ABSTRACT: Iron deficiency along with iodine deficiency affects the developing brains, physical and mental growth of the children. Multiple micronutrient deficiencies co-exists in developing countries at a higher rate due to monotonous diets with low nutrient density. Hence, food fortification has been used as a tool to increase the content of essential micronutrients. Aim Our study was aimed to assess the efficacy of DFS supplementation on iodine and hemoglobin status of the rural school children Methods Interventional study. Rural school children (6-15 yrs) randomly divided into experimental and control group. Experimental were supplemented with double fortified salt (DFS) and controls consumed adequately iodized salt (IS). Pre and post hemoglobin concentration (Hb) and urinary iodine (UI) recorded. Results Mean Hb increased in experimental (+0.42 g/dl) (p <0.001) and decreased in control group (-0.54 g/dl) (p<0.001). The intensity of anemia classification decreased in experimental group and increased in control group. UI increased significantly in both the groups (p<0.001). Hence, DFS is proven to be beneficial compared to IS. Key words: DFS, Anemia, iodine deficiency, UIE, School age, Children

INTRODUCTION

Iron is one of the most essential micronutrient at every stage of human life, other than iodine and Vitamin A. Almost one third of the world’s population suffer iron deficiency anemia (IDA). India has its major share with high prevalence (70-80%) of IDA in children (NFHS-III) (2005-2006). With respect to iodine deficiency disorders (IDD) prevalence, it is estimated that about 7.1 crore are suffering while 20 crore people are at the risk in India. Anemia affects the oxygen carrying capacity of the cells reflected as lower levels of hemoglobin and thereby reduces the work capacity of the children. Iron deficiency along with iodine deficiency affects the developing brains, physical and mental growth of the children (Zimmermann MB et al. 2005). As reported by WHO 2001, multiple micronutrient deficiencies coexists in developing countries at a higher rate due to monotonous diets based on staple foods of low nutrient density. Typical Indian diets contain adequate amounts of iron, but the bioavailability of iron from rice and wheat, the staple cereals of Indians goes down, since absorption gets affected by phytates and other inhibiting factors. In addition to that, the intake of meat products which are rich in heme iron is low. Hence, food fortification could be the practical approach to deliberately increase the content of an essential micronutrient, in order to improve the nutritional quality of the food supply and provide a public health benefit with minimal health risk (WHO-2004). As effectively advocated public health approaches towards the control and prevention of iron deficiency are the distribution of supplements of iron and fortification of foods with a suitable iron compound. Medical input of iron is recommended for a short-term measure for the correction of anemia, while fortified foods are used to improve the iron balance over a period of time and build up iron reserves. Though the supplements are in place, compliance is not at its optimal levels. India already has a program to supply iodized salt. Hence, double fortification of salt with iron and iodine makes eminent sense (Vinodkumar M., Rajgopalan S, 2007). In India, the efforts towards producing a stable formula containing iron first and later merging iodine and iron together were pioneered by Dr. Narsinga Rao in early 70s. As a sequel to the introduction of universal iodization of edible salt as a National Policy in the country, National Institute of Nutrition (NIN) evolved the concept of double-fortified salt (DFS) with iodine and iron for controlling the deficiencies of both these micronutrients in a single measure as “one intervention controlling two problems.”
Sustainability is the key to success of supplementation strategy and it is more likely to be achieved if the supplement has a low cost, is simple to distribute, easy to administer and prevent deficiency (Bhoite R., Iyer U, 2012). It has to be efficacious enough for the school setups where the children have short and long term vacations coupled with high rate of absenteeism in rural areas. In this case daily or weekly iron supplementation as tablets may affect the compliance. Hence, looking into the scenario NIN-DFS was chosen as an effective and feasible strategy to combat both deficiencies in children. The study was aimed to assess the impact of DFS supplementation on iodine and hemoglobin status of the rural school children.

