INVESTIGATIVE NUCLEAR MEDICINE

Effects of Dietary Magnesium Deficiency on Thallium-201 Kinetics and Distribution in Rat Myocardium: Concise Communication

Josep G. Llaurado, Jane A. Madden, and George A. Smith

Wood Veterans Administration Center, Marquette University, and The Medical College of Wisconsin, Milwaukee, Wisconsin

Kinetics and distribution of Tl-201 were studied in myocardium of rats with chronic dietary induced Mg deficiency. Rats were fed the Mg-deficient diet for 30 days and were then injected intravenously with 0.2 mCi of Tl-201. Comparable control animals were fed a standard laboratory diet. One-half hour after injection, rats were killed and a segment of myocardium was washed with nonradioactive Krebs solution in a special chamber. Radioactivity in the tissue was recorded continuously for 1 hr. A three-compartment model (extracellular, main intracellular, and subcellular) was found to describe adequately the kinetics of Tl-201. In myocardium of Mg-deficient animals, significant changes in values of transport rate constants and compartment sizes for Tl-201 indicated a moderate decrease in extracellular compartment and a threefold enlargement in subcellular compartment (presumably mitochondrial) at the expense of the main intracellular compartment, which underwent a marked reduction. Bulk Tl-201 uptake in myocardium of Mg-deficient rats was unchanged. The findings are interpreted as being consistent with mitochondrial alterations reported in Mg-deficient animals. Clinical implications are discussed.

J Nucl Med 24: 402–407, 1983

Magnesium, a predominantly intracellular cation, plays an important role in maintaining the integrity of the myocardium. Clinically, a Mg deficiency due to inadequate nutrition or abuse of diuretics or alcohol may reduce myocardial resistance to stress by other agents (*1*). Furthermore, experimental Mg deficiency has been shown to cause functional and histological cardiac damage (*1,2*).

There is evidence elicited by us (*3*) and others (*4–6*) that Mg deficiency induces marked changes in potassium kinetics and distribution within the myocardium. Thallium ion has been shown to resemble K physiologically, although significant kinetic differences have been found between them (*7,8*). However, in diagnostic cardiovascular applications of radionuclides, Tl ion can be considered an analog of K.

Received Sept. 11, 1982; revision accepted Dec. 17, 1982.

For reprints contact: Dr. Josep G. Llaurado, Nuclear Med. Service, Veterans' Hospital-115, Loma Linda, CA 92357.

Previously we reported (*9*) that Tl-201 is distributed throughout three compartments in the myocardium and that relative compartment sizes for Tl-201 change with regard to experimental condition (rest compared with exercise) and sampling time (0.5 hr compared with 3 hr after radionuclide injection). In view of the known effects of Mg imbalance on distribution of myocardial K, we undertook the present investigation to clarify the effects of dietary Mg deficiency upon Tl-201 kinetics in rat myocardium. Results indicate that chronically induced dietary Mg deficiency alters significantly the distribution of Tl-201 within myocardium.

MATERIALS AND METHODS

A group of male Wistar rats about 10 wk old was given a Mg-deficient diet* containing 3 ppm Mg as sole food for 30 days. Another group serving as control received a standard diet containing approximately 800 ppm Mg. Water for drinking (0.8 ppm Mg) was provided

ad libitum for both groups. On the 30th day of Mg-deficient diet administration, rats were anesthetized with intraperitoneally pentobarbital sodium (50 mg/kg) and injected intrajugularly with 0.2 mCi of ^{201}TlCl of specific activity greater than 200 mCi/mg in 0.2 ml of 0.9% NaCl.

One-half hour after injection, rats were guillotined and bled. The thoracic cavity was opened quickly and the heart excised. The anterior coronary artery served as a guide for consistent sampling of the same region of the left ventricle in all animals. Segments of left ventricle 0.5 mm in thickness were quickly obtained. Each segment (about 10 × 8 mm in cross section) was subjected to a continuous outflow with nonradioactive Krebs solution in an apparatus specially constructed in the laboratory (*10*). In the experiments with specimens from rats on the Mg-deficient diet, the Mg in the Krebs solution used for the continuous washing was adjusted to 0.6 mmole/l, which approximated the Mg concentration in plasma of these Mg-deficient rats.

