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Effect of Moringa Leaves Powder Consumption on Young Children Nutritional and Serum Retinol Status in Burkina Faso Rural Area

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Abstract: The promotion of the consumption of indigenous plant species with high nutritional value is an important nutrition intervention in Africa rural areas. The current student was a randomized control trial of two groups (∅Gṁ and ∅Gm) with a baseline and an endline evaluation after 12 weeks. A total of 119 pre-school children received Moringa leaf powder and changes in vitamin A and anthropometric indicators of children were assessed against changes for control group children. After 12 weeks, the mean WHZ reflecting acute malnutrition declined in both groups. The mean WHZ decreased from -2.31 z-score to -1.86 z-score in group 1 (∅Gṁ) and -2.20 z-score to -1.88 z-score in group 2 (∅Gm) receiving Moringa as a dietary supplement with a statistically significant decrease in groups (p <0.001). The mean serum retinol concentration in children was below the cut-off defining VA deficiency (<0.7 µmol.L^{-1}). The baseline prevalence of VA deficiency was 56.8% in group 1 and 53.8% in group 2. Mean retinol concentrations increased significantly from 0.64 µmol.L^{-1} to 0.73 µmol.L^{-1} (p <0.001) in Group 1 (∅Gṁ (Control) and from 0.64 µmol.L^{-1} to 0.74 ± 0.05 µmol.L^{-1} (p <0.001) in group 2 (∅Gm). Significant increase was observed regardless of gender in both groups. The change between groups was not statistically significant (p=0.838). Our findings showed that the intervention was not effective enough in the change in serum retinol status in children (p = 0.379). The change in serum retinol was significantly influenced by serum retinol concentration at baseline. Therefore, the promotion of Moringa leaf consumption should be complemented by additional approaches to increase VA intake, as well as through public health measures such as deworming programs, to enhance its effectiveness in the fight against VA deficiencies and many other micronutrients.

Keywords: Nutritional Status, Serum Retinol, Moringa Oleifera, Dietary Diversification

1. Introduction

Micronutrient deficiency including vitamin A remains a public health problem in Burkina Faso. Since the first International Conference on Nutrition in 1992 food based interventions for the fight against micronutrient deficiencies were recommended [1]. Therefore, the consumption of indigenous food including animal and plant species with high nutritional value in dietary habits is promotion.

Dietary diversification is particularly appropriate for the

control of vitamin A deficiency, because of the existence of a large number of foods rich in vitamin A and pro-vitamin A, and in addition to increasing vitamin A status, dietary diversification appears to result in a better overall nutritional status [2]. Wasanwisut *et al.* showed that daily supplementation of green leaves and orange fruits and vegetables, as well as fat increase both the serum retinol and the retinol content of lactating milk in northeastern Thailand [3]. Green plant foods rich in proteins, vitamins, minerals, polyphenols, like the leaves of Moringa oleifera Lam. (Moringaceae) can be used for the prevention and correction of acute malnutrition because of their exceptional nutritional qualities [4–6].

Previous studies have identified cooked leafy vegetable sauces as one of the main sources of micronutrients for school-age children in Burkina Faso. A preliminary study in Burkina Faso found that 85.5% of preschool children's mothers know Moringa and its associated beneficial effects, it is culturally accepted by most ethnic groups, but the processing methods are not adequate [7].

In view of the important results on the consumption of Moringa leaves powder in the nutritional recovery from severe acute malnutrition [6], in clinical studies of malnourished and HIV positive [8, 9], consumption of Moringa leaves powder should be encouraged to improve the nutritional status of children, and prevent against some micronutrient deficiencies as iron and Vitamin A in view of the nutritional value of its leaves commonly used in Burkina Faso [6].

Our study report a method of plant species (*Moringa oleifera*) intervention in the rural nutrition sector and to prevent malnutrition and specific micronutrient deficiency (vitamin A).

The objectives of this study was (i) to assess the effect of Moringa leaves powder consumption on the overall nutritional status of preschool children, (ii) to assess the effect of Moringa leaves powder consumption on serum retinol concentration in the same children.

2. Methodology

2.1. Study Design and Subjects

The study was conducted in the rural district of Gaongo, Province of Bazega in the south-central part of Burkina Faso. It was a randomized control study (ϕGm and ϕGm) with an endline evaluation after 12 weeks.

A total of 119 children aged 12-59 months were randomly selected and assigned in two groups. Group 1 (ϕGm) was on a regular diet (control sample) and Group 2 (ϕGm) received Moringa leaves powder (treatment sample).

