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The effect of anti epileptic drug therapy on serum 25-hydroxyvitamin D and parameters of calcium and bone metabolism—A longitudinal study

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ABSTRACT

Background: Chronic antiepileptic drug use is associated with bone loss. We sought to assess the longitudinal effect of antiepileptic drug on serum 25-hydroxyvitamin D [25(OH)D] levels and bone mineral metabolism markers.

Methods: Patients in the emergency services or those in neurology outpatient department with history of seizure were characterized and included in the study prospectively. Daily dietary intake of calories, calcium, phosphorus and phytates were characterized by dietary recall method. Base line bone mineral parameters – serum calcium, phosphorus, alkaline phosphatase (SAP), tartrate resistant acid phosphatase (TRACP), 25(OH)D levels, parathyroid hormone (PTH) and urinary calcium creatinine ratio (Ca.Cr), urinary calcium/kg/bodyweight (BW) and phosphate excretion index (PEI) were determined. Patients on AED therapy with normal 25(OH)D levels were followed up and were re-evaluated at the end of 6 months.

Results: The daily dietary calcium intake of the subjects was lower than the RDA (Recommended Dietary Allowance) by ICMR (Indian Council of Medical Research). The diet was high in phytates. Two-thirds of the recruited subjects were vitamin D deficient. Subjects with normal 25(OH)D levels at base line showed a significant fall of 25(OH)D levels, urinary calcium, urinary calcium/kg/BW and TRACP levels at the end of 6 months irrespective of the AED used or the plasma level of AED.

Conclusions: Hypovitaminosis D is common in our population. Subjects with normal 25(OH)D levels, irrespective of the type of antiepileptic medications even at sub-therapeutic serum levels of the drug, went into 25(OH)D deficiency and insufficiency states. Theoretically it can be worthwhile to supplement calcium and vitamin D even before initiation of antiepileptic therapy.

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1. Introduction

Epilepsy is a common neurological disorder affecting all age groups. It is one of the world's most prevalent non-communicable diseases.[1] In India, 90% of the estimated 5.5 million people with epilepsy are from rural population and three-fourths of them may be deprived of specific treatment as per the present standard. New cases are close to half a million each year and the incidence rates far exceed prevalence figures.[1] In India, treatment gap is gradually getting narrowed as community based approaches are offering effective treatment strategies.[2,3] A number of medications are currently used in the treatment of epilepsy. The older AED's are Phenobarbital (PB), Phenytoin (PHT), Carbamazepine (CBZ), Primidone (PRM), Sodium valproate (VPA) and Clonazepam (CZP). Newer AED's are Lamotrigine (LTG), Topiramate (TPM), Clobazam (CLB), Oxcarbazepine (OXC) and Levetericitam (LEV). Use of AED's has acute and chronic side effects. Chronic use of AED can affect bone health. Maintenance of bone health and bone density is a dynamic process. Reports of AED induced biochemical abnormalities suggestive of osteomalacia have been reported in the early 70s.[4–7] Recent studies have also shown that chronic use of AED has an adverse effect on bone mineral metabolism[8,9] and bone mineral density.[8,10]

There are no set guidelines whether to supplement calcium and vitamin D in a patient with epilepsy from day of initiation of AED therapy or whether a base line data of markers of bone formation, resorption and vitamin D levels are mandatory. Moreover, studies so far have been cross sectional studies or longitudinal studies with patients already on AED therapy. We therefore sought to assess the longitudinal effects of antiepileptic drug on serum 25-hydroxyvitamin D [25(OH)D] levels and parameters of bone mineral metabolism before and after AED therapy.