With a well scintillation detector attached to a spectrometer and printer, radioactivity in the myocardial segment was recorded before starting the washout (t_0 time) and every 10 sec thereafter for 1 hr. Although counts were obtained every 10 sec, the following times were chosen for the processing of data in the computer input: from 0 to 120 every 10 sec; from 120 to 360 every 20 sec; from 360 to 840 every 30 sec; from 840 to 1800 every 40 sec; from 1850 to 3600 every 50 sec. This point selection provides a greater statistical weight on the initial part of the outflow curve. This is desirable for two reasons: (a) higher radioactivity counts are more accurate; (b) since the curve declines faster at the beginning, important information might otherwise be missed.

The printout value of counts in the course of time for each experiment constituted the input data for the compartmental analysis model with transport rate constants (k_{ij}) as primary parameters used to describe the kinetics of Tl-201 in myocardium. The SAAM (Simulation, Analysis And Modeling) computer program (*11*) was used to solve the compartmental model. The model considers Tl-201 partitioned into a three-compartment model consisting of 1) extracellular (EC), 2) intracellular (IC), and 3) subcellular (SC) space (Fig. 1).

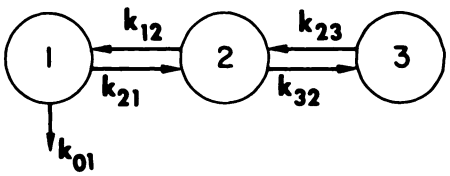

FIG. 1. Model of Tl-201 distribution in myocardium consists of three compartments: (1) extracellular (EC); (2) main intracellular (IC), and (3) subcellular (SC). Intercompartmental transport rate constants are symbolized by k_{ij}.

For further details see references (*12,13*). From numerical values for the k_{ij} it is possible to calculate (*12,13*) the relative compartment sizes (q_j/q_T) for Tl-201 at the beginning of the outflow (t_0).

In order to obtain a measure of total Tl-201 uptake, the remainder of rat's left ventricle was weighed and its activity counted. Results are expressed as percentage of activity in tissue to total injected dose, divided by left-ventricular mass.

Samples of plasma and left ventricular tissue were collected from the rats. Tissue samples were digested with 0.75 N HNO_3 for 48 hr. Both tissue and plasma samples were diluted appropriately and Mg concentration determined by atomic absorption spectrophotometry.

RESULTS

The adequacy of the Mg-deficient diet was assessed by measuring the Mg levels in plasma and heart tissue at the end of the diet period. Results are summarized in Table 1 where it can be seen that both plasma and heart tissue Mg concentrations were significantly lower in the Mg-deficient rats as compared with those fed the standard diet.

Typical examples of data plots for Tl-201 outflow from myocardium are shown for a rat fed the standard diet (Fig. 2) and for a rat fed the Mg-deficient diet (Fig. 3). Both experimental and theoretically calculated datum-point plots appear as a single curve (uppermost tracing) made up almost exclusively of coincidental points because adjacent points are nearly indistinguishable within the limits of resolution of the graph. The three curves labeled 1, 2, and 3 (Figs. 2 and 3) indicate

TABLE 1. COMPARATIVE VALUES OF Mg ION IN PLASMA AND MYOCARDIAL SEGMENTS OF RATS (MEAN ± s.e.)

Mg ion	Standard diet (N = 10)	Mg-deficient diet (N = 10)	p < *
Blood plasma mmole/l	1.0 ± 0.05	0.56 ± 0.04	0.0005
Left ventricular tissue mEq/kg wet mass	17.2 ± 0.30	9.85 ± 1.17	0.0005

* By Student's t-test, single tailed on account of expectancy of lower Mg concentrations in experimental group.

