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Vitamin D and Breast Cancer - Chemoprevention or Therapy?

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Abstract

Most evidence studies have concluded that there is an inverse association between blood levels of calcidiol and cancer incidence and survival. These findings especially apply in colorectal and breast cancer (BC). The phenomenon of the multidirectional activity of vitamin D is possibly due to the presence of the vitamin D receptor (VDR) in most non-skeletal human cells including cancer cells. The crucial is that a wide range of the genes regulated by VDR are related with cell proliferation, differentiation, angiogenesis and metastasis. The aim of this paper was to present recent data on the possible role of vitamin D as an chemopreventive or therapeutic agent against BC through considering the anticancer mechanisms induced by this vitamin as well as presenting the results of clinical studies on the impact of vitamin D status on BC incidence, survival and response to therapy. This review is based on an electronic search of articles in the PubMed database, including papers published mostly within last five years in that field and selected according to the following criteria: well commented studies concerning the association between vitamin D status and BC risk, response to therapy or survival, with the vitamin D supplementation outline and statistical data.

Introduction

Now we know that because of pleiotropic actions, bone homeostasis and mineralization is only one of the effects of vitamin D. Most evidence studies have concluded that there is an inverse association between blood levels of calcidiol and cancer incidence and survival. These findings especially apply in colorectal cancer and BC [1].

The phenomenon of the multidirectional activity of vitamin D is possibly due to the presence of VDR in most non-skeletal human cells including cancer cells [2,3]. VDR is a ligand-dependent transcription factor. When bound to its ligand, calcitriol, VDR dimerizes with the retinoid X receptor that allows the heterodimer to translocate into the nucleus and next to bind to vitamin D response elements in promoter regions inducing transcriptional regulation of target genes [1,3,4]. The crucial is that a wide range of the genes regulated by VDR are related with cell proliferation, apoptosis, differentiation, angiogenesis and metastasis [3].

The aim of this paper was to present recent data on the possible role of vitamin D as an chemopreventive or therapeutic agent against BC through considering the anticancer mechanisms induced by this vitamin as well as presenting the results of clinical studies on the impact of vitamin D supplementation on BC incidence and survival and response to anticancer therapy.

Literature Review

This review is based on an electronic search of articles in the PubMed database, including papers published mostly in the last five years up until 2018 in that field. The relevant papers are also included. All research articles were found with a combination of the following keywords: vitamin D and breast cancer, vitamin D and chemoprevention and breast cancer, vitamin D supplementation and breast cancer risk, vitamin D and anticancer therapy. Published articles were selected according to the following criteria: published in English, concerning the association between vitamin D status and BC risk, response to therapy or survival, with vitamin D supplementation outline, and statistical data. Studies with insufficient data were excluded.

Anticancer properties of vitamin D

The discovery that epithelial breast cells possess the same enzyme system as the kidney, where mainly is generated calcitriol, suggests the impact of vitamin D on BC cells. As was given by de La Puente-Yagüe et al. [5], human mammary cells cultured from normal breast tissue express VDR, 1α hydroxylase, CYP27B1 and the megalin-cubilin complex. This complex, among others, promotes the binding of 25(OH)D to vitamin D binding protein (VBP) [5]. VDR is a member of the nuclear family of receptors that also includes: estrogen and progesterone receptors (ER and PR), the androgen receptor and the T4/T3 receptor [1].

VDR has been implicated in cell cycle arrest, apoptosis and promotion of differentiation. This receptor affects cell proliferation, among others, *via* direct induction of growth arrest, indirect impact on the proteins, which affect G0/G1 cell cycle arrest or *via* suppression of human epidermal growth factor receptor 2 (HER2) as well as *via* the regulation of protooncogenes, such as c-Myc and c-Fos [4,6,7]. In BC cells, calcitriol induces apoptosis by stimulating Ca^{2+} release from intracellular stores [6]. According to Santos et al. [8], apoptosis induction by vitamin D in BC cells is associated also with decreased expression of mammalian target of rapamycin, which regulates glycolysis and cancer survival.

The growth factors and hormones up regulate cell proliferation and growth and in this way play a role in BC progression. According to Duffy et al. [1], VDR-mediated inhibition of ER^+ BC cells may be at least partly effected by downregulation of ER. In contrast, treatment with calcitriol was reported to induce ER expression in the ER- cells, SUM-229PE. This ability of calcitriol to convert ER- breast cancer cells to an ER^+ status, a potentially endocrine-sensitive would have major implications for the treatment of BC.

As was shown, calcitriol also is a potent inhibitor of tumor cell-induced angiogenesis in experimental models [9]. It inhibits vascular endothelial growth factor-induced endothelial cells tube formation *in vitro* and decreases tumor vascularization *in vivo* in mice bearing xenografts of BC cells over-expressing vascular endothelial growth factor. It can also directly inhibit the proliferation of endothelial cells leading to inhibition of angiogenesis [3,9]. Acting indirectly, calcitriol

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suppresses the expression of the proangiogenic factor interleukin 8 [9].

Chronic inflammation has been recognized as a risk factor for cancer development and many of the pro-inflammatory mediators which are then over expressed activate angiogenic processes and thereby promote tumor progression, metastasis and invasion [9]. Calcitriol, among others, suppresses the activation and signalling of nuclear factor κB, regulating the genes involved in inflammatory and immune responses and cellular proliferation [3]. Another mechanism of calcitriol that limit pro-inflammatory events is the increase of mitogen-activated protein kinase phosphatase-5 which indirectly decreases the pro-inflammatory cytokines, such as interleukin-6 [9].

The mechanisms underlying reduction of the invasive and metastatic potential of many malignant cells by calcitriol include a modulation of the expression of different surface proteins, such as induction of N-cadherin switching to E-cadherin, whose expression is inversely correlated to metastatic potential, downregulation of metalloproteinase-9 or an increase in tissue inhibitor of metalloproteinase-1 [4,9] . According to Santos et al. [8], significant reduction of cell migration and increased cell stiffness is probably a consequence of reversal of the epithelial to mesenchymal transition resulting in the increased E-cadherin and F-actin and reduced vimentin. 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 breast cancer (TNBC) cells, MDA-MB-231 migration and invasion through regulation not only E- and N- cadherins or metaloproteinase-9 but also through downregulation of P-cadherin expression and repression of lipocalin 2, one of the BC metastasis stimulator. In turn, Williams et al. [11] found a negative correlation between serum 25(OH)D levels and the ID1 expression, a gene involved in tumor progression and metastasis, in primary tumors from patients with BC. The other mechanisms induced by calcitriol to affect metastases include regulation of the key molecules involved in these processes, such as components of the plasminogen activator system or tenascin-C that promotes growth, invasion and angiogenesis [9].