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2. Methods

All subjects in the emergency services or the neurology outpatient department of a tertiary referral center with history of seizure and not on treatment were included in the study prospectively. After stabilization, a comprehensive proforma which included the semiology of seizure, family history of seizures, febrile seizures and general medical history was taken. Routine biochemical investigations including hemogram, renal function tests, liver function tests and serum electrolytes was done in all subjects. Electroencephalography (EEG) was done in all subjects. Neuroimaging with either Computerized tomography scan or Magnetic resonance imaging of the brain was done in selected subjects as per the patient's semiology. The epileptic seizure and epileptic syndrome was classified according to the International League Against Epilepsy (ILAE) classification of epileptic seizures and epileptic syndromes. Subjects newly initiated on AED were included in the study. Subjects who were already on AED, on drug therapy that interfere with vitamin D metabolism, those with hepatic, renal, dermatological disorders, chronic smokers and alcoholics, pregnant women, mentally challenged and institutionalized patients were excluded from the study. This study was supported by a grant from Indian Council of Medical Research (ICMR), New Delhi.

The study center is located 13.4°N latitude. The geography of all living residence of patients were around this latitude. The average duration of sunlight is around 8–10 h per day throughout the year. Winter is short with lowest temperatures ranging from 17 to 30 °C with scanty rainfall with cloud free sunshine of 8–10 h a day. Most often, there is little seasonal variation of the peak intensity of sunlight.[11] The UV index of the region is 6–10 throughout the year.[11]

The dietary assessment of total energy, calcium, phosphorous and phytates were documented by recalling the diet consumed in the previous 5–7 days. The documentation of dietary pattern was by a single observer. The validity and repeatability of documentation was rechecked at random by one of us (authors) over the period of the study. There was no significant error in documentation of diet history. From the raw weights; the total energy, calcium, phosphorous and phytate intakes were calculated using a published food composition table, detailing the nutritive value of Indian foods.[12]

For all subjects, peripheral venous blood samples were collected in the fasting state at 8 am in the morning without applying tourniquet for the estimation of serum calcium, phosphorus, alkaline phosphatase (SAP), creatinine, albumin and serum tartrate resistant acid phosphatase (TRACP). Samples were collected on ice for 25(OH)D and N-tact parathyroid hormone (PTH). The serum was separated in a refrigerated centrifuge at 4 °C and stored at −20 °C until the analysis for the estimation of 25(OH)D and PTH.

Serum calcium, phosphorus, alkaline phosphatase, creatinine and albumin levels; urinary calcium, phosphorous and creatinine were estimated by Beckman automated analyzer (CX 9, California, USA) using commercial kits. The detailed methodology is described in our previous publications.[11]

The normal laboratory range for serum calcium is 8.5–10.5 mg/dl, serum phosphorous 2.5–4.5 mg/dl and for SAP is <95 IU/l in adults and 40–390 IU/l in children before epiphyseal closure. From the calculated values, creatinine clearance (Cr.Cl), calcium creatinine ratio (Ca.Cr) and phosphate excretion index (PEI) were derived. The urinary calcium excretion per kilogram bodyweight (U.Ca/kg/BW) was calculated. Values less than 2 mg/kg/bodyweight was defined as hypocalciuria.

The 25(OH)D concentrations were measured in duplicate by competitive radioimmunoassay after acetonitrile extraction (DiaSorin, Stillwater, MN, USA, catalogue no. 68100E). The minimal detectable limit of 25(OH)D assay is 5.0 ng/ml. N-tact PTH was measured in duplicate by immunoradiometric assay (IRMA) (DiaSorin, Stillwater, MN, USA, catalogue no. 26100). The minimal detectable limit of N-tact PTH assay is 0.7 pg/ml. Vitamin D status was classified as vitamin D – deficient, – insufficient and – sufficient or replete on the basis of 25(OH)D concentrations of <20 ng/ml [25(OH)D deficiency], 20–30 ng/ml [25(OH)D insufficiency] and >30 ng/ml [25(OH)D replete state], respectively.[13–15] All subjects recruited and those followed on AED therapy were classified based on 25(OH)D levels. Subjects with 25(OH) D levels less than 20 ng/ml were excluded from the study and were supplemented with vitamin D (as it is unethical to restrict supplementation in the background of vitamin D deficiency and AED therapy, for fear of increased risk of fractures in the event of a seizure). All other subjects on AED were included in the study and followed up. They were evaluated every month for their seizure control and any other confounding factors. At the end of 6 months, the metabolic bone disease profile and serum levels of antiepileptic drugs (PHT, CBZ, and PB) were evaluated. There was no patient who declined to participate in the study.

