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Influence of parathyroid state on calcium uptake in bone

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Abstract

The exchange of calcium and strontium ions in bone was studied in control dogs, dogs made hypocalcemic by parathyroidectomy, and dogs rendered hypercalcemic by injection of parathyroid hormone. After injections of tracer into the tibial nutrient artery, extraction of tracer during transcapillary passage was measured and expressed as a fraction of 1. Extraction over the first 3 min in normal dogs was 0.46 ± 0.09 ($n = 6$), in hypocalcemic dogs it was increased to 0.53 ± 0.07 ($n = 6$), and in hypercalcemic dogs it was decreased to 0.39 ± 0.07 ($n = 5$). Subsequent washout was less rapid than normal in hypoparathyroid dogs and more rapid than normal in hyperparathyroid dogs. We conclude from this that the immediate volume of distribution in bone (or the number of available binding sites) for strontium diminishes as the parathyroid hormone level increases.

Keywords

blood flow; transport calcium tracers; parathyroid glands; ion regulation; volumes of distribution; calcium binding

The principal objective of this study was to clarify the role that bone may play in the control of the calcium ion level in blood. Two apparently contrary positions are held: one, that calcium homeostasis is mainly under renal or gut control (16, 18, 20), and the other, that there is a controlling physiologic mechanism in bone for ionic transport (14, 24). It is probable that both of these systems operate in parallel, but their relative importance remains unclear.

In a series of in vivo studies from this laboratory (3, 4, 7, 11, 17), it has been demonstrated that the principal mechanism of exchange between blood and extravascular fluid is by passive diffusion across the capillary wall, even for Sr^{2+} and Ca^{2+} , which concentrate in bone. This concept is also supported by evidence of Neuman et al. (15) from in vitro experiments. However, calcium and strontium differ from other solutes in being strongly retained in bone; that is, they have a large volume of distribution. It was the purpose of this investigation to use indicator-dilution studies to estimate the retention and washout of strontium and calcium in the canine tibial bone in the control situation and during hypocalcemia and hypercalcemia.

MATERIALS AND METHODS

The principal experiments were concerned with the effect that parathyroid hormone has on readily exchangeable calcium ions in bone. Preliminary validating experiments were undertaken so that the interpretation of the final experiments was accurate and justified.

The studies concerned the early phases of exchange, using the multiple indicator-dilution technique in two ways: first, to use the venous outflow curves to reveal the kinetics of rapid exchange with high resolution and then to use the residue curves (obtained by external gamma detection) for observing retention and rate of washout over the first hours after injection.

Primary Experiments

These experiments concerned the effect of parathyroid state on the rapid exchange of strontium. Three groups of dogs were studied: six normal dogs, six dogs made hypocalcemic by parathyroidectomy, and five dogs made hypercalcemic by injections of parathormone. The exchange of strontium was studied by observing 1) the immediate extraction during the first transcapillary passage, 2) the retention in bone during the first 3 min, and 3) the washout from the tibia over a 3-h period.

Parathyroidectomy was performed and the thyroid lobes preserved. The animals were allowed to stabilize before further study. That all the parathyroid tissue had been removed was demonstrated by the fall in the serum calcium level. Dogs were made hypercalcemic by injections of bovine parathyroid hormone (Eli Lilly), given as 10 U/kg, 3 times each day, every day for 3 or 5 days preceding the study.

General Experimental Procedure

All the animals studied were adult mongrel dogs weighing between 14 and 28 kg. Cannulation of the tibial nutrient artery and ipsilateral femoral vein was performed as previously described (8, 9), and the details for tibial perfusion have been reported previously (11). The dog was positioned so that each tibia could be monitored by the external detectors as described previously (9).

For Sr^{2+} , Ca^{2+} , and albumin, the relevant flow of solute-containing fluid is the plasma flow. In some of the experiments this plasma flow was changed while the total blood flow was kept nearly constant. This was done by diluting the blood; the perfusate calcium ion concentration was kept steady by the use of calcium gluconate-physiologic saline as the diluent. The plasma flow, F_s , in milliliters per minute per 100 g of bone, was $F_B (1 - \text{Hct})/W$, where F_B is the blood flow delivered by the Holter pump, Hct the hematocrit, and W the mass of the tibia perfused (11), as determined at the end of the experiment.

A 0.25-ml mixture of tracers, either albumin- ^{51}Cr or albumin- $^{99\text{m}}\text{Tc}$ and a tracer that concentrates in bone—strontium or calcium, or both—was injected into the nutrient artery. Femoral vein blood samples were collected every 5 s for 3 min, and washout was monitored with the external NaI detector for 3 h.

