

## Sunlight Exposure vs Oral Vitamin D Supplementation for Prevention of Vitamin D Deficiency in Infancy: A Randomized Controlled Trial

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**Objective:** To compare the efficacy of sunlight exposure and oral vitamin D3 supplementation to achieve vitamin D sufficiency in infants at 6 months of age.

**Design:** Open-label randomized controlled trial.

**Setting:** Public hospital in Northern India (28.7°N).

**Participant:** Breastfed infants at 6-8 weeks of age.

**Intervention:** Randomized to receive sunlight exposure (40% body surface area for a minimum of 30 minutes/week) or oral vitamin D3 supplementation (400 IU/day) till 6 months of age.

**Outcome:** Primary - proportion of infants having vitamin D sufficiency ( $>20$  ng/mL). Secondary - proportion of infants developing vitamin D deficiency ( $<12$  ng/mL) and rickets in both the groups at 6 months of age.

**Results:** Eighty (40 in each group) infants with mean (SD) age 47.8 (4.5) days were enrolled. The proportion of infants with

vitamin D sufficiency increased after intervention in the vitamin D group from 10.8% to 35.1% ( $P=0.01$ ) but remained the same in sunlight group (13.9%) and was significant on comparison between both groups ( $P=0.037$ ). The mean (SD) compliance rate was 72.9 (3.4)% and 59.7 (23.6)% in the vitamin D and sunlight group, respectively ( $P=0.01$ ). The geometric mean (95% CI) serum 25(OH) D levels in the vitamin D and sunlight group were 16.23 (13.58-19.40) and 11.89 (9.93-14.23) ng/mL, respectively; ( $P=0.02$ ), after adjusting baseline serum 25(OH)D with a geometric mean ratio of 1.36 (1.06-1.76). Two infants in sunlight group developed rickets.

**Conclusion:** Oral vitamin D3 supplementation is more efficacious than sunlight in achieving vitamin D sufficiency in breastfed infants during the first 6 months of life due to better compliance.

**Keywords:** Calcium deficiency, Compliance, Rickets, Ultraviolet rays.

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Vitamin D deficiency is a significant problem across all ages in India, especially infants due to associated maternal vitamin D deficiency, poor vitamin D content in breastmilk, and inadequate vitamin D content of traditional complementary foods in the first year of life [1,2]. Given the concern of increasing prevalence of vitamin D deficiency during infancy, Global Consensus [3], and the Indian Academy of Pediatrics (IAP)[4], recommend routine vitamin D supplementation to breastfed infants at a dose of 400 IU per day till 12 months of age. However, with an easier, natural, and inexpensive alternative for vitamin D being available through sunlight exposure, it becomes pertinent to explore the efficacy of this natural source in tropical countries like India.

The efficacy of vitamin D supplementation was reported to be higher than sunlight exposure with a mean difference ranging from 2.4-11 ng/mL in older children and

adults [5-9]. Similar comparative studies were not available in infants. An earlier study estimated that an afternoon sun exposure of 30 minutes per week for 16-18 weeks (starting from 6 weeks) over 40% of exposed body surface may be able to achieve sufficient serum 25(OH)D ( $\geq 20$  ng/mL) levels in infants at 6 months of age. The median observed sun-exposure duration was 17 minutes weekly over median 6% (range 2-40%) body surface area predominantly in summer months in northern India [10], similar to earlier conclusions on sunlight exposure in infants by Specker, et al. [11]. However, the adequacy of this duration of sunlight exposure in comparison to routine vitamin D supplementation has not been evaluated.

*Invited Commentary: Pages 835-36.*

We conducted this study to compare the efficacy of sunlight and oral vitamin D3 supplementation (400 IU/day) in achieving vitamin D sufficiency at 6 months of age. The

mean change in serum 25(OH)D levels and proportion of infants with vitamin D deficiency or rickets were also compared between both the groups.

## METHODS

This was an open-labeled randomized controlled trial conducted between November, 2018 and April, 2020, in a public hospital in Northern India (28.7°N). An approval from the ethics committee of the institute was obtained. Written informed consent was obtained from the parents for participation. The trial was registered with the clinical trial registry of India.

