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Research article

Investigating the Efficiency of Vitamin D administration with Buccal Spray in the Treatment of Vitamin D Deficiency in Children and Adolescents

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What is already known?
The main purpose of vitamin D therapy is to optimize serum 25-hydroxyvitamin D concentrations to improve bone homeostasis and decrease the risk of osteopenia and osteoporosis. Daily or weekly oral drops or a single large dose, either orally or through injection are used more frequently in the treatment of vitamin D deficiency. There are limited numbers of studies that were evaluated the effectiveness of buccal spray against other modes of vitamin D delivery in the treatment of vitamin D deficiency in children and the results of these studies are conflicting.

What this study adds?
Vitamin D3 supplementation with buccal spray and oral drops is equally effective in terms of raising serum 25-hydroxyvitamin D concentrations in short-term treatment of vitamin D deficiency.

Abstract
Aims: The aim of this study was to evaluate the efficiency of buccal spray form of vitamin D compared to single oral dose (stoss therapy) and oral drops therapy in the treatment of vitamin D deficiency.
Methods: Ninety healthy children and adolescents (3-18 years) with vitamin D deficiency [serum level of 25-hydroxyvitamin D (25OHD) < 12ng/ml] were randomized to receive vitamin D3 buccal spray (2000 U, n=30, group I) for 6-week period, oral drops (2000 U, n=30, group II) for 6-week period and a single oral dose (300 000 U) vitamin D 3 (n=30, group III). Serum calcium, phosphorus, ALP, PTH and 25OHD levels of the patients were measured at baseline and after the treatment (42^{th} day).
Results: All 3 groups had a significant increase in serum 25 hydroxyvitamin D (25OHD) concentrations ($p<0.001$). Serum 25OHD concentration in group I was 22.1 (17.8-28.2) ng/ml as compared to baseline value of 8.0±0.41ng/ml, with the mean increase of 15.6±1.3 ng/ml. On the other hand, in group II and group III, the mean serum 25OHD concentrations were 24.4 (20.6-29.6) ng/ml and 40.3 (29.4-58.4) ng/ml as compared to baseline value of 7.9±0.45 ng/ml and 7.6±0.47 ng/ml, with the mean increase of 17.3±1.1ng/ml, 34.3±3.2 ng/ml, respectively.
Conclusion: We conclude that vitamin D3 supplementation with buccal spray and oral drops is equally effective in terms of raising vitamin D concentrations in short-term treatment of vitamin D deficiency.
Keywords: Vitamin D, buccal spray, 25-hydroxyvitamin D, oral drops

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Introduction

Vitamin D is pro-hormone for active intestinal calcium absorption, and it plays a major role in maintaining calcium and phosphorous homeostasis and skeletal integrity (1). Deficiency of vitamin D leads to rickets, the failure of mineralization of growing bone in children and osteomalacia in adults (1). Meanwhile, it has been reported that vitamin D deficiency may be associated with chronic diseases such as cardiovascular diseases, diabetes, hypertension and autoimmune diseases etc. (2-10). So, treatment vitamin D deficiency and thus, maintenance of 25-hydroxyvitamin D (25OHD) level in normal range which was advised by expert committee to provide optimal tissue health is very important.

Numerous reported consensus reports on vitamin D therapy have been published by many organizations around the world (11-15). In these consensus reports, different treatment algorithms are recommended for vitamin D deficiency in children with healthy or chronic diseases (celiac disease, inflammatory bowel diseases, cystic fibrosis, etc.). For healthy children, different treatment regimens such as daily, weekly or a single dose (stoss) with cumulative vitamin D dose ranging from 84,000 to 600,000 IU are recommended (11-15). The recommended treatment duration of daily or weekly treatment regimens can range from 6 to 12 weeks. In these treatment protocols, vitamin D is still used as cholecalciferol (Vitamin D3) rather than ergocalciferol (vitamin D2), as oral low-dose long-term therapy or oral/ intramuscular high-dose injection (stoss therapy). However, both treatment protocols have their own disadvantages. Although low-dose long-term therapy varies depending on the dose, the treatment duration can be up to 3 months. This situation often causes problems in compliance with treatment. In addition, in cases of malabsorption such as celiac disease, a problem occurs in the dose adjustment required for the desired serum level due to insufficient absorption. Recently, novel treatment modalities have been developed including an oral spray, soft capsule, gels, and gums in the treatment of vitamin D deficiency (16). Most of the studies comparing different vitamin D treatment modalities were conducted with adults (1, 17, 18), and however, there are limited studies conducted in children (19,**20)**. In most of these studies, in which capsule, drop and spray forms of vitamin D were compared, it was shown that different treatment modes did not have superiority to each other (16-18), but in one, oral spray form was reported to be more effective (1). These studies are heterogeneous in terms of treatment dose and duration, population age, study design and health status, which make it difficult to draw assumptions from the results. In this study, we aimed to evaluate the efficiency of buccal spray form of vitamin D compared to single oral dose (stoss therapy) and oral drops therapy in the treatment of vitamin D deficiency.