Although many studies were taken to analyze the effect of vitamin D on the immune system, there are only a few reports on the effect of vitamin D on that system during progression and metastasis of solid tumors. In the light of evidence that calcitriol shows an immunosuppressive effect, it is very important to consider this action which is conflicting with other studies proving anticancer properties of calcitriol [12]. Pawlik et al. [12] who studied the impact of calcitriol and its analogs on 4T1 mouse mammary gland cancer observed that the most upregulated genes were related to Th2 and Treg cells which are involved in the immunosuppressive response. The authors 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. Very interesting role of vitamin D presented Thyer et al. [13]. On the basis of the study on human BC cells MCF-7, the authors demonstrated the interaction between VDR and VBP-derived macrophage activating factor (Gc-MAF) in the macrophage activation. As a result of such interaction, macrophages attacked human BC cells, inducing apoptosis and phagocytizing them. As was given by Saburi et al. [14] human macrophages activated *in vitro* with Gc-MAF (100 pg/mL) killed 60% and 86% of MCF-7 cells after 4h and 18 h of incubation, respectively. Although it has been demonstrated that macrophages activated by Gc-MAF bind to neoplastic cells *in vitro*, including BC cells, there is lack of clinical evidence about the action of activated macrophages to cancer cells [14]. According to the authors, differential responses to Gc-MAF that have been observed in human monocytes as well as in metastatic BC cells may be the result of individual VDR genotype. The same authors [14] also focus on the role of the VBP Gc-globulin (human group-specific component), the protein which apart from the storage and transport of vitamin D has an important function as a scavenger of extracellular G-actin to increase neutrophil chemotaxis and macrophage activation. By Saburi et al. [14], the modified Gc-globulin affects the activation and fortification of immune cells exhibiting anticancer activity.

Vitamin D status vs. BC risk and survival

The epidemiologic studies regarding the association between vitamin D and BC risk are on the one side promising and on the other side there are conflicting or inconclusive. The causes of the controversial results include: the degree of skin pigmentation, ER status, pre- or peri-menopausal period, BMI, potential modifying effect by VDR polymorphism that vary by ethnic groups or the circulating level of 25(OH)D [15].

Kim et al. [16] proved that race is an important factor which can determine the role of vitamin D in BC. The authors conducted a nested case-control study within Multiethnic Cohort Study of five race/ethnic groups between 2001-2006. Pre-diagnostic plasma levels of 25(OH) D were examined among 707 postmenopausal BC cases and matched controls. As was shown, 20 ng/mL increases of plasma 25(OH)D were inversely correlated with BC risk among white women and not among women in other race/ethnic groups (Table 1).

Kim and Je [15] on the basis of 30 prospective studies (nested case-control or cohort) focused on vitamin D as the intriquing factor which can influences BC survival. The authors found an overall non-significant inverse association between vitamin D intake or 25(OH)D blood levels and BC risk but in BC patients the risk of death from BC decreased by 42% for high *vs.* low 25(OH)D levels (Table 1). The authors considered strengths as well as weaknesses of their meta-analyses [16]. According to them, in the included studies, vitamin D intake or blood levels of 25(OH)D were measured before BC was diagnosed, so the possibility that cancer status affected vitamin D status was minimised. In the analyses of mortality from BC or overall mortality, the results were reported as compared with the healthy women. Most of the studies included in the meta-analysis used a single measurement at baseline which could lead to an underestimation of risk evaluation. The authors also pointed to the fact that unmeasured or residual confounding may affect the risk estimates in each study and thus pooled estimates in the meta-analyses. Similar observations to these presented by Kim and Je [15] in regard of positive role of vitamin D in BC made Hauser et al. [17] collecting medical records from adult solid tumor patients. Low 25(OH)D levels, i.e., vitamin D deficient and insufficient were highly prevalent in people with solid tumors including BC. The interesting relations in the effects of vitamin D blood levels and BC risk and prognosis revealed also Shirazi et al. [18] during the prospective, population-based cohort study. The significantly lower risk of ER-, PR- and tumor with high expression of Ki67-proliferation biomarker was found only in the group having vitamin D blood level ≥ 77 ≤ 97 nM/L. The groups with vitamin D blood level below or above this range had a relatively high risk of tumors with unfavourable prognosis (Table 1). A limitation of the study was that some tumor groups were relatively small.

According to the authors, these results suggest a U-shaped association between vitamin D blood levels and aggressive BC risk. Jeffreys et al. [19] in the cohort study confirmed that any vitamin D prescription, compared to never having been prescribed one was associated with a better survival from BC, however 3 or more

prescriptions was not (Table 1). Not fully consistent results were obtained by Bidgoli et al. [20] who did not show the differences in 25(OH)D levels between BC patients and healthy women and observed only slight associations between the lack of vitamin D and calcium supplementation or weekly egg consumption and premenopausal BC risk. The most important risk factor for BC incidence was the lack of sunlight exposure (Table 1).

From the studies presented by Cadeau et al. [21] it results the complex interactions between vitamin D intake, BMI and menopausal hormone therapy (MHT) use which affect postmenopausal BC risk. Authors found the increased postmenopausal BC risk associated with vitamin D supplementation in MHT never users with BMI< 25 kg/m^2, especially below 22 kg/m^2 than with BMI 22-24.9 kg/m^2. There was no association in women with higher BMI. Ever vitamin D supplementation was related to decreased BC risk in MHT ever users, regardless of BMI. As a summary, the increased risk associated with vitamin D supplementation in MHT never users was restricted to women with BMI<25 kg/m^2 (Table 1).

O'Brien et al. [22] confirmed that the beneficial effects of vitamin D supplementation can be related to menopausal status. The prospective cohort study conducted by the authors enrolled healthy women who had a sister with BC. It was found that 25(OH)D levels >38 ng/mL were associated with a 21% lower BC risk. Self-reported vitamin D supplementation ≥ 4 times/week was associated with an 11% lower hazard of BC wherein the inverse association was noted among postmenopausal women and a positive but statistically non-significant association among premenopausal women. Similar effects obtained Brisson et al. [23]-premenopausal female volunteers divided into four groups were supplemented with 1000 IU, 2000 IU or 3000 IU/day vitamin D or unsupplemented. The authors found that only supplementation with 3000 IU/day vitamin D was associated with a slightly smaller decline in breast density compared with placebo (Table 1). On the contrary, Fair et al. [24] in their cross-sectional study revealed the significant trends of decreasing breast density with increasing vitamin D and calcium intake in premenopausal but not among postmenopausal women, after statistical adjustment for age, race and BMI. Surprisingly, there was no association between serum vitamin D and breast density, regardless menopausal status (Table 1). Premenopausal status as more favourable for beneficial effects of vitamin D intake against BC risk found also Estèbanez et al. [25] in the meta-analysis on the effects of 25(OH)D blood levels and vitamin D intake, which included sixty eight studies published between 1998-2018, but only in the nested case-control study (Table 1). According to the authors, the observed effects dependent on menopausal status may result from the interaction between insulin-like growth factor-I (IGF-I) and vitamin D. As there is a physiological decline of IGF-I with aging, the interaction between IGF-I and vitamin D is likely to be stronger for pre- than for postmenopausal women.