We enrolled infants between 6-8 weeks of age during the first immunization visit from the outpatient clinic including those residing within a 5 km radius of the hospital to ensure uniform latitude, season, and geographical location. Infants who were born at term gestation (37-41 weeks), appropriate for gestational age, and exclusively breastfed were included. Any child with chronic systemic illness, congenital malformations, genetic syndrome, history of hospitalization in neonatal care unit for more than 48 hours, history of intake of calcium or vitamin D preparations, skin disorders, active rickets, mothers on vitamin D supplements, or history of intake of mega dose of vitamin D (6 lakh IU or >1000 IU/day) in preceding 12 weeks, and growth faltering as per WHO growth charts [12], were excluded. No changes in inclusion criteria were made after the commencement of the trial.

Participants were enrolled between March and July (during summer months) for a six month intervention period till September-December, keeping in mind the Indian equinox. The relevant clinical information on antenatal complications, maternal intake of calcium, or vitamin D supplements, and birth details of the baby were recorded. The socio-demographic characteristics of the household including the socioeconomic status and type of house were recorded. The infant's weight, length, and head circumference were recorded by standard techniques using calibrated equipment in a pre-designed proforma. WHO reference [12], was used for determining weight-for-age *z*-score (WAZ), length-for-age *z*-score (LAZ), weight-for-length *z*-score (WLZ) using AnthroCal software [13]. The feeding history of the infant with a history of intake of vitamin D supplements was recorded on the proforma. The infant was clinically examined for rickets by a pediatrician. They were clinically evaluated for skin color according to Fitzpatrick skin color scale into six types, where type I was the fairest and type VI was the darkest skin [14].

Infants were randomized using block randomization of varying block sizes ([www.randomization.com](http://www.randomization.com)) to receive

either of the following interventions till 6 months of age: *i*) Sunlight group: Sunlight exposure over 40% body surface area (BSA) for a minimum of 30 minutes/week, and *ii*) Vitamin D group: Oral vitamin D3 supplementation (400 IU/day). The random number sequence was generated by a third person not involved in the study. Allocation concealment was done by the sealed envelope technique which was prepared by a third person different from the person who generated the randomization sequence. This was an open-label study.

In the sunlight group, mothers were counseled to expose infants to direct (unfiltered) sunlight over 40% body surface area (BSA) for 30 minutes per week. The mothers were advised to lay infants in direct sunlight in a prone position wearing only diapers for 5 min/day preferably between 1000-1100 hours or 1430-1500 hours for 6 days every week excluding Sundays. Exposure on cloudy days was not advised; it was compensated by adding the duration of the missed day/s on the next day within seven days. Compliance was checked by a daily record of BSA exposure with mention of exact duration (in minutes) and time of the day (as per clock time) of sun exposure in a modified Lund and Browder chart [15] that was collected once a month. In the sunlight group, percent compliance was calculated by dividing the total duration (in minutes) for which the baby was exposed to sunlight by the total duration (in minutes) for which sunlight was prescribed.

In the vitamin D group, mothers were counseled to administer 400 IU/day of oral vitamin D3 as 0.5 mL/day (1 mL=800 IU in 30 mL pack, Arbivit-3 forte drops, Raptakos Brett & Co Ltd) using a standardized dropper supplied within the bottle. The drug was obtained from the manufacturers directly and distributed by the study investigators. The mothers were asked to report the missed doses by marking on calendar dates in the format provided to them and return with used supplement bottles during their monthly visits for measuring any leftover medicine. The percent compliance was calculated by dividing the number of days oral vitamin D3 was given in recommended dosage to the baby by the total number of days prescribed. They were not counseled for or against sunlight exposure but were asked to record the same.

Compliance in both the groups was ensured telephonically every week and was defined as low at <50%, moderate at 50-80%, and high at ≥80%. Infants were physically followed up at 10 and 14 weeks at consecutive immunization visits and then additionally at 6 months ( $\pm 2$  weeks) of age. Anthropometry of infants was performed at each visit. Infants were also examined for clinical signs of vitamin D deficiency/ rickets (anterior fontanelle measures, rachitic rosary, frontal bossing, and wrist widening).

Infants developing clinical rickets during follow-up were treated as per standard guidelines [4]. Mothers were asked to report adverse events, if any, at each visit.

