Published in final edited form as:
Arch Biochem Biophys. 2012 July 1; 523(1): 77–86. doi:10.1016/j.abb.2012.03.003.

Vitamin D and the Kidney

Rajiv Kumar[1,2,3], **Peter J. Tebben**[2,3], and **James R. Thompson**[4]

[1]Division of Nephrology, Mayo Clinic and Foundation, Rochester, Minnesota, 55905

[2]Division of Endocrinology, Diabetes, Metabolism, and Nutrition, Mayo Clinic and Foundation, Rochester, Minnesota, 55905

[3]Department of Medicine, Mayo Clinic and Foundation, Rochester, Minnesota, 55905

[4]Department of Biochemistry and Molecular Biology, [4]Physiology and Biomedical Engineering, Mayo Clinic and Foundation, Rochester, Minnesota, 55905

Abstract

The kidney is essential for the maintenance of normal calcium and phosphorus homeostasis. Calcium and inorganic phosphorus are filtered at the glomerulus, and are reabsorbed from tubular segments by transporters and channels which are regulated by 1α,25-dihydroxyvitamin (1α,25(OH)$_2$D) and parathyroid hormone (PTH). The kidney is the major site of the synthesis of 1α,25(OH)$_2$D under physiologic conditions, and is one of the sites of 24,25-dihydroxyvitamin D (24,25(OH)$_2$D) synthesis. The activity of the 25(OH)D-1α-hydroxylase, the mixed function oxidase responsible for the synthesis of 1α,25(OH)$_2$D, is regulated by PTH, 1α,25(OH)$_2$D, fibroblast growth factor 23 (FGF23), inorganic phosphorus and other growth factors. Additionally, the vitamin D receptor which binds to, and mediates the activity of 1α,25(OH)$_2$D, is widely distributed in the kidney. Thus, the kidney by regulating multiple transport and synthetic processes is indispensible in the maintenance of mineral homeostasis in physiological states.

Keywords

1α,25-Dihydroxyvitamin D; calcium; phosphorus; tubular reabsorption; epithelial calcium channel; plasma membrane calcium pump; calbindin; hydroxylase

1. INTRODUCTION

The kidney has a unique function in mineral homeostasis. Both calcium and phosphorus are filtered, reabsorbed and excreted in the urine to a varying degree, generally in amounts that reflect the endogenous requirements of the two substances. The vitamin D-endocrine system plays a key role in controlling the renal excretion of both calcium and phosphorus. The reabsorption of calcium in the kidney is controlled by several factors, including 1α,25-dihydroxyvitamin D (1α,25(OH)$_2$D) [1–5]. The kidney is the major site of synthesis of 1α,25(OH)$_2$D, the active, hormonal form of vitamin D [6–8]. The renal 25-hydroxyvitamin D$_3$ 1α–hydroxylase (1α–hydroxylase) and 25-hydroxyvitamin D-24-hydroxylase (24-hydroxylase), and other 1α,25-dihydroxyvitamin D$_3$ and vitamin D analog metabolizing

Corresponding author: Rajiv Kumar, MS 1-120, Mayo Clinic, 200 First street SW, Rochester, MN 55905, Telephone: 507-284-0020, Fax: 507-538-3954, rkumar@mayo.edu.

enzymes are expressed in kidney tissue [9–11]. The kidney expresses the vitamin D receptor (VDR) [11, 12]. The kidney expresses several 1α,25$(OH)_2$D-dependent proteins that are important in calcium reabsorption e.g. the plasma membrane calcium pump (PMCa) [13–15], the epithelial calcium channel (ECaC) [16], the sodium calcium exchanger [17], and the calbindins [13–15]. Finally, the kidney has an important role in the control of plasma phosphate, which following filtration in the glomerulus is reabsorbed in nephron segments at a rate influenced by many of the same hormones and factors involved in calcium regulation [18–23].

To understand how vitamin D influences the efficiency of calcium and phosphorus reabsorption, a brief review of normal calcium and phosphorus handling by the kidney follows.

2. CALCIUM AND PHOSPHORUS FILTRATION AND REABSORPTION IN THE KIDNEY

1.1 Calcium handling by the kidney

On account of protein-binding, slightly more than half of total plasma calcium (plasma concentration 10 mg/dL or 2.5 mM) is filtered at the glomerulus [24]. The concentration of calcium in the glomerular filtrate is the same as that of plasma ultra-filterable calcium [25–27]. Assuming a glomerular filtration rate of 140 L per day and an ultrafiltrable calcium of 5.5 mg/dL, about 8000 mg of calcium are filtered by the kidney in 24 hours; 98% of the filtered load of calcium is reabsorbed, resulting in an excreted calcium of about 150–200 mg/24 h [1, 24]. Fifty to sixty percent of the filtered load of calcium is reabsorbed in the proximal tubule [2, 3, 28] by sodium-dependent, para-cellular mechanisms. Inhibition of sodium-potassium ATPase activity by ouabain reduces the amount of calcium reabsorbed in the proximal tubule, as does the substitution of sodium with lithium [29]. A reduction of tubular sodium reabsorption by volume expansion inhibits proximal tubule calcium reabsorption, while an increase in sodium reabsorption associated with volume contraction, enhances proximal tubule calcium reabsorption [2, 29]. Importantly, calcium reabsorption in the proximal tubule is not altered by thiazide diuretics, hormones such as PTH or 1α, 25$(OH)_2D_3$, or by hydrogen ions [2, 3, 28, 29]. Vitamin D-dependent proteins that play a role in trans-cellular calcium transport, such as the calbindins, ECaCs, and the PMCa pump are either not expressed in the proximal tubule, or are expressed in low amounts when compared with the amounts expressed in the distal tubule. Minimal amounts of calcium are absorbed in the descending loop and the thin ascending limb of the loop of Henle. About 20% of the filtered load of calcium is reabsorbed in the thick ascending limb of the loop of Henle; another 10–15% of filtered calcium is reabsorbed in the distal tubule with the remaining 5% being reabsorbed in the collecting duct [2, 3, 28, 29]. The movement of calcium in the distal nephron is energy dependent and occurs against a concentration gradient; furthermore, the tubular lumen is electro-negative and becomes progressively more so towards the end of the distal tubule [30, 31]. In the distal nephron, calcium reabsorption can be dissociated from sodium reabsorption by thiazide diuretics which inhibit sodium reabsorption but enhance calcium reabsorption [30, 31]. In contrast to the proximal tubule, where hydrogen ion have no effect on calcium reabsorption, in the distal nephron hydrogen ions inhibit calcium reabsorption.

1.2 Phosphate handling by the kidney

Inorganic phosphate in the serum is freely filtered at the glomerulus [18–20]. About 80% of filtered phosphorus is reabsorbed in the proximal tubule [18]. The amount of phosphorus reabsorbed in the proximal tubule is greatest in the first half of the proximal tubule with some further phosphorus reabsorption occurring in the pars recta [18]. Little or no

phosphorus reabsorption occurs in the loop of Henle or the distal tubule. The reabsorption of phosphate is sodium-dependent and is mediated by sodium-phosphate co-transporters (Na-Pi IIa,SLC34A1; Na-Pi IIc, SLC34A3, and Pit-2, SLC20A2) [32, 33]. Na-Pi IIa activity is increased by a low phosphate diet and decreased by PTH.[34–37] The recently described phosphatonins, fibroblast growth factor-23 (FGF-23), MEPE and secreted frizzled related protein-4 (sFRP-4), inhibit sodium dependent phosphate transport [23, 38]. In opossum kidney (OK) cells, NaPi II is internalized from the cell membrane in response to FGF-23 and sFRP-4, similar to the effects of PTH [39, 40]. Additional factors involved in phosphorus reabsorption are noted in Table 1.

3. ROLE OF THE KIDNEY IN THE METABOLISM OF 25OH D

3.1. The 25-hydroxyvitamin D_3-1α-hydroxylase and the synthesis of 1α,25$(OH)_2D_3$

25-hydroxyvitamin D_3-1α-hydroxylase is a multi-component, cytochrome P-450 containing enzyme present in mitochondria of renal proximal tubular cells which transfers electrons from NADPH to the cytochrome P450, Cyp27B1 [7, 41–56]. The latter, using molecular oxygen, converts 25-hydroxyvitamin D_3 (25$(OH)D_3$) to 1α,25$(OH)_2D_3$ and water. Nephrectomy greatly decreases circulating 1α,25$(OH)_2D_3$ concentration *in vivo* except during pregnancy, granuloma-forming diseases, and lymphomas associated with the ectopic production of 1α,25$(OH)_2D_3$ [57–60]. While the kidney is the major site of 1α,25$(OH)_2D_3$ production, 25$(OH)D_3$-1α-hydroxylase activity has been found *in vitro* in several other cell types [49, 61–72]. *In vitro*, chick renal epithelial cells in culture, mammalian nephron segments and homogenates derived from avian and mammalian (mostly rodent) renal cells metabolize 25$(OH)D_3$ to 1α,25$(OH)_2D_3$ [73–77]. Proximal and distal tubular segments synthesize 1α,25$(OH)_2D_3$ [78, 79]. Zehnder *et al* have demonstrated the presence of 1α-hydroxylase mRNA and protein in the distal convoluted tubule, cortical collecting duct, thick ascending limb of the loop of Henle, and Bowman's capsule [79]. Recent experiments in which the 25$(OH)D_3$-1α-hydroxylase cytochrome P450 gene (*Cyp27B1*) was deleted in mice, point to the central role of this enzyme in vitamin D metabolism [80]. Table 2 summarizes some of the key factors known to regulate the activity of this enzyme *in vivo* and *in vitro*. The major regulators appear to be PTH, inorganic phosphorus and 1α, 25$(OH)_2D_3$ itself. Fibroblast growth factor 23 (FGF 23), and its binding protein Klotho that is produced mainly in the distal tubule, and which is essential in mediating FGF 23 activity, inhibit 25$(OH)D_3$-1α-hydroxylase activity and regulate the production of 1α,25$(OH)_2D_3$ [81–93]. Indeed, FGF 23 may mediate the effects of changing dietary phosphorus on 1α, 25$(OH)_2D_3$ production. Regulators such as 1α,25$(OH)_2D_3$ which alter the expression of the *Cyp27B1* gene have reciprocal effects on the expression of the *Cyp24A1* gene [94] that are mediated via vitamin D-response elements in the promoters of the respective genes [95, 96]. In cell culture, the extra-renal 25$(OH)D_3$-1α-hydroxylase is regulated by nitric oxide and by activation of toll-like receptors [66–72].

3.2 Modeling Cyp27B1 and Computational Docking Studies

Given its critical role in the synthesis of 1α,25$(OH)_2D_3$, it is important to determine how the Cyp27B1 cytochrome P450 binds 25$(OH)D_3$. Two energy-minimized homology models for Cyp27B1 were created by manual rebuilding from Cyp24A1 (open-cavity form, PDB ID 3K9V) [97] and Cyp2R1 (closed, PDB 3C6G [98] crystal structures, guided by I-TASSER and PHYRE2 modeling [99–101]. Figure 1A shows putative Cyp27B1 residues of the substrate binding cavity. The "yellow-carbon" Cyp27B1 residues differ from Cyp24A1's homologous amino acids.

Cyp27B1 models, the Cyp24A1 and Cyp2R1 structures and all ligand atom coordinates used were parameterized by Autodock Tools (http://autodock.scripps.edu/resources/adt), with

heme Fe charge explored from +0.2 to 1.0, both with & without a partially-charged oxygen bound to the Fe atom. Control Autodock Vina computations [102], chosen for high binding mode accuracy, correctly oriented & positioned Chaps within 0.59 Å RMSD of that in the Cyp24A1 crystal (chain A) [97] and 25(OH)D_3 within 0.99 Å RMSD of the 25(OH)D_3 observed in the Cyp2R1 crystal [103]. The search box covered all molecular extents, with num_mode and exhaustiveness at 500. Predictions concerning the plausibility of the binding modes, and signal-noise level of calculations, were based on computed free energy of binding, clustering of binding modes, proximity to key residues identified in literature, and distances between the known hydroxylated carbon atoms and heme Fe.

