The preparatory extraction with alcohol had also the advantage that some of the unpalatable and bitter substances are also removed so that the watery extract has a pleasant taste.

On the basis of these experiences, we tried to prepare a highly active fluidextract through concentration of an infusion, made from alcohol-treated leaves. The infusion was evaporated to one-fifth of its volume *in vacuo*, so that 1 cc. corresponded to 0.5 Gm. of leaves. It resulted in a fluidextract of syrupy consistency.

The results of the bioassay of this fluidextract are shown in Table IX.

Table IX.—Assay of Fluidextracts Prepared by a Method Discussed in this Paper

Extract	Dilution	Animals Used	No. of Positive Results	Positive in %
M	1:20	12	11	91
	1:30	12	3	25
	1:40	12	1	10
N	1:20	10	8	80
	1:30	10	3	30
	1:40	10	2	20
O	1:20	10	9	90
	1:30	10	4	40
	1:40	10	0	0

As 0.5 cc. of the 1:30 diluted extract equals 0.008 Gm. of leaves, 0.5 cc. of the undiluted extract is equal to 0.24 Gm. of leaves and 1 cc. equals 0.48 Gm. of leaves.

The data of Table IX show that the cathartic activity is also practically equal to the potency of 0.5 Gm. of leaves. This means that the extract prepared by the described method contains all of the cathartic activity of the original material, so that no loss has occurred during the process.

With the help of the bioassay, we have succeeded in showing that the U. S. P. Fluidextract of Senna is not satisfactory in activity and that the factors responsible for this low activity can easily be avoided.

SUMMARY

1. A simple method for the bioassay of senna leaves and senna preparations is described, based on the cathartic action of the senna principles on mice. The amount of experimental data obtained till now is not sufficient for the statistical evaluation of the method; it has been shown, however, that the discussed method is quite suitable for practical purposes.

2. With the help of this method, it has been shown that the low p_H and the alcoholic content of the U. S. P. Fluidextract of Senna diminish the cathartic activity of this preparation.

The author is indebted to Professor E. N. Gathercoal of the University of Illinois School of Pharmacy for his helpful interest in the present experiments.

REFERENCES

(1) Führer, H., *Arch. exp. Path. Pharmakol.*, 105 (1925), 251; Loewe, S., and Faure, G., *Ibid.*, 107 (1925), 170

(2) Munch C. J., "Bioassays: A Quantitative Handbook of Pharmacology," page 773. William and Wilkins Co., Baltimore, 1931.

(3) Straub, W., and Gebhardt, H., *Arch. exp. Path. Pharmakol.*, 181 (1936), 400.

Absorption and Toxicity of Sodium and Potassium Thiocyanates*

By Robert C. Anderson and K. K. Chen†

Since the observation of Pauli (1) in 1903 that sodium thiocyanate (synonymous with sulfocyanate or rhodanate) lowered the blood pressure in hypertension, numerous clinical reports have appeared in medical literature. Table I summarizes the articles of American and Canadian origin. Briefly speaking, sodium or potassium thiocyanate when properly used reduces both systolic and diastolic pressures in patients with hyperpiesis, and causes subjective improvements. Toxic symptoms of various forms, however, may occur frequently. Thus extensive cutaneous lesions have been repeatedly recorded by Logefeil (7), Weis and Ruedemann (25), Ayman (26), Tyrrell (27), Baker and Brunsting (28), Green and Snow (29), Healy (30) and others. Fatalities have been attributed to thiocyanate therapy—the latest being described by Healy (30), Goldring and Chasis (31) and Garvin (32). Weakness and pain resembling angina pectoris were illustrated by Palmer and Sprague (33). Careful analysis and classification of the various untoward effects from the use of thiocyanate were made by Goldring and Chasis (31), and particularly by Wald, Lindberg and Barker (34).

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Table I.—Summary of Clinical Results with Thiocyanates for the Treatment of Hypertension, Gathered from American Literature

Author	Salt of Thiocyanate Used	Dose	Number of Cases Treated	Result
Le Roy (2)	Sodium	0.004–0.08 4.i.d.	7	All favorable
Nichols (3)	Sodium	5 grains t.i.d.	12	Effective
Gager (4)	Potassium	1½ grains t.i.d.	35	Uniformly satisfactory; blood pressure reduced in all except 3; side reactions noted
Smith and Rudolf (5)	Sodium	2½ grains b.i.d. to 5 grains t.i.d.	72	Blood pressure of 66 cases lowered; toxic symptoms recorded
Palmer, Silver and White (6)	Potassium	1½ grains once to t.i.d.	59	More than 42% favorable; untoward but not alarming symptoms mentioned
Logefeil (7)	Potassium	0.32 Gm. t.i.d.	24	Blood pressure of 21 cases reduced; no effect in 2; toxic symptoms observed
Fineberg (8)	Potassium	1½–5 grains t.i.d.	58	With the larger dose, 57% showed a drop of blood pressure
Maguire (9)	Potassium	1½ grains t.i.d.	4	Blood pressure all lowered and condition improved
Borg (10)	Sodium or potassium	2–5 grains t.i.d.	24	92% of the cases showed systolic drop; only 2 cases failed; toxic effects listed
Ayman (11)	Potassium	0.1–0.2 Gm. t.i.d. or 4.i.d.	26	Large doses caused fall of blood pressure but with toxic symptoms; small doses were ineffective
Saleeby (12)	Potassium	1½ grains t.i.d.	88	Gratifying in 90% of the cases
Meakins and Scriver (13)	Sodium	4–12 grains daily	7	Blood pressure lowered in 3 cases; no effect in others
Egloff (14)	Potassium or sodium	0.6–1 Gm. daily	25	Blood pressure lowered only in 2 cases; uneffected in 23
Goldring and Chasis (15), (16)	Sodium or potassium	2½–25 grains daily	50	Effective in 31%; toxic manifestations in 17%
Palmer (17)	Potassium	1½–5 grains once to t.i.d.	35	Blood pressure in 31% of the cases; toxic symptoms in 9 cases
Bolatin (18)	Potassium	1½–5 grains t.i.d.	99	68.8% improvement in men, 23.5% improvement in women; untoward symptoms occurred in only 5 patients
Barker (19)	Sodium or potassium	0.3–1 Gm. daily	45	Relief in 35 patients; blood concentration of thiocyanate emphasized
Griffith and Lindauer (20)	Potassium	0.3–0.8 Gm. daily	16	Improvement in 10 cases
Steidl (21)	Potassium	0.3–0.8 Gm. daily	3	Blood pressure reduced in all 3
Massie, Ethridge and O'Hare (22)	Sodium	0.2–0.8 Gm. daily	14	All symptomatically improved; toxic reactions reported
Doles (23)	Sodium	2½–12 grains daily	300	Blood pressure lowered in cases of hypertension with macrocytosis of R.B.C.
Doles (24)	Sodium	7½ grains daily	10	All had macrocytosis of R.B.C.; blood pressure therefore all reduced

