The incidence of type 1 diabetes mellitus in European countries has been increasing yearly, reaching several folds in the last three decades. It is supposed that in 2020 the number of new cases of type 1 diabetes mellitus will be doubled in children under five years old, and the prevalence will increase by 1.7 times in children under 15 years old age. HbA1c is a good indicator of metabolic control in diabetic patients, and it is less affected by food intake and daily activity as it is in other tests in addition to its predictive value for microvascular complication. Vitamin D is a fat-soluble vitamin usually obtained from exposure to sunlight. Also, it is found in very few foods or is given as a dietary supplement. The consumption of
Methods

A prospective quasi-experimental study was conducted to show the effect of vitamin D supplementation on HbA1c level in type 1 diabetic patients with vitamin D deficiency before and after vitamin D supplementation. The study was conducted in Layla Qasim diabetic center in Erbil city, the capital city of Kurdistan region of Iraq, from 1st September of 2019 till 1st December of 2019.

A systematic sampling method was used to select patients from a total of 984 registered type 1 patients aged less than 18 years old at Layla Qasim diabetic center. Every 10th patient was selected from the list of registered type 1 patients. Out of 98 patients interviewed, only 76 were tested for the initial vitamin D level. The 22 patients not involved in the study either refused or were excluded. The exclusion criteria included chronic renal disease, celiac disease, patients already on vitamin D supplementation, on medications that interfere with vitamin D metabolism, hypothyroidism, any acute illness such as acute febrile illness, recent documented infection or inflammation and recent admission to hospital for diabetic ketoacidosis. Out of 76 patients, only 50 patients were vitamin D deficient (25OHD: < 20) and were recruited. No other data was collected about those patients with sufficient vitamin D.

Recruited patients were assigned to receive vitamin D according to the safe upper limit dose recommended by the American Diabetic Association (1,000–1,500 IU/day for infants, 2,500–3,000 IU/day for children aged 1–8 years, and 4,000 IU/day for children ≥ 9 years of age, adults). Vitamin D was provided in the form of syrup or tablet of the same manufacturer according to age and acceptance of the patient.

No further therapeutic interventions were performed, and the family was instructed that the child should continue the same dose of insulin and lifestyle till the end of the study.

Then after 12 weeks from starting the supplementation, vitamin D level and glycosylated hemoglobin (HbA1c) were measured again. They were compared with the initial readings to evaluate the effect of vitamin D supplementation on HbA1c level. Both serum 25-hydroxy vitamin D and HbA1c level were measured using Cobas 6000 (Roche Diagnostics, Hitachi, Tokyo, Japan).

HbA1c levels were divided into three categories: first tertile (HbA1c<7.5%), second tertile (HbA1c:7.6%-9.9%), and third tertile (HbA1c≥10%).
The body mass index (BMI) was calculated by dividing weight (kg) over squared height (m²). The BMI-z score was graded according to CDC growth charts. The research proposal was accepted by the ethical committee of the Kurdistan Board of Medical Specialties. Informed consent taken from the caregiver of patients before participating in the study. The anonymity of patients was preserved. Data analysis was performed by using the statistical package for the social sciences (SPSS version 24). Student's t-test was used to compare the means of two groups (gender, residency regarding HbA1c level, and serum 25(OH)D). One-way analysis of variance (one-way ANOVA) test was used to compare means of more than two groups (age groups, duration categories, BMI, insulin regimens, insulin dose, regarding HbA1c level and serum 25(OH)D). McNemar test was used to compare the percentage of patients' glycemic control group before and after vitamin D supplementation.

**Results**

This study recruited 50 type 1 diabetic patients with vitamin D deficiency: 19 males (38%) and 31 females (62%). The patients' age range was 2-17 years (52% aged 2-10 years, 32% aged 11-14 years, and 16% aged 15-17 years) with a mean of 10.14 ± 3.93 years. Forty-eight patients (96%) were from urban residence, while only two patients (4%) were from rural residence. At the stage of recruitment, the mean duration of diabetes was 3.77 ± 3.19 years, as shown in Table 1.