Irrespective of whether the Cyp27B1 model was derived from Cyp24A1 (open cavity) or Cyp2R1 (closed) structures, the 25(OH)D_3 docking modes deemed most plausible were not among those top ranked by Autodock Vina's binding-energy scoring function. Compare the highest-rank 25(OH)D_3 pose, −9.2 kcal/mol, in Figure 1B to the pose in Figure 1C (9^{th} in rank) that we find somewhat promising at −8.1 kcal/mol. Allowing cavity residues of Cyp27B1 to be flexible during computations did not improve the likely validity of top-scored poses. Interestingly, we found top binding modes for the product of Cyp27B1 metabolism, 1α,25$(OH)_2D_3$, more believable as the molecules were oriented with A-rings closest to Cyp27B1's heme Fe, though not in positions able to explain catalysis (best score −9.5 kcal/mol, not shown). Our top poses of 1α,25$(OH)_2D_3$ docked to the Cyp24A1 crystal structure, with best at −9.0 kcal/mol in Figure 1D, are acceptable predictions that appear nearly equivalent to a pose published by Rhieu et al. (Fig10 in [103]). Template-bias prevented our two homology models of Cyp27B1 from being useful prediction tools for understanding ligand-binding. Crystal structures of Cyp27B1 will be required to definitively establish its structure.

3.3 The 25(OH)D_3-24-hydroxylase and the synthesis of 24,25-dihydroxyvitamin D_3

The 25(OH)D_3-24-hydroxylase enzyme is a multi-component cytochrome P450 enzyme expressed in the kidney as well as many other tissues.[104–125]. The renal 24-hydroxylase is regulated by calcium and phosphorus such that elevated or normal calcium levels induce the 24-hydroxylase activity, whereas, low calcium levels inhibit it [116–118, 126]. Similarly, elevated serum phosphorus concentrations increase 24-hydroxylase activity and low phosphorus diets decrease the enzyme activity [127]. 24-hydroxylase enzyme activity and mRNA expression in the kidney is up-regulated by 1α,25$(OH)_2D_3$ [128–134]. The effect of 1α,25$(OH)_2D_3$ is blunted *in vitro* and *in vivo* by parathyroid hormone [128–131]. We used antibodies against the 24-hydroxylase cytochrome P-450 to examine the distribution of the enzyme in the human kidney, and found high concentrations of the cytochrome P-450 in distal tubular cells and lower amounts in the proximal tubule [11]. 24-Hydroxylase activity, however, is expressed primarily in the proximal tubule but is also present in more distal segments [135, 136]. It is responsible for the conversion of 25(OH)D_3 and 1α,25$(OH)_2D_3$ to 24,25-dihydroxyvitamin D_3 (24,25$(OH)_2D_3$) and 1α,24,25-trihydroxyvitamin D_3, respectively. There is conflicting evidence as to whether 25(OH)D_3 or 1α,25$(OH)_2D_3$ is the preferred substrate for 24-hydroxylase [137]. Some have reported 1α,25$(OH)_2D_3$ is the preferred substrate with a K_m approximately 10-fold lower than that for 25(OH)D_3 [138, 139], while others have found K_m values substantially lower for 25(OH)D_3 [140]. It has been suggested that 24,25$(OH)_2D_3$ has unique properties and actions in cartilage and bone [124, 125, 141, 142], but others have not confirmed these observations [143–149].

3.4 The Structure of the 25(OH)D_3-Cyp24A1

Recently, the structure of the Cyp24A1 in association with CHAPS was reported [97]. The crystal structure shows an open cleft leading to the active-site heme prosthetic group. The

entrance to the cleft is flanked by conserved hydrophobic residues on helices A' and G'. The determinants of adrenodoxin recognition are conserved residues from helices K, K", and L.

3.5 Other vitamin D metabolizing enzymes present in the kidney

The kidney is also capable of transforming $25(OH)D_3$ to several other compounds listed in Table 3 [150–170]. The specific physiological roles of these various metabolites are not known with certainty. Several polar metabolites of $1\alpha,25(OH)_2D_3$ are formed in the liver, including calcitroic acid and the glucuronide and sulfate conjugates of the hormone; these and small amounts of unchanged dihydroxylated and tri-hydroxylated metabolites of vitamin D are excreted in the urine [171–185]. Many of the transformations that occur with $25(OH)D_3$ also occur in the case of $1\alpha,25(OH)_2D_3$.

4. EFFECTS OF VITAMIN D, $25(OH)D_3$ AND $1\alpha,25(OH)_2D_3$ ON THE RENAL HANDLING OF CALCIUM AND PHOSPHORUS

In vitamin D deficiency urine calcium concentrations are low, whereas, with vitamin D excess or intoxication hypercalciuria is frequently present [1]. The excretion of calcium and phosphorus in the urine, however, also reflects the decreased or increased calcium absorption in the intestine, the presence of hypo- or hyper-calcemia, and the presence of diminished or elevated concentrations of circulating PTH. Short term infusions of vitamin D_3, $25(OH)D_3$ and $1\alpha,25(OH)_2D_3$ decrease phosphate, calcium and sodium clearance relative to the clearance of inulin in parathyroidectomzed dogs [186–189]. We performed similar studies examining the effects of $25(OH)D_3$ on renal bicarbonate and phosphate reabsorption [190]. Unlike these earlier studies, we observed that phosphate and bicarbonate reabsorption increased only in intact animals but not in parathyroidectomized animals suggesting the need for PTH. Our observations are similar to those of others [191]. Yamamoto *et al.* examined the effects of $1\alpha,25(OH)_2D_3$ in vitamin D-deficient rats, vitamin D-deficient rats supplemented with dietary calcium to normalize plasma calcium and PTH levels, and vitamin D-replete rats following thyro-parathyroidectomy and the infusion of graded amounts of calcium [192]. Urinary calcium excretion was lower in vitamin D-replete rats than in vitamin D-deficient rats suggesting that vitamin D increased the efficiency of renal calcium reabsorption in the absence of PTH. In a second group of experiments, rats treated in the manner noted above, were TPTX and infused with PTH. The results of this experiment show that a lower dose of PTH is needed to exhibit a comparable effect on renal calcium reabsorption in vitamin D-replete rats when compared to vitamin D-deficient rats. This finding may be explained by *in vitro* studies of distal convoluted tubule cells in which $1\alpha,25(OH)_2D_3$ increased PTH/PTHrp receptor mRNA levels [193]. Micropuncture experiments have demonstrated that $25(OH)D_3$ exerts its anti-phosphaturic and hypocalciuric effects in the distal tubule [194, 195]. These effects occur shortly after the administration of $25(OH)D_3$ and are therefore independent of conversion of $25(OH)D_3$ to $1\alpha,25(OH)_2D_3$. $1\alpha,25(OH)_2D_3$ enhances the reabsorption of calcium in distal tubular segments of the nephron [196]. These studies are consistent with the localization of many vitamin D responsive proteins exclusively in the distal nephron. *In vitro* studies show that vitamin D deficiency is associated with decreased calcium uptake in luminal and basolateral membranes derived from distal nephron segments [197]. Cultured rabbit connecting tubule cells show an increase in the transport of calcium when treated with $1\alpha,25(OH)_2D_3$ [198]. ECaC1 (TRPV5, present in the apical membrane of the distal convoluted tubule) and ECaC2 (TRPV6, localized to the principal cells of the cortical and medullary collecting ducts) protein and mRNA are diminished in vitamin D deficient rats and increased by $1\alpha,25(OH)_2D_3$ [199–201]. $1\alpha,25(OH)_2D_3$ increases mRNA and protein expression of the PMCa pump which is involved with active transport of calcium through the basolateral membrane of distal tubule cells [202].

A synthesis of the experimental results suggest that 1α,25$(OH)_2D_3$ has effects on the distal tubular reabsorption of calcium through several mechanisms (Figure 2). Expression of proteins responsible for distal tubule calcium uptake, intracellular trafficking, and basolateral transport of calcium are responsive to 1α,25$(OH)_2D_3$.

5. DISTRIBUTION AND REGULATION OF VITAMIN D-DEPENDENT PROTEINS IN THE KIDNEY

Several vitamin D-dependent proteins are expressed in the kidney and many of these play a role in calcium and phosphate transport. The VDR, calbindins, PMCa pump, and the ECaCs (TRPV5 and TRPV6) are all found in renal tubule cells and act co-ordinately in the regulation of calcium transport in the nephron [5, 11, 14, 15, 28, 193, 199, 201, 203–206]. Additional vitamin D responsive proteins involved in renal calcium and phosphate transport are listed in Table 4.

5.1 1α,25$(OH)_2D_3$ receptor (VDR) in the kidney

The VDR mediates many, if not all, of the effects of 1α,25$(OH)_2D_3$ in diverse organs [207–215]. The distribution of the VDR in the kidney has been assessed using a variety of techniques [135, 216–220]. A full discussion regarding its distribution is given in a separate contribution in this volume. Suffice it to say that the VDR is found in varying amounts in different species in proximal and distal tubules [11, 135, 217, 218, 221]. In the human kidney, we found that the VDR was present abundantly in the distal tubule and to a lesser extent in the proximal tubule. Cells expressing calbindin-D_{28K} also express the calcium pump and the epithelial calcium channel [199, 222]. Interestingly, not all cells in the distal tubule expressed the receptor. Acid secreting cells do not express the VDR in significant amounts. Taken together, the results are consistent with the notion that the VDR is present in significant amounts in the distal tubule where it regulates the amount and the activity of several vitamin D-dependent proteins such as the plasma membrane calcium pump, epithelial calcium channel, and calbindin D_{28K}. Although present in lesser amounts, the proximal tubule also expresses the VDR where it regulates the activity of 1α-hydroxylase and 24-hydroxylase. The VDR is detected in the developing rat kidney *in vivo* as early as day 15 *post-coitum* (p.c.) and in the cultured metanephros [12]. Significant amounts of the VDR are found in the metanephric mesenchyme and the ureteric bud. As the kidney matures, the VDR is observed in S-shaped and comma-shaped bodies and in the developing glomerulus, specifically in the parietal and visceral endothelial cells. VDR staining in the latter cells persists in the adult kidney as well. Similar patterns of VDR expression were found in the developing mouse kidney *in vivo* and in mouse metanephric cultures.

The VDR is regulated by several factors in diverse tissues [223]. Concentrations of the VDR in the kidney and parathyroid glands are mainly regulated by 1α,25$(OH)_2D_3$, PTH and dietary calcium. Exogenous administration of 1α,25$(OH)_2D_3$ in rats increases VDR levels in duodenal and renal tissues [224, 225]. When endogenous 1α,25$(OH)_2D_3$ concentrations are increased by adapting an animal to a low calcium diet (Table 5), VDR concentrations in the duodenum and kidney do not increase. The difference appears to be due to increases in the levels of PTH elicited by the low calcium diet and decreased PTH following 1α,25$(OH)_2D_3$ administration. Differences in *Cyp24A1* expression (decreased in the presence of a low calcium diet, and increased following the administration of 1α,25$(OH)_2D_3$) might also contribute to this difference in VDR expression. PTH has been shown to down-regulate VDR in osteosarcoma cells as well as block up-regulation of VDR in rats infused with both PTH and 1α,25$(OH)_2D_3$ [226]. These results demonstrate the opposing effects of PTH and 1α,25$(OH)_2D_3$ on VDR expression in the kidney.

In addition to the VDR, several vitamin D-dependent proteins are expressed in the kidney. The pattern of regulation of these proteins is of great interest, in as much as it casts light on the different mechanisms by which calcium is transported in the kidney. Those proteins involved in calcium transport include: calbindin D_{28K} and calbindin D_{9K}, the plasma membrane calcium pump, the epithelial calcium channel, and the calcium sensing receptor. Additionally, although not involved in the transport of calcium, the 24-hydroxylase and the sodium-phosphate type 2 co-transporter (NaPi 2) are also expressed in the kidney. All of these proteins appear to be regulated by $1\alpha,25(OH)_2D_3$.

5.2 Calbindins D and the kidney

The two forms of calbindin-D, namely, calbindin-D_{28K} and calbindin-D_{9K} are variably distributed in different tissues [227–244]. The proteins are classical E-F-hand proteins. Calbindin-D_{28K} has 6-EF hands and the calbindin-D_{9K} has 2 such motifs [245–264]. Calbindin D_{28K} binds 4 moles of calcium per mole of protein and calbindin D_{9K} two moles of calcium per mole of protein [227]. In the mouse kidney, both forms are present and are regulated by vitamin D. In other species, only calbindin-D_{28K} is expressed in the kidney. Both proteins undergo conformational change upon binding to calcium [254, 256, 265]. We recently solved the structure of Ca^{2+}-loaded calbindin-D_{28K} using NMR spectroscopy [266]. The protein is comprised of a single, globular fold consisting of six distinct EF-hand subdomains, which coordinate Ca^{2+} in loops on EF1, EF3, EF4 and EF5. Ran-binding protein M, *myo*-inositol monophosphatase, and procaspase-3-derived peptides interact with the protein on a surface comprised of alpha5 (EF3), alpha8 (EF4) and the EF2–EF3 and EF4–EF5 loops. The Ca^{2+}-dependent conformational change is probably what allows the protein to act as a modulator of effector proteins.