Material and Methods

This study was conducted in children with vitamin D deficiency aged between 3-18 years old who were diagnosed as in Dr. Behçet Uz Children Hospital between January - March 2020. The exclusion criteria were as follows: hepatic or renal failure, uncontrolled hypothyroidism or hyperthyroidism, systemic inflammatory or malignant disease, vegans, confirmed diagnosis of any of the following malabsorptive conditions such as ulcerative colitis, Crohn's disease or steatorrhea etc. Consumption of medication known to influence vitamin D metabolism such as bisphosphonates, glucocorticoids and anticonvulsants; and those who had been on a sun holiday in the 30 days prior to baseline measurements and also those planning a sun holiday during the time of the study, using the medication known to affect bone metabolism. Pregnant or desired to pregnant woman during the study period was also excluded. The Local Ethics Committee approved the study (Dr. Behçet Uz Children's Hospital, Clinical Research Ethics Committee, İzmir; approval number: 2018/17-12), and written informed consent was obtained from all individuals involved.

Collected Data at Baseline

Age, sex, height and weight of all cases were evaluated. A Harpenden stadiometer with sensitivity of 0.1 cm was used for measurement of height. Body weight measurement was performed using a scale with sensitivity of 0.1 kg (SECA, Hamburg, Germany). All measurements made by the same person. The patients took off their shoes and wore light clothes before the measurement. BMI was calculated by dividing the weight in kilograms by the square of the height in meters (m^2). BMI percentiles and z scores were determined by using data of Turkish children according to their age and sex (**21, 22**). The children with a body mass index (BMI) equal or greater than the 95th percentile were considered as obese.

Baseline fasting blood samples including serum calcium (Ca), phosphorus (P), alkaline phosphatase (ALP), 25OHD, albumin, blood urea nitrogen (BUN), serum creatinine, alanine aminotransferase (ALT), aspartate aminotransferase (AST) were collected. Serum 25OHD levels and Parathyroid hormone were measured by Electrochemiluminescence (ECLIA) assay method. This assay was carried out through quantitative determinations of total 25OHD in serum samples using a standard kit available from Abbott Architect system, USA. Serum calcium, phosphorus, alkaline phosphatase, albumin, BUN, serum creatinine, ALT, AST were also measured in duplicate and assessed using an Architect C system biochemistry analyzer (Abbott, USA)

In this study, we used the following classification: Serum vitamin D level > 20 ng / mL is "sufficient", below 12 ng / mL is deficiency, and 12-20 ng / mL is "insufficiency" (11, **23**). Written and signed consent form was obtained from the parents who met the criteria and agreed to participate in the study, and from both parents and children in those older than 12 years. The 90 participants were randomly divided into 3 groups. The first group was treated with buccal spray (Wellcare vitamin D3, 1 puff, 1000 U), the second group treated with vitamin D containing drops (Devit3 oral drop, 1 drop, app.133 U) and the third group was treated with vitamin D containing ampoule (Devit3 ampoule). Each group was containing 30 patients. In group I, patients received 2000 IU/day (2 puff) for 6 weeks; while in group II patients were applied 2000 IU/day (15 drops) for 6 weeks. The patients in group III are treated with a single dose of vitamin D (300.000 IU single oral dose). Twenty-nine of them were male, and 61 of the participants were female. There were 30 participants in each group. In group I there was 11 male, 19 female, there were 9 male and 21 female in both group II and group III. All patients participating in the study kept a record of the intake time and the amount of the medication they applied. They were told that if they forgot to take the drops or spray, they would take the missing dose when they remembered. The blood samples of all groups including serum calcium, phosphorus, alkaline phosphatase, 25OHD were measured at the end of the treatment period of 6-weeks (42th day).

