Association Of Vitamin D Deficiency And Pelvic Organ Prolapse In Postmenopausal Women: A Cross-Sectional Study

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Research article

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Abstract

Background: Vitamin D is vital for skeletal integrity as well as optimal muscle work. High incidence and prevalence of vitamin D deficiency as well as pelvic organ prolapse are found in postmenopausal women, thus raising the question of whether the entities could be related.

Methods: We compared 50 postmenopausal women with pelvic organ prolapse (POP) with 48 age- and weight-matched controls. The clinical assessment of the disorder was performed using the Pelvic Organ Prolapse Quantification system (POP-Q). A questionnaire was filled out by the participants and a blood sample was collected for 25-OH-vitamin D determination.

Results: The two groups did not differ significantly in body mass index, but differed significantly in vitamin D blood level concentrations, those being lower in POP patients. A significantly higher prevalence of vitamin D deficiency (25-OH-vitamin D< 50 nmol/l) was found in the test group compared to controls. Higher parity/vaginal deliveries and less caesarean sections were found in patients than in controls.

Conclusions: Vitamin D deficiency might be an important associated systemic factor associated to pelvic organ prolapse. The determination of vitamin D levels in postmenopausal women and replenishing its deficiency might also be of importance for the pelvic floor.

Background

Pelvic organ prolapse (POP) is affecting millions of women worldwide. It is caused by the weakening of the pelvic floor supportive tissue and occurs independently or coexists with other pelvic floor dysfunction, with a lifetime risk for surgery as high as 20% [1] and with a substantial reoperation rate due to recurrence [2]. The supportive soft tissues within the pelvic floor are a combination of muscles, fascias, and ligaments working together to keep the pelvic organs in place in a highly dynamic environment, to provide support and resist deformations [3]. Evidence suggests that weakness of the supportive tissues, either at the systemic or local level, may predispose to POP, suggesting that underlying supportive tissues of the pelvic floor are made of weak components and these components are more likely to fail or stretch leading to POP [4]. Various factors may affect the functional capacity of the pelvic floor. Knowing the prevalence of vitamin D deficiency, its recently observed potentially protective actions on tendons, ligaments and connective tissue [5] and its known effect on the longitudinal and striated muscle [6–8], the role of this factor became a subject of observation in few clinical studies in the last years, already bringing some promising, but inconsistent results [9–11].

In the last two decades, epidemiological extension of vitamin D deficiency in the common population has been confirmed. It has been estimated that 20–80% of US, Canadian and European elderly men and women are vitamin D deficient [12, 13]. Being a major factor in maintaining calcium and phosphorus homeostasis, vitamin D is involved in bone integrity. Its active form 1,25(OH)$_2$D$_3$ exerts its biological effects through vitamin D receptors (VDRs). These receptors are present also in the smooth and skeletal muscles [14, 6]. Through them vitamin D impacts in the proper
functioning of skeletal muscles by regulating calcium homeostasis to affect muscle contractility and by maintaining muscle cell environment against inflammation [7]. In case of deficiency, a smaller number of type 2 muscle fibres are present in the muscle [8], and muscle weakness is obvious not only in cases of overt deficiency, but also of insufficiency. In POP, fibrosis and the degradation of the connective tissue in the vaginal wall predominate and the aggravation of degenerative changes in the connective tissue lead to its progression [15]. Women with POP have more type III collagen than women without the disease, and the ratio of collagen I to collagen III is decreased [4].

We wanted to understand how important vitamin D deficiency/insufficiency is in the Slovenian female population suffering from POP. We expected it to be important in addition to the known factors influencing the pelvic floor, such as parity, exercise and chronic coughing. We conducted a prospective study on postmenopausal women with objective morphological evaluation of the pelvic floor status using the POP-Q system [16].

**Methods**

One hundred and two consecutive patients all attending the Gynaecological Outpatient Department at the University Medical Centre Ljubljana aged 50 to 74 years were assessed for either POP (test group) or other unrelated condition (control group). The exclusion criteria were conditions affecting the muscle function (asthma/chronic obstructive pulmonary disease, chronic cough, muscle or connective tissue disease, nerve disease, i.e. multiple sclerosis). Treated osteoporosis and taking more than 400 IU of vitamin D supplement daily were also exclusion criteria, making the bias in previous and current vitamin D status. Two participants were excluded from the analyses due to high-dose supplemental vitamin D treatment of osteoporosis, one from test and 1 from control group. Also, one patient was excluded from the control group due to multiple sclerosis, identified only after later review of the anamnestic questionnaire. One patient was too young to include in the study and thus excluded. Final number of participants was 98, among them 50 POP patients and 48 controls. The study was approved by the National Committee for Medical Ethics and written informed consent was obtained from all participants.

