Original Article

Bone Health in Young Adults with Type 1 Diabetes Mellitus in South India

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Abstract

Introduction: Patients with type 1 diabetes mellitus (T1DM) have a higher lifetime fracture risk. The rising obesity incidence in T1DM is causing hybrid diabetes. There is scarce data of bone health and body composition in T1DM from India. This South Indian study compared bone health and body composition of patients with T1DM to healthy age- and sex-matched controls. **Methods:** Fifty-one adults with T1DM and 52 healthy controls were enrolled and underwent dual-energy X-ray absorptiometry (DXA) for bone mineral density (BMD) and body composition. Bone turnover markers—C-terminal telopeptide of type 1 collagen (CTX) and procollagen type 1 N pro-peptide (P1NP), parathormone (PTH) and 25 hydroxy-vitamin D levels were measured in patients. **Results:** The mean age of patients with T1DM was 27 years, the median duration of illness was 15 years and the median glycated haemoglobin (HbA1c) was 8%. BMD at the lumbar spine and hip were similar among patients and healthy controls. Patients with T1DM had decreased levels of CTX compared to controls (cases 0.347 (0.233–0.463) ng/mL, controls 0.440 (0.320–0.684) ng/mL, $P = 0.005$), whereas both had similar levels of P1NP. One-third of patients with T1DM were obese. Patients with T1DM with disease duration more than 10 years showed higher abdomen fat% (<10 years 20.4% [14–39.5], 10–20 years 37.2% [30.1–41.9], >20 years 41.5% [36.4–42.7] $P = 0.013$) and trunk fat% (<10 years 22.6% [14.8–37.2], 10–20 years 36.7% [30.1–40.5], >20 years 37.6% [33.55–40.5]). **Conclusion:** Young adults with T1DM have normal BMD and low bone resorption markers compared to healthy controls, whereas truncal obesity increases with a longer duration of illness. This may indicate a change in T1DM bone health and body composition characters, probably due to better glycaemic management.

Keywords: Bone health, bone turn-over markers, DXA, type 1 diabetes mellitus

Introduction

Low bone mass in type 1 diabetes mellitus (T1DM) is poorly addressed in India, with insufficient awareness and literature. Patients with T1DM have a one- to twofold greater incidence of fractures across all skeletal sites despite no consistent decreased bone mineral density (BMD).[1-3] Due to rigorous insulin treatment and an unhealthy lifestyle, young patients with T1DM are at risk of obesity, often known as hybrid diabetes or double diabetes.[4,5] This study examined bone health and body composition using dual-energy X-ray absorptiometry (DXA) and bone turnover markers (BTM) in a cohort of young patients with T1DM and compared them with normal healthy controls.

Materials and Methods

This was a cross-sectional study conducted at the Type 1 Diabetes Clinic of the Department of Endocrinology of a tertiary referral university teaching hospital in South India from January 2020 to January 2022. Based on the mean comparison of BMD in an earlier publication in patients with T1DM and healthy controls and with 95% confidence and 80% power, the minimum sample size was 50 in each group.[3] Fifty-one patients with T1DM and 52 healthy controls were included in the study. Adults more than 20 years of age who were diagnosed with T1DM before the age of 20 years as per the American Diabetes Association (ADA) criteria and who had a minimum 5-year duration of diabetes were included. Patients

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with T1DM with chronic comorbidities that could affect BMD such as chronic kidney disease (eGFR <90 mL/min/m^2), celiac disease, or those who were on medications such as antiepileptic drugs or steroids were excluded from the study. Patients were categorised according to their BMI as lean (BMI <18.5 kg/m^2), normal (BMI = 18.5–22.9 kg/m^2), overweight (BMI = 2 3.0–24.9 kg/m^2) and obese (BMI ≥25.0 kg/m^2).

Apart from diabetic history, history pertaining to bone health, including past history of fractures, calcium intake as per the International Osteoporosis Federation (IOF) calcium calculator, and adequacy of sun exposure were obtained. The 24-h recall method was utilised to assess the calcium content in a regular diet. Dietary calcium intake was considered adequate if the diet contained calcium of ≥800 mg/day. Sun exposure was considered adequate if there was at least 0.5–1 minimal erythemal dose of exposure to sunlight three times per week of arms (18% body surface area), between 10 am–3 pm.[6]

Age-, sex- and body mass index (BMI)-matched healthy volunteers who did not have any chronic comorbidities, which could affect BMD such as diabetes, chronic kidney disease (eGFR <90 mL/min/m^2), celiac disease, and history-wise not presently on any medications/supplements who consented were taken as controls.