It was Barker (19), (35) who first advocated the determination of blood concentration of thiocyanate as a criterion for the safe administration of this potentially toxic drug. He found that the optimal concentration of the thiocyanate ion in the blood was 8 to 12 mg. per 100 cc. At such levels, toxic reactions were reduced to a minimum, while the blood pressure was effectively lowered in the majority of cases. The results of subsequent workers, such as Griffith, Lindauer and Campbell (20), Steidl, Steenken and Heise (21), Massie, Ethridge and O'Hare (22) and Binger (36) apparently all supported Barker's contentions.

The pharmacology and toxicology of thiocyanate up to 1923 were reviewed by Hunt (37). The acute lethal doses in experimental animals as compiled by Hunt are shown in Table II. An interesting fact is that thiocyanate normally occurs in human saliva, perhaps as a result of detoxification of cyanide split off from digestion of proteins. If a small amount of cyanide is ingested, it is converted into thiocyanate, as demonstrated by Bodansky (38). Thiocyanate is easily absorbed and excreted in urine and other body fluids (37). Wallace and Brodie (39), (40) concluded that thiocyanate, like the halides, was distributed in

Table II.—Acute Toxicity of Thiocyanates in Animals Recorded in Literature

Animal	Salt of Thiocyanate	Administration	Dose, Gm. per Kg.	Effect
Frogs	Potassium	Oral	0.3	Always fatal
Frogs	Potassium	Oral	0.2	Never fatal
Frogs	Potassium	By lymph sac	0.25–0.35	Lethal
Frogs	Potassium	Intramuscular	0.4	Always fatal
Frogs	Potassium	Intramuscular	0.05	Never fatal
Frogs	Potassium	Intramuscular	0.1–0.25	Variable
Pigeons	Potassium	Subcutaneous	0.5	Lethal
Pigeons	Potassium	Intramuscular	0.75	Lethal
White mice	Sodium	Subcutaneous	0.4–0.6	Lethal
White rats	Potassium	Subcutaneous	1.0	Lethal
Guinea pigs	Potassium, sodium or ammonium	Oral	0.6–0.8	Death in $4^1/_2$ to 24 hrs.
Guinea pigs	Potassium	Subcutaneous	0.75	Lethal
Guinea pigs	Sodium	Subcutaneous	0.5	Lethal
Rabbits	Potassium	Oral	0.5–0.91	Death in 6 hrs. to 4 days
Rabbits	Potassium	Oral	1.0	Always fatal
Rabbits	Potassium	Oral	0.5	Never fatal
Rabbits	Potassium	Subcutaneous	0.55	Always fatal
Rabbits	Potassium	Subcutaneous	0.4	Never fatal
Rabbits	Potassium	Intravenous	0.15	Always fatal
Rabbits	Potassium	Intravenous	0.06	Never fatal
Rabbits	Potassium	Intravenous	0.1	Variable
Rabbits	Sodium	Intravenous	2.6	Lethal
Dogs	Potassium	Intravenous	0.1	Lethal
Dogs	Sodium	Intravenous	0.8	Ineffective

the extracellular fluid. Although sodium thiocyanate depressed oxygen consumption of liver cells (rats), a concentration of 8 to 22 mg. per 100 cc. of blood had no effect as disclosed by Friend and Robinson (41).

Apparently the depressor action of small doses of thiocyanate per os can be more easily demonstrated in patients with hypertension than in animals with normal blood pressure. Its exact mechanism is little understood. Takacs (42) believed that it was due to vagal stimulation. Doles (23), (24) associated it with the chemical reaction between thiocyanate and the iron in the enlarged erythrocytes. While complete clarification of the phenomenon must depend upon further evidence, the following data were obtained to strengthen the toxicological knowledge of this drug.

EXPERIMENTAL

1. Acute Toxicity.—Both sodium and potassium thiocyanates were investigated. They were recrystallized from water, and their moisture content determined, respectively. The water of crystallization of the sodium constituted 3.525 per cent, and that of the potassium salt 0.455 per cent. All the doses were calculated according to the anhydrous forms.

As shown in Table III, potassium thiocyanate by intravenous injection is more than five times as toxic as sodium thiocyanate to mice when their median lethal doses (L. $D._{50}$) are compared. Clonic convulsions, gasping and deaths occurred within 2 to 3 minutes with the potassium salt, while the same were delayed with the sodium salt for 1 to 3 hours. The highly toxic action of the potassium ions upon the heart undoubtedly accounted for the difference.

When given by mouth, the differences in toxicity between the two compounds in both mice and rats are very slight. Statistically, such differences as recorded in Table III are not significant. These results substantiate Barker's statement (19) that there was no choice between the sodium and potassium thiocyanates. Theoretically, however, sodium thiocyanate contains 71.63 per cent of SCN^- while potassium thiocyanate has 59.76 per cent of SCN^- ions.