Strontium chloride (New England Nuclear, Boston) was administered in a dose of 5 μCi , and calcium-47 (Oak Ridge National Laboratory, Oak Ridge, TN), whose half-life is short (4.54 days), was employed to validate the use of ^{85}Sr . For a vascular reference tracer, albumin- ^{51}Cr (Squibb, Chicago) was used in experiments with ^{47}Ca alone, and albumin- $^{99\text{m}}\text{Tc}$ (supplied by the Nuclear Medicine Dept., Mayo Clinic) was used in experiments with ^{85}Sr , to minimize spillover from high-energy to low-energy windows. The dose for these vascular reference tracers was 5 μCi .

Tracer content of the tibia was estimated from the gamma activity detected with a 2.5-inch thallium-activated NaI crystal (Harshaw integral line scintillation type 125AF 1213, Solon, OH) placed over the midshaft. The remaining elements (preamplifier, amplifier, single-channel analyzer, scaler, data buffer, and timer) were supplied by Canberra Industries, Nuclear Division, Meriden, CT. The energy spectrum was determined for each isotope, and window settings were adjusted to span the main peaks. Spillover ratios were determined when more than two isotopes were counted together. For determining the gamma emission in samples of blood and bone, a Beckman 310 system (Beckman Instruments, Fullerton, CA) was used.

Analysis of Indicator-Dilution Curves

The instantaneous fractional extraction is determined by

$$E(t) = \frac{h_R(t) - h_D(t)}{h_R(t)} \quad (1)$$

where $h(t)$ is the fraction of the injectate appearing per unit time in the femoral vein outflow, R the vascular reference tracer, and D the diffusible or permeant tracer. The outflow dilution curve is calculated for each tracer by plotting $h(t)$ versus time, where

$$h(t) = C(t) \cdot (F_v / q_0) \quad (2)$$

in which q_0 is the total counts per minute from the injectate, F_v the femoral vein blood flow, and $C(t)$ the tracer concentration (cpm/ml) in the outflowing blood.

Calcium tracers have a prolonged retention in bone, and at a particular time, T , this retention can be quantitated by calculating a net extraction, $E_{\text{net}}(t)$, as in Eq. 3. After the reference tracer has washed out, this value is equivalent to the difference between the total dose injected and the integral of the amount in the outflow and is then the same as $R(t)$.

$$E_{\text{net}}(T) = \frac{\int_0^T [h_R(t) - h_D(t)] dt}{\int_0^T h_R(t) dt} \quad (3)$$

The fraction of the injected dose remaining in the tissue at any time is the residue function, $R(t)$, which is $1 - \int_0^T h(t') dt'$. It is determined directly by the surface detection of gamma emissions over the tibia

$$R(t) = \frac{C^*(t)}{C^*(0)} \quad (4)$$

where $C^*(0)$ is the peak activity over the tibia and $C^*(t)$ is the activity at any time, t , during the 3-h period of washout.

The fractional escape rate (FER) during the washout period is expressed as a ratio of the rate of loss of tracer divided by the amount still residing within the tissue at time t and is equal to

$$\text{FER} = \frac{\Delta R / \Delta t}{R(t)} \quad (5)$$

Capillary permeability-surface products, PS_c , were calculated by the standard formula

$$PS_c = -F_s \log_e (1 - E_{\max}) \quad (6)$$

where PS_c , ($\text{ml} \cdot \text{g}^{-1} \cdot \text{min}^{-1}$) is the product of the capillary permeability, P (cm/min), and the capillary surface area per unit mass of tissue (cm^2/g); F_s , is the flow of plasma ($\text{ml} \cdot \text{min}^{-1} \cdot 100 \text{ g}^{-1}$); and $E_{\max}$ is the smoothed maximum plateau value of the instantaneous extraction, $E(t)$.

$E_{\max}$ was obtained by drawing a smooth curve through $E(t)$ from the 15th to the 40th s and is the smoothed value at a time at or just past the peak of $h_R(t)$, when about half of the reference tracer has passed the sampling point. Because $E(t)$ was rather flat in this region, the choice of the particular time did not influence the estimates of PS_c . This method was detailed in a previous publication (11).

When flow is known, one may normalize the residue function, $R(t)$, from individual experiments by plotting $R(t)$ against $F_s \cdot t$; this product is the volume of plasma that has left the bone by time t after injection. Such a plot would cause all curves for one tracer to become superimposed on one another if the tracers were freely diffusible and the washout was limited solely by flow, as is the case with antipyrine (see Fig. 4 of Ref. 9).