3. Statistical methods

A statistical analysis was performed using SPSS package (version 11.5, SPSS Inc, Chicago, IL). Descriptive results are presented as mean ± standard error of mean (SEM). Lower and upper bound confidence intervals (CI) are given in parenthesis. Paired *t*-test was conducted to study the group before and after therapy. Pearson's rank test was used for the correlation analysis when necessary. Probability values <0.05 were considered significant. One-way analysis of variance (ANOVA) was used to estimate the differences between the study groups.

4. Results

A total of 144 subjects were evaluated during the study. The mean ± SEM age of the subjects was 29.3 ± 1.2 years (CI 27–32) (range 12–65 years). The M:F ratio was 1.5:1. Twelve patients (8.3%) had status epilepticus and 54 (37.5%) had cluster attacks. As per the ILAE 38 (26.4%) had localization related symptomatic epilepsy, 20 (13.9%) had localization related cryptogenic epilepsy, 85 (59%) had generalized Idiopathic epilepsies and one was unclassified (0.7%). Computerized tomography of the brain showed the following features: Gliosis 1; calcified lesion 15; old infarct 5; Neurocysticercosis 16.

The total calories (TC) consumed daily of the whole group was 2036 ± 16 (2006–2066) kcal/day, daily dietary calcium (DC) intake was 304 ± 4 (297–311) mg/day, daily dietary phosphorous was 647 ± 6.4 (634–660) mg/day and phytate to calcium ratio 0.48 ± 0.02 (0.46–0.51). The total calories and DC consumed by males were significantly higher than that of the females 2068 ± 20 vs. 1989 ± 24 kcal/day ($P < 0.01$) and 311 ± 4.6 vs. 295 ± 5 ($P < 0.03$), respectively. There was a positive correlation between the TC and DC ($r = 0.4$; $P < 0.001$); TC and dietary phytates ($r = 0.3$; $P < 0.001$) and DC with dietary phytates ($r = 0.6$; $P < 0.001$).

The biochemical indices of bone mineral metabolism are shown in Table 1. The serum albumin was in the normal range 3.82 ± 0.03 (gm/dl). 25(OH)D deficiency (<20 ng/ml) was seen in 68% ($n = 98$), 25(OH)D insufficiency (20–30 ng/ml) in 22% ($n = 32$) and 25(OH)D replete state (greater than 30 ng/ml) in 10% ($n = 14$) of the study group. The urinary calcium/kg/bodyweight was significantly ($P < 0.05$) different among various groups of 25(OH)D deficiency (Table 1) declining with severity of 25(OH)D deficiency. Similarly changes were observed with PEI also ($P < 0.001$).

N-tact PTH correlated negatively with 25(OH)D ($r = -0.18$; $P < 0.03$) (the relationship between N-tact PTH and 25(OH)D is

Table 1
Table depicting the biochemical and hormonal indices of bone mineral metabolism of the patients at recruitment.

