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# Chromium Metabolism

## A Literature Review

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## ABSTRACT

The trivalent state of chromium ($Cr^{3+}$) is that encountered in biological milieus and is responsible for its nutritional activity. The principal route by which trivalent chromium enters the body is the digestive system. Chromium in foods is present both in the inorganic form and as organic complexes. Intestinal absorption of chromium is low (0.5–2%), and the mechanism has not yet been fully elucidated. Absorbed chromium circulates as free $Cr^{3+}$, as $Cr^{3+}$ bound to transferrin or other plasma proteins, or as complexes, such as glucose tolerance factor (GTF)-Cr. Circulating trivalent chromium can be taken up by tissues, and its distribution in the body depends on the species, age, and chemical form. It is excreted primarily in the urine by glomerular filtration or bound to a low-mol-wt organic transporter. Chromium metabolism is still imperfectly understood. The use of $^{51}Cr$ has nevertheless furnished valuable data concerning its transport and excretion.

**Index Entries:** Chromium; metabolism; blood transport; urinary excretion; tissue distribution; radioisotopes.

The routes by which chromium (Cr) enters the body are the skin, the digestive tract, and the respiratory apparatus. The metabolism of this trace element is considerably influenced by the route of entry, but also by its oxidation level and the nature of its ligands.

## PHYSICOCHEMICAL PROPERTIES OF Cr

Cr is a shiny, hard, white metal, belonging to the first series of transition elements. Its atomic number and mass are 24 and 52 (51.9961), respectively. It has four stable isotopes:

$^{50}$Cr: 4.355%;
$^{52}$Cr: 83.779%;
$^{53}$Cr: 9.501%; and
$^{54}$Cr: 2.365% (IUPAC, 1984).

Five radioactive isotopes can be produced, but only $^{51}$Cr with a half-life of 27.8 d is marketed and used in tracer studies.

### *Oxidation States*

Cr can assume all oxidation states from $-2$ to $+6$, but the most often encountered are 0, $+2$, $+3$, and $+6$.

### *Divalent Cr*

By simple exposure to air, $Cr^{2+}$ is oxidized to $Cr^{3+}$, so the 2+ state does not exist in biological systems.

### *Hexavalent Cr*

In the hexavalent state, Cr is bound to oxygen in the form of chromate ($CrO_4^{2-}$) or dichromate ($Cr_2O_7^{2-}$), a powerful oxidizing agent. These ions are easily reduced to $Cr^{3+}$ in acid.

### *Trivalent Cr*

This is the most stable oxidation state existing in biological milieus. Trivalent Cr forms coordination complexes, complexes, or chelates. Its coordination number is 6, and it can bind to ligands to form hexacoordinate or octahedral complexes. Metal ligand bonds may involve oxygen, nitrogen, or sulfur. At neutral or basic pH, Cr—$H_2O$ bonds are modified and Cr—$OH^-$ bonds are created, leading to the formation of giant macromolecules (polymerization of hydrated Cr) that precipitate and are thus biologically inert. This phenomenon, called olation, is also favored by heat. Among the biological ligands that can prevent olation at physiological pH are pyrophosphate and certain amino acids, such as methionine, serine, glycine, leucine, lysine, and proline.

## ABSORPTION

Chromium is present in the diet both as the inorganic form and as organic complexes.

### Inorganic Cr

Elemental Cr arising from cooking and from manufacturing processes involving stainless-steel equipment is probably not absorbed and almost certainly has no nutritional value (*1*). The most usual ionic forms of Cr are $Cr^{3+}$ and $Cr^{6+}$. Hexavalent chromium is supplied to the organism in the diet or by inhalation of industrial contamination. Because of the rapid reduction of $Cr^{6+}$ in tissues and biological fluids, it is converted to $Cr^{3+}$, which is the biologically active form. We will thus be interested primarily in trivalent Cr.

### Organic Cr

The biologically active form of Cr, trivalent Cr combined with glutathione and with nicotinic acid (GTF), is present in the diet as brewer's yeast, in pig liver, and kidneys, or is synthesized from $Cr^{3+}$ in the organism (*1,2*). The exact structure of GTF remains to be fully elucidated. Regardless, it is essential that inorganic chromium be converted to a biologically active form in order to have a physiological role.

