

# Efficacy Study of Iodine Fortification of Milk on Iodine Status Markers: A Longitudinal Interventional, Controlled Study Among Schoolchildren in Morocco

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**Abstract**— Iodine deficiency is prevalent worldwide and can affect growth, mental development and learning ability of school children. In line with the Moroccan National Strategy for Nutrition 2011–2019, we undertook a study to evaluate the efficacy of the consumption of iodine fortified milk on iodine status of children living in rural region. The study is a longitudinal, interventional double-blinded (participants and assessors), controlled one concerning 200 children, aged 7 to 9 years, recruited from 3 schools in a rural, high altitude province in Morocco. The sample was divided in two groups: a non-fortified milk group (NFM, n=103) received 200ml of non-fortified Ultra High Temperature (UHT) milk and a fortified milk group (FM, n=50) received 200ml UHT milk fortified with iodine persulfate to cover 30% of Recommended Daily Intake. Urine samples were collected at baseline, 4th and 9th month and urinary iodine was determined spectrophotometrically using the Sandell-Kolthoff reaction. Severe iodine deficiency was prevalent among the FM group. After interventions at 4<sup>th</sup> and 9<sup>th</sup> month, there was a marked improvement in the case of severe iodine deficiency in the FM group (4.0% at baseline, and none

at 4<sup>th</sup> and 9<sup>th</sup> month). The NFM group had an important moderate decline from 3.9% at baseline to 1.9% at 9<sup>th</sup> month. For the moderate iodine deficiency, the prevalence was reduced from the baseline to the end of the study (9<sup>th</sup> month) ranging from 35.9% to 6.0% in both groups. Whereas for the mild iodine deficiency, the prevalence was increased at the 4<sup>th</sup> month to 63.7% and 47.7% respectively in NFM and FM groups and then decreased at 9<sup>th</sup> month to 23.3% and 22.0% respectively in NFM and FM groups. The consumption of fortified UHT milk seems to be an effective strategy to reduce iodine deficiency among schoolchildren in this rural region of Morocco. Trial registration: PACTR201410000896410. Registered 24 October 2014.

**Index Terms**— Iodine deficiency, Fortification, Moroccan schoolchildren

## I. INTRODUCTION

Worldwide, micronutrient malnutrition is considered as an important health problem affecting more than 2 billion persons [1]. Iodine is an essential micronutrient required for thyroid hormone synthesis, which in turn is necessary for normal physiological development and to control various functional processes [2]. Iodine deficiency disorders (IDD) cover a wide range of clinical conditions affecting the health and well-being of a large part of the world population. Goiter is considered the most visible and the less harmful manifestation of this deficiency. The most serious consequences involve brain damage, mental retardation, abortions and infant mortality [2], [3]. In Morocco, the severity of the IDD is considered as moderate. In 1993, the National Iodine Deficiency Disorders Control Program conducted a nationwide survey in children, the total goiter prevalence was 22% (95 % CI: 20.0–24.1) with extremes ranging from 0% to 77.4%. Moreover, average iodine excretion in urine was 86µg/l and 63% of the samples were below normal value (100µg/l). However, in some isolated mountain areas, IDD is more pronounced with high prevalence of goiter and very low concentrations of iodine excretion in urine [4].

To combat iodine deficiency (ID) and to limit the related functional disorders, the National Iodine Deficiency Disorders Control Program has set up an eradication strategy with two sets of action: 1. a short-and-medium term interventions involving supplementation of the most deficient

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populations with iodized oil; 2. a long-term intervention consisting of fortifying table salt with iodine (iodine content in dietary salt was set at 30mg/kg±10mg/kg) [5]. It's widely accepted that food fortification is one of the most cost-effective and sustainable strategies for increasing micronutrient intake, for the prevention and the control of micronutrient deficiencies in all the population [6], [7]. Moreover, given the diversity of food preferences and consumption patterns, locally and internationally, multiple products should be chosen from locally produced and commonly consumed foods as vehicles for fortification [8]. Due to the limited impact of these eradication actions and according to its strategy and orientations, the Foundation for Child Nutrition in Morocco has set up an alternative approach consisting of daily distribution of fortified milk in schools of the rural region of Morocco. Distributed milk is fortified with Iodine, Vitamin A, Vitamin D<sub>3</sub> and iron providing 30% of the recommended daily intake (RDI). Milk represents naturally a potential source of iodine, as was proved by several studies worldwide. A significant correlation between urinary iodine concentration and milk intake was observed among Italian schoolchildren consuming at least 200 ml of milk per day [9]. Moreover, milk consumption can positively influence the iodine uptake from dietary sources and/or iodine handling in the body [10]. The aim of our study is thus to determine the efficacy of the consumption of iodine-fortified milk in reducing IDD among school children in high altitude rural areas of Morocco. The results of our study will contribute in the development of national strategies, and policies aiming to reduce IDD, to improve child survival, and allow optimal physical and intellectual development.