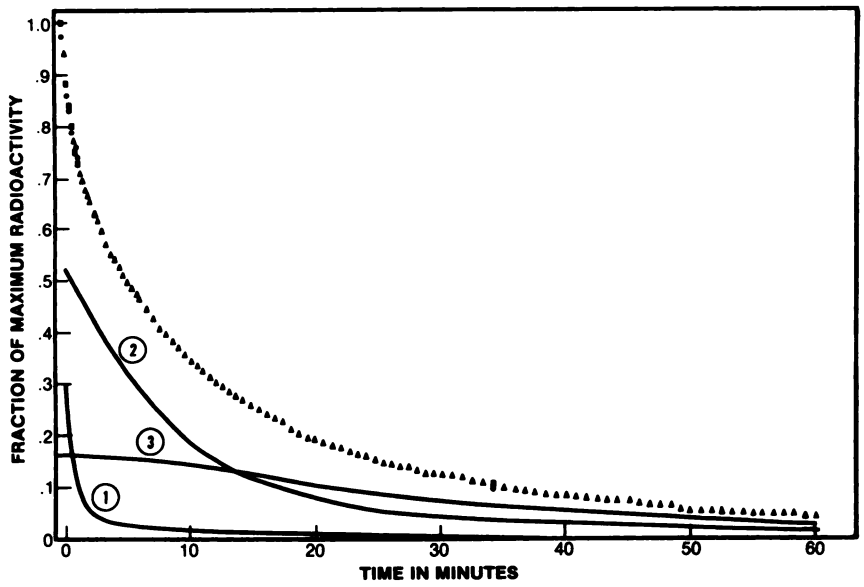

FIG. 2. Kinetics and distribution of Tl-201 in left-ventricular myocardial segment obtained 0.5 hr after i.v. injection of this tracer into rat fed a standard diet. Uppermost tracing is plot of Tl-201 outflow ((●)experimental; (■)theoretical; (▲)coincidental points) in total tissue segment where experimental and theoretically calculated curves appear as single curve owing to good agreement of the two sets of data. Three lower curves describing Tl-201 in each compartment are computer-simulated, and represent (1) extracellular, (2) main intracellular, and (3) subcellular compartments.

the fraction of radionuclide in each compartment as obtained from the computer output. These compartments represent EC, IC and SC spaces respectively in the proposed model. Note the marked preponderance, initially and throughout, of Compartment 3 (SC) in the specimen corresponding to the rat fed the Mg-deficient diet (Fig. 3).

In Table 2 values for the transport rate constants (k_{ij}) are compared. All, except k_{01}, which represents outflow of Tl-201 to the surrounding fluid, changed in a statistically significant manner for myocardial samples from Mg-deficient rats. Of particular interest are the 78% decrease in k_{23} and 79% increase in k_{32}, indicating an accumulation of Tl-201 in Compartment 3 (SC). Relative compartment sizes (q_j/q_T) are tabulated in the lower half of Table 2. Specimens from Mg-deficient rats showed significantly smaller Compartments 1 (EC) and 2 (IC), and a threefold enlargement of Compartment 3 (SC).

Dietary Mg deficiency did not alter the bulk Tl-201 uptake expressed as percent ratio of radioactivity in left-ventricular tissue to injected radioactivity per g of tissue (Table 2).

DISCUSSION

There is paucity of data regarding the influence of metabolic alterations on myocardial Tl-201 kinetics and distribution. An increase of myocardial bulk Tl-201 concentration in animals was noted with concomitant injection of Tl-201 and sodium bicarbonate (*14*); a recent study revealed that a drastic increase in K concentration of the perfusate medium modified the kinetics and compartmental distribution of Tl-201 in rabbit