### The Absorption Coefficient

The absorption coefficient of Cr salts is variable: approx 0.5% for $CrCl_3$ and acetate (*2*), <1% for $^{51}CrCl_3$, and approx 40% for $^{51}Cr^{3+}$-trisacetylacetonate in rats (*3*). The yield of absorption of inorganic Cr is low, from 0.4 to 3% or even lower, without taking into account the dose and nutritional status of Cr (*4*). It was long assumed that natural complexes of Cr were absorbed better than simple Cr salts, since they were in a biologically active form, e.g., brewer's yeast (*5*). Recently, however, it has been shown that inorganic Cr is equally well absorbed (*6*). Anderson and Bryden (*7*) measured urine levels and showed that absorption of Cr arising from beer was similar to that of endogenous dietary Cr. Cr is absorbed rapidly: Labeled Cr is extensively absorbed in < 15 min, and urinary excretion in humans is high during the first 2 h following ingestion (*4*). The absorption of dietary Cr is a function of daily dose supplied. At the daily dose of 10 μg, almost 2% of the Cr is absorbed. At the dose of 40 μg, absorption decreases to 0.5%, and at doses higher than 40 μg, absorption remains constant at 0.4% (*4*).

### Site and Mechanism

Cr is absorbed in the intestinal mucosa. In rats (in vitro with $^{51}CrCl_3$), the middle section of the small intestine was the most active segment for Cr absorption, followed by the ileum and the duodenum (*8*). In humans, the site of absorption also includes the jejunum (*9*).

The mechanism responsible for the intestinal absorption of Cr is not well known. It is not known if Cr is absorbed passively (by a concentra-

tion gradient) or if it is absorbed by transport proteins located in the intestinal mucosa.

The several studies dealing with the intestinal absorption of Cr are not in perfect agreement. Thus, some authors believe that the absorption of trivalent Cr is a process that cannot be saturated; hence, it is a passive diffusion process (*10,11*). Results of in vitro studies using the isolated intestine from deficient rats (*12*), however, showed that the percentage of trivalent Cr absorbed decreased when its concentration in the incubation medium increased. This observed saturation effect suggests that transport proteins participate in the absorption of Cr. The same authors reported that cellular energy was not required for Cr transport. They concluded that trivalent Cr was absorbed by facilitated transport, since transport proteins were involved but oxidative metabolism was not.

Using a more physiological technique called double perfusion (perfusion of the intestinal lumen and simultaneous, but distinct vascular perfusion), it was shown that inorganic trivalent Cr was absorbed by a nonsaturable passive diffusion process (*13,14*). The same author noted that, in certain conditions, Cr could be absorbed by binding to transport proteins (case of zinc and iron deficiencies). The transport proteins for these metals are believed to be less specific and would also transport trivalent Cr.

## Interactions

### *With Amino Acids*

The absorption of Cr is facilitated by certain amino acids, such as histidine, which chelates Cr in the small intestine (*12*). These amino acids prevent the precipitation of Cr at the basic pH of the intestine and thereby increase its absorption.

### *With Vitamins*

Other factors have recently been demonstrated. They are required for Cr absorption and act in synergy with the element: nicotinic acid (in humans) (*15*) and ascorbic acid (in guinea pigs) (*16*).

### *With Dietary Carbohydrates*

Starch facilitates the absorption of Cr more than glucose, fructose, and sucrose (in rats)(*17*).

### *With Plasma Proteins*

Cr absorption can be influenced by the plasma proteins transferrin and albumin (*13*), as is the case for iron and zinc (*18*).

### *With Metals*

Metals can form complexes with Cr and modify its absorption. Zinc, vanadium, and iron decrease the absorption of Cr (*19–21*).

An oral dose of zinc decreased the absorption of $^{51}Cr$ in zinc-deficient rats, and Cr decreased the absorption of $^{65}Zn$ in zinc-deficient rats (*19*). This suggests that Cr and zinc can also be metabolized by a common mechanism in the intestine. The absorption of Cr is also influenced by vanadium. Large quantities of Cr prevented the reduced growth and death of chicks with high dietary intakes of vanadium (*20*). Iron-deficient animals absorbed more Cr than iron-replete animals (*21*). The administration of iron to these animals inhibited the absorption of Cr. Thus, Cr and iron must also share a common gastrointestinal transport mechanism.

