® Postepy Hig Med Dosw (online), 2020; 74: 191-197
e-ISSN 1732-2693

www.phmd.pl
Review

Received: 27.06.2019
Accepted: 07.11.2019
Published: 09.06.2020

Anticancer activity of vitamin D – molecular mechanisms

Mechanizmy przeciwnowotworowej aktywności witaminy D

Beata M. Gruber-Bzura

Department of Biochemistry and Biopharmaceuticals, National Medicines Institute, Warsaw, Poland

Summary

A large number of studies have pointed to the relations between blood levels of 25-hydroxy vitamin D with cancer incidence and survival. The phenomenon of the multidirectional activity of vitamin D is possibly due to the presence of VDR in most nonskeletal human cells, including cancer cells. A wide range of the genes regulated by VDR are related with cell proliferation, apoptosis, differentiation, angiogenesis and metastasis. In some preclinical studies, colon, lung and BC have all demonstrated downregulation of VDR expression as compared to normal cells, and well-differentiated tumors have shown more VDR expression when compared to their poorly differentiated counterparts. Generally, higher tumor VDR expression has been noted as correlating with better prognosis in cancer patients. However, vitamin D pathway genetic polymorphisms also may influence cancer risk. *VDR* polymorphisms have received the most attention, but this influence has also been observed in genes related to vitamin D metabolism or signalling, such as: *CYP27B1*, *CYP24A1*, *VDBP* or *RXRA*. Even though the associations between most of them and cancers were not significant, some studies show that *VDR* polymorphisms may be a better or poor prognostic factor to assess the risk of cancer. The aim of this paper was to present the molecular pathways affected by vitamin D, which are included in carcinogenesis. The literature survey comprised of research compiled from mostly the last five years and it proves vitamin D as the most phenomenal among other vitamins.

Keywords: **vitamin D • anticancer activity • vitamin D pathway genetic polymorphisms**

GICID 01.3001.0014.1882
DOI: 10.5604/01.3001.0014.1882
Word count: 5839
Tables: –
Figures: –
References: 44

Author's address: Beata M. Gruber-Bzura, Department of Biochemistry and Biopharmaceuticals, National Medicines Institute, Chełmska 30/34 Str., 00-725 Warsaw, Poland; e-mail: b.gruber@nil.gov.pl

Abbreviations: **BC** – breast cancer; **CaR** – calcium-sensing receptor; **COX-2** – cyclooxygenase-2; **CC** – colon cancer; **CRC** – colorectal carcinoma; **CYP24A1** – 24-hydroxylase; **CYP27B1** – 1α-hydroxylase; **ER** – estrogen receptor; **IL** – interleukin; **K_{Ca}** – Ca^{2+} activated K^+ channels; **miRNA** – micro RNA; **MKP5** – mitogen-activated protein kinase phosphatase-5; **MM** – malignant melanoma; **NFκB** – nuclear transcription factor κB; **NSCLC** – non-small cell lung cancer; **OPN** – osteopontin; **PC** – prostate cancer; **PTH** – parathyroid hormone; **RXR** – retinoid X receptor; **SNP** – single nucleotide polymorphism; **VDR** – vitamin D receptor; **VDBP** – vitamin D binding protein; **VDRE** – vitamin D response elements; **VEGF** – vascular epithelial growth factor

INTRODUCTION

The term Vitamin D covers a group of steroid-like molecules that includes cholecalciferol (vitamin D_3), ergocalciferol (vitamin D_2), calcidiol (25-hydroxy vitamin D, 25(OH)D) and calcitriol (1,25-dihydroxy vitamin D, 1,25α$(OH)_2$D) [16]. The main source of vitamin D_3 (90%) is skin synthesis from 7–dehydrocholesterol upon solar ultraviolet B light. Cholecalciferol and ergocalciferol (formed in yeast and mushrooms exposed to UV radiation from ergosterol) are transported to the liver to be converted into the major circulating form, i.e. 25(OH) D by mitochondrial and microsomal CYP24A1. Next, calcidiol bound to the VDBP is transferred to the kidneys and hydroxylated into calcitriol, the active form of vitamin D, by CYP27B1, an enzyme that is stimulated by PTH and by serum increased Ca^{2+}. Many extrarenal tissues, including osteoclasts, skin, colon, brain and macrophages, also express this enzyme and, therefore, vitamin D can act in a paracrine and autocrine manner in these tissues [14, 15, 19, 35]. CYP27B1 is tightly regulated in a negative feedback loop. Calcitriol both inhibits renal 1α-hydroxylase and stimulates the 24-hydroxylase enzymes, thus maintaining 25(OH) D within limited boundaries and preventing excessive vitamin D activity [35]. 1,25α$(OH)_2$D is further hydroxylated by CYP24A1, forming two inactive metabolites, i.e. 24,25$(OH)_2$D and 1α,24,25$(OH)_3$D [14]. *CYP24A1* encodes a protein which inactivates calcitriol and calcidiol [16].