According to Madden et al. [26], time of initiation of vitamin D supplementation is novel and could have significant clinical implications including the immediate prescribing of vitamin D supplements after BC diagnosis. In the large national BC cohort the authors found that *de novo* vitamin D use within 6 months after diagnosis is associated with 49% reduction in BC-specific mortality and that vitamin D supplemented>6 months after diagnosis results only in 20% reduction in BC- mortality (Table 1). The lack of the data concerning 25(OH)D blood levels at BC diagnosis, of course, makes impossible to highlight the only role of supplements in a reduction of BC mortality but, on the other side, the authors argue that vitamin D healthy blood level of >30 ng/mL is not possible to obtain without supplementation.

On the contrary to the above mentioned, mostly promising results which point the beneficial role of vitamin D supplementation in BC, Manson et al. [27] did not confirm any relationships between vitamin D intake and BC risk. The authors presented the results from randomized, placebo-controlled trial. As was shown, vitamin D intake 2000 IU/day was not associated with a lower cancer risk of any type (Table 1).

According to Grant [28], for BC only case-control studies consistently find inverse correlations between 25(OH)D and BC and 25(OH)D concentration values are only useful for short follow-up times for BC since it develops rapidly. Moukayed and Grant [29] claim that the prospective studies generally fail to find significant inverse correlations between 25(OH)D concentration and BC incidence because, apart from BC develops rapidly, the point is that a single 25(OH)D concentration measurement rapidly loses predictive ability. Grant et al. [30] suggest that the most likely reason for the failure of randomized controlled trials (RCTs) on the relationship between 25(OH)D blood and BC risk is inappropriate design, conduct, analysis and interpretation of the trials and that the most RCTs use principles designed to test pharmaceutical drugs which incorporate the assumption that RCTs is the sole source of the agent and that dose-response relationships are linear. Both assumptions are not true for vitamin D. In turn, the problem of the case-control studies may be that disease state, its treatment or disease-related behavioral changes may affect 25(OH)D concentration leading to reverse causation [22,29]. Bias from reverse causation can be avoided by prospective studies [22]. Welsh et al. [31] point to the fact that BC is heterogeneous and because of that, analysis of VDR actions in specific molecular subtypes of BC should be considered to clarify all conflicting data obtained in the clinical studies. Another point which should be regarded in such studies is the seasonal variations in vitamin D status. It can be speculated that women who are vitamin D deficient in summer are more likely to be deficient year round, enhancing the BC risk relative to those who are deficient only in winter [31].

Vitamin D and BC therapy

As was pointed by Madden et al. [26], there are the evidences to suggest that vitamin D supplementation used in conjunction with standard therapies may reduce BC recurrence and improve survival. According to Jacobs et al. [32], the results for prognosis and survival provide a more consistent picture than for vitamin D and BC incidence. As mostly, well-conducted observational studies are controlled for the confounders which may affect the relationship between 25(OH)D and BC survival, as for example, BMI, physical activity or cancer stage at diagnosis.

The topic which is the most often undertaken by the authors is the role of vitamin D in therapy of TNBC, the leading cancer in women [33]. BCs are categorized into three subtypes, ER+, HER2+ and TNBC. Treatment of ER+ and HER2+ BCs has been successful through targeted therapy with anti-estrogen and anti-HER2 drugs. Due to the lack of these targets, neo-adjuvant therapy is used for treatment of TNBC that is associated with aggressive phenotype, poorer prognosis and the high rate of relapse compared to other BC subtypes. Besides, almost half of the ER+ tumors eventually become resistant to anti-estrogens [34,35].

As was demonstrated by Chiang et al. [10], calcitriol and another vitamin D analog MART-10 (19-nor-2α-(3-hydroxypropyl)-1α, 25(OH)$_2$D) could effectively repress TNBC cells migration and invasion with analog more effective. These compounds induced cadherin switching and down regulated P-cadherin expression in MDA-MB-231 cells as well as repressed lipocalin 2, one of BC

Author, year	Type of study/ Country	Population	Vitamin D supplementation	25(OH)D serum concentration	Results	OR, CI, p-value	Adjustment factors
Kim et al., 2014 [16]	Nested case-control cohort study/Hawaii, Los Angeles	n=215 000/707 postmenopausal BC cases and matched controls/five race/ ethnic groups	-	Measure of pre- diagnostic plasma levels of 25(OH)D	20 ng/mL increases of plasma 25(OH)D levels were inversely associated with BC risk among white women; for women other than non- Hispanic whites such association was not significant	OR=0.28, 95% CI: 0.14-0.56	BMI, multivitamin or calcium supplements, number of live births, family history of BC, season, sunburn and engagement in strenuous sport
Kim et Je, 2014 [15]	prospective nested case-control or cohort studies/South Korea	n=31 867 BC incidences BC mortality and overall mortality: n=6092 BC patients	>500 IU/day *vs.* <148 IU/day - -	- - >29.1 ng/mL *vs.* <21 ng/mL <20.7ng/mL *vs.* 20.7 ng/mL	Not significant risk of BC incidence Lower mortality from BC Lower overall mortality	RR=0.95, 95% CI:0.88-1.01 p=0.09 RR=0.92, 95% CI:0.83-1.02 RR=0.58, 95% CI:0.40-0.85 RR=0.61, 95% CI:0.48-0.79	Menopausal status, BMI, physical activity
Bidgoli, Azarshab, 2014 [20]	Case-control study/Iran	n=60 BC premenopausal newly diagnosed patients/n=116 controls	BC patient's *vs.* control group Egg consumption > 3/week in control group *vs.* egg consumption <3/week in BC premenopausal patients group Un-supplemented BC premenopausal patients (1.67% supplemented with calcium) *vs.* control group supplemented with vitamin D (18.46%) and calcium (18.1%)/ dose unknown Lack of sun exposure in BC patients (98.33%) *vs.* lack of sun exposure in control group (85.3%)	15.2 ± 8.15 ng/mL *vs.* 15.5 ± 7.45 ng/mL - - Both groups vitamin D deficient	No significant differences in 25(OH)D blood levels between BC Premenopausal patients and control group Slightly reduced BC risk Slightly increased BC risk Increased risk of BC incidence	- OR=0.232, 95% CI:0.065-0.806 p=0.023 OR=1.115, 95% CI:1.049-1.187 p=0.009 OR=10.131, 98% CI:0.314-78.102 p=0.007	Age, reproductive features, history of pregnancy, menstrual disorders, BMI
Jeffreys et al., 2015 [19]	Retrospective cohort study/UK	n=21 932 women ≥ 55 y old, at least 5 y with the first diagnosis of BC, colorectal, lung, ovarian or uterine cancer and at least 5 y prior to diagnosis	Pre-diagnostic vitamin D supplementation, 3 prescriptions/ dose unknown *vs.* 1 or 2 prescriptions Pre-diagnostic vitamin D supplementation, any prescriptions/ dose unknown *vs.* no prescription	- -	No association with survival from any of the cancers studied Better survival from BC	- HR=0.78, 95% CI:0.70-0.88	BMI, alcohol drinking, smoking status, deprivation
Fair et al., 2015 [24]	Cross-sectional study/USA	n=57 premenopausal w omen / n=106 postmenopausal women	Premenopausal women total vitamin D intake <191.56 IU/ day/n=12 191.56-568.8 IU/day/n=24 ≥ 568.9 IU/day/n=21 Postmenopausal women total vitamin D intake <191.56 IU/ day/n=36 191.56-568.8 IU/day/n=36 ≥ 568.9 IU/day/n=34	–	Premenopausal women level of breast density 33.0% level of breast density 30.9% level of breast density 23.9% Postmenopausal women level of breast density 20.8% level of breast density 20.0% level of breast density 16.5%	95% CI:23.9-42.1 95% CI:24.4-37.4 95% CI:17.1-30.7 p=0.03 95% CI:15.8-25.8 95% CI:14.8-25.1 95% CI:10.8-22.2 p=0.67	BMI, race, age, calcium intake