## II. METHODS

### A. Subjects

This is a longitudinal interventional, double-blinded (participants and assessors) and controlled study that was conducted between February and October 2012, in a rural mountainous region. A total of 200 schoolchildren aged 7-9 yrs were recruited from three primary satellite-schools (Abu Antar, Iwariden and Iminifri). Participants from the Abu Antar School in the district of Ouaoula were recruited to receive a fortified milk (FM group), and participants from the two other schools, Iwariden and Iminifri schools in the district of Tifni near Damnate were recruited to receive the non-fortified milk (NFM group), located at 52 km from the FM group, to avoid errors in the dispatch of milk batches. At recruitment, a meeting with parents and school's officials was undertaken to explain the purpose of the study and to get the parent's consents. To be included in the study, children had to be aged between 7 and 9 years and should not take supplements during the study period. Children with severe malnutrition needing nutritional rehabilitation, or having chronic or severe illness requiring hospitalization or treatment were excluded from the study. The study was conducted under the ethical approval of the Ministry of

Education of Morocco. A written informed consent was obtained from each parent of recruited children.

### B. Sample size

The calculation of sample size was based on the standard deviation (2.5µg/l) of the rate of urinary excretion of iodine [11]. To observe a difference of 2µg/l with 5% level of significance and 90% power between the intervention group and placebo, and after accounting for 15% dropouts, sample size of 40 children per group was required [12].

### C. Milk

The milk used in the study was whole, flavored with vanilla and sterilized by ultra-high temperature (UHT). It was developed and produced specifically for this study by the Foundation for Child Nutrition. The firm produced 2 batches of milk that were distributed in schools during the 9 months of this study. Both fortified and non-fortified milk were identical in taste and smell, and the containers had the same appearance and packaging. The inter laboratory control was performed with Laboratoire Aquitaine Analyses (AQUANAL, France), to determine the quality and quantity of nutrients of each milk before their use in the study (table I).

**Table I Analyzed composition of fortified and non-fortified milk provided to the study participants**

| Nutritional composition     | Non-fortified milk |           | Fortified milk   |           |
|-----------------------------|--------------------|-----------|------------------|-----------|
|                             | Content 200ml      | for % RDI | Content 200ml    | for % RDI |
| Energy (Kcal)               | 154.8              | -         | 154.8            | -         |
| Protein (g)                 | 5.8                | -         | 5.8              | -         |
| Lipids (g)                  | 6                  | -         | 6                | -         |
| Carbohydrates (g)           | 19.44              | -         | 19.44            | -         |
| Fat (%)                     | 5.8                | -         | 5.8              | -         |
| Calcium (mg)                | 240                | 30        | 240              | 30        |
| Iron (mg)                   | <0.4               | <3        | 4.2 <sup>a</sup> | 30        |
| Iodine (µg)                 | 20.8               | 14        | 45 <sup>b</sup>  | 30        |
| Vitamin D <sub>3</sub> (µg) | <1                 | <10       | 3                | 30        |
| Vitamin A (µg)              | 54                 | 7         | 240 <sup>c</sup> | 30        |

<sup>a</sup>Iron: Ferrous sulfate, <sup>b</sup>Iodine: potassium iodide and <sup>c</sup>Vitamin A: Retinyl palmitate.

### D. Study design

At recruitment, socio-economic status (SES) of each participant was assessed and anthropometric parameters were measured. Children from both groups were followed up for 9 months and received daily in the morning 200 ml of UHT milk during the study period including weekends and holidays. In each school, the milk was delivered to children by the director and teachers, and during holidays children accompanied by their parents received a weekly supply. At baseline and after 4 and 9 months of intervention, urine sample from each participant was collected to evaluate iodine excretion.

### E. Socio-economic assessments

Data regarding SES were collected only at baseline from parents in both groups, using questionnaires including level of parental education, household size, total monthly expenses and monthly food expenditure.

