



# Widespread vitamin D deficiency among adults from northern Poland (54°N) after months of low and high natural UVB radiation

Powszechny niedobór witaminy D u dorosłych z województwa pomorskiego po miesiącach niskiego i wysokiego promieniowania UVB

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## Abstract

**Introduction:** Widespread vitamin D deficiency has been reported worldwide and evidence of its deleterious effects on health has been accumulating. There is insufficient data concerning vitamin D status among the Polish adult population.

The aim of the study was to determine vitamin D status and factors influencing it — UVB exposure, supplementation, and diet — among adults in northern Poland following months of low and high natural UVB radiation.

**Material and methods:** Adults were recruited in an outpatient clinic. All were examined twice in 2012: in winter (in February, March and the first half of April), and autumn (between 25 September and 8 November). Questionnaire examinations were performed, and serum concentrations of 25-hydroxy-vitamin D (25(OH)D), calcium, phosphorus, parathyroid hormone (PTH), and alkaline phosphatase (ALP) were determined.

**Results:** 40 men and 69 women participated in the study (age  $48.4 \pm 15$  years, body mass index (BMI)  $25.9 \pm 4.3$  kg/m<sup>2</sup>, mean  $\pm$  SD). Mean 25(OH)D serum level in winter was  $13.3 \pm 6.6$  ng/mL, 81.1% of participants were vitamin D-deficient. Mean 25(OH)D concentration in autumn was  $22.8 \pm 7.9$  ng/mL (42.2% of subjects were vitamin D-deficient). Median 25(OH)D and PTH concentrations between the two examination periods differed significantly (11.9 vs. 22.1 ng/mL, and 46.3 vs. 32.2 pg/mL, respectively).

In autumn, negative correlations were found between: 25(OH)D and PTH serum levels, 25(OH)D and BMI values. When compared to respective counterparts, participants declaring vitamin D supplementation, and sunbed use had significantly higher median 25(OH)D concentrations.

**Conclusions:** UV exposure during the summer was insufficient to provide adequate vitamin D status for almost half of the participants by as soon as the early autumn. Our results suggest all-year-round vitamin D supplementation should be widely implemented.

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**Key words:** vitamin D deficiency; cholecalciferol; parathyroid hormone; ultraviolet rays; adult

## Streszczenie

**Wstęp:** Liczne doniesienia naukowe mówią o powszechnym niedoborze witaminy D na całym świecie i jego negatywnych skutkach zdrowotnych. Dane dotyczące stanu zaopatrzenia w witaminę D wśród dorosłych Polaków są niewystarczające.

Celem pracy było określenie stanu zaopatrzenia w witaminę D i czynniki nań wpływające — ekspozycję na promieniowanie UVB, suplementację i dietę — u dorosłych z województwa pomorskiego po miesiącach niskiego i wysokiego promieniowania UVB.

**Materiały i metody:** Dorosłych probantów zrekrutowano w przychodni. Wszystkich przebadano dwukrotnie w 2012. roku: zimą (od lutego do połowy kwietnia) i jesienią (od 25. września do 8. listopada). Przeprowadzono badania ankietowe; określono stężenia surowicze 25-hydroksywitaminy D (25(OH)D), wapnia, fosforu, parathormonu (PTH) i fosfatazy alkalicznej (ALP).

**Wyniki:** 40 mężczyzn i 69 kobiet wzięło udział w badaniu (wiek  $48,4 \pm 15$  lat, indeks masy ciała (BMI)  $25,9 \pm 4,3$  kg/m<sup>2</sup>,  $\bar{x} \pm$  odchylenie standardowe). Średnie stężenie 25(OH)D w zimie wyniosło  $13,3 \pm 6,6$  ng/ml, 81,1% badanych miało niedobór witaminy D. Średnie stężenie 25(OH)D jesienią wyniosło  $22,8 \pm 7,9$  ng/ml (42,2% badanych miało hipowitaminozę D). Uzyskano istotne różnice median stężeń 25(OH)D i PTH między okresami badań (odpowiednio 11,9 wobec 22,1 ng/ml oraz 46,3 wobec 32,2 pg/ml).

Jesienią uzyskano ujemne korelacje między stężeniami 25(OH)D i PTH oraz 25(OH)D i wartościami BMI. Wyższą medianę stężeń 25(OH)D odnotowano u osób deklarujących suplementację (wobec tych negujących) oraz korzystanie z solarium (porównując z niekorzystającymi).