Calbindin D_{28K} and calbindin-D_{9K} are $1\alpha,25(OH)_2D_3$-regulated proteins [258–264, 267]. Their expression is reduced in the kidneys of 1α–hydroxylase knock-out mice [268]. Calbindin D_{28K} and calbindin-D_{9K} expression is normalized by treatment with $1\alpha,25(OH)_2D_3$, however, only calbindin D_{28K} expression is increased when 1α–hydroxylase knock-out mice are fed a high calcium diet. In a mouse VDR-KO model, renal calbindin D_{9K} expression is nearly abolished whereas calbindin D_{28K} expression returns to control values as the animals age [269]. Calbindin-D_{9K} induction by $1\alpha,25(OH)_2D_3$ *in vitro* is absent in VDR null cells [270]. *In vitro,* PTH has a synergistic effect with $1\alpha,25(OH)_2D_3$ on calbindin D_{9K} expression [270]. In contrast, calbindin D_{28K} expression is enhanced by PTH infusion without elevations in $1\alpha,25(OH)_2D_3$ concentrations [271]. Preincubation with calbindin D_{28K} increases calcium uptake in luminal membranes while calbindin D_{9K} preincubation increases calcium uptake in basolateral membranes [272, 273]. Calbindin D_{28K} knock-out mice fed a high calcium (1%) diet have an elevated urinary calcium/creatinine ratio; curiously, no differences in serum calcium or PTH are noted compared to wild-type littermates [274, 275]. The elevated urinary calcium/creatinine ratio in calbindin D_{28K} knock-out mice is not apparent after fasting [274]. This is consistent with the finding that fractional excretion of calcium is not altered in calbindin D_{28K} knock-out mice fed a normal calcium diet [276]. The effects of calbindin D_{28K} on renal calcium conservation in mice are modest, perhaps because of compensatory increases in calbindin D_{9K}.

5.3. The plasma membrane calcium pump

In the adult human kidney, epitopes for the calcium-magnesium ATPase (calcium pump) are expressed in the basolateral membrane of distal tubular cells [14, 15, 28, 203, 204]. Similar patterns of expression were apparent in the rat [203] and rabbit kidney [277]. Not all cells of the distal tubule stained positively for the calcium pump; cells expressing acid secreting, carbonic anhydrase positive cells do not express the plasma membrane calcium pump, whereas the other cells of the distal tubule express the PMCa. Calbindin D_{28K} is co-

expressed with the PMCa pump. Our studies have been confirmed by others [277]. The PMCa pump is widely distributed in a large number of other calcium transporting tissues many of which display vitamin D-dependent calcium transport [13, 278–283].

The PMCa is regulated by 1α,25$(OH)_2D_3$. In MDBK (bovine distal tubule) cells, 1α,25$(OH)_2D_3$ treatment increases PMCa pump mRNA and protein [202]. Vitamin D deficiency decreases PMCa pump activity in the distal tubule of the kidney [272, 273]. We have shown that the PMCa pump mRNA and protein expression in the intestine increase after the administration of 1α,25$(OH)_2D_3$ [279, 284, 285]. The increase occurs within 3–6 hours and is dose-dependent. Dietary calcium and phosphorus depletion also increase the amount of PMCa pump expressed in the intestine. Thus, in the intestine and in intestinal cell basolateral membranes, 1α,25$(OH)_2D_3$ increases the synthesis of the PMCa pump. The mechanism by which up-regulation of PMCa pump activity occurs in the kidney and intestine is uncertain. 1α,25$(OH)_2D_3$-mediated increases in PMCa pump synthesis or stimulation by calbindin D_{9k} or calbindin D_{28k} are possibilities [272, 273]. There is also evidence that stimulation of the calcium sensing receptor (CaSR) decreases calcium absorption by inhibiting PMCa pump activity [286].

5.4. The epithelial calcium channel/vanilloid-receptor related transient receptor potential channels 5 and 6

Hoenderop *et al* described epithelial calcium channels (ECaCs) which are expressed in the apical membrane of the distal tubule and principal cells of the collecting duct and are distinct from previously described calcium channels [16, 200, 222, 287, 288]. There are at least two members in this family of calcium channels, ECaC-1/TRPV5 and ECaC-2/TRPV6 [288]. ECaC1/TRPV5 expression is limited to the kidney while ECaC2/TRPV6 is expressed in several other tissues [287–290]. The ECaCs/TRPV channels have six putative transmembrane spanning domains, including a pore-forming hydrophobic region between transmembrane domains 5 and 6 [16]. Several putative vitamin D response elements (VDRE) have been identified within the promoter region of the human TRPV5 channel. Hoenderop *et al* also demonstrated that ECaC1/TRPV5 mRNA and protein levels are increased to near control levels after vitamin D rescue in rats fed a vitamin D deficient diet [199]. In a study using 1α-hydroxylase knock-out mice, a greater than 50% reduction in ECaC-1/TRPV5 expression was found compared to control mice [268]. Additionally, renal ECaC-1/TRPV5 expression in 1α-hydroxylase KO mice is normalized after treatment with 1α,25$(OH)_2D_3$ [268]. Similar findings were seen when examining calbindin D_{28K} expression which co-localizes to the same distal tubule cells as ECaC-1/TRPV5 [222, 268]. However, ECaC-1/TRPV5 is not regulated through vitamin D effects on calbindin D_{28K}. Calbindin D_{28K} KO mice and cyclosporine A induced down-regulation of calbindin D_{28K} has no effect on ECaC-1/TRPV5 expression [274]. Others have suggested that calcium also regulates ECaC-1 expression. Quantitative PRC techniques showed reduced expression of ECaC-1 in VDR KO mice compared to control mice. When fed high calcium diets, VDR KO mice had normalization of ECaC-1 concentrations [290].

V. CONCLUSION

The kidney plays a vital role in the conservation of calcium and phosphorus. Besides being the site of synthesis of 1α,25$(OH)_2D_3$, the kidney responds to the hormone by increasing the efficiency of calcium and phosphorus reabsorption. Elements of the calcium transport systems including calbindin D_{28K}, calbindin D_{9K}, the epithelial calcium channel, and the plasma membrane calcium pump all localize to the distal portion of the nephron and are regulated directly or indirectly by 1α,25$(OH)_2D_3$.

HIGHLIGHTS

- The kidney is essential for the appropriate regulation of calcium and phosphorus homeostasis.
- It is the site of synthesis of the active, hormonal form of vitamin D, 1α, $25(OH)_2D$, as well as, the site of degradation of the hormone.
- Many proteins involved in the transport of calcium are regulated by 1α, $25(OH)_2D$.

Acknowledgments

Supported by NIH grants AR058003 and AR060869 and a grant from the Ralph and Marion C. Falk Foundation (to RK).

ABBREVIATIONS

1α,25(OH)$_2$D	1α,25-dihydroxyvitamin D
ECaC	epithelial calcium channel
PMCa	plasma membrane calcium pump
VDR	vitamin D receptor

REFERENCES

1. Metabolic bone and stone disease. New York: Churchill Livingstone, Edinburgh; 1993.
2. Friedman PA, Gesek FA. Physiological Reviews. 1995; 75:429–471. [PubMed: 7624390]
3. Borke JL, Penniston JT, Kumar R. Seminars in Nephrology. 1990; 10:15–23. [PubMed: 2153313]
4. Johnson JA, Kumar R. Current Opinion in Nephrology & Hypertension. 1994; 3:424–429. [PubMed: 8076146]
5. Kumar R. Journal of Cellular Biochemistry. 1995; 57:392–398. [PubMed: 7768975]
6. Fraser DR, Kodicek E. Nature. 1970; 228:764–766. [PubMed: 4319631]
7. Gray R, Boyle I, DeLuca HF. Science. 1971; 172:1232–1234. [PubMed: 4325104]
8. Shultz TD, Fox J, Heath H 3rd, Kumar R. Proc Natl Acad Sci U S A. 1983; 80:1746–1750. [PubMed: 6572938]
9. Satomura K, Seino Y, Yamaoka K, Tanaka Y, Ishida M, Yabuuchi H, DeLuca HF. Kidney Int. 1988; 34:712–716. [PubMed: 3199682]
10. Tanaka Y, DeLuca HF. Am J Physiol. 1984; 246:E168–E173. [PubMed: 6538071]
11. Kumar R, Schaefer J, Grande JP, Roche PC. American Journal of Physiology. 1994; 266:F477–F485. [PubMed: 8160797]
12. Johnson JA, Grande JP, Roche PC, Sweeney WE Jr, Avner ED, Kumar R. American Journal of Physiology. 1995; 269:F419–F428. [PubMed: 7573491]
13. Borke JL, Eriksen EF, Minami J, Keeting P, Mann KG, Penniston JT, Riggs BL, Kumar R. Journal of Clinical Endocrinology & Metabolism. 1988; 67:1299–1304. [PubMed: 2461388]
14. Borke JL, Minami J, Verma A, Penniston JT, Kumar R. Journal of Clinical Investigation. 1987; 80:1225–1231. [PubMed: 2445778]
15. Borke JL, Minami J, Verma AK, Penniston JT, Kumar R. Kidney International. 1988; 34:262–267. [PubMed: 2460662]
16. Muller D, Hoenderop JG, van Os CH, JMB R. Nephrology Dialysis Transplantation. 2001; 16:1329–1335.

17. Windhager EE, Frindt G, Milovanovic S. Ann N Y Acad Sci. 1991; 639:577–591. [PubMed: 1785887]
18. Berndt, TJ.; Knox, FG. The Kidney : physiology and pathophysiology. Seldin, DW.; Giebisch, GH., editors. New York: Raven Press; 1992. p. 2511-2532.
19. Knox, FG.; Haramati, A. The Kidney : physiology and pathophysiology. Seldin, DW.; Giebisch, GH., editors. New York: Raven Press; 1985. p. 1351-1396.
20. Knox FG, Haas JA, Berndt T, Marchand GR, Youngberg SP. American Journal of Physiology. 1977; 233:F150–F153. [PubMed: 888956]
21. Schiavi SC, Moe OW. Current Opinion in Nephrology & Hypertension. 2002; 11:423–430. [PubMed: 12105393]
22. Kumar R. Current Opinion in Nephrology & Hypertension. 2002; 11:547–553. [PubMed: 12187320]
23. Berndt T, Craig TA, Bowe AE, Vassiliadis J, Reczek D, Finnegan R, Jan De Beur SM, Schiavi SC, Kumar R. Journal of Clinical Investigation. 2003 In press.
24. Kumar, R. Fluids and electrolytes. Kokko, JP.; Tannen, RL., editors. Philadelphia: Saunders; 1996. p. 391-419.
25. Neuman, WF.; Neuman, MW. The chemical dynamics of bone mineral. Chicago: University of Chicago Press; 1958.
26. Moore EW. Journal of Clinical Investigation. 1970; 49:318–334. [PubMed: 4983663]
27. Raman A. Clinical Biochemistry. 1971; 4:141–146. [PubMed: 5143530]
28. Kumar R, Penniston JT, Borke JL. New Physiol Sci. 1988; 3:219–222.
29. Friedman PA. New Physiol Sci. 1988; 3:17–21.
30. Costanzo LS, Windhager EE. Am J Physiol. 1978; 235:F492–F506. [PubMed: 727266]
31. Costanzo LS, Windhager EE, Ellison DH. J Am Soc Nephrol. 2000; 11:1562–1580. [PubMed: 10991552]
32. Murer H, Hernando N, Forster I, Biber J. Curr Opin Nephrol Hypertens. 2001; 10:555–561. [PubMed: 11496046]
33. Picard N, Capuano P, Stange G, Mihailova M, Kaissling B, Murer H, Biber J, Wagner CA. Pflugers Arch.
34. Traebert M, Roth J, Biber J, Murer H, Kaissling B. American Journal of Physiology - Renal Fluid & Electrolyte Physiology. 2000; 278:F148–F154.
35. Murer H, Lotscher M, Kaissling B, Levi M, Kempson SA, Biber J. Kidney Int. 1996; 49:1769–1773. [PubMed: 8743494]
36. Keusch I, Traebert M, Lotscher M, Kaissling B, Murer H, Biber J. Kidney International. 1998; 54:1224–1232. [PubMed: 9767538]
37. Lotscher M, Scarpetta Y, Levi M, Halaihel N, Wang H, Zajicek HK, Biber J, Murer H, Kaissling B. Journal of Clinical Investigation. 1999; 104:483–494. [PubMed: 10449440]
38. Yamashita T, Konishi M, Miyake A, Inui K, Itoh N. Journal of Biological Chemistry. 2002; 277:28265–28270. [PubMed: 12032146]
39. O'Brien S, Bowe AE, Hernando N, Biber J, Murer H, Kumar R, Breusegem SY, Barry NP, Levi M, Pragnell M, Schiavi SC. Journal of the American Society of Nephrology. 2003 ASN abstract # 555303 (in press).
40. O'Brien S, Bowe AE, Pragnell M, Kumar R, Schiavi SC. Journal of Bone & Mineral Research. 2003 ASBMR Abstract.
41. Ghazarian JG, DeLuca HF. Archives of Biochemistry & Biophysics. 1974; 160:63–72. [PubMed: 4151327]
42. Ghazarian JG, Schnoes HK, DeLuca HF. Biochemistry. 1973; 12:2555–2558. [PubMed: 4711462]
43. Gray RW, Omdahl JL, Ghazarian JG, DeLuca HF. The Journal of biological chemistry. 1972; 247:7528–7532. [PubMed: 4404596]
44. Pedersen JI, Ghazarian JG, Orme-Johnson NR, DeLuca HF. Journal of Biological Chemistry. 1976; 251:3933–3941. [PubMed: 819434]
45. Yoon PS, DeLuca HF. Biochemistry. 1980; 19:2165–2171. [PubMed: 6246921]