Bone homeostasis and mineralization is only one of the effects of vitamin D. This vitamin is also associated with diabetes, cardiac and gastrointestinal diseases. Most recent evidence has found that vitamin D has a role in thyroid function, oxidative stress and immune modulation [19, 35]. A large number of studies have pointed to the relations between blood levels of 25(OH) D with cancer incidence and survival. Multidirectional activity of vitamin D is possibly due to the presence of VDR in most nonskeletal human cells, including cancer cells [14, 19]. When bound to its ligand, calcitriol, VDR, a ligand-dependent transcription factor, dimerizes with RXR, which allows the heterodimer to translocate into the nucleus and, next, to bind to VDRE in promoter regions, inducing transcriptional regulation of target genes [14, 16, 35]. What is crucial to recognize is that a wide range of the genes regulated by VDR is recognize with cell proliferation, apoptosis, differentiation, angiogenesis and metastasis [14].

Calcitriol modulates transcriptional, post-transcriptional and post-translational mechanism, such as pre-mRNA splicing, epigenetic regulation and protein degradation in many cell types [23]. As was presented by Khatun et al. [23], calcitriol-induced transcription correlates with the chromatin accessibility of VDR binding regions. Besides, 1,25$(OH)_2$D epigenetically regulated tumor-related VDR target genes through DNA methylation and histone modifications. In turn, protein degradation has been shown to be regulated by calcitriol-modulated proteases and protease inhibitors and specific miRNA processing by enhancing the expression of Dicer in cancer cells.

As was presented by Crew [14], in BC cell lines exposed to calcitriol and in feeding studies of cholecalciferol in mouse models of BC, vitamin D was shown to affect signaling pathways in differentiation, alter metabolism, remodel extracellular matrix and innate immunity. Alimirah et al. [2] reported a positive correlation between VDR status and the expression of suppressor of fused gene, a hedgehog pathway inhibitor. It is due to the inhibition of miRNA-199a/miRNA-214 clusters, a well-known tumor promoter which inhibits the suppressor of fused gene. In some preclinical studies, BC, colon and lung cancers have all demonstrated the downregulation of VDR expression as compared to normal cells and well-differentiated tumors have shown more VDR expression when compared to their poorly differentiated counterparts. Higher tumor VDR expression has been noted as correlated with better prognosis in cancer patients [14].

The aim of this paper was to present the molecular pathways affected by vitamin D, which are included in carcinogenesis. The literature survey comprised of last five years and it proves vitamin D as the most phenomenal among others vitamins.

ANTICANCER PROPERTIES OF VITAMIN D

Cell proliferation, apoptosis and differentiation

Calcitriol regulates cancer transformation and malignancy through signaling cross talk with, and downregulation of mitogenic hormones or growth factors [33]. VDR has been implicated in cell cycle arrest, apoptosis and promotion of differentiation. This receptor affects cell proliferation *via* p21-activated kinase (PAK1), p27 (CDKN1B), p53 (*TP53*) retinoblastoma 2 (Rb(p130)) insulin-like growth factor binding protein5 (*IGFBP5*) or *via* direct induction of growth arrest and DNA damage-inducible 45 alpha proteins (GADD45-alpha), a regulator of NF-κB, which affects G0/G1 cell cycle arrest. According to Zeichner et al. [44], the induction of VDR is associated with suppression of human epidermal growth factor receptor 2 (HER2) through the ErbB2/AKT/ERK pathway. Calcitriol can suppress cell cycle proliferation genes encoding cyclins D1, A1, D3 and E1 and upregulate p21 (*CDKN1A*) [37]. In PC cells, it causes G1/G0 arrest in a p53-dependent manner by increasing the expression of the cyclin-dependent kinase inhibitors $p21^{Waf/Cip1}$ and $p27^{Kip1}$ and by causing the hypophosphorylation of the retinoblastoma protein (pRb) [27]. The molecules that are a target for vitamin D action are also c-Myc and c-Fos, the known protooncogenes involved in the cell cycle regulation and overexpressed in several tumours. Vitamin D suppresses the expression of c-Myc and promotes the increased expression of its antagonist, the transcriptional repressor, MAX dimerization protein 1 (MAD1/MXD1) [35]. Arrest of the cell cycle at the G0/G1 transition phase

can also be mediated concurrently with apoptosis generated *via* increased of proapoptotic proteins, BAX/BCL and mitochondrial release of cytochrome *c*. In some cells, calcitriol directly activates caspases to induce apoptosis. It enhances the expression of p73, a p53 homolog, which has been shown to be associated with apoptosis induction in some human and murine tumor system [14, 16, 27, 33]. In keratinocytes, calcitriol promotes the repair of DNA damage induced by UV radiation and increases p53 expression in damaged cells [35].

According to Jeong et al. [22], calcitriol can inhibit Wnt/B-catenin proliferation pathway in mouse breast tumor-initiating stem cells in a dose-dependent manner. It was associated with the significant decrease in the proliferative genes, such as *Lef-1, Axin2, Cdh1, TCf4.* The authors noted that mice fed with vitamin D-deficient diets displayed accelerated tumor growth and apperance. Calcitriol supplementation also proved to have a higher response with respect to tumor reduction than was the case in mice treated with radiation alone.

In many neoplastic cells, calcitriol induces differentiation. The mechanisms of the prodifferentiation effects are specific to the cell type and include the following: β-catenin, Jun-N-terminal kinase (JNK), phosphatidyl inositol 3-kinase, NFκB and the regulation of the activity of other transcription factors, as the activator protein-1 (AP-1) complex and CCAAT/enhancer-binding protein (C/EBP) [27].