**Wnioski:** Ekspozycja na promienie UV w lecie była niewystarczająca by zapewnić odpowiedni poziom witaminy D już wczesną jesienią u niemal połowy badanych. Wyniki wskazują na konieczność wprowadzenia powszechnej całorocznej suplementacji witaminy D u dorosłych. (Endokrynol Pol 2015; 66 (1): 30–38)

**Słowa kluczowe:** niedobór witaminy D; cholekalcyferol; parathormon; promienie ultrafioletowe; dorośli

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## Introduction

Vitamin D deficiency has been recorded across the globe and evidence of its deleterious effects on both skeletal and extraskeletal health has been accumulating [1, 2]. Associations between hypovitaminosis D and numerous diseases, including cardiovascular, neoplastic, infectious and autoimmune, have been put forward and investigated [2, 3].

Vitamin D status in the body is reflected by the concentration of its most abundant circulating and storage form: 25-hydroxy-vitamin D (25(OH)D). While there is no consensus as to the optimal serum levels of 25(OH)D, many authors agree that deficiency is defined as 25-hydroxy-vitamin D concentrations lower than 20 ng/mL [4–8]. Vitamin D status was defined here with the following 25(OH)D serum levels: sufficiency at 30 to 80 ng/mL, insufficiency at 20 to 30 ng/mL, and deficiency at less than 20 ng/mL.

The main sources for vitamin D in humans are: synthesis in the skin after exposure to sunlight, diet, and supplements. In many countries, including Poland, certain foods are fortified with vitamin D. However, natural UVB radiation is vitamin D's most important source [1]. While it has been shown that incidental sun exposure (2–3 times a week for 5–30 minutes each time) may be sufficient to maintain vitamin D adequacy throughout the year, epidemiologic data worldwide records hypovitaminosis D, suggesting widespread insufficient exposure to UVB radiation [1, 9].

In the northern hemisphere, 25(OH)D serum levels vary seasonally: its highest values are recorded in August and September, and its lowest in February [10, 11]. In Europe at latitudes above 51 degrees, there is insufficient natural UV light for cutaneous vitamin D synthesis from September to March [12]. It has been suggested that a peak 25(OH)D serum level of at least 32 ng/mL in summer is required to maintain non-deficient (> 20 ng/mL) vitamin D status during winter [13].

In this study, vitamin D status and other parameters of calcium-phosphate homeostasis were assessed among urban adults in northern Poland following months of high and low natural UVB radiation, *i.e.* in autumn and in winter. To assess 25-hydroxy-vitamin D serum concentration as well as other calcium-phosphate metabolism parameters, and behavioural factors influencing vitamin D status among adults from northern Poland in two repeated examinations: after months with low and high natural UVB radiation.

## Material and methods

The study was approved by the independent bioethics committee of the Medical University of Gdańsk, Poland.

All subjects were examined twice, in two examination periods, or series, in 2012: from February to mid-April (the winter series of the study), *i.e.* in the months after and during low natural UVB radiation, and from 25<sup>th</sup> September to 8<sup>th</sup> November (the autumn series) — following high ultraviolet radiation in summer.

### Recruitment of subjects

Adult participants were enrolled in the study in an outpatient clinic in Gdansk. These included the clinic's employees and their family members, 20 employees of two local companies producing mechanical and electrical equipment, and individuals who presented for doctor's appointments and laboratory examinations. All subjects received an information sheet concerning the study and signed a consent form for participation. There were no exclusion criteria.

Participants received their laboratory results in person or by post within a month of each study series.

### Questionnaire examinations

A questionnaire was filled out by study subjects in both examination series. Participants declared their socioeconomic status, frequency of intake (per week) of foods rich in vitamin D (fish, eggs) and calcium (dairy products), vitamin D supplementation, exposure to UVB (sun-tanning and sunbed usage), health status (self-assessed), diseases (hypertension, diabetes mellitus, asthma, digestive system and kidney diseases, depression, neoplasms, sarcoidosis, rheumatoid arthritis, osteoporosis, psoriasis), chronic and sporadic medication, and symptoms of vitamin D deficiency (weakness, muscle and bone pain).

### Control group

Based on the questionnaire data, a control group of healthy subjects was selected. This comprised individuals who denied receiving chronic medication (apart from oral contraceptives in the case of one female) or having been diagnosed with chronic diseases, and who declared themselves able to climb at least two flights of stairs without the need to rest.