**Table 1** Association between patients' variables with initial serum 25(OH)D

| Variables          | Mean ± SD* | No (%) | 25(OH)D (Mean ± SD) | P value |
|--------------------|------------|--------|---------------------|---------|
| Age (years)        |            |        |                     |         |
| 2-10               | 10.14 ± 3.93 | 26(52%) | 10.26 ± 3.47        | 0.541   |
| 11-14              | 10.00 ± 3.47 | 16(32%) |                     |         |
| 15-17              | 11.02 ± 4.05 | 8(16%)  |                     |         |
| Gender             |            |        |                     | <0.001  |
| Male               | 14.31 ± 3.19 | 19(38%) |                     |         |
| Female             | 8.67 ± 2.56  | 31(62%) |                     |         |
| Residency          |            |        |                     | 0.663   |
| Urban              | 10.77 ± 3.86 | 48(96%) |                     |         |
| Rural              | 12.00 ± 7.07 | 2(4%)   |                     |         |
| BMI (kg/m²)        | 18.25 ± 2.87 |        |                     | 0.189   |
| Normal             | 11.11 ± 3.95 | 43(86%) |                     |         |
| Overweight         | 9.00 ± 3.41  | 7(14%)  |                     |         |
| Duration (years)   | 3.77 ± 3.19  |        |                     | 0.405   |
| <4                 | 9.96 ± 3.70  | 30(60%) |                     |         |
| 4-8                | 10.26 ± 3.26 | 15(30%) |                     |         |
| 9-12               | 10.54 ± 5.27 | 5(10%)  |                     |         |
| Insulin regimen    |            |        |                     | 0.192   |
| Multiple injections| 37.56 ± 11.67 | 43(86%) |                     |         |
| Premixed two dose injection | 36.67 ± 19.15 | 6(12%)  |                     |         |
| Insulin pump       | 36.67 ± 19.15 | 1(2%)   |                     |         |

*Standard deviation, *Body mass index
Association between patients’ variables with initial HbA1c and serum 25(OH)D is shown in Table 1 and Table 2. The initial mean level of serum 25(OH)D among females (8.67 ± 2.56 ng/mL) was lower than males (14.31 ± 3.19 ng/mL). A statistically significant association was found between gender and initial serum 25(OH)D level \((P < 0.001)\). However, no association was found between the initial HbA1c level and gender \((P = 0.245)\).

No significant difference was found among age groups regarding initial serum 25(OH)D \((P = 0.541)\) and initial HbA1c \((P = 0.342)\). Body mass index was not associated with the initial serum 25(OH)D level \((P = 0.189)\) and initial HbA1c \((P = 0.545)\) value, respectively. There was no statistically significant association between initial serum 25(OH)D level and the initial HbA1c with the residency of the patients \((P = 0.663 \text{ and } 0.107, \text{ respectively})\).

About insulin regimens, 86% of patients were on multiple injections (mean 37.56±11.67), followed by 12% of patients on premixed two dose insulin injections (mean 36.67±19.15). The insulin pump was used only by 2% (mean of 65 units), as is shown in Table 1. No statistically significant difference was found among insulin regimens regarding initial HbA1c \((P = 0.178)\) and initial serum 25(OH)D \((P = 0.192)\), as it is shown in Table 2 and 1, respectively.

**Table 2** Association between patients’ variables with initial HbA1c

| Variables          | HbA1c (Mean ± SD) | \(P\) value |
|--------------------|-------------------|-------------|
| Age (years)        |                   |             |
| 2-10               | 8.15 ± 0.38       | 0.342       |
| 11-14              | 8.37 ± 0.58       |             |
| 15-17              | 8.28 ± 0.55       |             |
| Gender             |                   |             |
| Male               | 8.34 ± 0.45       | 0.245       |
| Female             | 8.18 ± 0.51       |             |
| Residency          |                   |             |
| Urban              | 8.22 ± 0.34       | 0.107       |
| Rural              | 8.80 ± 0.30       |             |
| BMI\(^*\) (kg/m2) |                   |             |
| Normal             | 8.29 ± 0.49       | 0.545       |
| Overweight         | 7.91 ± 0.27       |             |
| Duration (years)   |                   |             |
| <4                 | 8.22 ± 0.50       | 0.528       |
| 4-8                | 8.34 ± 0.45       |             |
| 9-12               | 8.06 ± 0.48       |             |
| Insulin regiment   |                   |             |
| Multiple injections| 37.56±11.67       | 0.178       |
| Premixed two dose injection | 36.67±19.15 |             |
| Insulin pump       | 65                |             |

\(*\text{Standard deviation, } ^*\text{ Body mass index}\)
The mean dose of vitamin D supplementation was 3860 ± 995 IU/day. The difference in the mean HbA1c level before (8.24 ± 0.49) and after (7.93 ± 0.67) vitamin D supplementation was statistically significant ($P = 0.032$). Also, the percentage of patients’ glycemic control groups was statistically significant after vitamin D supplementation ($P < 0.001$), as shown in Table 3.