An apparent tissue-blood partition, λ , for calcium tracer was determined from the ratio of activity in bone to that in blood while it was recognized that this does not represent an equilibrium, but only a near equilibrium between blood and more accessible or exchangeable bone calcium at chosen times after injection.

$$\lambda = \frac{\text{cpm 1 ml bone}}{\text{cpm 1 ml blood}} \quad (7)$$

In preliminary experiments not reported in detail here, we had observed that perfusing the tibial nutrient artery at 1 and 2 $\text{ml} \cdot 100 \text{ g}^{-1} \cdot \text{min}^{-1}$ had a definite effect on the rate of washout of ^{85}Sr from the tibial diaphysis. Thus, even though collateral flow from epiphyseal or metaphyseal sources sufficed for washout, it appeared that some minimal nutrient artery

flow was needed to create a nearly normal enough situation so that the question of flow limitation to washout could be examined.

The examination for a flow effect was then performed in a situation in which we attempted to keep the overall perfusion of the vascular bed constant by keeping the total blood flow and pressure constant. Changes in the relevant perfusate flow were then induced by changing the hematocrit, as will be described later; this allowed us to look for changes in washout rate with little or no alteration in the status of the local vascular bed.

Other preliminary experiments were done as controls for certain aspects of the primary experiments. These were as follows.

Comparison of ^{85}Sr and ^{47}Ca —It was desirable in these experiments to use ^{85}Sr as a calcium tracer because of its ready availability and relatively long half-life (64 days). However, this substitution required validation, for it is known in tissues such as the heart (10) that strontium shows some differences from calcium in transport rates and in its binding to contractile proteins. The experiments were performed on five dogs with the use of both ^{47}Ca and ^{85}Sr simultaneously, and then the extraction and the volume of distribution of each were compared.

Distribution of tracer between bone tissue and marrow—To demonstrate that the tracer was in bone tissue and not in marrow or other tissues peripheral to the bone, we removed the bones in four dogs at the end of the 3-h experiment, separated the various fractions of the bone, and counted them for tracer individually. Recirculation of tracer was analyzed from data obtained from the opposite tibia.

Influence of flow on strontium washout—Because parathyroid hormone is a known vasodilator (2), it is necessary to know its effect on calcium homeostasis independent of blood flow. In seven animals (four controls, one parathyroidectomized, and two given parathyroid extract), the effects of varying the plasma flow through the nutrient artery on the washout of strontium were examined.

RESULTS

Comparison of ^{85}Sr and ^{47}Ca

The experiments performed were in two classes: 1) outflow dilution curves and extraction and 2) apparent volumes of distribution after prolonged whole-body equilibration.

Outflow dilution curves and extractions—In four dogs, injections of multiple tracers were made into the tibial nutrient artery without continuous perfusion of the nutrient artery; thus, there was dependence on collateral circulation. The tracers injected were chromium-labeled albumin, ^{47}Ca , and ^{85}Sr injected as an extended bolus over several seconds at a constant rate. The outflow indicator-dilution curves were obtained by sequential sampling from the femoral vein, and both instantaneous and net extractions were calculated from these curves as a function of time. An example is shown in Fig. 1. In this experiment the strontium and calcium curves showed about 50% extraction when the dilution curves were at

a peak. The instantaneous extractions, $E(t)$, diminished gradually from the 30th s to the point at which backdiffusion occurs, where the dilution curves of Sr and Ca cross albumin. The net extractions, which are an average extraction compared with the reference albumin, showed only a very gradual diminution even after the instantaneous extraction was negative; this finding indicated that the return of tracer that had escaped into the tissues and reentered the bloodstream was very low. For this particular set of data, the net extraction at the end of 3 min was still 45%, which would indicate that the residue function at this time for strontium and for calcium would have been 45% or slightly higher in accordance with Eqs. 3 and 4.

Apparent volumes of distribution after prolonged whole-body equilibration—

After intravenous injection of the ^{47}Ca and ^{85}Sr , samples were taken from bone and blood at 15–150 min. The results are presented in Table 1. [In this comparison it is important that calcium and strontium uptake by erythrocytes either be negligible or be the same; the evidence is that Ca^{2+} exchange in human erythrocytes is exceedingly low (21).] For this test, it is not important that equilibration of these tracers between plasma and bone be complete, but rather that the equivalence between calcium and strontium be reasonably close. As seen from Table 1, this is apparently the case; the rates of uptake of calcium and strontium by the bone are similar.