### Laboratory examinations

Blood drawn from subjects was centrifuged for ten minutes at 3,500 g. Acquired serum was transferred to separate tubes and frozen at –20°C. Tubes were transported in dry ice to the Central Diagnostic Laboratory of the Medical University of Gdańsk hospital. Serum concentrations of 25-hydroxy-vitamin D and parathyroid hormone were determined, using, respectively, a DiaSorin® Liaison® analyser with the '25OH vitamin D TOTAL' assay, and a Siemens IMMULITE® 1000 Immunoassay System with a dedicated assay. An Abbott

Table I. Descriptive statistics of acquired laboratory results

Tabela I. Statystyki opisowe uzyskanych wyników laboratoryjnych

## A. Winter data

	Age (years)	BMI [kg/m <sup>2</sup> ]	Ca [mg/dL]	P [mg/dL]	ALP [U/L]	PTH [pg/mL]	25(OH)D [ng/mL]
n (40 men)	109 (40)	103 (39)	108 (40)	108 (40)	108 (39)	108 (40)	109 (40)
mean	48	26.1	9.5	3.5	67.8	48.2	13.3
SD	15.2	4.5	0.5	0.5	30.3	18.4	6.6
SEM	1.5	0.5	0.04	0.05	2.90	1.8	0.63
Median	48	25.5	9.5	3.6	61	46.2	11.9
Q1	35	23.2	9.2	3.2	52	33.3	8.9
Q3	61	28.7	9.8	3.9	75	58.9	16.2
IQR	26	5.5	0.6	0.7	23	25.6	7.3
Min.	21	18.7	8.4	1.9	32	16.0	3.9
Max.	86	40.9	11.5	4.6	279	113.0	42.6

## B. Autumn data

	Age (years)	BMI [kg/m <sup>2</sup> ]	Ca [mg/dL]	P [mg/dL]	ALP [U/L]	PTH [pg/mL]	25(OH)D [ng/mL]
n [total(men)]	109 (40)	104 (40)	107 (40)	107 (40)	108 (39)	108(40)	109 (40)
mean	48.5	25.76	9.5	3.57	66.2	36.5	22.8
SD	15.0	4.29	0.6	0.61	20.50	16.2	7.9
SEM	0.9	0.25	0.05	0.06	1.18	1.6	0.5
Median	48	25.2	9.5	3.6	63	32.0 (*)	22.1 (*)
Q1	35	22.7	9.2	3.1	53	24	16.2
Q3	62	27.8	9.9	4.0	77.8	45.3	27.7
IQR	27	5.1	0.8	0.9	24.8	21.3	11.5
Min.	21	18.4	7.6	2.0	31	4	9.5
Max.	86	38.6	10.6	5.0	170	84	44.4

\*  $p < 0.0001$  for both PTH and 25(OH)D median concentrations in autumn vs. winter (Mann-Whitney U test and Wilcoxon matched-pairs signed rank test)

Architect® analyser was applied for spectrophotometric determination of alkaline phosphatase activity, calcium and phosphorus concentrations.

**Statistical analysis**

Statistical evaluation was performed using Graphpad Prism 5 (GraphPad Software, Inc) software. Non-parametric tests were used for group comparisons since 25(OH)D, PTH, age and BMI values did not follow a Gaussian distribution (as verified with the Shapiro-Wilk test). Also, Spearman rank correlations were calculated. Significance level was set at 0.05.

**Excluded data**

Certain obtained laboratory results were excluded from the statistical analysis:

— ALP activities in a male with chronic cholelithiasis (279 and 283 U/L),

— PTH, calcium and phosphorus of two females due to a strong suspicion of primary hyperparathyroidism. All other data of these subjects was included in the analysis.

**Results**

Descriptive statistics of data on the laboratory results acquired in both study series are shown in Tables IA and B. Laboratory reference ranges were as follows — calcium: 8.5–10 mg/dL, phosphorus: 2.3–4.7 mg/dL, ALP: 40–150 U/L, PTH: 10–62 pg/mL, 25(OH)D: 30–80 ng/mL.

Since peak annual 25(OH)D serum levels have been previously recorded in August and September for the northern hemisphere [10, 11], we checked whether subjects tested in each week of the autumn examination series (25<sup>th</sup> September to 1<sup>st</sup> October, 2<sup>nd</sup> October to 8<sup>th</sup> October etc.) had different median 25(OH)D concentra-

tions. No statistically significant differences were found. The same result (no differences) was obtained in the case of the winter examinations.

### Demographic data and health status of study participants

109 adults were enrolled in the study, 40 men and 69 women, aged  $48 \pm 15.2$  years (mean  $\pm$  standard deviation). Median age for men was statistically lower at 40, interquartile range (IQR) 30, than that for women: 52, IQR 23 years, Mann-Whitney U test,  $p < 0.05$ . There were 13 participants aged over 65 (the Polish retirement age) – nine women and four men.

96 participants lived in highly urbanised communities (all at  $54^\circ\text{N}$ , in and around the Tri-City area, *i.e.* Gdańsk, Sopot, and Gdynia).