Statistical analysis

Statistical analyses of the data were performed using SPSS 20.0 for Windows (IBM Corp., Armonk, NY, USA). Distribution of data was evaluated using the Kolmogorov–Smirnov test. For comparison of more than two groups, one-way ANOVA or

Kruskall-Wallis test were used according to normal distribution of the data. If a significant difference was found in the comparison of more than two groups, Mann Whitney U test with Bonferroni correction or Tukey test were performed as post-hoc test to determine where the differences truly originated. In the comparison of two dependent groups (for pre- and post-treatment measurements), paired-t test or Wilcoxon test were performed according to normal distribution of the parameters. The chi-square test was used to compare the categorical variables. Spearman's rho correlation was used to identify the associations between BMI SDS, post-treatment serum 25OHD levels and the amount of increase in 25OHD levels variables. Categorical data were expressed as frequency (%), while numerical data were expressed as median (25-75th percentile) or mean ± standard deviation. A value of $p<0.05$ was considered significant.

RESULTS

The mean ages were 12.1±4.1, 10.8±3.6, 11.9±3.9 in group I, group II and group III, respectively ($p > 0.05$). The anthropometric and demographic data of participants are shown in Table 1. There was no statically significant difference for sex, age, weight, height, weight SDS, height SDS, BMI SDS between the 3 groups ($p > 0.05$).

Baseline Ca, P, ALP, PTH, 25OHD values and Ca, P, ALP, PTH, 25OHD values after the treatment were shown in Table 2. All participants were normo-calcemic therefore none of them received calcium. There was no statically significant difference between the baseline Ca, P, ALP, PTH and Ca, P, ALP and PTH levels after the treatment in all three groups. On the other hand, both the post-treatment levels of serum 25OHD and the amount of increase in serum 25OHD levels after treatment were significantly higher in group III ($p < 0.001$).

When the baseline and post-treatment values of the parameters were compared, while serum 25OHD levels were increased and PTH levels decreased in all three groups ($p<0.05$) and serum ALP levels were decreased in only group I ($p<0.05$), no statistically significant change in serum Ca and P levels were found.

At the end of the treatment, 20 (66.7%) patients in group I, 26 (86.7%) patients in group II and 27 (90%) patients in group III had normal (>20 ng / mL) serum 25OHD levels ($p=0.044$). In the remaining patients, serum 25OHD levels were in the range of 12-20 ng / mL. Six cases in group I, and 3 cases in each of group 2 and 3 were obese ($p = 0.421$). Serum 25OHD level at the end of treatment was sufficient in 4 of 6 obese patients in group I, in all obese patients in group II, and in 2 of 3 obese patients in group III ($p > 0.05$). There was no correlation between the amount of increase in 25OHD level and BMI SDS in groups I, II and III ($p > 0.05$).

Discussion

In the current study, we found that 300 000 IU oral single dose treatment was superior to 2000 IU of oral drop vitamin D3 daily for six-weeks or 2000 IU of buccal spray vitamin D3 for six weeks treatments in increasing serum 25OHD levels. Moreover, oral drop and buccal spray treatments were found to be similarly effective in raising vitamin D3. In three groups with similar baseline serum 25OHD levels, however, the percentage of normalizing serum 25OHD levels (>20 ng/mL) was lower in buccal spray treatment (66.7%) compared to oral drops (86.7%) or 300 000 IU oral single dose (90%) treatments. Malabanan et al. reported that vitamin D supplementation using 50,000 IU weekly for 8 weeks is successful in the treatment of vitamin D deficiency in older children and adolescents (**24**). In another study conducted in healthy infants and young children with hypovitaminosis D, patients were divided into 3 different groups that received 2,000 IU oral vitamin D2 daily, 50,000 IU vitamin D2 weekly or 2,000 IU vitamin D3 daily, and these three regimens were compared. All three treatment regimens were applied for 6 weeks. The regimens were shown to give equivalent results in the short-term treatment of hypovitaminosis D among healthy infants and young children (**25**). Pappa et al. found that both 2,000 IU of daily vitamin D_3 and 50,000 IU of weekly vitamin D_2 were superior to 2,000 IU of daily vitamin D_2, all taken orally for 6 weeks, in raising serum 25OHD concentration in young patients with inflammatory bowel disease and vitamin D insufficiency (**26**). When all these studies and the current study are evaluated, an inference can be made that 2,000 IU oral vitamin D3 per day, 50,000 IU oral weekly treatment for 6-8 weeks and 300 000 IU oral single dose treatment are effective in the treatment of vitamin D deficiency.