				Premenopausal women <17.55 ng/mL/ n=20 17.56 - 28.6 ng/ mL / n=16 ≥ 28.7 ng/mL / n=21 Postmenopausal women < 17.55 ng/mL / n=35 17.56-28.6 ng/mL / n=38 ≥ 28.7ng/m L/ n=33	Premenopausal women level of breast density 29.7% level of breast density 26.4% level of breast density 25.0% Postmenopausal women level of breast density 19.4% level of breast density 23.4% level of breast density 20.2%	 95% CI:21.3-38.0 95% CI:17.7-35.2 95% CI:16.3-33.6 p=0.69 95% CI:13.5-25.4 95% CI:17.5-29.3 95% CI:14.1-26.3 p=0.20	
Shirazi et al., 2016 [18]	Nested case-control study based on prospective, cohort study/ Sweden	n=17035 / n=764 BC cases	-	≤ 76 nM/L and ≥ 98 nM/mL ≥ 77 nM/L and ≤ 97 nM/L	high risk of tumors with unfavorable prognosis the lowest overall risk of BC lower risk of ER- tumors lower risk of PR- tumors high expression Ki67 tumors	OR=0.97; 95% CI:0.75-1.25 OR=0.77, 95% CI:0.59-1.00 OR=0.46, 95% CI:0.23-0.94 OR=0.66, 95% CI:0.46-0.96 OR=0.57, 95% CI:0.36-0.90	Age, menopausal status, hormone replacement therapy, socio-economic index
Cadeau et al., 2016 [21]	Prospective cohort study/ France	n=57 403 postmenopausal women/n=2482 BC cases	<200 IU/day combined with calcium/dose unknown	-	Decreased postmenopausal BC risk in MHT ever users with BMI <25 kg/m^2 Decreased postmenopausal BC risk in MHT BMI ≥ 25 kg/m^2 Increased postmenopausal BC risk in MHT never users with BMI <25 kg/m^2 and stronger with BMI <22 kg/ m^2 than with BMI = 22-24.9 kg/m^2	HR=0.84, 95% CI:0.70-0.99 HR=0.87, 95% CI:0.62-1.23 HR=1.51, 95% CI:1.13-2.02 HR=1.62, 95% CI:1.11-2.35 HR=1.35, 95% CI:0.84-2.17	--
O'Brien et al., 2017 [22]	Prospective cohort study/USA	n=50884 healthy women/n=35074, 35-74 y old who had a sister diagnosed with BC/84% non-Hispanic white/85% w ell educated	> 38 ng/mL	-- Regular supplement use/ dose unknown (≥ 4 times/week) Regular supplement use Regular supplement use Regular supplement use	21% lower BC hazard 11% lower BC hazard inverse association with BC risk among postmenopausal women inverse association with BC risk among postmenopausal women per 100 IU increase positive association with BC risk but non-significant among premenopausal women positive association with BC risk among premenopausal women per 100 IU increase	adjusted HR=0.79, 95% CI:0.63-0.98 HR=0.89, CI:0.81-0.99 HR=0.84, CI:0.75-0.94 HR=0.98,95% CI:0.96-1.00 HR=1.17, 95% CI:1.43 p=0.008 HR=1.06, 95% CI:1.01-1.10	sunlight-related variables, exogenous hormone use, history of osteoporosis, education, BMI

Brisson et al., 2017 [23]	Double-blind, placebo-controlled parallel group trial/ Canada	n=306 premenopausal women supplemented/n=99 placebo	1000 IU / day for one year 2000 IU / day for one year 3000 IU / day for one year placebo	- - - -	-5.5% ± 0.5% reduction of percent mammographic breast density -5.9% ± 0.5% reduction of percent mammographic breast density -3.8% ± 0.5% reduction of percent mammographic breast density -5.7% ± 0.5% reduction of percent mammographic breast density	mean difference in change in percent mammographic breast density for increments of 1000 IU/day- 0.53,95% CI: 0.07-0.99, p=0.02	Serum 25(OH) D concentration, BMI, calcium intake, month of mammography, type of mammography, percent breast density
Madden et al., 2018 [26]	Cohort study/ Ireland	n=5417/women 50-80 y old with invasive BC not supplemented before diagnosis	-	> 400 IU after 6 months following diagnosis > 400 IU within 6 months following diagnosis	20% reduction in BC-specific mortality in *de novo* users *vs.* non-users 49% reduction in BC-specific mortality in *de novo* user's *vs.* non-users	HR=0.80, 95% CI:0.64-0.99 p=0.048 HR=0.51, 95% CI:0.34-0.74 p<0.001	age, smoking status, tumor stage, ER, PR and HER2 status, surgery after diagnosis, receipt of chemotherapy
Manson et al., 2019 [27]	Randomized, placebo-controlled study/ USA	n=25 871 healthy black and white participants including women ≥ 55 y old	2000 IU/day *vs.* placebo	--	Site-specific cancer incidence BC incidence Death from cancer	HR=0.96, 95% CI:0.88-1.06 p=0.47 HR=1.02, 95% CI:0.79-1.31 HR=0.83, 95% CI:0.67-1.02	--

BC-breast cancer; OR-odd ratio; CI-confidence interval; BMI-body mass index; RR-relative risk; HR-hazard ratio; ER-estrogen receptor; PR-progesterone receptor; MHT-menopausal hormone therapy; HER-human epidermal growth factor receptor.