2. Prolonged Administration.—A group of 10 female rats was each given by mouth 100 mg. of sodium thiocyanate per Kg. of body weight, daily except Saturdays and Sundays for 12 weeks. A similar group was treated with 100 mg. of the potassium salt per Kg., and a third group was used as controls. Before the end of the experiment, 4 rats from the control group, 4 from the sodium thiocyanate group and 2 from the potassium thiocyanate group died accidentally from aspiration. Figure 1A shows the average weight curves of the three groups of animals that survived the entire period of 12 weeks. It is obvious that neither sodium thiocyanate nor potassium thiocyanate inhibited the growth of rats.

The experiment was repeated with double the dose of thiocyanates, that is, 200 mg. per Kg., respectively, in a series of 30 rats divided equally into three groups. As in the previous study, the third group served as controls. The duration of observation in this case was 8 weeks. One rat was lost from a broken leg in the potassium thiocyanate group and three accidentally from the control group. The three average weight curves show a great similarity (Fig. 1B), indicating that in rats sodium or potassium

Table III.—Acute Toxicity of Sodium and Potassium Thiocyanates in Mice and Rats

Animal	Salt of Thiocyanate	Administration	Dose, mg. per Kg.	Number Died/ Number Used	L. D.$_{50}$ ± S. E., mg. per Kg.
Mice	Sodium	Intravenous	453	0/6	
Mice	Sodium	Intravenous	482	5/7	483.5 ± 9.3
Mice	Sodium	Intravenous	521	4/5	
Mice	Sodium	Intravenous	579	5/5	
Mice	Potassium	Intravenous	49.8	0/5	
Mice	Potassium	Intravenous	74.7	3/10	88.2 ± 5.8
Mice	Potassium	Intravenous	99.5	6/10	
Mice	Potassium	Intravenous	139.4	10/10	
Mice	Sodium	Oral	579	2/5	
Mice	Sodium	Oral	627	3/5	598.4 ± 18.3
Mice	Sodium	Oral	675	5/5	
Mice	Sodium	Oral	724	5/5	
Mice	Potassium	Oral	547	1/5	
Mice	Potassium	Oral	597	3/5	594.4 ± 27.0
Mice	Potassium	Oral	647	4/5	
Mice	Potassium	Oral	697	4/5	
Rats	Sodium	Oral	675	1/5	
Rats	Sodium	Oral	772	3/5	764.7 ± 50.9
Rats	Sodium	Oral	868	4/5	
Rats	Sodium	Oral	965	4/5	
Rats	Potassium	Oral	697	1/5	
Rats	Potassium	Oral	796	3/5	
Rats	Potassium	Oral	896	2/5	854.1 ± 66.6
Rats	Potassium	Oral	995	3/5	
Rats	Potassium	Oral	1095	4/5	
Rats	Potassium	Oral	1195	5/5	

thiocyanate in the daily dosage of 200 mg. per Kg. does not impair the animals' growth.

A group of 11 dogs was employed for repeated medication with thiocyanates by mouth—six of which were given various doses of potassium thiocyanate daily except Saturdays and Sundays, and the other five different doses of sodium thiocyanate in the same manner. Since dogs tend to vomit with large doses of thiocyanates, enteric coated tablets of both salts were prepared and administered, each containing 200 mg. Blood concentrations of each animal were determined semiweekly or weekly by a micromethod to be described in the section on absorption.

According to the chronological data in Table IV, it becomes obvious that repeated ingestion of thiocyanate in the dosage of 100 mg. per Kg. or more, with one exception, caused a progressive loss of, weight, appearance of toxic symptoms such as apathy, head-droop, and ataxia, and ultimate death. The blood concentration of thiocyanate in each dog

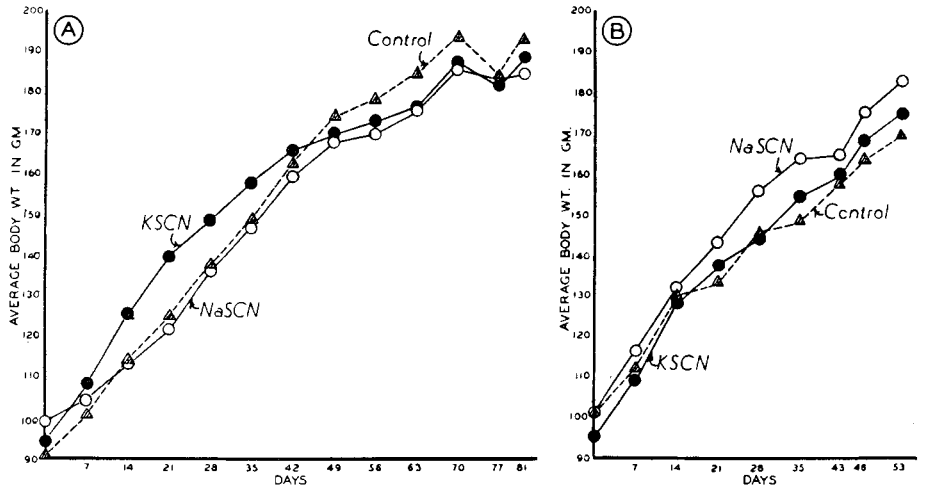

Fig. 1.—Prolonged Administration of Thiocyanate by Mouth in Rats.

A. Daily dose of 100 mg. per Kg. for a period of 12 weeks except Saturdays and Sundays. The curve through hollow circles represents the average weight of 6 rats on sodium thiocyanate, that through solid circles that of 8 animals on potassium thiocyanate, and that through triangles that of 6 control rats.

