

Impact of vitamin D fortified food on quality of life and emotional difficulties among adolescents – A randomized controlled trial

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ABSTRACT

Background: The action of vitamin D on health related quality of life (HRQoL) and emotional difficulties of individuals has been extensively studied in several epidemiological studies. However, studies focusing on the effect of vitamin D fortification on these aspects among adolescents are limited. **Aim:** This study aims to explore the influence of vitamin D fortification on the vitamin D status and the HRQoL and emotional difficulties among adolescents. **Methods:** A total of 71 healthy adolescents consented to be a part of the randomized controlled trial conducted in two schools in Chennai, India. They were allocated and blinded into two groups: experimental (one vitamin D fortified laddu (a homemade eatable with natural supplements) daily, containing 1000 IU vitamin D₃) or the control group (one unfortified laddu daily) for 12 weeks following random assignment. The vitamin D status, HRQoL and emotional difficulty parameters of anxiety and stress of the participants were evaluated both at baseline and at 12 weeks. Data were analyzed using SPSS version 12. **Results:** Vitamin D status of the adolescents significantly improved post supplementation ($Z=-6.28$; $P=0.00$). However, despite of significant improvement, no significant changes were observed in the other parameters. **Conclusion:** These findings demonstrate that although vitamin D fortification helps in improving the vitamin D status of adolescents, it does not particularly benefit their HRQoL and emotional difficulties of anxiety and stress of adolescents.

Key words: Vitamin D fortified laddu, Vitamin D status, Quality of life, Emotional difficulties, adolescents

Adolescence is marked by a period of transition in the emotional, mental and behavioural spheres^[1-4] thus, making it essential to study the health related quality of life (HRQoL) and emotional difficulties experienced in this phase^[2,3]. The prevalence of behavioural and emotional problems among adolescents ranges from 16.5% to 40.8% in different parts of the world and in India it is in the range of 13.7% to 50%^[1].

Vitamin D has been widely identified as a neurosteroid with multiple actions in the brain during the past 20 years^[5]. It has long been recognized that out of the many areas in the body, the brain can also produce 1,25 dihydroxy vitamin D, the active form of vitamin D^[6]. Sunlight is considered to be the most abundant source of vitamin D as it is naturally synthesized in the skin on exposure to ultraviolet B rays^[7]. Surprisingly although India is located between 8.4° and 37.6° north latitude and majority of the occupants reside in regions with good and sufficient sunshine throughout the year, vitamin D deficiency is widespread among 70-100% Indians^[8-12]. In north India, vitamin D deficiency was reported among 93.7% children and adolescents^[13]. Thirty four percent of children in South Chennai were also shown to have hypovitaminosis^[14].

Scientists have suggested vitamin D supplementation to have biological plausibility in promoting increased well-being and

physical activity of individuals^[15]. Epidemiological evidences indicate its role in emotional difficulties such as depression, anxiety and stress and HRQoL among individuals^[16-18]. The link between vitamin D deficiency and headache^[19], muscular pain, psychotic features among mentally ill^[17], reduced physical activity^[20], lower back pain^[21], increased emotional and peer relationship problems among adolescents have been explored in several studies^[22]. Vitamin D supplementation has resulted in lower rates of psychosis and depression among adolescents^[17,23]. However, conflicting results have been observed in this area. In a study conducted among children in North India, vitamin D did not seem to have any link with neurodevelopment when compared with Vitamin B₁₂^[24]. Trials focusing the action of vitamin D supplementation on HRQoL and emotional difficulties of adolescents is lacking in India. To bridge this gap, the present study was designed to evaluate whether vitamin D fortified food will help in improving the HRQoL and emotional difficulties among adolescents.

MATERIAL AND METHODS

A randomized controlled trial was employed to study the effect of the vitamin D fortified food ('laddu' (a homemade

sweet loaded with natural supplements) a cereal legume based preparation) on the HRQoL and emotional difficulties of adolescents. The study was carried out as per the guidelines stated by the Indian Council Medical Research and was approved by the Institutional Ethics Committee (IEC/14/DEC/113/26). The study has been registered under the Clinical Trial Registry of India (CTRI/2017/04/008408).

Participants were healthy adolescents recruited from two schools in Chennai. Inclusion criteria were both boys and girls of 11-16 years with low serum 25OHD levels and complete willingness to participate in the intervention. Individuals were excluded if they reported any ailments like epilepsy, migraine, intestinal malformation; hypercalcemia or were taking D₃ supplements (current or recent within one year). Informed consent and assent forms were distributed to the parents and their wards respectively and only those whose signed forms were obtained, were included for the baseline screening.