46. Yoon PS, Deluca HF. Arch Biochem Biophys. 1980; 203:529–541. [PubMed: 6779710]
47. Yoon PS, DeLuca HF. Methods Enzymol. 1980; 67:430–440. [PubMed: 6767897]
48. Yoon PS, Rawlings J, Orme-Johnson WH, DeLuca HF. Biochemistry. 1980; 19:2172–2176. [PubMed: 6246922]
49. Fu GK, Lin D, Zhang MY, Bikle DD, Shackleton CH, Miller WL, Portale AA. Molecular Endocrinology. 1997; 11:1961–1970. [PubMed: 9415400]
50. Fu GK, Portale AA, Miller WL. DNA Cell Biol. 1997; 16:1499–1507. [PubMed: 9428799]
51. Monkawa T, Yoshida T, Wakino S, Shinki T, Anazawa H, Deluca HF, Suda T, Hayashi M, Saruta T. Biochem Biophys Res Commun. 1997; 239:527–533. [PubMed: 9344864]
52. St-Arnaud R, Messerlian S, Moir JM, Omdahl JL, Glorieux FH. J Bone Miner Res. 1997; 12:1552–1559. [PubMed: 9333115]
53. Takeyama K, Kitanaka S, Sato T, Kobori M, Yanagisawa J, Kato S. Science. 1997; 277:1827–1830. [PubMed: 9295274]
54. Fraser DR, Kodicek E. Nature. 1970; 228:764–766. [PubMed: 4319631]
55. Lawson DE, Fraser DR, Kodicek E, Morris HR, Williams DH. Nature. 1971; 230:228–230. [PubMed: 4323313]
56. Shultz TD, Fox J, Heath H 3rd, Kumar R. Proceedings of the National Academy of Sciences of the United States of America. 1983; 80:1746–1750. [PubMed: 6572938]
57. Weisman Y, Vargas A, Duckett G, Reiter E, Root AW. Endocrinology. 1978; 103:1992–1996. [PubMed: 748031]
58. Tanaka Y, Halloran B, Schnoes HK, DeLuca HF. Proceedings of the National Academy of Sciences of the United States of America. 1979; 76:5033–5035. [PubMed: 291919]
59. Bell, NH. Vitamin D : basic and clinical aspects. Kumar, R., editor. Boston Hingham, MA: Nijhoff ; Distributors for North America Kluwer Boston; 1984. p. 581-589.
60. Golconda MS, Larson TS, Kolb LG, Kumar R. Mayo Clinic Proceedings. 1996; 71:32–36. [PubMed: 8538229]
61. Schuster I, Egger H, Bikle D, Herzig G, Reddy GS, Stuetz A, Stuetz P, Vorisek G. Steroids. 2001; 66:409–422. [PubMed: 11179750]
62. Diaz L, Arranz C, Avila E, Halhali A, Vilchis F, Larrea F. Journal of Clinical Endocrinology & Metabolism. 2002; 87:3876–3882. [PubMed: 12161526]
63. Zehnder D, Evans KN, Kilby MD, Bulmer JN, Innes BA, Stewart PM, Hewison M. American Journal of Pathology. 2002; 161:105–114. [PubMed: 12107095]
64. Howard GA, Turner RT, Sherrard DJ, Baylink DJ. Journal of Biological Chemistry. 1981; 256:7738–7740. [PubMed: 6973569]
65. Dusso AS, Finch J, Brown A, Ritter C, Delmez J, Schreiner G, Slatopolsky E. Journal of Clinical Endocrinology & Metabolism. 1991; 72:157–164. [PubMed: 1986015]
66. Adams JS, Beeker TG, Hongo T, Clemens TL. J Bone Miner Res. 1990; 5:1265–1269. [PubMed: 1963733]
67. Adams JS, Gacad MA, Diz MM, Nadler JL. J Clin Endocrinol Metab. 1990; 70:595–600. [PubMed: 2106525]
68. Adams JS, Ren SY. Endocrinology. 1996; 137:4514–4517. [PubMed: 8828516]
69. Adams JS, Ren SY, Arbelle JE, Horiuchi N, Gray RW, Clemens TL, Shany S. Endocrinology. 1994; 134:2567–2573. [PubMed: 8194484]
70. Adams JS, Ren SY, Arbelle JE, Shany S, Gacad MA. Endocrinology. 1995; 136:2262–2269. [PubMed: 7536666]
71. Adams JS, Sharma OP, Gacad MA, Singer FR. J Clin Invest. 1983; 72:1856–1860. [PubMed: 6688814]
72. Wu S, Chun R, Gacad MA, Ren S, Chen H, Adams JS. Endocrinology. 2002; 143:4135. [PubMed: 12239126]
73. Henry, HL. Vitamin D: Basic and clinical aspects. Kumar, R., editor. Boston: Nijhoff ; Distributors for North America Kluwer Boston; 1984. p. 152-174.

74. Turner, RL. Vitamin D: Basic and clinical aspects. Kumar, R., editor. Boston: Nijhoff ; Distributors for North America Kluwer Boston; 1984. p. 175-196.
75. Rasmussen H, Wong M, Bikle D, Goodman DB. Journal of Clinical Investigation. 1972; 51:2502–2504. [PubMed: 4344734]
76. Trechsel U, Bonjour JP, Fleisch H. Journal of Clinical Investigation. 1979; 64:206–217. [PubMed: 109470]
77. Henry HL. Journal of Biological Chemistry. 1979; 254:2722–2729. [PubMed: 429315]
78. Bland R, Zehnder D, Hughes SV, Ronco PM, Stewart PM, Hewison M. Kidney International. 2001; 60:1277–1286. [PubMed: 11576342]
79. Zehnder D, Bland R, Walker EA, Bradwell AR, Howie AJ, Hewison M, Stewart PM. Journal of the American Society of Nephrology. 1999; 10:2465–2473. [PubMed: 10589683]
80. Panda DK, Miao D, Tremblay ML, Sirois J, Farookhi R, Hendy GN, Goltzman D. Proceedings of the National Academy of Sciences of the United States of America. 2001; 98:7498–7503. [PubMed: 11416220]
81. Saito H, Kusano K, Kinosaki M, Ito H, Hirata M, Segawa H, Miyamoto K, Fukushima N. Journal of Biological Chemistry. 2003; 278:2206–2211. [PubMed: 12419819]
82. Shimada T, Hasegawa H, Yamazaki Y, Muto T, Hino R, Takeuchi Y, Fujita T, Nakahara K, Fukumoto S, Yamashita T. J Bone Miner Res. 2004; 19:429–435. [PubMed: 15040831]
83. Shimada T, Kakitani M, Yamazaki Y, Hasegawa H, Takeuchi Y, Fujita T, Fukumoto S, Tomizuka K, Yamashita T. J Clin Invest. 2004; 113:561–568. [PubMed: 14966565]
84. Shimada T, Mizutani S, Muto T, Yoneya T, Hino R, Takeda S, Takeuchi Y, Fujita T, Fukumoto S, Yamashita T. Proceedings of the National Academy of Sciences of the United States of America. 2001; 98:6500–6505. [PubMed: 11344269]
85. Shimada T, Urakawa I, Yamazaki Y, Hasegawa H, Hino R, Yoneya T, Takeuchi Y, Fujita T, Fukumoto S, Yamashita T. Biochem Biophys Res Commun. 2004; 314:409–414. [PubMed: 14733920]
86. Urakawa I, Yamazaki Y, Shimada T, Iijima K, Hasegawa H, Okawa K, Fujita T, Fukumoto S, Yamashita T. Nature. 2006; 444:770–774. [PubMed: 17086194]
87. Kuro-o M. Pflugers Arch. 2010; 459:333–343. [PubMed: 19730882]
88. Kuro-o M. Pediatr Nephrol. 2010; 25:583–590. [PubMed: 19626341]
89. Kurosu H, Ogawa Y, Miyoshi M, Yamamoto M, Nandi A, Rosenblatt KP, Baum MG, Schiavi S, Hu MC, Moe OW, Kuro-o M. Journal of Biological Chemistry. 2006; 281:6120–6123. [PubMed: 16436388]
90. Medici D, Razzaque MS, Deluca S, Rector TL, Hou B, Kang K, Goetz R, Mohammadi M, Kuro OM, Olsen BR, Lanske B. J Cell Biol. 2008; 182:459–465. [PubMed: 18678710]
91. Nakatani T, Sarraj B, Ohnishi M, Densmore MJ, Taguchi T, Goetz R, Mohammadi M, Lanske B, Razzaque MS. FASEB J. 2009; 23:433–441. [PubMed: 18835926]
92. Razzaque MS, Lanske B. Trends Mol Med. 2006; 12:298–305. [PubMed: 16731043]
93. Razzaque MS, Sitara D, Taguchi T, St-Arnaud R, Lanske B. FASEB J. 2006; 20:720–722. [PubMed: 16436465]
94. Kumar R. Physiological Reviews. 1984; 64:478–504. [PubMed: 6324253]
95. Turunen MM, Dunlop TW, Carlberg C, Vaisanen S. Nucleic Acids Res. 2007; 35:2734–2747. [PubMed: 17426122]
96. Kumar R, Iachini DN, Neilsen PM, Kaplan J, Michalakas J, Anderson PH, May BK, Morris HA, Callen DF. Molecular and cellular endocrinology. 325:46–53. [PubMed: 20450955]
97. Annalora AJ, Goodin DB, Hong WX, Zhang Q, Johnson EF, Stout CD. J Mol Biol. 2010; 396:441–451. [PubMed: 19961857]
98. Strushkevich N, Usanov SA, Plotnikov AN, Jones G, Park HW. J Mol Biol. 2008; 380:95–106. [PubMed: 18511070]
99. Kelley LA, Sternberg MJ. Nat Protoc. 2009; 4:363–371. [PubMed: 19247286]
100. Roy A, Kucukural A, Zhang Y. Nat Protoc. 2010; 5:725–738. [PubMed: 20360767]
101. Zhang Y. Proteins. 2007; 69(Suppl 8):108–117. [PubMed: 17894355]

