Archives of Biochemistry and Biophysics 523 (2012) 73–76

Contents lists available at SciVerse ScienceDirect

Archives of Biochemistry and Biophysics

journal homepage: www.elsevier.com/locate/yabbi

Review

Recent advances in our understanding of 1,25-dihydroxyvitamin D_3 regulation of intestinal calcium absorption

Sylvia Christakos *

Department of Biochemistry and Molecular Biology, University of Medicine and Dentistry of New Jersey, New Jersey Medical School, Newark, NJ 07103, United States

ARTICLE INFO

Article history:
Available online 2 January 2012

Keywords:
Intestine
Transport
Calcium
1,25-Dihydroxyvitamin D_3
Transient receptor potential vanilloid type 6 (TRPV6)
Calbindin
Plasma membrane calcium pump PMCA1b
Claudin-2
Claudin-12
Cadherin-17

ABSTRACT

Calcium is required for many cellular processes including muscle contraction, nerve pulse transmission, stimulus secretion coupling and bone formation. The principal source of new calcium to meet these essential functions is from the diet. Intestinal absorption of calcium occurs by an active transcellular path and by a non-saturable paracellular path. The major factor influencing intestinal calcium absorption is vitamin D and more specifically the hormonally active form of vitamin D, 1,25-dihydroxyvitamin D_3 ($1,25(OH)_2D_3$). This article emphasizes studies that have provided new insight related to the mechanisms involved in the intestinal actions of $1,25(OH)_2D_3$. The following are discussed: recent studies, including those using knock out mice, that suggest that $1,25(OH)_2D_3$ mediated calcium absorption is more complex than the traditional transcellular model; evidence for $1,25(OH)_2D_3$ mediated active transport of calcium by distal as well as proximal segments of the intestine; $1,25(OH)_2D_3$ regulation of paracellular calcium transport and the role of $1,25(OH)_2D_3$ in protection against mucosal injury.

Introduction

1,25-Dihydroxyvitamin D_3 ($1,25(OH)_2D_3$)[1], the hormonally active form of vitamin D, is the most significant factor controlling intestinal calcium absorption [1,2]. Two modes of intestinal calcium absorption have been proposed: one is a saturable, active, transcellular process and the other mode is non-saturable, paracellular, occurs at luminal concentrations of calcium >2–6 mmol/l and is driven, at least in part, by the integrity of tight junctions [3,4]. Calcium absorption occurs in each segment of the small intestine and is determined by the rate of absorption and the transit time through each segment. In mammals most of the ingested calcium is absorbed in the lower segments of the small intestine (mostly in the ileum) where the transit time is longer compared to more proximal segments. Only about 8–10% of calcium absorption takes place in the mammalian duodenum although its capacity to absorb calcium is more rapid than any other segment [3–7]. As the body's demand for calcium increases as a result of a diet deficient in calcium or due to growth, pregnancy or lactation, synthesis of $1,25(OH)_2D_3$ is increased resulting in the stimulation of active calcium transport [1,2]. Although the duodenum has been a focus of research related to $1,25(OH)_2D_3$ mediated active calcium absorption, $1,25(OH)_2D_3$ regulation of calcium absorption in the ileum, cecum and colon has also been reported [8–14]. The vitamin D receptor (VDR) is expressed in all segments of the small and large intestine [15–17] and two targets of vitamin D (the calcium binding protein calbindin-D_{9k} and the epithelial calcium channel TRPV6) are present in all segments of mouse and rat intestine [18,19]. Thus, although calcium is absorbed most rapidly in the duodenum compared to other segments, $1,25(OH)_2D_3$ action in the distal segments of the intestine also contributes to the calcium absorptive process.

$1,25(OH)_2D_3$ regulated transcellular intestinal calcium absorption

Transcellular calcium transport is believed to be comprised of three $1,25(OH)_2D_3$ regulated steps: (1) the entry of calcium from the intestinal lumen across the brush border membrane; (2) the transcellular movement of calcium through the cytosol of the enterocyte; and (3) the energy requiring extrusion of calcium against a concentration gradient at the basolateral membrane [3,4]. It has been reported that only 2–4 h after $1,25(OH)_2D_3$ treatment to vitamin D deficient or normal animals is overall intestinal calcium absorption significantly increased ([20]; Fig. 1).