### F. Anthropometric measurements

Body weight was measured to the nearest 0.1 Kg using an electronic scale (type Seca 750, Germany) with minimal clothing and no shoes. Height was measured to the nearest 0.1 cm using a Stadiometer (Seca type Fazzini 2 meters) without shoes. Body mass index (BMI) was calculated as a ratio of weight in kg by squared height in m<sup>2</sup>. To assess nutritional status of children, the dates of birth of the children were obtained from the school register. According to the World Health Organization, stunting and thinness were defined as Height-for-Age (HAZ) and Body Mass Index-for-Age (BAZ); Z-scores <-2, respectively. Overweight was defined as Body Mass Index-for-Age (BAZ); Z-scores >+2, and obesity as Body Mass Index-for-Age (BAZ); Z-scores >+3. Z-scores were calculated using software Anthro+ [13].

### I. Urine sampling and iodine analysis

Random urine samples were collected in the morning, between 10 a.m. and 11 a.m. to assess urinary iodine [14]. These samples were aseptically collected in 40 ml capped polypropylene tubes, aliquoted in 4 ml cryovial tubes and stored at -20°C until analysis. Urinary iodine was determined spectrophotometrically using the Sandell-Kolthoff reaction [15]. Briefly, samples of 250µl urine were digested with Ammonium persulfate at 95°C; Arsenious acid and Ceric ammonium sulfate are then added. The decrease in yellow color over a fixed time period is then measured by spectrophotometry (JENWAY 6505 UV/Vis Spectrophotometer) and plotted against a standard curve that ranged from 20 to 200µg/l [16]. According to the level of iodine in urine, the ID is classified into three classes; normal iodine status: >100µg/l; mild iodine deficiency: 50-99µg/l; moderate iodine deficiency: 20-49µg/l and severe iodine deficiency: <20µg/l [17].

Rate of improvement was calculated to assess the evolution of iodine status after 4 and 9 months of intervention.

Rate of improvement at 4<sup>th</sup> month = [(prevalence of iodine deficiency at baseline (T0) – prevalence of iodine deficiency at 4<sup>th</sup> month (T4)) / prevalence of iodine deficiency at baseline (T0)] x 100.

Rate of improvement at 9<sup>th</sup> month = [(prevalence of iodine deficiency at baseline (T0) – prevalence of iodine deficiency at 9<sup>th</sup> month (T9)) / prevalence of iodine deficiency at baseline (T0)] x 100.

### J. Statistical analyses

A per-protocol analysis was conducted on the data of participants who were fully compliant, predefined as attendance of the three urine samplings. All statistical analyses were performed using Statistical Package for the Social Sciences (SPSS, version 20.0). Anthropometric Z-scores were calculated using WHO standards. The distribution normality of quantitative variables was tested by Kolmogorov -Smirnov test. The variables normally distributed were presented as mean±standard deviation, and those non-normally distributed as median (interquartile at 25 and 75%). The nominal variables were presented as proportion and 95% Confidence Interval (Lower-Upper).

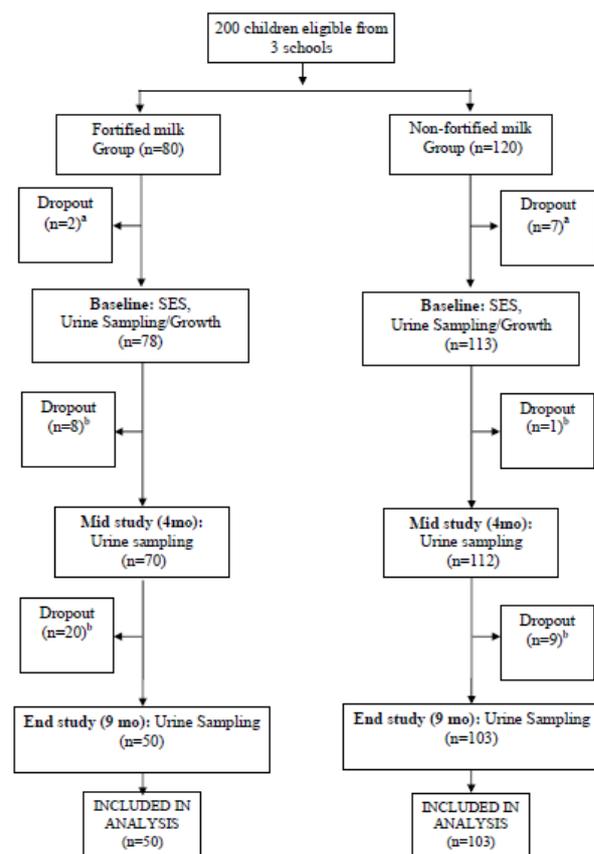
95% Confidence Intervals were determined using the Bootstrap technique based on 1000 bootstrap samples. Mann-Whitney test was used to compare medians between NFM and FM group for variables non-normally distributed. Chi-square test was used to test independence between nominal variables. In the case of cells with a theoretical frequency n < 5, we take the p-value of Fisher. Repeated measures ANOVA as an equivalent of paired t-test was used to determine the statistical significant of the mean differences in the iodine status markers within each group before and after intervention. Two-sided p-values <0.05 were considered significant.