The adolescents were randomly divided to the experimental or control group to receive either vitamin D fortified snack or the unfortified snack respectively. These were cereal pulse based ladduus prepared with *Eleusine coracana L* (finger millet), *Pennisetum typhoideum* (pearl millet) grains and *Vigna mungo* (black gram dhal) mixed together in the ratio of 34%:17%:8% respectively along with ghee and sugar. Vitamin D premix containing cold water soluble form of vitamin D₃ was procured from Hexagon Nutrition Pvt Ltd and was used for fortification. Each fortified ladduu had 1000IU of vitamin D₃. The non-fortified ladduus had the same ingredients as that of the fortified ladduu but was without vitamin D₃.

The vitamin D₃ fortified and the unfortified ladduus were prepared by the investigator twice in a week under hygienic conditions. The ladduus were wrapped in aluminium foils and stored in HDPE container. The participants in control and experimental group were given one non-fortified and one fortified ladduu each day respectively for a period of 12 weeks. At the school, the ladduus were distributed to the participants daily and the ladduus were consumed under direct supervision. Calculated portions of ladduus were given on occasion of holidays.

After the initial biochemical assessment each participant was assigned a number and it was entered into the randomization software based on which they were divided into the two study groups. The participants were blinded to the assigned groups throughout the research period.

Outcome measures: The parameters of HRQoL and anxiety and stress of the adolescents were evaluated at the initiation and at the end of 12 week of supplementation period with the help of World Health Organization (WHO) QOL BREF questionnaire and the Depression, Anxiety and Stress (DASS 21) questionnaire respectively. The WHO QOL-BREF instrument consists of four domains namely, physical, psychological, social and environment domain containing 26 items in all which produce a generic HRQoL score^[25] which defines the QoL of the individual. This has been validated for Indian population^[26]. Permission to use the instrument was sought from the WHO.

Physical health domain includes facets such as daily living activities, dependency on medicines, liveliness and exhaustion, vigour, aches and discomfort, sleep and capacity to work^[25]. Psychological health domain incorporates aspects related to individual's feelings regarding his body and appearance, positive and negative emotions, self-respect, personal convictions, thinking, learning, memory and attentiveness^[25]. Social health domain covered the areas of personal relationships and social support^[25]. Environmental health domain focuses on environmental factors that determine the health of an individual such as physical welfare, freedom and financial assets. It also comprises of aspects involving health and social care such as reachability to hospitals, home setting, learning opportunities, involvement in and favourable occasions for enjoyable activities and conveyance^[25].

The emotional aspects of anxiety and stress among the adolescents were evaluated using the Scale - 21 Items (DASS-21). DASS- 21 comprises of three self-report scales, each of which contain seven items. These in combination ultimately measure depression, anxiety and stress. Scores for anxiety and stress were calculated by totalling the scores for the relevant items^[27]. Anxiety rating scale assesses the physical and emotional experiences of an individual when anxious. The cut off scores for this domain are 0-7 (normal), 8-9 (mild anxiety), 10-14 (moderate), 15-19 (severe), and >20 (extremely severe)^[27]. Stress rating scale analyses the stress of an individual by assessing aspects such as difficulties in relaxing, frequency of being easily worried/distressed, angry/over-reactive and impatient. The cut off scores for the categories of normal, mild, moderate, severe and extremely severe for this scale are 0-14, 15-18, 19-25, 26-33, and >34 respectively^[27].

Quantification of Vitamin D status: A phlebotomist was employed to collect blood samples at baseline and 12 week for the assessment of serum 25OHD which was assessed using the Microparticle Enzyme Immunoassay (MEIA) technique at the Central Laboratory of the Institute.

Sample Size Calculation

Based on the study by Khadgawat *et al.* we assumed that post supplementation, the mean percentage change in serum 25(OH)D would be -5.25 % and 177.29% in the control and the experimental group respectively. A sample size of 35 in each arm would be sufficient to detect this difference with 95% confidence interval and 90% power. Kolmogorov Smirnov test was used to assess the normality of continuous variables. Impact of supplementation on the vitamin D status, quality of life and emotional disturbances of the adolescents was evaluated using Mann Whitney U test. Less than 5% was set as the cut off point to determine the significance level.

RESULTS

Enrolment for the study was done between October 2016 and October 2017 after the target sample size was achieved.

Permission to carry out the trial was granted by two schools out of the 20 schools approached by the investigator. The informed consent and assent forms were distributed to 121 children belonging to the age group of 11-16 years. A total of 80 children consented to participate in the study. Two were excluded as they had sufficient levels of vitamin D. Among the 78, 39 children each were randomly assigned to both the study groups respectively.