### *With Chelating Substances*

Phytates significantly decreased the absorption of Cr in the intestine of rats, both in vivo and in vitro (*8*), whereas oxalate led to a significant increase. Other chelators, such as citrate and EDTA, are apparently without effect.

### *Physiological Factors*

Age: There are no modifications (*9*). Sex: Absorption is higher in women (0.93 ± 0.06%) than in men (0.64 ± 0.05%) (*22*), but this is related to the generally lower caloric content of the diet of women.

## BLOOD TRANSPORT

Absorbed Cr is bound to transferrin (siderophilin) (*21*). Transferrin possesses two binding sites, A and B, with different affinities for iron as a function of pH (experiments in vitro). It has been shown that Cr binds exclusively to site B. Thus, there is antagonism between $Cr^{3+}$ and $Fe^{3+}$ at high concentrations of iron (hemochromatosis). Recent work has verified that trivalent Cr has a high binding affinity for plasma transferrin (*23,24*). A low-mol wt Cr binding substance (LMCr) with a high affinity for $Cr^{3+}$ has been reported (*24*). This substance is responsible for a reversible and significant transfer of Cr to transferrin and not to albumin. Although transferrin and albumin affect Cr absorption, transferrin binds newly absorbed Cr, whereas albumin takes up the relay for transferrin when it is saturated. At very high concentrations, Cr can bind nonspecifically to other plasma proteins, such as γ- and β-globulins and lipoproteins (*21*). The level of Cr in the bloodstream is not a faithful reflection of the nutritional status of Cr, since it is not in equilibrium with body stores.

## TISSUE FATE

The distribution of Cr in body tissues has been determined in humans after autopsy, and in animals after the injection or ingestion of radioactive $^{51}Cr$.

## In Humans

The total quantity of Cr in the human body has been estimated at 1.7 mg. In addition, there are factors that modify the tissue concentrations of Cr in humans.

### The Geographic Locale

This is a point requiring confirmation, since the data available are old and used poorly adapted assay methods. Above all, sample collection and analysis posed problems of contamination (steel knives and steel contains 18% Cr).

### Subject Age

The concentrations of Cr in the lungs, aorta, heart, and spleen decrease considerably during the first months of life, whereas the liver and kidneys retain their neonatal concentrations up to the age of 10 yr (*25*). This decrease of the Cr concentrations with age is less pronounced in samples coming from countries other than the United States (*26*). The lungs are the only tissue in which an increase in the Cr content with age has been reported (after 20 yr), undoubtedly as a result of atmospheric pollution.

### Presence or Absence of Diabetes

Type I diabetics have modified tissue levels of Cr. Morgan (*27*) reported that the hepatic Cr content in diabetics is lower than that of control subjects. Schroeder et al. (*25*) similarly reported that the pancreas of diabetics contained less Cr than that of nondiabetics. Eatought et al. (*28*), on the contrary, found no difference in the Cr contents of the liver, pancreas, or spleen between diabetic and nondiabetic Pima Indians. The incidence of diabetes in this population is 50%, however, so the nondiabetic population may have included predisposed subjects, especially since the mean age in this group was 40 yr in comparison to 61 yr in the diabetic group.

## In Animals

Tissue distribution is modified as a function of the chemical form, age, species, and the presence or absence of diabetes.

### Chemical Form

Visek et al. (*29*) found that, after iv injection in rats, almost 100% of the $^{51}Cr$ administered in the form of sodium chromate was localized in the reticuloendothelial system, but only 55% of $^{51}Cr$ administered as $CrCl_3$ was in the liver. When $^{51}CrCl_3$ was buffered with acetate or citrate, <5% reached the liver, and most was excreted in the urine. When Cr was administered in the form of chromate, 25% reached the liver, but hepatic radioactivity rapidly decreased. Aside from the oral route, the tissue distribution of Cr is not modified as a function of the route of injection (iv, ip, intratracheal). Edel et al. (*30*) showed that $Cr^{6+}$ in the form of

sodium chromate was preferentially bound to the kidneys in comparison to other organs in rats, whereas $Cr^{3+}$ in the form of $^{51}CrCl_3$ was bound primarily in the liver and spleen. No difference was noted in the lungs as a function of valence.