B. Daily dose of 200 mg. per Kg. for a period of 8 weeks except Saturdays and Sundays. The curves have similar designations. Ten rats survived with sodium thiocyanate, 9 with potassium thiocyanate and 7 as controls.

was consistently high, exceeding 20 mg. per 100 cc. most of the time. Four dogs succumbed in spite of withdrawal of the thiocyanate upon the appearance of toxic manifestations. Dog numbered 72 was the only one that survived 8 doses of 104.2 mg. of potassium thiocyanate per Kg. of body weight. Its blood concentration of thiocyanate finally surpassed 20 mg. per 100 cc. until the drug was withdrawn. Pathological examination of the dead animals by Dr. Paul N. Harris revealed no uniform lesion which could account for their death. In fact, the organs of the dog numbered 14 appeared normal.

Table IV.—Prolonged Administration of Thiocyanates in Dogs by Mouth

Dog Number	Sex	Date	Body Weight, Kg.	Salt of Thiocyanate	Dose, mg. per Kg.	Total Amount, Gm.	Blood Concentration as SCN^-, mg. per 100 Cc.	Remarks
14	F	6- 6-39	9.2	Potassium	108.7	1.0	..	
		6- 9-39	..	Potassium	...	3.0	26.0	
		6-13-39	9.0	Potassium	...	5.0	27.0	
		6-16-39	..	Potassium	...	8.0	..	Dead. No pathological lesions could be detected
63	F	6- 6-39	11.3	Potassium	106.2	1.2	..	
		6- 9-39	..	Potassium	...	3.6	22.0	Dose withheld; unable to stand up
		6-12-39	..	Potassium	...	..	..	Drug resumed
		6-13-39	10.2	Potassium	...	4.8	12.0	
		6-16-39	..	Potassium	...	8.4	19.0	In poor condition
		6-21-39	..	Potassium	...	13.2	21.0	In poor condition
		6-22-39	9.4	Potassium	...	..	..	Drug discontinued; unable to stand, head drooping, eyes glassy
		6-23-39	..	Potassium	...	..	..	Dead. Central necrosis and multiple cavernous hemangiomata of liver, focal necrosis of adrenal cortex, pulmonary edema, mucosal polyps of stomach
65	F	6- 6-39	6.0	Potassium	100	0.6	..	
		6- 9-39	..	Potassium	...	1.8	13.0	
		6-13-39	5.5	Potassium	...	3.0	21.0	
		6-16-39	..	Potassium	...	4.8	24.0	
		6-21-39	..	Potassium	...	6.6	20.0	
		6-22-39	5.2	Potassium	...	..	..	Drug discontinued; unsteady
		6-27-39	4.4	Potassium	...	6.6	31.0	In worse condition
		6-28-39	..	Potassium	...	..	..	Dead. Chronic interstitial nephritis, slight fatty metamorphosis of liver
72	F	6- 9-39	9.6	Potassium	104.2	1.0	..	
		6-13-39	9.2	Potassium	...	2.0	14.0	
		6-16-39	..	Potassium	...	5.0	22.0	
		6-21-39	..	Potassium	...	8.0	23.0	
		6-22-39	9.4	Potassium	...	..	..	Drug discontinued for fear of toxicity
		6-27-39	9.2	Potassium	...	8.0	11.0	
		6-30-39	..	Potassium	...	8.0	4.0	
		7- 3-39	8.6	Potassium	...	..	..	
		7- 7-39	..	Potassium	...	8.0	2.0	
		7-14-39	8.9	Potassium	...	..	..	
		7-17-39	..	Potassium	20.8	..	..	Drug resumed
		7-21-39	8.8	Potassium	...	8.8	5.0	
		7-28-39	9.0	Potassium	...	9.8	6.0	
		8- 4-39	8.9	Potassium	...	10.8	8.0	
		8-11-39	8.9	Potassium	...	11.8	6.0	
		8-17-39	8.8	Potassium	...	12.6	4.0	
		8-25-39	8.8	Potassium	...	13.8	..	
		9- 1-39	9.2	Potassium	...	14.8	2.0	
		9- 8-39	9.4	Potassium	...	15.8	6.0	
		9-15-39	9.1	Potassium	...	16.8	6.0	Medication stopped
		10- 4-39	9.4	Potassium	...	..	..	In excellent condition throughout
24	F	6-26-39	8.2	Potassium	24.4	0.2	..	
		6-30-39	..	Potassium	...	0.8	7.5	
		7- 3-39	7.0	Potassium	...	1.0	..	
		7- 7-39	..	Potassium	...	1.8	2.0	
		7-14-39	7.3	Potassium	...	2.8	7.0	
		7-21-39	7.2	Potassium	...	3.8	7.0	
		7-28-39	7.1	Potassium	...	4.8	6.0	
		8- 4-39	7.1	Potassium	...	5.8	10.0	
		8-11-39	7.1	Potassium	...	6.8	10.0	
		8-17-39	7.1	Potassium	...	7.8	12.0	

Table IV (*Continued*)