During two consecutive years of sun-deprived winter months from November to April, 50 women with POP and 48 controls were recruited. All participants completed anamnestic questionnaire. A blood sample was taken in the morning hours for detecting the value of 25-OH-D$_3$ in the serum; serum calcium and phosphorus were also routinely measured. One of the gynaecologists (M.B. or M. Ba.) performed a gynaecological examination using the Pelvic Organ Prolapse Quantification System (POP-Q) as an objective, site-specific examination system describing and quantifying the location of different points along the vaginal wall for staging the degree of POP [16]. The control group of age- and weight-matched women had no POP.

Vitamin D – more precisely 25-OH-D$_3$ – was determined from two batches (April 2018 and 2019), minimizing interassay variability. The sera were frozen until the time of analysis. Serum 25-OH-D$_3$ (in the following text 25-OH-D, also vitamin D) levels were measured using a direct competitive
chemiluminescence immunoassay (CLIA). During the first incubation, 25-OH-D is dissociated from its binding protein and binds to the specific antibody in the solid phase. After 10 minutes the tracer (vitamin D linked to an isoluminol derivative) is added. After a second 10-minute incubation, the unbound material is removed with a wash cycle. Subsequently, the starter reagents are added to initiate a flash chemiluminescent reaction. The light signal is measured by a photo multiplier as relative light units and is inversely proportional to the concentration of 25-OH-D present in calibrators, controls and samples. The LIASON 25-OH-D assay has an analytical sensitivity of 10 nmol/l. Intraassay precision/coefficient of variability (CV) is 2 to 4% and interassay CV is 7%.

Twenty-five-hydoxy-vitamin D values of < 50 nmol/l are considered to be deficient, values < 25–30 nmol/l are associated with osteomalacia. However, values 50–74 nmol/l are considered to be insufficient, especially in certain risk patient subgroups. Values 75–125 nmol/l are considered normal.

Statistical analyses were performed using SPSS version 25 statistical program. Descriptive statistics (mean, standard deviation, median) were calculated for continuous variables. Frequency distributions were obtained for categorical data (descriptive statistics). After prior testing for normal distribution of parametric variables, the Student’s t-test for independent samples was applied to compare differences between test and control group. For descriptive statistics, Chi square test was used. Pearson and Spearman correlation coefficients were calculated. Multivariate discriminant analyses were performed to search for the prediction value of dependent variables for POP. A p-value < 0.05 was considered statistically significant in all the calculations.

**Results**

Ninety-eight postmenopausal women aged from 49 to 75 (mean age 60.7 years, median 60.0 years) were recruited, 50 of them suffered of POP; the other 48 were weight-matched controls of the same age span. All women were of good health. Thirteen patients had well controlled hypertension, as did 11 controls (p = 0.72). Five patients and 1 control had type 2 diabetes (p = 0.09), on oral medication without any significant diabetic complications. 6 participants had osteoporosis and were treated in the past.

Their menopausal age was 1 to 30 yrs (mean 10.5, median 10.0) and did not differ significantly between the test and control group. Personal anthropometric characteristics of our patients are shown in Table 1.
Table 1
Personal characteristics of participants.