There are studies supporting that new treatment modality such as buccal spray are as effective as oral drops in this treatment (1, 16-19). Satia et al. (1) compared the absorption of vitamin D3 through the oral route by comparing buccal spray and gelatin capsule in healthy adults and patients with malabsorption disease. All participants in groups were randomized to receive either the vitamin D3 buccal spray (2 sprays each of 500 IU) or soft gelatin capsule containing vitamin D3 (1000 IU) for 30 days. After the completion of the 30-day treatment, all participants were given a 30-day washout. In the second period, crossover fashion was made and those participants who had received the buccal spray formulation (vitamin D3) in period I received the soft gelatin capsule formulation in period II and vice versa. In this study, the superiority of vitamin D3 delivery via buccal spray compared to capsules a reported positive finding the superiority of vitamin D3 delivery via buccal spray to capsules in both healthy subjects as well as in patients with intestinal malabsorption syndrome was reported. On the other hand, the trial has limitations regarding the washout duration. Todd et al. (17) compared the efficacy of vitamin D_3 liquid capsules and oral spray solution in increasing wintertime total 25OHD concentrations in a randomized, open-label, cross-over trial in healthy adults. In this randomized, open-label, cross-over trial study, 22 healthy adults received 3000 IU (75 µg) vitamin D_3 daily for 4 weeks in either capsule or oral spray form. After 10 days wash-out period, participants received opposite treatment for 4 weeks. They demonstrated that oral spray vitamin D3 is just as effective as capsule supplementation at increasing total serum 25OHD concentrations in the healthy adult population. Penagini et al. (19) demonstrated that vitamin D3 supplementation with buccal spray and oral drops is equally effective in the short-term treatment of vitamin D deficiency in a population of children with neuro disabilities. In this study 12 patients were received receive vitamin D3 buccal spray 800 IU/daily (n = 12) and 12 patients were received oral drops 750 IU/daily for 3 months during winter. Williams et al (18) conducted a randomized, placebo-controlled, three-arm parallel design study in healthy volunteers to compare the rate of change of vitamin D status in response to vitamin D3 (3000 IU/day) supplementation in capsule and sublingual spray preparations over a 6-week period. They suggested that a sublingual vitamin D spray is an effective mode of delivery for supplementation in a healthy population and the capsule and spray were equally efficacious. When all these studies considered, only Satia advocated for superiority of buccal spray of vitamin D against the other modes of delivery in increasing serum 25OHD concentrations. Recent systematic reviews demonstrated that the administration of vitamin D3 by

buccal spray is not different from other supplementation methods in increasing serum plasma 25OHD levels (16, **27**). In line, the small number of randomized controlled trials and the high degree of clinical heterogeneity of included patients did not allow for a guiding conclusion from the results (16). In the study of Unsur (20), in which evaluated infants using 400 IU / day vitamin D supplementation as oral drops or buccal spray forms at the first year of life, it was reported that the serum 25OHD levels measured at the age of one year were higher and the frequency of vitamin D deficiency was lower in infants using buccal spray than those using oral drops. In the current study, the group receiving stoss vitamin D had a significantly higher mean increase than both groups receiving buccal spray or oral drops, however there was no significant difference in terms of increase in 25OHD levels between the group receiving buccal spray and oral drops. On the other hand, when 25OHD levels of the three groups was evaluated at the end of the treatment, the frequency of the patients who reached normal 25OHD level in the patients using buccal spray was found to be lower than those using oral drops for the same dose and duration (66.7% vs 86.7%). It can be concluded that in cases with a 25OHD level of <12 ng / mL, 2000 IU / day 6-week spray therapy may be insufficient.