Venous blood (2 mL) from mother and infant was drawn at baseline for estimation of serum 25(OH)D levels; and from the infant at the end of 6 months (3 mL) for estimation of serum 25(OH)D, serum calcium, serum phosphorus, and serum alkaline phosphatase (ALP). Serum calcium, phosphorus, and ALP levels were measured at baseline within 24 hours of collection. Serum was stored at -20 degrees Celsius for estimation of baseline serum 25(OH)D levels and were measured with serum 25(OH)D levels collected at six months. Additionally, in the vitamin D group, a spot urinary sample was taken for determining urinary calcium creatinine ratio (U-CaCr) to detect hypercalciuria. Serum calcium, phosphorus, and ALP were estimated by UniCelDxC 600 automatic analyzer (Beckman Coulter India, Pvt. Ltd.) Total serum 25(OH)D was estimated by the Beckmann Coulter radio-immunoassay method. Urinary calcium creatinine ratio (U-CaCr) was measured in a spot freshly void urine sample by UniCelDxC 600 automatic analyzer (Beckman Coulter India Pvt. Ltd.) using the Colorimetric method. A U-CaCr cut-off of >0.6 mg/mg was taken as abnormal. Vitamin D status was defined as sufficient if >20 ng/mL, insufficient 12-20 ng/mL, and deficient <12 ng/mL [3,4].

The outcomes were measured at 6 months  $\pm$  2 weeks of age. The primary outcome was proportion of infants having serum 25(OH)D levels >20 ng/mL. The secondary outcomes were serum 25(OH)D levels, the proportion of infants developing clinical rickets, and vitamin D deficiency (<12 ng/mL).

In a previous study [16], the observed proportion of infants being administered 400 IU of oral vitamin D3 daily (from birth) to achieve serum 25(OH)D level  $\geq$ 20 ng/mL at 6 months of age was 97.5%. Considering less compliance in our population, we expected 85% to achieve serum 25(OH)D level >20 ng/mL at 6 months. With 15% absolute difference with sunlight group on either side of oral vitamin D group, type-I error 5% and 80% power, we needed 36 participants per group (non-inferiority). Assuming 10% attrition, we planned to enrol 40 infants per group.

**Statistical analysis:** Data were analyzed by using SPSS software (version 11). Descriptive data were presented as mean or median (IQR). Baseline continuous characteristics were compared between the groups using unpaired t-test and categorical with Chi-square/Fisher exact test. Normality of serum 25(OH)D data was tested using the Shapiro Wilk test and found right-skewed distribution. The natural log transformation was applied to make distribution normal and

results are reported in geometric mean with a 95% confidence interval. Analysis of covariance was applied to compare serum 25(OH)D at 6 months between the groups taking baseline serum 25(OH)D as a covariate and also with compliance. The interaction between the groups and covariates was tested and subgroup analysis was performed in case of a significant interaction. McNemar test was used to compare the proportions within the group before and after the intervention and Chi-square/Fisher exact test was performed to compare two groups. Intention to treat analysis was used for comparison between intervention and control groups. The missing values were substituted by the baseline value of serum 25(OH)D for final analysis. Pearson correlation was used to check the correlation between two continuous variables. *P*-value <0.05 was considered significant.

## RESULTS

Participant enrolment and follow-up are depicted in **Fig. 1**. A total of 80 (49 boys) infants were enrolled [mean (SD) age 47.8 (4.5) days; birth weight 2805.8 (313.6) g]. Infants

**Table I Baseline Parameters in Sunlight and Vitamin D Groups**

<i>Parameters</i>	<i>Sunlight (n=40)</i>	<i>Vitamin D (n=40)</i>
Age at enrolment (d)	47.9 (4.7)	47.7 (4.3)
Birth weight (g)	2790.3 (314.6)	2821.3 (315.8)
Gestation (wk)	37.8 (1.1)	37.9 (1.0)
<i>Anthropometry</i>		
Weight at enrolment (g)	4375.5 (421.0)	4415.8 (383.8)
Length at enrolment (cm)	55.8 (1.6)	56.0 (1.6)
Weight for age z-score	-0.96 (0.69)	-0.82 (0.63)
Length for age z-score	-0.28 (0.80)	-0.04 (0.82)
Weight for length z-score	-1.03 (0.76)	-1.04 (0.65)
<i>Skin Fitzpatrick score<sup>a</sup></i>		
Type III	6 (15)	9 (22.5)
Type IV	26 (65)	25 (62.5)
Type V	8 (20)	6 (15)
<i>Socioeconomic status<sup>a</sup></i>		
Upper middle	2 (5)	2 (5)
Lower middle	8 (20)	6 (15)
Upper lower	20 (50)	27 (67.5)
Lower	10 (25)	5 (12.5)
Maternal serum 25(OH) D (ng/mL) <sup>b</sup>	9.76 (8.81-10.80)	9.31 (8.27-10.47)
Infant serum 25(OH) D (ng/mL) <sup>b</sup>	10.93 (9.24-12.93)	10.87 (9.44-12.52)
<i>Month of enrolment<sup>a</sup></i>		
March to May	35 (87.5)	37 (92.5)
June-July	5 (12.5)	3 (7.5)