A fasting venous blood sample was obtained from patients with T1DM for performing biochemical investigations, which included serum calcium, phosphorus, 25 hydroxy-vitamin D (here-after mentioned simply as vitamin D), parathormone (PTH) and bone turnover marker profile, which included procollagen type 1 N pro-peptide (P1NP) and C-terminal telopeptide of type 1 collagen (CTX). Serum calcium and phosphorus were analysed using the Roche Cobas c analyser using the colourimetric method. The normal range for serum total calcium was 8.6–10 mg/dL and the normal range for serum phosphorus was 2.6–4.5 mg/dL. iPTH was analysed with the Rosche Cobas e 602 immunoassay analyser using the sandwich principle (electrochemiluminescence assay [ECLIA]). The functional sensitivity of the kit was 6.0 pg/mL (0.64 pmol/L). The normal range for PTH was 15–65 pg/mL. For vitamin D analysis, samples were processed in the Rosche Cobas e 602 immunoassay analyser using the sandwich principle (ECLIA) and the functional sensitivity of the kit was 4.01 ng/mL. Ranges for vitamin D were taken from the Endocrine Society guidelines that define normal and deficiency of vitamin D levels as ≥20 ng/mL and <20 ng/mL, respectively. The total P1NP was analysed using the Roche Cobas E 100 Test, cat. no-03141071190 Elecsys analyser and the method used was ECLIA. Inter and Intra-assay CV for Total P1NP was 2.6% and 4.1%, respectively. This P1NP assay detects both trimeric and monomeric fractions present in blood and is therefore called total P1NP. CTX was analysed using the Roche Cobas E 100 Test Cat no-11972308122 Elecsys analyser and the method used was ECLIA. Inter and Intra-assay CV for CTX 3.2% and 6.5%, respectively. The functional sensitivity of the kit was 0.07 ng/mL.

BMD at femur and spine and body composition was performed using the Lunar Prodigy Advance Dual-Energy X-ray absorptiometry (DXA) System (Analysis Version 14.10) in both cases and controls. BMD is expressed in g/cm^2 and in age- and sex-matched values (Z scores). A Z score less than –2 was taken as low bone mass. Body composition parameters tested were total fat %, abdomen fat %, trunk fat % and lean mass%.

Statistical analysis

Statistical analysis was conducted using IBM SPSS 20.0 (SPSS Inc, Chicago, USA). For all continuous variables, the results are given as mean ± SD/median (Q1, Q3), and for categorical variables as frequency and percentage. The Kolmogorov–Smirnov test for normality was applied to check the distribution of variables.

To test the statistical significance of the difference in the proportion of categorical variables between the groups, a Chi-square with continuity correction test was applied. To test the statistical significance of the difference in the median of numerical variables between groups, the Mann–Whitney *U* test was applied. To test the statistical significance of the median difference of numerical variables among categorized variables, the Kruskal–Wallis test was applied and multiple comparison tests were conducted using the Bonferroni test. A *P* value < 0.05 was considered statistically significant.

Ethical aspect

The study was approved by the Institutional Ethics Committee Amrita Institute of Medical Sciences, with approval letter number IRB-AIMS-2020-228, issued on 21-07-2020. Written informed consent was taken from all subjects before recruitment in the study. All study procedures followed the guidelines laid down in the Declaration of Helsinki 1964 and as revised later.

Results

Fifty-one adults with T1DM and 52 age-, sex- and BMI-matched healthy controls were included. Baseline characteristics of patients with T1DM are shown in Table 1. The median (Q1-Q3) age among cases was 27.83 years (22.1–31.7) and among controls was 27 years (22.9–33.3). Among the cases, 27 (53%) were males and 24 (47%) were females and among the controls, 27 (52%) were males and 25 (48%) were females. The median (Q1-Q3) of BMI among cases was 23.3 kg/m^2 (19.5–25.8) and among controls was 23 kg/m^2 (19.6–25.5). The number of normal, overweight and obese individuals were 7, 18, 7 and 19 among the cases and 4, 21, 12 and 15 among the controls respectively. The PTH (cases $P = 0.653$), CTX (cases $P = 0.476$, controls $P = 0.140$), and total P1NP (cases $P = 0.140$, controls $P = 0.296$) values were found to be normally distributed.