Four children in experimental and three in control were lost to follow up. Thus, the results for a total of 71 children are reported. Participant selection and flow is represented as a CONSORT diagram in Fig 1. The mean age of the consented participants was 12.49 ± 1.20 years, and almost half of them were girls (54.9%; 39/71). The baseline values for all the outcome variables is described in Table 1. None of the outcome variables showed any significant difference except for the physical health domain between both the study groups at baseline. Participants in the experimental group exhibited higher score for physical health domain than those in the control group ($Z=-2.17$; $P=0.03$).

The average serum 25 (OH) D levels of the adolescents was 19.5 ± 5.64 ng/mL (95% CI 16.65 to 20.17 ng/mL). According to the Lip's classification^[9], hypovitaminosis was observed among 97.26% (71 out of 73) of the adolescents with 56.8% (41 out of 73) being deficient and 40.7% (29 out of 73) being insufficient with mean S.25 (OH) D being 18.71ng/mL.

A significant difference was observed in the vitamin D status between the experimental and the control group over the period of 12 weeks ($Z=-6.28$; $P=0.00$) with the children in the vitamin D fortified group exhibiting an increase from a mean of 18.4 ng/mL at baseline to 31.2 ng/mL while those in non-fortified group showing no particular difference (baseline 20.47 ng/mL; follow-up 21.2 ng/mL).

Mann Whitney U test was used to study the effect of vitamin D fortification on the different domains of WHOQOL BREF questionnaire and the DASS 21 questionnaire. While adolescents in both the groups exhibited similar scores in the social and environmental domains before and after supplementation, an improvement was seen in the physical and psychological health domains and the anxiety and the stress scores in both the groups but, without any significant difference. Similarly, the anxiety and the stress scores also showed no significant difference before and after supplementation [Table 2].

DISCUSSION

We believe this is the first Indian study which explored the effect of vitamin D fortified food on the quality of life and emotional difficulties of adolescents. Our results suggest that HRQoL and emotional difficulties of anxiety and stress of adolescents are not particularly benefitted by vitamin D fortification or increased serum levels of 25OHD.

A potential link between vitamin D status and HRQoL of apparently healthy male adults^[16] and also among children and adolescents with lower back pain^[21], have been indicated previously. Few authors have also proposed that vitamin D status and adolescent health could be associated with each other^[28,29]. However, there is limited evidence on the impact of vitamin D supplementation on the quality of life of adolescents.

In our study although vitamin D supplementation did seem to have positive impact on the physical and psychological health domains of the adolescents belonging to the experimental group, adolescents in the control group also seemed to show improvement in those domains. The control group were given the same laddu