### *Function of the Species*

Kraintz and Talmage (*31*) injected $^{51}CrCl_3$ in rats, and reported that 40% of the dose had been excreted by the kidneys 24 h later and that the highest remaining activity was in the bone marrow, indicating that $^{51}Cr$ was deposited in the reticuloendothelial system, similar to small colloids. In rabbits, however, the highest concentration of $^{51}Cr$ was in the spleen. Berggren and Flatt (*32*) showed that laboratory mice accumulated inorganic Cr in the exocrine and endocrine pancreas.

### *Function of Age*

The uptake of $^{51}Cr$ decreased with age in mice in certain tissues, in particular the liver, the testes, and fatty tissue of the epididymis. Hopkins (*33*) also observed differences in tissue retention as a function of age, after injecting physiological quantities of $^{51}Cr$ as $^{51}CrCl_3$. Adult rats retained less $^{51}Cr$ in the bone, but more in the spleen, kidneys, and testes, in comparison to immature rats. Wallach and Verch (*34*) reported less tissue retention of Cr in older rats, except in the spleen, and above all a major decrease in the bone. This is consistent with age leading to a modification of tissue Cr by lowering cellular Cr and transport, except for bone, where the mechanism is different, since Cr in this tissue is extracellular. The same authors (*35*) also showed a correlation between body wt and tissue retention of $^{51}Cr$: The uptake of $^{51}Cr$ by heavier rats was reduced by 10–30%. There is no difference in tissue distribution related to sex, dose, or preceding dietary regimes deficient or replete in Cr.

### *Function of the Presence or Absence of Diabetes*

Mathur and Doisy (*36*) reported that the tissue distribution of $^{51}Cr$ between diabetic and normal rats was similar, but that after homogenization the liver of the diabetic animals contained more $^{51}Cr$ in the nuclear and supernatant fractions than in the microsomal and mitochondrial fractions. The same profile was obtained in normal rats nourished with a high-fat diet. When the rate of hepatic lipogenesis is normal or high, however, Cr is apparently mobilized from the nuclear fraction to the microsomal fraction of liver cells. Raz et al. (*37*) reported increased levels of hepatic Cr in hyperinsulinic and hyperglycemic sand rats (*Psammomys obesus*) in comparison to normal and hyperinsulinic rats.

## Excretion

Absorbed Cr is excreted principally in the urine, and in small quantities in the hair, sweat, and bile (*33,38*). Since Cr is absorbed little, the major route of elimination after *per os* absorption is fecal. The kidneys are

a major route of elimination after injection, since at least 80% of the dose is eliminated via the kidneys (*39*). The presence of Cr in the feces after injecting $^{51}Cr$ has been noted (*29,33*), but the physiological significance is unknown. Urinary excretion, the major route of elimination of body Cr, is a good reflection of the ingestion of Cr, but not necessarily of its body status (*40, 41*).

The exact mechanisms of renal Cr metabolism are not known. The first studies suggested that 5–40% of plasma Cr III could be ultrafiltered and that 60–95% of ultrafiltered Cr was reabsorbed by the renal tubules (*42*). Ultrafiltrable Cr in the plasma is used as a marker for glomerular filtration. Stable complexes of Cr, such as Cr-EDTA, which is practically not reabsorbed, can also be used to indicate the rate of glomerular filtration. These initial studies of $^{51}Cr$ metabolism by the kidneys used only the isotope intravenously. It was shown that the distribution of Cr in the serum was different depending on whether the isotope was added directly or was absorbed *per os*. Donaldson et al. (*43*) showed that ultrafiltrable $^{51}Cr$ in the plasma of dogs was included between 9–19% if the isotope was given by gavage and only 2–3% if given parenterally. In addition, the same author believed that urinary Cr was equal to filtered Cr, so the predominant mechanism would be glomerular filtration without reabsorption. Wu and Wada (*44*) recently characterized a low-mol-wt substance (mol wt 1500) found in the urine of humans and rats that apparently binds trivalent Cr chemically. It is similar to a Cr-binding substance found in the liver of dogs and rats, as well as in other organs. This third hypothesis does not involve a classical renal mechanism, but rather one of binding. Water diuresis tests in ADH (vasopressin)-deficient rats did not affect the body concentrations of $^{51}Cr$, consistent with the possibility of proximal tubule reabsorption of Cr (*45*). Tests of isotonic saline diuresis have not shown an effect on urinary excretion, body retention of $^{51}Cr$, or any considerable effect on its tissue distribution. These findings also suggest that reabsorption does not occur in the collecting tubule or in the distal tubule, whereas proximal reabsorption is possible (*46*).