102. Trott O, Olson AJ. J Comput Chem. 2010; 31:455–461. [PubMed: 19499576]
103. Rhieu SY, Annalora AJ, Gathungu RM, Vouros P, Uskokovic MR, Schuster I, Palmore GT, Reddy GS. Arch Biochem Biophys. 2011; 509:33–43. [PubMed: 21338573]
104. Rubin LP, Yeung B, Vouros P, Vilner LM, Reddy GS. Pediatric Research. 1993; 34:98–104. [PubMed: 8356026]
105. Staal A, van den Bemd GJ, Birkenhager JC, Pols HA, van Leeuwen JP. Bone. 1997; 20:237–243. [PubMed: 9071474]
106. Armbrecht HJ, Hodam TL, Boltz MA, Partridge NC, Brown AJ, Kumar VB. Endocrinology. 1998; 139:3375–3381. [PubMed: 9681485]
107. Kurahashi I, Matsunuma A, Kawane T, Abe M, Horiuchi N. Endocrine Journal. 2002; 17:109–118.
108. Nishimura A, Shinki T, Jin CH, Ohyama Y, Noshiro M, Okuda K, Suda T. Endocrinology. 1994; 134:1794–1799. [PubMed: 8137744]
109. Kreutz M, Andreesen R, Krause SW, Szabo A, Ritz E, Reichel H. Blood. 1993; 82:1300–1307. [PubMed: 8394753]
110. Demers C, Lemay J, Hendy GN, Gascon-Barre M. Journal of Molecular Endocrinology. 1997; 18:37–48. [PubMed: 9061605]
111. Theodoropoulos C, Demers C, Delvin E, Menard D, Gascon-Barre M. Clinical Endocrinology. 2003; 58:489–499. [PubMed: 12641633]
112. Furuichi T, Kawata S, Asoh Y, Kumaki K, Ohyama Y. Life Sciences. 1998; 62:453–459. [PubMed: 9449236]
113. Akeno N, Saikatsu S, Kawane T, Horiuchi N. Endocrinology. 1997; 138:2233–2240. [PubMed: 9165006]
114. Tanaka Y, Lorenc RS, DeLuca HF. Archives of Biochemistry & Biophysics. 1975; 171:521–526. [PubMed: 1200639]
115. Tanaka Y, DeLuca HF, Ikekawa N, Morisaki M, Koizumi N. Archives of Biochemistry & Biophysics. 1975; 170:620–626. [PubMed: 1190783]
116. Holick MF, Baxter LA, Schraufrogel PK, Tavela TE, DeLuca HF. Journal of Biological Chemistry. 1976; 251:397–402. [PubMed: 1245479]
117. Holick MF, Schnoes HK, DeLuca HF, Gray RW, Boyle IT, Suda T. Biochemistry. 1972; 11:4251–4255. [PubMed: 4342902]
118. Knutson JC, DeLuca HF. Biochemistry. 1974; 13:1543–1548. [PubMed: 4150615]
119. Kumar R, Schnoes HK, DeLuca HF. Journal of Biological Chemistry. 1978; 253:3804–3809. [PubMed: 649607]
120. Garabedian M, Du Bois MB, Corvol MT, Pezant E, Balsan S. Endocrinology. 1978; 102:1262–1268. [PubMed: 744024]
121. Kulkowski JA, Chan T, Martinez J, Ghazarian JG. Biochemical & Biophysical Research Communications. 1979; 90:50–57. [PubMed: 227393]
122. DeLuca H. Nutrition Reviews. 1979; 37:161–193. [PubMed: 542234]
123. Boyle IT, Gray RW, DeLuca HF. Proceedings of the National Academy of Sciences of the United States of America. 1971; 68:2131–2134. [PubMed: 4332247]
124. Ornoy A, Goodwin D, Noff D, Edelstein S. Nature. 1978; 276:517–519. [PubMed: 723936]
125. Henry HL, Taylor AN, Norman AW. Journal of Nutrition. 1977; 107:1918–1926. [PubMed: 903834]
126. Tanaka Y, Castillo L, DeLuca HF. Proceedings of the National Academy of Sciences of the United States of America. 1976; 73:2701–2705. [PubMed: 1066683]
127. Tanaka Y, Deluca HF. Archives of Biochemistry & Biophysics. 1973; 154:566–574. [PubMed: 4691503]
128. Shinki T, Jin CH, Nishimura A, Nagai Y, Ohyama Y, Noshiro M, Okuda K, Suda T. Journal of Biological Chemistry. 1992; 267:13757–13762. [PubMed: 1618870]
129. Zierold C, Mings JA, DeLuca HF. Proceedings of the National Academy of Sciences of the United States of America. 2001; 98:13572–13576. [PubMed: 11698670]

130. Zierold C, Reinholz GG, Mings JA, Prahl JM, DeLuca HF. Archives of Biochemistry & Biophysics. 2000; 381:323–327. [PubMed: 11032422]
131. Matsumoto T, Kawanobe Y, Ogata E. Biochimica et Biophysica Acta. 1985; 845:358–365. [PubMed: 3924117]
132. Armbrecht HJ, Boltz MA. FEBS Letters. 1991; 292:17–20. [PubMed: 1959602]
133. Gray RW, Omdahl JL, Ghazarian JG, Horst RL. Steroids. 1990; 55:395–398. [PubMed: 2281516]
134. Chen ML, Boltz MA, Armbrecht HJ. Endocrinology. 1993; 132:1782–1788. [PubMed: 7681765]
135. Kawashima H, Torikai S, Kurokawa K. Proceedings of the National Academy of Sciences of the United States of America. 1981; 78:1199–1203. [PubMed: 6940135]
136. Yang W, Friedman PA, Kumar R, Omdahl JL, May BK, Siu-Caldera ML, Reddy GS, Christakos S. American Journal of Physiology. 1999; 276:E793–E805. [PubMed: 10198318]
137. Omdahl JL, Morris HA, May BK. Annual Review of Nutrition. 2002; 22:139–166.
138. Akiyoshi-Shibata M, Sakaki T, Ohyama Y, Noshiro M, Okuda K, Yabusaki Y. European Journal of Biochemistry. 1994; 224:335–343. [PubMed: 7925346]
139. Burgos-Trinidad M, DeLuca HF. Biochimica et Biophysica Acta. 1991; 1078:226–230. [PubMed: 2065089]
140. Taniguchi T, Eto TA, Shiotsuki H, Sueta H, Higashi S, Iwamura T, Okuda KI, Setoguchi T. Journal of Bone & Mineral Research. 2001; 16:57–62. [PubMed: 11149490]
141. Seo EG, Norman AW. Journal of Bone & Mineral Research. 1997; 12:598–606. [PubMed: 9101371]
142. Henry HL, Norman AW. Science. 1978; 201:835–837. [PubMed: 684411]
143. St-Arnaud R, Arabian A, Travers R, Barletta F, Raval-Pandya M, Chapin K, Depovere J, Mathieu C, Christakos S, Demay MB, Glorieux FH. Endocrinology. 2000; 141:2658–2666. [PubMed: 10875271]
144. Kobayashi Y, Taguchi T, Terada T, Oshida J, Morisaki M, Ikekawa N. Tetrahedron. 1979; 22:2023–2026.
145. Yamada S, Ohmori M, Takayama H. Tetrahedron. 1979; 21:1859–1862.
146. Halloran BP, DeLuca HF, Yamada S, Takayama H, Jee WS. Calcified Tissue International. 1981; 33:489–497. [PubMed: 6274486]
147. Tanaka Y, DeLuca HF, Kobayashi Y, Taguchi T, Ikekawa N, Morisaki M. Journal of Biological Chemistry. 1979; 254:7163–7167. [PubMed: 457674]
148. Mathews CHE, Parfitt AM, Brommage R, De Luca HF. American Society for Bone and Mineral Research (abstract). 1983:S66.
149. Tanaka Y, Pahuja DN, Wichmann JK, De Luca HF, Kobayashi Y, Taguchi T, Ikekawa N. Archives of Biochemistry & Biophysics. 1982; 218:134–141. [PubMed: 7149723]
150. Takasaki Y, Horiuchi N, Takahashi N, Abe E, Shinki T, Suda T, Yamada S, Takayama H, Horikawa H, Masumura T, Sugahara M. Biochemical & Biophysical Research Communications. 1980; 95:177–181. [PubMed: 7417247]
151. Takasaki Y, Suda T, Yamada S, Takayama H, Nishii Y. Biochemistry. 1981; 20:1681–1686. [PubMed: 7225351]
152. Wichmann JK, Schnoes HK, DeLuca HF. Biochemistry. 1981; 20:7385–7391. [PubMed: 6275880]
153. Redel J, Bazely N, Tanaka Y, DeLuca HF. FEBS Letters. 1978; 94:228–230. [PubMed: 700144]
154. Partridge JJ, Shiuey SJ, Chadha NK, Baggiolini EG, Blount JF. Journal Am Chem Society. 1981; 103:1253–1255.
155. Cesario M, Guilhem J, Pascard C, Redel J. Tetrahedron. 1978; 12:1097–1098.
156. Cesario M, Guilhem J, Pascard C, Redel J. Tetrahedron. 1980; 21:1588.
157. Horst RL, Littledike ET. Biochemical & Biophysical Research Communications. 1980; 93:149–154. [PubMed: 6246894]
158. Gray RW, Caldas AE, Weber JL, Ghazarian JG. Biochemical & Biophysical Research Communications. 1978; 82:212–218. [PubMed: 208553]

159. Tanaka Y, Shepard RA, DeLuca HF, Schnoes HK. Biochemical & Biophysical Research Communications. 1978; 83:7–13. [PubMed: 697824]
160. Horst RL. Biochemical & Biophysical Research Communications. 1979; 89:286–293. [PubMed: 475814]
161. Wichmann JK, DeLuca HF, Schnoes HK, Horst RL, Shepard RM, Jorgensen NA. Biochemistry. 1979; 18:4775–4780. [PubMed: 228698]
162. Ishizuka S, Ishimoto S, Norman AW. FEBS Letters. 1982; 138:83–87. [PubMed: 7067832]
163. Tanaka Y, Wichmann JK, Schnoes HK, DeLuca HF. Biochemistry. 1981; 20:3875–3879. [PubMed: 6268149]
164. Horst RL, Pramanik BC, Reinhardt TA, Shiuey SJ, Partridge JJ, Uskokovic MR, Napoli JL. Biochemical & Biophysical Research Communications. 1982; 106:1006–1011. [PubMed: 6288026]
165. Partridge, JJ.; Chadha, NK.; Shiuey, SJ.; Wovkulich, PM.; Uskokovic, MR.; Napoli, JL.; Horst, RL. Vitamin D: Endocrinological aspects and their clinical applications. Norman, AW., editor. New York: Walter de Gruyter; 1982. p. 1073-1078.
166. Morris DS, Williams DH, Norris AF. Journal of Organic Chemistry. 1981; 46:3422–3428.
167. Morris DS, Williams DH, Norris AF. Journal Chem Soc Chem Comm. 1981; 9:424–425.
168. Tanaka Y, Wichmann JK, Paaren HE, Schnoes HK, DeLuca HF. Proceedings of the National Academy of Sciences of the United States of America. 1980; 77:6411–6414. [PubMed: 6935655]
169. Horst RL, Reinhardt TA, Napoli JL. Biochemical & Biophysical Research Communications. 1982; 107:1319–1325. [PubMed: 6291527]
170. Horst RL, Reinhardt TA, Pramanik BC, Napoli JL. Biochemistry. 1983; 22:245–250. [PubMed: 6297543]
171. Kumar, R. Vitamin D: Basic and clinical aspects. Kumar, R., editor. Boston: Martinus Nirjhoff; 1984. p. 69-90.
172. Frolik CA, DeLuca HF. Journal of Clinical Investigation. 1972; 51:2900–2906. [PubMed: 4342957]
173. Frolik CA, DeLuca HF. Journal of Clinical Investigation. 1973; 52:543–548. [PubMed: 4685080]
174. Kumar R, Harnden D, DeLuca HF. Biochemistry. 1976; 15:2420–2423. [PubMed: 1276151]
175. Esvelt RP, Schnoes HK, DeLuca HF. Biochemistry. 1979; 18:3977–3983. [PubMed: 486408]
176. Ohnuma N, Kruse JR, Popjak G, Norman AW. Journal of Biological Chemistry. 1982; 257:5097–5102. [PubMed: 6896055]
177. Ohnuma N, Norman AW. Journal of Biological Chemistry. 1982; 257:8261–8271. [PubMed: 6896331]
178. Reinhardt TA, Napoli JL, Beitz DC, Littledike ET, Horst RL. Biochemical & Biophysical Research Communications. 1981; 99:302–307. [PubMed: 6263274]
179. Kumar R, Nagubandi S, Mattox VR, Londowski JM. Journal of Clinical Investigation. 1980; 65:277–284. [PubMed: 7356679]
180. Kumar R, Nagubandi S, Londowski JM. Digestive Diseases & Sciences. 1981; 26:242–246. [PubMed: 6894570]
181. Litwiller RD, Mattox VR, Jardine I, Kumar R. Journal of Biological Chemistry. 1982; 257:7491–7494. [PubMed: 6896328]
182. Gray RW, Caldas AE, Wilz DR, Lemann J Jr, Smith GA, DeLuca HF. Journal of Clinical Endocrinology & Metabolism. 1978; 46:756–765. [PubMed: 263717]
183. Seeman E, Kumar R, Hunder GG, Scott M, Heath H 3rd, Riggs BL. Journal of Clinical Investigation. 1980; 66:664–669. [PubMed: 7419714]
184. Wiesner RH, Kumar R, Seeman E, Go VL. Journal of Laboratory & Clinical Medicine. 1980; 96:1094–1100. [PubMed: 7430765]
185. Harnden D, Kumar R, Holick MF, Deluca HF. Science. 1976; 193:493–494. [PubMed: 941020]
186. Puschett JB, Beck WS Jr. Science. 1975; 190:473–475. [PubMed: 1166316]
187. Puschett JB, Beck WS Jr, Jelonek A, Fernandez PC. Journal of Clinical Investigation. 1974; 53:756–767. [PubMed: 4359939]