Parameter	Whole group ($n=144$)		25(OH)D <20 ng/ml ($n=98$)		25(OH)D 20–30 ng/ml ($n=32$)		25(OH)D >30 ng/ml ($n=14$)	
	Mean	SEM	Mean	SEM	Mean	SEM	Mean	SEM
S.Ca (mg/dl)	9.78 ± 0.04 (9.70–9.86)		9.8 ± 0.05 (9.7–9.9)		9.77 ± 0.08 (9.61–9.94)		9.76 ± 0.14 (9.45–10.1)	
S.Phos (mg/dl)	3.85 ± 0.1 (3.7–4)		3.75 ± 0.1 (3.57–3.92)		3.95 ± 0.17 (3.6–4.3)		4.3 ± 0.21* (3.84–4.75)	
SAP (IU/l)	95.24 ± 5.44 (84.5–106)		94.54 ± 6.7 (81.27–107.80)		86 ± 8.52 (68.56–103.37)		120.50 ± 23.32 (70.13–171)	
TRACP (IU/l)	8.63 ± 0.68 (7.29–9.97)		8.58 ± 0.73 (7.13–10)		7.15 ± 1.58 (3.92–10.38)		12.17 ± 3.07 (5.53–18.8)	
25(OH)D (ng/ml)	16.9 ± 0.7 (15.4–18.4)		11.9 ± 0.5** (10.9–12.8)		24.7 ± 0.5** (23.6–25.8)		35.4 ± 1** (33.3–37.6)	
N-tact PTH (pg/ml)	25.1 ± 2.2 (20.8–29.4)		26 ± 3 (20.2–31.8)		24. ± 2.7 (18.6–29.5)		21 ± 6.4 (7.1–34.8)	
S.Cr (mg/dl)	0.86 ± 0.03 (0.8–0.93)		0.87 ± 0.04 (0.8–0.95)		0.89 ± 0.06 (0.8–1.00)		0.77 ± 0.06 (0.7–0.9)	
ALB (gm/dl)	3.82 ± 0.03 (3.76–3.87)		3.81 ± 0.03 (3.74–3.87)		3.81 ± 0.06 (3.69–3.94)		3.91 ± 0.08 (3.73–4.10)	
U.CA (mg/day)	107 ± 6 (96–119)		103 ± 7 (90–117)		120 ± 12 (96–144)		107 ± 19 (65–149)	
U.PHO	381 ± 19 (344–418)		395 ± 23 (349–442)		375 ± 38 (297–453)		301 ± 51 (191–410)	
U.Ca/Wt (mg/day)	2.04 ± 0.11 (1.81–2.26)		1.90 ± 0.13* (1.64–2.17)		2.28 ± 0.25* (1.76–2.80)		2.37 ± 0.35* (1.61–3.12)	
Ca.Cr	0.12 ± 0.01 (0.11–0.14)		0.11 ± 0.01 (0.10–0.13)		0.14 ± 0.02 (0.11–0.17)		0.15 ± 0.02 (0.10–0.19)	
PEI	−0.05 ± 0.01 (−0.06 to −0.03)		−0.03 ± 0.01* (−0.05 to −0.01)		−0.06 ± 0.01* (−0.09 to −0.03)		−0.11 ± 0.02* (−0.14 to −0.08)	

Mean ± SEM all such values; values in the parenthesis – confidence intervals. 25(OH)D: 25-hydroxyvitamin D; N-tact PTH: intact PTH; S.Ca: serum calcium; S.Phos: serum phosphorus; SAP: serum alkaline phosphatase; S.Cr: serum creatinine; S.Alb: serum albumin; U.Ca: urinary calcium; U.Ca/Wt: urinary calcium/kg/bodyweight; Ca.Cr: calcium to creatinine ratio; U.Phos: urinary phosphorous; and PEI: phosphate excretion index.
The main effect of 25(OH)D on age is not significant ($F=0.057$; $P=0.94$) and on gender is not significant ($F=0.52$; $P=0.47$). The interactions of age and gender on 25(OH)D is not significant ($F=0.023$; $P=0.98$).
* $P<0.05$.
** $P<0.001$.

well modeled by a 2nd degree curve with a downward slope $R^2 = 0.9818$; $P < 0.0001$); with serum calcium ($r = -0.24$; $P < 0.01$) and PEI ($r = -0.26$; $P < 0.002$). A positive correlation was observed between serum phosphorous vs. 25(OH)D ($r = 0.21$; $P < 0.01$); and urinary calcium/kg/bodyweight ($r = 0.19$; $P < 0.03$). TRACP correlated positively with serum albumin ($r = 0.22$; $P < 0.01$) and negatively with dietary phytates ($r = -0.32$; $P < 0.001$) and phytate to calcium ratio ($r = -0.2$; $P < 0.03$).