Angiogenesis and antiinflammatory effects

Angiogenesis is the process of forming new blood vessels from existing vasculature. VEGF is the most potent stimulator of angiogenesis. As was noted in PC cells, calcitriol reduces VEGF expression through transcriptional repression of hypoxia-inducible factor-1 (HIF-1) [14, 27]. As was shown, it is a potent inhibitor of tumor cell-induced angiogenesis in experimental models [27]. It inhibits VEGF-induced endothelial cells tube formation *in vitro* and decreases tumor vascularization *in vivo* in mice bearing xenografts of BC cells over-expressing VEGF. It can also directly inhibit the proliferation of endothelial cells, leading to the inhibition of angiogenesis [14, 27]. Acting indirectly, calcitriol suppresses the expression of the proangiogenic factor IL-8. Chung et al. [12] in the tumors in the *VDR* knockout mice reported increased expression of proangiogenic factors, such as HIF-1α, VEGF, angiopoetin-1 and platelet-derived growth factor (PDGF). The other mechanism by which calcitriol inhibits angiogenesis is suppression of COX-2 expression [27].

One of the risk factors for cancer development is chronic inflammation. It generates many proinflammatory mediators which activate angiogenic processes and thereby promote tumor progression, metastasis and invasion. It is, therefore, the case that a proved decrease in the developing several cancers can be associated with the intake of antioxidants and nonsteroidal antiinflammatory drugs (NSAIDs) [27]. As was shown, calcitriol regulates the expression of several prostaglandin pathway genes, including decreasing *COX-2* and increasing 15-hydroxyprostaglandin dehydrogenase (*15-PGDH)*. It also decreases the expression of EP and prostaglandin F receptors and in this manner indirectly attenuates stimulation of growth [27]. Calcitriol-mediated reduction in prostaglandins provides an indirect downregulation of aromatase expression. Calcitriol also can indirectly regulate inflammatory, immune responses and cellular proliferation *via* suppression of NFκB or through the increase of MKP5 [14]. MKP5 causes dephosphorylation and inactivation of the p38 stress-induced kinase resulting in a decrease of proinflammatory cytokines, such as IL-6 [27]. As was shown in PC cells, the loss of MKP5 might occur during cancer progression and, therefore, calcitriol up-regulated MKP5 was seen only in primary cells normal or cancer, but not in the established cell lines derived from metastatic PC [27].

Metastases

Reduction of the invasive and metastatic potential of many malignant cells by calcitriol has been proven in murine models of prostate and lung cancers [27]. The mechanisms underlying this effect include a modulation of the expression of different surface proteins, N-cadherin switching to E-cadherin, whose expression is inversely correlated to metastatic potential, metalloproteinase-9 (MMP-9) or tissue inhibitor of metalloproteinase-1 (TIMP-1) [27, 35]. Calcitriol also inhibits expression of *ID1*, a gene involved in tumor progression and metastasis [35]. Williams et al. [42] found a negative correlation between serum 25(OH)D levels and the *ID1* expression in primary tumors from patients with BC. The other mechanisms induced by calcitriol to affect metastases include the regulation of the key molecules involved in these processes, such as components of the plasminogen activator (PA) system or tenascin-C, an extracellular matrix protein that promotes growth, invasion and angiogenesis and the expression of α6 and β4 integrins [27]. Significant reduction of cell migration and increased cell stiffness is probably a consequence of a reversal of the epithelial to mesenchymal transition, resulting in increased E-cadherin and F-actin and reduced vimentin [40]. Chiang et al. [10] demonstrated that calcitriol and its analog MART-10 (19-nor-2α-(3-hydroxypropyl)-1α,25(OH)$_2$D) effectively repress triple negative BC MDA-MB-231 cells migration and invasion through regulation, not only E and N-cadherins or MMP-9 but also through the down-regulation of P-cadherin expression and repression of LCN2, one of the BC metastasis stimulator.

Immune system

Pawlik et al. [36] noticed that the immunomodulating role of calcitriol and its analogs may play different roles at different stages of tumor progression and the cytokine profile activated or not by these compounds is dependent on the cell type. Thus, at an early stage of mouse mammary carcinoma 4T1 progression, in spleno-

cytes and/or lymph nodes, an increased of mRNA of Th2 cytokines, such as IL -4, -5, -9 and -13 was observed. In splenocytes alone, at an early stage of tumor progression, the levels of tumor necrosis factor α (TNFα), IL -12b and -12 rb2 were increased, but during the advanced phase of the experiment, calcitriol decreased their levels and stably increased the expression of IL -2 [36]. Several studies have demonstrated that calcitriol induces IL-10 expression, which has immunosuppresive activity [7, 21, 36]. In 4T1 tissues, Pawlik et al. [36] observed the upregulation of IL-10 during the early stages of tumor growth. In plasma of 4T1 bearing mice, administered with calcitriol, the Ly6G-6C^{+}SCC^{high} granulocytes, the primary source of tumor growth factor -β (TGF-β) was found to be increased [36]. The increased percentage of mature granulocytes and the producion of TGF-β may be responsible for the enhancement of metastasis [3, 25, 36]. As was shown by Anisiewicz et al. [3], calcitriol and its analogs enhanced the metastatic potential of 4T1 tumors without influencing the growth of primary tumor, by inducing the secretion of OPN, which is known to contain VDRE in the promoter region of *Spp1*, stimulates tumor growth and promotes metastasis by influencing tumor angiogenesis [36]. According to Anisiewicz et al. [3], the proved metastatic potential of 4T1 cancer cells was the result of the OPN overexpression, which prevailed over the decreasing tumor TGF-β level and tumor VEGF deprivation observed upon treatment with calcitriol and its analogs. Apart from these procarcinogenic immune processes induced by calcitriol and its analogs, Pawlik et al. [36] in the tumor tissue also noted the changes considered to be involved in antitumor immune response, such as, enhanced secretion of interferon -γ (IFN γ) or decreased expression of IL-1β after treatment with calcitriol and its analogs. According to the authors of this study, the contradictory processess induced by these compounds encourage further studies to define the role of vitamin D and carcinogenesis.