188. Puschett JB, Moranz J, Kurnick WS. Journal of Clinical Investigation. 1972; 51:373–385. [PubMed: 4333022]
189. Puschett JB, Fernandez PC, Boyle IT, Gray RW, Omdahl JL, DeLuca HF. Proceedings of the Society for Experimental Biology & Medicine. 1972; 141:379–384. [PubMed: 5082314]
190. Siegfried, D.; Kumar, R.; Arruda; Kurtzman, N. Influence of vitamin D on bicarbonate reabsorption. In: Massry, SG., editor. Proceedings of the 2nd International Congress on Phosphate. Heidelberg: Plenum press; 1976.
191. Popovtzer MM, Robinette JB, DeLuca HF, Holick MF. Journal of Clinical Investigation. 1974; 53:913–921. [PubMed: 4812447]
192. Yamamoto M, Kawanobe Y, Takahashi H, Shimazawa E, Kimura S, Ogata E. Journal of Clinical Investigation. 1984; 74:507–513. [PubMed: 6086715]
193. Sneddon WB, Barry EL, Coutermarsh BA, Gesek FA, Liu F, Friedman PA. Cellular Physiology & Biochemistry. 1998; 8:261–277. [PubMed: 9792954]
194. Winaver J, Sylk DB, Robertson JS, Chen TC, Puschett JB. Mineral Electrolyte Metab. 1980; 4:178–188.
195. Sutton, RAL.; NLM, W.; Dirks, JS. Dirks, 25-hydroxyvitamin D_3 (25(OH)D_3): Enhancement of distal tubular calcium reabsorption in the dog, 8th annual meeting of the american society of nephrology; 1975. p. 8
196. Harris CA, Seely JF. Clinical Res. 1979; 27:417A.
197. Bouhtiauy I, Lajeunesse D, Brunette MG. Endocrinology. 1993; 132:115–120. [PubMed: 8419116]
198. Bindels RJ, Hartog A, Timmermans J, Van Os CH. American Journal of Physiology. 1991; 261:F799–F807. [PubMed: 1659224]
199. Hoenderop JG, Muller D, Van Der Kemp AW, Hartog A, Suzuki M, Ishibashi K, Imai M, Sweep F, Willems PH, Van Os CH, Bindels RJ. Journal of the American Society of Nephrology. 2001; 12:1342–1349. [PubMed: 11423563]
200. Nijenhuis T, Hoenderop JG, van der Kemp AW, Bindels RJ. J Am Soc Nephrol. 2003; 14:2731–2740. [PubMed: 14569082]
201. Woudenberg-Vrenken TE, Bindels RJ, Hoenderop JG. Nature reviews. 2009; 5:441–449.
202. Glendenning P, Ratajczak T, Dick IM, Prince RL. Archives of Biochemistry & Biophysics. 2000; 380:126–132. [PubMed: 10900141]
203. Borke JL, Caride A, Verma AK, Penniston JT, Kumar R. American Journal of Physiology. 1989; 257:F842–F849. [PubMed: 2556040]
204. Johnson JA, Kumar R. Seminars in Nephrology. 1994; 14:119–128. [PubMed: 8177979]
205. Canaff L, Hendy GN. Journal of Biological Chemistry. 2002; 277:30337–30350. [PubMed: 12036954]
206. Wald H, Dranitzki-Elhalel M, Backenroth R, Popovtzer MM. Pflugers Archiv - European Journal of Physiology. 1998; 436:289–294. [PubMed: 9594030]
207. Ozono K, Sone T, Pike JW. Journal of Bone & Mineral Research. 1991; 6:1021–1027. [PubMed: 1665675]
208. Pike JW. Annual Review of Nutrition. 1991; 11:189–216.
209. Hughes MR, Malloy PJ, O'Malley BW, Pike JW, Feldman D. Journal of Receptor Research. 1991; 11:699–716. [PubMed: 1653354]
210. Feldman D, Malloy PJ. Molecular & Cellular Endocrinology. 1990; 72:C57–C62. [PubMed: 1963156]
211. Malloy PJ, Hochberg Z, Tiosano D, Pike JW, Hughes MR, Feldman D. Journal of Clinical Investigation. 1990; 86:2071–2079. [PubMed: 2174914]
212. Hughes M, Malloy P, Kieback D, McDonnell D, Feldman D, Pike JW, O'Malley B. Advances in Experimental Medicine & Biology. 1989; 255:491–503. [PubMed: 2618873]
213. Ritchie HH, Hughes MR, Thompson ET, Malloy PJ, Hochberg Z, Feldman D, Pike JW, O'Malley BW. Proceedings of the National Academy of Sciences of the United States of America. 1989; 86:9783–9787. [PubMed: 2557627]

214. Sone T, Scott RA, Hughes MR, Malloy PJ, Feldman D, O'Malley BW, Pike JW. Journal of Biological Chemistry. 1989; 264:20230–20234. [PubMed: 2555349]
215. Hughes MR, Malloy PJ, Kieback DG, Kesterson RA, Pike JW, Feldman D, O'Malley BW. Science. 1988; 242:1702–1705. [PubMed: 2849209]
216. Stumpf WE, Sar M, Narbaitz R, Reid FA, DeLuca HF, Tanaka Y. Proceedings of the National Academy of Sciences of the United States of America. 1980; 77:1149–1153. [PubMed: 6928665]
217. Stumpf WE, Sar M, Reid FA, Tanaka Y, DeLuca HF. Science. 1979; 206:1188–1190. [PubMed: 505004]
218. Kawashima H, Kurokawa K. Journal of Biological Chemistry. 1982; 257:13428–13432. [PubMed: 6754714]
219. Liu L, Ng M, Iacopino AM, Dunn ST, Hughes MR, Bourdeau JE. Journal of the American Society of Nephrology. 1994; 5:1251–1258. [PubMed: 7873736]
220. Liu L, Khastgir A, McCauley JM, Dunn ST, Morrissey JH, Christakos S, Hughes MR, Bourdeau JE. American Journal of Physiology. 1996; 270:F677–F681. [PubMed: 8967346]
221. Solvsten H, Fogh K, Svendsen M, Kristensen P, Astrom A, Kumar R, Kragballe K. Journal of Investigative Dermatology. Symposium Proceedings. 1996; 1:28–32.
222. Hoenderop JG, van der Kemp AW, Hartog A, van de Graaf SF, van Os CH, Willems PH, Bindels RJ. Journal of Biological Chemistry. 1999; 274:8375–8378. [PubMed: 10085067]
223. Kumar R. Nephrology Dialysis Transplantation. 1996; 11(Suppl 3):6–10.
224. Goff JP, Reinhardt TA, Beckman MJ, Horst RL. Endocrinology. 1990; 126:1031–1035. [PubMed: 2153516]
225. Uhland-Smith A, DeLuca HF. Biochimica et Biophysica Acta. 1993; 1176:321–326. [PubMed: 8385999]
226. Reinhardt TA, Horst RL. Endocrinology. 1990; 127:942–948. [PubMed: 2164926]
227. Gross M, Kumar R. American Journal of Physiology. 1990; 259:F195–F209. [PubMed: 2201202]
228. Desplan C, Heidmann O, Lillie JW, Auffray C, Thomasset M. Journal of Biological Chemistry. 1983; 258:13502–13505. [PubMed: 6315698]
229. Desplan C, Thomasset M, Moukhtar M. Journal of Biological Chemistry. 1983; 258:2762–2765. [PubMed: 6298202]
230. Fullmer CS, Wasserman RH. Proceedings of the National Academy of Sciences of the United States of America. 1987; 84:4772–4776. [PubMed: 3474624]
231. Hofmann T, Kawakami M, Hitchman AJ, Harrison JE, Dorrington KJ. Canadian Journal of Biochemistry. 1979; 57:737–748. [PubMed: 476518]
232. Horrocks, WD. Advances in Inorganic Biochemistry. Eichorn, GL.; Marzitti, LG., editors. New York: Elsevier; 1982. p. 201-261.
233. Hunziker W. Proceedings of the National Academy of Sciences of the United States of America. 1986; 83:7578–7582. [PubMed: 3463988]
234. Hunziker W, Schrickel S. Molecular Endocrinology. 1988; 2:465–473. [PubMed: 2843757]
235. Kumar R, Wieben E, Beecher SJ. Molecular Endocrinology. 1989; 3:427–432. [PubMed: 2710141]
236. Lomri N, Perret C, Gouhier N, Thomasset M. Gene. 1989; 80:87–98. [PubMed: 2792772]
237. Parmentier M, Lawson DE, Vassart G. European Journal of Biochemistry. 1987; 170:207–215. [PubMed: 3691519]
238. Takagi T, Nojiri M, Konishi K, Maruyama K, Nonomura Y. FEBS Letters. 1986; 201:41–45. [PubMed: 3754823]
239. Tsarbopoulos A, Gross M, Kumar R, Jardine I. Biomedical & Environmental Mass Spectrometry. 1989; 18:387–393. [PubMed: 2765697]
240. Wilson PW, Harding M, Lawson DE. Nucleic Acids Research. 1985; 13:8867–8881. [PubMed: 3841205]
241. Yamakuni T, Kuwano R, Odani S, Miki N, Yamaguchi Y, Takahashi Y. Nucleic Acids Research. 1986; 14:6768. [PubMed: 3755822]

242. Darwish HM, Krisinger J, Strom M, DeLuca HF. Proceedings of the National Academy of Sciences of the United States of America. 1987; 84:6108–6111. [PubMed: 3476932]
243. Fullmer CS, Wasserman RH. Journal of Biological Chemistry. 1981; 256:5669–5674. [PubMed: 7240164]
244. MacManus JP, Watson DC, Yaguchi M. Biochemical Journal. 1986; 235:585–595. [PubMed: 3741407]
245. Szebenyi DM, Obendorf SK, Moffat K. Nature. 1981; 294:327–332. [PubMed: 7312031]
246. Elms TN, Taylor AN. Journal of Dental Research. 1987; 66:1431–1434. [PubMed: 3476614]
247. Dorrington KJ, Kells DI, Hitchman AJ, Hartison JE, Hofmann T. Canadian Journal of Biochemistry. 1978; 56:492–499. [PubMed: 667695]
248. O'Neil JD, Dorrington KJ, Hofmann T. Canadian Journal of Biochemistry & Cell Biology. 1984; 62:434–442. [PubMed: 6432301]
249. Chiba K, Ohyashiki T, Mohri T. Journal of Biochemistry. 1983; 93:487–493. [PubMed: 6841348]
250. Vogel HJ, Drakenberg T, Forsen S, O'Neil JD, Hofmann T. Biochemistry. 1985; 24:3870–3876. [PubMed: 4052373]
251. Shelling JG, Sykes BD. Journal of Biological Chemistry. 1985; 260:8342–8347. [PubMed: 3924913]
252. Bredderman PJ, Wasserman RH. Biochemistry. 1974; 13:1687–1694. [PubMed: 4831357]
253. Fullmer CS, Edelstein S, Wasserman RH. Journal of Biological Chemistry. 1985; 260:6816–6819. [PubMed: 3997849]
254. Gross MD, Nelsestuen GL, Kumar R. Journal of Biological Chemistry. 1987; 262:6539–6545. [PubMed: 3571269]
255. Gross MD, Kumar R, Hunziker W. Journal of Biological Chemistry. 1988; 263:14426–14432. [PubMed: 3049577]
256. Veenstra TD, Gross MD, Hunziker W, Kumar R. Journal of Biological Chemistry. 1995; 270:30353–30358. [PubMed: 8530460]
257. Kumar R, Hunziker W, Gross M, Naylor S, Londowski JM, Schaefer J. Archives of Biochemistry & Biophysics. 1994; 308:311–317. [PubMed: 8311469]
258. Wasserman RH, Corradino RA, Fullmer CS, Taylor AN. Vitamins & Hormones. 1974; 32:299–324. [PubMed: 4376296]
259. Christakos S, Brunette MG, Norman AW. Endocrinology. 1981; 109:322–324. [PubMed: 7238412]
260. Taylor AN, Wasserman RH. Archives of Biochemistry & Biophysics. 1967; 119:536–540. [PubMed: 4293192]
261. Taylor AN, McIntosh JE, Bourdeau JE. Kidney International. 1982; 21:765–773. [PubMed: 6180216]
262. Roth J, Thorens B, Hunziker W, Norman AW, Orci L. Science. 1981; 214:197–200. [PubMed: 7025212]
263. Roth J, Brown D, Norman AW, Orci L. American Journal of Physiology. 1982; 243:F243–F252. [PubMed: 7051856]
264. Thomasset M, Parkes CO, Cuisinier-Gleizes P. American Journal of Physiology. 1982; 243:E483–E488. [PubMed: 7149019]
265. Venters RA, Benson LM, Craig TA, Bagu J, Paul KH, Kordys DR, Thompson R, Naylor S, Kumar R, Cavanagh J. Analytical biochemistry. 2003; 317:59–66. [PubMed: 12729601]
266. Kojetin DJ, Venters RA, Kordys DR, Thompson RJ, Kumar R, Cavanagh J. Nature structural & molecular biology. 2006; 13:641–647.
267. Hemmingsen C, Staun M, Nielsen PK, Olgaard K. Pharmacology & Toxicology. 2002; 91:111–115. [PubMed: 12427110]
268. Hoenderop JG, Dardenne O, Van Abel M, Van Der Kemp AW, Van Os CH, St - Arnaud R, Bindels RJ. FASEB Journal. 2002; 16:1398–1406. [PubMed: 12205031]
269. Li YC, Bolt MJ, Cao LP, Sitrin MD. American Journal of Physiology - Endocrinology & Metabolism. 2001; 281:E558–E564. [PubMed: 11500311]