		8-25-39	7.2	Potassium	...	8.8	..	
		9- 1-39	7.4	Potassium	...	9.8	2.0	
		9- 8-39	7.6	Potassium	...	10.8	5.0	
		9-15-39	7.3	Potassium	...	11.8	5.0	Medication stopped
		10- 4-39	7.4	Potassium	...	..	..	In excellent condition throughout
32	M	6-26-39	9.2	Potassium	21.7	0.2	..	
		6-30-39	..	Potassium	...	0.8	10.0	
		7- 3-39	8.2	Potassium	...	1.0	..	
		7- 7-39	..	Potassium	...	1.8	2.0	
		7-14-39	8.0	Potassium	...	2.8	9.5	
		7-21-39	7.6	Potassium	...	3.8	5.0	
		7-28-39	7.7	Potassium	...	4.8	6.0	
		8- 4-39	7.7	Potassium	...	5.8	6.0	
		8-11-39	7.7	Potassium	...	6.8	6.0	
		8-17-39	7.8	Potassium	...	7.6	4.0	
		8-25-39	7.9	Potassium	...	8.8	..	
		9- 1-39	8.2	Potassium	...	9.8	4.0	
		9- 8-39	8.6	Potassium	...	10.8	5.0	
		9-15-39	8.3	Potassium	...	11.8	5.0	Medication stopped
		10- 4-39	9.0	Potassium	...	..	..	In excellent condition throughout
60	F	6- 6-39	11.6	Sodium	103.4	1.2	..	
		6- 9-39	..	Sodium	...	3.6	26.0	
		6-13-39	10.0	Sodium	...	6.0	27.0	
		6-16-39	..	Sodium	...	9.6	32.0	
		6-21-39	..	Sodium	...	13.2	35.0	
		6-22-39	7.6	Sodium	...	..	..	Drug discontinued; wabbly; tremors
		6-23-39	..	Sodium	...	..	..	Dead. Bronchopneumonia, pulmonary edema, emaciation, slight cloudy swelling of kidneys, congestion of liver and kidneys
61	F	6- 6-39	9.8	Sodium	102	1.0	..	
		6- 9-39	..	Sodium	...	3.0	26.0	
		6-13-39	8.8	Sodium	...	5.0	27.0	
		6-16-39	..	Sodium	...	8.0	24.0	
		6-21-39	..	Sodium	...	11.0	37.5	
		6-22-39	8.0	Sodium	...	11.0	..	Drug discontinued; ataxia
		6-27-39	7.0	Sodium	...	11.0	27.0	
		6-29-39	..	Sodium	...	..	..	Dead. Pulmonary edema and congestion, acute purulent bronchitis
59	F	6- 6-39	8.0	Sodium	100	0.8	..	
		6- 9-39	..	Sodium	...	2.4	17.0	
		6-10-39	..	Sodium	...	..	..	Dead. No autopsy was performed
25	F	6-26-39	12.8	Sodium	31.25	0.4	..	
		6-30-39	..	Sodium	...	1.6	17.0	
		7- 3-39	11.8	Sodium	...	2.0	..	
		7- 7-39	..	Sodium	...	3.6	11.0	
		7-14-39	11.1	Sodium	...	5.6	20.5	
		7-21-39	10.3	Sodium	...	7.6	20.5	Head drooping
		7-28-39	10.0	Sodium	...	9.6	27.0	Apparently recovering
		8- 4-39	9.4	Sodium	...	11.6	34.0	Toxic symptoms appearing again
		8- 8-39	9.2	Sodium	...	12.0	..	Dead. Slight cloudy swelling of kidneys, fatty metamorphosis of liver, congestion of abdominal viscera
6	F	6-26-39	8.4	Sodium	23.8	0.2	..	
		6-30-39	..	Sodium	...	0.8	5.0	
		7- 3-39	7.4	Sodium	...	1.0	..	
		7- 7-39	..	Sodium	...	1.8	5.0	
		7-14-39	7.2	Sodium	...	2.8	9.5	
		7-21-39	7.0	Sodium	...	3.8	9.5	
		7-28-39	7.3	Sodium	...	4.8	11.0	
		8- 4-39	7.4	Sodium	...	5.8	8.0	
		8-11-39	7.4	Sodium	...	6.8	8.0	
		8-17-39	7.6	Sodium	...	7.6	6.0	
		8-25-39	7.8	Sodium	...	8.8	..	
		9- 1-39	8.2	Sodium	...	9.8	4.0	
		9- 8-39	8.0	Sodium	...	10.8	8.0	
		9-15-39	8.0	Sodium	...	11.8	10.0	Medication stopped
		10- 4-39	8.0	Sodium	...	..	..	In excellent condition throughout

Three dogs including the one numbered 72 were given small repeated doses of potassium thiocyanate, varying from 20.8 to 24.4 mg. per Kg., and two other dogs were fed sodium thiocyanate in the daily dosage of 23.8 and 31.25 mg. per Kg., respectively. As with other animals, medication was omitted on Saturdays and Sundays. The dog numbered 25 on the larger dose of sodium thiocyanate died in a little over 6 weeks. Its blood concentration gradually rose from 17 to 34 mg. per 100 cc. Four dogs, numbered 6, 24, 32 and 72, therefore, survived more than 12 weeks, and were in excellent health when the experiment was concluded. Their blood concentration at no time exceeded 12 mg. per 100 cc. when the small doses were employed.

The above results perhaps suggest that dogs have a relatively similar susceptibility to thiocyanate as men. In any event, they prove the correctness of Barker's contention (19) that blood concentration can serve as a criterion of thiocyanate therapy. When the blood concentration consistently exceeds 20 mg. per 100 cc. or more, dangerous symptoms and finally death ensue.

3. Absorption.—In Barker's clinical study of thiocyanate in patients, he employed 5-cc. samples of serum or plasma (19). In experimental animals smaller volumes of blood would be more desirable, so that they can be repeatedly bled without harm. A micromethod was thus devised based upon the principle of Barker's procedure (19). To determine the blood concentration of thiocyanate, a volume of 14.9 cc. of water was measured to an appropriate glass tube, and 0.1 cc. of blood was drawn and mixed with it. After hemolysis was complete, 3 cc. of a