Distribution of Tracer in Bone and Marrow at End of Washout

To enable us to make the appropriate interpretation of blood-bone exchanges, it was critical to demonstrate that the tracer being observed was in bone and not in marrow or other tissues peripheral to the bone. In four of the dogs in which ^{85}Sr injections were made into the nutrient artery for examination of the kinetics of washout from the bone, both tibias were removed after the dogs had been killed at the end of the 3-h period of washout, and counts were obtained from tibial bone and from various fractions. The influence of whole-body recirculation of tracer was studied by dissection of the opposite tibias in the same fashion. The data were obtained in two hypoparathyroid dogs, one control dog, and one hyperparathyroid dog. In all four of these experiments, the nutrient artery was perfused with whole blood via a pump at constant flow rate for the 3-h period. The results are shown in Table 2.

The effects of whole-body recirculation and secondary deposition in the tibia can be shown by comparing the counts obtained over the experimental tibia, column 4, with those obtained from the opposite tibia, column 5. It may be seen that the opposite tibia, representing recirculation, shows count rates that are above background; however, they average less than 5% of the experimental tibia and thus indicate minimal interference from recirculation of tracer.

For determination of the marrow contribution to external counting, the tibia was split lengthwise, the fatty marrow was removed and counted (column 7) cancellous bone was curetted out and counted (column 8), and the amount remaining in the tibia cortical bone was counted (column 6). The results show that only a small portion was in marrow, in agreement with our previous findings (17) and with the observations of Bosch (1).

Influence of Flow on Strontium Washout

The question of concern was whether varying the flow of “solute-containing mother fluid,” the plasma flow, F_s , affected the rate of tracer washout or whether the washout was mainly barrier-limited for tracer that had exchanged with the extravascular regions of the bone.

The results in one normal dog are shown in Fig. 2. The total blood flow was almost constant over a 3-h period, with $F_B = 2.5 \text{ ml} \cdot 100 \text{ g}^{-1} \cdot \text{min}^{-1}$. At $t = 30 \text{ min}$, the hematocrit was lowered to 6%; at $t = 75 \text{ min}$, it was increased to 38%; and at $t = 130 \text{ min}$, it was decreased again to 14%. The changes in F_s (solid line) were -35% at 75 min and $+26\%$ at $t = 130 \text{ min}$; neither of these changes resulted in definitive changes in the fractional escape rate, FER, which indicates that the degree of flow limitation to ^{85}Sr washout was minor and that the main barrier to washout was a diffusive or membrane-transport process. This was true in the three other controls and also in the one animal made hypoparathyroid and the two made hyperparathyroid.

Effect of Parathyroid State on Rapid Exchange of Strontium

Data on strontium extraction, retention, and washout are shown in Table 3. These results are analyzed in terms of the initial extraction and retention and then in terms of the retention at longer times and the measurement of washout.

First-pass extraction: $E_{\max}$ and PS_c —The $E_{\max}$ of strontium can be estimated near the peak of the albumin reference indicator-dilution curve obtained from the venous outflow and used together with the known flow to calculate the permeability-surface area product, PS_c , (Eq. 6). In the control state, the initial extractions averaged 0.60 ± 0.14 (SD, $n = 6$), and the PS products averaged $0.89 \pm 0.24 \text{ ml} \cdot 100 \text{ g}^{-1} \cdot \text{min}^{-1}$.

In the hypoparathyroid state, the extractions were modestly increased but not significantly, as was also true for the estimates for PS_c . As with the controls, the PS_c estimates were calculated on the basis that the pump perfusion flow of plasma is the total flow through the region that has washed out. This flow is very close to cortical bone blood flow in the awake, standing dog, which is $2.5 \text{ ml} \cdot 100 \text{ g}^{-1} \cdot \text{min}^{-1}$ of whole blood (13).

In the hyperparathyroid animals, the instantaneous extractions, $E(t)$, were perhaps a little less than the values in the hypoparathyroid dogs; accordingly, the mean PS_c was somewhat less than in the hypoparathyroid dogs, but the values were not significantly different from either those in the hypoparathyroid dogs or those in the control animals.

We conclude from these experiments that capillary permeability-surface area products are not influenced by the parathyroid state. Lumping all of the observations together provides estimates of PS_c for strontium of $1.11 \pm 0.25 \text{ ml} \cdot 100 \text{ g}^{-1} \cdot \text{min}^{-1}$ ($n = 17$).

Retention and net extraction, E_{net} , at 3 min—The computation of E_{net} is given in Eq. 3, and an example is shown in the inset graph in Fig. 1. The values of E_{net} at $t = 3 \text{ min}$ were chosen as representative of the early extraction during the first few minutes. Although E_{net} was calculated from the outflow indicator-dilution curves by comparison with the albumin reference curve, it should give a value at $t = 3 \text{ min}$ that is very close to $R(t)$ at that same

time, since most of the albumin has washed out by the end of 3 min. Thus, E_{net} gives an indication of retention of the tracer by the bone, and, because there has been substantial time available for exchange, it is an indicator of processes going on beyond the capillary membrane and is not influenced solely by the capillary permeability. Values for E_{net} differ statistically in the hypoparathyroid state compared with the hyperparathyroid state. The scatter among the control values was such that there was no statistically significant difference from either hypocalcemic or hypercalcemic dogs.