{Two years back, at the time of recruitment of patients for the study previous classification of vitamin D status was used. Vitamin D status was classified as vitamin D – deficient, – insufficient and – sufficient or replete on the basis of 25(OH)D concentrations of <10 ng/ml [25(OH)D deficiency], 10–20 ng/ml [25(OH)D insufficiency] and >20 ng/ml [25(OH)D replete state], respectively.}[16] Using old classification 25(OH)D deficiency, 25(OH)D insufficiency and 25(OH)D replete state was seen in 25%, 44%, 31%, respectively. With the new classification[13–15] being presently followed (as defined above), the data was analyzed using the latest cut off.

Thus 46 subjects [25(OH)D replete and insufficient subjects] qualified for the longitudinal study. Only 31 subjects reported for follow-up. They were assessed at the end of 6 months for their bone mineral parameters along with serum antiepileptic drug levels. They were also analyzed based on the type and daily dose of AED used. None of them had clinical signs of drug toxicity. None of them had clinical history of fracture during 6 months follow-up period. Six months after therapy serum 25(OH)D levels declined from 29 ± 1.3 to 17.2 ± 1.6 ng/ml ($P < 0.001$) (Table 2). Ninety-four percent of them had either vitamin D deficiency or insufficiency. Of them 25(OH)D deficiency was found in 65% ($n = 20$), 25(OH)D insufficiency in 29% ($n = 9$) and normal 25(OH)D levels in 6% ($n = 2$) (Fig. 1a and b). The decline in 25(OH)D levels were observed in all subjects irrespective of the AED used (Table 3). There was a significant fall in urinary calcium ($P < 0.001$), U.Ca/kg/BW ($P < 0.001$). The TRACP levels decreased significantly ($P < 0.02$). There was no significant change in N-tact PTH, SAP and PEI (Table 2). There was no change in physical activity in these patients during the course of AED therapy.

5. Discussion

In the present study, all the patients had low dietary calcium intake compared to Recommended Daily/Dietary Allowance (RDA) as per ICMR guidelines. Previous studies from south India, especially, Andhra Pradesh have documented low dietary calcium intake in the population studied.[7,12] This is supported by studies from other parts of the country[17,18] as well on immigrant Indians in USA.[19] RDA has been revised and redefined as the Dietary Reference Intake (DRI).[20] The RDA of calcium for Indians recommended by the Indian Council of Medical Research (ICMR) is lower than the recently revised DRI.[21–23]

At recruitment, before initiation of AED therapy 90% of the subjects had varying degrees of 25(OH)D deficiencies. Only 10% had normal 25(OH)D levels. The linear decline in urinary calcium in relation to 25(OH)D levels ($P < 0.05$) document the indirect evidence of hypovitaminosis D. The normal TRACP levels suggest that bone resorption was not altered.

Subjects on AED therapy, when evaluated at the end of 6 months showed a significant fall of 25(OH)D levels, urinary calcium, urinary calcium/kg/BW and TRACP levels irrespective of the type of AED used (Table 2). Earlier studies in children[24] and adults[25] have shown decreased urinary excretion of calcium in patients on PHT, VPA, CBZ. Ninety percent of the subjects studied longitudinally developed either vitamin D deficiency or insufficiency at the end of 6 months of therapy (Fig. 1a and b, Table 3).

Table 2
Table depicting the biochemical and hormonal indices of bone mineral metabolism at recruitment and 6 months after anti epileptic therapy (AED) therapy.