* Address: UMDNJ-New Jersey Medical School, Department of Biochemistry and Molecular Biology, 185 South Orange Ave., Newark, NJ 07193, United States. Fax: +1 973 972 5594.
E-mail address: christak@umdnj.edu

[1] *Abbreviations used:* $1,25(OH)_2D_3$, 1,25-dihydroxyvitamin D_3; TRPV6, transient receptor potential vanilloid type 6; VDR, vitamin D receptor; KO, knock out; WT, wild type.

0003-9861/$ - see front matter © 2011 Elsevier Inc. All rights reserved.
doi:10.1016/j.abb.2011.12.020

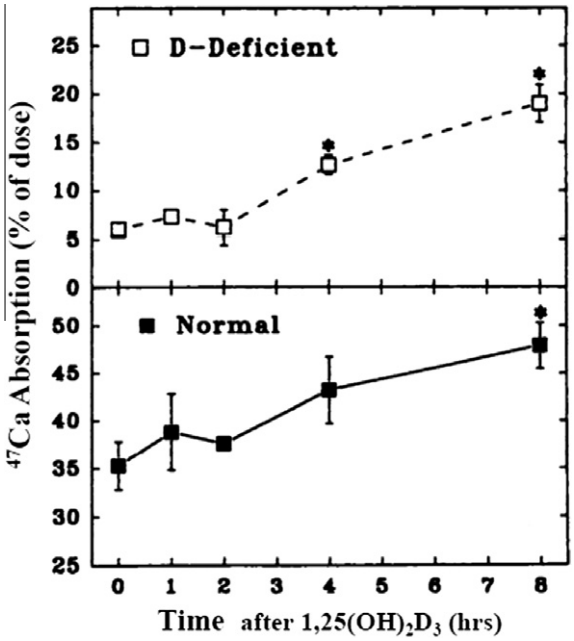

Fig. 1. Time dependent effect of 1,25(OH)$_2$D$_3$ on duodenal absorption of ^{47}Ca in vitamin D deficient (top panel) and vitamin D replete (bottom panel) chicks. Chicks were injected (i.v.) with 1ug 1,25(OH)$_2$D$_3$ and duodenal calcium absorption was measured by the *in situ* ligated loop procedure. * Significant difference from zero time controls ($p < 0.05$). (from Wasserman [3] with permission)

Although early studies noted that calcium uptake by brush border membrane vesicles isolated from vitamin D replete animals is enhanced compared to uptake by vesicles isolated from vitamin D deficient animals [21–23], the molecular basis of vitamin D dependent calcium entry was not known. In 1999 the apical epithelial calcium channel TRPV6 was cloned from rat duodenum, suggesting one mechanism of calcium entry [24]. TRPV6 is a membrane protein containing six transmembrane domains, a putative N-linked glycosylation site and a long C terminal tail [24,25]. TRPV6 is expressed in villi tips but not in villi crypts and is strongly calcium selective [25]. Calmodulin associates with the C terminal end of TRPV6 enabling acceleration of the rate of TRPV6 current inactivation [26]. Association of TRPV6 with S100A10-annexin 2 complex and Rab11a has been reported to be required for targeting and retention of TRPV6 to the plasma membrane and recycling of TRPV6, respectively [27,28]. These TRPV6 associated proteins may represent additional components of regulated calcium entry into the intestinal cell. TRPV6 and the calcium binding protein calbindin-D$_{9k}$ are colocalized in the intestine and both are induced by 1,25(OH)$_2$D$_3$, under low calcium conditions and at weaning [29]. In VDR KO mice, TRPV6 mRNA is reduced in the duodenum by more than 90% and there is a 50% reduction in calbindin-D$_{9k}$ mRNA [30,31]. These studies provide indirect evidence for a role of TRPV6 and calbindin-D$_{9k}$ in 1,25(OH)$_2$D$_3$ regulated intestinal calcium absorption. However, studies in TRPV6 knock out (KO) mice as well as in calbindin-D$_{9k}$ KO mice indicate that active calcium transport still occurs in these mice, suggesting compensation by other calcium channels and other calcium binding proteins [32–34]. Although TRPV6 may be redundant, transgenic mice overexpressing TRPV6 in the intestine have been reported to develop hypercalcemia, hypercalciuria and soft tissue calcification, indicating a direct role for TRPV6 in the process of intestinal calcium transport [35]. Transgenic expression of TRPV6 is accompanied by an increase in calbindin-D$_{9k}$ [35]. In addition, unlike TRPV6 or calbindin-D$_{9k}$ single KO mice which transport calcium in response to 1,25(OH)$_2$D$_3$ similar to wild type (WT) mice, 1,25(OH)$_2$D$_3$ mediated intestinal calcium transport is reduced by 60% in TRPV6/calbindin-D$_{9k}$ double KO mice [32]. Findings in the transgenic mice as well as in the double KO mice suggest that TRPV6 and calbindin-D$_{9k}$ can act together in certain aspects of the intestinal absorptive process. Early studies (prior to the identification of TPRV6) showed that a significant fraction of total intestinal calbindin is associated with intestinal brush borders and binds to "a specific protein" [36]. It is indeed possible, but has not as yet been investigated, that calbindin-D$_{9k}$ binds to TRPV6 and that a principal function of calbindin is as a modulator of TRPV6 calcium influx (facilitating high calcium transport rates by preventing calcium dependent inactivation). Only recently has it been reported that calcium binding proteins in addition to calmodulin can bind to calcium channels, indicating differential adjustment of calcium influx by different calcium binding proteins [37,38]. Calbindin-D$_{9k}$ may also act as a calcium buffer preventing toxic levels of calcium from accumulating in the intestinal cell. With regard to calcium in the cytosol, calcium may be bound to calbindin as well as to other calcium binding proteins. Calcium may also be sequestered by the endoplasmic reticulum and then could be released in the proximity of the basolateral membrane. At the basolateral membrane calcium is extruded by the intestinal plasma membrane ATPase (PMCA1b) (Fig. 2). Previous studies have shown that 1,25(OH)$_2$D$_3$ and low calcium stimulate the synthesis of PMCA1b [39,40]. Although it has been suggested that the sodium calcium exchanger present at the intestinal basolateral membrane (isoform NCX1) may also play a role in calcium extrusion, this cotransporter, at least in rat intestine, is not regulated by 1,25(OH)$_2$D$_3$ [41].

