Low levels of calcium or vitamin D – which is more important in systemic lupus erythematosus patients? An extensive data analysis

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Abstract

Objective

Several reports have indicated an association between systemic lupus erythematosus (SLE) and low levels of vitamin D. We examined several blood work parameters in SLE patients and controls and performed an extensive data analysis in order to investigate the links between blood levels of calcium, vitamin D, and SLE disease.

Methods

4,278 SLE patients and 16,443 age and sex-matched controls were selected from a national health insurer database in Israel. Patients with no blood work results or having renal disease were excluded. Retrospective data from five consecutive years of routine blood work results were then analysed for mean serum calcium, albumin, albumin-corrected calcium, vitamin D levels, and the presence of a hypocalcaemic episode (Corrected Ca <8.5 mg/dL).

Results

The mean levels of corrected serum calcium levels were slightly higher among SLE patients than controls (9.23±0.34 vs. 9.19±0.36 mg/dL p≤.001 respectively). In contrast to results of published studies, SLE patients had slightly higher levels of 25(OH)-vitamin D (SLE patients: 22.2±9.06 ng/ml, controls: 20.0±8.76 ng/ml, p≤.001). The most impressive finding entailing SLE patients was that they were twice as likely to experience episodes of hypocalcaemia in comparison to controls (SLE patients: 13.8%, controls: 6.4%, OR 2.34; 95% CI 2.33-2.83).

Conclusion

Calcium levels may play a significant role in the SLE disease process, more than originally thought, since SLE patients are at a higher risk for hypocalcaemic events. Specific changes in vitamin D and calcium homeostasis in SLE patients may be responsible for the severity of symptoms. Further research is required to determine the role of calcium supplementation.

Key words

calcium, vitamin D, hypocalcemia, lupus, systemic lupus erythematosus, autoimmune diseases

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Introduction

Systemic lupus erythematosus (SLE) is a multi-systemic autoimmune disease (1, 2). Like many other autoimmune diseases its aetiology is unclear, and several predisposing factors have been investigated extensively over the years (3). Medications, smoking, ultra-violet (UV) radiation exposure, and viral infections have been among the environmental factors suggested to influence the aetiology and clinical course of SLE (4, 5). Additionally, vitamin D is also known to be involved in SLE disease process (6, 7).

Vitamin D is known to have an immunomodulatory role in various processes of the immune system (7-9). Previous studies suggested that vitamin D deficiency is more common in SLE patients than in healthy subjects (7, 10-12). Vitamin D deficiency was also previously linked to a higher disease activity, inflammatory marker levels and titer of autoantibodies (10, 13-15). However, interventional studies with vitamin D supplementation are inconsistent in regards to its efficacy, and one study did manage to show amelioration in disease activity after vitamin D supplementation (16, 17).

Vitamin D is a key regulator of calcium and phosphate stores in the human body; It increases the absorption of calcium and phosphate from the intestine by 30–40% and 80% respectively (18). Vitamin D deficiency may greatly influence calcium status. We can postulate that some symptoms of SLE that are related to vitamin D deficiency may actually arise from the low levels of serum calcium, and therefore, by correcting the calcium levels we may influence the course of the disease.

There is little evidence, if any, on calcium levels and hypocalcaemia events among SLE patients. Our goal in this project was to examine if there is an association between SLE disease, serum calcium concentration, hypocalcaemic events and vitamin D levels in a large-scale population-based study.

Methods

The data for this study were collected from the database of "Clalit Health services" (CHS), the largest state-mandated health service organisation in Israel. CHS has over four million members, which constitute over 50% of Israel's population. All members over 18 years of age diagnosed with SLE were included (n= 5,018) and each member was matched with four age and sex-matched controls (n=20,090).

The exclusion criteria included: renal failure and the lack of recent biochemical analysis results. Thus, subjects with renal failure (defined as a single creatinine measurement over 1.5 mg/dL), as well as thus with no biochemical analysis results (serum vitamin D, albumin and calcium levels) in recent years were excluded from the analysis, resulting in a total of 4,278 SLE patients and 16,443 (Tables I-II). We have also categorised the vitamin levels into three categories, normal levels (>30 ng/ml), insufficiency (20–30 ng/ml) and deficiency (<20 ng/ml).

The CHS computerised database includes all medical records of its members and encompass past and present diagnoses. The diagnosis of SLE was determined by physicians either in a community-based clinic, or in a hospital throughout the country.

The mean levels of serum calcium, albumin and vitamin D were calculated for each patient from all blood results taken over the course of five years, between January 1st 2010 and December 31st 2014. The serum calcium levels of the subjects were corrected to serum albumin levels, using a standard correction formula. Mean levels of serum calcium and vitamin D were compared between patients and controls using a Student's t-test. Odds ratios (OR) were calculated with 95% confidence interval. Additionally, hypocalcaemia episode was defined as any measurement of corrected serum calcium level under 8.5 mg/dL. The incidence of events of hypocalcaemia was compared between SLE patients and controls using a Chi-square test. Statistical analysis was performed using R Statistical Software (v. 3.2.2; R Foundation for Statistical Computing, Vienna, Austria).