It is well-known that there are various factors that affect the effectiveness of vitamin D therapy other than the route of administration or dose. Dark skinned children, reduced sunlight exposure due to constant use of sunscreens or lifestyle factors, covering clothing for religious or cultural reasons, chronic illness, obesity, malabsorption syndromes (cystic fibrosis, inflammatory bowel diseases, celiac disease ect.), drugs such as anticonvulsants, systemic glucocorticoids, antiretroviral therapy, systemic antifungals can affect the success of treatment (**28**). The Institute of Medicine does not take BMI into account in recommendations for vitamin D treatment, however the Clinical Practice Guidelines by the Endocrine Society recommend obese subjects be given two to three times more vitamin D to satisfy their body's vitamin D requirement. In a study in adults, it is identified that supplementation efficiency is associated with body mass index. In participants with normal body weight higher change in serum 25OHD level was observed (**29**). Ekwaru et al (**30**). recommended 2 to 3 times higher vitamin D supplementation for obese subjects and 1.5 times higher for overweight subjects relative to normal weight subjts. Although the association between vitamin D deficiency and obesity and obesity-related diseases has been confirmed by numerous studies, the existence of a causal relationship is still unclear. In the current study, we didn't detect significant relationship between obesity and the success of vitamin D therapy.

Our study has some limitations to be acknowledged. The first limitation of our study was the small sample size. Despite the weaknesses of the present study was its small sample size, the current study was conceived as a pilot study to assess the 3 different ways of vitamin D administration, buccal spray, oral drops and oral stoss vitamin D. The second limitation of our study was that we didn't know the vitamin D binding protein (VDBP) variations of our patients. Genetic variants not only affect vitamin D metabolism, it also affects the VDBP lead to different phenotypes of the protein with different affinities to 25-OHD and 1,25-(OH)2 D3 (23). Genetic polymorphisms of DBP can also alter the protein concentration in blood (31). Furthermore, assessment of VDBP polymorphisms may be useful to adjust treatment in individuals with an insufficient response to vitamin D supplementation. Genetic factors may be taken into account in the future design of personalized supplementation. Additionally, while all patients are at the same latitude, the impact of intake of vitamin D containing foods, duration of breastfeeding, clothing, and exposure to sunlight were not considered. Finally, the patients' compliance to treatment (especially those receiving daily oral or buccal vitamin D treatment) were evaluated on the basis of their own statements. However, most of the patients we included in the study were adolescents and it is well-known that low adherence to treatment at this age is very common, which might be negatively impact on the results.

Conclusion

We conclude that a single dose 300 000IU vitamin D3 formulation was able to increase mean serum vitamin D3 concentration significantly higher compared to 2000 IU /day for six weeks either buccal spray or oral drops in both healthy children and adolescents. Vitamin D3 supplementation with buccal spray and oral drops is equally effective in terms of raising vitamin D concentration in the short-term treatment of vitamin D deficiency. However, in cases with a serul level of 25OHD < 12 ng / mL, 2000 IU / day 6-week spray therapy may be insufficient.

References

1. Satia MC, Mukim AG, Tibrewala KD, Bhavsar MS. A randomized two way cross over study for comparison of absorption of vitamin D3 buccal spray and soft gelatin capsule formulation in healthy subjects and in patients with intestinal malabsorption. Nutr J 2015;14:114.
2. Garland CF, French CB, Baggerly LL et al. Vitamin D supplement doses and serum 25-hydroxyvitamin D in the range associated with cancer prevention. Anticancer Res 2011;31:617–622.
3. Grant WB. Relation between prediagnostic serum 25-hydroxyvitamin D level and incidence of breast, colorectal, and other cancers. J Photochem Photobiol B 2010;101:130–136.
4. Anderson JL, May HT, Horne BD et al. for the Intermountain Heart Collaborative (IHC) Study Group: Relation of vitamin D deficiency to cardiovascular risk factors, disease status, and incident events in a general healthcare population. Am J Cardiol 2010;106:963–968.
5. Mitri J, Muraru MD, Pittas AG. Vitamin D and type 2 diabetes: a systematic review. Eur J Clin Nutr 2011;65:1005–1015.
6. Khan H, Kunutsor S, Franco OH et al. Vitamin D, type 2 diabetes and other metabolic outcomes: a systematic review and meta-analysis of prospective studies. Proc Nutr Soc 2012;30: 1–9.
7. Pittas AG, Nelson J, Mitri J, Hillmann W, Garganta C, Nathan DM, Hu FB, Dawson-Hughes B; Diabetes Prevention Program Research Group. Plasma 25-hydroxyvitamin D and progression to diabetes in patients at risk for diabetes: an ancillary analysis in the Diabetes Prevention Program. Diabetes Care 2012;35(3):565-73.
8. Littorin B, Blom P, Schölin A, Arnqvist HJ, Blohmé G, Bolinder J, Ekbom-Schnell A, Eriksson JW, Gudbjörnsdottir S, Nyström L, Ostman J, Sundkvist G. Lower levels of plasma 25-hydroxyvitamin D among young adults at diagnosis of autoimmune type 1 diabetes compared with control subjects: results from the nationwide Diabetes Incidence Study in Sweden (DISS). Diabetologia 2006;49(12):2847-52.
9. Raghuwanshi A, Joshi SS, Christakos S. Vitamin D and multiple sclerosis. J Cell Biochem 2008;105:338–343.

