The Effect of Thiocyanate Concentration on Thiocyanate Distribution and Excretion[1] (35468)

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The metabolism of thiocyanate (SCN^-) has been of interest to many investigators for over a century. Although the physiological presence of this anion in biological fluids was established early (1, 2), subsequent studies tended to categorize thiocyanate as a drug, where it gained recognition as an antihypertensive agent (3) and as an antithyroid agent (4). More recent investigations have been redirected toward understanding the metabolism of thiocyanate (5–9) and the possible functions of this anion in normal biological processes (10). In the course of one of these investigations (9), it was desirable to study the volume of distribution of endogenous thiocyanate so that we might calculate the total amount of thiocyanate in the animal. The results of that investigation are reported here and show that (1) the measured volume of distribution of thiocyanate is decreased by high plasma concentrations of thiocyanate, such as used in the conventional measurement of the extracellular fluid space (11, 12), and that (2) large doses of thiocyanate have a much faster rate of disappearance from the plasma than does endogenous thiocyanate at normal concentration.

Methods. Male albino Sprague-Dawley rats (Holtzman and Co., Madison, Wis.), weighing between 340 and 360 g each, were maintained on Purina Laboratory Chow and water *ad libitum.* Five rats were injected intraperitoneally with a 1.0 ml tracer dose of $^{35}SCN^-$ (50 μg/1.8 μCi/1.0 ml). Four additional rats were given the above tracer dose of $^{35}SCN^-$, but with 5.0 mg of SCN^- carrier added to each injection, as NH_4SCN, to achieve a SCN^- concentration of approximately 5 mg/100 ml of plasma. Blood samples were analyzed from each rat at 1, 2, 4, 6, and 25 hr after injection, by digesting 0.100 ml of plasma in 1.0 ml of NCS solvent.[3] After digestion, 10 ml of toluene scintillation solution (13) was added and the radioactivity in each sample was counted twice for a total of 20 min. Chromatography of the injection solution, using an ethanol:1 *M* ammonium acetate (7.5:3.0) solvent (5) showed that 99.7% of the radioactivity moved as SCN^-. Previous experiments have shown that the radioactivity in the plasma remains as $^{35}SCN^-$ over the time period of the experiments reported here (9).

The volume of distribution of thiocyanate was estimated by plotting the plasma concentration of radioactivity on a semilog plot against time. Correction for the urinary excretion and metabolism of thiocyanate was made by extrapolating the semilog disappearance curve for plasma radioactivity, using the 6- to 25-hr slope, to zero time (12). The precision of this method is limited to some extent by lack of information regarding the dynamics of individual thiocyanate compartments (14), but is believed sufficient to warrant the conclusions drawn here.

Results. The pertinent data from the experiment are summarized in Table I, and the plasma disappearance curves of the injected $^{35}SCN^-$ are shown in Fig. 1. The radioactivity in the plasma of rats receiving only the tracer $^{35}SCN^-$ decreased with a half-life of 4.42 days. In the rats which received the

[1] Supported by Grants K6-AM-40 and AM-04689 from the NIH and AEC-AT(40-1)-1643 from the United States Atomic Energy Commission.

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TABLE I. Effect of Plasma Thiocyanate Concentration on the Measured Volume of Distribution and Plasma Disappearance of Thiocyanate.

	Dose injected	
	Tracer	Carrier
No. of rats	5	4
Body wt (g)	356[a] ±6	351 ±4
Injected cpm	3,340,000	3,340,000
cpm/ml plasma at zero time (extrapolated)	24,400 ±946	32,600 ±4500
Calculated vol of thiocyanate space (ml)	136.9 ±5.2	103.6 ±13.1
Thiocyanate space as percentage of body wt	38.4 ±1.2	29.5 ±3.8
Half-life of plasma $^{35}SCN^-$ (days)	4.42[b]	0.31 ±0.08

[a] Average values ± 1 standard deviation.

[b] Calculated from slope connecting average points. One of five rats did not show a decrease in plasma radioactivity from 6 to 25 hr. This is likely due to a redistribution of thiocyanate during and following the taking of blood samples. A similar finding has been reported and was caused by fasting rats for 1 day (9).

5-mg carrier dose of thiocyanate, the plasma radioactivity fell more rapidly, with a half-life of 0.31 days. The difference in slopes of the disappearance curves shows clearly that thiocyanate, when present in high plasma concentrations, is handled dramatically different from thiocyanate present in physiological concentrations.

Discussion. The use of thiocyanate as a tool in the endocrine research laboratory is a frequent occurrence (15, 16). In addition, numerous researches are related to the metabolism of thiocyanate (5–10) and its possible contribution to the etiology or complication of endemic goiter (17, 18). Hence it is becoming more important to appreciate three basic facts about thiocyanate metabolism: (i) thiocyanate is physiologically present in plasma in concentrations a hundred times greater (or more) than the inorganic iodide concentration (7–9); (ii) the dynamics of thiocyanate, when present at greatly elevated plasma concentrations, are markedly different from the dynamics of endogenous thiocyanate at normal concentration; and (iii) the dynamics of thiocyanate at either plasma concentration are altered by the presence of particular anions, such as perchlorate or nitrate (8, 19).

