The importance of vitamin D in the pathology of bone metabolism in inflammatory bowel diseases

Iwona Krela-Kaźmierczak[1], Aleksandra Szymczak[1], Liliana Łykowska-Szuber[1], Piotr Eder[1], Kamila Stawczyk-Eder[1], Katarzyna Klimczak[1], Krzysztof Linke[1], Wanda Horst-Sikorska[2]

[1]Department of Gastroenterology, Human Nutrition and Internal Diseases, Poznan University of Medical Sciences, Poznan, Poland
[2]Department of Family Medicine, Poznan University of Medical Sciences, Poznan, Poland

Corresponding author:
Piotr Eder MD, PhD
Department of
Gastroenterology,
Human Nutrition
and Internal Diseases
Poznan University
of Medical Sciences
49 Przybyszewskiego St
60-355 Poznan, Poland
Phone: +48 698 050 797
E-mail: piotr.eder@op.pl

Abstract

Etiological factors of bone metabolism disorders in inflammatory bowel diseases have been the subject of interest of many researchers. One of the questions often raised is vitamin D deficiency. Calcitriol acts on cells, tissues and organs through a vitamin D receptor. The result of this action is the multi-directional effect of vitamin D. The reasons for vitamin D deficiency are: decreased exposure to sunlight, inadequate diet, inflammatory lesions of the intestinal mucosa and post-gastrointestinal resection states. This leads not only to osteomalacia but also to osteoporosis. Of significance may be the effect of vitamin D on the course of the disease itself, through modulation of the inflammatory mechanisms. It is also necessary to pay attention to the role of vitamin D in skeletal pathology in patients with inflammatory bowel diseases and thus take measures aimed at preventing and treating these disorders through the supplementation of vitamin D.

Key words: vitamin D, Crohn's disease, ulcerative colitis, osteoporosis, osteopenia.

Introduction

Non-specific inflammatory bowel diseases (IBD), which include Crohn's disease (CD) and ulcerative colitis (UC), have become a significant problem in recent years due to the significant increase in morbidity. Typically, these are diseases of the gastrointestinal tract, but many patients experience extra-intestinal symptoms, among which bone mineralisation disorders and decrease in bone mass are of major clinical importance. The incidence of low bone mineral density in patients with IBD is currently estimated at 31–59% [1]. The pathogenesis of these disorders appears to be multifactorial, and vitamin D and calcium deficiencies, chronic inflammatory processes and the use of glucocorticoids have been mentioned as the main causes [2–4].

The aim of this systematic review was to present the influence of vitamin D on bone metabolism in IBD. The following databases were searched: MEDLINE, PubMed from 1980 to 2013. We reviewed papers describing the environmental, genetic and epidemiologic association of vitamin D and IBD and bone metabolism using the following keywords:

AMS

"osteoporosis in inflammatory bowel diseases", "vitamin D in inflammatory bowel diseases", "bone metabolism in inflammatory bowel disease". Both human and animal studies published were examined.