MATERIALS AND METHODS

Design A longitudinal intervention study

Study population

Primary school children (6-15 years) were enrolled from the rural schools of Waghodia, in Vadodara district, Gujarat. At baseline four schools were selected purposively (based on the availability of iodized salt) where n=1184 children were enrolled, and n=947 could complete the study due to various reasons of drop out (frequent absenteeism, not willing to participate, missing data, parent’s withdrawal of consent to name a few). Two schools were chosen as experimental group (n=442) (IS availability >50%), who were given DFS as an intervention and the other two as control group (n=505) where the children were recommended to continue consuming adequately iodized salt (IS) (IS availability >90%). Hence, DFS supplementation and Nutrition health education (NHE) were the two strategies used in the study. The study was carried out between March 2010-April 2011.

Data collection and supplementation

The children were explained the purpose of the study and the class teachers were involved for repout build up with younger children (6-9 yrs). At baseline anthropometry (height and weight), urinary iodine excretion (UIE) and hemoglobin (Hb) measurements were recorded. Weight was taken with the help of digital bathroom scale with least count of 0.01 Kg. The bathroom scale was calibrated before use. Height was taken by fiber glass tape as that was the feasible method in the field level in rural area. DFS packets (1kg) were distributed among experimental group during first week of every month and average required quantity for the family could be derived within first two months. From the third month the children received adequate packets of salt to suffice their family intake and so as to improve compliance. The intervention period was 9 months, after which the post data collection for both groups repeating anthropometric measurements, UIE and Hb sample analysis were carried out.

Methods of estimation

Urine (10-50 ml) and blood samples (0.02 µl) were drawn from the subjects at the time of enrollment and at the end of supplementation period to assess their iodine (by UIE µg/L) and iron status (Hb g/dl); Urine samples were stored at 25°C and 2 ml of toluene was added as preservative to each sample. Urinary iodine (UI) excretion was measured (at 405 nm) by modified simple microplate method (Ohashi T et al. 2000) using ELIZA reader (Tecan Autria GMBH, Europe) at ICCIDD Laboratory, New Delhi. Blood samples were collected (dry blood spots) (Brahmam GNV) for hemoglobin estimation using cyan met-hemoglobin method. The hemoglobin (Hb) measurements were performed on the samples within few hours of blood collection and were read on spectrophotometer (spectronic 20D) at 304 nm.

Definitions

IDA was defined using UNICEF/UNU/WHO (2001) criteria for Hb concentration (g/dl) in children (≥11.5: Normal, 10.0-11.49 g/dl: Mild, 7.0-9.9 g/dl: Moderate and <7 g/dl: Severe). ID was defined using WHO (2007) criteria for UIE in children (≥100 µg/L: Normal, 50-99 µg/L: Mild, 20-49 µg/L: Moderate and ≤20 µg/L: Severe) Z score was used for defining underweight, stunted, and wasted based on CDC growth standards (2005). (>2SD: marginally malnourished, -2SD to -3SD – moderately malnourished, <-3SD – severely malnourished)

Statistical analysis

The data was processed, entered and analyzed in the Statistical Package for Social Sciences for windows version 15.00 (SPSS 15.0). Growth indices were analyzed in Epi-Info, Version 3.5.3. Simple Statistical analysis was performed using Chi-square (X²) when appropriate for categorical data. Results of anthropometry and hemoglobin were reported as mean (±sd). UI was reported as median. For comparing data between paired samples, paired ‘t’ test or Mann Whitney ‘U’ test was used. 25th and 75th percentile was calculated to reveal ±2SD from the mean/median values. A two-tailed p value at <0.05 was considered statistically significant.
Ethical statement

The study approval was obtained from the ethical committee of the home institution ethical board in compliance with the guidelines issued by Indian council of Medical research (No. F. C. Sc FN ME70).

Written permissions from all the school principals and district education officer were availed to carry out the work. All the children from 1st to 6th standard were enrolled for the study. Written consent from the parents of the children (in local language) and oral consent from the children was also availed.

RESULTS

Mean age of the children was 9± 2 yrs. After the completion of study period, mean height of the children increased by 6.36 ±1.86 cm [122 (initial) - 128.42 (final)] and weight by 3.40 ± 1.80 kg [20.83 (initial) - 24.23(final)]. Overall prevalence of underweight was 72% and stunting was 46% at baseline which decreased significantly (p<0.01) to 35.1% and 13.0% respectively.