General health was self-assessed as 'satisfactory' by 24, 'good' by 57, and 'very good' by 24 participants. Three subjects reported being able to climb only one flight of stairs without resting, all others reported at least two. 12 study participants were active, and 18 former, smokers. 32 exercised more frequently than once a month. 44 subjects did not take any chronic medication. 67 study participants denied suffering or having suffered from any of the diseases mentioned in the questionnaire, nine confirmed osteoporosis, three — rheumatoid arthritis, four — type 2 diabetes mellitus, two — asthma, 13 — gastrointestinal system diseases, seven — depression, two — kidney disease, five — psoriasis, and five — cancers (one skin, one prostate, one kidney, two cervical).

A control group of 40 healthy study participants, 25 men and 15 women, was selected, as described above.

**Table II.** Percentages of subjects according to 25(OH)D concentration ranges

**Tabela II.** Odsetek badanych w zależności od zakresów stężeń 25(OH)D

25(OH)D [ng/mL]	% of subjects in winter	% of subjects in autumn
$\geq 30$	1.8	14.7
20–30	10.1	43.1
10–20	50.5	39.4
$< 10$	37.6	2.8

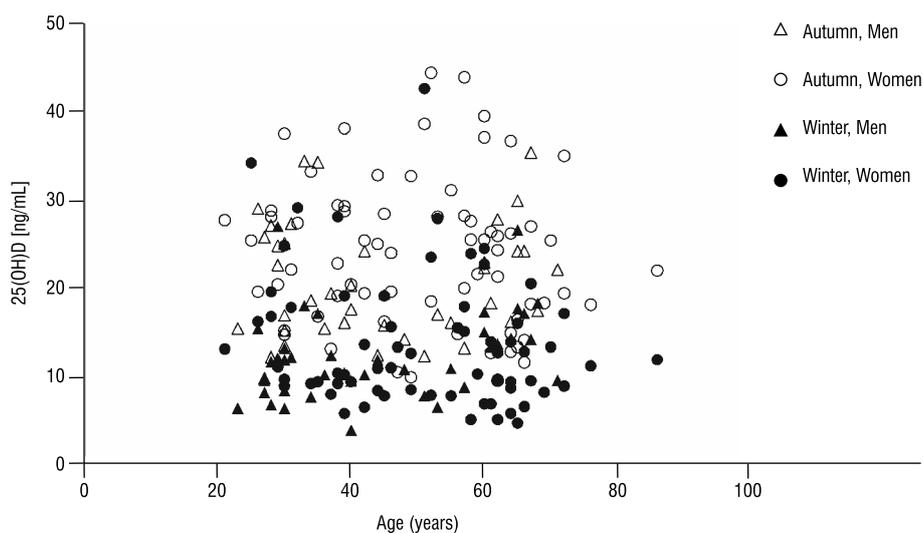
The number of examined subjects in both winter and autumn was 109

Their median age (35, IQR 12 years) was significantly lower than that of the remaining subjects (58). No statistically significant differences were found in comparisons of BMI values, 25(OH)D, PTH, Ca, P, and ALP levels between healthy controls and other subjects in either examination period.

### Vitamin D status

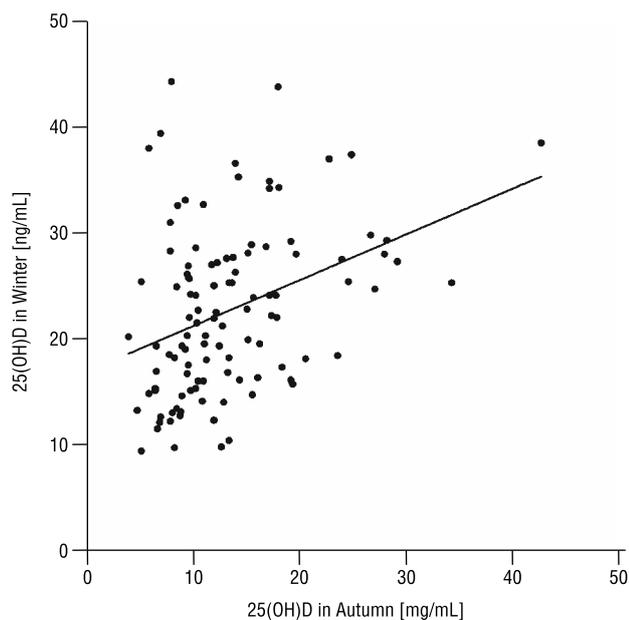
In winter 88.1%, and in autumn 42.2%, of subjects were vitamin D-deficient. Sufficiency (25(OH)D  $\geq 30$  ng/mL) was recorded in 1.8% and 14.7% of study participants in the respective study series (Table II and Fig. 1). Median 25(OH)D concentration was higher in autumn compared to winter ( $p < 0.0001$ , Table I).