Table 1. The selected human studies on the associations between 25(OH)D serum concentration or vitamin D supplementation and BC risk and survival published between years 2014-2019.

metastasis stimulator. Furthermore, MART-10 downregulated matrix metalloproteinase-9 activity and attenuated F-actin as well as calcitriol. According to Maaty et al. [36], in TNBC cells MDA-MB-231, MDA-MB-468 and HCC-1143 calcitriol regulates energy metabolism. On the opposite, one year earlier, Richards et al. [37] found that vitamin D at the high concentrations inhibited BC cell line MCF-7 but not the TNBC cell lines. As regards BC ER-, Santos-Martinez et al. [38] found that calcitriol is able to induce the expression of a functional ERα that is mediated through VDR. Calcitriol-induced ERα restored the response to anti-estrogens by inhibiting cell proliferation and the calcitriol-treated cells in the presence of anti-estrogen IC-182,780 resulted in a significant reduction of some cell proliferation regulators [38].

Neoadjuvant chemotherapy (NAC) has become a standard of care in locally advanced BC, especially for patients with large tumor size, lymph node metastasis, HER2 overexpression, TNBC subtype or inflammatory BC [39].

As was given by Thakkar et al. [34], approximately two-thirds of TNBCs express VDR and/or androgen receptor (AR) and it is possible that TNBCs co-expressing AR and VDR could be treated by targeting both of these hormone receptors. The authors provided that treatment of 15 BC cell lines including the cell lines which expressed AR and VDR receptors with AR or VDR agonists inhibited cell viability in a receptor-dependent manner and their combination appeared to inhibit cell viability. Apart from, the agonists induced differentiation and inhibited cancer stem cells [34]. Interestingly, cell viability was further decreased when AR/VDR agonists were combined with chemotherapeutic drugs. Among others, the authors shown that combination of calcitriol and Taxol resulted in an additive or synergistic decrease in cell proliferation and viability in two different TNBC cell lines, positive for VDR [34]. Similar results were obtained for TNBC cells positive for AR and VDR. The combination of AR- and VDR- targeted therapy with Taxol or cisplatin had an additive effect in reducing cell viability [34].

Some associations between vitamin D and tumor subtypes as hormone receptor (HR)-/HER2+ and TNBC were noted by Viala et al. [39]. The authors conducted a retrospective, observational, multicenter study which included 327 women treated with NAC with adjunction of therapies for HER2+ subtype. As was shown, vitamin D deficiency (measured as <20 ng/mL) was associated with the odds of not attaining pathologic complete response (pCR). The 5-year date of relapse (PFS) was 92 and 79% in the vitamin D deficient and in the sufficient group, respectively for patients with HER2+ tumors (p=0.20) while 5-year PFS rates in the HR+/HER2- cohort were 78 and 89%, respectively (p=0.056). A non-significant trend was observed in the TNBC subgroup (60.4% *vs.* 72.3%, respectively (p=0.3)). According to the authors, the lack of statistical significance could be explained by the relatively small number of patients in the TNBC cohort (n=90).

The promising studies were presented by Zeichner et al. [7] who performed a retrospective review of patients who received vitamin D supplementation during trastuzumab-based chemotherapy for HER2+ non metastatic BC and patients who were unsupplemented. As was found, vitamin D intake was associated with improved disease-free survival (DFS). Larger tumor size was associated with worse DFS and there was no overall survival based on any of the categories, including vitamin D supplementation, age at diagnosis or lymphovascular invasion (Table 2).

Charehbili et al. [40] during NEOZOTAC phase III trial studied the relationship between vitamin D serum level that was measured at baseline and before the last cycle of chemotherapy and pCR or pathological good response (defined as >90% decrease in tumor cellularity or total absence of invasive tumor cells in the breast only) in breast and lymph nodes. Admittedly, as was found, there was no association between baseline vitamin D levels and pCR, even including the season in which the baseline vitamin D was measured whereas the positive changes in vitamin D levels were significantly associated with pathological good response and the expected response was observed more often in women with end of treatment vitamin D levels >50.99 nM/L than in those with vitamin D level <50.99 nM/L [40] (Table 2).

Similar results obtained Chiba et al. [41] in retrospective cohort study with patients with operable BC. The authors, after adjusting for the effects of cohort clinical stage and receptor status showed that vitamin D deficiency (defined as <20 ng/mL serum levels measured before NAC) increased the odds of not attaining a pCR (Table 2).

On the contrary, Kim et al. [42] investigating the changes in serum 25(OH)D levels before and after NAC and the associations of this level with pCR and survival in 374 BC patients found that the patients with either pre- or post-NAC sufficient 25(OH)D levels accounted for 23.8% and the overall pCR rate was 25.9%. Most patients showed 25(OH)D deficiency at diagnosis and 65.8% showed decreased serum 25(OH) D levels after NAC (as was obtained also in other studies) [39,43]. As was concluded, the changes in calcidiol status were associated with postmenopausal status, molecular phenotype, baseline summer examination and rural residence but not with pCR. No association between survival and calcidiol status was found [42]. Similarly, Clark et al. [44] did not prove that vitamin D improves response to NAC. The authors during a retrospective cohort study showed that pre-treatment vitamin D levels had no impact on tumor response to NAC or short-term prognosis. Vitamin D level was not associated with attaining pathologic residual cancer burden 0/1 after NAC with anthracycline and taxane and was not associated with a 3-year relapse-free survival (RFS). However, surprisingly, the lower level of 25(OH)D correlated with higher tumor Ki67 proliferation biomarker adjusting for race (Table 2). As was given by the authors, no correlation between vitamin D and NAC response can be linked to the absence of HER2+ patients in the study. By Clark et al. [43], suggestion that vitamin D higher levels may suppress proliferation of BC is speculative as well as ascertainment that insufficient or deficient vitamin D levels do not impair or predict the efficacy of NAC in BC patients. However, the authors pointed to the fact that their study regarded vitamin D cut off level 30 ng/mL and they would not be able to identify the potential association with BC risk at higher serum concentrations of 25(OH)D to optimize response to NAC [44]. Furthermore, in these studies potentially important confounding variable, such as smoking status, physical activity, diabetes mellitus 2 status or vitamin D supplements were not regarded and the vitamin D status was not known at the time of NAC. The results described above comply with these shown by Lohmann et al. [45] one year later. The authors during randomized clinical trial with BC patients treated with NAC did not find the evidence that vitamin D blood level is associated with RFS, BC-specific survival (BCSS) and overall survival (Table 2). The authors pointed to the strenghts of the study such as the large number of patients, the high quality of data collection, the long follow-up (5 years) and that this was a multicenter international trial. According to the authors, the inverse association between vitamin D blood levels and the improved survival of BC patients treated with NAC which is shown in the observational studies may result from methodology. Besides, it is crucial to consider all confounds which can affect vitamin D blood levels and which are associated with BC outcomes, such as age, BMI, physical activity or good overall health.