10. Pappa HM, Gordon CM, Saslowsky TM, Zholudev A, Horr B, Shih MC, Grand RJ. Vitamin D status in children and young adults with inflammatory bowel disease. Pediatrics 2006;118(5):1950-61.
11. Munns CF, Shaw N, Kiely M, Specker BL, Thacher TD, Ozono K, Michigami T, Tiosano D, Mughal MZ, Mäkitie O, Ramos-Abad L, Ward L, DiMeglio LA, Atapattu N, Cassinelli H, Braegger C, Pettifor JM, Seth A, Idris HW, Bhatia V, Fu J, Goldberg G, Sävendahl L, Khadgawat R, Pludowski P, Maddock J, Hyppönen E, Oduwole A, Frew E, Aguiar M, Tulchinsky T, Butler G, Högler W. Global Consensus Recommendations on Prevention and Management of Nutritional Rickets. J Clin Endocrinol Metab 2016;101:394-415.
12. Holick MF, Binkley NC, Bischoff-Ferrari HA, Gordon CM, Hanley DA, Heaney RP, Murad MH, Weaver CM; Endocrine Society. Evaluation, treatment, and prevention of vitamin D deficiency: an Endocrine Society clinical practice guideline. J Clin Endocrinol Metab. 2011 Jul;96(7):1911-30. doi: 10.1210/jc.2011-0385. Epub 2011 Jun 6. Erratum in: J Clin Endocrinol Metab 2011;96(12):3908.
13. Braegger C, Campoy C, Colomb V, Decsi T, Domellof M, Fewtrell M, Hojsak I, Mihatsch W, Molgaard C, Shamir R, Turck D, van Goudoever J; ESPGHAN Committee on Nutrition. Vitamin D in the healthy European paediatric population. J Pediatr Gastroenterol Nutr 2013;56(6):692-701.
14. Saggese G, Vierucci F, Prodam F, Cardinale F, Cetin I, Chiappini E, De' Angelis GL, Massari M, Miraglia Del Giudice E, Miraglia Del Giudice M, Peroni D, Terracciano L, Agostiniani R, Careddu D, Ghiglioni DG, Bona G, Di Mauro G, Corsello G. Vitamin D in pediatric age: consensus of the Italian Pediatric Society and the Italian Society of Preventive and Social Pediatrics, jointly with the Italian Federation of Pediatricians. Ital J Pediatr 2018;44(1):51.
15. Wagner CL, Greer FR; American Academy of Pediatrics Section on Breastfeeding; American Academy of Pediatrics Committee on Nutrition. Prevention of rickets and vitamin D deficiency in infants, children, and adolescents. Pediatrics 2008;122(5):1142-52. Erratum in: Pediatrics 2009;123(1):197.
16. Grammatikopoulou MG, Gkiouras K, Nigdelis MP, Bogdanos DP, Goulis DG. Efficacy of Vitamin D3 Buccal Spray Supplementation Compared to Other Delivery Methods: A Systematic Review of Superiority Randomized Controlled Trials. Nutrients. 2020;12(3):691.
17. Todd JJ, McSorley EM, Pourshahidi LK, Madigan SM, Laird E, Healy M, Magee PJ. Vitamin D3 supplementation in healthy adults: a comparison between capsule and oral spray solution as a method of delivery in a wintertime, randomised, open-label, cross-over study. Br J Nutr 2016;116(8):1402-1408.
18. Williams CE, Williams EA, Corfe BM. Rate of change of circulating 25-hydroxyvitamin D following sublingual and capsular vitamin D preparations. Eur J Clin Nutr 2019;73(12):1630-1635.