Retention at longer times: $R(t)$ —Residue functions, $C^*(t)/C^*(0)$, were recorded from the 17 animals with known perfusion rates through the nutrient artery. Because there may be some influence of flow on the washout rate—indicating some flow limitation to washout rather than purely barrier limitation, the residue functions, $R(t)$, are presented in two ways in Fig. 3. In Fig. 3A, the values of $R(t)$ are plotted as a function of time. In an attempt to account for the partial influence of flow on the exchange, these same curves are plotted in Fig. 3B as a function of the accumulated volume that is being pumped through the bone, that is, the product of the plasma flow, F_s , times time. The values of $R(t)$ at a particular phase of the washout at which time the product $F_s \cdot t = 0.8$ ml/g are given in Table 3 and plotted in Fig. 4A. The data of the table and the figures demonstrate that hypoparathyroid animals retain strontium for a longer time than do hyperparathyroid animals. The data are summarized in Fig. 4. At a time when $F_s \cdot t = 2$ ml/g, the hypoparathyroid animals still retained, on the average, 24% of the injected tracer, whereas the hyperparathyroid animals retained 13% of the tracer. The controls were in between, with a retention of 18%.

These data confirm, from a very different kind of experimental viewpoint, what was predicted by the values for E_{net} , namely, that the hypoparathyroid animals retained more tracer than did the hyperparathyroid animals.

Fractional escape rate (FER)—Table 3 shows the rates of washout values for FER at a succession of intervals. In the first minute and also to some extent in the first 30 min, the FER reflects what one would predict from E_{net} and the observed residue functions at $F_s \cdot t = 2$ ml/g. However, at very long times, as shown in the last column of the table during the period from 60 to 180 min, the FER in the three groups became statistically indistinguishable. This would suggest that most of the tracer that had been taken up and retained had been handled by the transport mechanism during the early times—for the most part in the first 30 min. Because all of these FER values were similar during the 2nd and 3rd h, there was actually more tracer being washed out of the hypoparathyroid animals during this time than there was from the hyperparathyroid animals, which had lost the major fraction much earlier.

DISCUSSION

The preliminary experiments, in which calcium and strontium were used simultaneously, indicate that their handling by bone tissue is essentially the same. Comparison of the outflow dilution curves and the apparent volumes of distribution showed no significant differences. This evidence justifies the use of strontium as an adequate imitator of calcium behavior in bone. This has been shown previously by Eisenberg (5).

Studies on the distribution of strontium in the whole tibia demonstrate that most of the tracer was in bone and only a small portion was in marrow.

The third set of preliminary experiments were important in eliminating the vasodilator effect of parathyroid hormone as the mechanism for its effect on the ion exchange in bone (2).

The most important finding in this study was the difference between dogs made hypocalcemic (parathyroidectomized) and dogs made hypercalcemic by bovine parathyroid extract. In the dogs with tibias perfused with their own blood, 3-min extraction was less rapid and initial washout of the calcium tracer, ^{85}Sr , was more rapid in the hypercalcemic dogs than in the hypocalcemic dogs. Because the capillary PS_c in these two groups was similar, obviously there was reduced retention in the extra-vascular volume of distribution of the hypercalcemic dogs. The data suggest that in dogs with induced hyperparathyroidism (hypercalcemia) the volume of distribution of the calcium tracer, ^{85}Sr , is decreased either by a reduction in the number of available binding sites in the matrix or by stimulation of an active pumping mechanism that returns the tracer from a controlled region of sequestration to the perivascular region. Alternatively, there could be a reduction of the rate of pumping into a site of sequestration, allowing an increase in the net efflux. Either would allow more return to the effluent blood.

These findings are compatible with the observation of Talmage et al. (23), who showed that the plasma clearance of ^{45}Ca by thyroparathyroidectomized rats was less rapid after parathyroid hormone injections at 1 h.

A further possibility is that elevation of serum phosphate might in some fashion affect the transcapillary movement of the strontium ion, perhaps by binding this calcium tracer in the interstitial fluid space. However, the serum phosphate of both dogs made hypocalcemic by parathyroidectomy or hypercalcemic by short-term injections of parathyroid extract are substantially the same (Table 3).