270. Cao LP, Bolt MJ, Wei M, Sitrin MD, Chun Li Y. Archives of Biochemistry & Biophysics. 2002; 400:118–124. [PubMed: 11913978]
271. Hemmingsen C, Staun M, Lewin E, Nielsen PK, Olgaard K. Journal of Bone & Mineral Research. 1996; 11:1086–1093. [PubMed: 8854244]
272. Bouhtiauy I, Lajeunesse D, Christakos S, Brunette MG. Kidney International. 1994; 45:461–468. [PubMed: 8164434]
273. Bouhtiauy I, Lajeunesse D, Christakos S, Brunette MG. Kidney International. 1994; 45:469–474. [PubMed: 8164435]
274. Lee CT, Huynh VM, Lai LW, Lien YH. Kidney International. 2002; 62:2055–2061. [PubMed: 12427129]
275. Sooy K, Kohut J, Meyer M, Yeh C, Hou T, Christakos S. J Bone Min Res. 1999; 14(Suppl 1):S211.
276. Lutz W, Berndt T, Knox FG, Kumar R. FASEB Journal. 2000; 14:A343.
277. Doucet A, Katz AI. American Journal of Physiology. 1982; 242:F346–F352. [PubMed: 6175229]
278. Borke JL, Caride A, Verma AK, Penniston JT, Kumar R. Pflugers Archiv - European Journal of Physiology. 1990; 417:120–122. [PubMed: 1705698]
279. Wasserman RH, Smith CA, Brindak ME, De Talamoni N, Fullmer CS, Penniston JT, Kumar R. Gastroenterology. 1992; 102:886–894. [PubMed: 1311274]
280. Borke JL, Caride A, Verma AK, Kelley LK, Smith CH, Penniston JT, Kumar R. American Journal of Physiology. 1989; 257:c341–c346. [PubMed: 2475024]
281. Borke JL, Caride AJ, Yaksh TL, Penniston JT, Kumar R. Brain Research. 1989; 489:355–360. [PubMed: 2525946]
282. Wasserman RH, Smith CA, Smith CM, Brindak ME, Fullmer CS, Krook L, Penniston JT, Kumar R. Histochemistry. 1991; 96:413–418. [PubMed: 1660861]
283. Kumar R, Haugen JD, Medora M, Yamakawa K, Hruska KA. Journal of Bone & Mineral Research. 1993; 8:S386.
284. Cai Q, Chandler JS, Wasserman RH, Kumar R, Penniston JT. Proceedings of the National Academy of Sciences of the United States of America. 1993; 90:1345–1349. [PubMed: 7679502]
285. Wasserman RH, Chandler JS, Meyer SA, Smith CA, Brindak ME, Fullmer CS, Penniston JT, Kumar R. Journal of Nutrition. 1992; 122:662–671. [PubMed: 1311756]
286. Blankenship KA, Williams JJ, Lawrence MS, McLeish KR, Dean WL, Arthur JM. American Journal of Physiology - Renal Fluid & Electrolyte Physiology. 2001; 280:F815–F822.
287. Muller D, Hoenderop JG, Meij IC, van den Heuvel LP, Knoers NV, den Hollander AI, Eggert P, Garcia-Nieto V, Claverie-Martin F, Bindels RJ. Genomics. 2000; 67:48–53. [PubMed: 10945469]
288. Hoenderop JG, Nilius B, Bindels RJ. Annual Review of Physiology. 2002; 64:529–549.
289. Peng JB, Hediger MA. Current Opinion in Nephrology & Hypertension. 2002; 11:555–561. [PubMed: 12187321]
290. Weber K, Erben RG, Rump A, Adamski J. Biochemical & Biophysical Research Communications. 2001; 289:1287–1294. [PubMed: 11741335]
291. Murayama A, Takeyama K, Kitanaka S, Kodera Y, Kawaguchi Y, Hosoya T, Kato S. Endocrinology. 1999; 140:2224–2231. [PubMed: 10218975]
292. Shinki T, Ueno Y, DeLuca HF, Suda T. Proceedings of the National Academy of Sciences of the United States of America. 1999; 96:8253–8258. [PubMed: 10393981]
293. Adams ND, Gray RW, Lemann J Jr. Calcified Tissue International. 1979; 28:233–238. [PubMed: 228816]
294. Bilezikian JP, Canfield RE, Jacobs TP, Polay JS, D'Adamo AP, Eisman JA, DeLuca HF. New England Journal of Medicine. 1978; 299:437–441. [PubMed: 210383]
295. Boyle, IT.; Gray, RW.; Omdahl, JL.; DeLuca, HF. Calcium control of the in vivo biosynthesis of 1,25-dihydroxyvitamin D_3: Nicolaysen's endogenous factor. In: S, T., editor. Proceeding of the International Symposium on Endocrinology. London: Heinemann; 1972. p. 468-476.
296. Drezner MK, Neelon FA, Haussler M, McPherson HT, Lebovitz HE. Journal of Clinical Endocrinology & Metabolism. 1976; 42:621–628. [PubMed: 1083395]

297. Fraser DR, Kodicek E. Nature - New Biology. 1973; 241:163–166.
298. Garabedian M, Holick MF, Deluca HF, Boyle IT. Proceedings of the National Academy of Sciences of the United States of America. 1972; 69:1673–1676. [PubMed: 4340153]
299. Omdahl JL, Gray RW, Boyle IT, Knutson J, DeLuca HF. Nature - New Biology. 1972; 237:63–64.
300. Gray RW, Wilz DR, Caldas AE, Lemann J Jr. Journal of Clinical Endocrinology & Metabolism. 1977; 45:299–306. [PubMed: 885994]
301. Yoshida T, Yoshida N, Monkawa T, Hayashi M, Saruta T. Endocrinology. 2001; 142:1720–1726. [PubMed: 11316734]
302. Zhang MY, Wang X, Wang JT, Compagnone NA, Mellon SH, Olson JL, Tenenhouse HS, Miller WL, Portale AA. Endocrinology. 2002; 143:587–595. [PubMed: 11796514]
303. Kawashima H, Torikai S, Kurokawa K. Nature. 1981; 291:327–329. [PubMed: 7231553]
304. Bland R, Walker EA, Hughes SV, Stewart PM, Hewison M. Endocrinology. 1999; 140:2027–2034. [PubMed: 10218951]
305. Lund B, Sorensen OH, Bishop JE, Norman AW. Journal of Clinical Endocrinology & Metabolism. 1980; 50:480–484. [PubMed: 6892641]
306. Yoshida N, Yoshida T, Nakamura A, Monkawa T, Hayashi M, Saruta T. Journal of the American Society of Nephrology. 1999; 10:2474–2479. [PubMed: 10589684]
307. Adams ND, Gray RW, Lemann J Jr. Journal of Clinical Endocrinology & Metabolism. 1979; 48:1008–1016. [PubMed: 571872]
308. Sauveur B, Garabedian M, Fellot C, Mongin P, Balsan S. Calcified Tissue Research. 1977; 23:121–124. [PubMed: 196720]
309. Weber HP, Gray RW, Dominguez JH, Lemann J Jr. Journal of Clinical Endocrinology & Metabolism. 1976; 43:1047–1055. [PubMed: 993311]
310. Castillo L, Tanaka Y, DeLuca HF, Sunde ML. Archives of Biochemistry & Biophysics. 1977; 179:211–217. [PubMed: 843083]
311. Adams ND, Garthwaite TL, Gray RW, Hagen TC, Lemann J Jr. Journal of Clinical Endocrinology & Metabolism. 1979; 49:628–630. [PubMed: 573283]
312. Kumar R, Cohen WR, Epstein FH. New England Journal of Medicine. 1980; 302:1143–1145. [PubMed: 7366639]
313. Spanos E, Colston KW, Evans IM, Galante LS, Macauley SJ, Macintyre I. Molecular & Cellular Endocrinology. 1976; 5:163–167. [PubMed: 955248]
314. Eskildsen PC, Lund B, Sorensen OH, Bishop JE, Norman AW. Journal of Clinical Endocrinology & Metabolism. 1979; 49:484–486. [PubMed: 468984]
315. Gertner JM, Horst RL, Broadus AE, Rasmussen H, Genel M. Journal of Clinical Endocrinology & Metabolism. 1979; 49:185–188. [PubMed: 457839]
316. Menaa C, Vrtovsnik F, Friedlander G, Corvol M, Garabedian M. Journal of Biological Chemistry. 1995; 270:25461–25467. [PubMed: 7592714]
317. Condamine L, Menaa C, Vrtovsnik F, Vztovsnik F, Friedlander G, Garabedian M. Journal of Clinical Investigation. 1994; 94:1673–1679. [PubMed: 7929846]
318. Kumar, R.; Merimee, TJ.; Silva, P.; Epstein, FH. The effect of chronic growth hormone excess or deiciency on plasma 1,25-dihydroxyvitamin D levels in man. In: Norman, AW.; Schaefer, K.; von Herrath, D.; Grigoleit, HG.; Coburn, JW.; De Luca, HF.; Mawer, EB.; Suda, T., editors. Proceedings of the fourth workshop on Vitamin D. New York: de Gruyter, Elmsford; 1979. p. 1005-1009.
319. Bianda T, Glatz Y, Bouillon R, Froesch ER, Schmid C. Journal of Clinical Endocrinology & Metabolism. 1998; 83:81–87. [PubMed: 9435420]
320. Carre M, Ayigbede O, Miravet L, Rasmussen H. Proceedings of the National Academy of Sciences of the United States of America. 1974; 71:2996–3000. [PubMed: 4528777]
321. Favus MJ, Walling MW, Kimberg DV. Journal of Clinical Investigation. 1973; 52:1680–1685. [PubMed: 4718960]
322. Feher JJ, Wasserman RH. Endocrinology. 1979; 104:547–551. [PubMed: 221183]

323. Akeno N, Matsunuma A, Maeda T, Kawane T, Horiuchi N. Journal of Endocrinology. 2000; 164:339–348. [PubMed: 10694374]

324. Bouillon R, Muls E, De Moor P. Journal of Clinical Endocrinology & Metabolism. 1980; 51:793–797. [PubMed: 6893457]

325. Pahuja DN, De Luca HF. Archives of Biochemistry & Biophysics. 1982; 213:293–298. [PubMed: 6977339]

326. Jastrup B, Mosekilde L, Melsen F, Lund B, Sorensen OH. Metabolism: Clinical & Experimental. 1982; 31:126–132. [PubMed: 6978987]