## III. RESULTS

### A. Participants

A total of 200 eligible children were recruited from the 3 schools. The flow diagram of the study is reported in Fig.1. Overall, 47 participants withdrew from the study, 9 at recruitment because they didn't give their consents, 9 and 29 after 4 and 9 months of intervention respectively, as they refused to continue the study, were relocated out of the study area and changed school, or were excluded from the study for non-respect of the study protocol (he ceded or shared his bottle of milk to third party, etc...)

Figure 1 Flow chart



Dropouts were concerned: <sup>a</sup> Dropout = participants refused to give their consents and <sup>b</sup> Dropout= participants refusal to continue the study, relocation out of the study area and change of school or exclusion from the study for non-respect

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of the study protocol (a child ceded or shared his bottle of milk to third party, etc...).

## B. Household profiles

The socioeconomic characteristics of both groups are reported in table II. Overall, the study population has limited resources. The majority of parents are illiterates or at low level of education. Families mainly consist of 4 to 7 persons. For both groups, more than 50% of families live with less than 195 US \$ per month, and more than 60% spend monthly less than 110 US \$ for food.

## C. Baseline data

The table III illustrates the anthropometric data, the growth parameters of both groups at baseline. Overall, all recruited children were within the normal range of growth curve. In fact, the mean age was  $8.0 \pm 0.7$  years with a mean weight of  $23.2 \pm 2.7$  kg and average height of  $122.2 \pm 5.8$  cm. No statistically significant difference was recorded between the two groups. For malnutrition, the percentages of underweight (WAZ), stunting (HAZ) and wasting (BAZ) in children were 4.6% 9.9% and 0.7%, respectively. No child was overweight or obese (table III).

## D. Effect of the intervention on iodine status

For the participants who were fully compliant ( $n = 153$ ), a larger beneficial effect was seen using per-protocol analyses at 4 and 9 months. The evolution of the median urinary iodine for both groups over 9 months of intervention was reported in table IV. At baseline the median concentration of iodine was  $59.6 \mu\text{g/l}$  for the whole population and a significant difference was reported between both groups;  $55.4 \mu\text{g/l}$  and  $69.1 \mu\text{g/l}$  for the NFM and the FM groups respectively. During the intervention study, a significant improvement of urinary iodine status was observed for the two groups but was more pronounced in the FM group (Mann-Whitney U,  $p < 0.05$ ) (table IV).

The iodine status for both FM and NFM groups was evaluated at 4 months and 9 months of intervention, results are reported in table V. At the beginning of the study, 89.3% of children from the NFM group and 72% from FM group had an ID, with iodine concentration  $< 100 \mu\text{g/l}$ . After 4 months of intervention, an evident increase in iodine status was observed for both groups and was more pronounced in the FM group. Indeed, the rate of improvement was 21.1% in FM group whereas it was limited to 14.3% in the NFM group ( $\chi^2$ ,  $p < 0.05$ ). After 9 months of intervention, 32% of children from the NFM group and 28% of children from the FM group exhibited yet an ID, with improvement rates exceeding 60%. However, no significant difference was observed between both intervention groups ( $\chi^2$ ,  $p > 0.05$ ) (table V).

The distribution of iodine deficient children according to urinary excretion of iodine severity and its evolution during the intervention period is reported in table VI. Data analysis clearly demonstrated the amelioration of iodine status in both groups. Severe and mild iodine deficiencies

