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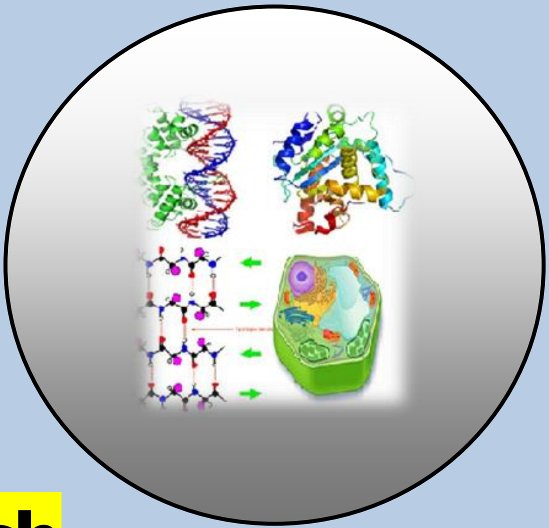

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RESEARCH PAPER

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Intake of Powdered Tea Supplemented with *Moringa oleifera* Leaf Attenuates Some Haematological Parameters in Malnourished Children

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ABSTRACT

Intake of tea supplemented with Moringaoleifera is common in many rural communities in Nigeria whereas there is paucity of information on the effect of this on haematological indices of malnutrition. This study used 124 pupils consisting of 62 malnourished and 62 non-malnourished pupils recruited from 3 primary schools in Malete, Nigeria. Collection of anthropometric data of the pupils was done using administered questionnaire. The height, weight, plasma haemoglobin (Hb), Red Blood Cell Count (RBC), White Blood Cell Count (WBC), Mean Corpsular Haemoglobin Concentration (MCHC), Platelet Count and Packed Cell Volume (PCV) of each student was also determined. The malnourished pupils were thereafter randomly assigned to groups A and B of 31 pupils each. Group A was placed on tea supplemented with moringa while group B was placed on tea alone. Treatments were done for 8 weeks after which blood was withdrawn from all the pupils and plasma haematological parameters determined. Results indicate that malnourished pupils has reduced Body Mass Index, Hb, RBC, platelet count, MCHC and elevated WBC when compared with the non malnourished pupils. Supplementation with moringa leaves powder and intake of tea alone did not significantly alter the PCV, Hb and RBC values. The plasma platelet count, MCHC and WBC were however elevated in malnourished pupils placed on tea supplemented with Moringa oleifera but not in the pupils placed on tea alone. The study reports that supplementation of tea with Moringa oleifera improves haematological indices of malnutrition and perform better than intake of tea alone.
Key words: Moringa oleifera, Anaemia, Manutrition, haematological parameters and Body mass Index.

