cient to make a total of 30 cc. Dilute with water to about 75 cc. Digest gently over a low flame for 30 to 50 minutes. Filter the hot solution into a 400-cc. beaker. Wash the filter free from iodides. Add a slight excess of silver nitrate solution (5 cc. of 10%) and stir. Add 25 cc. of concentrated nitric acid, stir, cover the beaker with a watch glass and boil gently till the liquid becomes clear. Filter on a tared gooch crucible, wash with water. If chlorides are present wash with ammonia water. Dry at 120° to 130° C. and weigh. Silver iodide × 0.9677 equals mercuric iodide.

CONCLUSIONS.

1. The incomplete reduction of mercuric iodide in alkaline solution with lactose as a reducing agent is due to incomplete peptization and occlusion.

2. Thorough mixing of the lactose in a dry condition together with proper manipulation in moistening it with the alkali solution prepares the mercury compound for complete reduction.

3. The iodine is completely changed to a soluble iodide which can be separated and determined as silver iodide.

4. A method is submitted, the procedure of which is simple and rapid, and the principles are sound. Apparatus common to every laboratory may be used.

5. The limits of accuracy are within one per cent which is satisfactory for control and regulatory purposes.

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THE TOXICITY OF THALLIUM SULPHATE.*

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The introduction in 1920 of proprietary rat poisons containing thallium compounds as the toxic ingredients directed attention to the toxicity of these products. The literature upon thallium is meager, so this investigation was undertaken to obtain definite knowledge of the toxicity of thallium compounds.

Thallium occurs in various ores which usually also contain sulphur. The chemical symbol is Tl; the atomic weight is 204.0. It forms two series of white, water-soluble salts: thallous, in which the metal is monovalent; and thallic, in which it is trivalent. The monovalent compounds are considered to be more stable and are used in pharmaceutical practice. The acetate (CH_3COO-Tl) has been used as a depilatory, and to check the night sweats in phthisis: the sulphate, Tl_2SO_4, is most commonly employed in the preparation of rat poisons.

Thallium was discovered by Crookes in 1863 by the characteristic green line in the spectrum (19, 20). Olmer and Tian (40) claim that the limit of detection with a spectroscope is 1:500,000,000 (2 micrograms per liter). It may be determined quantitatively by fusion with sodium carbonate, acidification with hydrochloric acid, adding an excess of potassium chlorate and boiling to remove excess

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chlorine, addition of sodium thiosulphate and potassium iodide, and titration of the excess sodium thiosulphate with iodine solution.

TOXICITY TO BACTERIA.

Thallium has a strong oligodynamic action on *B. Coli*, typhoid, dysentery, streptococci and staphylococci, but not on molds (6). When a piece of metallic thallium is placed in the center of an agar plate containing viable organisms, a clear zone of dead bacteria is produced over a zone 7 to 14 millimeters in diameter. This action disappears when air is removed, but returns on reëxposure, suggesting that air oxidizes Tl to Tl_2O, which dissolves to form TlOH (14). Concentrations of thallium acetate or carbonate as strong as 1:1000 (1000 milligrams per liter) are required to inhibit the growth of *B. Coli*.

TOXICITY TO ANIMALS.

Lamy (34) offered milk containing 5 grams of thallium sulphate to two young puppies. They drank only a small quantity, refusing the remainder, and died in 4 days in spite of all efforts to save them. Six ducks, two hens and a dog consumed a portion of the remaining solution. During the day the dog became restless, salivated and refused food; that night constipation and respiratory distress developed. The hind legs of the dog and the legs of the ducks and hens became paralyzed. All animals were dead or moribund 64 hours after consuming the poisoned milk. Ten milligrams of thallium sulphate killed another dog in 40 hours with the same symptoms. One gram of thallium sulphate killed a dog in 5 days, whereas 1.5 grams of lead acetate failed to kill (29), which led to the conclusion (41) that thallium was more toxic than lead. Doses up to 350 milligrams of thallium sulphate intravenously to rabbits interferred markedly with pulmonary circulation; 750 milligrams killed (1).

The flesh of a hen poisoned with thallium was fed to eleven rats, all of which died (Kobert, 33). Direct application of a thallium paste killed rats in 2 to 4 days (45). A mother rat was fed large doses of thallium for 4 days when it died. The nursing young were at once transferred to a normal lactating rat and given her milk, but all showed the typical effects of thallium poisoning (Ehrhardt, 28).