The research protocol was approved by the Ethical Committee for Health Research of Burkina Faso. Informed consent was obtained from parents prior to children's enrolment. At baseline all participants received 500 mg anthelmintic drug (mebendazole) in order to reduce parasite load. The last VA and anthelmintic supplementation in the study area was six months before the intervention. The amount of Moringa leaves powder to be incorporated was determined according to the daily intake recommended by

FAO [10, 11] and organoleptic tests carried out in the health centers [12]. Thus, participants in the treatment group (ϕGm) received 30 g of Moringa oleifera leaves powder daily during meals for 12 weeks. Moringa leaf powder was used to sprinkle meals before eating. The 30 g were distributed among the meals consumed during the day. This strategy was more conducive to consumption and avoided discards.

Weekly appointments were held to check the nutritional status of the children, motivate the mothers in relation to their children's diets, but also to make them aware of their general health status.

2.2. Data Collection

2.2.1. Socio-Economic Characteristic

Socio-economic characteristics including marital status, mother's level of education, religion, ethnicity, main professional activities, sources of income, sources of drinking water, and sanitation at the household level were collected through a semi-structured interview. In addition, data on the level of knowledge and use of vitamin A rich foods rich such as Moringa were also collected.

2.2.2. Anthropometric Measurements and Morbidity

For each children, sex, age, weight and height a well as morbidity indicators were recorded. Age was determined using official documents such as birth certificate or equivalent, health record, birth record if available. If not, the calendar of events was used to estimate the age of the children.

Anthropometric measures including height and weight of each participant were determined using standard techniques [13]. The child weight was measured using a UNISCALE type SECA Gmbh & co. kg, model 881 1021659 with an accuracy of 100 g. The height was measured using a wooden SHORR measuring board graduated in centimeter with precision to the millimeter. Arm circumference measurement was performed using the mnemonic tape [14]. These parameters were compared against the WHO recommended standards for different age to determine the anthropometric status of children.

Morbidity data include occurrence of diarrhea and/or fever in the previous 15 days [15].

2.2.3. Food Diversity

Food consumption data was collected using the 24-hour recall. Foods have were classified into six food groups as recommended by WHO. Dietary diversity and food variety score was evaluated using the country-specific WHO guide [16, 17]. Dietary diversity indicator is the sum of scores in the six food groups and is therefore ranged from 0-6. The Minimum Dietary diversity (MDD) indicator is calculated based on consumption of at least four of the following six food groups as recommended by WHO and included: (1) starchy foods (cereals, roots and tubers), (2) legumes and nuts, (3) dairy products (milk, yogurt and Cheese), (4) Eggs, meat, fish, poultry and offal), (5) vitamin A-rich fruits and vegetables; and (6) other fruits and vegetables [18]. Response options were marked and one point (1) was assigned if the food was consumed and zero points (0) if food not consumed

[19]. The dietary diversity indicator was the sum of the scores of the six food groups from 0 to 6. The food variety was calculated as the number of foods items consumed over a 24-hour period [20, 21]. Minimum Dietary Diversity (MDD) is calculated on the basis of a consumption of at least four of the six food groups. Consumption ≤ 3 food groups was considered to be low dietary diversity, while 4 groups of foods considered as minimum dietary diversity and ≥ 5 food groups such as high dietary diversity [18].

2.2.4. Serum Retinol Concentration

At baseline and endline, venous blood samples were taken early in the morning, from fasted children using a needle fitted to a 5 ml non-heparinised tube. Blood was protected from light, placed on ice and transported immediately to the medical laboratory of the District Hospital. Blood samples were then centrifuged at 1000 g for 10 min at 4°C; serum was separated into aliquots, the tubes wrapped in aluminium foil to protect the contents from light and stored at − 18°C until analysis. Serum retinol concentration was measured in duplicate, using high-performance liquid chromatography [22], at the laboratory of the Unité de Formation et de Recherche en Sciences de la Santé in Burkina Faso. We used WHO cut-off to define low and very low serum retinol as <0.7 μmol.L^{-1} and <0.35 μmol.L^{-1} respectively.

2.3. Data Analyses

Analyzes were performed using SPSS software for Windows version 24.0. Group differences in socio-economic and anthropometric variables without normal distribution were tested using the Mann-Whitney U-test for continuous variables and the chi-square test was used for categorical variables. The McNemar test was used to compare changes in the prevalence of low serum retinol and dietary diversity within groups for categorical variables. The paired t test or Wilcoxon test was used to assess changes in mean serum retinol concentration, depending on whether or not the variable was normally distributed. The differences between group 1 (ΦGṁ) and group 2 (ΦGm) for serum retinol concentration were tests using the independent samples t test. Multiple linear regression analysis was used to appraise the effect of the treatment on serum retinol concentration. For all tests, a p-value less than 0.05 was considered significant

3. Results

Characteristics of the study participants and their households at baseline are summarized in Table 1. For most characteristics, the two groups were nearly similar. In the control group (ΦGṁ), 84.9% of households had agriculture as their main source of income, as compared to 79.3% in treatment group (ΦGm).