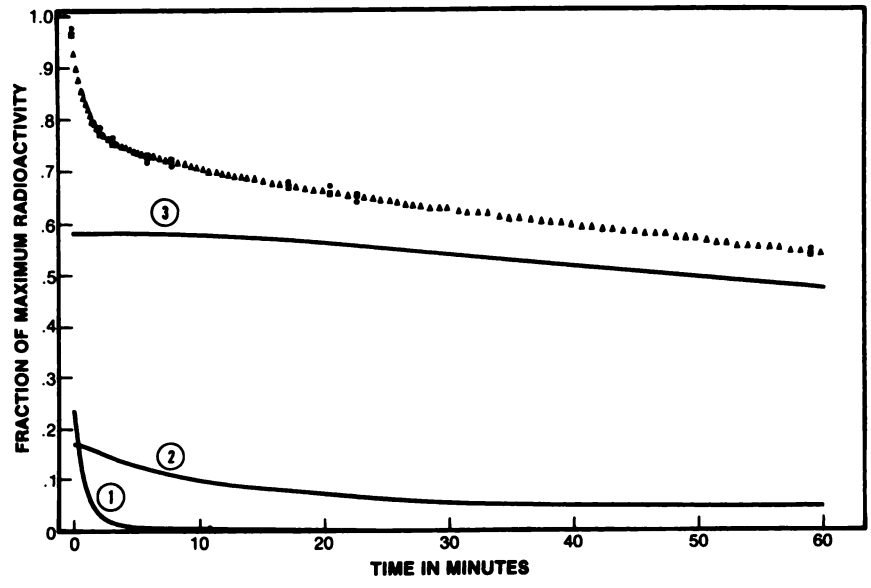

FIG. 3. Curves and symbols as in Fig. 2, but from rat maintained on a magnesium-deficient diet. Note higher values initially and throughout the study for Compartment 3 (SC), compared with those for rats fed a standard diet (Fig. 2).

TABLE 2. VALUES OF TRANSPORT RATE CONSTANTS* (k_{ij} sec^{-1}) AND RELATIVE COMPARTMENT SIZES† (q_i/q_T) FOR Tl-201 IN MYOCARDIAL SEGMENTS OF RATS (MEAN ± s.e.)

Parameter of interest	Standard diet (N = 10)	Mg-deficient diet (N = 10)	p < ‡
10^3k_{01}	19.5 ± 1.47	21.1 ± 1.70	NS
10^3k_{12}	2.12 ± 0.10	1.45 ± 0.15	0.002
10^3k_{21}	2.89 ± 0.22	0.98 ± 0.085	0.001
10^3k_{23}	0.59 ± 0.048	0.13 ± 0.016	0.001
10^3k_{32}	0.24 ± 0.016	0.43 ± 0.044	0.001
q_1/q_T	0.34 ± 0.023	0.25 ± 0.024	0.02
q_2/q_T	0.46 ± 0.018	0.17 ± 0.013	0.001
q_3/q_T	0.20 ± 0.023	0.58 ± 0.032	0.001
$(a_{LV}\cdot 100)/(a_D\cdot m_{LV})$§	0.71 ± 0.04	0.79 ± 0.03	NS

* Transport rate constant (k_{ij}) is the fraction of Tl-201 in compartment j (2nd subscript) that enters compartment i (1st subscript) in unit time. Values of k_{ij}, which have the dimension sec^{-1}, are multiplied by 10^3 to facilitate comparison.

† Relative compartment sizes are calculated as ratios between appropriate values of k_{ij}(12,13). Each q_i symbolizes the quantity (activity) of Tl-201 in its compartment and q_T the total quantity in the tissue at the beginning of the outflow (t_0).

‡ By Student's t-test, double tailed.

§ Percent ratio of radioactivity in left-ventricular tissue (a_{LV}) to injected dose (a_D) divided by mass of tissue (m_{LV}) in g (a measure of bulk uptake).

ventricle (*15*). Therefore, we felt that an investigation in which the compartmental distribution of Tl-201 within the myocardium under dietary mineral ion deficiencies, particularly of Mg, would be of value.

In the present study it is noteworthy that in Mg-deficient rats total Tl-201 remained unaltered in myocardium whereas marked changes occurred in the compartmental distribution of Tl-201. These findings reveal the importance of the cellular factors in Tl-201 distribution, and illustrate the usefulness of investigating organ compartmental distribution of a given electrolyte (or chemical substance in general) as well as total amount. In this context, we note that long ago Davson and Danielli (*16*) and Adolph (*17*) pointed out that the partition of electrolytes between extra- and intracellular fluids is of greater physiological importance than the total amount.

Extensive evidence concerning the validity, accuracy and limitations of studying compartmental Tl-201 by the present method has been presented earlier (*8,9*). It was shown therein that most of the thallium in myocardium is normally intracellular ($q_2 + q_3$). In Mg-deficient rats there was a significant reduction (from 0.34 to 0.25, Table 2) of Tl-201 in the extracellular space (q_1). Quantitatively, however, the most remarkable effect of dietary Mg deficiency was the translocation of Tl-201 (from 0.20 to 0.58, Table 2) into Compartment 3 (q_3) representing subcellular space mostly at the expense of the main intracellular space (q_2).