By doing linear regression, it has been found that the change in the serum 25(OH)D level with the vitamin D supplementation was significantly associated with the grade of the response of post HbA1c level (the more increase in the serum 25(OH)D level, the lower post HbA1c level) ($B = -0.29$, $P = 0.003$), as shown in Table 4.

### Table 3 Glycemic control before and after vitamin D supplementation

| Glycemic control groups | Before supplementation | After supplementation | $P$ value |
|-------------------------|------------------------|-----------------------|-----------|
|                         | Count | (%) | Count | (%) |           |
| First tertile           | 8     | (16.0) | 12      | (24.0) | $<0.001$ |
| Second tertile          | 11    | (22.0) | 37      | (74.0) |           |
| Third tertile           | 31    | (62.0) | 1       | (2.0)  |           |
| Pre- HbA1c* (Mean ± SD) | 8.24 ± 0.49 | | Post- HbA1c (Mean ± SD) | 7.93 ± 0.67 | 0.032 |

* Glycosylated hemoglobin.

### Table 4 Regression analysis of the change in the serum 25(OH)D level predicting post HbA1c level

| HbA1c level after vitamin D supplementation | B     | 95 % CI     | SE B | $\beta$ | T     | $P$ value |
|--------------------------------------------|-------|-------------|------|---------|-------|-----------|
| (Constant)                                 | 8.941 | 8.33-9.545  | 0.300| 0.300   | 29.814| 0         |
| Vit D3 difference                          | -0.29 | -0.47-(-0.010) | 0.009| -0.410  | -3.112| 0.003     |

B: Unstandardized beta, CI: Confidence interval for unstandardized beta, SE B: Standard error for the unstandardized beta, $\beta$: Standardized beta, t: t-test statistic, $R^2=0.168$, Adjusted $R^2= -0.151$
Diabetes mellitus can lead to the development of several complications. Studies have shown the importance of good glycemic control in reducing the risk of micro and macro vascular complication in type 1 and type 2 diabetes mellitus. Risk for the development of diabetic complications, morbidity, and mortality can be minimized by decreasing HbA1c level. Serum 25(OH)D before vitamin D administration were lower among females (8.67 ± 2.56) ng/mL than males (14.31 ± 3.19) ng/mL (P <0.001) (table 1). A similar figure was also reported by Al-Agha. However, Mutlu found no difference in Serum 25(OH)D between males and females. This could be attributed to the difference in the cultural influences and traditions. Besides gender, none of other demographic (age, residence) and clinical factors (BMI and duration of disease) were found to have an effect on and initial serum 25(OH)D and initial HbA1c. Many studies did not find an association between demographic and clinical factors mentioned above with Serum 25(OH)D and HbA1c among patients less than 18 years old. Our results showed that mean pre intervention HbA1c level improved significantly from (8.24 ± 0.49) to (7.93 ± 0.67) after supplementation (P = 0.032) (Table3). A similar result was reported by Mohammadian that HbA1c level improved (from 9.73±1.85 to 8.55±1.91) after vitamin D supplementation (P <0.0001). A similar figure was also found by Aljabri. In addition to this result, our study showed improvement in patients number and percent in all glycemic groups (first tertile<7.5%, second tertile 7.6%-9.9%, third tertile ≥10%) after vitamin D supplementation (Table 3). In contrast, a study by El Baba showed no significant relationship between glycemic control and variation of vitamin D level. Vitamin D deficiency may contribute to the development of insulin resistance and the impairment of the secretion of insulin. Vitamin D reduces the excessive release of insulin (in reaction to increased blood sugar) by decreasing insulin resistance. Thus, it improves insulin sensitivity. Vitamin D deficiency is also correlated with β-cell dysfunction. Vitamin D enhances the conversion of pro-insulin as well as β-cells capacity for insulin production. In our study, improvement of glycemic control was observed only after 12 weeks; we do not know if the improvement of glycemic control after vitamin D supplementation would carry on further. This study had a number of limitations. The design did not include a control group (no placebo control). It is not known if the patients or their caregivers did any change of insulin dose without informing us during or after the vitamin D supplementation. Finally, the study had a small number of patients recruited.