As was shown by O'Brien et al. [34], immune-related properties of vitamin D may be associated with methylation of CpGs in the vitamin D-related genes, with potential links to immune funcion-related genes. The authors demonstrated an interaction between 25(OH) D and methylation at cg21201924 in relations to BC risk (relative risk, RR = 1.22, 95% CI: 1.10–1.34; p = 7x10-5), indicating a larger methylation-BC hazard ratio in those with high serum 25(OH)D concentrations.

Vitamin D and calcium vs. antineoplastic effects

The interaction between vitamin D and calcium arises from the regulating function of calcium in the turnover of vitamin D in the organism. As was mentioned earlier, the renal synthesis of CYP27B1 is modulated by circulating Ca^{2+} and PTH [37].

Calcitriol may upregulate expression of the extracellular CaR, which transduces changes in extracellular fluid Ca^{2+} concentrations to stimulatory and inhibitory G proteins in a variety signalling pathways. The CaR is expressed by many normal and neoplastic cells [37]. As was given by Peterlik et al. [37], in a population-based cross-sectional study on calcium and vitamin D status of healthy adults, low daily calcium consumption reported in 81% of the cohort was consistent with vitamin D insufficiency in 26% of participants. Lin et al. [28] found that higher intakes of total calcium and vitamin D were associated with a lower risk of pre menopausal BC (RR = 0.65, 95% CI: 0.42–1.00 for vitamin D intake and RR = 0.61, 95% CI: 0.40–0.92 for calcium). Similarly Mc Cullogh et al. [31], who analysed data from nearly 70 000 postmenopausal women participating in the Cancer Prevention Study II Nutrition Cohort and found a negative correlation between the risk of BC (RR = 0.80) and intake of calcium (>1250 mg/day compared to <500 mg/day). The association was stronger (RR = 0.67) in women with tumor ER(+). Significant trends of decreasing breast density (high density is a strong risk factor) with increasing vitamin D and calcium intake observed also Fair et al. [18], but only among pre- not post menopausal women. The authors conducted a cross-sectional analysis of diet and serum vitamin D in relations to breast density in screening population with a large proportion of African American women.

A special role in cancerogenesis is played by K_{Ca}, affecting cell proliferation, differentiation, migration and apoptosis in various cell types by regulating Ca^{2+} signaling [23]. The amplification of *KCNMA1*, encoding one of K_{Ca} has been correlated with a high tumor stage and poor prognosis in BC. As was found by the authors, VDR agonists transcriptionally repressed larger conductance K_{Ca} in human BC cells MDA-MB-453 and promoted K_{Ca} protein degradation.

However, we should consider that the results obtained in the studies concerning the anticancer role of vitamin D are not unequivocal. The results obtained by Manson et al. [30] during randomized, placebo-controlled trial of vitamin D at a dose of 2000 IU per day and omega-3 fatty acids at a dose of 1g per day for the prevention of cancer and cardiovascular disease do not directly indicate the universal anticancer effect of vitamin D supplementation. The studies were conducted with 25.871 participants, including 5.106 black participants. As a result, supplementation with vitamin D did not affect the incidence of BC, prostate or CRC cancers. The invasive cancer of any type developed in 1.617 participants, with no significant differences between the vitamin D group (793 incidences) and the placebo group (824 incidences) (for vitamin D group *vs.* placebo group: hazard ratio, HR = 0.96, 95% CI: 0.88–1.06, P = 0.47). Also, the rate of death from cancer of any type did not differ significantly between the groups (HR = 0.83, 95% CI: 0.67–1.02). Nevertheless, in an analysis that excluded 1 year or 2 years follow up, the rate of death from cancer was significantly lower with vitamin D than with placebo (HR = 0.79, 85% CI: 0.63–0.99; HR = 0.75, 95% CI: 0.59–0.96, respectively).

VITAMIN D PATHWAY GENETIC POLYMORPHISM AND CANCER

Vitamin D pathway genetic polymorphisms may influence cancer risk. *VDR* polymorphisms are the best studied. Polymorphisms have also been found for genes related to vitamin D metabolism and signaling, such as *CYP27B1, CYP24A1*, *DBP, RXRA, CRP2, CASR, CUBN* and *DHCR7* [4, 13]. However, the associations between most of them and cancers were not significant.