Results

Our study involved a total of 20,721 subjects, 4,278 SLE patients and 16,443 controls after the exclusion of subjects

Table I. Demographic and clinical characteristics of the study population.

Variable	Controls n=16443	SLE patients n=4278	OR	*p*-values
Age	47.8 ± 14.6	51.1 ± 16.2	1.01 (1.01;1.02)	<0.001
Gender:				
Female	13992 (85.1%)	3583 (83.8%)	1	1
Male	2451 (14.9%)	695 (16.2%)	1.11 (1.01;1.21)	0.031
BMI	27.0 ± 5.94	27.0 ± 6.07	1.00 (0.99;1.00)	0.556
SES:				
Low	6851 (41.7%)	1696 (39.7%)	1	1
Medium	6233 (37.9%)	1645 (38.5%)	1.07 (0.99;1.15)	0.099
High	3345 (20.4%)	935 (21.9%)	1.13 (1.03;1.24)	0.008

SLE: systemic lupus erythematosus; BMI: body mass index, kg/m^2; SES: socioeconomic status; OR: odds ratio.

Table II. Blood work results of SLE patients and controls.

Variable	Controls n=16443	SLE patients n=4278	OR	*p*-values
Calcium (mg/dL)	9.19 ± 0.36	9.23 ± 0.34	1.35 (1.22;1.49)	<0.001
Creatinin (mg/dL)	0.70 ± 0.15	0.72 ± 0.16	1.76 (1.41;2.20)	<0.001
Albumin (g/dL)	4.25 ± 0.31	4.13 ± 0.34	0.32 (0.29;0.36)	<0.001
Hypocalcaemia event	1052 (6.4%)	589 (13.8%)	2.34 (2.10;2.60)	<0.001
Vitamin D categories (ng/ml)	20.0 ± 8.76	22.2 ± 9.06	1.03 (1.02;1.03)	<0.001
Deficiency (<20)	3422 (52.1%)	1018 (40.7%)	1	1
Insufficiency (20-30)	2337 (35.5%)	1037 (41.5%)	1.49 (1.35;1.65)	<0.001
Normal (>30)	815 (12.4%)	445 (17.8%)	1.84 (1.60;2.10)	<0.001

Vitamin D levels in ng/mL, ref. 30-100, hypocalcaemia event is defined as one or more events of documented corrected calcium <8.5 mg/dL.

with renal failure and with no blood work results (vitamin D, albumin and calcium levels) (Table I). Although initially matched for age, controls were slightly younger than SLE patients, due to exclusion of subjects from the analysis.

The mean levels of corrected serum calcium levels were slightly higher among SLE patients than controls (SLE patients: 9.23±0.34 mg/dL, controls: 9.19±0.36 mg/dL, $p \leq .001$). SLE patients in our study had higher mean levels of vitamin D than controls, although both groups had vitamin D insufficiency (22.2 *vs*. 20.0 ng/ml, respectively) (Table II). Dividing the vitamin D levels into three categories we found normal levels (17.8% of SLE patients *vs*. 12.4% of controls, $p<0.001$), insufficiency levels (41.5% of SLE patients *vs*. 35.5% of controls, $p<0.001$), and deficiency levels (40.7% of SLE patients *vs* 52.1% of controls, $p<0.001$). Our most interesting finding arose from our analysis of single events of hypocalcaemia. We detected 13.8% of SLE patients had at least one event of corrected serum calcium levels below 8.5 mg/dL, in comparison to only 6.4% in the controls (OR 2.34, 95% CI 2.10–2.60) (Table II).

Discussion

We examined the relationship between hypocalcaemic events, total serum calcium, and vitamin D levels in SLE patients. The link between low levels of vitamin D, specifically 25(OH)-vitamin D and SLE has been established in several previous studies (19–21). In these studies the percentage of SLE patients who were 25(OH)-vitamin D deficient or insufficient was between 65%–77%. In contrast, vitamin D deficiency in the general population varied greatly due to the age and exposure to sunlight. In one study from Boston, employing records from a bone health clinic, 41% of their patient population was found to be deficient of 25(OH)-vitamin D (22). In other studies it has been estimated that 25(OH)-vitamin D deficiency affects more than one billion people worldwide (18, 23, 24).

Vitamin D and calcium homeostasis are integrally linked, and calcium role in the pathogenesis of SLE warrants investigation. We aimed to determine the association between serum calcium, vitamin D levels, hypocalcaemic events and SLE. Our study included nearly 4,300 patients with SLE and around 17,000 controls, and we found that SLE patients were twice as likely to experience at least one hypocalcemic event (Corrected Ca<8.5mg/dl) on a routine blood work (OR 2.34) (Table II). Interestingly, in contrast to most results of published articles, we found that the mean levels of 25(OH)-vitamin D were higher in the SLE patient than controls (SLE patients: 22.2±9.06, controls: 20.0±8.76 ng/ml, $p \leq .001$) (Table II). Additionally when the levels of vitamin D were divided into three categories we have noticed that normal, insufficient levels were observed more commonly in SLE patients but not deficient levels that were more common in controls.