BMD at both the lumbar spine and hip were comparable among cases and controls and so were the calcium intake and sunlight exposure as shown in Tables 2 and 3. There was no correlation between body composition and BMD.

Females with T1DM had significantly lower femoral neck BMD compared to males, 0.910 g/cm^2 (0.795–0.945) vs. 0.990 g/cm^2 (0.93–1.08) (P = 0.001) respectively, whereas in the lumbar spine, it was comparable 1.155 g/cm^2 (1.044–1.237) vs. 1.200 g/cm^2 (1.110–1.290) (P = 0.224), respectively. This difference was not there between males and females among controls (femur neck BMD in males 0.982 [0.900–1.077] g/cm^2 vs. 0.907 (0.837–1.020) g/cm^2 in females, P = 0.066). The factor significantly affecting femur neck BMD in females with T1DM was poor dietary calcium (798 mg/day vs. 850 mg/day, P = 0.021) apart from the expected factors such as height (169.5 [165.5–174] cm in males vs. 154 (152.7–159) cm in females, P = 0.00), weight (68 [59–80] kg in males vs. 51.7 [47.8–59.4] kg in females, P = 0.00) and lean body mass percentage (65.28 [63.50–71.45]) in males vs. 57.72 [54.35–62.55] in females, P = 0.00). Age at diabetes onset or duration of diabetes did not significantly correlate with BMD (lumbar spine BMD in diabetes duration <10 years 1.15 [0.99–1.33], 10–20 years 1.17 [1.00–1.25], >20 years 1.20 [1.15–1.25], P = 0.353; femur neck BMD in diabetes duration <10 years 0.95 [0.89–1.035], 10–20 years 0.92 [0.80–0.99], >20 years 1.04 [0.96–1.10], P = 0.062]. Body composition was not found to be significantly different among patients with T1DM compared to healthy controls. Bone resorption marker CTX was significantly low in patients with T1DM compared to controls, whereas the formation marker PINP was comparable in the two groups as shown in Figure 1.

Table 1: Baseline characteristics of patients with T1DM

Baseline characteristics	Cases *n* (%)=51
Duration of T1DM* (years)	
<10	7 (13.7)
10-20	33 (64.7)
>20	11 (21.5)
Median (Q1, Q3)	15 (11-20)
HbA1c† (%)	
<7	11 (21.6)
7-10	32 (62.7)
>10	8 (15.7)
Median (Q1, Q3)	8 (7.3 – 9.2)
Insulin dose (U/kg)	
<0.5	6 (11.8)
≥0.5	45 (88.2)
Median (Q1, Q3)	0.7 (0.7–0.9)
25 OH vitamin D (ng/mL)	
<20	40 (78.4)
≥20	11 (21.6)
Median (Q1, Q3)	20.70 (13.1–26.1)
Median PTH‡ levels(pg/mL) (Q1, Q3)	35.7 (22.1–54)
Median serum calcium (mg/dL) (Q1, Q3)	9.2 (9–9.5)
Median serum phosphorus (mg/dL) (Q1, Q3)	3.5 (3.2–3.7)

*T1DM=Type 1 Diabetes Mellitus, HbA1C†=Glycated Haemoglobin, PTH‡=Parathormone

Table 2: Dietary calcium, sunlight exposure in cases and controls

	Cases *n* (%)=51	Controls *n* (%)=52	*P*
Dietary calcium (mg/day)			
<800	18 (35.3)	13 (25)	0.255
≥800	33 (64.7)	39 (75)	
Median calcium intake (mg/day) (Q1, Q3)	825 (776-882)	842 (793.25-880)	
Sun exposure			
Adequate	18 (35.3)	15 (28.8)	0.483
Inadequate	33 (64.7)	37 (71.2)	

In Table 4, a comparison of the duration of diabetes with body composition parameters is shown. There was a significant effect of duration of diabetes and abdomen fat% (P = 0.013), pairwise comparison showed a significant difference between those with <10 years diabetes duration and 10–20 years duration, with P = 0.033. This difference was even more significant between those with <10 years of diabetes duration and ≥20 years duration, with P = 0.003, whereas the difference between groups with a duration of 10–20 years and >20 years was not significant as shown in Figure 2. Trunk fat% as studied across the duration of diabetes showed that the median (Q1, Q3) among those with a duration <10 years was 22.6% (14.8–37.2), in those with a duration between 10–20 years was 36.7% (30.1-40.5), and among those with duration >20 years the value was 37.6% (33.55-40.5). There was a statistically significant association between the duration of diabetes and trunk fat % (P = 0.047). On pairwise comparisons of duration of diabetes with trunk fat%, those with <10 years diabetes duration were statistically significantly different from 10–20 years duration, with P = 0.043 and ≥20 years duration (P = 0.014).