Reimers et al. [39] found that one of the *CYP24A1* polymorphism, rs6068816, was associated with a 72% reduction in BC risk (TT *vs.* CC, OR=0.28, 95% CI: 0.10–0.76, p_{trend} = 0.01). The inverse correlation for plasma 25(OH)D with BC risk was noted among women with the common allele for *CYP24A,* rs927650 (p for interaction on a multiplicative scale = 0.01). With regards to *VDBP* polymorphism, the authors observed some variations in plasma 25(OH)D levels across the genotype for both *VDBP* polymorphisms: rs7041 and rs4588. However, the association between both *VDBP* variants and BC incidence was not clearly proved. Crew [14] mentioned the studies in which a significant inverse association between *VDBP* polymorphism and BC risk was noted independently on 25(OH) D levels. Kong et al. [24] investigated the role of genetic polymorphisms involved in the vitamin D pathway in NSCLC. In the case-control study, the authors found that SNPs rs3782130 (*CYP27B1*), rs7041 (group-specific component, *GC*), rs6068818 and rs4809957 (*CYP24A1*) are associated with NSCLC risk and that SNP rs3782130 may affect gene expression and patient survival. In turn, Vidigal et al. [41] proved that dietary, lifestyle and polymorphisms in *VDR* (ApaI), rs6013897, rs158552, rs17217110 in *CYP24A1* and rs10877012 in *CYP27B1* were associated with a higher risk of CRC.

VDR is one of the super families of steroid/thyroid hormone/retinoid nuclear receptors and consists of two domains which bind to DNA or vitamin D. It is highly polymorphic, with at least 618 reported variants, mostly undetectable. The most frequently studied *VDR* polymorphism are FokI (rs10735810) on exon 2, ApaI (rs7975232) and BsmI (rs1544410) on intron 8 and TaqI (rs731236) on exon 9 [14, 35].

Chen et al. [9] in a large nested case-control study found a positive association between the ff genotype of FokI and BC risk (multivariate odds ratio, OR = 1.34, 95% CI: 1.06–1.69), whereas eight other studies found no association with this genotype. In turn, Guy et al. [20] found that FokI genotype influenced BC risk when accounting for other *VDR* polymorphisms in haplotype combinations. Abbas et al. [1] found that TaqI polymorphism was associated with a significantly increased risk for ER+ tumors (OR = 1.18, 95% CI:1.00–1.38) as compared to carriers with non-carriers, but not for ER- tumors. Haplotype analysis revealed the haplotype FtCA containing TaqI allele is associated with a significantly greater BC risk as compared with the most frequent haplotype FTCG (OR = 1.43, 95% CI: 1.00–2.05). Simultaneously, no significant interaction between *VDR* genotypes or haplotypes and 25(OH)D was noted. During a meta-analysis of 67 independent studies, Raimondi et al. [38] found that *VDR* FokI and BsmI polymorphisms might modulate the risk of BC, skin cancer and PC. For the genotype FokI ff compared with FF carriers, the authors noted a significantly higher risk of PC, BC, CRC, skin and ovary and non-Hodgkin lymphoma, which was also associated with vitamin D levels. A significant reduction in PC risk was observed for carriers of BsmI Bb compared with bb genotype.

As was presented by Crew [14], the TT genotype of the TaqI polymorphism has been associated with reduced circulating levels of vitamin D. In women with high calcium intake, this genotype was associated with lower BC risk as compared to women with a tt or TT genotype and low calcium intake, which indirectly points to the positive role of calcium in chemoprevention. Reimers et al. [39] in a population-based case control study of 967 incident BC cases and 993 controls showed that the TT genotype of the TaqI polymorphism was associated with a 26% BC risk reduction (TT *vs.* CC, OR = 0.74, 95% CI: 0.56–0.98 , p_{trend} = 0.01).

A reliable meta-analysis was presented by Lu et al. [29]. The authors searched, among others, PubMed, ISI web of science and EMBASE. The studies have met such criteria as prospective nested case-control or cohort study and detailed data of OR and 95% CI. Eight studies were included in the meta-analysis. As a result, no significant association between FokI, BsmI, TaqI and ApaI polymorphisms and BC risk was showed.

Chandler et al. [8] conducted a Mendelian randomization analysis of the relationship between a vitamin D genetic score (GRS), comprised of five SNPs of vitamin D status in the *DHCR7, CYP2R1* and *GC* genes and cancer risk among women. Analysis was performed including 23.294 women of European ancestry who were cancer-free at baseline and followed for 20 years for incident cancer, including BC, CRC and lung cancers. A GRS for higher circulating 25(OH)D was not associated with cancer incidence or mortality. According to the authors, the known SNPs account for only about 5% of the variance in 25(OH)D and highlight the complex trait of circulating 25(OH)D. The authors suggest that the majority of the genetic effects for circulating calcidiol may be related to rare variants or structural variants other than SNPs or gene-environment interactions. The authors do not exclude the possibility that other genetic variants affect circulating 25(OH)D and cancer risk through entirely different mechanisms independent of 25(OH)D levels.

Köstner et al. [26] performed an analysis of studies evaluating the association between *VDR* polymorphism Fok1, Bsm1, Taq1, Apa1 and Cdx2, poly(A) and BgI1, as well as some haplotype combinations and cancer risk. As was shown, data indicating an association of *VDR* polymorphism and cancer risk were strongest for BC (Bsm1, Fok1), PC and malignant melanoma (MM) (Fok1). An association