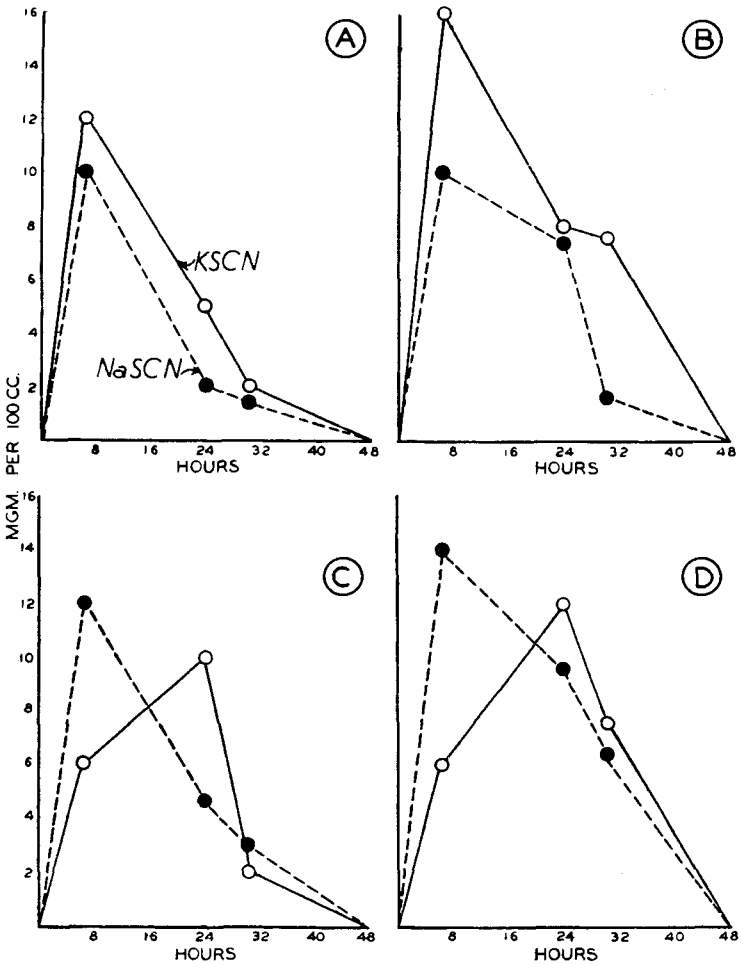

Fig. 2.—Blood Concentration of Thiocyanate in Rabbits following a Single Dose by Mouth.

A. Rabbit numbered 4371, female, weighing 1.918 Kg.
B. Rabbit numbered 4372, male, weighing 1.865 Kg.
C. Rabbit numbered 4373, male, weighing 1.915 Kg.
D. Rabbit numbered 4374, female, weighing 1.775 Kg.

The experiment was carried out as a cross-over test. Rabbits A and B were first given sodium thiocyanate dispensed in capsules, and rabbits C and D, potassium thiocyanate; after one week's rest, the medication was reversed. The dose of each salt was 100 mg. per Kg.

30 per cent solution of trichloracetic acid were added to precipitate the proteins. The whole was filtered in 3 to 5 minutes, and 9 cc. of the filtrate were measured into a small test-tube. Upon the addition of 1 cc. of a 5 per cent solution of ferric nitrate, an amber color developed. It was then examined in an electric photometer (photolometer), and the amount read off from a curve previously determined with known quantities of thiocyanate.

A study of both sodium and potassium thiocyanates in 14 unanesthetized rabbits revealed that when a single dose of 100 mg. per Kg. was administered by mouth, it slowly but steadily made its presence in the blood stream, reaching the peak in about 6 hours or later, and remained in the circulation for approximately 48 hours. The highest level varied from 10 to 16 mg. per 100 cc. as shown in Fig. 2, illustrating the results of four cross-over tests. If the dose of either the sodium or the potassium salt was increased to 200 mg. per Kg., the maximal blood concentration rose to 20 to 24 mg. per 100 cc., and it fell to zero in most cases by the

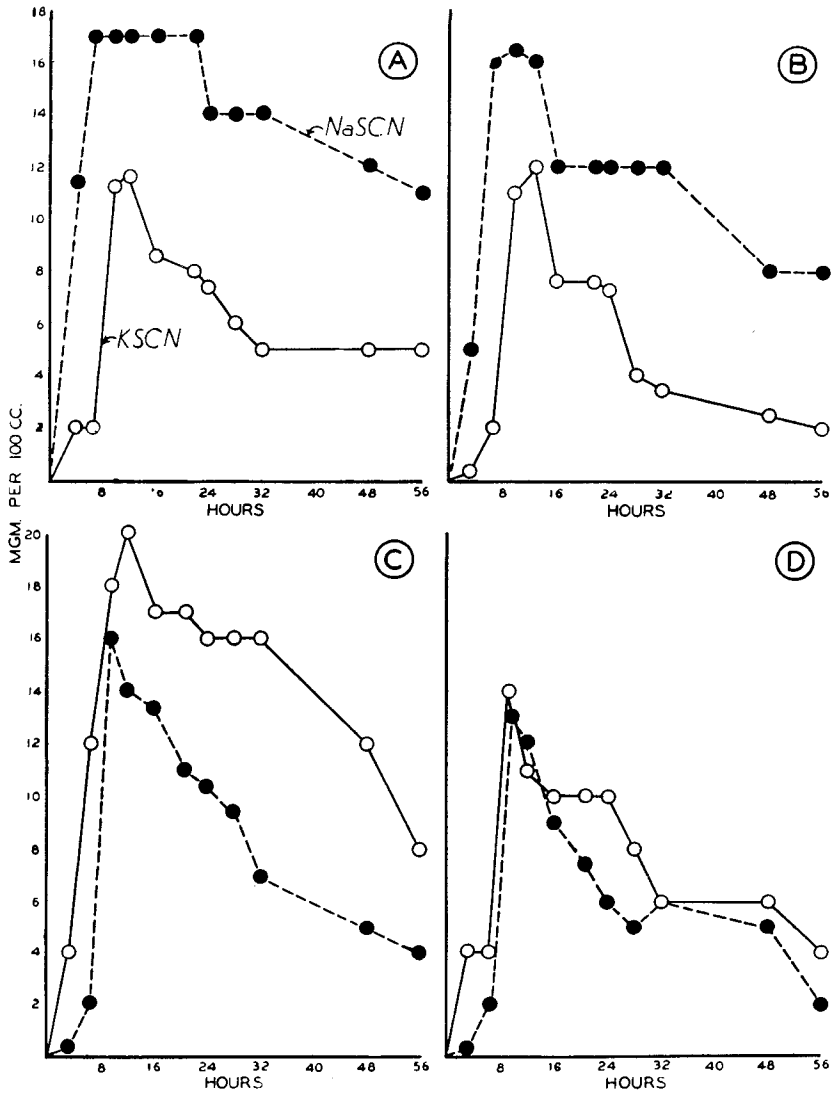

Fig. 3.—Blood Concentration of Thiocyanate in Dogs following a Single Dose by Mouth.