HbA1c was not significantly associated with BMD (lumbar spine [P = 0.302], femur neck [P = 0.064]) or with BTM (CTX [P = 0.205], P1NP [P = 0.712]). No association of

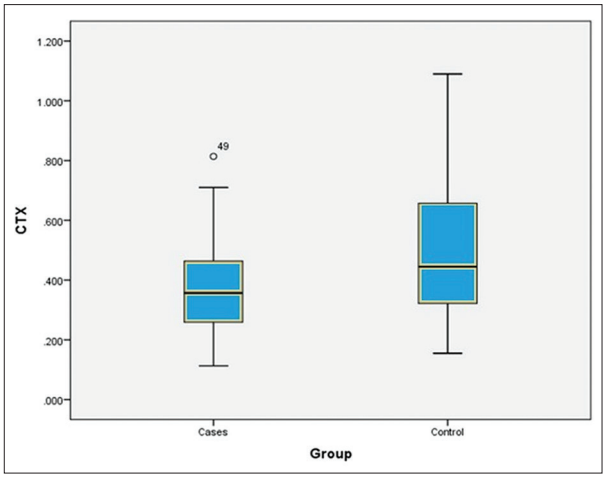

Figure 1: Comparison of CTX* (bone resorption marker) between cases and controls. Figure 1 footnotes: Abbreviation CTX* = C-terminal telopeptide of type 1 collagen. Bone resorption marker CTX was significantly low in T1DM compared to controls

Table 3: BMD, Body Composition and BTM in cases and controls

	Cases *n*=51	Controls *n*=52	*P*
Lumbar spine BMD* g/cm^2			
Median (Q1, Q3)	1.17 (1.059–1.240)	1.138 (1.046–1.183)	0.084
Femur Neck BMD* g/cm^2			
Median (Q1, Q3)	0.943 (0.831–1.044)	1.138 (0.880–1.058)	0.062
Lumbar Spine Z-score			
≥-1 (%)	42 (82.4)	41 (78.8)	
-1 to -2 (%)	9 (17.6)	11 (21.2)	
<-2 (%)	0	0	
Femur Neck Z-score			
≥-1	36 (70.6)	38 (73.1)	
-1 to -2	14 (27.5)	12 (23.1)	
<-2	1 (2)	2 (3.8)	
Body composition Median (Q1, Q3)			
Total fat %	34 (28.2–38.4)	35.9 (28.8–39.5)	0.415
Abdomen fat %	38.7 (29.4–41.9)	39.7 (33.2–47.5)	0.098
Trunk fat %	36.8 (29.2 – 39.6)	39.7 (33.2–47.5)	0.337
Lean mass%	63 (58.14 - 68.10)	60.5 (57.04–67.22)	0.248
Bone mineral content g/cm^2	3.5 (3.3–4.02)	3.5 (3.18–3.96)	0.841
Total body BMD* g/cm^2	1.133 (1.038–1.206)	1.11 (1.063–1.215)	0.856
Bone turnover markers Median (Q1,Q3)			
CTX† (ng/mL)	0.347 (0.233-0.463)	0.440 (0.320 – 0.684)	0.005
P1NP‡ (µg/L)	53.13 (38.79-72.11)	55.2 (38.79 – 72.11)	0.258

*BMD=Bone Mineral Density, CTX†=C-terminal telopeptide of type 1 collagen, P1NP‡=Procollagen type 1 N pro-peptide

Table 4: Comparison of Duration of Diabetes with Body Composition parameters

	Duration (years) median (Q1, Q3)			*P*
	<10 *n*=7	10-20 *n*=33	≥20 *n*=11	
Total fat %	25 (16.70-34.4)	35.5 (29.4-39.2)	34 (30.3-38.95)	0.152
Abdomen fat%	20.4 (14-39.5)	37.2 (30.1-41.9)	41.5 (36.4-42.7)	0.013
Trunk fat%	22.6 (14.8-37.2)	36.7 (30.1-40.5)	37.6 (33.55-40.5)	0.047
Lean Mass%	70.97 (63.2-79.1)	62.3 (57.2-66.7)	63 (57.6-66.4)	0.112
BMC* %	4.02 (3.68-4.7)	3.5 (3.3-4.1)	3.4 (3.05-3.71)	0.099