Yao et al. [46] proved the importance of menopausal status to reveal the role of vitamin D in BC therapy. During prospective cohort study, the authors investigated a serum biomarker of 25(OH)D status measured at the time of BC diagnosis, with prognosis and found that serum 25(OH)D concentrations were lower in women with advanced stage tumors and the lowest in premenopausal women with TNBC. The calcidiol levels were inversely associated with hazards of disease progression and death, even after adjustment for clinical prognostic factors. Among premenopausal women, there were also associations with overall survival, BCSS and IDFS (invasive disease-free survival) (Table 2).

What about the "optimal" dose of vitamin D as beneficial for BC patients?

Despite of the equivocal results concerning the beneficial impact of vitamin D on BC prevention or therapeutic efficacy, which were presented in this article, the common conclusion is that the sufficient serum level of 25(OH)D is one of the factors which define our self-defence against neoplastic changes and successful therapy when it is needed. The additional benefit of vitamin D supplementation is to maintain the required serum concentration of 25(OH)D during chemotherapy which can decrease its level [39].

The key points are: "if there is the correlation between the beneficial effects of vitamin D and the vitamin D intake?" and "if are the optimal doses for chemoprevention or BC therapy support?" According to Kim et al. [16] it is plausible that the association of serum 25(OH)D with BC risk is non-linear, and a minimum threshold is needed for vitamin D to exert a protective effect. On the opposite, according to de La Puente-Yagüe et al. [5], the concentrations of calcidiol necessary to mediate the anti-cancerogenic effects are well above the physiological range and are associated with undesirable effects *in vivo*. According to Crew et al. [47], data from observational studies suggest that optimal levels for BC prevention exceed 40-50 ng/mL. As was highlighted by Kim and Je [15] for every 100 IU of vitamin D, blood 25(OH)D levels increase by 1 ng/mL while a healthy level >30 ng/mL of calcidiol is difficult to achieve without supplementation >1000 IU per day.

Some studies, presented above, showed that the relationship between vitamin D status health outcomes is a U-shaped and, that, for example, chemopreventive benefits from vitamin D supplementation are associated with rather lower vitamin D intake, such as less than 1000 IU per day, at least in premenopausal women [23]. These results prove that there is no the one, common daily dose of vitamin D having chemopreventive and anticancer effects. Variability in response to vitamin D intake may be due to other factors, such as menopausal status, BC phenotype, VDR signalling and heterogeneity, BMI or race [48]. The other point is the time of initiation of vitamin D supplements that could have significant clinical implications, especially in the case of applied chemotherapy [26]. Truly, the sufficient evidences on the benefits of different times of initiation of other adjuvant interventions including vitamin supplementation are lacking [26].

As was presented in this article, many studies found the association between vitamin D status and lower risk of BC incidence or between vitamin D supplementation used in conjunction with standard therapies and lowered BC recurrence or improved survival. The most important, phenomenal and promising is that because CYP27B1 was found in skin, colon, prostate and a breast cancer that allows calcitriol act in an autocrine or paracrine manner against cancerous transformation.

Author, year	Type of study/ Country	Population	Antitumor therapy	Vitamin D supplementation or 25(OH)D serum concentration	Results	HR, CI, p-value	Adjustment factors
Clark et al., 2014 [44]	Retrospective cohort study/ USA	n=82/BC HER2- cases	All patients anthracy cline and 90% anthracy cline with taxane	70% of patients <30 ng/mL	Not associated with RCB after NAC associated with higher biomarker Ki67 adjusting f or race not associated with 3 year RFS	OR=1.01, 95% CI:0.96-1.05 OR=0.95, 95% CI:0.90-0.99 HR=0.98, 95% CI:0.95-1.02	Hormone receptor status, BMI, race
Zeichner et al., 2015 [7]	Retrospective study/USA	n=134/HER2+ non metastatic BC patients who received vitamin D supplementation during therapy *vs.* n=112/HER2+ non-metastatic BC patients who were not supplemented	trastuzumab	10472 IU/week or 1500 IU/day *vs.* BC patients un-supplemented	Improved DFS a trend toward improved DFS despite large tumor size, increased number of metastatic lymph nodes, presence of LIV DFS: 32.6 months *vs.* 25.5 months OS: 43.8 months *vs.* 32.8 months	HR=0.36, 95%CI:0. 15- 0.88, p=0.026 HR=0.66, 95%CI: 0.41-1.06, p=0.09 p=0.022 p=0.07	Age at diagnosis, tumor size, PR and ER status, type of chemotherapy used, number of metastatic lymph nodes, diabetes mellitus type 2 development, BMI, heart failure, smoking status, radiation therapy, type of surgery, percentage of recurrences
Charehbili et al., 2016 [40]	Randomized, multicenter study / Holland	n=250/early BC II/ III stage/HER2-/ NAC with or without zoledronic acid	-	BC patients in zoledronic acid group supplemented with vitamin D 400 IU / day and calcium 500 mg/day	no association with pCR no association with pathological good response	OR=1.00, 95% CI:0.97-1.03, p=0.92 OR=1.00, 95% CI:0.97-1.02, p=0.66	age, ER status, N status, BMI, season of treatment
Yao et al., 2017 [46]	Prospective cohort study / USA	n=1666 / BC cases	-	≥ 25.10 ng/mL *vs.* <16.75 ng/mL Premenopausal women ≥ 25.10 ng/mL *vs.* <16.75 ng/mL	increased OS increased OS BCSS IDFS	HR=0.72, 95% CI:0.54-0.98 HR=0.45, 95% CI:0.21-0.96 HR=0.37, 95% CI:0.15-0.93 HR=0.58, 95% CI:0.34-1.01	Age, menopausal status, BMI, race / ethnicity , socioeconomic status, physical activity , smoking, supplementary and dietary vitamin D intake, season of blood collection, tumor stage, grade
Chiba et al., 2018 [41]	Retrospective cohort study/ France	n=67 patients/USA and n=77 patients/ France/clinical I and III stage BC	Anthracycline, taxane, trastuzumab for HER+ patients	<20 ng/mL *vs.* sufficient vitamin D serum level	increased no attaining pCR	OR=2.68, 95% CI:1.12-6.41, p=0.03	age, receptor status, clinical stage, BMI, disease type

BC-breast cancer; HER-human epidermal growth f actor receptor; RCB-pathologic residual cancer burden; NAC-neoadjuvant therapy ; RFS-relapse-free survival; OR-odd ratio; CI-confidence interval; HR-hazard ratio; DFS-disease-free survival; LIV-lymph vascular invasion; OS-overall survival; PR-progesterone receptors; pCR-pathological complete response; ER-estrogen receptors; N status-node status; BCSS-breast cancer-specific survival; IDFS-invasive disease-free survival.

