

The Effect of Calcium Infusions, Parathyroid Hormone, and Vitamin D on Renal Clearance of Calcium.* (28042)

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The fundamental function of the parathyroid glands is the homeostatic regulation of the ionized calcium in body fluids. It has been conclusively demonstrated that parathyroid hormone causes mobilization of calcium from the skeleton and an increase in renal clearance of phosphorus. Recent studies strongly suggest that this hormone enhances both the intestinal absorption(1,2) and the renal tubular reabsorption(3,4,5,6) of calcium.

Earlier reports from this laboratory(4,5) demonstrated that there was a direct relationship between the filtered load and the renal clearance of calcium. For any given filtered load of this ion an increase in the circulating level of parathyroid hormone lowers renal clearance of calcium expressed as per cent of the filtered load excreted. These studies suggested that the increase in renal clearance of calcium in the hypoparathyroid human receiving Vit. D(7) was not due to a direct effect of the vitamin on the renal tubule but to the increase in serum calcium and filtered load which followed administration of the vitamin. The present experiments were undertaken to compare the effects of calcium infusions, parathyroid extract, and Vit. D₂ on renal clearance of calcium over a comparable range of serum calcium or filtered load.

Methods. The studies were performed in 2 post-surgical-hypoparathyroid females with normal renal function. Patient R.R., age 34, was hypoparathyroid 13 years and had never received supplemental Vit. D. Patient B.W., age 29, was hypoparathyroid 10 years and discontinued the daily dose of 4 cc of AT-10 four weeks prior to these studies.

The patients were placed on a constant calcium and phosphorus intake and experiments were carried out in the following order: Approximately one-half hour clearance periods were obtained during an infusion of 1.7 g of calcium ion as calcium chloride over

a 6-hour period. Three days later, after the serum calcium returned to the previous baseline levels, they were given progressively increasing doses of parathyroid extract (Parathyroid, Eli Lilly) up to a maximum of 350 units every 8 hours intramuscularly. Approximately one-half hour clearance periods were obtained twice daily over a range of serum calcium comparable to those achieved during the calcium infusion. After the parathyroid extract was discontinued and serum calcium once again returned to the baseline levels, the patients were placed on 1.5 g of oral calcium daily, and progressively increasing doses of Vit. D₂ up to 150,000 units, for patient R.R. and 400,000 units for patient B.W. Clearances were obtained as described above during administration of parathyroid extract.

Calcium clearance was determined by dividing quantity of calcium excreted per minute in the urine by the serum diffusible calcium(8). The latter figure was determined separately for the calcium infusion, the parathyroid extract and the Vit. D administration by calculating the mean diffusible serum calcium from blood samples obtained during each experimental condition. The calcium clearance ratio was calculated by dividing the observed calcium clearance by the simultaneously measured endogenous creatinine

clearance $\left(\frac{C_{Ca}}{C_{Cr}} \times 100 \right)$, or per cent of the

filtered load of calcium excreted. This calculation standardized the clearance for variations in size of the individuals and corrected for changes in the filtered load not due to changes in plasma concentration of diffusible calcium.

The methods for carrying out renal clearances and chemical analyses have been described(5).

Results. The results are illustrated in Fig. 1 and 2. In both subjects during the cal-

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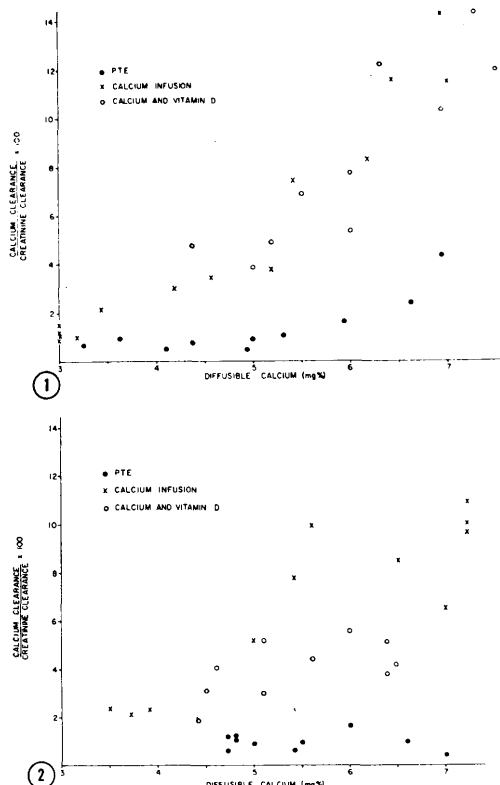


FIG. 1. Patient R.R. Effect of a calcium infusion, parathyroid extract (Parathyroid, Eli Lilly) and Vit. D₂ administration on serum diffusible calcium and calcium clearance ratio.