Effect of other hormones (estrogen, prolactin, glucocorticoids) and the effect of aging on active intestinal calcium transport

During pregnancy and lactation active intestinal calcium transport is increased [42–44]. It has been reported that estrogens and prolactin, independent of vitamin D, stimulate active intestinal calcium transport [45,46]. Mechanisms include induction of TRPV6 by estrogen via estrogen receptor α and induction of TRPV6 by prolactin [45,47]. Prolactin also has cooperative effects with 1,25(OH)$_2$D$_3$

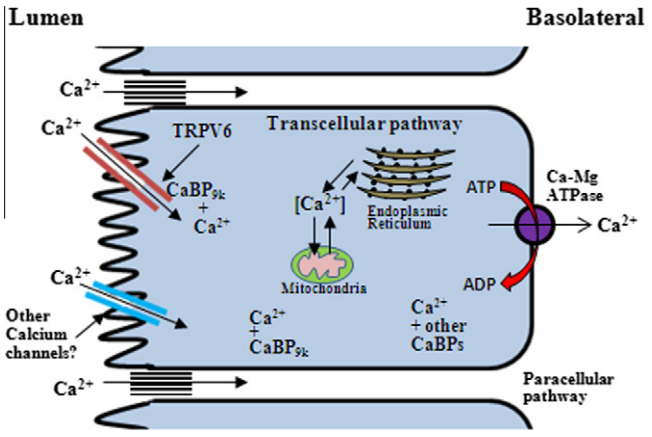

Fig. 2. Model of vitamin D mediated intestinal calcium absorption. The traditional model of transcellular calcium transport consists of influx through an apical calcium channel (TRPV6), diffusion through the cytosol and active extrusion at the basolateral membrane by the plasma membrane ATPase (PMCA1b). Although entry of calcium has been reported to involve TRPV6, other calcium channels may also be involved. Calcium binding proteins including calmodulin and calbindin-D$_{9k}$ (CaBP$_{9k}$) may be important for fine tuning calcium channel activity. In the cytosol calcium may be bound to calbindin as well as other calcium binding proteins. Calbindin as well as other calcium binding proteins may act to prevent toxic levels of calcium from accumulating in the cell. Calcium may also be sequestered by intracellular organelles i.e. endoplasmic reticulum and mitochrondria which could also contribute to the protection of the cell against excessively high calcium. Increasing evidence supports regulation by 1,25(OH)$_2$D$_3$ of paracellular calcium transport.