Impact on iron status

Overall initial mean hemoglobin of the children was 9.17±1.22 g/dl which decreased significantly (p<0.05) to 9.08 ± 0.88 g/dl towards the end. Hence, entire population fell under moderately anemic category (WHO-2001) at both time points. Group wise distribution showed significant (p<0.001) increase (+0.42g/dl) in Hb concentration of the experimental group after 9 months of supplementation [8.67 (initial) – 9.09 (final)], however it decreased significantly (p<0.001) in control group (-0.54 g/dl) [Table 1].

Table 1: Impact on iron status of the children

<table>
<thead>
<tr>
<th>Group</th>
<th>Hemoglobin concentration (g/dl)</th>
<th>N</th>
<th>Initial</th>
<th>Final</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>(Mean±SD) (25th-75th percentile)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Experimental</td>
<td>8.67 ± 1.24 (7.94-9.51)</td>
<td>442</td>
<td>9.09 ± 0.87c (8.55-9.66)</td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>9.62 ± 1.03a (8.99-10.28)</td>
<td>505</td>
<td>9.07 ± 0.90d (8.50-9.59)</td>
<td></td>
</tr>
</tbody>
</table>

a,b Values differed between both the groups at significance level (p<0.001)
c Initial and final values differed in experimental group at significance level (p<0.001)
d Initial and final values differed in control group at significance level (p<0.001)

Initially Hb conc. varied significantly (p<0.001) between both the groups which remained non significant towards the end. Baseline prevalence of moderate anemia in both the groups was >60% which increased significantly to >80% (p<0.01) towards the end [Figure 1(A) and (B)]. There was a percent distribution shift from severe to moderate anemia in experimental group where as in control group, the shift was from mild to moderate category.

Figure 1: Percent distribution for IDA classification

(A) Experimental group

(B) Control group
**Impact on iodine status**

Overall median UIE of the children at both time points showed iodine sufficiency among study population [145.91 µg/l (initial) - 204 µg/l (final)]. This classified the population as iodine sufficient (WHO-2007). In experimental group, the UI concentration increased significantly (p<0.001) after supplementation [132.31 (initial) – 177 (final)]. Control group also showed significant increase (p<0.001) towards the end. The UI concentration of control group was observed to be significantly higher (p<0.05) compared to experimental group at both time points [Table 2].

<table>
<thead>
<tr>
<th>Group</th>
<th>Urinary iodine concentration (µg/l) (Median) (25th-75th percentile)</th>
<th>N</th>
<th>Initial</th>
<th>Final</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Experimental</td>
<td></td>
<td>442</td>
<td>132.31 (71.38-220.25)</td>
<td>177.02* (124.16-270.49)</td>
</tr>
<tr>
<td>Control</td>
<td></td>
<td>505</td>
<td>158.18 (101.82-257.90)</td>
<td>238.22 b,d (150.60-348.64)</td>
</tr>
</tbody>
</table>

* Initial and final values differed in experimental group at significance level(p<0.001)
* Initial and final values differed in experimental group at significance level(p<0.001)
* Initial values differed between both the groups at significance level (p<0.001)
* Final values differed between both the groups at significance level (p<0.001)

Iodine sufficiency was observed to be >60% in both the groups which increased significantly (p<0.01) (>80%) towards the end [Figure 2 (A) and (B)]. In experimental group the percentage of iodine sufficient children increased by 13% [63.3(initial) – 86.4 (final)], whereas the increase was 17% [75.84 (initial) – 92.5 (final)] in control group. Rest of the deficiency categories also showed decrease in percent prevalence in both the groups.