A positive correlation was found between autumn and winter 25(OH)D concentrations: Spearman rank order correlation coefficient equalled 0.37,  $p < 0.05$  (Fig. 2). Assuming participants' habits concerning UVB



**Figure 1.** 25(OH)D serum levels in winter and autumn versus age. Median 25(OH)D concentrations were higher in autumn compared to winter,  $p < 0.0001$  (Mann-Whitney U test and Wilcoxon matched-pairs signed rank tests)

**Rycina 1.** Stężenie 25(OH)D zimą i jesienią w odniesieniu do wieku. Mediana stężeń 25(OH)D jesienią była wyższa niż zimą,  $p < 0.0001$  (test U Manna-Whitneya i test Wilcoxon dla par obserwacji)



**Figure 2.** 25(OH)D serum levels for each study participant in autumn versus winter. Spearman rank correlation coefficient  $r = 0.37$ ,  $p < 0.0001$ , 95% confidence interval: 0.19–0.53

**Rycina 2.** Stężenia 25(OH)D każdego probanta jesienią wobec zimy. Współczynnik korelacji rang Spearmana  $r = 0.37$ ,  $p < 0.0001$ , 95% przedział ufności: 0.19–0.53

exposure in summer did not change between 2011 and 2012, the correlation of winter vitamin D serum levels with those of autumn hints strongly at the importance of sunlight on 25(OH)D status.

### PTH and 25(OH)D

In winter, 20% of subjects (22 out of 109) had PTH concentrations above the upper reference range limit of 62 pg/mL (PTH values of 97, 113 and 123 were found in three women, while the remaining elevated PTH concentrations were lower than 85 pg/mL). In the autumn examination series, nine study participants had elevated PTH serum values (with only one subject with a value exceeding 85 pg/mL: 117 pg/mL).

A negative correlation (Spearman rank correlation coefficient  $r = -0.31$ ,  $p < 0.05$ ) was found between PTH and 25(OH)D concentrations in the autumn examination.

Median PTH concentration was lower in autumn than in winter (Table I).

In both study series (winter and autumn), vitamin D supplementation did not affect subjects' PTH concentrations.

### Calcium, phosphate and ALP data

No statistically significant differences were found in ALP activity, calcium, and phosphorus concentrations between measurement series, between men and women, in respective age groups (Table I). Nor were

**Table III.** 25(OH)D serum level and vitamin D supplementation  
**Tabela III.** Stężenie 25(OH)D a suplementacja witaminy D

		Supplementation	
		No	Yes
Winter	n [total (men)]	74 (29)	27 (6)
	25(OH)D [ng/mL]		
	Mean	12.4	16
	SD	5.9	8
Autumn	n [total (men)]	79 (34)	30 (1)
	25(OH)D [ng/mL]		
	Mean	22.2	24.3
	SD	7.5	9
	Median	22.1	24.9 (*)
	IQR	11.2	12.5

\* $p < 0.05$  for 25(OH)D median concentrations of Vitamin D supplementing vs not-supplementing subjects (Mann-Whitney U test)

any correlations found between these three parameters and 25(OH)D, PTH, BMI, and age.

### Vitamin D supplementation

Participants who declared vitamin D supplementation had higher median 25(OH)D levels in both measurement series (*i.e.* winter and autumn, see Table III).

Significantly more women (29) *vs.* men (only one) declared taking vitamin D supplements in the autumn examination: both in general and in all age-groups (Table IVb).

Despite the fact that the vast majority of study participants were vitamin D-deficient in the winter period, the number of subjects supplementing vitamin D decreased slightly in the autumn series of the study compared to winter (Table V).

There were no differences in median 25(OH)D concentrations in persons who declared higher or lower frequency of supplementation (answers available in the questionnaire were: '5–7 times weekly', '< 5 times weekly' and '< 9 times per month').

### UVB exposure

In the winter examination series, participants who declared recent (*i.e.* after 1<sup>st</sup> October) natural UVB exposure had higher median 25(OH)D concentration compared to those who declared no sun-tanning or who had tanned before this date: 19.1 ( $n = 13$ , IQR = 15.2) *vs.* 13.5 ( $n = 33$ , IQR = 5.7 ng/mL) (Mann-Whitney U test,  $p < 0.05$ ). However, among the sun-tanning-declaring subjects there were two women who also

Table IV. 25(OH)D serum levels and BMI values in men and women in the winter (A) and autumn (B) examination series

Tabela IV. Stężenia 25(OH)D i wartości BMI mężczyzn i kobiet w serii zimowej (A) i jesiennej (B) badania