Although the kinetic approach of compartmental analysis does not directly identify cytological components, independent workers have shown that heart mitochochondria are organelles capable of considerable thallous-ion uptake (*18*). Furthermore, ultramicroscopy has revealed that Mg deficiency for as little as 12–14 days causes mitochondrial swelling (*19*). The finding that Compartment 3 (subcellular) showed a considerable thallous-ion accumulation in myocardium of chronically Mg-deficient rats is consistent with the above observations (*18,19*). Loss of Mg from heart mitochondria is most likely responsible for inability to maintain physiological ionic gradients between these organelles and main cytoplasm. These defects may impair metabolic and respiratory functions of the mitochondria, producing myocardial alterations seen in Mg-depleted patients and animals (*20*). It has been found very recently that Mg can exert a protective effect from ischemia on myocardium (*21*).

It is also of interest that the changes in sizes of compartment induced by Mg deficiency—decreased EC (q_1) and IC (q_2), with considerable increase of SC (q_3)—are qualitatively in the same direction as those seen after strenuous exercise (*8,9*); again, mitochondria have shown to enlarge markedly after exercise (*22,23*). Quantitatively, however, the change in SC Tl-201 distribution induced by Mg deficiency is far more pronounced than that seen after exercise (*8,9*).

One of the factors that seemingly has limited the widespread clinical use of thallium-201 chloride as a myocardial imaging agent is the poor correlation of its results with other findings. In this context, studies on kinetics and distribution of Tl-201 in myocardium and/or other tissues may throw light on the mechanism(s) whereby this ion is handled by cardiac muscle.

For instance, the increased size of Compartment 3 after exercise (*9*) is due principally to increased k_{32}, the transport rate constant representing transfer from Compartment 2 into 3, while the same result in Mg deficiency can be to a large extent attributed to decreased k_{23}, the transport rate constant representing transport from compartment 3 into 2. Thus, with regard to mechanisms, these findings may suggest that exercise determines an increased uptake of Tl- 201 in Compartment 3 (presumably mitochondria) whereas Mg deficiency induces a fixation or delayed release of Tl-201 in this Compartment 3.

Because distribution of Tl-201 in myocardium is related to both regional blood flow and efficiency of extraction by myocardial cells (*24*), one would expect that the larger the proportion of myocardial Tl-201 contained in the SC or innermost compartment, the longer it would remain in the myocardium. This situation has been proven to occur when animals are injected with Tl-201 after strenuous exercise (*9*). The clinical counterpart of this phenomenon is the fact that vigorous exercise before, and for a short time after, Tl-201 injection into the subject gives better myocardial images than those obtained at rest.

Since we have shown experimentally that in chronic dietary Mg deficiency (a) bulk Tl-201 uptake in myocardium is unchanged, and (b) the subcellular (SC) distribution of Tl-201 resembles that in myocardium after strenuous exercise, these facts provide solid grounds for the conclusion that Mg deficiency seen clinically is not likely to impair the quality of heart scintigraphy; it also may explain why poor quality Tl-201 scintigrams in Mg-deficient patients have not been clinically reported in spite of the relatively frequent occurrence of Mg deficiency in the age group incurring higher incidence of coronary artery disease.

FOOTNOTE

* Teklad Rest Diets, Madison, WI.

ACKNOWLEDGMENTS

This work was largely supported by research funds from the Veterans Administration. Dr. Jane A. Madden, was Young Investigator of the National Institutes of Health-National Heart, Lung and Blood Institute during this work. We are very grateful for the secretarial assistance of Genevieve Wollinka.