Supplementing vitamin D to type 1 diabetic patients with vitamin D deficiency leads to improvement in glycosylated hemoglobin level. Also, we concluded that vitamin D level is affected by the gender of the patient. However, neither the patients’ age, BMI, nor the duration of diabetes affects vitamin D level or HbA1c level.

None.

None declared.

1. Kahanovitz L, Sluss PM, Russell SJ. Type 1 Diabetes - A Clinical Perspective. Point Care. 2017; 16(1):37–40. https://doi.org/10.1097/POC.0000000000000125.
2. Katsarou A, Gudbjörnsdóttir S, Rawshani A, Dabelea D, Bonifacio E, Anderson BJ, et al. Type 1 diabetes mellitus. Nat Rev Dis Primers. 2017; 3(1):1–7. https://doi.org/10.1038/nrdp.2017.16.
3. Rewers M, Ludvigsson J. Environmental risk factors for type 1 diabetes. Lancet. 2016; 387 (10035):2340–8. https://doi.org/10.1016/S0140-6736(16)30507-4.
4. Patterson CC, Gyürüs E, Rosenbauer J, Cinek O, Neu A, Schober E, et al. Trends in childhood type 1 diabetes incidence in Europe during 1969-2008: evidence of non-uniformity over time in rates of increase. Diabetologia. 2012; 55(8):2142–7. https://doi.org/10.1007/s00125-012-2571-8.

5. Harvard Medical School. Type 1 Diabetes Mellitus: What is it, Boston, USA: Harvard Medical School; 2018. https://www.health.harvard.edu/a_to_z/type-1-diabetes-mellitus-a-to-z. Accessed 2 Apr 2020.

6. Florkowski C. HbA1c as a Diagnostic Test for Type 1 Diabetes Mellitus - Reviewing the Evidence. Clin Biochem Rev. 2013; 34(2):75–83.

7. National Institutes of Health. Vitamin D: Fact Sheet for Health Professionals USA: NIH. https://ods.od.nih.gov/factsheets/VitaminD-HealthProfessional. Accessed 8 Apr 2020.

8. Christakos S, Hewison M, Gardner DG, Wagner CL, Sergeev IN, Rutten E, et al. Vitamin D: beyond bone. Ann N Y Acad Sci. 2013; 1287(1):45–58. https://doi.org/10.1111/nyas.12129.

9. Hummel D, Aggarwal A, Borka K, Bajna E, Kállay E, Horváth HC. The vitamin D system is deregulated in pancreatic diseases. J Steroid Biochem Mol Biol. 2014; 144(Pt B):402–9. https://doi.org/10.1016/j.jsbmb.2014.07.011.

10. Sung CC, Liao MT, Lu KC, Wu CC. Role of vitamin D in insulin resistance. J Biomed Biotechnol. 2012; 2012:634195. https://doi.org/10.1155/2012/634195.

11. Benetti E, Mastrocola R, Chiazzra F, Ningro D, D’Antona G, Bordano V, et al. Effects of vitamin D on insulin resistance and myosteatosis in diet-induced obese mice. PLoS One. 2018; 13(1):e0189707. https://doi.org/10.1371/journal.pone.0189707.

12. Ali R, Fawzy I, Mohsen I, Settin A. Evaluation of vitamin D receptor gene polymorphisms (Fok-I and Bsm-I) in TIDM Saudi children. J Clin Lab Anal. 2018; 32(5):e22397. https://doi.org/10.1002/jcla.22397.

13. Yang CY, Leung PS, Adamopoulos IE, Gershwin ME. The implication of vitamin D and autoimmunity: a comprehensive review. Clin Rev Allergy Immunol. 2013; 45(2):217–26. https://doi.org/10.1007/s12016-013-8361-3.

14. Abd-Allah SH, Pasha HF, Haggrass HA, Alghobashy AA. Vitamin D status and vitamin D receptor gene polymorphisms and susceptibility to type 1 diabetes in Egyptian children. Gene 2014; 536(2):430–4. https://doi.org/10.1016/j.gene.2013.12.032.

15. Sahin OA, Goksen D, Ozpinar A, Serdar M, Onay H. Association of vitamin D receptor polymorphisms and type 1 diabetes susceptibility in children: a meta-analysis. Endocr Connect. 2017; 6(3):159–71. https://doi.org/10.1530/EC-16-0110.