In summary, urinary elimination has not been totally elucidated. Is it glomerular filtration alone, is there proximal reabsorption, and what is the situation concerning binding to a low-mol-wt substance?

## DYNAMIC STUDY OF Cr METABOLISM BY COMPARTMENTAL ANALYSIS

Metabolic studies involving the iv administration of $^{51}CrCl_3$ to humans and to animals have shown that Cr III is rapidly eliminated from the blood and that most is excreted in the urine. During the first few days after administration, however, elimination is far from being total.

### *In Animals*

Mertz et al. (*10*) found that the elimination of $^{51}Cr$ administered intravenously was independent of the quantity injected and of the nutritional status of the animals (rats). They described body retention of Cr in terms of the sum of three compartments having three distinct phases with half-lives of 0.5, 5.9, and 83.4 d (72 d of observation). The quantity of residual Cr at the end of the observation period was still 13.5% of the dose injected.

Onkelinx (*47*) studied the metabolism of Cr after the iv injection of small quantities of $^{51}CrCl_3$ to rats of different ages. He observed that approx 93% of the $^{51}Cr$ was bound to proteins between 3–96 h and defined a three-compartment model (11 d of observation). The following hypothesis was formulated: Extracellular Cr is in equilibrium with two tissue compartments having different rates of Cr exchange. In addition, Cr can also enter nonexchangeable tissues. The influence of age on Cr metabolism was shown by compartmental analysis: There was an age-related decrease of the three components of excretory clearance, fu (urine), fd (feces), and fs (body sink). This indicates that Cr plays a physiological role and that its metabolism is subjected to regulation. In particular, the age-dependent fs component (elimination from relatively nonexchangeable pools) is a possible explanation for the fact that Cr levels in human tissues decrease with age.

Similarly to Onkelinx, Jain et al. (*48*) proposed a model in which extracellular Cr was in equilibrium with rapidly and slowly exchanging cellular pools and an internal tissue Cr pool with very slow exchange. All tissues contained pools that were exchangeable at varying rates, but the liver contained a larger fraction of the exchangeable pool than the pancreas. The picture was more complex for the kidney, with not only a large, rapidly exchanging pool, but also an internal pool in which $^{51}Cr$ taken up was not released rapidly.

### *In Humans*

Very few studies have been carried out in humans: to our knowledge only those of Doisy et al. (*38*) and Lim et al. (*49*). Doisy et al. studied the kinetics of $^{51}Cr$ in different types of populations: normal, the elderly, and types I and II diabetics. *Per os* absorption showed that plasma and urinary Cr in type I diabetics were much higher than that of the other groups, which were similar to the normal group. Following iv administration, urinary excretion by type I diabetics was higher than that of the other groups. In addition, external counting revealed an increase in the liver and in the precordial region around days 4–7.

Lim et al. compared the kinetics of $^{51}Cr$ after iv injection in normal subjects in comparison to those with hemochromatosis. In six subjects (three normal and three with hemochromatosis), the principal sites of Cr

concentrations were the liver, spleen, soft tissues, and bone. After ultracentrifuging plasma, two phases were found: One contained proteins binding $^{51}Cr$, e.g., transferrin, and another contained $^{51}Cr$ free or bound to low-mol-wt molecules (<90,000). Thus, 80% of injected radioactivity was in the plasma, and 20% was in the supernatant in hemochromatosis subjects, whereas in normal subjects, only 4% of radioactivity was in the supernatant, thereby confirming the role of transferrin in Cr transport. Lim et al. proposed a *physiological model* (Fig. 1) for $Cr^{3+}$ transport, including one central plasma compartment with two subcompartments called BB (Cr bound to proteins) and BF (free Cr), and peripheral compartments, including the liver and spleen. There are exchanges between BB and BF, favoring BB. The liver and spleen are organs whose function is primarily storage, although the metabolic processes of storage differ between the two. Uptake is primarily the role of adipose and muscle tissue. The term of uptake indicates only an accumulation with a short or medium half-life. A bone compartment was defined from scintigraphic images. A hypothetical "other organs" compartment corresponds to the fraction that was not detected.