of *VDR* polymorphism and cancer prognosis were strongest for PC (Fok1), BC (Bsm1, Taq1), MM (Bsm1) and renal cell carcinoma. Cho et al. [11] in a Korean case-control study of 695 CC patients and 1.397 controls confirmed the association between *VDR* FokI polymorphism and CC risk. Köstner et al. [26] and Cho et al. [11] agree that the same *VDR* polymorphism has a different effect depending on the type of cancer or on the aggressiveness of the tumor. Indeed, the effects observed by Eom et al. [17] in a hospital case-control study with the 715 pairs of newly diagnosed gastric cancer patients and controls suggest that the genetic polymorphisms of vitamin D-related genes do not modulate the effect of vitamin D with respect to gastric carcinogenesis, even though vitamin D intake is significantly correlated with the circulating 25(OH)D levels (but not with $1.25(OH)_2D$). The relationship between genotype and type of cancers was proved also by Beysel et al. [6], who conducted a case-control study on 165 patients with papillary thyroid cancer and 172 controls. *VDR* polymorphisms BsmI, ApaI, and TaqI were not associated with an increased cancer risk. Otherwise, FokI CT/TT genotype was related with an increased papillary thyroid cancer risk (CT vs CC: OR = 1.71, 95% CI:1.15–2.76, p = 0.028; TT vs CC: OR = 2.44, 95% CI: 1.29–4.62, p = 0.005; CT/TT vs CC: OR =1.88, 95% CI = 1.20–2.96, p = 0.006; CT/CC vs TT: OR = 1.80, 95% CI: 1.05–3.20, p = 0.041). Additionaly, *VDR* FokI T allele and TT genotype correlated with aggresiveness of papillary thyroid cancer. Thus, the authors suggested a new role of *VDR* FokI polymorphisms as a poor prognostic factor to assess the high risk of papillary thyroid cancer. Yu et al. [43] during meta-analysis conducted on 7 eligible studies noted that the *VDR* TaqI T allele carriers were at increased risk of lung cancer (OR = 1.25, 95% CI: 1.04–1.50). Compared with the TaqI TC+CC genotype, the TT genotype was positively associated with lung cancer risk (OR = 1.42, 95% CI:1.11–1.82). On the opposite, BsmI A allele was negatively related to the cancer risk and ApaI or FokI were reported as not associated with lung cancer risk. Lack of associations between *VDR* FokI polymorphism, low serum vitamin D and a risk of cancer was noted also by Mohamed et al. [32] as concerned pancreatic cancer in Egyptian patients. The study included rather small group of 47 cancer cases and 37 controls. In turn, Baykan et al. [5] did not demonstrate a statistically significant correlation between plasma vitamin D levels, ApaI, BsmI, FokI and TaqI polymorphisms and urothelial type bladder cancers in a Turkish population. Their study included 101 patients and 109 control subjects.

According to Crew [14], the discrepancies in the results obtained in the studies on the association between vitamin D pathway genetic polymorphism and cancer risk may be due to ethnic variation in the frequency of *VDR* gene polymorphisms and also may be due to the potential gene-environment interactions polymorphisms and factors, such as vitamin D and calcium intake, blood levels of 25(OH)D and sunlight exposure.

According to Pandolfi et al. [35], even the mechanisms through which vitamin D works are still not totally understood, it appears that its levels have a positive impact on the type and outcome of the tumor.

REFERENCES

[1] Abbas S., Nieters A., Linseisen J., Slanger T., Kropp S., Mutschelknauss E.J., Flesch-Janys D., Chang-Claude J.: Vitamin D receptor gene polymorphisms and haplotypes and postmenopausal breast cancer risk. Breast Cancer Res., 2008; 10: R31

[2] Alimirah F., Peng X., Gupta A., Yuan L., Welsh J., Cleary M., Mehta R.G.: Crosstalk between the vitamin D receptor (VDR) and miR-214 in regulating SuFu, a hedgehog pathway inhibitor in breast cancer cells. Exp. Cell Res., 2016; 349: 15–22

[3] Anisiewicz A., Pawlik A., Filip-Psurska B., Turlej E., Dzimira S., Milczarek M., Gdesz K., Papiernik D., Jarosz J., Kłopotowska D., Kutner A., Mazur A., Wietrzyk J.: Unfavorable effect of calcitriol and its low-calcemic analogs on metastasis of 4T1 mouse mammary gland cancer. Int. J. Oncol., 2018; 52: 103–126

[4] Arem H., Yu K., Xiong X., Moy K., Freedman N.D., Mayne S.T., Albanes D., Arslan A.A., Austin M., Bamlet W.R., Beane-Freeman L., Bracci P., Canzian F., Cotterchio M., Duell E.J., et al.: Vitamin D metabolic pathway genes and pancreatic cancer risk. PLoS One, 2015; 10: e0117574

[5] Baykan O., Akgul M., Uren N., Yaman A., Tinay I., Erqul E., Sazci A., Turkeri L., Haklar G.: The relationship between urothelial type bladder cancer, plasma 25-hydroxyvitamin D levels, and vitamin D receptor ApaI BsmI FokI, and TaqI polymorphisms. Clin. Lab., 2019; 65(4)

[6] Beysel S., Eyerci N., Pinarli F.A., Apaydin M., Kizilgul M., Caliskan M., Ozcelik O., Kan S., Cakal E.: VDR gene FokI polymorphism as a poor prognostic factor for papillary thyroid cancer. Tumour Biol., 2018; 40: 1010428318811766

[7] Boonstra A., Barrat F.J., Crain C., Heath V.L., Savelkoul H.F., O'Garra A.: 1α,25-dihydroxyvitamin D3 has a direct effect on naive CD4+ T cells to enhance the development of Th2 cells. J. Immunol., 2001; 167: 4974–4980