A. Dog numbered 26, female, weighing 6 Kg.
B. Dog numbered 30, male, weighing 5 Kg.
C. Dog numbered 31, female, weighing 7.4 Kg.
D. Dog numbered 36, female, weighing 6.8 Kg.

The experiment was also one of a cross-over test. Dogs A and B first received potassium thiocyanate in form of enteric coated tablets, and dogs C and D sodium thiocyanate similarly dispensed; after one week's rest, the order of drugs was reversed. The dose in each instance was 100 mg. per Kg.

end of 96 hours. The highest concentration in the blood after a large dose of potassium thiocyanate, that is, 300 mg. per Kg., in 2 rabbits was 24 and 30 mg. per 100 cc., respectively, and the drug remained in the circulation for more than a week. Two other animals receiving sodium thiocyanate in the amount of 300 mg. per Kg. died on the fifth day. Their maximal blood levels of thiocyanate reached 43 and 46 mg. per 100 cc., respectively. Undoubtedly, weight for weight, sodium thiocyanate containing more SCN^- ions is potentially more toxic than potassium thiocyanate. At any rate, these results further support Barker's emphasis (19) that the blood concentration serves as a measure of the toxicity of thiocyanate.

A similar series of cross-over experiments was carried out in dogs. On account of vomiting, enteric coated tablets of both salts were again employed. The dose in each case was 100 mg. per Kg. The peak level of blood thiocyanate, as shown in Fig. 3, occurred in all cases with one exception more than 8 hours after the administration of the drug. It should be noted that in dogs either compound in the dosage of 100 mg. per Kg. remained in the blood stream for more than 3 days—longer than in rabbits.

SUMMARY

The toxicity of sodium and potassium thiocyanates has been studied in experimental animals. When injected intravenously in mice the potassium salt is much more toxic than the sodium salt; but when given by mouth in rats and mice the median lethal doses of both compounds are comparable, weight for weight, although the sodium salt has a higher content of SCN^- ions.

In rats, daily administration of either sodium or potassium thiocyanate in the dosage of 100 and 200 mg. per Kg. for 12 and 8 weeks (except Saturdays and Sundays), respectively, causes no inhibition of their growth as evidenced by their weight curves.

Dogs are more susceptible to thiocyanate than mice and rats. Daily doses of either sodium or potassium thiocyanate equal to or exceeding 100 mg. per Kg. produce with few exceptions rapid loss of weight, appearance of very toxic symptoms and, finally, death. Their blood concentration consistently runs more than 20 mg. per 100 cc. No uniform pathological lesions can be made out to account for their death. Daily doses of 20.8 to 24.4 mg. per Kg. may be administered for a period of 12 weeks without apparent impairment of dogs' health—medication being omitted on Saturdays and Sundays. In our series, one dog given sodium thiocyanate in the amount of 31.25 mg. per Kg. died in about 6 weeks after the commencement of the drug. Its blood concentration of thiocyanate gradually rose to 34 mg. per 100 cc.

A micromethod for the determination of blood thiocyanate employing samples of 0.1 cc. has been devised. This becomes helpful in the study of absorption.

In rabbits, a single dose of 100 mg. per Kg. (either the sodium or the potassium salt) gives rise to a maximal blood concentration of 10 to 16 mg. per 100 cc., and its presence in the circulation is no longer detectable after 48 hrs. A dose of 200 mg. per Kg. raises the peak level of blood concentration to 20 to 24 mg. per 100 cc. It disappears from the blood stream in approximately 4 days. A dose of potassium thiocyanate in the amount of 300 mg. per Kg. is followed by a maximal blood concentration of 24 to 30 mg. per 100 cc. It remains in the blood for more than a week. The same dose of sodium thiocyanate kills rabbits after the blood concentration reaches a high level of 43 to 46 mg. per 100 cc.

In dogs, a single dose of 100 mg. per Kg. (either sodium or potassium thiocyanate) brings about a maximal blood concentration of 12 to 20 mg. per 100 cc. It remains in the blood stream for more than 3 days.

Our results strongly support Barker's advocation that the safety and toxicity of thiocyanate can be measured by the determination of blood concentration.

BIBLIOGRAPHY

(1) Pauli, W., "Über Ionenwirkungen und ihre therapeutische Verwendung," *Münch. med. Wochschr.*, 50 (1903), 153.

(2) Le Roy, B. R., "The Thiocyanates in Solution, the Natural Physiological Solvents of the Body," *N. Y. Med. J.*, 90 (1909), 802.

(3) Nichols, J. B., "The Pharmacologic and Therapeutic Properties of the Sulfocyanates," *Am. J. Med. Sci.*, 170 (1925), 735.

(4) Gager, L. T., "The Incidence and Management of hypertension. With a Note on Sulphocyanate Therapy," *J. Am. Med. Assoc.*, 90 (1928), 82.

(5) Smith, A. G., and Rudolf, R. D., "The Use of Sulphocyanate of Soda in High Blood Pressure," *Can. Med. Assoc. J.*, 19 (1928), 288.

(6) Palmer, R. S., Silver, L. S., and White, P. D., "Clinical Use of Potassium Sulfocyanate in Hypertension: A Preliminary Report of 59 Cases," *New England J. Med.*, 201 (1929), 709.

(7) Logefeil, R. C., "Observations on the Use of Potassium Sulphocyanate (Rhodan) in the Treatment of Essential Hypertension, *Minnesota Med.*, 12 (1929), 151.

(8) Fineberg, M. H., "Potassium Thiocyanate in the Treatment of Patients with Hypertension," *J. Am. Med. Assoc.*, 94 (1930), 1822.

(9) Maguire, L. M., "Potassium Sulphocyanate in the Treatment of Hypertension," *U. S. Veterans' Bur. Med. Bull.*, 6 (1930), 978.