One study evaluated bone mineral density and biochemical parameters of bone metabolism in ambulatory premenopausal female patients with SLE, and reported that levels of 25(OH)-vitamin D were not different than healthy controls, but the levels of corrected serum calcium were found to be higher in the SLE group (25). A study performed in Texas found that only 20% of their SLE patients had deficient levels of 25(OH)-vitamin D (26). Both our patients and control populations had mean levels of 25(OH)-vitamin D which are classified as insufficient (<30 ng/mL). Low levels of 25(OH)-vitamin D are fairly common in countries with large percentages of individuals who cover their body for religious reasons, in particular the Middle East (27, 28). In the "CARMA" study, which as a cross-sectional study performed to investigate the association between 25-(OH)-vitamin D levels and the clinical characteristics of patients with chronic inflammatory rheumatic diseases (CIRD) the following median levels of vitamin D were reported; 20.4 (14.4–29.2) ng/ml in rheumatoid arthritis (RA), 20.9 (13.1–29.0) in ankylosing spondylitis (AS), 20.0 (14.0–28.8) in psoriatic

arthritis (PsA), and 24.8 (18.4–32.6) ng/ml in non-CIRD patients and concluded that patients with RA had higher rates of 25(OH)D deficiency compared to non-CIRD controls (29). In contrast to this finding in our study we reported higher levels of vitamin D in SLE patients compared to controls. This probably reflects the results of a better medical education and higher rates of calcium and vitamin D supplementation in SLE patients. Interestingly, a study conducted by Tepper *et al*. in Israel in 2014 (30) the authors reported that that age, sun-exposure, seasonality did not significantly affect vitamin D levels.

In patients with SLE, those with higher levels of 25(OH)-vitamin D were found to have less disease activity than their deficient counterparts according to the SLE disease activity index (SLEDAI) (13, 16, 31). Several theories exist as to why vitamin D plays a protective role in SLE and other autoimmune diseases; Lack of calcitriol is responsible for the pathological activation of the Th-1 response leading to autoimmunity (32). Studies *in vitro* demonstrated that lymphocytes cultured with calcitriol had reduced frequencies of IL-2-producing cells in the CD4+ as well as in the CD8+ population and revealed that about 70% of IL-6-producing CD4+ and CD8+ cells were also positive for IL-2, but more than 90% were negative for IFN gamma, IL-4, or IL-13 (33).

An alternative theory suggests that the vitamin D receptor (VDR) is somehow altered in SLE and other autoimmune diseases patients, leading to pathological activation of the immune system which may also interfere with calcium homeostasis (34–38). A recent meta-analysis included studies that associated the VDR gene BsmI, FokI, ApaI or TaqI polymorphism with SLE risk. This meta-analysis comprised eleven studies, with a total of 1621 cases and 1883 controls. It showed that the BsmI B allele was associated with the onset of SLE in the overall population (OR 1.726, 95%CI 1.214–2.455) and in Asians (OR 1.952, 95%CI 1.135–3.355). FokI FF genotype was correlated with the susceptibility of SLE for Asians (OR 1.469, 95%CI 1.005–2.148). FokI T/C and TaqI polymorphisms were not associated with SLE risk for Caucasians. There was no significant association between the ApaI polymorphism and SLE risk for the overall population, Asians and Caucasians (35). Calcitriol treatment has been shown to improve significantly murine SLE (39). VDR polymorphisms may explain why adequate levels of 25(OH)-vitamin D do not necessarily lead to improvement in SLE; Studies performed in VDR knocked out mice showed that their inflammatory bowel disease (IBD) was more severe than in normal counterparts, suffered from a plethora of immune abnormalities as well as hypocalcaemia typical of rachitic animals, and when calcium levels were restored, the immune defects were resolved (40, 41).

Drugs prescribed for lupus can influence conversion of 25(OH)-vitamin D to calcitriol as well as affect the intestinal calcium absorption. Corticosteroids have been employed in the treatment for SLE for several decades, and were reported to reduce calcium absorption (42). One mechanism reported to affect the hemostasis of calcium and vitamin D is by increasing the conversion of calcitriol to a more polar and inactive form and to decrease the absorption in the intestines (43). Hydroxychloroquine (HCQ) is another drug commonly employed in SLE which affects 1-alpha hydroxylase activity is. In patients with sarcoidosis HCQ was shown to greatly reduce the levels of calcitriol without affecting levels of serum 25(OH)-vitamin D (44). Both corticosteroids and HCQ could lower the levels of calcium leading to hypocalcaemia through decreased absorption without significantly altering serum levels of 25(OH)-vitamin D.

In conclusion, calcium levels may play a more significant role in the SLE disease process than originally thought. SLE patients are at a higher risk for hypocalcaemic events. Specific changes in vitamin D and calcium homeostasis in SLE patients may be responsible for the severity of disease symptoms. Our data may support the need for calcium supplements and not only for vitamin D in order to prevent events of hypocalcaemia.

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