Swain and Bateman (46) reported that the lethal dose of thallium as thallous acetate when injected into the lymph sacs of 300-gram toads was approximately 16.7 milligrams per kilogram; death was due to respiratory failure. Immersed in a 1:2500 solution of thallium as thallous chloride (400 milligrams per liter), two toad fish and a rock cod died in 4 days; 800 milligrams per liter killed in 20 hours. Injected into the lymphatic trunk immediately posterior to the lateral fin, 10 to 20 milligrams per kilogram killed cod and blue fish in 1 to 4 days. Subcutaneous injections of 20 milligrams per rat, 15 milligrams per guinea-pig and 20 milligrams per rabbit caused death in 2 to 6 days. (Since the body weights of the test animals are not given, the doses per kilogram cannot be determined from the report.) Oral administration of 13 milligrams per kilogram and larger doses to dogs caused death in 7 to 15 days; 45 milligrams per kilogram killed in 4 days. The administration of 200 milligrams over a period of 13 days produced the same effect upon a dog as did the same quantity over 4 days to another dog. Swain and Bateman conclude that there is no tolerance for thallium but a cumulation of action, and

that thallium is more toxic than lead, being about as potent as "arsenic" (arsenious oxide). The first symptoms of thallium intoxication were lack of coördination of the hind legs of quadrupeds. Trembling, emesis, congestion of the entire gastrointestinal tract, constipation or diarrhea, albuminuria and finally, diffuse alopecia were noted. A blue line appeared upon the margins of the gums. Kidney lesions were observed in dogs receiving 200 milligrams (17 milligrams per kilogram).

In a very thorough series of studies by Buschke and co-workers (4 through 17), thallium poisoning of rats and warm-blooded animals was connected with its action upon the various endocrine glands, especially the ovaries or testicles, thyroid and suprarenals. Disturbances in calcium metabolism produced rickets; the addition of calcium to perfusion solutions counteracted thallium poisoning of isolated frog hearts. Dal Collo (21, 22) obtained parenchymatous and vascular nephritis with characteristic epithelial lesions in rabbits. No relation or interdependence could be established between alopecia, hypertrophic processes, bone lesions and cataracts in the eyes of rats by Mamoli (36). Olivier (39) confirmed previous reports that small doses of thallium fed over a long period of time caused inflammatory proliferations of the mucosa of the oesophagus and cardiac end of the stomach.

When rats receive a single large dose of thallium acetate, or daily doses as small as 0.1 milligram for some time, alopecia is produced. Buschke and coworkers, and later Dixon (25) showed that this was due to its action on the autonomic nervous system, and that local applications of thallium salves did not cause loss of hair in the treated area. The hairs of the scalp and the chest under direct control of the sympathetic nervous system are loosened readily. The eyebrows, eyelashes, hairs of the snout and other hairs not under control of the autonomic nervous system are not affected. All animals with the possible exception of the guinea-pig are susceptible to thallium alopecia. The hair becomes less glossy, and is easily rubbed off after about the 7th day. Complete loss of hair usually occurs between the 14th and the 19th days. Growth of new hair starts about the 4th week, and is complete by the end of the second month. Interruption of growth of the hair was found to be due to failure of transition of the large polygonal cells to the stratified hair cells. Thallium was found to selectively increase the reactivity of the entire autonomic nervous system, just as strychnine increases the activity of the spinal cord. Thallium is eliminated in the urine; spectroscopically, it has been found in every tissue of the body.

TOXICITY TO MAN.

Lamy (34) developed general lassitude and pains in the lower limbs which he attributed to thallium vapors, although Crookes (19) stated that he was not affected by the fumes, nor by the ingestion of 65 to 130 milligrams of various thallium salts. The application of a thallium paste to the chest of a 27-year old man produced violent pains in the extremities, alopecia, persistent albuminuria, stomatitis and general depression; these symptoms continued for about a month. Twenty micrograms of thallium per liter were found in the spinal fluid 25 days after application of the paste (Olmer and Tian, 40).

Ten milligrams of thallium nitrate per kilogram taken by mouth by a 34-year old printer with suicidal intent caused the same symptoms; in addition, emesis and achlorhydria were marked. New hair returned in 8 weeks, but 11 weeks were

required for the return of the gastric acidity to normal (Buschke, Peiser and Klopstock, 16).

A 2.5-year old child ate an undetermined amount of a bait made by smearing thallium paste upon bread, which had been exposed as a rat poison. Rapid collapse and vomiting were followed by partial recovery, but the child died in 29 hours. Post-mortem examination showed nothing abnormal except marked irritation of the stomach and small intestines. Spectroscopically thallium was demonstrated in various tissues. A 4.5-year old child also ate some of the same bait, but apparently a smaller quantity, as it survived (Lühring, 35). Bullard (3), Brieger (2), and Redlich (43) also report instances of poisoning from thallium.