*Data expressed as mean (SD), <sup>a</sup>no.(%) or <sup>b</sup>Geometric mean (95% CI). 25(OH)D-25 hydroxyvitamin D. All *P*>0.05.*

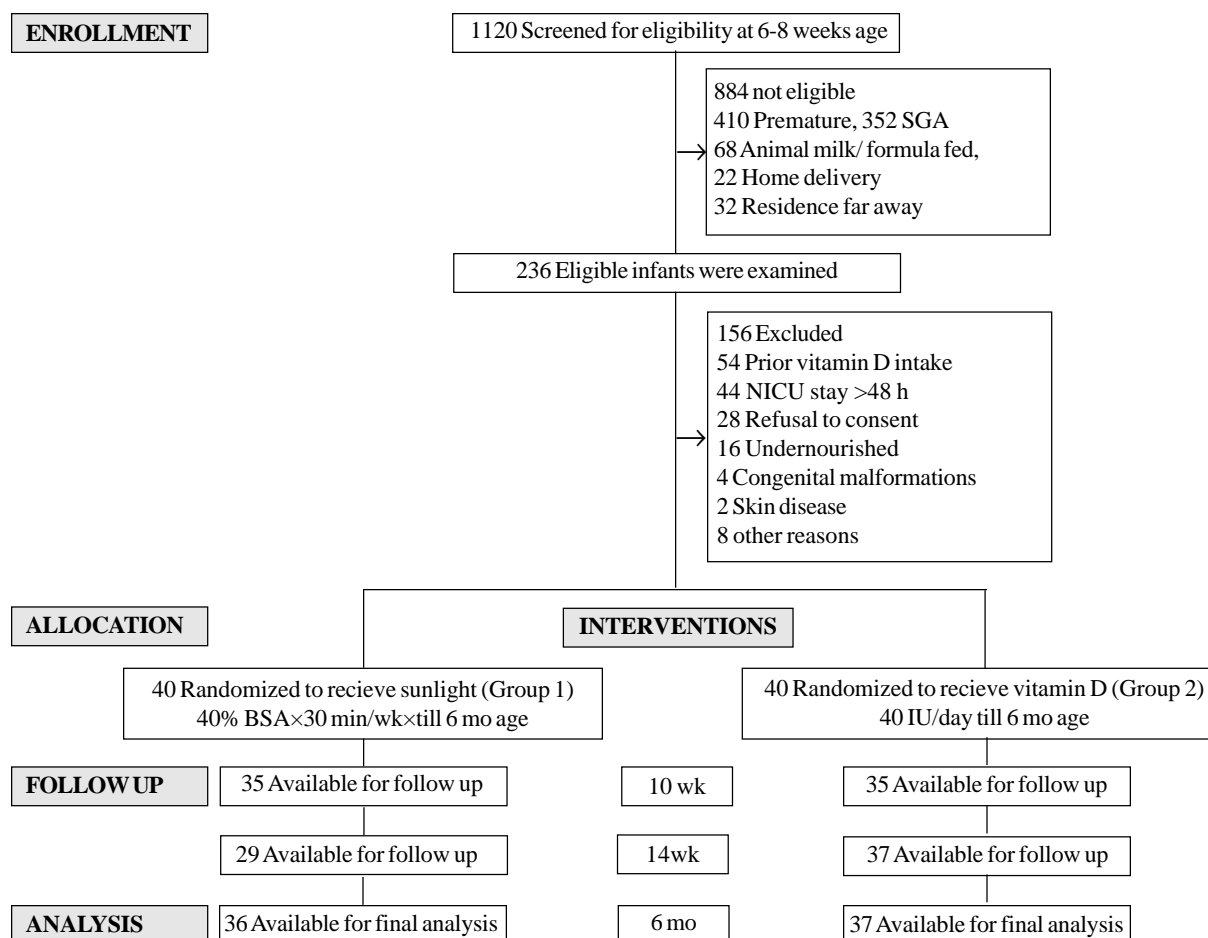


Fig. 1 Study flow chart

belonged to Fitzpatrick skin type III 15 (18.8%), IV 51 (63.8%), or V 14 (17.5%).