(10) Borg, J. F., "Experiences in the Use of the Thiocyanates," *Minnesota Med.*, 13 (1930), 293.

(11) Ayman, D., "Potassium Thiocyanate in the Treatment of Essential Hypertension," *J. Am. Med. Assoc.*, 96 (1931), 1852.

(12) Saleeby, P. J., "Some Observations on the Effects of Potassium Sulphocyanate on Elevated Blood Pressure," *New Orleans Med. & Surg. J.*, 83 (1930–1931), 93.

(13) Meakins, J. C., and Scriver, W. de M., "The Treatment of Hypertension," *Can. Med. Assoc. J.*, 25 (1931), 285.

(14) Egloff, W. C., Hoyt, L. H., and O'Hare, J. P., "Observations on Thiocyanate Therapy in Hypertension," *J. Am. Med. Assoc.*, 96 (1931), 1941.

(15) Goldring, W., and Chasis, H., "The Use of Sulphocyanate in the Treatment of Hypertension," *N. Y. State J. Med.*, 31 (1931), 1322.

(16) Goldring, W., and Chasis, H., "Thiocyanate Therapy in Hypertension," *Arch. Internal. Med.*, 49 (1932), 934.

(17) Palmer, R. S., "The Hypotensive Action of Potassium Sulphocyanate in Hypertension," *Am. J. Med. Sci.*, 184 (1932), 473.

(18) Bolatin, M. T., "Potassium Sulphocyanate in Hypertension," *Illinois Med. J.*, 62 (1932), 557.

(19) Barker, M. H., "The Blood Cyanates in the Treatment of Hypertension," *J. Am. Med. Assoc.*, 106 (1936), 762.

(20) Griffith, J. Q., Jr., Lindauer, M. A., and Campbell, R., "Thiocyanate Therapy in Hypertension Including a New Micro Method for Determining Blood Thiocyanate," *Am. Heart J.*, 14 (1937), 710.

(21) Steidl, J., Steenken, W., Jr., and Heise, F. H., "Influence of Sulphocyanates on Tuberculosis," *Am. Rev. Tuberc.*, 38 (1938), 50.

(22) Massie, E., Ethridge, C. B., and O'Hare, J. P., "Thiocyanate Therapy in Vascular Hypertension," *New England J. Med.*, 219 (1938), 736.

(23) Doles, H. M., "Treatment of Hypertension," *South. M. J.*, 31 (1938), 1225.

(24) Doles, H. M., "The Mode of Action of Sodium Sulfocyanate in Reducing Hypertension," *South. M. J.*, 32 (1939), 299.

(25) Weis, C. R., and Ruedemann, R., "Exfoliative Dermatitis from Potassium Sulphocyanate Therapy," *J. Am. Med. Assoc.*, 93 (1929), 988.

(26) Ayman, D., "Exfoliative Dermatitis from Potassium Thiocyanate," *J. Am. Med. Assoc.*, 93 (1929), 1671.

(27) Tyrrell, J. D., "Severe Exfoliating Dermatitis from Sodium Sulphocyanate Therapy," *Can. Med. Assoc. J.*, 22 (1930), 80.

(28) Baker, T. W., and Brunsting, L. A., Dermatitis Medicamentosa Resulting from Administration of Sulfocyanates in the Treatment of Hypertension," *J. Am. Med. Assoc.*, 108 (1937), 549.

(29) Green, M. E., and Snow, J. S., "Thiocyanate Dermatitis," *Arch. Internal Med.*, 64 (1939), 579.

(30) Healy, J. C., "Therapeutics and Toxicology of the Sulphocyanates," *New England J. Med.*, 205 (1931), 581.

(31) Goldring, W., and Chasis, H., "Thiocyanate Therapy in Hypertension," *Arch. Internal Med.*, 49 (1932), 321, 934.

(32) Garvin, C. F., "The Fatal Toxic Manifestations of the Thiocyanates," *J. Am. Med. Assoc.*, 112 (1939), 1125.

(33) Palmer, R. S., and Sprague, H. B., "Four Cases Illustrating the Untoward Symptoms Which May Be Produced by the Use of Potassium Sulphocyanate in the Treatment of Hypertension," *M. Clin. North America*, 13 (1929), 215.

(34) Wald, M. H., Lindberg, H. A., and Barker, M. H., "The Toxic Manifestations of the Thiocyanates," *J. Am. Med. Assoc.*, 112 (1939), 1120.

(35) Barker, M. H., "The Use of Cyanates in the Treatment of Hypertension," *Wisconsin M. J.*, 36 (1937), 28.

(36) Binger, M. W., personal communication.

(37) Hunt, R., "Rhodanwasserstoff," *Heffters Handbuch der experimental len Pharmakologie*, 1 (1923), 817.

(38) Bodansky, M., "The Conversion of Cyanide into Thiocyanate in Man and in Alkaline Solutions of Cystine," *J. Pharmacol.*, 37 (1929), 463.

(39) Wallace, G. B., and Brodie, B. B., "The Distribution of Administered Iodide and Thiocyanate in Comparison with Chloride, and Their Relation to Body Fluids," *J. Pharmacol. and Exper. Therap.*, 61 (1937), 397.

(40) Wallace, G. B., and Brodie, B. B., "The Distribution of Iodide, Thiocyanate, Bromide and Chloride in the Central Nervous System and Spinal Fluid," *J. Pharmacol. and Exper. Therap.*, 65 (1939), 220.

(41) Friend, D. G., and Robinson, R. W., "Action of Sodium Thiocyanate upon Tissue Oxygen Consumption," *J. Lab. & Clin. Med.*, 24 (1939), 832.

(42) Takacs, I., "Versuche mit Rhodansalzen," *Ztschr. f. d. ges. exper. Med.*, 50 (1926), 432.

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Do not overlook sending along an abstract with the paper which you are presenting at the Richmond meeting.