Kaps (32) summarizes the symptoms produced in fatal subacute thallium poisoning as: (1) a short period of excessive gastrointestinal pain, emesis, nausea, colic and diarrhea which soon changes to obstinate constipation; (2) disorders of the central and vegetative nervous systems, as conjunctivitis, blepharitis, alopecia totalis, acute suppurative dermatitis of the face, sensitiveness of the muscles and joints which is increased upon pressure, retrobulbar neuritis, amaurosis and a decline in psychic functions to complete dementia; (3) degeneration of the heart, liver and kidneys. The mechanism of thallium poisoning following subcutaneous injection must be sought in the action on the central and vegetative nervous systems, with secondary disturbances of the nervous regulation of the endocrine glands.

In a survey of industrial poisonings among thallium workers, Meyer (37) reports reduction in vision, cataract, nephritis, alopecia and endocrine disturbances of various sorts.

The control of night sweats of phthisis by thallium acetate was found efficacious by Combenale (18), Huchard (30) and Buschke (10). However, the development of alopecia after 270 milligrams (Dubreille, 27); (Jeanselme, 31) and of pains in the legs and in peripheral nerves after 100 milligrams (Vassaux, 47) directed attention to its dangers and it has been abandoned. In the treatment of alopecia areata and ringworm in children, thallium acetate has been successfully employed in doses of 8 milligrams per kilogram (38). Great care is necessary, as toxic reactions have been obtained with doses as small as 4 milligrams per kilogram by Davies (23, 24). Dowling (26) reports 78 toxic reactions in 90 children, and Peyri (42) had 19 reactions in 74 children, following doses of 8 milligrams per kilogram. The conclusion reached by Sollmann (44) in his discussion of thallium seems justified: "The therapeutic use has repeatedly caused severe poisoning and is scarcely justified."

FEEDING EXPERIMENTS: TOXICITY OF THALLIUM SULPHATE TO RATS.

A sample of C. P. thallium sulphate was powdered and thoroughly mixed with screened rat food normally fed to our laboratory animals. Concentrations of bait were prepared containing from 0.25 to 10 per cent of thallium; in most experiments, 0.5 per cent of thallium was used. All rats tested were held in the laboratory and fed control food for at least a week, in order to obtain some degree of uniformity in condition of the animals. The day before they were to be used, each rat was placed in an individual cage and offered water but no food. On the morning of the experiment, each rat was weighed, and a suitable quantity of bait offered in a glass sponge cup. So far as feasible, it was our effort to offer each animal one-half to one per cent of its body weight of bait, previous experiments with other

poisons having shown that this quantity of food is usually consumed within 10 to 30 minutes. Frequent inspections were made after feeding to determine the rapidity with which the rats started to eat, and the time required to completely consume the poisoned food. Any uneaten food was removed after several hours and weighed. The day following the feeding of the poisoned bait, all animals were offered normal food and drinking water. All rats dying within 5 days (occasionally a longer time was employed) which showed symptoms of thallium poisoning, were considered as fatalities from the quantity of thallium consumed.

The Minimum Lethal Dose (MLD) was considered to be that dose of thallium which caused the death of all, or practically all test animals within the specified time. Doses somewhat larger than the accepted MLD must kill all test animals; doses somewhat smaller (20 to 25 per cent) kill less than half of the test animals in general practice. The MLD is considered to be accurate within 10 to 20 per per cent, which is the usual range of variability of rats to rat poisons.

Thallium sulphate, having the formula Tl_2SO_4, contains 81 per cent of thallium, so that 1.24 grams of thallium sulphate were taken as the equivalent of 1.00 gram of thallium in the preparation of all lots of rat food. Seventy-two white rats, weighing between 103 and 197 grams, were fed specified doses of thallium sulphate between December 4 and December 17, 1924. The poisoned baits were consumed with the same eagerness as control food containing no thallium. In most instances all food was consumed within 15 to 30 minutes after exposure. The results obtained in this series of feeding experiments are reported in Table 1. The dose of 25 milligrams of thallium per kilogram killed 73 per cent of the rats tested. All larger doses, commencing with 35 milligrams per kilogram (an increase of 40 per cent in dosage), killed all rats. The next lower dose tried, 20 milligrams per kilogram (a decrease of 20 per cent in dosage), killed but one-third of the test animals. Accordingly, 25 milligrams per kilogram was selected as the minimum lethal dose. This value was confirmed in a further series of feeding experiments in May 1927, upon 15 white rats weighing between 125 and 175 grams, with the results reported in Table 2.

With a minimum lethal dose (MLD) of 25 milligrams per kilogram, thallium is as toxic as strychnine when fed to rats.