were reduced for both groups. Moreover, there was complete

|                                  | Total<br>(n=191)<br>% (95% CI) | Non-fortified<br>group (n=113)<br>% (95% CI) | Fortified group<br>(n=78)<br>% (95% CI) |
|----------------------------------|--------------------------------|--|---|
| <b>Level of education</b>        |                                |  |   |
| <b>Mother</b>                    |                                |  |   |
| Illiterate                       | 94.6 (91.0-97.6)               | 98. (95.0-100.0)                             | 89.4 (81.8-95.5)                        |
| Primary                          | 4.2 (1.2-7.2)                  | 1.0 (0.0-3.0)                                | 9.1(3.0-15.2)                           |
| Secondary                        | 1.2(0.0-3.0)                   | 1.0 (0.0-3.0)                                | 1.5 (0.0-4.5)                           |
| <b>Father</b>                    |                                |  |   |
| Illiterate                       | 59.4 (52.1-66.7)               | 59.6 (49.5-68.7)                             | 59.1 (47.0-69.7)                        |
| Primary                          | 32.1 (24.9-39.4)               | 32.3 (23.2-41.4)                             | 31.8 (21.2-43.9)                        |
| Secondary                        | 7.9 (4.2-12.1)                 | 8.1 (3.0-13.1)                               | 7.6 (1.5-15.2)                          |
| College                          | 0.6 (0.0-1.8)                  | 0.0 (0.0-0.0)                                | 1.5 (0.0-4.5)                           |
| <b>Household size</b>            |                                |  |   |
| ≤4 people                        | 27.1 (21.1-33.7)               | 26.0 (18.0-33.0)                             | 28.8 (19.7-39.4)                        |
| 4 to 7 people                    | 56.6 (49.4-63.9)               | 59.0 (50.0-68.0)                             | 53.0 (42.4-63.6)                        |
| ≥7 people                        | 16.3 (11.4-21.1)               | 15.0 (9.0-21.0)                              | 18.2 (12.1-25.8)                        |
| <b>Total monthly expenses</b>    |                                |  |   |
| Lower to 122 US \$               | 24.8 (18.8-31.5)               | 28.0 (21.0-35.0)                             | 20.0 (12.3-27.7)                        |
| 122 to 195 US \$                 | 29.7 (23.0-35.8)               | 23.0 (16.0-31.0)                             | 40.0 (29.2-50.8)                        |
| 196 to 244 US \$                 | 25.5 (19.7-31.5)               | 29.0 (21.0-37.0)                             | 20.0 (12.3-27.7)                        |
| 244 to 366 US \$                 | 12.1 (7.9-16.4)                | 12.0 (7.0-17.0)                              | 12.3 (6.2-18.5)                         |
| Higher to 366 US \$              | 7.9 (4.2-11.5)                 | 8.0 (4.0-12.0)                               | 7.7 (1.5-15.4)                          |
| <b>Monthly expenses for food</b> |                                |  |   |
| Lower to 110 US \$               | 63.0 (55.8-70.3)               | 60.0 (50.0-69.0)                             | 67.7 (56.9-78.5)                        |
| 110 to 147 US \$                 | 18.2 (12.7-24.2)               | 23.0 (15.0-31.0)                             | 10.8 (3.1-20.0)                         |
| 147 to 195 US \$                 | 9.7 (5.5-13.9)                 | 7.0 (3.0-12.0)                               | 13.8 (6.2-23.1)                         |
| 196 to 305 US \$                 | 8.5 (4.2-12.7)                 | 10.0 (4.0-16.0)                              | 6.2 (1.5-12.3)                          |
| Higher to 305 US \$              | 0.6 (0.0-1.8)                  | 0.0 (0.0-0.0)                                | 1.5 (0.0-4.6)                           |

disappearance of severe deficient cases in the FM group after 4 months of intervention. In the NFM group, we have noted the apparition of two cases (1.9%) with severe urinary excretion of iodine at the end of the study (table VI).

Table VII, shows the paired comparison for urine parameters, comparing FM and NFM groups. The analysis of differences of averages of urinary excretion of iodine between the beginning (T0), after 4 months of intervention (T4) and at the end of the study (T9) showed an improvement of iodine status in both groups between T0 and T9 ( $-66.8 \pm 67.3 \mu\text{g/l}$  in the case of NFM group and  $-73.0 \pm 77.3 \mu\text{g/l}$  in the case of the FM group).

## Table II Socio-economic status at baseline in non-fortified and fortified groups

There was also an improvement between T0 and T4 ( $-18.2 \pm 31.1 \mu\text{g/l}$  in the case of NFM group and  $-20.2 \pm 50.8 \mu\text{g/l}$  in the case of FM group). Interestingly, even though not statistically significant, a higher improvement was observed in the case of FM group ( $2.0 \pm 8.3 \mu\text{g/l}$  after 4 months of intervention and  $6.2 \pm 12.2 \mu\text{g/l}$  after 9 months) in comparison with the NFM group.

## IV. DISCUSSION

During the last decades, Morocco has implemented a large strategy to control ID, based especially on salt iodization. In this context, to ensure the sustainability and success of salt iodization program, the Ministry of Health and the Ministry of Energy and Mines have adopted in 1995 (revised in 2009) a regulatory text making the salt iodization obligatory. This text was to fix the declaration conditions for the manufacture of dietary salt, the specifications of this salt and the equipment used for its iodization [18], [19]. Nowadays, and after 20 years of salt fortification with iodine in Morocco, the results are far from being satisfactory. However, iodine

salt fortification is well documented worldwide and all the encountered problems were reported in both developing and developed countries [20]-[25].