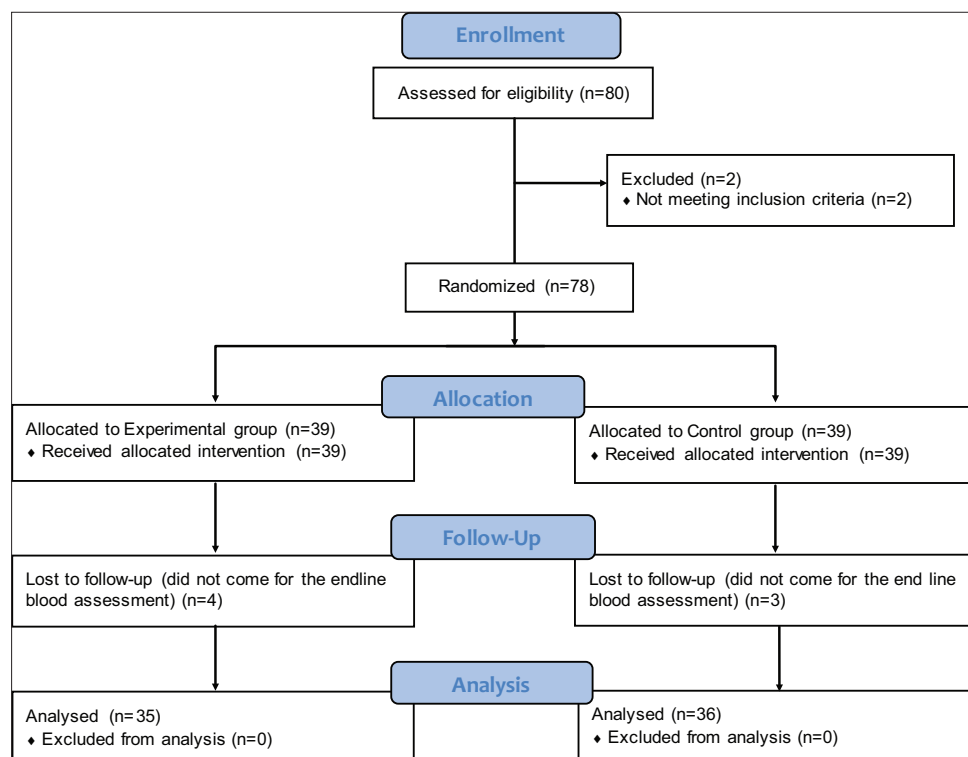


Figure 1: Flow chart of participant and enrolment flow

Table 1: Baseline characteristics of the study participants

Characteristics median (IQR)	Experimental group (n=35)	Control group (n=36)	Statistic
Age in years	12.00 (1)	12.00 (1)	Z=-1.15, P=0.246
Gender (% , girls)	54.28 (19/35)	55.55 (20/36)	$\chi^2=1.28$, P=0.262
Serum 25(OH) D, mean (SD)	18.43±5.06	20.46±6.05	t=1.53, P=0.129
Physical health domain	63.0 (13)	56 (7)	Z=-2.17, P=0.03*
Psychological health domain	69 (9)	69 (7.5)	Z=-1.55, P=0.120
Social health domain	75 (6)	75 (0)	Z=-0.63, P=0.52
Environmental health domain	75 (9)	66 (13.75)	Z=-0.62, P=0.52
Anxiety score	14 (16)	12.0 (8.5)	Z=-0.52, P=0.59
Stress score	10 (11)	9 (12)	Z=-0.36, P=0.71

*Indicates significant difference

Table 2: Data for differences in the outcome parameters pre and post supplementation among adolescents in the experimental versus control group

Outcome parameters (Difference pre and post supplementation) (Median, IQR)	Experimental group	Control group	Z value	P value
S.25OHD (ng/mL)	11.1 (9.2)	0.15 (4.6)	-6.28	0.000*
Physical health domain	0 (6)	0 (6.25)	-0.43	0.966
Psychological health domain	0 (3)	0 (0)	0.147	0.14
Social health domain	0 (0)	0 (0)	0.04	0.96
Environmental health domain	0 (0)	0 (0)	0.00	1.00
Anxiety score	-1 (2)	-2 (5)	-0.68	0.49
Stress score	0 (2)	0 (2)	-0.56	0.51

*Indicates significant difference

without the vitamin D. Since the laddu contained fibre, protein and Ca and was nutritionally dense, it probably had a positive impact on the quality of life of the adolescents which suggests that any healthy food does have a positive role in improving the quality of life of adolescents.

In relation to anxiety rating, no difference was observed in their level of anxiety post 12 weeks of supplementation. With regard to the stress scores, adolescents in the experimental group with severe and extreme severe stress had a mean reduction of -1.5 in their scores, as against their counterparts in the control group with -.5 reduction. However, the differences were not significantly different.

Our findings in the current study are in line with the observation in few other studies. e.g. Dean *et al.* reported that supplementation with vitamin D failed to affect emotional functioning and cognition among healthy young adults and the NHANES study that was conducted among adolescents and young adults did not report any significant improvement in the cognition among both the groups on vitamin D supplementation^[30].

However two randomized controlled trials have reported a significant reduction in the symptoms of depression, anxiety and stress among the participants after supplementation with high dose of vitamin D^[23,30]. In the current study, the investigators did not feel the need to fortify with higher doses of vitamin D as the used dosage which was within the Endocrine Society Guidelines^[31], sufficient enough to significantly improve the serum levels of 25OHD^[32]. Among women with type 2 diabetes and depressive symptoms weekly vitamin D supplementation of 50000 IU

resulted in reduced anxiety and improved mental health status^[33]. All of these studies have one major limitation, i.e., absence of a control group. Further randomized controlled trials in target specific population could help in deciding if supplementation with vitamin D is required for improvement for emotional health and wellbeing. To date however, no study has provided a strong evidence that vitamin D supplementation significantly reduces quality of life and emotional health^[30]. Our findings, must be considered in the wake of the large number of investigations indicating an association between low vitamin D status with emotional difficulties among adolescents with depression or psychotic illness or dialysis thereby leading to recommendations for widespread supplementation^[17,18,22,23,34] Hence, further studies with larger sample sizes and larger treatment period are warranted to affirm the role of vitamin D in improving quality of life and emotional difficulties among adolescents.

Our study has few limitations- Parathyroid hormone was not assessed for the subjects and a larger sample size could not be employed due to poor acceptance from schools (1%; 2 out of 20) and parents since the study involved blood withdrawal. Our study subjects included apparently healthy children without any medical complications and hence the results of this study cannot be generalized to individuals with emotional disorders.

CONCLUSION

This study observes and concludes that the quality of life and emotional difficulties of anxiety and stress of vitamin D deficient

children can be improved by nutritional intervention alone, without any particular need for vitamin D fortification. Further studies can be carried out on apparently healthy children versus those known to have emotional disorders to understand the impact of vitamin D in such conditions.

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