Metabolism of vitamin D$_3$

Vitamin D belongs to a heterogeneous group of chemical compounds, which includes, among others, ergocalciferol (vitamin D$_2$) and cholecalciferol (vitamin D$_3$). The main sources of vitamin D in humans are cutaneous synthesis under the influence of sunlight and through food supply [5]. During exposure to sunlight, 7-dehydrocholesterol present in the skin absorbs UVB radiation with a wavelength of 290–315 nm and is converted to previtamin D$_3$, which is thermodynamically unstable and rapidly isomerises at body temperature (37°C) to vitamin D$_3$ [6]. This transformation takes place during the first 30 min of solarisation, after which biologically inactive compounds are formed. The skin synthesis is affected by many factors, including the angle of incidence of the sun rays (latitude, season and time of day), the thickness of the ozone layer, the skin melanin content, age, body weight and the use of protective creams with anti-UVB filter [7–10]. From the skin, vitamin D is transported to the liver, bound with vitamin D binding protein (DBP). Vitamin D is found only in a few food products; the major sources are oily fish and cod-liver oil. In many countries, vitamin D is used for the supplementation of generally available food products such as breakfast cereals, bread, margarine, milk and orange juice [1, 11]. The absorption of vitamin D contained in food takes place in the jejunum and terminal ileum. From the enterocytes, vitamin D passes into the circulatory system, and then to the liver, where hydroxylation takes place at position 25 of the side chain, leading to the formation of 25-hydroxyvitamin D. This transformation is catalysed by the cytochrome P450 group of enzymes. The hepatic metabolite is then converted in the kidneys to the more active form of vitamin D [1,25(OH)$_2$D] and to [24,25(OH)$_2$D]. The whole process is regulated by the respective hydroxylases (CYP27B1), the actions of which are dependent on the parathyroid hormone, 1,25(OH)$_2$D$_3$, calcium and phosphorus. CYP27B1 expression takes place also in the bone tissue, lungs, liver, skin and macrophages [12].

Vitamin D$_3$ deficiency in patients with inflammatory bowel diseases

Vitamin D deficiencies seem to be a worldwide problem. It is estimated that up to a billion people may have significantly reduced levels of serum 25(OH)D. This is the most accurate functional indicator used to evaluate the degree of the body demand for vitamin D, due to its long half-life and the dependence of its concentration on cutaneous synthesis and intestinal absorption [13]. A 25(OH)D concentration of less than 30 ng/ml in adults is regarded as a deficiency. According to the most recent figures, 22–70% of patients with CD and approximately 45% of patients with UC have vitamin D deficiency [14]. Low levels of vitamin D have also been found in patients with new onset IBD. The data show that as many as 10.9% of patients may have severe vitamin D deficiency (< 10 ng/dl) [15]. The main reasons for vitamin D deficiency in IBD have been given as: decreased exposure to sunlight, inadequate diet, smoking, increased disease activity, inflammatory lesions of the intestinal mucosa and post-gastrointestinal resection states. Researchers have reported significantly reduced levels of vitamin D in patients in the northern hemisphere especially in the winter months, while other publications tend to also postulate the tendency to reduced exposure to the sunlight during the summer months compared to healthy subjects [16]. Patients with IBD tend to avoid foods rich in vitamin D (fatty fish and to a lesser extent dairy products), which may aggravate their symptoms. Patients with lesions in the proximal part of the small intestine have difficulties digesting lactose and therefore often avoid milk and milk products. Patients with IBD often have disorders of the digestion and absorption of lipids, and therefore vitamin D. States of postintestinal resection interfere with the enterohepatic circulation of lipid compounds. The location of inflammatory lesions in the terminal ileum in CD also inhibits the absorption of vitamin D [12]. Inflamed intestinal mucosa predisposes to the loss of proteins, including DBP [17]. In healthy subjects, the main causes of deficiencies are reduced cutaneous synthesis and insufficient intake of vitamin D in food products and diet supplements.

The action of vitamin D is mediated by the vitamin D receptor (VDR) [18]. It is a class II nuclear receptor and shows expression in more than 30 different types of cells and tissues. Vitamin D receptor with a molecular weight of 48 kDa consists of an N-terminal DNA-binding domain with zinc fingers and a C-terminal ligand binding domain, which is formed from α-helices. After the binding of calcitriol with VDR, there occurs a heterodimerisation with the retinoid X receptor (RXR). The next stage is the binding with regulatory proteins (activating or inhibiting) in promoters of target genes, resulting in regulation of the transcription of these genes [19, 20]. Vitamin D receptor activates osteocalcin, osteopontin, calbindin and RANKL genes, among others, and inhibits parathormone (PTH), interleukin-2 (IL-2), IL-12, interferon (IFN)-γ and

nuclear factor (NF)-κB genes [21]. The VDR gene is located on chromosome 12. Literature data contain five known polymorphisms of this gene – BsmI, ApaI, TaqI, FokIa and poly A. Four of them (excluding poly A) play an important role in bone mineral density, osteoporosis, and the occurrence of autoimmune diseases [22]. The presence of TaqI tt alleles in European populations is associated with a higher risk of the occurrence of CD [23].