327. Kumar R, Cohen WR, Silva P, Epstein FH. Journal of Clinical Investigation. 1979; 63:342–344. [PubMed: 429557]

328. Pike JW, Parker JB, Haussler MR, Boass A, Toverud SV. Science. 1979; 204:1427–1429. [PubMed: 451573]

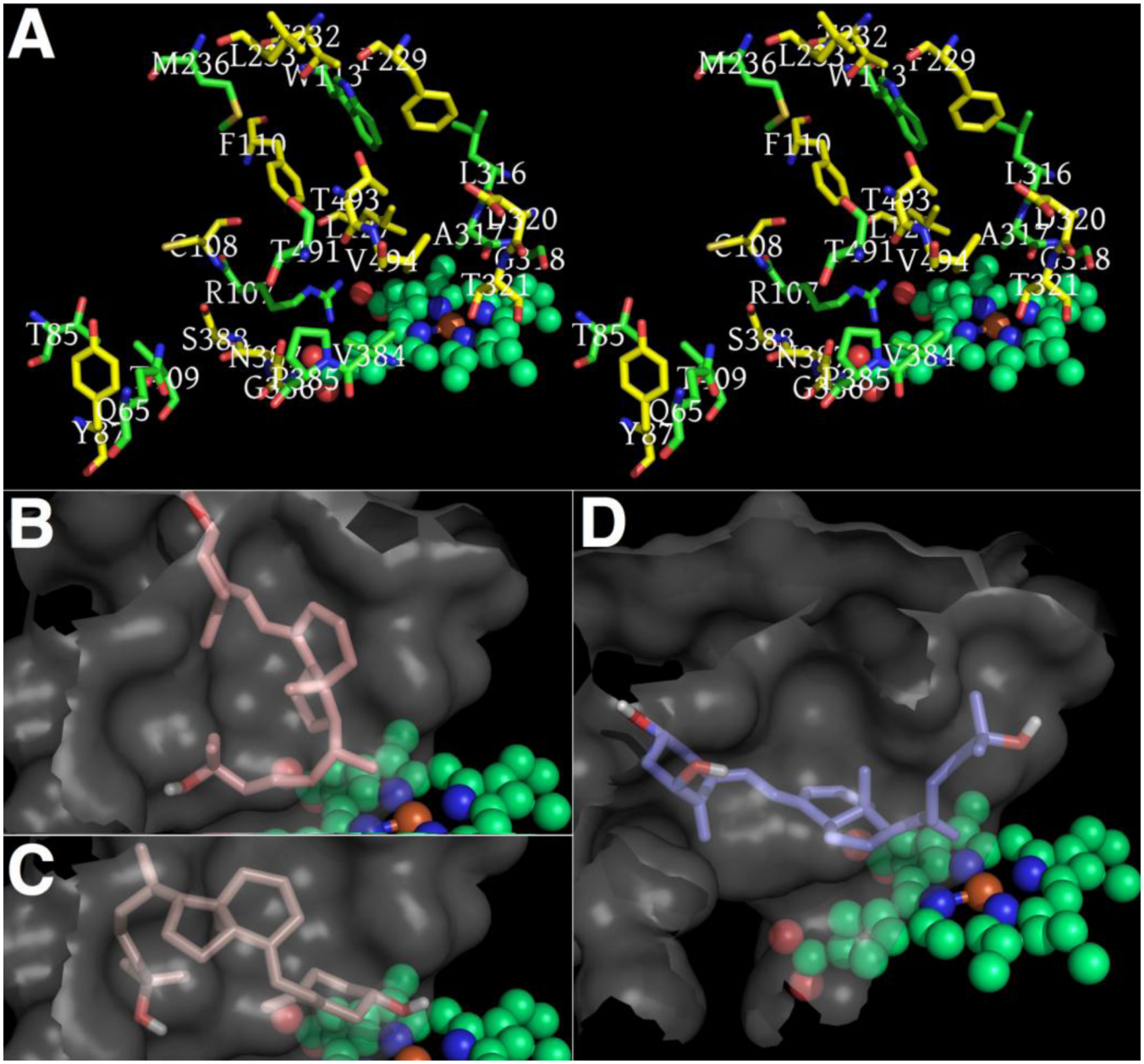

Figure 1.

A. Stereo view of the Cyp27B1 model's substrate cavity residues above the heme cofactor. **B.** Highest-scoring 25(OH)D_3 dockings in the Cyp27B1 homology model did not seem plausible, though possible binding modes, see **C**, were among the top 10 results. **D.** In contrast, top 3 and many lower scoring dockings of 1α,25(OH)$_2$$D_3$ inside the Cyp24A1 crystal structure seemed credible.

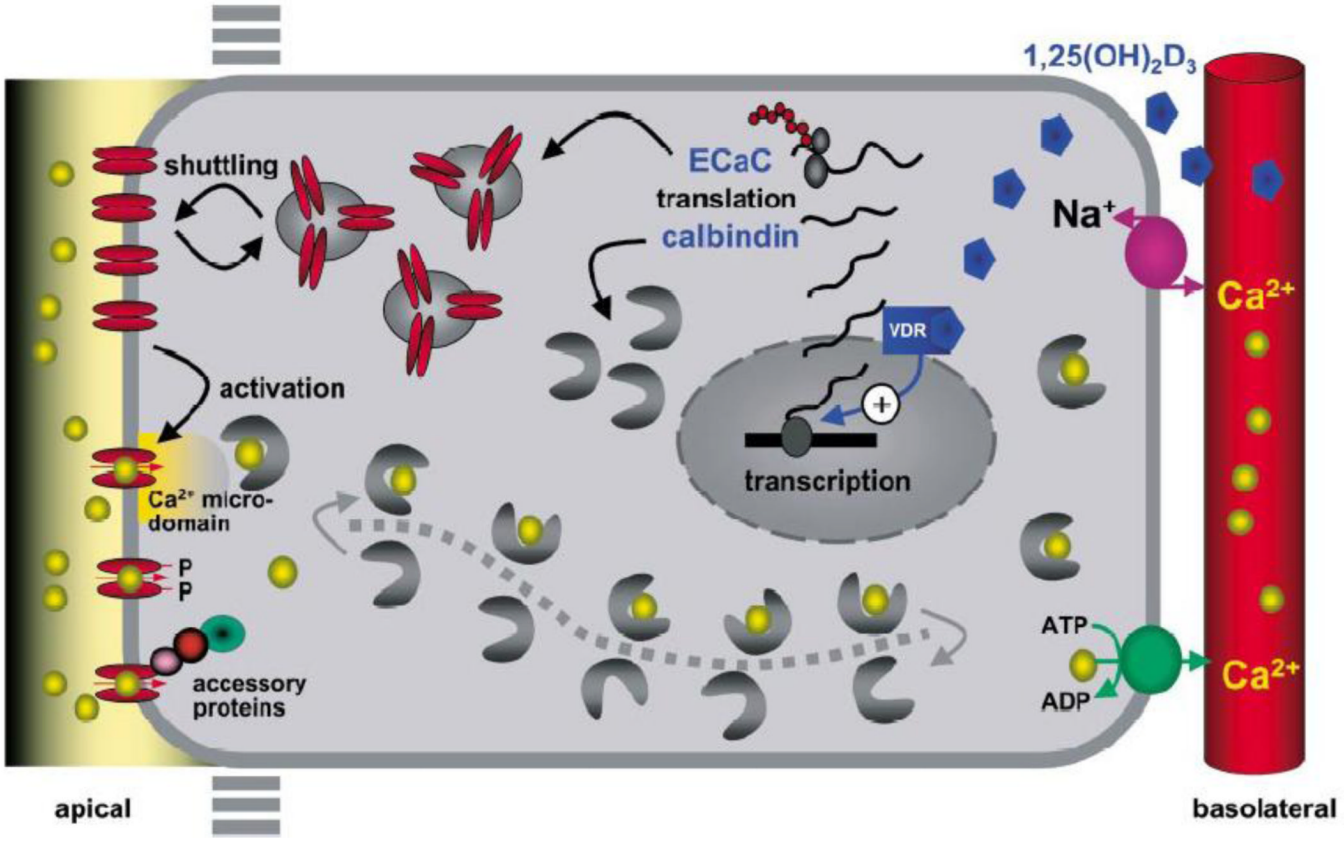

Figure 2.

Integrated model of active Ca^{2+} reabsorption in the distal part of the nephron. Apical entry of Ca^{2+} is facilitated by ECaC. Ca^{2+} then binds to calbindin-D_{28K}, and this complex diffuses through the cytosol to the basolateral membrane, where Ca^{2+} is extruded by a Na^{+}/Ca^{2+} exchanger and a plasma membrane Ca^{2+}-ATPase. The individually controlled steps in the activation process of the rate limiting Ca^{2+} entry channel include 1α,25$(OH)_2D_3$-mediated transcriptional and translational activation, shuttling to the apical membrane, and subsequent activation of apically located channels by ambient Ca^{2+} concentration, direct phosphorylation and/or accessory proteins. (With permission from Hoenderop, et al [268])

Table 1

Factors that alter renal phosphate excretion

Increase	Decrease
1. High phosphate diet	1. Low-phosphate diet
2. Parathyroid hormone	2. Parathyroidectomy
3. Calcitonin	3. Thyroxine
4. Chronic vitamin D	4. Acute vitamin D
5. Glucagon	5. Insulin
6. Glucocorticoids	6. Growth hormone
7. Volume expansion	7. Volume contraction
8. Increased pCO_2	8. Decreased pCO_2
9. Chronic Acidosis	
10. Starvation	
11. Diuretics	
12. “Phosphatonin”	
FGF-23	
sFRP-4	

TABLE 2

Effect of increased level or activity of various factors on 1α,25$(OH)_2D_3$ concentration or 1α-hydroxylase activity

Factor	Animals	Humans	Ref.
Parathyroid hormone	↑	↑	[27, 75, 291–299]
Serum inorganic phosphorus	↓	↓	[127, 300–302]
1α,25$(OH)_2D_3$	↓	↓	[291, 303]
Calcium (direct)	?	↓	[304, 305]
Calcitonin	↑,↓,0	↑	[24, 75, 291, 292, 306, 307]
Hydrogen ion	↓	0	[293, 308, 309]
Sex steroids	↑	↑	[126, 310]
Prolactin	↑	0	[311–313]
Growth hormone and insulin-like growth factor-1	↑	↑,↓,0	[225, 305, 314–319]
Glucocorticoids	↓,0	↑,↓,0	[183, 320–323]
Thyroid hormone	?	↓*	[324–326]
Fibroblast growth factor 23/Klotho axis	↓	?	[81–93]
Frizzled related protein 4	↓	?	[23]
Pregnancy	↑	↑*	[327, 328]

↑ Stimulation or increase; ↓, suppression or decrease; 0, no effect; ?, effect not known.

* Effects may be secondary to changes in calcium, phosphorus or parathyroid hormone. (With permission, modified from Kumar R[94])

Table 3

The metabolism of $25OHD_3$ by the kidney

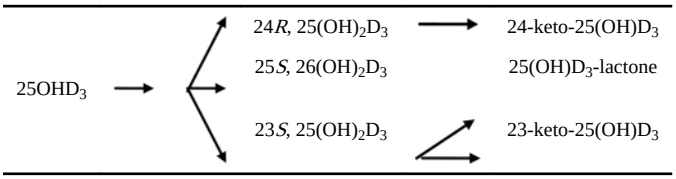

Table 4

Vitamin D responsive proteins in the kidney

Vitamin D receptor
24-hydroxylase
Plasma membrane calcium pump
Epithelial calcium channels, TRPV5 and TRPV6
Calbindin D_{28K}
Calbindin D_{9K}
Calcium sensing receptor
PTH/PTHrp receptor
Sodium-phosphate co-transporter type 2

TABLE 5

Effect of dietary calcium on unoccupied VDR content in rat duodenum and kidney

		Day			
	Diet	**2**	**7**	**14**	**21**
VDR					
Duodenum	1% calcium	341±26	197±17	202±17	259±26
	0.02% calcium	365±27	226±28	221±16	267±28
Kidney	1% calcium	ND	163±11	165±9	124±8
	0.02% calcium	ND	120±4 *	131±10 *	77±3 *
Calcium (mg/dL)	1% calcium	9.98±0.18	9.82±0.08	9.16±0.16	9.52±0.41
	0.02% calcium	9.45±0.12	9.15±0.17 *	9.09±0.14	8.70±0.29 *
Phosphorus (mg/dL)	1% calcium	8.40±0.17	8.88±0.22	8.73±0.17	7.92±0.29
	0.02% calcium	8.15±0.22	8.65±0.13	8.37±0.28	8.41±0.23
1α, 25-$(OH)_2D_3$ (pg/mL)	1% calcium	153±11	113±13	139±9	160±32
	0.02% calcium	180±20	392±45 **	682±44 **	829±59 **

Values are mean ± SEM.

Unoccupied vitamin D receptor content expressed as fmols [^{3}H]1α,25$(OH)_2D_3$ bound per mg cytosol protein.

* P<0.05;

** P<0.001.

ND = not done.

Modified from Goff JP, et al[224]

Table 6

Distribution of plasma membrane calcium pump in transporting epithelia as assessed by immunohistochemistry

Tissue	Source	Cell Type	Location in Cell	Reference
Kidney	Rat, human	Distal convoluted tubule, principal cell	Basolateral	[14, 15, 28, 203, 204]
Intestine	Rat, chick	Absorptive cell	Basolateral	[278, 279]
Trophoblast	Rat, human	Syncytiotrophoblast	Basal	[280]
Choroid plexus	Cat Human	Choroid plexus Secretory cell	Apical	[281]
Shell gland	Chick	Principal cell	Apical	[282]
Bone	Human	Osteoblast	Not vectorially oriented	[13]
Bone	Chick	Osteoclast	Not vectorially oriented	[283]