REFERENCES

1. SEELIG MS: Magnesium deficiency loss from myocardium. In *Magnesium Deficiency in the Pathogenesis of Disease.* New York, Plenum Medical Book Co., 1980, pp 185-205
2. RAAB W: Cardiotoxic effects of emotional, socioeconomic, and environmental stresses. In *Recent Advances in Studies on Cardiac Structure and Metabolism. 1. Myocardiology.* Bajusz E, Rona G, Eds. Baltimore, University Park Press, 1972, pp 707-713
3. MADDEN JA, SMITH GA, LLAURADO JG: Myocardial potassium kinetics in rats on magnesium deficient diet. *J Am Coll Nut* 1:111, 1982
4. DYCKNER T, WESTER PO: Ventricular extrasystoles and intracellular electrolytes before and after potassium and magnesium infusions in patients on diuretic treatment. *Am Heart J* 97:12-18, 1979
5. ALTURA BM, ALTURA BT: Magnesium ions and contraction of vascular smooth muscles: relationship to some vascular diseases. *Fed Proc* 40:2672-2679, 1981
6. SHINE KI, DOUGLAS AM: Magnesium effects on ionic exchange and mechanical function in rat ventricle. *Am J Physiol* 227:317-324, 1974
7. LLAURADO JG, MADDEN JA, SMITH GA: Differences between ^{201}Tl and ^{43}K tissue distribution in rested and exercised rats. *Nucl Med* 20:40-45, 1981
8. LLAURADO JG, SMITH GA, MADDEN JA: Effects of exercise on kinetics and distribution of K-43 and Tl-201 in isolated myocardium: Concise communication. *J Nucl Med* 22:441-446, 1981
9. LLAURADO JG, SMITH GA, MADDEN JA, et al: Partition of thallium-201 in isolated myocardial tissue of rats previously injected at rest or after exercise. *J Nucl Med* 20:1136-1141, 1979
10. LLAURADO JG: Digital computer simulation as an aid to the study of arterial wall Na kinetics. *J Appl Physiol* 27:544-550, 1969
11. BERMAN M: Compartmental analysis in kinetics. In *Computers in Biomedical Research.* Vol. 2, Stacy RW, Waxman BD, Eds. New York, Academic Press, 1965, pp 173-201
12. LLAURADO JG: Relationship between kinetics of inflow and outflow as the basis of a computer simulation for solving compartmental models: example of electrolyte transfers in cardiovascular tissues. In *Dynamic Studies with Radioisotopes in Medicine*, Vienna, International Atomic Energy Agency, 1971, pp 13-26
13. MADDEN JA, SMITH GA, LLAURADO JG: Myocardial kinetics and organ distribution of thallium-201 in two different age groups of rats. *J Gerontol* 37:16-23, 1982
14. HETZEL KR, WESTERMAN BR, QUINN JL, et al: Myocardial uptake of thallium-201 augmented with bicarbonate: Concise communication. *J Nucl Med* 18:24-25, 1977
15. KRIVOKAPICH J, SHINE KI: Effects of hyperkalemia and glycoside on thallium exchange in rabbit ventricle. *Am J Physiol* 240:H612-H619, 1981
16. DAVSON H, DANIELLI JF: *The permeability of natural membranes*, Cambridge, Great Britain, Cambridge University Press, 1952, p 177
17. ADOLPH EF: *Origins of physiological regulations.* New York, Academic Press, 1968, p 43
18. BARRERA H, GOMEZ-PUYOU A: Characteristics of the movement of K^+ across the mitochondrial membrane and the inhibitory action of Tl^+. *J Biol Chem* 250:5370-5374, 1975
19. NAKAMURA M, NAKATINI M, KOIKE M, et al: Swelling of heart and liver mitochondria from magnesium deficient rats and its reversal. *Proc Soc Exp Biol* 108:315-319, 1961
20. HEGGTVEIT HA: The cardiomyopathy of Mg-deficiency. In *Electrolytes and Cardiovascular Diseases. 1.* Bajusz E, Ed. New York, S. Karger, 1965, jpp 204-220
21. BERSOHN MM, SHINE KI, STERMAN WD: Effect of increased magnesium on recovery from ischemia in rat and rabbit hearts. *Am J Physiol* 242:H89-H93, 1982
22. LAGUENS RP, GOMEZ-DUMM CLA: Fine structure of

myocardial mitochondria in rats after exercise for one-half to two hours. *Circ Res* 21:271–279, 1967

23. KING DW, GOLLNICK PD: Ultrastructure of rat heart and liver after exhaustive exercise. *Am J Physiol* 218:1150–1155, 1970

24. POHOST GM, ALPERT NM, INGWALL JS, et al: Thallium redistribution: mechanisms and clinical utility. *Semin Nucl Med* 10:70–93, 1980