Effect of vitamin D on the bone

Vitamin D acts directly and indirectly on the bone. It directly acts on osteoblasts, osteocytes and osteoclasts. $1,25(OH)_2D$ regulates the transcription of genes as well as the differentiation and mineralisation of osteoblasts. It has an effect on the production of type-I collagen and numerous non-collagenous proteins (osteocalcin, osteopontin, bone sialoprotein BSP1). Osteocalcin and osteoopontin are responsible for the binding of calcium and hydroxyapatite, thereby contributing to their deposition in the bone matrix. Sialoprotein also contributes to the organisation of the bone matrix [24]. Under the influence of vitamin D, osteocytes produce FGF23 – phosphatonin, which regulates the renal hydroxylation of $1,25(OH)_2D_3$, inhibits the reabsorption of phosphates and reduces the synthesis of PTH. Vitamin D is also an essential factor necessary for the proper differentiation and functioning of osteoclasts. It stimulates the preosteoblasts to secrete the macrophage-colony stimulating factor (M-CSF), which in turn activates the preosteoclasts to proliferation and prevents their apoptosis. RANK found on the membrane of preosteoclasts bind with RANKL released by preosteoblasts. This results in activation of the differentiation pathway and activation of osteoclasts, leading to the formation of a mature cell. Osteoprotegerin protein (OPG) from the osteoblasts may bind with RANKL and inhibit its binding with RANK, thereby inhibiting the maturation pathway of the osteoclast. Vitamin D causes an increase in the expression of RANKL in osteoblasts and inhibits the expression of OPG, thereby stimulating osteoclastogenesis [25].

Vitamin D has indirect action on bones by regulating the calcium and phosphate metabolism of the body. The main organs involved in this regulation are the intestine and kidneys. Vitamin D acts on the intestinal epithelium in a number of ways, each of which stimulates the process of calcium resorption from the intestines. The absorption of calcium from the intestine takes place on the basis of transcellular and paracellular transport. Transcellular transport dominates in the proximal section of the small intestine and paracellular transport in the jejunum and the terminal section of the small intestine (locations characteristic of CD). Vitamin D regulates the transcellular transport processes, enhancing the expression of the epithelial calcium receptor (TRPV6), calbindin and calcium binding protein (CaBP). The more calcium there is in the intestinal lumen, the greater is the synthesis of CaBP. Paracellular transport takes place based on changes in the permeability of the cell membrane of the enterocyte for calcium ions. $1,25(OH)_2D$, interacting with PTH, stimulates the reabsorption of calcium and phosphate in the kidneys, from the tubular fluid into the blood; it also has a stimulating effect on the activity of 24-hydroxylase, and an inhibitory effect on the activity of 1α-hydroxylase [12].

Vitamin D and the immune system in inflammatory bowel diseases

Many clinical studies have demonstrated the crucial role of vitamin D as a modifiable environmental factor of autoimmune diseases. It has an effect on the immune cells, which include T-lymphocytes, B-lymphocytes and dendritic cells, among others [26]. Each of these cell types demonstrates VDR expression, and produces 1α-hydroxylase and 24-hydroxylase, which has an effect on the local production of active $1,25(OH)_2D$. Its autocrine and paracrine functions remain under the strict control of the immune system and depend on an adequate supply of circulating 25(OH)D.