## A. 25(OH)D, BMI and vitamin D supplementation in winter

Sex	Age group (years)	Both					Men			
		All	< 40	40–60	> 60	All	< 40	40–60	> 60	
25(OH)D [ng/mL]	n	109	69	20	24	25	40	20	9	11
	Mean	13.3	13.8	15.6	14.6	11.6	12.5	11.7	9.9	16.1
	SD	6.6	7.4	8	8.4	5.3	5.1	4.9	4.3	4.3
	Median	11.9	11.2	12.1	13	9.7	11.9	11.1	9.2	15(*)
	IQR	7.3	7.9	9.8	8.3	5.7	6.2	4.5	3.1	4
BMI [kg/m <sup>2</sup> ]	n	103	64	20	20	24	39	20	8	11
	Mean	26.1	25	23.7	24.3	26.8	28	25.9	30.8	29.7
	SD	4.5	4.2	3.5	3.7	4.6	4.5	4.1	5	2.9
	Median	25.5	24.8	23.5	23.3	25.9	27.2(#)	25.3	30.4(##)	29.1(###)
	IQR	5.7	5.3	4.4	5.1	3.2	6.7	3.9	5.3	5.2
Vit. D supplements [n]		27	21	4	5	12	6	2	1	3

\*p &lt; 0.05; #p &lt; 0.005; ##p &lt; 0.005; ###p &lt; 0.05

## B. 25(OH)D, BMI and vitamin D supplementation in autumn

Sex	Age group (years)	Both					Men			
		All	< 40	40–60	> 60	All	< 40	40–60	> 60	
25(OH)D [ng/mL]	n	109	69	20	24	25	40	19	10	11
	Mean	22.8	24	24.9	25.3	22.1	20.7	21.2	16.3	23.7
	SD	7.9	8.4	7.2	9.1	8.5	6.6	7.3	3.7	5.7
	Median	22.1	24.3	26.4	25.2	21.3	19(*)	19.4	16(**)	22.9
	IQR	11.5	10.2	9.7	10.3	11.3	9.4	11.7	4.4	9.5
BMI [kg/m <sup>2</sup> ]	n	109	69	20	24	25	40	19	10	11
	Mean	25.9	24.8	24	24	26.1	27.9	25.5	29.9	30.2
	SD	4.3	3.8	3.8	3.2	4.1	4.5	4.5	3.9	3.3
	Median	25.4	24.2	23.3	23	26	27(#)	24.2	29.4(##)	30.8(###)
	IQR	5.1	4.9	4.9	4.8	3	7.1	3.4	4.9	5.8
Vit. D supplements (n)		30	29	7	8	14	1(\$)	1(\$)	0(\$)	0(\$)

\*p &lt; 0.05; #p &lt; 0.001; \*\*p &lt; 0.005; ##p &lt; 0.001; ###p &lt; 0.01; \$ significantly more women than men took vitamin D supplements in general and in all age groups, p &lt; 0.05. Symbols indicate statistically significant comparisons between men and women in respective age groups (Mann-Whitney U test)

used a sunbed after 1<sup>st</sup> October. When these two female participants were excluded from the above comparison, there was no statistically significant difference between those who declared sun- (but not sunbed-) tanning and those who did not.

Significantly higher median 25(OH) serum level was demonstrated in those who declared sunbed tan-

ning after 1<sup>st</sup> October *vs.* those who either did not tan or tanned before 1<sup>st</sup> October: 24.8 (n = 5, IQR 11.9) *vs.* 11.7 ng/mL (n = 87, IQR 6.6), p < 0.05.

There were no differences in 25(OH)D concentrations related to declared frequency and duration of UVB exposure in the autumn study series (frequency of sun exposure lasting more than 15 minutes daily as

**Table V.** Vitamin D-deficient and vitamin D-supplementing subjects according to changes in 25(OH)D levels between winter and autumn examination series**Tabela V.** Osoby z hipowitaminozą D i osoby suplementujące witaminę D w zależności od zmiany stężenia 25(OH)D między zimową i jesienną serią badania

Change in 25(OH)D [ng/mL]	< 2	2–6	6–10	10–14	14–18	≥ 18
Percentage of subjects (%)	12.4	22.5	16.9	16.9	15.7	14.6
Number of subjects	11	20	15	15	15	13
No. of Vit. D-deficient subjects in winter	9	19	15	15	14	13
No. of Vit. D-deficient subjects in autumn	10	17	10	3	0	0
No. of Vit. D supplementing subjects in winter	5	4	6	2	4	5
No. of Vit. D supplementing subjects in autumn	3	3	5	2	6	5

Participants who reported sunbed- or sun-tanning after 1 October in the winter examination series were not included in this table; thus data of 89 subjects is presented here: 53 women (mean age 50, SD 14; median age 57, IQR 22 years) and 36 men (mean and median age 56, SD 17 and IQR 30 years). Vitamin D deficiency was defined as serum 25(OH)D concentrations lower than 20 ng/mL

well as duration of sun exposure in preceding months were investigated in the questionnaire).