In Morocco the lack of quality in the process of salt fortification could be explained by a combination of several factors mainly the absence of mastery in salt fortification, the origin and granulometric quality of the salt, the practice of retail, the mode of preservation, the storage period after iodization and the nature of material of packaging [26], [27]. An epidemiologic survey conducted in 2003 in the province of Larache, Morocco, revealed that 90% of people have iodine deficiency and interestingly only 15% of salt consumed by households contained iodine and none of them responded to the technical specifications [28].

**Table III Characteristics of growth parameters of the studied population at baseline**

| Variables                | Total                | Non-fortified group  | Fortified group     |
|--------------------------|----------------------|----------------------|---------------------|
| <b>Growth parameters</b> |                      |                      |                     |
|                          | Mean±SD or % (n=153) | Mean±SD or % (n=103) | Mean±SD or % (n=50) |
| Age, yrs                 | 8.0±0.7              | 8.1±0.7              | 8.0±0.8             |
| Weight, kg               | 23.2±2.7             | 23.1±2.8             | 23.3±2.5            |
| WAZ                      | -0.7±0.8             | -0.7±0.8             | -0.5±0.8            |
| WAZ <-2SD, %             | 4.6                  | 5.9                  | 2                   |
| Height, cm               | 122.2±5.8            | 121.8±6.1            | 122.9±5.1           |
| HAZ                      | -0.8±0.9             | -1.0±0.9             | -0.6±0.9            |
| HAZ<-2SD,%               | 9.9                  | 12.7                 | 4.1                 |
| BMI, kg/m <sup>2</sup>   | 15.5±1.0             | 15.5±1.0             | 15.4±1.0            |
| BAZ                      | -0.2±0.6             | -0.2±0.6             | -0.2±0.7            |
| BAZ <-2SD, %             | 0.7                  | 0                    | 2                   |
| BAZ W>2SD, %             | 0                    | 0                    | 0                   |

Results are presented as mean ± standard deviation or proportion (%).

BMI, body mass index; WAZ, z-score of weight for age; HAZ, z-score of height for age; BAZ, z-score of body mass index for age; Z-scores were determined according to [13].

Thus, in the absence of an effective quality control system of salt iodization, iodine salt fortification remains unsatisfactory, and consequently iodine deficiency disorders will continue to affect Moroccan population health. To control iodine deficiency disorders, alternative approaches were developed, focusing on fortification of basic food such as dairy products, these approaches are currently considered as an attractive public health strategy, as compared to micronutrient supplementation, without requiring major changes in existing consumption patterns [29]-[31]. On the other hand, it's widely accepted that in mountain and continental areas, ID is more pronounced and is accompanied by serious functional and physiological disorders especially goiter. In Morocco, a regional prevalence survey done in mountainous areas of Azilal in 1992 revealed that 65% of children examined were goitrous [32], [4].

**Table IV Evolution of the median urinary iodine for both groups over 9 months**

| Total (n=153) | Non-fortified group (n=103) | Fortified group(n=50) | p-values |
|---------------|-----------------------------|-----------------------|----------|
|---------------|-----------------------------|-----------------------|----------|

|                                   | Median (Q1-Q3)     | Median (Q1-Q3)     | ) Median (Q1-Q3)   |       |
|-----------------------------------|--------------------|--------------------|--------------------|-------|
| Before intervention, Iodine, µg/l | 59.6 (42.8-79.1)   | 55.4 (40.1-71.2)   | 69.1 (50.0-112.2)  | 0.010 |
| After 4 months, Iodine, µg/l      | 74.2 (55.8-105.5)  | 72.2 (55.5-99.5)   | 89.3 (60.2-138.4)  | 0.034 |
| After 9 months, Iodine, µg/l      | 125.3 (83.9-166.2) | 116.4 (80.9-150.7) | 138.8 (95.7-211.8) | 0.044 |

Results are presented as median (interquartiles at 25 and 75%). P-values were determined using Mann-Whitney test for medians.