<table>
<thead>
<tr>
<th></th>
<th>POP patients (No = 50)</th>
<th>Controls (No = 48)</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>AGE (yrs +/- SD)</td>
<td>62.6 +/- 9.3</td>
<td>58.7 +/- 7.2</td>
<td>0.024</td>
</tr>
<tr>
<td>BMI (kg/m^2 +/- SD)</td>
<td>27.6 +/- 3.8</td>
<td>27.1 +/- 5.2</td>
<td>0.65</td>
</tr>
<tr>
<td>MENARCHE (age +/- SD)</td>
<td>13.4 +/- 1.6</td>
<td>12.9 +/- 1.7</td>
<td>0.17</td>
</tr>
<tr>
<td>PARITY (no. +/-SD)</td>
<td>2.3 +/- 0.8</td>
<td>1.8 +/- 0.7</td>
<td>0.005</td>
</tr>
<tr>
<td>Vaginal deliveries (no. +/-SD)</td>
<td>2.2 +/- 0.8</td>
<td>1.7 +/- 0.9</td>
<td>0.02</td>
</tr>
<tr>
<td>Caesarean section (no.)</td>
<td>0.02 +/- 0.1</td>
<td>0.1 +/- 0.4</td>
<td>0.15</td>
</tr>
<tr>
<td>MENOPAUSE (age +/- SD)</td>
<td>50.5 +/- 3.6</td>
<td>50.1 +/- 3.2</td>
<td>0.53</td>
</tr>
<tr>
<td>POSTMENOP. AGE (yrs +/- SD)</td>
<td>12.3 +/- 8.7</td>
<td>8.7 +/- 7.1</td>
<td>0.03</td>
</tr>
</tbody>
</table>

Due to a wide range of participants a statistically important difference resulted in the mean age between test and control group (62.6 vs. 58.7 years).

Mean 25-OH-D level in patients was 42.9 compared to 50.9 in controls, p = 0.049, reaching statistically significant difference. Table 2 include laboratory results of our participants: measurements of 25-OH-D, serum Ca and P.

Table 2
Results of vitamin D determination and biochemical measurements in participants.

<table>
<thead>
<tr>
<th></th>
<th>POP patients (No = 50)</th>
<th>Controls (No = 48)</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>25-OH-D (nmol/l +/- SD)</td>
<td>42.9 +/- 18.8</td>
<td>50.9 +/- 21.1</td>
<td>0.049</td>
</tr>
<tr>
<td>Ca (mmol/l +/- SD)</td>
<td>2.32 +/- 0.13</td>
<td>2.33 +/- 0.1</td>
<td>0.69</td>
</tr>
<tr>
<td>P (mmol/l +/- SD)</td>
<td>1.13 +/- 0.23</td>
<td>1.13 +/- 0.18</td>
<td>0.98</td>
</tr>
</tbody>
</table>

Values for Ca and P were all in a normal range.

Groups significantly differed in vitamin D levels: the test group had significantly lower blood vitamin D levels. However, we searched for the clinical significance of vitamin D levels and searched for vitamin D deficiency in our participants. Indeed, patients had a significantly higher prevalence of vitamin D deficiency, as shown in Table 3 and Fig. 1. There were 33 (66.0%) patients with vitamin D deficiency compared to 21 (43.8%) in controls. In the study group there were also 15 (30.0%) vitamin D insufficient
subjects and only 2 patients (4%) with normal vitamin D levels compared to 20 vitamin D insufficient controls (41.6%) and 7 controls (14.6%) with normal vitamin D levels (< 0.05).

Table 3
Results of anamnestic questionary on life facts and habits.

<table>
<thead>
<tr>
<th></th>
<th>POP patients YES/NO (No.)</th>
<th>Controls YES/NO (No.)</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Physical exercising</td>
<td>28/22</td>
<td>26/22</td>
<td>0.85</td>
</tr>
<tr>
<td>Sexual intercourses</td>
<td>16/14</td>
<td>21/27</td>
<td>0.23</td>
</tr>
<tr>
<td>Pelvic floor exercises of Kegel</td>
<td>31/19</td>
<td>28/20</td>
<td>0.71</td>
</tr>
<tr>
<td>Hormone replacement therapy</td>
<td>6/44</td>
<td>1/47</td>
<td>0.06</td>
</tr>
<tr>
<td>Smoking</td>
<td>9/41</td>
<td>7/41</td>
<td>0.64</td>
</tr>
<tr>
<td>Coffee</td>
<td>6/35/9*</td>
<td>1/40/7*</td>
<td>0.07</td>
</tr>
</tbody>
</table>

*high intake/moderate intake (3 coffees or less)//no coffee

The results of POP-Q test showed that 15 patients had a stage 2 prolapse, 32 a stage 3 prolapse and 3 patients a stage 4 prolapse. The control group included 31 participants without prolapse as well as 17 participants with stage 1 asymptomatic physiological prolapse considered normal in women with parity history and evaluated for a condition other than pelvic floor disfunction (PFD) [17]. Participants did not differ significantly in the incidence of vitamin D deficiency at different prolapse stages (p = 0.21), but bivariate analyses between degree of POP (0 to 4) and vitamin D levels showed significant moderately high negative ccorelation (r= -0.24, p = 0.018, n = 98).