The experiment reported herein demonstrates that the rate of disappearance of thiocyanate from the plasma is a function of the plasma concentration of thiocyanate. Previous experiments have shown that normal rats have 0.5–0.6 mg of SCN^-/100 ml of serum

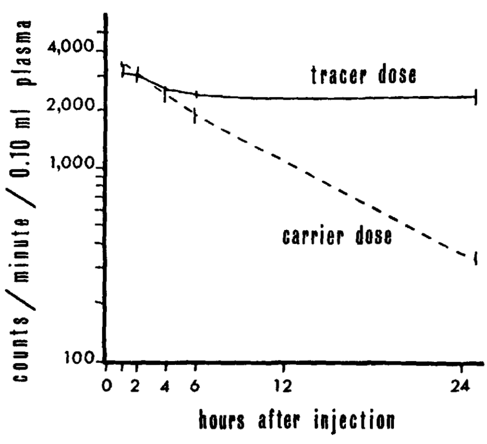

FIG. 1. The disappearance of plasma radioactivity after injecting (at zero time) $^{35}SCN^-$ as a tracer dose (50 μg/1.8 μCi) or with 5.0 mg of carrier SCN^-. Lines connect the average points, vertical bars represent ± 1 standard deviation.

and this has a biological half-life of 2 to 4 days (9); this rate is slightly faster than the half-life of 4.42 days observed in the five rats reported here which received only the tracer dose of $^{35}SCN^-$. In the present experiments the rats given $^{35}SCN^-$ with 5 mg of carrier thiocyanate had an estimated initial serum SCN^- of 5 mg/100 ml which was lost from the plasma with a half-life no more than 0.31 days.

Why is the disappearance rate of plasma thiocyanate a function of the plasma thiocyanate concentration? The available evidence suggest at least two contributing factors: (i) changes in the proportions of the total-body thiocyanate distributed in the available thiocyanate compartments, and (ii) the capacity of the kidney to reabsorb the thiocyanate filtered.

Changes in the proportions of the total-body thiocyanate distributed in the available thiocyanate compartments are well documented, and it is recognized that the ability of compartments to maintain a concentration gradient for thiocyanate against plasma diminishes as the plasma concentration of thiocyanate increases (20–22). Under these conditions, any specific traps for thiocyanate may become saturated and a greater proportion of the total-body thiocyanate exist within the vascular and interstitial fluid volume and this is available for rapid excretion in the urine. Normally, the plasma thiocyanate concentration in rats is 500 to 700 μg/100 ml of plasma (7–9), and a fraction of the body thiocyanate is distributed into areas of increased concentration (*i.e., gastric juice*) and this is not as readily available for excretion. The data presented in Table I support such a concept and show that the *measured* volume of distribution of thiocyanate is less at the higher plasma concentrations of thiocyanate. This decrease in measured volume is a reflection of the relative lack of extravascular compartments, which ordinarily contain concentrations of thiocyanate greater than that of plasma, and which, when present, give a high "thiocyanate volume" when measured by isotope dilution.

Secondly, the capacity of the kidney to reabsorb many polyatomic anions decreases as the anion concentration in the glomerular filtrate increases. This phenomenon has been observed for perchlorate (23, 24), nitrate (24), and thiocyanate (24, 25), as well as for chloride (24). In addition, there is a competition between these anions for being passively reabsorbed, so that the excretion of one is enhanced by a high concentration of a second (8, 23, 26).

A third possibility suggested to explain the rapid excretion of anions when present at high plasma concentrations is that the binding of the anion to plasma proteins is less at high concentrations of the anion, thereby leaving a greater proportion of the anion free and available for filtration through the glomerulus. While this hypothesis has some basis (27, 28), the available evidence suggests that this mechanism is relatively unimportant under conditions of normal blood pH (23, 28).

One of the more important and current implications of the present and cited data lies in the area of using anions such as thiocyanate or perchlorate in the study of iodine metabolism and thyroid physiology. It is critical in the interpretation of such studies to recognize that perchlorate, for example, alters drastically the extrathyroidal dynamics of thiocyanate which is itself actively metabolized by the thyroid (5, 6). It is reasonable to expect that a disregard of the interrelations between these or similar anions will, on occasion, lead to invalid conclusions. It is therefore important when using one or more of these anions to recognize these interrelations and the possible effects of such in the experiment and subsequent conclusions.

Summary. It has been observed in rats that the measured volume of distribution of thiocyanate is decreased by high plasma concentrations of thiocyanate, and that large doses of thiocyanate have a faster rate of disappearance from the plasma than does endogenous thiocyanate at normal concentrations. These observations are interpreted as results of (i) the saturation of thiocyanate-concentrating compartments, and (ii) the inability of the kidney tubule to reabsorb large amounts of filtered thiocyanate.

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Received Nov. 20, 1970. P.S.E.B.M., 1971, Vol. 136.