BMC*=Bone Mineral Content

abdomen fat percentage, a measure of central adiposity, with insulin dose requirements was observed (P = 0.870). Also, insulin dose did not have a significant association with lumbar spine BMD (P = 0.828) or femur neck BMD (P = 0.636). No significant association was found between vitamin D levels or PTH and BMD (lumbar spine BMD [P = 0.323] and [P = 0.392], respectively, femur neck BMD [P = 1.00 and 0.953], respectively). The association of BMI with spine BMD and femur neck BMD among cases was not found to be statistically significant with P = 0.068 and 0.113, respectively. There was no significant association of presence or absence of diabetes-related complications with lumbar spine BMD (P = 0.231) or femur neck BMD (P = 0.149).

Discussion

This cross-sectional case–control study of bone health and body composition of patients with T1DM showed that in comparison to age-, sex-, and BMI-matched healthy controls, the BMD of young adults with T1DM with a median diabetes duration of 15 years was not different but resorption bone makers were low and there was more of abdominal obesity in T1DM with increasing duration of diabetes.

Among cases, more than 80% of participants had Z score of ≥−1 at the spine and more than 70% had the same at the femur neck, and this was not statistically different from that observed in the controls. Not a single research participant had a lumbosacral spine BMD Z score lower than −2. The majority of past research on patients with T1DM discovered a somewhat reduced BMD in the hip and lumbar spine.[7] Patients with T1DM in this cohort showed lower femur neck BMD than spine BMD, a finding that did not achieve statistical significance, most likely as a result of the smaller study sample.

There was a statistically significant difference in BMD between male and female patients with T1DM at the femur neck, with the former having better BMD than the latter. However, there was no significant difference in BMD at the spine or femur

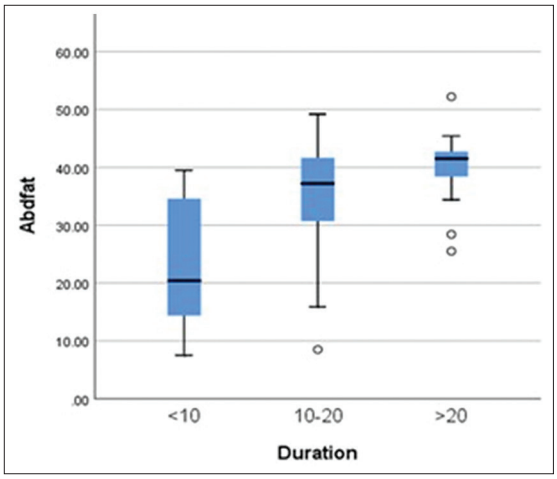

Figure 2: Comparison of abdomen fat percentage with duration of diabetes (in years) among cases. Figure 2 Footnotes: Abbreviation Abdfat = Abdomen fat percentage. The difference in abdomen fat percentage was significant between those with <10 years diabetes duration and >20 years duration, with $P = 0.003$; while the difference between groups with the duration of 10-20 years and >20 years was not significant

neck among identical sexes between cases and controls. There was no statistically significant difference in BMD between the male and female controls also. When comparing female patients with T1DM to male patients with T1DM, the lower femur neck BMD difference was found to be substantially associated with lower lean mass, lower calcium consumption, lower weight, and lower height. Mastrandrea *et al*.[8] found that T1DM females continued to have low bone mineral density (BMD) during a 2-year follow-up period. This finding was considered to be one of the likely contributing factors to the greater incidence of osteoporotic fractures among postmenopausal T1DM women.

Similar results were also found in a meta-analysis conducted by Vestergaard *et al*.[1] The result of this study must have also been influenced by a small sample size. Strotmeyer *et al*. showed that in patients with T1DM, BMD was worse with longer duration of the disease and advancing age.[9] According to the HbA1c measured during the bone health assessment, there was no significant association between BMD and glycaemic status. Vestergaard *et al*.'s[1] meta-analysis also shows a similar lack of association between HbA1C and BMD. This finding may suggest that BMD is not directly impacted by the state of metabolic control at the moment. There is evidence that suggests the health of bones is impacted by a persistent hyperglycaemic state.[10,11] This lack of an association between glycaemic status and BMD may have resulted from the fact that previous glycaemic status as indicated by older HbA1Cs was not considered in the analysis.