Table 2. The selected human studies on the possible associations between vitamin D supplementation and chemotherapy for BC published between years 2014-2018.

Furthermore, still new data attain to prove the anticancer effects of vitamin D, such as its potential role in sensitizing of drug-resistant cancer cells [49].

Conclusion

On the basis of the studies to date, it seems to be undoubtedly that vitamin D is a potentially modifiable risk factor to target as a strategy for BC prevention and treatment. Of course, it does not mean that more is always better. According to Kim and Je the current evidences do not support use of high dose vitamin D regimens to get benefits for BC survival and more large randomised clinical trials should be conducted to provide evidence having implications for clinical practice. Consistently with the last supplementation guideline, obtaining and maintaining higher 25(OH)D concentrations than 30-50 ng/mL is not advisable. This range is recommended to ensure the balanced extra-skeletal effects of vitamin D.

Conflict of Interest

The author declares no conflict of interest.

References

1. Duffy MJ, Murray A, Synnott NC, O'Donovan N, Crown J (2017) Vitamin D analogues: potential use in cancer treatment. Crit Rev Oncol Hematol 112: 190-197.

2. Crew KD (2013) Vitamin D: Are we ready to supplement for breast cancer prevention and treatment? ISRN Oncol 2013: 483687.

3. Gruber-Bzura B (2018) Vitamin D and influenza – Prevention or therapy? Int J Mol Sci 19: 2419.

4. Pandolfi F, Franza L, Mandolini C, Conti P (2017) Immune modulation by vitamin D: special emphasis on its role on prevention and treatment of cancer. Clin Ther 39: 884-893.

5. De La Puente-Yagüe M, Cuadrado-Cenzual MA, Ciudad-Cabañas MJ, Hernández- Cabria M, Collado-Yurrita L (2018) Vitamin D: and its role in breast cancer. Kao Hsiung J Med Sci 34: 423-427.

6. Peterlik M, Grant WB, Cross HS (2009) Calcium, vitamin D and cancer. Anticancer Res 29: 3687-3698.

7. Zeichner SB, Koru-Sengul T, Shah N, Liu Q, Markward NJ, et al. (2015) Improved clinical outcomes associated with vitamin D supplementation during adjuvant chemotherapy in patients with HER2+ nonmetastatic breast cancer. Clin Breast Cancer 15: e1-11.

8. Santos JM, Khan ZS, Munir MT, Tarafdar K, Rahman SM, et al. (2018). Vitamin D3 decreases glycolysis and invasiveness, and increases cellular stiffness in breast cancer cells. J Nutr Biochem 53: 111-120.

9. Krishnan AV, Trump DL, Johnson CS, Feldman D (2012) The role of vitamin D in cancer prevention and treatment. Rheum Dis Clin North Am 38: 161-178.

10. Chiang KC, Yeh TS, Chen SC, Pang JHS, Yeh CN, et al. (2016) The vitamin D analog, MART-10, attenuates triple negative breast cancer cells metastatic potential. Int J Mol Sci 17: 606.

11. Williams JD, Aggarwal A, Swami S, Krishnan AV, Ji L, et al. (2016) Tumor autonomous effects of vitamin D deficiency promote breast cancer metastasis. Endocrinology 157: 1341- 1347.

12. Pawlik A, Anisiewicz A, Filip-Psurska B, Nowak M, Turlej E, et al. (2018) Calcitriol and its analogs establish the immunosuppressive microenvironment that drives metastasis in 4T1 mouse mammary gland cancer. Int J Mol Sci 19: 2116.

13. Thyer L, Ward E, Smith R, Fiore MG, Magherini S, et al. (2013) A novel role for a major component of the vitamin D axis: vitamin D binding protein-derived macrophage activating factor induces human breast cancer cell apoptosis through stimulation of macrophages. Nutrients 5: 2577-2589.

14. Saburi E, Saburi A, Ghanei M (2017) Promising role for Gc-MAF in cancer immunotherapy from bench to bedside. Caspian J Intern Med 8: 228-238.

15. Kim Y, Je Y (2014) Vitamin D intake, blood 25(OH)D levels, and breast cancer risk or mortality: a meta-analysis. Br J Cancer 110: 2772-2784.

16. Kim Y, Franke A, Shvetsov YB, Wilkens LR, Cooney RV, et al. (2014) Plasma 25- hydroxyvitamin D3 is associated with decreased risk of postmenopausal breast cancer in whites: A nested case-control study in the multiethnic cohort study. BMC Cancer 14: 29.

17. Hauser K, Walsh D, Shrotriya S, Karafa M (2014) Low 25-hydroxyvitamin D levels in people with a solid tumor cancer diagnosis: the tip of the iceberg? Support Care Cancer 22: 1931-1939.

18. Shirazi L, Almquist M, Borgquist S, Malm J, Manjer J (2016) Serum vitamin D (25OHD3) levels and the risk of different subtypes of breast cancer: A nested case-control study. Breast 28: 184-190.

19. Jeffreys M, Redaniel MT, Martin RM (2015) The effect of pre-diagnostic vitamin D supplementation on cancer survival in women: A cohort study within the UK Clinical Practice Research Datalink. BNC Cancer 15: 670.

20. Bidgoli SA, Azarshab H (2014) Role of vitamin D deficiency and lack of sun exposure in the incidence of premenopausal breast cancer: A case control study in Sabzevar, Iran. Asian Pac J Cancer Prev 15: 3391-3396.

21. Cadeau C, Fournier A, Mesrine S, Clavel-Chapelon F, Fagherazzi G, et al. (2016) Postmenopausal breast cancer risk and interactions between body mass index, menopausal hormone therapy use, and vitamin supplementation: evidence from the E3N cohort. Int J Cancer 139: 2193-2200.

22. O'Brien KM, Sandler DP, Taylor JA, Weinberg CR (2017) Serum vitamin D and risk of breast cancer within five years. Environ Health Perspect 125:1-9.

