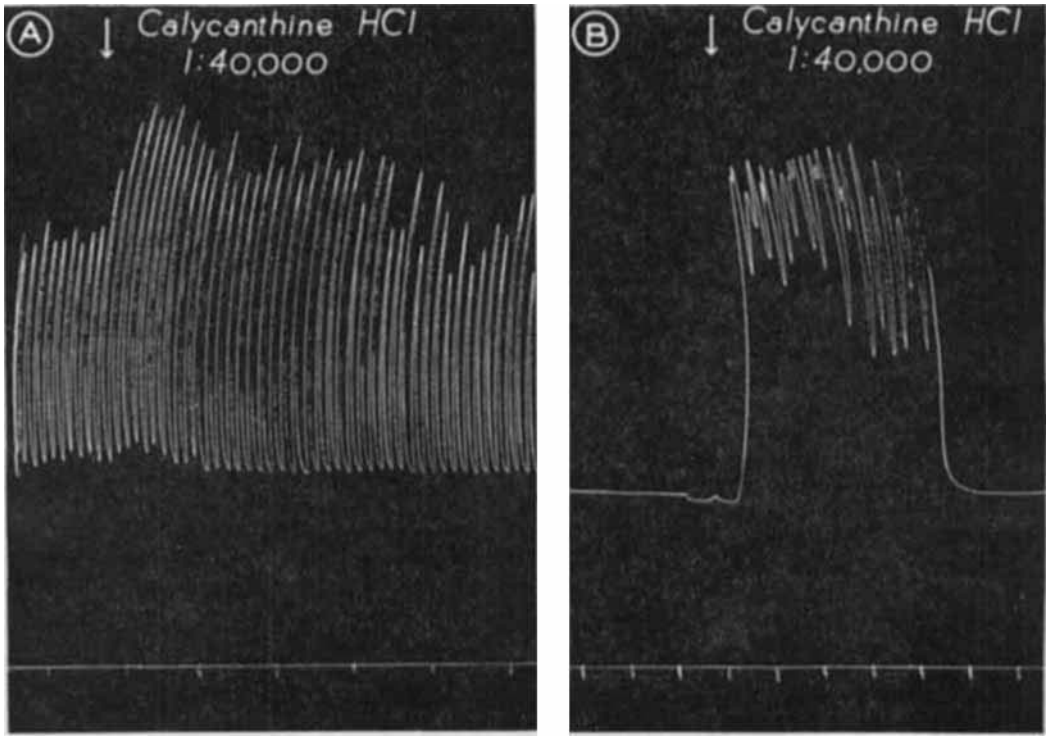


Fig. 3.—Action of calycanthine on isolated smooth muscle organs.

A.—A strip of isolated rabbit's intestine, immersed in Tyrode's solution at 38° C., was stimulated by calycanthine HCl in the concentration of 1:40,000.

B.—A strip of isolated rabbit's uterus, immersed in Tyrode's solution, was contracted by calycanthine HCl in the same concentration.

stimulation. Calycanthine caused a very feeble contraction of the isolated guinea pig's uterus in the concentration of 1:10,000.

SUMMARY

1. The acute toxicity of calycanthine hydrochloride has been determined in mice, rats, rabbits and frogs. Toxic doses cause stimulation in the first three species of animals, but depression and prostration in frogs.

2. Calycanthine hydrochloride induces hyperglycemia in rabbits when injected intravenously. No apparent changes in the counts of erythrocytes and leucocytes take place.

3. Calycanthine hydrochloride reduces blood pressure and depresses the cardiac contraction in anesthetized cats. Fall of blood pressure has also been observed in anesthetized dogs.

4. Calycanthine hydrochloride produces stimulation of the isolated rabbit's intestines and uterus. It has only slight effect upon the isolated guinea pig's uterus.

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Ketone Alcohols*,†

I. Derivatives of 2-Methyl-pentanol-2-one-4

By Clifton Eugene Miller‡

It was reported in a previous paper (1) that preliminary pharmacological tests indicated that the dibromoureide of 2-methyl-pentanol-2-one-4 (diacetone alcohol) possessed hypnotic action. It seemed of interest therefore to prepare other derivatives and to determine, if possible, whether the degree of hypnosis might be increased or diminished by the substitution of various groups or atoms. This paper presents an introductory report on the preparation of certain acetyl, chlorine, nitrogen and sulfur derivatives of 2-methyl-pentanol-2-one-4.

* A contribution from the Department of Pharmaceutical Chemistry, North Dakota Agricultural College, Fargo, N. D.

† Presented to the Scientific Section of the A. PH. A., Detroit meeting, 1941.

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EXPERIMENTAL

Preparation of 2-Methyl-2-chloro-pentanone-4.—One mole of 2-methyl-pentanol-2-one-4 and three moles of hydrochloric acid, sp. gr. 1.175, were placed