Assuming that isotopic strontium is a valid tracer in bone for calcium, an alternative to a theory of cellular control would be that changes in blood flow control calcium efflux from bone; that is, in the hyperparathyroid state flow is increased and in the hypoparathyroid state flow is decreased. If flow dominated the exchange, the plots of residue versus volume of effluent (Fig. 3B) should all be superimposed; however, they are not, and this indicates that permeation (or extravascular diffusion) is important in limiting washout. This in turn implies that calcium binding to surfaces or more active extrusion is a more likely mechanism.

It may be argued that these findings are contrary to those of Eisenberg and Gordan (6) and Massin et al. (12), in which expanded pools and turnover of calcium tracer (stable strontium) or ^{45}Ca were observed in patients with long-term hyperparathyroidism or decreased pool size with hypoparathyroidism. However, our dogs had received parathyroid extract for a short period of time only. Longer periods of treatment with parathyroid extract cause bone remodeling, oxygen consumption, and clearance of ^{85}Sr to increase (22)—findings from this laboratory that are not contradictory to the observations of Eisenberg and Gordan (6) or Massin et al. (12). Kinetic analysis by Reeve (19) suggested that chronic parathyroid hormone administration decreased the transit time for calcium in the exchangeable pools, an

observation that is explained by our observations of reduced strontium retention in the extravascular volume of distribution of the hypercalcemic dogs.

The data presented show that parathyroid hormone is able to affect the rapid component of calcium exchange in bone. This is not achieved by influencing the capillary wall but by affecting the processes of absorption or transport beyond the capillary. The effects of parathyroid hormone on the retention of strontium and calcium in the first few minutes is dramatic. The hormone has the effect of reducing the immediate volume of distribution of tracer ions; this occurs, we suggest, by the release of accessible calcium ions into the circulation, and this reduces the readily exchangeable calcium ion pool in the skeleton. The shift in the ionic balance toward the interstitial fluid would have the effect of raising the plasma level of calcium.

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References

1. Bosch WJ. Plasma ^{45}Ca clearance by the tibia in the immature dog. *Am J Physiol.* 1969; 216:1150–1157. [PubMed: 5797296]
2. Charbon GA, Hulstaert PF. Augmentation of arterial hepatic and renal flow by extracted and synthetic parathyroid hormone. *Endocrinology.* 1974; 95:621–626. [PubMed: 4850699]
3. Cofield RH, Bassingthwaite JB, Kelly PJ. Strontium-85 extraction during transcapillary passage in tibial bone. *J Appl Physiol.* 1975; 39:596–602. [PubMed: 1104562]
4. Davies DR, Bassingthwaite JB, Kelly PJ. Transcapillary exchange of strontium and sucrose in canine tibia. *J Appl Physiol.* 1976; 40:17–22. [PubMed: 1248978]
5. Eisenberg E. Effect of intravenous phosphate on serum strontium and calcium. *N Engl J Med.* 1970; 282:889–892. [PubMed: 5434935]
6. Eisenberg E, Gordan GS. Skeletal dynamics in man measured by nonradioactive strontium. *J Clin Invest.* 1961; 40:1809–1825. [PubMed: 13889673]
7. Hughes SPF, Davies DR, Bassingthwaite JB, Knox FG, Kelly PJ. Bone extraction and blood clearance of diphosphonate in the dog. *Am J Physiol.* 232Heart Circ Physiol. 1977; 1:H341–H347.
8. Kelly PJ, Bassingthwaite JB. Studies on bone ion exchanges using multiple-tracer indicator-dilution techniques. *Federation Proc.* 1977; 36:2634–2639. [PubMed: 913623]
9. Kelly PJ, Yipintsoi T, Bassingthwaite JB. Blood flow in canine tibial diaphysis estimated by iodoantipyrine- ^{125}I washout. *J Appl Physiol.* 1971; 31:38–47. [PubMed: 5556961]
10. Kerrick WGL, Hoar PE, Malencik DA, Stamps L, Fischer EH. Characterization of Ca^{2+} - and Sr^{2+} -activated tension in functionally skinned chicken fibers of normal and dystrophic skeletal and normal cardiac muscle. *Pfluegers Arch.* 1979; 381:53–62. [PubMed: 573454]
11. Lemon GJ, Davies DR, Hughes SPF, Bassingthwaite JB, Kelly PJ. Transcapillary exchange and retention of fluoride, strontium, EDTA, sucrose, and antipyrine in bone. *Calcif Tissue Int.* 1980; 31:173–181. [PubMed: 6770980]
12. Massin JP, Vallee G, Savoie JC. Compartmental analysis of calcium kinetics in man: application of a four-compartmental model. *Metabolism.* 1974; 23:399–415. [PubMed: 4825298]
13. Morris MA, Kelly PJ. Use of tracer microspheres to measure bone blood flow in conscious dogs. *Calcif Tissue Int.* 1980; 32:69–76. [PubMed: 6775781]