**DISCUSSION**

Evidence based studies have reported that, physical growth and cognitive development in children are faster during early years of life, and that by the age of four years, 50% of the adult intellectual capacity has been attained and before thirteen years, 92% of adult intellectual capacity is attained (Vernon P.E. 1976). However, it is affected significantly due to micronutrient deficiencies, especially iron and iodine. Anemia affects mental and physical performances of the school children irrespective of their genders. School age is a period of rapid growth, where iron requirement for both girls and boys increase. The minimum daily dietary iron requirements are 12-15 mg/day for this age group (Bhoite R., Iyer U,
2012). However, it is surprising and moreover disappointing to know that in rural settings the children cannot meet the RDA for iron despite fully fledged MDM programs and government schemes for IFA supplementation at regular intervals. This is majorly due to lack of compliance and unawareness regarding the beneficial effects. In such a situation blanket coverage for DFS supplementation could provide partial support to the daily dietary need for iron along with meeting complete requirement of iodine. In our study, DFS could bring remarkable impact on hemoglobin status of the experimental group compared to control group. This could have been due to sustained iron release from DFS and thus the children could achieve their daily RDA for iron along with iodine. At baseline, the experimental group children could meet RDA for Iron ranging between 17-47%, which increased to 35-120% (p<0.001) towards the end due to addition of DFS in their daily diet. On the other hand, control group did not show remarkable rise in iron intake despite giving regular NHE sessions. Hence, DFS was proven to be the best cost effective measure for improving iron intake and thereby Hb concentrations in the supplemented children.

Salt is a daily diet necessity for all and it was supplemented for entire family so none of the experimental group consumed their homemade meals without DFS. Incorporating DFS to Mid Day Meal (MDM) recipes in experimental schools, also proved to be beneficial. Indeed it was interesting to note that the percent of anemic children in both the groups did not change since it was very high (98-99%), but the percent distribution under anemia classification showed a positive shift towards mild category from moderate and none of the children remained severely deficient among experimental group. However, in control group there was a significant rise in moderately anemic subjects who shifted from mild category towards the end. These findings are in compliance with earlier reports (Asibey-Berko, E et al, 2007) for an eight months DFS trial in rural Ghana, the prevalence of anemia in children decreased by 21.7% (p<0.02) and no change in control group was observed. These transitions were negative in control group since almost 20% of the children suffered moderate anemia at the end who were mildly anemic at baseline. Similar pattern was also observed (UNICEF Report 1989-92) in a UNICEF study in Delhi among the children aged 6-15 years for anemia who belonged to the orphanages, where population was wheat based. The results revealed that, in experimental group, male children showed high, clear significant decline in the prevalence of anemia from 29.5% to 4.5% (p<0.001) and non significantly in the girls from, 28.9% to 25%. However, there was significant increase in the children belonged to control group with rise of 7% in boys and 23% in girls at the end (p<0.001).

Impact on iodine status

DFS being an adequate source of iodine due to optimal fortification level (40 ppm) likewise IS, showed a significant (p<0.001) positive shift in the proportion of normal children based on UIE. The experimental population, where the availability of iodized salt was lower than the control group and thus chosen for DFS supplementation groups, showed an increase in UI sufficiency to > 80%. However, in control group where the availability of iodized salt was almost 100% but due to improper dietary - cooking - storage practices of iodized salt, they were not able to achieve 100% iodine sufficiency. After NHE as an effective intervention, control group also showed remarkable improvement in UI levels with sufficiency at >90%. In continuation to our findings, a study conducted in south India (Andersson M et al, 2008), amongst school children revealed a significant (p<0.001) increase in median urinary iodine excretion at the end of 10 months of supplementation compared to the baseline values. These values were 182, 143 and 133 µg/L in IS, DFS1 and DFS2 groups respectively at baseline and they increased to 355, 166 and 252 µg/L towards the end. There are more supporting evidences available on the impact of DFS or IS supplementation resulting in increased UI concentration in both the groups (Nair KM et al, 1998, Zimmermann M et al, 2002, 2003, 2004).

CONCLUSION

Thus, we conclude that the experimental group had an improvement in iodine and hemoglobin status. DFS supplemented children are in transition phase of iron deficiency to sufficiency with significant improvement in Hb concentrations and decrease in the intensity of the iron deficiency categories (shift from moderate to mild) contrary to the control group. Hence, we recommend that, the population who are iron and iodine insufficient, efforts to achieve sufficiency through immediate actions should be initiated by DFS incorporation in the diet of the entire family to have a sustained liberation of iron from the salt in the gut. This would meet partial RDA for iron and complete RDA for iodine of adults too at an economic cost. We also recommend long term efficacy trials on consumption of DFS in various age groups to have a complete scenario of the entire population. Further, in vitro- vivo bioavailability studies on DFS in various regional food patterns should be conducted.
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