Vitamin D deficiency, insufficiency and sufficiency at baseline were seen in 57 (71.2%), 22 (27.5%), and 1 (1.3%) mothers, and in 49 (61.2%), 21 (26.3%), and 10 (12.5%) infants, respectively. The baseline characteristics of infants in the two groups are shown in **Table I**. All infants were exclusively breastfed till six months of age and did

not receive calcium or vitamin D supplements from any other source.

The mean (SD) compliance was 72.9 (3.4)% in the vitamin D group and 59.7 (23.6)% in the sunlight group ( $P=0.01$ ) and 17 (45.9%) and 6 (16.7%) infants had  $\geq 80\%$  compliance in both groups, respectively. The mean (SD) WAZ score at follow-up in two groups was comparable [-0.19 (0.56) and -0.24 (0.66), respectively;  $P=0.726$ ]

Table II Vitamin D Status Before and After Intervention in Sunlight and Vitamin D Group

Vitamin D status	Group	Before intervention	After intervention	P value
Vitamin D sufficient <sup>a</sup>	Sunlight (n= 36)	5 (13.9)	5 (13.9)	1.0
	Vitamin D (n= 37)	4 (10.8)	13 (35.1)	0.01
Vitamin D deficient <sup>b</sup>	Sunlight (n= 36)	22 (61.1)	17 (47.2)	0.45
	Vitamin D (n=37)	22 (59.5)	13 (35.1)	0.04

Values in no. (%). Vitamin D sufficient as serum 25 (OH) D levels  $>20$  ng/mL. Vitamin D deficient as serum 25 (OH) D levels  $<12$  ng/mL.

<sup>a</sup>P value between two groups before intervention  $P=0.74$  after intervention  $P=0.037$ ; <sup>b</sup>P value between two groups before intervention  $P=0.88$ , after intervention  $P=0.294$ .

Vitamin D status of participants at baseline and after the intervention is compared within group and between groups in **Table II**. In the sunlight group, five infants with serum 25(OH)D levels >20 ng/mL had >50% compliance. In the vitamin D group, 11 out of 13 (84.6%) infants with serum 25(OH)D levels >20 ng/mL had ≥80% compliance. Values of four and three participants were missing in the sunlight and vitamin D group, respectively, that were substituted for baseline values for statistical analysis. On analyzing the primary outcome by replacing the missing values, there was no change in the results.

We did not observe any significant correlation between the log-transformed maternal serum 25(OH)D values and serum 25(OH)D values of the infant, both before ( $r=-0.128$ ;  $P=0.258$ ) and after intervention ( $r=-0.089$ ;  $P=0.456$ ). There was no statistical significant association found for change in vitamin D and both serum calcium and phosphorus in both groups at six months (data not shown).

The mean (SD) serum calcium in the sunlight and vitamin D group were 8.86 (1.03) and 9.08 (0.93) mg/dL, respectively ( $P=0.335$ ); serum phosphorus 5.24 (0.87) and 5.57 (0.75) mg/dL, respectively ( $P=0.09$ ); and serum ALP 226.6 (75.6) and 245.3 (48.91) IU/L, respectively ( $P=0.158$ ).

The geometric mean serum 25(OH)D level at 6 months of age was higher ng/mL in the vitamin D group than sunlight group after adjusting baseline serum 25(OH)D level as a covariate with a geometric mean ratio of 1.36 (95% CI: 1.06-1.76) (**Table III**). Analysis of covariance (ANCOVA) found a significant interaction between the group and compliance percentage ( $P=0.045$ ) revealing the dependence of vitamin D values at 6 months on the compliance percentage (**Web Fig. 1**). A post-hoc analysis was performed to assess the effect of vitamin D by dividing the study subjects into ≥80% (high) and <80% (low and moderate) compliance. The ANCOVA models were performed taking serum 25(OH)D levels at 6 months as dependent and group as the fixed factor with baseline serum 25(OH)D levels as the covariate (**Table III**).