The results of the anamnestic questionnaire regarding personal habits, physical activity, pelvic floor muscle training, sexual activity and hormone replacement therapy are shown in Table 4. Physical activity was evaluated (from sedentary lifestyle to intensive physical excercise) and no important difference between the patients and controls were observed. Dividing the participants into two groups regarding sufficient and insufficient physical activity (Table 4) did not bring any significant difference between patients and controls, either. Smoking habits were comparable in both groups. However, more patients than controls were taking hormone replacement therapy, almost reaching statistical importance (p = 0.06).
Table 4
Prevalence of vitamin D deficiency in POP patients vs. controls.

<table>
<thead>
<tr>
<th></th>
<th>POP patients</th>
<th>Controls</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vitamin D deficiency</td>
<td></td>
<td></td>
</tr>
<tr>
<td>YES (No.)</td>
<td>33 (66.0%)</td>
<td>21 (43.8%)</td>
</tr>
<tr>
<td>NO (No.)</td>
<td>17 (34.0%)</td>
<td>27 (56%)</td>
</tr>
<tr>
<td>Total (No.)</td>
<td>50</td>
<td>48</td>
</tr>
</tbody>
</table>

Chi² (1) = 4.90, p = 0.027

Regarding the pattern of POP, 28 patients had cytocele, 20 patients the combination of cystocele and rectocele, but 1 patient had rectocele only. In 1 patient with POP there was an isolated apical prolapse without cysto- or rctocele. Patients with a cystocele were more prone to vitamin D deficiency (p < 0.05), but patients with a rectocele (p = 0.78) were not (Fig. 2, Fig. 3).

Multivariate discriminant analyses extracted two important predictive variables from the set of dependent variables (vitamin D, age, serum Ca, serum P, body mass index) to predict having POP or no. These are age and vitamin D. The multivariate discriminant analysis on these two variables and POP (Wilks λ = 0.905, p = 0.009, structure matrix coefficients regarding mathematical function of POP, are: for age = 0.72, for vitamin D= -0.63) showed, that vitamin D and age are predictive factors (without gynaecological examination) for correctly placing patient in POP group in 58%, and in 71% for predicting that women belong to control group.

Discussion

The study demonstrates that vitamin D deficiency is an independent and important factor in POP in female postmenopausal population as hypothesised. Significantly lower vitamin D levels were found in POP group and a significant prevalence of vitamin D deficiency was found in patients with POP compared to controls. A significant correlation was found between vitamin D deficiency and cystocele, but not vitamin D deficiency and rectocele. Among the possible factors affecting the pelvic floor, parity and vaginal deliveries showed significant importance as expected, since caesarian section appears to be protective against POP.

Our results regarding vitamin D are consistent with some previous publications. Badalian and co. [9] found that higher vitamin D levels are associated with decreased risk of pelvic floor dysfunction in women. The study included women of all ages; PFD was evaluated by anamnestic questionnaire. Based on a cohort of 349 participants Parker-Autry et al. [10] concluded that insufficient vitamin D was
associated with increased colorectal symptoms and greater impact of urinary incontinence on the quality of life. On the other hand, no significant correlation was found between lower urinary symptoms and vitamin D deficiency in the research by Aydogmus and coworkers [11], but it stressed the necessity of further investigation of pelvic floor integrity and functions. Interestingly, an American study [18] showed a potential association between vitamin D and the development of urinary incontinence in a racially diverse cohort of older men and women. A very recent study by Kaur et al. [19] confirms that vitamin D levels were associated with a decreased risk of pelvic floor disorders in geriatric females. In their research, Hyung Ahn et al. [20] examined vitamin D levels and performed a VDR genotype analysis. The presence of a certain sequence of the vitamin D receptor (VDR) polymorphism (Apal and Bsmi) was associated with PFD in vitamin D deficient subjects.