19. Penagini F, Borsani B, Maruca K, Giosia V, Bova S, Mastrangelo M, Zuccotti GV, Mora S. Short-Term Vitamin D3 Supplementation in Children with Neurodisabilities: Comparison of Two Delivery Methods. Horm Res Paediatr 2017;88(3-4):281-284.
20. Unsur EK. Vitamin D levels of the healthy infants using oral spray or drop form of vitamin D supplement in the first year of life. North Clin Istanb 2021;8(1):31-36.
21. Kondolot M, Balci E, Ozturk A, Mazicioglu MM, Hatipoglu N, Kurtoglu S, Ustunbas HB. Body mass index percentiles for Turkish children aged 0-84 months. Ann Hum Biol 2011;38(6):676-80.
22. Ozturk A, Mazicioglu MM, Hatipoglu N, Budak N, Keskin G, Yazlak Z, Balci N, Yildiz H, Yildiz K, Ustunbas HB, Kurtoglu S. Reference body mass index curves for Turkish children 6 to 18 years of age. J Pediatr Endocrinol Metab 2008;21(9):827-36.
23. IOM (Institute of Medicine) (2011) Dietary reference intakes for calcium and vitamin D. The National Academies Press, Washington
24. Malabanan A, Veronikis IE, Holick MF. Redefining vitamin D insufficiency. Lancet 1998;351(9105):805-6.
25. Gordon CM, Williams AL, Feldman HA, May J, Sinclair L, Vasquez A, Cox JE. Treatment of hypovitaminosis D in infants and toddlers. J Clin Endocrinol Metab 2008;93(7):2716-21.
26. Pappa HM, Mitchell PD, Jiang H, Kassiff S, Filip-Dhima R, DiFabio D, Quinn N, Lawton RC, Varvaris M, Van Straaten S, Gordon CM. Treatment of vitamin D insufficiency in children and adolescents with inflammatory bowel disease: a randomized clinical trial comparing three regimens. J Clin Endocrinol Metab 2012;97(6):2134-42.
27. Pritchard L, Lewis S, Hickson M. Comparative effectiveness of vitamin D supplementation via buccal spray versus oral supplements on serum 25-hydroxyvitamin D concentrations in humans: a systematic review protocol. JBI Database System Rev Implement Rep 2019;17(4):487-499.
28. Saggese G, Vierucci F, Prodam F, Cardinale F, Cetin I, Chiappini E, De' Angelis GL, Massari M, Miraglia Del Giudice E, Miraglia Del Giudice M, Peroni D, Terracciano L, Agostiniani R, Careddu D, Ghiglioni DG, Bona G, Di Mauro G, Corsello G. Vitamin D in pediatric age: consensus of the Italian Pediatric Society and the Italian Society of Preventive and Social Pediatrics, jointly with the Italian Federation of Pediatricians. Ital J Pediatr 2018;44(1):51.
29. Žmitek K, Hribar M, Hristov H, Pravst I. Efficiency of Vitamin D Supplementation in Healthy Adults is Associated with Body Mass Index and Baseline Serum 25-Hydroxyvitamin D Level. Nutrients 2020;12(5):1268.
30. Ekwaru JP, Zwicker JD, Holick MF, Giovannucci E, Veugelers PJ. The importance of body weight for the dose response relationship of oral vitamin D supplementation and serum 25-hydroxyvitamin D in healthy volunteers. PLoS One 2014;9(11):e111265.
31. Powe CE, Evans MK, Wenger J, Zonderman AB, Berg AH, Nalls M, et al. Vitamin D-binding protein and vitamin D status of black Americans and white Americans. N Engl J Med 2013;369: 1991–2000.