**Table V Prevalence of iodine deficiency in both groups over 9 months of intervention**

|   | Total (n=153) (%) (95%CI) | Non-fortified Group (n=103) (%) (95%CI) | Fortified group (n=50) (%) (95%CI) | p-values |
|---|---------------------------|---|------------------------------------|----------|
| <b>Before intervention</b>                  |                           |   |                                    |          |
| Deficiency, <100µg/l                        | 83.7 (78.4-88.9)          | 89.3 (84.5-94.2)                        | 72.0 (62.0-82.0)                   | 0.007    |
| <b>Intervention at 4<sup>th</sup> month</b> |                           |   |                                    |          |
| Deficiency, <100µg/l                        | 70.5 (63.7-77.4)          | 76.5 (68.6-83.3)                        | 56.8 (43.2-70.5)                   |          |
| Rate of improvement (T0-T4/T0)*100          | 15.8%                     | 14.3%                                   | 21.1%                              | 0.049    |
| <b>Intervention at 9<sup>th</sup> month</b> |                           |   |                                    |          |
| Deficiency, <100µg/l (95%CI)                | 30.7 (24.2-73.3)          | 32.0 (24.3-40.8)                        | 28.0 (18.0-38.0)                   |          |
| Rate of improvement (T0-T9/T0)*100          | 63.3%                     | 64.2%                                   | 61.1%                              | 0.75     |

Results are presented as proportion and 95% Confidence Interval (Lower-Upper). P-values were determined using Chi-square test (the Chi-square value was corrected for cells with a theoretical frequency less than 5)

T0: prevalence of iodine deficiency at baseline; T4: prevalence of iodine deficiency at 4<sup>th</sup> month; T9: prevalence of iodine deficiency at 9<sup>th</sup> month.

Thus, in collaboration with the foundation for child nutrition, a leading manufacture in the distribution of dairy products in Morocco, we have undertaken the current study to evaluate the efficacy of the consumption of iodine fortified milk among schoolchildren aged 7 to 9 years old, living in rural mountain region of Morocco. The choice of this age segment was justified by the fact that school age children undergo a high degree of mental and physical development, and that ID is particularly prevalent at this age [10]. Primarily, our study showed that at baseline, 83.7% of children had ID.

Among them, 30.7% had a moderate deficiency and 3.9% a severe deficiency. These results confirm the fact that the strategy of iodine salt fortification did not attain its objectives in this region of Morocco. To our knowledge, this is the first efficacy study of iodine fortification of milk that was conducted in Morocco. In this nutritional

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intervention, the FM group showed higher improvement of iodine status in comparison with the NFM group ( $2.0 \pm 8.3 \mu\text{g/l}$  after 4 months and  $6.2 \pm 12.2 \mu\text{g/l}$  after 9 months of intervention). Our results are in agreement with previous studies conducted worldwide, using different fortification vehicles. Indeed, in Bangladesh, the consumption of the yoghurt fortified in iodine, reduced iodine deficiency in school children after one year of intervention [8].

In the same way, a study undertaken in the Philippines showed that the consumption of a drink enriched in iodine during 16 weeks had significant effects on iodine status of the school children [33]. Similar results were found in an Indian study on the consumption of a drink enriched with iodine during 14-months period which has led to the reduction of the iodine deficiency in children aged 6 to 16 years [34].

Moreover, in Durban, South Africa, a positive effect of the consumption of biscuits fortified with iodine on the performances and the health of schoolchildren aged 6-11y after twelve months was reported [35]. Furthermore, similar results were found with the consumption of bread fortified with iodine in Tasmania [36].

**Table VI Prevalence of different classes of iodine deficiency in both groups over 9 months**

|                                     |                     | Severe deficiency, <math> < 20 \mu\text{g/l}</math> (%)(95% CI) | Moderate deficiency, <math> 20-49 \mu\text{g/l}</math> (%)(95%CI) | Mild deficiency, <math> 50-99 \mu\text{g/l}</math> (%)(95%CI) |
|-------------------------------------|---------------------|---|---|---|
| <b>Total</b><br>N=153               | Before intervention | 3.9 (1.3-7.2)   | 30.7 (23.5-37.3)  | 49.0 (41.8-56.2)  |
|                                     | After 4 months      | 0.0 (0.0-0.0)   | 11.6 (7.5-15.8)   | 58.9 (52.1-66.4)  |
|                                     | After 9 months      | 1.3 (0.0-3.3)   | 6.5 (3.3-10.5)  | 22.9 (17.3-28.8)  |
| <b>Non fortified group</b><br>N=103 | Before intervention | 3.9 (1.0-7.8)   | 35.9 (28.2-43.7)  | 49.5 (40.3-58.3)  |
|                                     | After 4 months      | 0.0 (0.0-0.0)   | 12.7 (7.8-17.6)   | 63.7 (55.9-72.5)  |
|                                     | After 9 months      | 1.9 (0.0-4.9)   | 6.8 (2.9-10.7)  | 23.3 (15.5-30.1)  |
| <b>Fortified group</b><br>N=50      | Before intervention | 4.0 (0.0-10.0)  | 20.0 (12.0-28.0)  | 48.0 (35.8-60.0)  |
|                                     | After 4 months      | 0.0 (0.0-0.0)   | 9.1 (4.5-15.9)  | 47.7 (36.4-59.1)  |
|                                     | After 9 months      | 0.0 (0.0-0.0)   | 6.0 (0.0-14.0)  | 22.0 (14.0-32.0)  |

Results are presented as proportion and 95% Confidence Interval: % (Lower-Upper). Deficiency categories were defined according to [17].