[8] Chandler P.D., Tobias D.K., Wang L., Smith-Warner S.A., Chasman D.I., Rose L., Giovanucci E.L., Buring J.E., Ridker P.M., Cook N.R., Manson J.E., Sesso H.D.: Association between vitamin D genetic risk score and cancer risk in a large cohort of U.S. women. Nutrients, 2018; 10: E55

[9] Chen W.Y., Bertone-Johnson E.R., Hunter D.J., Willet W.C., Hankinson S.E.: Associations between polymorphisms in the vitamin D receptor and breast cancer risk. Cancer Epidemiol. Biomarkers Prev., 2005; 14: 2335–2339

[10] Chiang K.C., Yeh T.S., Chen S.C., Pang J.H., Yeh C.N., Hsu J.T., Chen L.W., Kuo S.F., Takano M., Kittaka A., Chen T.C., Sun C.C., Juang H.H.: The vitamin D analog, MART-10, attenuates triple negative breast cancer cells metastatic potential. Int. J. Mol. Sci., 2016; 17: E606

[11] Cho Y.A., Lee J., Oh J.H., Chang H.J., Sohn D.K., Shin A., Kim J.: Vitamin D receptor FokI polymorphism and the risk of colorectal cancer, inflammatory bowel disease, and colorectal adenoma. Sci. Rep., 2018; 8: 12899

[12] Chung I., Guangzhou H., Seshadri M., Gillard B.M., Yu W., Foster B.A., Trump D.L., Johnson C.S.: Role of vitamin D receptor in the antiproliferative effects of calcitriol in tumor-derived endothelial cells and tumor angiogenesis in vivo. Cancer Res., 2009; 69: 967–975

[13] Clendenen T.V., Ge W., Koenig K.L., Axelsson T., Liu M., Afanasyeva Y., Andersson A., Arslan A.A., Chen Y., Hallmans G., Lenner P., Kirchhoff T., Lundin E., Shore R.E., Sund M., et al.: Genetic polymorphisms in vitamin D metabolism and signaling genes and risk of breast cancer: a nested case-control study. PLoS One, 2015; 10: e0140478

[14] Crew K.D.: Vitamin D: are we ready to supplement for breast cancer prevention and treatment? ISRN Oncol., 2013; 2013: 483687

[15] de La Puente-Yagüe M., Cuadrado-Cenzual M.A., Ciudad-Cabañas M.J., Hernández-Cabria M., Collado-Yurrita L.: Vitamin D: and its role in breast cancer. Kaohsiung J. Med. Sci., 2018; 34: 423–427

[16] Duffy M.J., Murray A., Synnott N.C., O'Donovan N., Crown J.: Vitamin D analogues: potential use in cancer treatment. Crit. Rev. Oncol. Hematol., 2017; 112: 190–197

[17] Eom S.Y., Yim D.H., Kim D.H., Yun H.Y., Song Y.J., Youn S.J., Hyun T., Park J.S., Kim B.S., Kim Y.D., Kim H.: Dietary vitamin D intake and vitamin D related genetic polymorphisms are not associated with gastric cancer in a hospital-based case-control study in Korea. J. Biomed. Res., 2018; 32: 257–263

[18] Fair A.M., Lewis T.J., Sanderson M., Dupont W.D., Fletcher S., Egan K.M., Disher A.C.: Increased vitamin D and calcium intake associated with reduced mammographic breast density among premenopausal women. Nutr. Res., 2015; 35: 851–857

[19] Gruber-Bzura B.M.: Vitamin D and influenza – prevention or therapy? Int. J. Mol. Sci., 2018; 19: E2419

[20] Guy M., Lowe L.C., Bretherton-Watt D., Mansi J.L., Peckitt C., Bliss J., Wilson R.G., Thomas V., Colston K.W.: Vitamin D receptor gene polymorphisms and breast cancer risk. Clin. Cancer Res., 2004; 10: 5472–5481

[21] Heine G., Niesner U., Chang H.D., Steinmeyer A., Zügel U., Zuberbier T., Radbruch A., Worm M.: 1,25-dihydroxyvitamin D3 promotes IL-10 production in human B cells. Eur. J. Immunol., 2008; 38: 2210–2218

[22] Jeong Y., Swami S., Krishnan A.V., Wiliams J.D., Martin S., Horst R.L., Albertelli M.A., Feldman B.J., Feldman D., Diehn M.: Inhibition of mouse breast tumor-initiating cells by calcitriol and dietary vitamin D. Mol. Cancer Ther., 2015; 14: 1951–1961

[23] Khatun A., Fujimoto M., Kito H., Niwa S., Suzuki T., Ohya S.: Down-regulation of Ca2+ -activated K+ channel KCa 1.1 in human breast cancer MDA-MB-453 cells treated with vitamin D receptor agonists. Int. J. Mol. Sci., 2016; 17: 2083

[24] Kong J., Xu F., Qu J., Wang Y., Gao M., Yu H., Qian B.: Genetic polymorphisms in the vitamin D pathway in relation to lung cancer risk and survival. Oncotarget, 2015; 6: 2573–2582

[25] Kowanetz M., Wu X., Lee J., Tan M., Hagenbeek T., Qu X., Yu L., Ross J., Korsisaari N., Cao T., Bou-Reslan H., Kallop D., Weimer R., Ludlam M.J., Kaminker J.S., et al.: Granulocyte-colony stimulating factor promotes lung metastasis through mobilization of Ly6G+Ly6C+ granulocytes. Proc. Natl. Acad. Sci. USA, 2010; 107: 21248–21255