Descriptive	Before TRT ($n=31$)		6 months after TRT ($n=31$)		Paired differences	
	Mean	SEM	Mean	SEM	Mean	SEM
S.Ca (mg/dl)	9.64 ± 0.14 (9.35–9.94)		9.78 ± 0.13 (9.51–10.05)		−0.14 ± 0.11 (−0.37 to 0.09)	
S.Phos (mg/dl)	4.33 ± 0.18 (3.97–4.69)		4 ± 0.18 (3.68–4.43)		0.27 ± 0.17 (−0.08 to 0.63)	
SAP (IU/ml)	94 ± 12.70 (68.10–119.97)		78.20 ± 9.50 (58.81–97.60)		15.83 ± 10.35 (−5.31 to 36.97)	
TRACP (IU/l)	13.43 ± 2.38 (7.82–16.86)		7.20 ± 0.81 (5.48–8.58)		6.23 ± 2.51* (1.05–11.40)	
25(OH)D (ng/ml)	29 ± 1.34 (26.34–31.82)		17.20 ± 1.57 (13.98–20.41)		11.88 ± 1.67** (8.47–15.30)	
N-tact PTH (pg/ml)	25.36 ± 8 (9.17–41.56)		28.54 ± 4.64 (19.06–38.03)		−3.46 ± 4.50 (−12.66 to 5.74)	
U.Ca (mg/day)	118 ± 12.42 (98.09–154.25)		67 ± 9.35 (46.40–84.02)		51 ± 11** (28.61–73.43)	
U.Phos (mg/day)	377 ± 45 (291.09–480.38)		349.06 ± 46.76 (244.20–433.70)		28.39 ± 50.33 (−75.06 to 131.83)	
U.Cal/Wt (mg/kg/BW)	2.56 ± 0.27 (1.96–3.05)		1.39 ± 0.15 (1.07–1.71)		1.17 ± 0.27** (0.61–1.73)	
Ca.Cr	0.15 ± 0.02 (0.11–0.18)		−0.01 ± 0.08 (−0.17 to 0.15)		0.16 ± 0.09 (−0.03 to 0.34)	
PEI	−0.09 ± 0.02 (−0.12 to −0.05)		−0.06 ± 0.02 (−0.10 to −0.03)		−0.02 ± 0.02 (−0.06 to 0.02)	

Mean ± SEM all such values; values in the parenthesis – confidence intervals. 25(OH)D: 25-hydroxyvitamin D; N-tact PTH: intact PTH; S.Ca: serum calcium; S.Phos: serum phosphorus; SAP: serum alkaline phosphatase; S.Cr: serum creatinine; S.Alb: serum albumin; U.Ca: urinary calcium; Ca.Cr: calcium to creatinine ratio; U.Phos: urinary phosphorous; and PEI: phosphate excretion index.
The interaction of 25(OH)D levels after AED treatment is significant ($R^2=0.787$) with (a) type of AED × gender ($F=3.268$; $P=0.05$); (b) gender × age ($F=6.953$; $P=0.022$); type of AED × age × gender ($F=9.411$; $P=0.01$).
The interactions of TRACP after AED treatment is significant ($R^2=0.884$) with (a) type of AED × gender ($F=3.987$; $P=0.04$); (b) age × gender ($F=7.773$; $P=0.02$); (c) type of AED × age × gender ($F=8.9$; $P=0.01$).
* $P<0.05$.
** $P<0.001$.

The 25(OH)D levels of the subjects on AED therapy were correlated with the serum AED levels (Table 3). Even at sub-therapeutic plasma level of the changes in calcium and bone metabolism were observed.

Two mechanisms are proposed for vitamin D inactivation by AEDs – hepatic enzyme induction and pregnane X receptor (PXR) and SXR activation.[26–28] Vitamin D (D2 and D3) are normally taken up by the liver and transformed into 25(OH)D by the mitochondrial vitamin D hydroxylase CYP27A. The AED binds to the SXR and activates it. The complex binds to RXR which then interacts with the vitamin D responsive element for the 24-OHase.[27–32] This enhances the destruction of 25(OH)D and $1,25(OH)_2D$. The accelerated clearance will promote all normal physiological adaptive mechanisms in response to progressive 25(OH)D insufficiency and consequent secondary hyperparathyroidism. Hypovitaminosis D leads to decreased calcium absorption in the intestine. Thus in the background of low dietary calcium intake, hypovitaminosis D induced by AED will have a detrimental effect on bone mineral metabolism. There are also genetic factors that may influence the susceptibility of individuals to the effect of AED on vitamin D and bone metabolism.