As mentioned above, median 25(OH)D levels were higher in the autumn examinations (compared to winter series). To investigate the influence of natural, ambient UVB radiation on vitamin D status, we compared 25(OH)D levels recorded in both examination series only among those subjects who did not sunbathe or used a sunbed in the 4–5 months prior to the winter examinations, since we assumed they might have had 'unnaturally' higher vitamin D status in winter; there were 20 subjects who declared such UV exposure and they were excluded from this analysis. Among the remaining participants (*i.e.* not UVB-exposed after the 1<sup>st</sup> October that preceded winter testing), between the winter and autumn examinations an increase in 25(OH)D serum levels of at least 10 ng/mL was recorded in 43 participants, and of 2 to 10 ng/mL in 35 individuals (Table V). The effect of ambient sunlight on vitamin D status is apparent in the vast majority of subjects. However, there were 11 study participants whose 25(OH)D levels were almost unchanged — most remained vitamin D-deficient in autumn too (Table V).

### Diet

There were no differences in 25(OH)D concentration depending on declared vitamin D-rich foods and/or dairy intake (data not shown).

### Age, sex, BMI and 25(OH)D serum levels

A higher median 25(OH)D concentration was found in women compared to men in the autumn examination: 24.3 (IQR = 10.20) *vs.* 19 ng/mL (IQR = 9.35). In the 40–60 year-olds group, the same observation was made (25.2 *vs.* 16 ng/mL) (Table IVB). No differences in 25(OH)D levels between participants of opposite sexes were found in the winter series of examinations, with

the exception of the eldest study participants, where median vitamin D concentration was higher in men (Table IVA).

In the same group of 89 subjects mentioned above (excluding participants who sunbathed and/or used a sunbed between 1<sup>st</sup> October and the winter examination series), the median change in 25(OH)D concentrations between the two series of measurements (winter *vs.* autumn) was higher for women (totalling 53), at 11 ng/mL, IQR 11.7, than for men (*n* = 36): 6.8, IQR 9 ng/mL, *p* < 0.05 (Mann-Whitney U test).

Women (all and those older than 40) had lower median BMI than men (in age-matched groups) (Table IV). There was a weak negative correlation between the 25(OH)D concentrations in autumn and BMI values (Spearman rank order correlation coefficient *r* was –0.25, *p* < 0.05).

Also, BMI negatively correlated with the change in 25(OH)D concentration values recorded in winter and autumn for each participant: *r* = –0.31; after excluding 20 study participants, who reported sun-tanning or sunbed use after 1<sup>st</sup> October in the winter measurement series, the coefficient was –0.40 (*p* < 0.05, Spearman rank order correlation).

### Discussion

The results obtained here demonstrate widespread vitamin D deficiency in both winter (88.1%) and autumn (42.2%) measurement series among enrolled subjects. Similar data has been reported in other studies involving white adults from Poland and from other countries in the temperate climate zone [10–12].

In a study by Webb et al., 109 ambulatory participants (85 female, 24 male, mean age 44, IQR 34–51 years, median BMI 24.9, IQR 22.7–28 kg/m<sup>2</sup>), living in Greater Manchester (53.48°N) had the lowest annual

mean 25(OH)D serum levels in February:  $18.3 \pm 8.7$  ng/mL (while for March ca. 20 ng/mL), which is comparable to the results of our study [11]. For October, these authors reported a mean 25(OH)D serum level of approximately 28 ng/mL, which was similar to our result for the six-week autumn period (late September to early November). One possible reason for differences in recorded concentrations are different study groups, *i.e.* participants older than 60, who have lower vitamin D synthesis capability, were excluded by Webb et al. [14].

In another British study, 365 healthy postmenopausal women recruited by Mavroeidi et al. in Aberdeen (57°N) were examined consecutively at fixed, three-monthly intervals [15]. Apart from taking cod liver oil, participants supplementing vitamin D were excluded. Mean winter (December, January, February) and spring (March, April, May) 25(OH)D serum concentrations were ca. 16 ng/mL, while autumn levels were ca. 20.2 ng/mL, which correspond quite well with our data reported for women aged 60 and above.

In Poland, 132 citizens of Krakow (50°N): 82 females and 50 males aged 41–81 years (median 62) were examined during “winter” (the exact time span was not reported, unfortunately) [16]. Median 25(OH)D serum level was 16.7 ng/mL and vitamin D deficiency was recorded in 90.2% of study participants.

In another Polish study, surprisingly minor seasonal variations in mean vitamin D concentrations, 25.5 and 23.6 ng/mL in summer and winter respectively, were found in 50 healthy, pregnant women [17]. Summer concentrations were similar to those found here, in particular for women aged below 40.