23. Brisson J, Bérubé S, Diorio C, Mâsse B, Lemieux J, et al. (2017) A randomized double-blind placebo-controlled trial of the effect of vitamin D3 supplementation on breast density in premenopausal women. Cancer Epidemiol Biomarkers Prev 26: 1233-1241.

24. Fair AM, Lewis TJ, Sanderson M, Dupont WD, Fletcher S, et al. (2015) Increased vitamin D and calcium intake associated with reduced mammographic breast density among premenopausal women. Nutr Res 35: 851-857.

25. Estébanez N, Gómez-Acebo I, Palazuelos C, Llorca J, Dierssen-Sotos T (2018) Vitamin D exposure and risk of breast cancer: a meta-analysis. Sci Rep 8: 9039.

26. Madden JM, Murphy L, Zgaga L, Bennett K (2018) De novo vitamin suplement use post-diagnosis is associated with breast cancer survival. Breast Cancer Res 172: 179-190.

27. Manson JE, Cook NR, Lee IM, Christen W, Bassuk SS, et al. (2019) Vitamin D supplements and prevention of cancer and cardiovascular disease. N Engl J Med. 380: 33-44.

28. Grant WB (2015) 25-hydroxyvitamin D and breast cancer, colorectal cancer, and colorectal adenomas: case-control versus nested case-control studies. Anticancer Res 35: 1153-1160.

29. Moukayed M, Grant WB (2017) 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 18: 167-182.

30. Grant WB, Boucher BJ, Bhattoa HP, Lahore H (2018) Why vitamin D clinical trials should be based on 25-hydroxyvitamin D concentrations. J Steroid Biochem Mol Biol 177: 266-269.

31. Welsh JE (2018) Vitamin D and breast cancer: past and present. J Steroid Biochem Mol Biol 177: 15-20.

32. Jacobs ET, Kohler LN, Kunihiro AG, Jurutka PW (2016) Vitamin D and colorectal, breast, and prostate cancers: a review of the epidemiological evidence. J Cancer 7: 232-240.

33. Kutlehria S, Behl G, Patel K, Doddapanaeni R, Vhora I, et al. (2018) Cholecalciferol- PEG conjugate based nanomicelles of doxorubicin for treatment of triple-negative breast cancer. AAPS Pharm Sci Tech 19: 792-802.

34. Thakkar A, Wang B, Picon-Ruiz M, Buchwald P, Ince TA (2016) Vitamin D and androgen receptor-targeted therapy for triple-negative breast cancer. Breast Cancer Res Treat 157: 77-90.

35. Shan NL, Wahler J, Lee HJ, Bak MJ, Gupta SD, et al. (2017) Vitamin D compounds inhibit cancer stem-like cells and induce differentiation in triple negative breast cancer. J Steroid Biochem Mol Biol 173: 122-129.

36. Abu El=Maaty MA, Dabiri Y, Almouhanna F, Blagojevic B, Theobald J, et al. (2018) Activation of pro-survival metabolic networks by 1,25(OH)2D3 does not hamper the sensitivity of breast cancer cells to chemotherapeutics. Cancer & Metabolism 6: 11.

37. Richards SE, Weierstahl KA, Kelts JL (2015) Vitamin D effect on growth and vitamin D metabolizing enzymes in triple-negative breast cancer. Anticancer Res 35: 805-810.

38. Santos-Martinez N, Diaz L, Ordaz-Rosado D, Garcia-Quiroz J, Barrera D, et al. (2014) Calcitriol restores antiestrogen responsiveness in estrogen receptor negative breast cancer cells: A potential new therapeutic approach. BMC Cancer 14: 230.

39. Viala M, Chiba A, Thezenas S, Delmond L, Lamy PJ, et al. (2018) Impact of vitamin D on pathological complete response and survival following neoadjuvant chemotherapy for breast cancer: a retrospective study. BMC Cancer 18: 770.

40. Charehbili A, Hamdy NAT, Smit VTHBM, Kessels L, Van Bochove A, et al. (2016) Vitamin D (25-OH D3) status and pathological response to neoadjuvant chemotherapy in stage II/III breast cancer: data from the NEOZOTAC trial (BOOG 10-01). The Breast 25: 60- 74.

41. Chiba A, Raman R, Thomas A, Lamy PJ, Viala M, et al. (2017) Serum vitamin D levels affect pathologic complete response in patients undergoing neoadjuvant systemic therapy for operable breast cancer. Clin Breast Cancer 18: 144-149.

42. Kim JS, Haule CCh, Kim JH, Kim SM, Yoon KH, et al. (2018) Association between changes in serum 25-hydroxyvitamin D levels and survival in patients with breast cancer receiving neoadjuvant chemotherapy. J Breast Cancer 21: 134-141.

43. Isenring EA, Teleni L, Woodman RJ, Kimlin MG, Walpole E, et al. (2018) Serum vitamin D decreases during chemotherapy: an Australian prospective cohort study. Asian Pac J Clin Nutr 27: 962-967.

44. Clark AS, Chen J, Kapoor S, Friedman C, Mies C, et al. (2014) Pretreatment vitamin D level and response to neoadjuvant chemotherapy in women with

breast cancer on the I-SPY trial (CALGB 150007/150015/ACRIN6657). Cancer Med 3: 693-701.

45. Lohmann AE, Chapman JAW, Burnell MJ, Levine MN, Tsvetkova E, et al. (2015) Prognostic associations of 25 hydroxy vitamin D in NCIC CTG MA.21, a phase III adjuvant randomized clinical trial of three chemotherapy regimens in high-risk breast cancer. Breast Cancer Res Treat 150: 605-611.

46. Yao S, Kwan ML, Ergas IJ, Roh JM, Cheng TYD, et al. (2017) Higher serum levels of vitamin D at diagnosis are associated with better survival in a prospective cohort of 1,666 women with breast cancer: A case-cohort analysis in the Pathways Study. JAMA Oncol 3: 351-357.

47. Crew KD, Xiao T, Thomas PS, Terry MB, Maurer M, et al. (2010) Safety, feasibility, and biomarker effects of high-dose vitamin D supplementation among women at high risk for breast cancer. Int J Food Sci Nutr Diet S1:001 1-9.

48. Crew KD, Shane E, Cremers S, McMahon DJ, Irani D, et al. (2009) High prevalence of vitamin D deficiency despite supplementation in premenopausal women with breast cancer undergoing adjuvant chemotherapy. J Clin Oncol 27: 2151-2156.

49. Pludowski P, Holick MF, Grant WB, Konstantynowicz J, Mascarenhas MR, et al. (2018) Vitamin D supplementation guidelines. J Ster Biochem Mol Biol 175: 125-135.