It may be noted that death usually occurred on the second or third day after feeding thallium baits, unless the quantity of poison taken was excessive. Ten times the MLD killed only 1 rat of 5 within 1 day; forty times the MLD killed only 4 of 5 rats in 1 day. Thallium is a certain, but not a rapid, rat poison.

FEEDING EXPERIMENTS: TOXICITY OF COMMERCIAL THALLIUM PREPARATIONS.

A commercially prepared sample of grain with a thallium coating was fed to 7 wild rats weighing between 78 and 355 grams. Six rats died on the third day, the other on the fourth day after feeding. The doses of grain consumed were equivalent to 3 to 7.5 grams of grain per kilogram. Doses equivalent to 2.5 to 27.25 grams of grain per kilogram were fed to 12 white rats weighing between 157 and 210 grams. One rat died on the second day, 10 on the third and the other on the fourth day after feeding. Since all animals died in each test, it is not possible to ascertain the MLD, except that it is probably in the neighborhood of 2.5 grams of grain per kilogram.

A commercial thallium paste distributed as a rat poison was incorporated with rat food and offered to 3 white rats weighing about 200 grams; it was not readily eaten, but all rats consumed enough poison to cause death on the third day. Smeared on the backs of 6 other white rats, 3 died on the fourth day and the other three on the fifth day after application. A series of 6 commercial samples of thallium products were tested which had been used by field agents of the Bureau of Biological Survey. One sample was taken readily by prairie dogs, one indifferently well and two were refused, when the samples were mixed with the same baits and tested at the same places and under essentially the same conditions. Thirty-six rats weighing between 145 and 355 grams were all killed by doses of 20 and 40 milligrams per kilogram. The MLD was not determined accurately but is practically the same as that of CP thallium sulphate. No appreciable differences in rate of consumption of the different baits could be detected.

TOXICITY TO RABBITS: INTRAVENOUS INJECTIONS.

To determine the toxicity to rabbits, aqueous solutions containing 0.5 or 1.0 per cent of thallium sulphate were injected into the marginal ear veins of Belgian hares weighing between 1530 and 2250 grams. Some injections were made in 1924, and others in 1927 to confirm the original findings. The results obtained in both series of injections are consolidated in Table 3. The MLD was found to be 25 milligrams per kilogram, the same figure as obtained in feeding experiments upon rats. Respiratory failure was the cause of death. Post-mortem examinations failed to show any unusual reactions except marked congestion of the kidneys, and hyperemia of the entire gastrointestinal tract.

CONCLUSIONS.

1. Thallium compounds are very toxic to animals. The Minimum Lethal Doses when fed to rats, or intravenously injected into rabbits, are 25 milligrams of thallium per kilogram body weight.

2. Although occasionally given to children in doses of 8 milligrams per kilogram, it has caused toxic reactions even in doses half that size.

3. Thallium affects the sympathetic nervous system, thereby causing alopecia, pains in the muscles and nerves of the legs, and disturbances of the endocrine glands, particularly the ovaries or testicles.

TABLE 1. THE TOXICITY OF THALLIUM AS SULPHATE TO WHITE RATS.

Feeding experiments in December 1924.

Dose of thallium. mg./Kg.	No. rats fed.	No. rats dying after... days. 1.	2.	3.	4.	5.	Total.	Per cent dying.
10	10	0	0	0	0	0	0	0
15	8	0	1	0	0	0	1	12
20	8	0	2	1	0	0	3	37
25	11	0	1	2	4	1	8	73
35	5	0	3	2	0	0	5	100
50	5	0	0	1	2	2	5	100
75	5	0	0	3	2	0	5	100
150	5	0	3	2	0	0	5	100
250	5	1	4	0	0	0	5	100
500	5	2	3	0	0	0	5	100
1000	5	4	1	0	0	0	5	100

4. Thallium is cumulative in action; tolerance does not develop in animals. Thallium is a certain, but not a rapid, poison for rats.

TABLE 2. THE TOXICITY OF THALLIUM AS SULPHATE TO WHITE RATS.

Feeding experiments in May 1927.

Dose of thallium. mg./Kg.	No. rats fed.	No. rats dying after... days. 1.	2.	3.	4.	5.	Total.	Per cent dying.
20	5	0	0	1	2	0	3	60
25	5	0	0	1	4	0	5	100
30	5	0	0	0	5	0	5	100

TABLE 3. THE TOXICITY OF THALLIUM AS SULPHATE TO RABBITS. INTRAVENOUS INJECTIONS.

Dose of thallium. mg./Kg.	Body weight of rabbit, Grams.	Result.
10	2250	Survived
25	1800	Died after 1 day
25	2035	Died after 2 days
25	2220	Died after 2 days
50	1700	Died after 1 day
75	1500	Died after 1 day
100	1530	Died after 1 day

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