**Table VII Paired comparison for urine parameters between baseline and end study comparing both groups**

| Variables                         | Non-fortified group(n=103)<br>Mean± SD | Fortified group (n=50)<br>Mean± SD | Mean Diff ± SD | 95% CI |       | p-values |
|-----------------------------------|--|------------------------------------|----------------|--------|-------|----------|
|                                   |  |                                    |                | Lower  | Upper |          |
| Iodine (T0-T4)<br>$\mu\text{g/l}$ | -18.2 ± 31.1                           | -20.2 ± 50.8                       | 2.0±8.3        | -14.5  | 18.5  | 0.81     |
| Iodine (T0-T9)<br>$\mu\text{g/l}$ | -66.8 ± 67.3                           | -73.0 ± 77.3                       | 6.2±12.2       | -17.9  | 30.2  | 0.61     |

Results are presented as mean ± standard deviation and 95% Confidence interval (CI). P-values were determined using paired t test analysis.

T0: before intervention; T4: after 4 months of intervention; T9: after 9 months of intervention.

In our study, the prevalence of moderate iodine deficiency decreased and the severe form was completely eliminated in the FM group over 9 months of intervention. For the NFM group, we also observed a decrease in the prevalence of moderate and severe forms of ID. Although the amelioration of the prevalence of iodine deficiency was more pronounced among the FM than the NFM group, the difference was not statistically significant between both groups. Similar results were found in Vietnam and the lack of significant improvement between regular and fortified groups after 6 months of intervention was explained by the fact that possibly milk provision alone could positively influence the iodine uptake from dietary sources and/or iodine handling in the body [10].

Thus, in our study the improvement of iodine status among NFM group could be explained by the consumption of non-fortified milk which contains naturally iodine at a level of  $10.4 \mu\text{g}/100 \text{ ml}$  (Table I). The daily consumption of fortified UHT milk seems to be an effective strategy to fight against iodine deficiency among school children in mountainous and rural regions of Morocco. Thus, the generalization of the distribution of fortified UHT milk under school feeding programs will be of great benefit to the National program of fight against micronutrient deficiency in Morocco and to attain the objectives of the National Iodine Deficiency Disorders Control Program, aiming to eliminate iodine deficiency by 2019 [37]. In addition, several actions should be undertaken as the implementation of a surveillance system to monitor the rate of urinary iodine excretion and to control the quality of the fortified product. These actions should be associated to an extensive program of training, awareness and education of the entire population.

The main strengths of the study concern the evaluation of the status of schoolchildren living under normal conditions as they did not undergo deworming before or during the intervention. However, the major limitations of the study are the small size of the study population due to recruitment of children from only 3 schools and the lack of the assessment of iodine concentration in milk prior to the intervention.

## V. CONCLUSION

In conclusion, the school children from high altitude rural areas of Morocco have an unbalanced nutritional profile resulting from a monotonous diet of poor quality which can cause various health hazards. Our study showed that the consumption of fortified milk led to a clear improvement in iodine status and proved to be efficient to maintain adequate reserves of body iodine. Thus, milk can be successfully used as a vehicle for iodine fortification in school feeding programs. Adequate and sustainable iodine fortification of milk is very beneficial for health and will be of great interest for the psychomotor and cognitive development of schoolchildren.

## ABBREVIATIONS

IDD: Iodine Deficiency Disorders;  
ID: Iodine Deficiency;  
RDI: Recommended Daily Intake;  
FM: fortified milk;  
NFM: Non-fortified milk;  
UHT: Ultra-High-Temperature;  
SES: Socio economic Status;  
BMI: Body Mass Index;  
SPSS: Package for the Social Sciences;  
CI: Confidence Interval;  
WAZ: Weight for age z-scores;  
BAZ: Body mass index for age z-scores;  
HAZ: Height for age Z-scores;  
SD: Standard Deviation.

## COMPETING INTERESTS

The authors declare that they have no competing interests.

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