14. Neuman WF. The milieu interieur of bone: Claude Bernard revisited. *Federation Proc.* 1969; 28:1846–1850. [PubMed: 5353625]
15. Neuman WF, Neuman MW, Myers CR. Blood: bone disequilibrium. III. Linkage between cell energetics and Ca fluxes. *Am J Physiol. 236Cell Physiol.* 1979; 5:C244–C248.
16. Nordin BEC, Marshall DH, Peacock M, Robertson WG. Plasma calcium homeostasis. *Excerpta Med Int Congr Ser.* 1974; 346:239–253.
17. Paradis GR, Bassingthwaighe JB, Kelly PJ. Inhibition of transport of ^{47}Ca and ^{85}Sr by lanthanum in canine cortical bone. *J Appl Physiol.* 1974; 36:221–225. [PubMed: 4811382]
18. Parsons JA, Reit B. Chronic response of dogs to parathyroid hormone infusion. *Nature London.* 1974; 250:254–257. [PubMed: 4851028]
19. Reeve J. The turnover time of calcium in the exchangeable pools of bone in man and the long-term effect of a parathyroid hormone fragment. *Clin Endocrinol Oxford.* 1978; 8:445–455.
20. Reynolds JJ. Bone turnover, vitamin D and plasma calcium homeostasis. *Excerpta Med Int Congr Ser.* 1974; 346:254–261.
21. Schatzmann HJ, Vincenzi FF. Calcium movements across the membrane of human red cells. *J Physiol London.* 1969; 201:369–395. [PubMed: 4238381]
22. Sim FH, Kelly PJ. Relationship of bone remodeling, oxygen consumption, and blood flow in bone. *J Bone Jt Surg Am Vol.* 1970; 52:1377–1389.
23. Talmage RV, Doppelt SH, Fondren FB. An interpretation of acute changes in plasma ^{45}Ca following parathyroid hormone administration to thyroparathyroidectomized rats. *Calcif Tissue Res.* 1976; 22:117–128. [PubMed: 1000348]
24. Talmage, RV.; Meyer, RA, Jr. *Handbook of Physiology Endocrinology Parathyroid Gland.* Vol. vii. Washington, DC: Am. Physiol. Soc; 1976. Physiological role of parathyroid hormone; p. 343-351.

FIG. 1. Normalized outflow dilution curves from dog tibia after injection of albumin, strontium, and calcium into tibial nutrient artery. Curves for strontium and calcium for $h(t)$, $E(t)$, and $E_{\text{net}}(t)$ are all essentially similar. Extraction curve does not start at zero because first calculated point is not zero.

FIG. 2. Effect of changing solute flow on rate of ^{85}Sr washout from tibia is minimal. The fractional escape rate (FER) is calculated by Eq. 5, over 5-min intervals. In the experiment, no apparent change in FER occurred with changes in flow.

FIG. 3.

Effect of parathyroid state on washout of ^{85}Sr from dog tibia. *A*: residue functions recorded from hypoparathyroid and hyperparathyroid dogs; *B*: same curves plotted by scaling *abscissa* of each curve by flow. $F_s \cdot t$ is volume of perfusate, $V(t)$ (ml/g of tibial bone), passing through nutrient artery since tracer injection.

FIG. 4.

Strontium retention in tibial bone is greater than normal in hypoparathyroid dogs and less in hyperparathyroid dogs. Data are from Table 3. Bars are ± 1 SD and rectangles ± 1 SE. Retention is residue function, $R(t)$, after a bolus injection into tibial nutrient artery following washout (A) with 0.8 ml plasma per gram of bone and (B) with 2.0 ml plasma per gram of bone.

TABLE 1Apparent volume of distribution for ^{47}Ca and ^{85}Sr in dogs

Time after Injection, min	<i>Dog G 183</i>		<i>Dog G 214</i>	
	^{47}Ca	^{85}Sr	^{47}Ca	^{85}Sr
15	0.27	0.26	0.09	0.08
45	0.59	0.60	0.29	0.27
90	0.78	0.77	0.15	0.15
150	1.07	1.02	0.38	0.37

Values are the ratio, cpm per ml bone/cpm per ml blood.

TABLE 2

Distribution of tracer

(1) Dog	(2) Parathyroid State	(3) Background	(4) Final Counts over Expt Tibia *	(5) Opposite Tibia *	(6) Isolated Tibia *	(7) Isolated Marrow *	(8) Isolated Cancellous Bone *
G 867	Hypo	35	1,405	35	1,265	45	85
G 537	Hypo	30	1,920	70	1,865	100	570
G 862	Control	30	890	90	870	70	190
G 864	Hyper	30	1,470	40	1,370	40	160

Values are counts per minute (cpm). Numbers in parentheses are the numbers of the columns.