INTRODUCTION

Moringa oleifera (horseradish) belongs to the family *Moringaceae.* It is a tree with sparse foliage leavescommonlyplanted in residential compounds in western Nigeria and/or used as aesthetic barricade to prevent wind-storm in Northern Nigeria (Keay, 1989).The plant is commonly referred to as horseradishtree. *M. oleifera*is well known for its nutritional values aswell as medicinal properties by many traditional communities. The leaf is used asvegetable in soup preparation when mixedwith other spices. Medicinally, the plant had been reported to exhibit anti-inflammatory, antihypertensive and antiulcerogenic potentials(Pal et al., 1995; Ezeamuzie et al., 1996). The efficacy of the plant as anti–bacterial agent particularly against *Bacillus subtilis, Mycobacterium phlei, Staphylococcus aureus, Salmonella* and Shigellaspecies has also been reported (Caceres et al., 1991; Sofowora, 1993). Reports indicated that most parts of the plant were applied as traditional therapies especially as arbotifacient and infertility control (Watt and Breyer, 1962; Oommachan and Khan, 1981). Important phytochemical that has been identified in the plant include moringine, moringinine alkaloids and pterygospermine alkaloids (Bharah et al., 2003). But the latter active component is abundant in the flower (Fahey, 2005). The leaves also contain isothiocyanate (Fahey, 2005) while some other study has reported the presence of fatty acids and fixed oils in the seeds (Foidl et al., 2001; Abdulkarim et al., 2005). Moringa is an important food source for people, especially in many rural communities in Nigeria. Reports from previous studies indicate that the leaves of moringa have immense nutritional potential as it contain amino acid, vitamins and minerals (Sawadogo et al. 2004; Urbain et al., 2013). This thus makes the plant a candidate for ameliorating malnutrition. During the recovery of moderate or severe and particularly clinical cases of malnutrition, it is important to improve the nutritional quality (energy density conditions, macronutrients and micronutrients) of complementary food with low-cost, locally available ingredients consistent with local cultural food habits (Sawadogo et al., 2004, Bruyeron et al., 2010). In order to make the intake of moringa leaves appealing to taste, the common traditional practice in most homes in Nigeria is to take powdered *Moringa oliefera* leaves with tea. As at the time of this study, no report has been cited in the literature that investigated the combined effect of intake of *Moringa* leaves with tea on the haematological parameters which are established indices of malnutrition. The goal of this study therefore is to assess the alteration that occur in these parameters in malnutrition and the benefits of intake of tea supplemented with *Moringa oleifera* leaves in ameliorating them.

MATERIAL AND METHODS

Plant collection and preparation of *Moringa oliefera* leaves powder

Fresh leaves of *M.oleifera* were harvested from a local garden in Malete, Nigeria. The leaves were identified and authenticate that the herabarium of Plant and Environmental Biology Department, Kwara State University, Malete, Nigeria where a voucher number (MOL14 /I) was assigned anda voucher specimen deposited. The leaves were initially washed in tap water, followed with distilled water and then shade-dried for one week. They were subsequently pulverized by grinding and passed through #40 mesh sieves. Ten (10g) of powdered *Moringa oliefera* leaves was mixed with 50ml of tea (comprising of cowbell milk and Milo). Natural drinking water was used as diluents. Each of the selected children was then allowed to take 1 cup of the combined tea and *Moringa oliefera* leaves once daily for 8 weeks.

Experimental Protocol

A total of 124 children wererecruited for this study. These pupils were all selected from 3 primary schools around Malete. The schoolswere: Pilot Primary School, Malete (60 children), Community Primary School Gbugudu (42 pupils), Elemere Community Primary School (22 children). The mean age of the pupils was 8.5 years and ranged from 9 to 13 years. The collection of anthropometric data of the pupils was done from questionnaireadministered to the pupils with written informed consent of parents or guardians following an explanation of the interest of the study. Upon pupil enrollment, information about family history, disease status, family demographics, and socioeconomic status was acquired in an interview with the children's mother and pupil in order to determine their nutritional and illness status (presence of oedema and diarrhea). The height and weight of each pupil was also measured. All selected parameters were first recorded in the pupils. The identified malnourished pupils were then randomly assigned into two groups A and B using computer assigned number. Group A (31 pupils) was placed on tea supplemented with *Moringa oliefera* leaves, while group B (31 pupils) was placed on tea alone.The daily dose of 10 g of powdered moringa dry leaves was decided upon based on previous report (Fuglie, 2001). The monitoring of the children was carried out during the day and in the afternoon they went home with their parent/guardian. Treatment was done for 8 weeks after which the experiment was terminated

The experimental procedure and protocol used in the study were approved by the ethical committee on the use of animal and human subjects, College of Pure and Applied Sciences, Kwara State University, Malete, Nigeria. The guidelines were in accordance with the Internationally Accepted Principles for Laboratory Use (WHO, 1998).

Blood Collection

Both prior to intervention and eight (8) weeks after continuous intake of *Moringa oleifera* leaves and tea supplements, 4 ml of blood samples were collected from each subject into EDTA specimen bottles using 5ml syringes.