The other mechanisms of AED induced bone loss include direct impact of the drug on intestinal calcium transport.[33] Physiological concentrations of vitamin D_3 are necessary for maintenance of aromatase activity necessary in osteoblasts.[34–36] In the present group of subjects, enzyme inducing AED (PHT, PB, CBZ) and enzyme inhibiting AED (VPA) affected the 25(OH)D metabolism alike at sub-therapeutic doses of AED. This leads us to speculate that bone mineral metabolism is affected on those with AED at sub-therapeutic plasma concentrations of the drug level. The other mechanisms postulated include inducers of cytochrome P450 enzyme system (PB, PHT, CBZ),[4,37] reduced intestinal calcium absorption (PHT),[33] impaired response to parathyroid hormone (PB, PHT), secondary hyperparathyroidism,[38] poor vitamin K status (PHT),[39] calcitonin deficiency,[40] hepatic enzyme inhibition (VPA),[41] and by induction of the 25-hydroxyvitamin D-24-hydroxylase by the steroid hormone xenobiotic receptor.[26] Multi-drug therapy is shown to be associated with high risk of bone mineral metabolism abnormalities than monotherapy.[9,42]

Low vitamin D status in patients with epilepsy (PWE) has various management implications. Low serum 25(OH)D concentrations are associated with a higher risk for hip fracture.[43,44] PWE have an increased incidence of falls, seizure related injuries and fractures.[45] The falls may be seizure related or non seizure related injuries. Vitamin D deficiency may also lead to proximal muscle weakness and falls. Yet another study found that the risk of fracture may be more common in women on long term AED.[46] The spectrum of epilepsy is varied, with some patients having mental and motor handicap which might require institutionalization. In such cases, the physical activity and exposure to sunlight is restricted. AED associated side effects such as drowsiness; ataxia and tremor may contribute to gait disturbances with consequent increased risk of falls and fractures. All these factors add to AED induced alterations in 25(OH)D. Modification of life style, good exercise programme and nutrition supplementation can lead to healthy bones.[47] Calcium and vitamin D supplementation exerts its beneficial effects by counterbalancing the heightened vitamin D requirement induced by AEDs. Bone markers like osteocalcin and

Table 3
25(OH)D and PTH levels baseline and after 6 months of therapy on various AED with corresponding serum drug levels and the daily dose of AED used.

Drug	25(OH)D ng/ml Baseline	25(OH)D ng/ml AFT TRT*	N-tact PTH pg/ml Baseline	N-tact PTH pg/ml AFT TRT*	Drug level μg/ml	Drug dose mg/day
CBZ ($n=7$)	32.5 ± 3	18.4 ± 4.4	14.22 ± 3	23.3 ± 2.7	7.60 ± 4.4	400 ± 87
CLB ($n=3$)	25 ± 5	14.37 ± 4.5	16.6 ± 1.3	37.5 ± 3.8	–	15 ± 5
PB ($n=2$)	21 ± 1	8.9 ± 0.4	20.1 ± 7.7	33.2 ± 3.8	9.70	90 ± 30
PHT ($n=14$)	28.1 ± 2.2	17.3 ± 2.3	19.2 ± 3.6	19.5 ± 4.3	10.6 ± 7	257 ± 14
VPA ($n=3$)	31.4 ± 2.2	21.77 ± 4.5	95 ± 77	64 ± 43	–	617 ± 73
Poly therapy ($n=2$)	31 ± 2	22.3 ± 3.7	22.3 ± 4.8	29.5 ± 8.6	–	

Mean ± SEM all such values; values in the parenthesis – confidence intervals. (Normal serum range of drug levels: CBZ: 4–11 μg/ml; PHT: 10–20 μg/ml; PB: 15–40 μg/ml.).
* $P < 0.05$.

cross laps would have thrown more light but were limited by constraints of funds. Bone histomorphometry would have also helped. Because of ethical considerations it could not be done.