A *functional model* (Fig. 2) was established:

Ninety-five percent of Cr is bound to plasma proteins and 5% is free,

Free Cr is rapidly transformed to bound Cr with a half-life of 4.6 min, and bound Cr is transformed more slowly to the free form, with a half-life of 1.4 h.

Free Cr is eliminated by the kidneys, with a half-life of 3.5 h.

The bound fraction is in equilibrium with three pools whose nature is unknown: One compartment is small (0.13 μg), with very rapid rates of exchange, i.e., a half-life of 12.1 min for transfer into the pool and 5.2 min for transfer out of the pool. Another compartment is of moderate size (0.8 μg), with an influx half-life of 0.84 d and an efflux half-life of 2.2 d. Finally, the third compartment is slow and large (24.4 μg), with influx and efflux half-lives of 4.2 d and 315 d, respectively.

The kinetics of Cr and its tissue distribution were relatively similar in the two groups, although there were differences in Cr transfer parameters between the two groups. This suggests that some parameters depend on physiological conditions.

## CONCLUSION

The metabolism of Cr is imperfectly understood. There remain a number of shadowy zones, in particular the mechanisms of intestinal

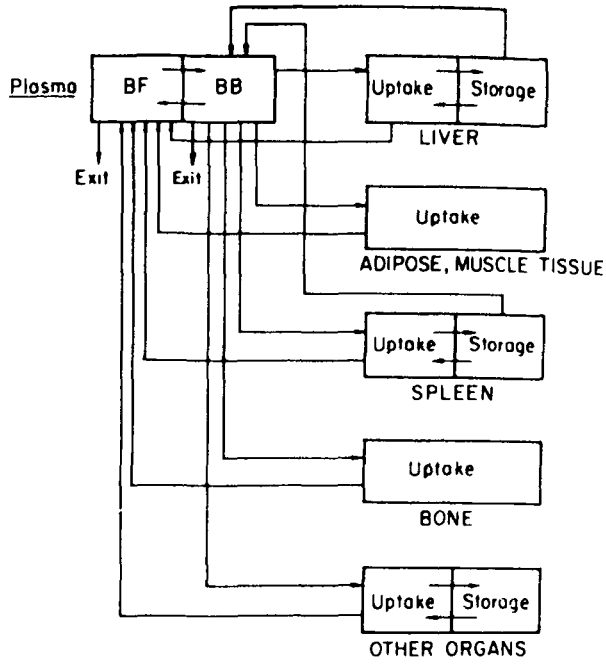


Fig. 1. *Physiological* model for transport of Cr III (*49*).

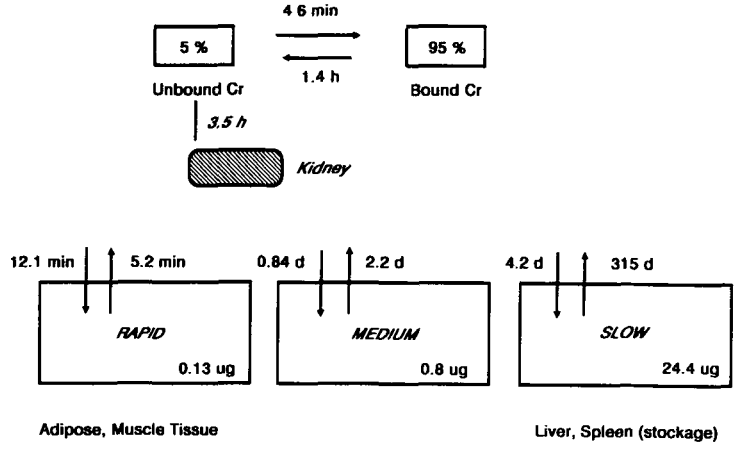


Fig. 2. *Functional* model for transport of Cr III (*49*).

absorption and urinary excretion. Studies carried out with radioactive $^{51}Cr$ have led to the elucidation of transport and have provided several hypotheses concerning urinary elimination. Future work will require the use of $^{51}Cr$-labeled compounds with very high specific activity. Another possibility not yet explored to our knowledge is the use of stable isotopes.

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