In our study, vitamin D supplementation clearly had a positive effect on 25(OH)D serum levels (Table III). Although we did not record the doses of vitamin D in supplements taken by study participants, it can be stated that supplementation was insufficient. In the above-mentioned Polish study involving healthy pregnant women, a 1 ng/mL difference in mean serum 25(OH)D level was found in winter in favour of those who took supplements, while there was a 4.1 ng/mL difference in summer. 71% of the women took a daily vitamin D<sub>3</sub> dose of 400 IU, the rest 200, 250, 500 or 800 IU. In the Endocrine Society Clinical Practice Guidelines from 2011, an estimate is given of a 1 ng/mL increase in serum 25(OH)D level for each supplemented 100 IU of vitamin D [5]. However, it is possible that this increase is significantly underestimated, as discussed by McKenna and Murray [18].

In our study, apart from the results regarding sunbed tanning, the questionnaire data concerning sunlight did not statistically significantly indicate its importance on vitamin D status. While this is understandable in the case of the February to April examination, since only

13 participants reported sun-tanning after 1<sup>st</sup> October (*i.e.* in the 4-5 month period preceding the winter examination), in the case of summer data the questions assessing sunlight exposure used in our examination were probably too general. These were: the amount of time spent outdoors of either < 30, 30–60 or > 60 minutes; and the number of days per week in which there had been sun exposure of at least 15 minutes in the preceding months.

On the other hand, in both series of the study, apart from 109 subjects analysed here, we recruited participants who were examined once only (either in winter or in autumn). In both examination series, significant differences were found in serum 25(OH)D levels depending on declared sun exposure. In the case of the winter series, a total of 448 subjects was recruited (including the 109 analysed here); the paper reporting our findings was accepted for publication in “Endokrynologia Polska”. Autumn measurements included 304 participants.

Questionnaires have also been used to assess the influence of sunlight on 25(OH)D levels by other authors. Vitamin D status in Italian hospital employees in summer could be predicted using a scoring system which assessed the sun-exposed skin area and the amount of time spent outdoors in the preceding week [19]. Also, a simple question posed to adolescent girls and postmenopausal women: “Do you avoid the sun, do you sometimes stay in the sun, or do you prefer to stay in the sun?” was sufficient to indicate statistically higher 25(OH)D values in the third group compared to the others [20].

However, the results of the present study also indirectly hint at the importance of natural UVB exposure on vitamin D status. Firstly, a positive, moderate correlation in the Spearman rank test was found between 25(OH)D concentrations in autumn and winter of every study participant. Taking into account the insufficient supplementation (in both study periods), it is the natural sun exposure which may account for this result. Secondly, among subjects who had higher 25(OH)D concentration variation ( $\geq 10$  ng/mL) between winter and autumn, only three out of 43 were vitamin D-deficient in autumn; whereas 80.4% of participants whose concentrations changed by less than 10 ng/mL were deficient after months of high UVB radiation (see Table V). Finally, a moderate negative correlation between BMI and change in 25(OH)D concentrations was found, and, also, lower median 25(OH)D concentration and higher median BMI were recorded for men compared to women in autumn. These findings can be explained by the fact that vitamin D synthesised due to UVB (sunlight) exposure becomes diluted in adipose tissue [21, 22].

As previously reported, our data also showed a negative correlation between 25(OH)D and PTH

concentrations (in the case of the autumn examination) [23, 24]. Spearman rank correlation coefficient of  $-0.31$  was recorded here; weaker correlations:  $-0.13$  and  $-0.21$  were found in winter by, respectively, Napiórkowska et al. and Trofimiuk-Muldner, Kieć-Klimczak, and Hubalewska-Dydejczyk [25, 26].

Although renal function was not evaluated in our study, which makes excluding secondary hyperparathyroidism due to renal disease impossible, only two study participants reported having (or having had) “kidney disease”; one had increased serum PTH concentration.

## Conclusions

Our study delivers important data on the vitamin D status of urban adults in northern Poland (at the latitude of  $54^{\circ}\text{N}$ ).

We demonstrated that vitamin D deficiency among mostly young and middle-aged (67% of subjects were less than 60 years old), non-osteoporotic subjects was common already in autumn, *i.e.* 8–10 weeks following the presumed nadir 25(OH)D concentrations achieved in August to early September, and widespread in winter.

Also, similarly to other studies, our findings show vitamin D deficiency must be considered a viable cause of elevated parathyroid hormone concentrations.

Bearing in mind the accumulating evidence of deleterious health outcomes of vitamin D deficiency, further large-scale studies are necessary to ascertain the scope of deficiency in Europe and to implement effective prophylaxis, monitoring and treatment programmes.

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