T helper (Th) lymphocytes play a key role in the antigen-specific immune response. There are two populations of these lymphocytes: Th1 and Th2 cells, which during the "normal" immune response remain in balance. Th1 cells secrete IFN-γ, IL-1 and the tumour necrosis factor (TNF), and their activation is characteristic of the host response to the existence of a tumour or intracellular pathogens such as viruses. Th2 lymphocytes secrete IL-4, IL-5, IL-10 and IL-13. The signal for their stimulation is the presence of extracellular pathogens – bacteria or parasites [27]. $1,25(OH)_2D$ acts on both populations of lymphocytes, and the activation of CD4+ lymphocyte causes a five-fold increase in the expression of VDR [28]. Vitamin D inhibits the proliferation of Th1 lymphocytes and reduces the production of IFN-γ and IL-2, and by acting on the Th2 phenotype enhances the expression of IL-4. In autoimmune diseases, Th1 cells are directed against the host proteins, resulting in the occurrence of pathological symptoms [29]. Many *in vivo* studies have shown the effectiveness of $1,25(OH)_2D$ in suppressing immune responses dependent on the secretion of IL-2 and IL-4 [30]. Th17 cells are a subpopulation of CD4+ lymphocytes, which play a significant role in inflammation and which may lead to tissue damage; their role in the pathogenesis of CD is currently being widely discussed. Vitamin D inhibits the autoimmune response and

thus the destruction of tissues by inhibiting the secretion of interleukins by the Th17 cells [31, 32]. $1,25(OH)_2D$ acting on mononuclear cells also inhibits the production of IL-1, IL-6 and IL-12, which are among the most potent proinflammatory cytokines, as well as stimulating the expression of anti-inflammatory compounds: cytotoxic T-lymphocyte antigen 4 (CTLA-4), FoxP3, IL-10 and TGF-β by regulatory lymphocytes (Treg). Vitamin D also has a direct effect on lymphocyte B cells, interfering with their process of differentiation and the production of immunoglobulins. Calcitriol has an effect on the maturation of dendritic cells by reducing the expression of the costimulatory molecules – CD40, CD80, CD86 and MHC class II proteins [26, 33]. In both types of IBD, there are abnormalities in the regulation of T helper lymphocytes. Inflammatory infiltrates in IBD are dominated by Th1 lymphocytes. An impaired local immune response to bacterial antigens in genetically predisposed patients has also been mentioned as one of the etiological factors in IBD. Vitamin D stimulates the transformation of Th1 phenotype to the Th2 form. A groundbreaking study by Cantorna *et al.* showed that the active metabolite of vitamin D prevents and relieves the symptoms of IBD in mice lacking IL-10 or VDR [34, 35]. In patients with CD, vitamin D deficiency is, therefore, not only a consequence but also a cause of inflammation, which may lead to loss of bone mass through an immune response mediated by Th1 lymphocytes. A negative effect of interleukins on bone mass through the stimulation of osteoclast activity has also been postulated [36]. Tilg *et al.* have shown that pro-inflammatory cytokines (TNF-α, IL-1, IL-6 and IFN-γ), which participate in inflammatory processes in the intestines, can also cause bone erosion and the loss of bone mass, and that these effects are exerted by osteoclasts [37].

Effect of vitamin D on bone mineral density

There are a few studies concerned with the relationship between serum 25(OH)D and bone mineral density (BMD) in patients with IBD. The results of these reports vary – some researchers state that there is a relationship between vitamin D level and bone mineral density in adults [38, 39], while other researchers do not find such a relationship – patients who have low BMD in the spine, measured using the DEXA method, have normal vitamin D levels [40].

Summary

Vitamin D is undoubtedly of great importance in IBD: it may affect their pathogenesis, course and the condition of bones in patients. There is still an unsatisfactory explanation of the mechanisms of these actions. The results of future stud-

ies should help to fully define the role of vitamin D in bone metabolism in patients with IBD and be the starting point in establishing guidelines for the supplementation of its possible deficiencies in prophylaxis and treatment.

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Conflict of interest

The authors declare no conflict of interest.

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