Regarding food consumption, there was no significant difference. An average diversity score of 2.82 ± 0.9 was evaluated in control group (ΦGṁ) against 3.2 ± 1.0 in treatment group (ΦGm), with non-significant difference (p = 0.758) between the two groups. Statistical analyzes showed a non-significant difference in nutritional status (WAZ (p = 0.734), HAZ (p = 0.518), WHZ (p = 0.792)) between the groups respectively before the intervention. Our results show that 36.7% and 38.3% of children respectively 60% and 63% had diarrhea and fever respectively in control group (ΦG ṁ) and in treatment group (ΦGm).

Table 1. Socio-economic characteristics of the study population at baseline.

Households characteristics	Group 1 (ΦGṁ) n=59	Group 2 (ΦGm) n=60	p-value
Marital status (%)	90.6	94.8	
Polygamy (%)	58.5	66.7	
Type and education level (%)			
No school	50.8	60.3	
Koranic school	18.9	12.1	
literate	20.8	22.4	
Primary	3.8	1.7	
Secondary	5.7	3.4	
Superior	0	0	
Main source of household income (%)			
Without income	11.3	10.3	
Trade	3.8	6.9	
Agriculture	84.9	79.3	
Fruit and vegetable growing (%)	24.5	34.5	
Knowledge of Moringa (%)	78.0	73.6	
Individual characteristics (children)			
Average age (months)	19.88	20.27	-
Sex (% boys)	50.0	56.7	-
Low dietary diversity (%)	27.7	11.3	-
DDS mean ±SD	2.82±0.9	3.2±1.0	0.758
FVS mean ±SD	3.67±1.6	3.71±1.4	-
Diarrhea (%)	36.7	38.3	-
Fever (%)	60.0	63.3	-
HAZ mean ±SD	-2.33±1.50	-2.15±1.61	0.518
WHZ mean ±SD	-2.31±0.84	-2.20±0.81	0.792
WAZ mean ±SD	-2.89±0.88	-2.72±0.98	0.734

Table 2 below shows the results on serum retinol and the anthropometric status of pre-school children in rural area of Gaongo. Between the baseline and the end of the intervention the nutritional status of the children improved in both groups. The average WHZ index reflecting acute malnutrition increased in both groups. It increased from -2.31 z-score to -1.86 z-score in control group (ɸGṁ) and from -2.20 z-score to -1.88 z-score in treatment group (ɸGm) and the increase was statistically significant in the groups (p <0.001). However, the slight increase observed on height for age z-score (HAZ) is not statistically significant (p = 0.168 for control group (ɸGṁ)) and p = 0.337 for treatment group (ɸGm).

The study found that the mean serum retinol concentration in pre-school children was below the threshold for vitamin A deficiency (0.7 µmol.L^{-1}) in both groups. After intervention,

mean serum retinol concentrations increased significantly from 0.64 µmol.L^{-1} to 0.73 µmol.L^{-1} (p <0.001) in control group (ɸGṁ) and 0.64 µmol.L^{-1} to 0.74 ± 0.05 µmol.L^{-1} (p <0.001) in treatment group (ɸGm). There was a significant increase in mean serum retinol concentration (p <0.001) by sex in both groups. The change between the two groups was not statistically significant.

The study assessed the proportion of children in vitamin A declines in rural areas. At baseline, the proportion of children with serum retinol deficiency was 56.8% in control group (ɸGṁ) and 53.8% in treatment group (ɸGm), decreasing to 22% and 19.2% respectively (p <0.05) after intervention. However, no group recorded an average serum retinol concentration of less than 0.35 µmol.L^{-1}, defining a very deficient situation.