There are certain methodological limitations in the study. A larger number of patients in different categories of drugs used, closer follow-up (earlier than 6 months) and a control group would have added strength of this study. The high prevalence of low vitamin D at study entry may just be a mere illustration of the prevalent low 25(OH)D levels in the population. But the most significant information of the study is that those with normal 25(OH)D levels irrespective of the type of antiepileptic medications went into various of 25(OH)D deficiency and insufficiency states at sub-therapeutic levels of the drug in a period of 6 months. However, the available data leads us to speculate that the diet in person with epilepsy need to be supplemented with calcium and vitamin D at initiation of AED therapy in India where there is low dietary calcium intake and wide spread vitamin D deficiency.[48] There are yet no consensus guidelines as to whether a baseline screening for the bone mineral indices is essential. Our study shows that if the patients had not been screened for their bone health, we would have missed the majority of patients with low 25(OH)D levels and would have continued on AED which would prove detrimental. A dropout rate of 30% ($n = 15$) in our study could be due to their low socioeconomic status. This also highlights the treatment gap in epilepsy in India which could be due to economic, social and cultural reasons. Any changes in bone mineral density (BMD) measurements are observed only after 18 months of therapy. Hence biochemical and hormonal parameter evaluation is very important.

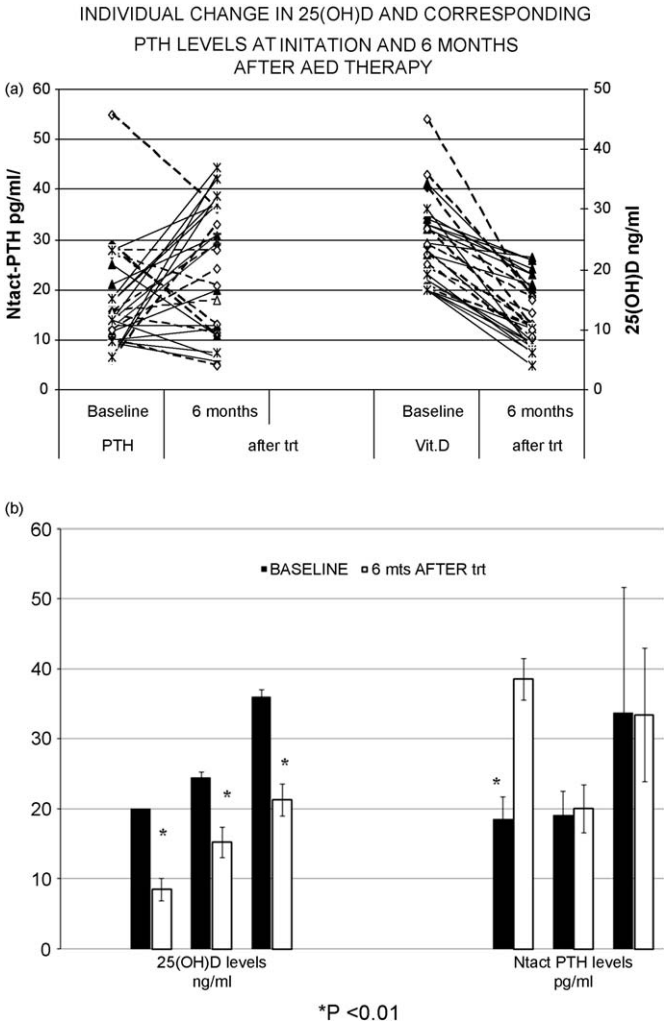

Fig. 1. (a) Individual changes in 25(OH)D and corresponding PTH levels at baseline and 6 months after AED therapy. (b) The mean and SEM of the 25(OH)D levels and their corresponding PTH levels at baseline and 6 months after AED therapy. The categorization into three groups 25(OH)D deficiency (Gr-1), sufficiency (Gr-2) and replete state (Gr-3) are based on the 25(OH)D levels at the end of AED therapy.

To the best of our knowledge, this is the first study to document the changes in bone mineral parameters at baseline and on follow-up with AED therapy along with the plasma AED levels in India. The accumulated evidence shows that there is a need for more bone health awareness and counseling of patients regarding calcium and vitamin D intake, sunlight exposure and physical activity. Good bone health practices should be a part of discussion with the patient before initiation of AED. Further multicentre prospective studies and guidelines are needed for treating bone metabolism in epilepsy.

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