[26] Köstner K., Denzer N., Müller C.S., Klein R., Tilgen W., Reichrath J.: The relevance of vitamin D receptor (VDR) gene polymorphisms for cancer: a review of the literature. Anticancer Res., 2009; 29: 3511–3536

[27] Krishnan A.V., Trump D.L., Johnson C.S., Feldman D.: The role of vitamin D in cancer prevention and treatment. Rheum. Dis. Clin. North Am., 2012; 38: 161–178

[28] Lin J., Manson J.E., Lee I.M., Cook N.R., Buring J.E., Zhang S.M.: Intakes of calcium and vitamin D and breast cancer risk in women. Arch. Intern. Med., 2007; 167: 1050–1059

[29] Lu D., Jing L., Zhang S.: Vitamin D receptor polymorphism and breast cancer risk: A meta-analysis. Medicine, 2016; 95: e3535

[30] Manson J.E., Cook N.R., Lee I.M., Christen W., Bassuk S.S., Mora S., Gibson H., Gordon D., Copeland T., D'Agostino D., Friedenberg G., Ridge C., Bubes V., Giovanucci E.L., Willett W.C., et al.: Vitamin supplements and prevention of cancer and cardiovascular disease. N. Engl. J. Med., 2019; 380: 33–44

[31] McCullough M.L., Stevens V.L., Diver W.R., Feigelson H.S., Rodriguez C., Bostick R.M., Thun M.J., Calle E.E.: Vitamin D pathway gene polymorphisms, diet, and risk of postmenopausal breast cancer: A nested case-control study. Breast Cancer Res., 2007; 9: R9

[32] Mohamed A.A., Aref A.M., Talima S.M., A Elshimy R.A., Gerges S.S., Meghed M., Zahran F.E., El-Adawy E.H., Abd-Elsalam S.: Association of serum level of vitamin D and VDR polymorphism Fok1 with the risk or survival of pancreatic cancer in Egyptian population. Indian J. Cancer, 2019; 56: 130–134

[33] Moukayed M., Grant W.B.: The roles of UVB and vitamin D in reducing risk of cancer incidence and mortality: a review of the epidemiology, clinical trials, and mechanisms. Rev. Endocr. Metab. Disord., 2017; 18: 167–182

[34] O'Brien K.M, Sandler D.P, Xu Z., Kinyamu H.K., Taylor J.A., Weinberg C.R.: Vitamin D, DNA methylation, and breast cancer. Breast Cancer Res., 2018; 20: 70

[35] Pandolfi F., Franza L., Mandolini C., Conti P.: Immune modulation by vitamin D: Special emphasis on its role on prevention and treatment of cancer. Clin. Ther., 2017; 39: 884–893

[36] Pawlik A., Anisiewicz A., Filip-Psurska B., Nowak M., Turlej E., Trynda J., Banach J., Gretkierewicz P., Wietrzyk J.: Calcitriol and its analogs establish the immunosuppressive microenvironment that drives metastasis in 4T1 mouse mammary gland cancer. Int. J. Mol. Sci., 2018; 19: E2116

[37] Peterlik M., Grant W.B., Cross H.S.: Calcium, vitamin D and cancer. Anticancer Res., 2009; 29: 3687–3698

[38] Raimondi S., Johansson H., Maisonneuve P., Gandini S.: Review and meta-analysis on vitamin D receptor polymorphisms and cancer risk. Carcinogenesis, 2009; 30: 1170–1180

[39] Reimers L.L., Crew K.D., Bradshaw P.T., Santella R.M., Steck S.E., Sirosh I., Terry M.B., Hershman D.L., Shane E., Cremers S., Dworakowski E., Teitelbaum S.L., Neugut A.I., Gammon M.D.: Vitamin D-related gene polymorphisms, plasma 25-hydroxyvitamin D, and breast cancer risk. Cancer Causes Control., 2015; 26: 187–203

[40] Santos J.M., Khan Z.S., Munir M.T., Tarafdar K., Rahman S.M., Hussain F.: Vitamin D3 decreases glycolysis and invasiveness, and increases cellular stiffness in breast cancer cells. J. Nutr. Biochem., 2018; 53: 111–120

[41] Vidigal V.M., Silva T.D., de Oliveira J., Pimenta C.A., Felipe A.V., Forones N.M.: Genetic polymorphisms of vitamin D receptor (VDR), CYP27B1 and CYP24A1 genes and the risk of colorectal cancer. Int. J. Biol. Markers, 2017; 32: e224–e230

[42] Williams J.D., Aggarwal A., Swami S., Krishnan A.V., Ji L., Albertelli M.A., Feldman B.J.: Tumor autonomous effects of vitamin D deficiency promote breast cancer metastasis. Endocrinology, 2016; 157: 1341–1347

[43] Yu Z.H., Chen M., Zhang Q.Q., Hu X.: The association of vitamin D receptor gene polymorphism with lung cancer risk: an update meta -analysis. Comb. Chem. High Throughput Screen, 2018; 21: 704–710

[44] Zeichner S.B., Koru-Sengul T., Shah N., Liu Q., Markward N.J., Montero A.J., Glück S., Silva O., Ahn E.R.: Improved clinical outcomes associated with vitamin D supplementation during adjuvant chemotherapy in patients with HER2+ nonmetastatic breast cancer. Clin. Breast Cancer, 2015; 15: e1–e11

The author has no potential conflicts of interest to declare.