Table1. Anthropometric and Demographic Data of Participants				
Parameters	**Group I**	**Group II**	**Group III**	**p**
Sex (n) Male Female	30 11 (36.7%) 19 (63,3%)	30 9 (30%) 21 (70%)	30 9 (30%) 21 (70%)	0.816[a]
Age (years)	12.1±4.1	10.8±3.6	11.9±3.9	0.411[b]
Weight (kg)	45.2 (30.6-56.0)	40.6 (29.8-56.6)	47.2 (28.2-56.4)	0.823[c]
Height (cm)	153.1 (134.8-160.3)	148.6 (126.1-159.5)	149.1 (123.8-160.0)	0.873[c]
Weight SDS	-0.05±0.24	0.02±0.20	-0.13±0.17	0.868[b]
Height SDS	-0.40±0.19	-0.12±0.18	-0.31±0.18	0.561[b]
BMI SDS	0.13±0.22	0.72±0.21	-0.50±0.18	0.815[a]
Data were presented as mean±SD or median (25-75th percentiles), [a]chi-square, [b]One-way-ANOVA, [c]Kruskal Wallis				

Table2. Baseline and post-treatment laboratory characteristics of patients				
Parameters	**Baseline values**	**p**	**Post-treatment values**	**p**
25OHD (ng/mL)				
Group I (Buccal spray) n=30	8,0±0.41		22.1(17.8-28.2)[d]	
Group II (Oral drops) n=30	7.9±0.45	0.852[a]	24.4 (20.6-29.6)[d]	<0.001[b]
Group III (Oral stoss) n=30	7.6±0.47		40.3 (29.4-58.4)[d,f]	
25OHD >20 ng/mL				
Group I (Buccal spray) n=30	0 (0 %)		20 (66.7%)	0.044[c]
Group II (Oral drops) n=30	0 (0 %)	-	26 (86.7%)	
Group III (Oral stoss) n=30	0 (0 %)		27 (90%)	
Serum PTH (pg/mL)				
Group I (Buccal spray) n=30	60.1 (47.0-71.2)		51.4 (36.2-65.7)[d]	
Group II (Oral drops) n=30	60.0 (54.0-69.6)	0.273[b]	47.1 (35.1-61.7)[d]	0.585[b]
Group III (Oral stoss) n=30	53.7 (37.3-76.1)		50.4 (34,3-68.6)[d]	
Serum ALP (U/L)				
Group I (Buccal spray) n=30	176.0 (81.2-229)		140.5 (71.7-216.5)[c]	
Group II (Oral drops) n=30	192.5 (102.7-240.7)	0.590[b]	188.5 (108.2-254)	0.118[b]
Group III (Oral stoss) n=30	186.5 (71.0-240.0)		165.0 (70.0-234.5)	
Serum calcium (mg/dL)				
Group I (Buccal spray) n=30	9.9 (9.7-10)		9.8±0.05	0.928[a]
Group II (Oral drops) n=30	9.9 (9.6-10.1)	0.234[b]	9.8±0.07	
Group III (Oral stoss) n=30	10.0 (9.7-10.5)		9.8±0.06	
Serum phosphorus (mg/dL)				
Group I (Buccal spray) n=30	4.3 (4.0-4.8)		4.4 (4.0-5.0)	
Group II (Oral drops) n=30	4.5 (4.1-5.1)	0.789[b]	4.6 (4.4-5.1)	0.405[b]
Group III (Oral stoss) n=30	4.4 (3.7-4.9)		4.4 (4.0-4.8)	
Change in 25OHD (ng/mL)				
Group I (Buccal spray) n=30		-	15.6±1.3	
Group II (Oral drops) n=30	-		17.3±1.1	<0.001[a]
Group III (Oral stoss) n=30			34.3±3.2[e]	
Change in 25OHD (%)				
Group I (Buccal spray) n=30		-	214 (110-294)	
Group II (Oral drops) n=30	-		224 (137-334)	<0.001[c]
Group III (Oral stoss) n=30			445 (221-727)	

Data were presented as mean±SD or median (25-75th percentiles), [a]One-way-ANOVA, [b]Kruskal Wallis test, [c]Chi-square
[d]Wilcoxon test ($p<0.05$); comparison variables between baseline and post-treatment value
[e]Tukey test ($p<0.05$); post-hoc test for ANOVA
[f]Mann Whitney U test with Bonferroni correction ($p<0.017$); post-host test to determine the predominance for non-parametric three group comparisons