To avoid seasonal variations in vitamin D concentrations we opted for the determination of 25-OH-D during winter time. Our study and control groups were ment to be of comparable age and they were weight-matched, both being important factors in vitamin D concentrations. Although the mean age of patients and controls was 62.6 years and 58.7 years, which appeared to be statistically significantly different (p < 0.05), the difference was not important for the subjects and questions from our study since we searched in the span of postmenopausal women aged between 50 and 75 years. Postmenopausal age was different between groups as expected, due to chronological age difference. Of the most importance is that our participants were weight-matched, because of the known significant inverse correlation between vitamin D levels and BMI [21, 22]. Since vitamin D is soluble in body fat, the reduced bioavailability of vitamin D results in obesity [23]. Parity differed among the two groups (2.26 vs. 1.81, p < 0.01) regarding the mean number of deliveries as well as the number of vaginal deliveries (p < 0.05), both being known as risk factors for POP. Unsignificantly higher prevalence of caesarean sections was observed in the control group (p = 0.15), also a known fact that caesarean section is protective for POP. There was no statistical difference in the timing of menarche or in age at the menopause.

PFD-POP was objectified using the POP-Q examination system in the present study. The numerical quantification of the disorder encountered for a homogeneous and stable recruitment of participants, where the subjective factor was minimized. Previous investigations were not based on POP-Q assessment [9, 10], except for the study of Kaur and coworkers [19], and Aydogmus and coworkers [11]. However, none of them assessed the possible differences among the different compartments of the prolapse. The limitation of this study was that the mean age of patients and controls appeared statistically significantly different (p < 0.05). Since we searched in the span of postmenopausal women, we assume the difference not to be of an utmost importance for the question of our study.

In our study cystocele was associated with vitamin D deficiency, whereas rectocele was not. This was not necessarily a surprise taking into account that abnormal pelvic floor muscles are observed more often in women with anterior prolapse than with posterior prolapse [24], and that anterior prolapse in a great number of cases is also linked to the descent of the apical vaginal support, hence sharing a more complex mechanism in terms of different pelvic support structures involved [25]. Additionally, women with cystocele have the most compliant anterior and posterior vaginal wall support systems when
compared to women with rectocele and normal support [4]. As vitamin D plays a role in different support tissues (i.e. striated and smooth muscles, tendons, fascias and connective tissue), we speculate that the likelihood of clinical manifestation of its deficiency is more likely to be pronounced in pelvic floor conditions where as many of those support tissues and structures are involved.

Both of our groups were comparable in physical activity, habits and rare use of hormone replacement therapy. We found the participating women to be well informed about the importance of physical exercise. 56% of patients and 54.2% of controls had sufficient physical exercise daily.

Smoking as a negative factor for bladder health [26] was present in 16 women, comparable in patients and controls.

Vitamin D supplementation was prescribed to all participants with vitamin D deficiency or insufficiency in this study. In an Indian study [19], the regression of symptoms of urinary incontinence as well as the positive effect on the symptoms of PFD were observed after 6 months of treatment.

**Conclusions**

Our study proved, there is a significant association between vitamin D deficiency and POP in postmenopausal women. We believe that vitamin D deficiency might be an important associated systemic factor in POP in postmenopausal female population. The anterior vaginal wall seems more susceptible for vitamin D deficiency. Educating women about vitamin D supplementation should be an important part of a conservative approach to the prevention and treatment of POP.

**Abbreviations**

POP - pelvic organ prolapse

POP-Q - Pelvic Organ Prolapse Quantification system

VDR - vitamin D receptor

PFD - Pelvic floor dysfunction

**Declarations**

- Ethics approval and consent to participate: approved by the National Committee for Medical Ethics on 19th September 2017 (ID: 0120-487/2017/7)
- Consent for publication: Not applicable
- Availability of data and materials: The datasets used and/or analysed during the current study are available from the corresponding author on reasonable request.
- Competing interests: The authors declare that they have no competing interests
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Authors’ contributions: Legan: Protocol and project development, Data analysis, manuscript writing and editing; M. Blaganje: Protocol development, Data collection, Manuscript writing and editing; J. Osredkar: Protocol development, Manuscript editing; M. Barbič: Data collection, Manuscript editing

Acknowledgements: Not applicable

References


**Figures**
Figure 1

Prevalence of vitamin D deficiency in prolapse patients vs. controls.
Figure 2

Prevalence of vitamin D deficiency in cystocele patients
Vitamin D deficiency comparable in rectocele patients and controls (p = 0.78)

Figure 3

Prevalence of vitamin D deficiency in rectocele patients