FIG. 2. Patient B.W. Effect of a calcium infusion, parathyroid extract (Parathyroid, Eli Lilly) and Vit. D₂ administration on serum diffusible calcium and calcium clearance ratio.

cium infusion and under the influence of Vit. D, as the filtered load of calcium increased the calcium clearance ratio progressively increased. For any given serum calcium (filtered load) the clearance ratios obtained during administration of parathyroid extract were significantly lower. In subject B.W. (Fig. 2), despite the increase in filtered load the parathyroid hormone effect was so pronounced that a significant increase in the calcium clearance ratio did not occur.

Discussion. These experiments clearly confirm the ability of parathyroid hormone to prevent or markedly diminish the rise in renal clearance ratio of calcium that occurs with increasing filtered loads. This is in contrast to the effects of a calcium infusion or Vit. D₂ (Fig. 1 and 2). The clearance ratios during the Vit. D administration were equal

to or slightly lower than those obtained at comparable blood levels during the calcium infusion. Recently Gordon *et al.* (9) stated that parathyroid extract did not decrease the renal clearance of calcium because calcium clearances observed in hyperparathyroid patients with *elevated* serum calciums were not lower than in normal subjects at *normal* calcium levels. These authors disregarded the specific effect of the filtered load of calcium on its renal clearance. They actually demonstrated that despite elevated serum calcium values, hyperparathyroid patients had calcium clearances only slightly higher than the normal subjects. In striking contrast were the markedly elevated clearances in normal subjects who had their serum calcium raised to approximately comparable levels by a calcium infusion.

Summary. The comparative effects of calcium infusions, parathyroid extract, and Vit. D on renal clearance of diffusible calcium in 2 hypoparathyroid subjects were studied. For any given filtered load of calcium, parathyroid extract significantly decreased the renal clearance ratio of this ion. A direct effect of Vit. D on the calcium clearance ratio could not be demonstrated.

ADDENDUM. The authors have had the opportunity to repeat the same study in one additional subject with idiopathic hypoparathyroidism. The results are similar to those found in the two individuals described above. For example, at a serum diffusible calcium of about 5.9 mg%, the calcium clearance ratios during parathyroid extract and vitamin D administration and calcium infusion were 0.8, 7.5 and 6.3 respectively.

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Hemagglutination: A Rapid Method for Differentiating *Vibrio cholerae* and El Tor Vibrios. (28043)

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At present, El Tor vibrios are differentiated from *Vibrio cholerae* in the laboratory primarily by their ability to hemolyze goat or sheep erythrocytes(1). However, the results of hemolysis tests have been extremely variable depending on the modification of the test used and the nature of the strains being tested. For example, strains from the 1961 epidemic in the Philippines and Hong Kong were non-hemolytic at 72 hours of incubation in "the original Greig test"(2) used by Goodner and his co-workers (personal communication) although other investigators have found these strains to be hemolytic in modifications of the test using 24 hour cultures (3,4). Some reason for this discrepancy was suggested by the work of de Moor(5) who reported the disappearance of hemolysin from broth cultures of the Celebes El Tor vibrios between the second and fourth days of incubation. This observation was repeated recently by Feeley and Pittman(6) who found the Philippine and Hong Kong cultures to be hemolytic or non-hemolytic depending on the time the broth cultures were tested. This characteristic varied somewhat with the different media used. de Moor(5) also had noted that the hemolytic activity of the Celebes vibrios increased during laboratory maintenance. Recently, some strains from the 1962 resurgence of cholera El Tor in the Philippines were found to be non-hemolytic when first isolated, but the strains revealed their hemolytic capacity in subsequent tests (C. Z. Gomez, personal communication).

The interpretation placed on the variable

results of this single biological test and the implications thereof have had far reaching consequences since, in 1957, the Committee on International Quarantine stated its opinion that El Tor infection should not be included in the term "cholera"(7). As a result of the widespread epidemic of cholera caused by El Tor vibrios in the Far East in 1961, it is now acknowledged that "cholera, under the definition of quarantinable diseases in Article 1 of the International Sanitary Regulations, should include cholera due to the El Tor vibrio"(7).

Although it is now recognized that epidemic cholera may be caused by either *V. cholerae* or El Tor vibrios, for epidemiologic and other considerations it is still important to be able to differentiate between the 2 groups of organisms. Since other methods which have been devised for that purpose have proved unsatisfactory(8), a new method more reliable than the hemolysis test is still needed. Consistent results have been obtained by use of the group IV cholera bacteriophage(9) which is lytic for *V. cholerae* but does not lyse El Tor vibrios(8,10). However, the requisite phage and facilities for its propagation and maintenance are not universally available. Further, the results of this test are not available until overnight incubation following the primary isolation.

In preliminary studies, freshly isolated El Tor vibrios were observed to cause hemagglutination of chicken red blood cells in slide tests while the laboratory strains of *V. cholerae* tested were unable to do so. Subsequent