16. Al-Agha AE, Ahmad IA. Association among vitamin D deficiency, type 1 diabetes mellitus and glycemic control. J Diabetes Metab. 2015; 6(594). https://doi.org/10.4172/2155-6156.1000594.

17. Savastano S, Cadario F, Genoni G, Bellomo G, Bagnati M, Secco G, et al. Vitamin D deficiency and glycemic status in children and adolescents with type 1 diabetes mellitus. PLoS One. 2016; 11(9):e0162554. https://doi.org/10.1371/journal.pone.0162554.

18. Elsayed AM, Mohamed GA. Vitamin D deficiency and its correlation to hemoglobin A1C in adolescent and young adult type 1 diabetes mellitus patients. AAMJ. 2016; 14(2):76–80. https://doi.org/10.4103/1687-1693.192643.

19. Shin YH, Shin HJ, Lee YJ. Vitamin D status and childhood health. Korean J Pediatr. 2013;56(10):417–23. https://doi.org/10.3345/kjp.2013.56.10.417.

20. Martin T, Campbell RK. Vitamin D and diabetes. Diabetes Spectr. 2011; 24(2):113–8.

21. Kliegman RM. Nelson Textbook of Pediatrics. 20th ed. Philadelphia: Elsevier; 2016. P. 2777.

22. American Diabetes A. 2. Classification and Diagnosis of Diabetes: Standards of Medical Care in Diabetes-2018. Diabetes Care 2018; 41(Suppl 1):S13–27. https://doi.org/10.2337/dc18-S009.

23. Centers for Disease Control and Prevention, National Center for Health Statistics. CDC growth charts: United States. http://www.cdc.gov/growthcharts/. Accessed 30 May 2020.

24. Bergenstal RM. Glycemic variability and diabetes complications: does it matter? Simply put, there are better glycemic markers! Diabetes Care. 2015; 38:1615–21. https://doi.org/10.2337/dc15-er09.

25. Aljabri KS, Bokhari SA, Khan MJ. Glycemic changes after vitamin D supplementation in patients with type 1 diabetes mellitus and vitamin D deficiency. Ann Saudi Med. 2010; 30(6):454–8. https://doi.org/10.4103/0256-4947.72265.

26. Mutlu A, Mutlu GY, Özsu E, Çizmecioglu FM, Hatun Ş. Vitamin D deficiency in children and adolescents with type 1 diabetes. J Clin Res Pediatr Endocrinol. 2011; 3(4):179–83. https://doi.org/10.4274/jcrpe.430.

27. Omar M, Nouh F, Younis M, Younis M, Nabil N, Saad M, et al. Culture, Sun Exposure and Vitamin D Deficiency in Benghazi Libya. J Adv Med Med Res. 2018; 25(5):1–13. https://doi.org/10.9734/JAMMR/2018/39562.

28. Nansel TR, Lipsky LM, Iannotti RJ. Cross-sectional and longitudinal relationships of body mass index with glycemic control in children and adolescents with type 1 diabetes mellitus. Diabetes Res Clin. 2013; 100(1):126–32. https://doi.org/10.1016/j.diabres.2012.12.025.
29. Mohammadian S, Fatahi N, Zaeri H, Vakili MA. Effect of vitamin D3 supplement in glycemic control of pediatrics with type 1 diabetes mellitus and vitamin d deficiency. JCDR. 2015; 9(3):SC05–7. https://doi.org/10.7860/JCDR/2015/10053.6683.

30. El Baba K, Zantout MS, Akel R, Azar ST. Seasonal variation of vitamin D and HbA(1c) levels in patients with type 1 diabetes mellitus in the Middle East. Int J Gen Med. 2011; 4:635–8. https://doi.org/10.2147/IJGM.S23548.

31. Kavadar G, Demircioglu DT, Ozgonenel L, Emre TY. The relationship between vitamin D status, physical activity and insulin resistance in overweight and obese subjects. Bosn J Basic Med Sci. 2015; 15(2):62–6. https://doi.org/10.17305/bjbms.2015.399.

32. Karnchanasorn R, Ou HY, Chiu KC. Plasma 25-hydroxyvitamin D levels are favorably associated with β-cell function. Pancreas. 2012; 41(6):863–8. https://doi.org/10.1097/MPA.0b013e31823c947c.