Two infants in the sunlight group developed rickets (diagnosed clinically and confirmed with biochemical and

radiological tests) while none developed rickets in the vitamin D group. Four infants (baseline mean (SD) serum 25(OH)D 13.12 (3.6)ng/mL) in the vitamin D group had U-CaCr >0.6 mg/mg with mean (SD) serum calcium 9.25 (0.54) mg/dL and serum 25(OH)D levels 20.97 (10.0) ng/mL, without any clinical or biochemical evidence of vitamin D toxicity.

## DISCUSSION

The present study showed a higher proportion of infants with vitamin D sufficiency after daily vitamin D3 supplementation for six months than daily sunlight exposure. Vitamin D sufficiency in both groups was significantly related to compliance which was better in the vitamin D group than the sunlight group.

Studies have shown that serum vitamin D level increases after both, sunlight and supplementation. A systematic review reported a significant increase in serum 25(OH)D levels after exposure to artificial UV light which was dependent on UV-B dose and baseline serum 25(OH)D [17]. A recent study showed that infants with <30 min/day sunlight exposure were five times likely to develop vitamin D insufficiency than those with >30 min/day [18].

However, vitamin D3 supplementation results in a more predictable increase in serum 25(OH)D levels than sunlight as seen in adults [19-21]. A systematic review of seven studies in adults reported a higher mean difference of 8.56 nmol/L (95% CI 4.15-12.97) with vitamin D supplementation (400-5000 IU/day) than sunlight (duration 8-48 weeks). This difference remained statistically significant irrespective of the duration of therapy and was higher with the use of natural sunlight [9]. In another RCT from China, adolescents with low baseline serum 25(OH)D (15.5-16.5 nmol/L), were randomized to receive oral 800 IU/day vitamin D3 or more than 30 min/day of outdoor exposure between 9 am and 3 pm and compared with a control group. A significantly higher reduction was observed in the proportion of subjects with vitamin D deficiency in the first two groups [8]. The present study also reported greater efficacy of vitamin D3 supplementation than sunlight exposure to achieve a higher proportion of vitamin D sufficiency in infants.

**Table III Serum 25(OH)D Levels Six Months After Intervention Among Children in the Two Groups**

	Sunlight group		Vitamin D group		OR (95% CI)	P value
	n	25 (OH) D levels	n	25 (OH) D levels <sup>a</sup>		
All children <sup>a</sup>	36	11.89 (9.93-14.24)	37	16.23 (13.58-19.40)	1.36 (1.06-1.76)	0.02
≥80% compliance	6	15.61 (11.36-21.44)	17	24.28 (20.11-29.31)	1.57 (1.08-2.27)	0.03
<80% compliance	30	11.25 (9.30-13.56)	20	11.54 (9.18-14.53)	1.03 (0.77-1.37)	0.86

Vitamin D levels as geometric mean (95% CI) in ng/mL. <sup>a</sup>Interaction between group and baseline serum 25(OH)D was not significant ( $P>0.05$ ).

Harsher summer in Northern India probably limits time spent outdoors, while increasing air pollution with poor UV index limits sunlight penetration during winters even when sun exposure is preferable [22]. A cross-sectional survey from Ethiopia reported poor knowledge, attitude, and practices towards sunlight exposure of infants in mothers [23]. Most families being nuclear in structure can offer limited time for supervised sun exposure for their infants. These variances are unlikely to be seen with vitamin D3 supplementation suggesting it to be a more reliable and effective strategy. Also, rickets developed in infants in the sunlight group unlike in the vitamin D group reiterating the role of vitamin D supplementation in the prevention of rickets as reported in an earlier study [24]. However, vitamin D supplementation carries its concerns of added costs, adherence, and risk of side-effects/toxicity unlike sunlight exposure which is a natural and effective endogenous source of vitamin D. There may occur inter-personal variations in adherence to universal vitamin D supplementation practices [25]. Moreover, at present the Government agencies do not endorse any routine infant supplementation.

The lack of direct observation of interventions, lack of objective measurement of sunlight with the solar meter or quantification of standard erythemal dose, and collection of information on sunlight exposure on questionnaire alone may be perceived as the limitations in the present study. Repeat urine samples were not collected to confirm elevated calcium excretion as part of this study. However, this study explores the role of sunlight in Indian infants where sunlight is replete and may be utilized for optimizing serum 25(OH)D levels after accounting for environmental and personal host factors.