***Table 2.** Changes in anthropometric status and serum retinol before and after intervention of the study population.*

	Baseline	Group 1 (ɸGṁ)			Group 2 (ɸGm)			
		End line	Change †	*p*-value	Baseline	End line	Change †	*p*-value
Nutritional status*								
HAZ (mean ± SD)	-2.33±1.5	-2.22±1.4	+0.93±0.43‡	0.168	-2.15±1.6	-2.12±1.6	+0.05±0.40‡	0.337
WHZ (mean ± SD)	-2.31±0.8	-1.86±0.7	+0.31±0.51‡	0.001	-2.20±0.8	-1.88±0.8	+0.30±0.63‡	<0.001
Serum retinol status								
Serum retinol concentration* (µmol.l^{-1})								
Total	0.64±0.1	0.73±0.07	+0.93±0.05‡	<0.001	0.64±0.1	0.74±0.05	0.90±0.007‡	<0.001
Boys	0.62±0.1	0.71±0.08		<0.001	0.65±0.1	0.74±0.04		<0.001
Girls	0.66±0.09	0.75±0.06		<0.001	0.64±0.1	0.74±0.04		<0.001
Serum retinol concentration* <0.70 µmol.l^{-1} (%)								
Total	56.8	22.0	- 34.4	<0.05	53.8	19.2	-34.6	<0.05
Boys	61.9	31.6			58.6	24.1		
Girls	52.2	13.6			47.8	13.0		
Serum retinol concentration* <0.35 µmol.l^{-1} (%)	0	0	-		0	0	-	

ɸGṁ= Group of children (witness) not receiving Moringa

ɸGm= Group of children receiving 30 g Moringa leaves powder per day

* Values are means ± standard deviation

† Difference of means or proportion (after intervention minus baseline)

‡ Comparison of difference not significantly between ɸGṁ and ɸGm

Table 3 shows the predictors of serum retinol change in the study population. Our results show that treatment was not a significant determinant in the change in serum retinol status (p = 0.379). However, a high serum retinol concentration, dietary diversity, or household size were associated with the slight increase in serum retinol observed after intervention.

***Table 3.** Potentials predictors of serum retinol change in pre-school children.*

	Linear regression		*p*-value
	Coefficient of regression	Standard error	
Treatment group	0.453	0.037	0.379
Retinol pre intervention	0.362	0.053	<0.001
Frequency of consumption of VA[*] rich foods	0.014	0.013	0.317
Dietary diversity[**]	0.020	0.009	0.03
Household size	0.002	0.001	0.003

* Vitamine A

** Minimum dietary diversity

4. Discussion

This study was carried out to assess the effectiveness of Moringa leaves powder consumption in improving the anthropometric status and serum retinol concentration of pre-school children. Findings suggest significant improvement in

WHZ and WAZ in both groups, but a non-significant difference in nutritional status (WAZ (p = 0.734), HAZ (p = 0.518), WHZ (p = 0.792)) between the groups respectively before the intervention were found. After intervention, the comparison of difference of the three nutritional index was not significant between control group and treatment group

(WAZ (p = 0.510), HAZ (p = 0.473), WHZ (p = 0.318)). The slight deterioration in children's health status during follow-up in both groups would explain the good nutritional status despite the low dietary diversity observed in the study area and specifically among the children studied. Indeed, an extensive descriptive analysis of daily diet of preschool children of the study area showed that the foods most commonly eaten by these children are cereals and tubers, vegetables (especially leafy vegetables sauces), and the leafy vegetables used prepare sauce, was mainly hibiscus, baobab, corte, okra, tomato, onion [7].

There were very high levels of VA deficiency among pre-school children up to 56.8% and 53.8% in control Group and treatment group respectively, which is a major public health concern. VA inadequacy levels much higher than the 36.2% low serum concentration reported by Nana in preschool children in rural Burkina Faso [23].

After 12 months of intervention, the proportions of children with low serum concentration decreased significantly in both groups. Despite the significant decrease in control group (ϕGm̀), the proportion of 22% still remains above the 20% threshold according to WHO. In treatment group (ϕGm) the proportion of children with VA deficiency fell below the 20% threshold but still remains above 15%. As suggested by Wasanwisut the intervention was considered effective since the deficiency rate was down to 15% or less in all intervention groups, considering significantly difference [24].

Factors other than VA intake may also contribute to low serum retinol values, and these should not be overlooked. One of those factors is underlying infection, which is known to reduce serum retinol and makes serum retinol non-specific of VA deficiency [25, 26]. Data on children's health status, including diarrhoea and fever, did not show significantly correlation with serum retinol. It is important to mention that all children were dewormed with mebendazole 500mg at the beginning of the intervention. Another factor that may explain the persistence of more than 10% low serum retinol values is the presence of concurrent nutritional deficiencies which may act as limiting factors. Indeed, it is recognized that zinc deficiency is widespread among children around the world and is known to affect growth [27] and interacts with VA [28].

In both groups, sex appears to be a determinant of serum retinol concentration across the high proportions of VA deficiency in boys compared to girls. This result could be due to their vulnerability compare to female. Previous studies have reported that boys have the highest risk of vitamin A deficiency even though the reasons for their vulnerability remain largely unexplained [29].