* Background subtracted.

Fractional escape rate or η (t)

TABLE 3

Dog	Plasma Flow, ml · 100 g ⁻¹ · min ⁻¹	Day of Study	E _{max} Sr	E _{net} (3 min)	PS _e , ml · 100 g ⁻¹ · min ⁻¹	[Ca], mg/dl	[PO ₄], mg/dl	R(t), V = 0.8 ml/g	FER × 10 ⁻³ /min				R(t), V = 2.0 ml/g
									0–1 min	1–30 min	30–60 min	60–180 min	
Control*													
G 783	0.65		0.68	0.57	0.74	9.9	4.9	0.42	143.2	18.2	5.7	2.9	0.31
G 705	0.92		0.61	0.41	0.87	10.0	3.4	0.18	393.3	30.9	6.4	2.4	
G 662	1.46		0.35	0.32	0.63	10.2	4.2	0.18	418.2	34.6	13.7	4.3	0.11
G 862	1.03		0.52	0.51	0.76	10.1	5.1	0.34	135.4	27.0	11.0	4.0	0.23
G 816	1.07		0.75	0.41	1.25	10.4	5.4	0.23	508.4	25.0	6.5		
H 80	1.03		0.66	0.53	1.11	10.2	3.5	0.15	479.0	42.2	11.3		0.08
Mean ± SD	1.02 ± 0.26		0.60 ± 0.14	0.46 ± 0.09	0.89 ± 0.24	10.13 ± 0.18	4.41 ± 0.85	0.23 ± 0.11	346.3 ± 165.5	29.6 ± 8.3	9.1 ± 3.3	3.4 ± 0.9	0.18 ± 0.107
Hypoparathyroid													
G 704	0.92	3	0.73	0.63	1.20	8.8	6.6	0.37	169.3	21.0	5.8	2.4	0.29
G 815	1.04	3	0.75	0.51	1.44	7.2	5.9	0.25	257.3	30.5	7.9	3.1	0.18
G 753	1.55	3	0.69	0.54	1.82	6.5	6.0	0.27	356.6	27.5	10.5	4.5	0.19
G 537	1.19	3	0.63	0.54	1.18	7.5	6.9	0.40	317.4	16.8	4.8	1.9	0.33
G 867	1.22	4	0.68	0.50	1.39	8.4	6.0	0.27	302.3	32.3	7.8	2.7	0.18
H 22	1.07	7	0.50	0.43	0.74	6.3	5.0	0.35	239.1	17.6	7.4	3.2	0.27
Mean ± SD	1.17 ± 0.22		0.66 ± 0.09	0.53 ± 0.07 [†]	1.30 ± 0.36	7.45 ± 1.00	6.06 ± 0.66	0.32 ± 0.06 [†]	273.7 ± 66.3 [†]	24.3 ± 6.7	7.4 ± 2.0	3.0 ± 0.9	0.24 ± 0.065
Hyperparathyroid													
G 745	0.83	5	0.53	0.36	0.63	14.7	5.5	0.19	421.5	34.0	10.9	2.8	0.13
G 864	1.44	5	0.53	0.29	1.09	13.1	5.5	0.21	498.2	31.7	6.2	3.1	0.15
G 584	1.30	3	0.66	0.49	1.75	14.7	6.0	0.18	354.9	37.7	7.6	3.2	0.14
G 543	1.35	5	0.68	0.39	1.54	13.9	6.9	0.07	315.0	64.8	12.6	3.7	0.05
H 184	0.89	3	0.53	0.41	0.67	13.5	5.3	0.22	454.5	27.4	8.2	1.6	0.16
Mean ± SD	1.16 ± 0.28		0.59 ± 0.08	0.39 ± 0.07 [†]	1.14 ± 0.50	13.98 ± 0.72	5.84 ± 0.65	0.17 ± 0.06 [†]	408.8 ± 74.1 [†]	39.1 ± 14.8	9.1 ± 2.6	3.1 ± 0.4	0.13 ± 0.044

Day of study refers to day after removal of parathyroid glands or start of parathyroid extract. $E_{\max}$ Sr, maximum extraction of strontium; E_{net} , net extraction; PS_e , capillary permeability-surface products; $R(t)$, residue function; FER, fractional escape rate.

* Previously reported in part (Ref. 11).

[†] $P < 0.01$, hypoparathyroid compared with hyperparathyroid.