We conclude that oral vitamin D3 supplementation of 400 IU/day was more efficacious than daily sunlight exposure to achieve vitamin D sufficiency in infants; the effect was mainly modified through better compliance to oral vitamin D3 supplementation than sunlight. Daily vitamin D supplementation has added costs with the need for administration, while sunlight exposure is an unpredictable source of vitamin D. Further epidemiological studies are needed to evaluate the practical implementation and cost-effectiveness of these public health strategies.

*Ethics approval:* Ethics Committee of University College of Medical Sciences; No. HR/2018/36/105, dated Oct 15, 2018.

*Note:* Additional material related to this study is available with the online version at [www.indianpediatrics.net](http://www.indianpediatrics.net)

*Contributions:* PG, DS: Study conception, data analysis, manuscript review and final editing; AG, AD: Data collection and analysis, manuscript drafting and writing; RKM: Data analysis, manuscript writing and review; PD: Study conception,

manuscript drafting; SVM: Laboratory analysis, manuscript review and editing; All authors have read and approve the final contents of the manuscript.

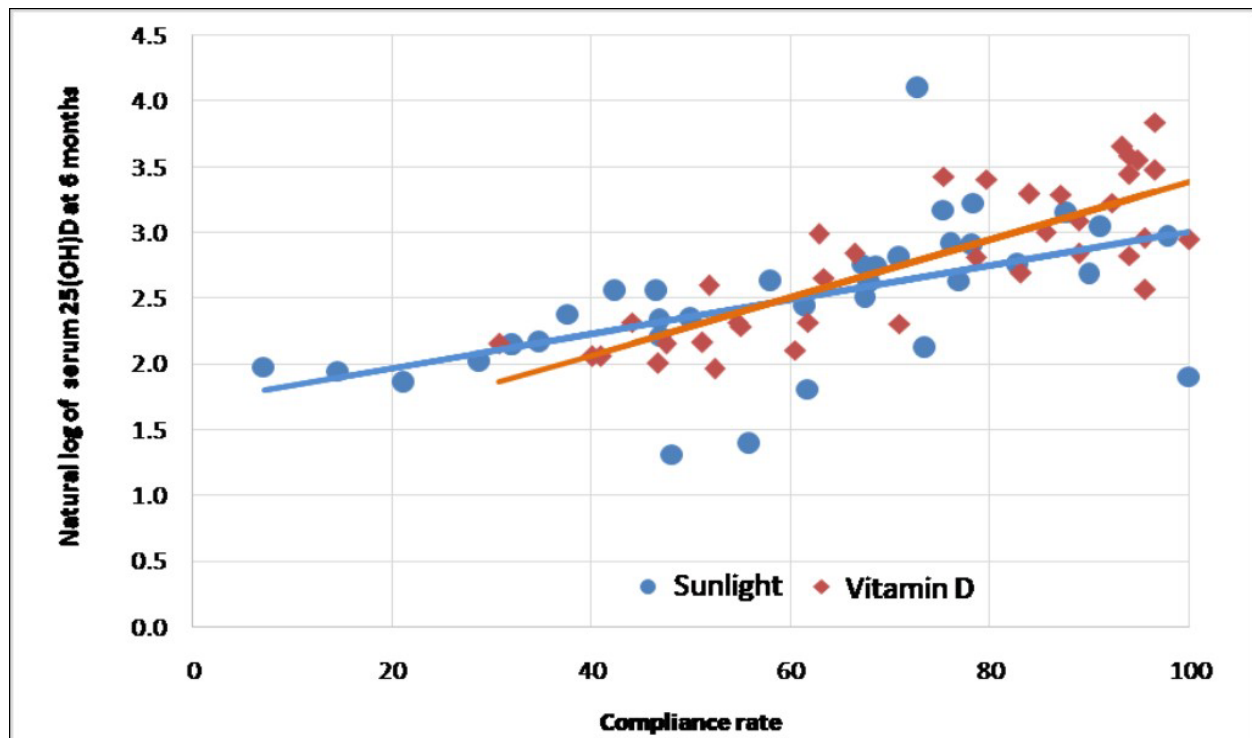
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*Note:* Additional material related to this study is available with the online version at [www.indianpediatrics.net](http://www.indianpediatrics.net)

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Web Fig. 1 Interaction between compliance rate and the natural log of infant's serum 25(OH)D at 6 months.