A mean serum retinol concentration below the threshold defining a VA deficiency ($<0.7\ \mu mol.L^{-1}$) is recorded in both groups before the intervention. Significant improvement in serum retinol was observed after intervention in both groups. However, the comparison between the two groups (ϕGm̀ and ϕGm) showed that the difference is not statistically significant (p=0.838).

In the absence of an effective control group under well-controlled conditions, it is difficult to attribute the decrease in VA deficiency (from 22% to 20%) observed in treatment Group (ϕGm) to the intervention, but considering the low dietary diversity and the almost homogeneous food variety in the studied community and in both groups, a minor effect on the consumption of Moringa leaves would be likely.

Our findings showed that the intervention was not effective enough in the change in serum retinol status in children. The change in serum retinol was significantly influenced by serum retinol concentration at baseline.

These results corroborates previous studies [2, 23]. It could probably be explained by the duration of the intervention and the poorly controlled feeding of children in both groups.

High dietary diversity and household size was also associated with the slight increase in serum retinol. Similar results for household size were reported by Nana in pre-school children [23]. Kjolhede et al. reported similar results from Java; an obvious explanation, according to the authors, would be that the more people have to be fed in the family, the less VA per person is available [30].

To better conclude on the impact of Moringa on serum retinol status, the study should be repeated in a long-term controlled trial by combining oil in food consumption.

Although dietary diversity has been associated with a slight increase in serum retinol, our previous results showed that three major foods make up the bulk of the daily diet of preschool children in the area, namely, Tô (Tô is the staple food in Burkina Faso, consisting of a cereal-based thick porridge, and is adapted to several types of sauces), porridge and rice. Our previous results also showed that only a small proportion of same preschool children consumed fruits and vegetables rich in vitamin A, meat, fish and eggs [7]. Low dietary diversity is a major problem in developing countries whose daily diet is usually based on starchy foods (cereals, tubers) with one or two additional components [31]. In the developing world, multiple micronutrient deficiencies generally coexist [32] and combined supplementation becomes an appealing strategy, although controlled studies have indicated a likely competition in absorption of iron and zinc [33] and the possible adverse effect of iron supplementation on VA status [34].

In view of the important results on the consumption of Moringa leaves powder in the nutritional recovery from severe acute malnutrition, consumption of Moringa leaves powder should be encouraged to improve the nutritional status of children, and prevent against some micronutrient deficiencies as iron and Vitamin A in view of the nutritional value of its leaves commonly used in Burkina Faso [6].

In the study area, among leafy vegetables, Moringa oleifera is the most available (during dry or rainy season) but more rarely consumed because of eating habits. Previous study showed that 85.5% of preschoolers' mothers know the beneficial effects of Moringa leaves on health [7].

Therefore, the promotion of Moringa leaves consumption should be complemented by additional approaches to

increase VA intake, as well as through public health measures such as deworming programs, to enhance its effectiveness in the fight against VA deficiencies and many other micronutrients.

5. Conclusion

Our study has, to a lesser extent, assessed the effect of Moringa leaves powder consumption on the nutritional status and serum retinol concentration of pre-school children in rural Burkina Faso. According to nutritional status, between the baseline and the end line of the intervention the anthropometric status of the children improved in both groups, but a non-significant difference in nutritional status between the groups after the intervention were found. Regarding food consumption, there was no significant difference. Dietary diversity score was evaluated in control group (ϕG$\dot{m}$) and treatment group (ϕGm), with non-significant difference between the two groups.

Significant improvement in serum retinol was observed after intervention in both groups. However, the comparison of difference between the two groups (ϕG$\dot{m}$ and ϕGm) showed that the difference is not statistically significant. There was a significant increase in mean serum retinol concentration by sex in both groups. The change between the two groups was not statistically significant.

Our findings showed that the intervention was not effective enough in the change of serum retinol status in children. However, a high serum retinol concentration, dietary diversity, or household size were associated with the slight increase in serum retinol observed after intervention.

The results showed that Moringa is interesting in improving the nutritional status of children even if its effect on the improvement of serum retinol status is limited. It seems compelling to supplement with additional approaches over a longer period, under controlled conditions to better appreciate the effect of Moringa in improving the nutritional status and in VA status and other micronutrient of vulnerable children.

Acknowledgements

Conflicts of Interest

The authors declare that they have no conflicts of interest.

Limitations

The main limitation of the study was the poor control of the different quantity of food consumed by preschool children, which made it impossible to assess the adequacy ratio of nutrients.

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