While P1NP did not substantially differ across the groups, the current study revealed considerably decreased bone resorption indicators among patients compared to controls. This suggests that bone turnover may be decreased in patients with T1DM, although bigger cohorts are needed to validate this. Decreased levels of osteocalcin, a measure of bone formation, and CTX, a marker of bone resorption, have been shown in a previous study to indicate decreased rates of bone remodelling in patients with T1DM.[7] Low bone turnover markers may be associated with inadequate metabolic regulation according to a study by Galluzzi *et al*.[12] Nonetheless, the current study did not yield a comparable result.

In patients with T1DM, anthropometry, BMI and body composition metrics did not correlate with BMD. Although BMI is not an exact indicator of body fat, the inclusion of BMI-matched controls may account for the negligible variation in body fat and abdomen fat percentages between case and controls. The duration of diabetes was found to significantly correlate with the trunk fat percentage and abdomen fat percentage of T1DM cases. This increment in abdomen and trunk fat was found to be very significant two decades after diabetes diagnosis and there was no significant difference within the first decade. The impact of patients' diet and lifestyle choices, the duration of intensive insulin therapy, on central obesity was not investigated. The individuals may be at risk for hybrid diabetes due to the growing body fat percentage over time. A substantial association between central adiposity and metabolic syndrome was discovered by Momesso *et al*.,[13] indicating a higher metabolic risk in this group of patients. As the duration of diabetes extended, there was a tendency for patients with T1DM BMC and lean mass to decrease; however, this difference could not achieve statistical significance.

Although 78% of patients with T1DM had 25 hydroxyvitamin D levels that were below normal, there was no discernible difference in BMD between those patients and those whose levels were normal. This could be due to normal serum calcium, phosphorus, and PTH levels as well as an appropriate calcium consumption comparable to that of the healthy controls. Joshi *et al.* showed similar results.[14]

Strengths of the study

The study's strength is that all T1DM participants had full bone health assessments, which included BMD, BTM, calcium profile, vitamin D, and PTH. The use of age-, sex-, and BMI-matched healthy controls is another strength. A thorough medical history concerning sun exposure and calcium consumption was also evaluated. Because body composition had not been taken into account in most previous investigations, its addition strengthened the research.

Limitations of the study

The small sample size is the primary limitation of the current work. Serum PTH, calcium, phosphorus, and vitamin D levels were not measured in the controls. The research was conducted using the most recent available HbA1c. Therefore, the impact of previous inadequate glycaemic management was overlooked. These may have affected the factors related to body composition and bone health. More effective methods to evaluate bone microarchitectures, such as Trabecular Bone Score (TBS) or peripheral quantitative computed tomography (pQCT) were also not performed.

Conclusion

According to our research, the young adults with T1DM had normal BMD compared to healthy age and sex-matched controls, with the exception that females had lower hip BMD compared to males. The T1DM patients had decreased bone resorption markers and near-equivalent amounts of bone formation markers as healthy controls. With longer duration of the disease, a tendency to central adiposity was demonstrated in the study. These findings may indicate a change in T1DM natural history of bone health due to better glycaemic control, probably adequate calcium intake and good sunlight exposure. More research is needed to further validate these findings. Maintaining ideal body weight, normal vitamin D levels and adequate calcium intake need to be emphasised for better bone health and body composition of T1DM patients.

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Author contribution

Akhila Bhandarkar has contributed to the concept, design, and definition of intellectual content, performed literature search, clinical studies, data acquisition, data analysis, statistical analysis, contributed to manuscript preparation, manuscript editing, and manuscript review and will be the guarantor for the manuscript. Nisha Bhavani has contributed to the concept, design, and definition of intellectual content, literature search, clinical studies, data acquisition, data analysis, and statistical analysis, contributed to manuscript preparation, manuscript editing, and manuscript review and will also be the guarantor for the manuscript. Praveen V.P. has contributed to the concept, definition of intellectual content, performed literature search, clinical studies, data acquisition, data analysis, contributed to manuscript preparation and manuscript review. Harish Kumar has contributed to concept, definition of intellectual content, performed literature search, clinical studies, data acquisition, data analysis, and contributed to manuscript preparation and manuscript review.

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Conflicts of interest

There are no conflicts of interest.

Data availability

The data that support the findings of this study are available on request from the corresponding author.

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