Haematological Analysis

Haemoglolin concentration (Hb%) was measured by Drabkin's colorimetric method (cyanomethemolgobin formation) and packed cell volume (PCV) was estimated by scale of microhameatocrit reader(Duncan et al., 1994).The concentration of red blood cells and white Blood Cells were measured in the given blood sample and was expressed as volume of cells (Coles 1986). The concentration of blood platelet was determined by conventional method while the mean corpuscular hemoglobin consternation (MCHC) was calculated using the formula

MCHC (g/dl) = Hb x 100/PCV

The body mass index (BMI) was calculated using the formulae

BMI (Kg/m^2) = Weight (Kg)/ $Height^2$ (m)

All the parameters studied were measured in triplicate.

Statistical analysis

Results are expressed as the Mean ± SD. Statistical significance was evaluated by one-way analysis of variance (ANOVA) using SPSS version 10 and the individual comparisons were obtained by the Duncan multiple range test (DMRT) (Duncan, 1957). A value of $P<0.05$ was considered to indicate a significant difference between groups.

RESULTS

The demographic status of the study population is shown in Table 1. A total of 130 pupils were recruited for the study. Sixty five (65) of these pupils were classified as malnourished children while 65 represent the control subject. The mean age of the malnourished children (10.7 years) was not significantly different from the mean age of the control subject (10.3 years). Forty one (41) representing 63.07% of the malnourished pupils were female while 36 (53.39%) of the control subjects were females. Among the malnourished children, 24 (36.92%) were males while 29 (44.61%) of the control pupils were males. The mean body mass index (BMI) of the malnourished pupils was 16.46±2.11 which is lower than the BMI of 20.51±3.21 obtained for the control subjects. Thirty four (52.31%) of the mothers of the malnourished pupils had no formal education whereas 15 (23.07%) of the control subjects mothers had no formal education. The result also indicates that 18 (27.69%) and 13 (20.00%) of the mothers of the malnourished pupils had primary and secondary school education respectively whereas 27 (41.53%) and 23 (35.38%) of the mothers of the control subjects had primary and secondary school education respectively. The result thus indicates that in comparison with the control subjects, most of the mothers of the malnourished pupils had no formal education whereas most of the mothers of the control subjects had primary education.

Results of the haematological analysis of the study population (Table 2) showed that the packed cell volume (PCV) of the malnourished children was 33.24±1.44% which was not significantly different from 37.90±2.57% observed for the control population. The observed haemoglobin (Hb) value was 14.35±1.90g/dl and 10.01±1.23 g/dl for the non-malnourished and malnourished population respectively and the difference was significant ($p<0.05$). Similarly, the Red Blood Cell (RBC), platelet count and Mean Corpsular Haematocrit Concentration (MCHC) was observed to be lower in the malnourished population compared with the non-malnourished control pupils. The WBC value of 8.72±3.21 ($x10^9L$) observed in the malnourished population was however higher than the value of 5.89±2.41 ($x10^9L$) observed in the non- malnourished population.

Table 3 is the result of the haematological parameters after intervention. No significant difference ($p>0.05$) was observed in the PCV value of the malnourished children placed on moringa supplemented tea and those not supplemented with moringa. When these values were compared with the baseline, they were also observed not to be significantly different. Supplementation of the tea of malnourished pupils with moringa, was also observed not to have significantly ($p>0.05$) altered the heamoglobin value. The observed value of 10.09±1.31 was not different from the baseline value of 11.01±1.23 g/dl. This observed value was also not significantly ($p>0.05$) different from the value of 12.07±1.01g/dlobserved when diet was not supplemened with moringa. Supplementation of the tea with moringa in malnourished pupils however raised the platelet count from the baseline value of 224.46±33.67 to 302.45±45.13 ($x10^9L^{-1}$). The platelet count of 227.31±10.12 ($x10^9L^{-1}$) observed in malnourished pupils whose tea was not supplemented with moringa was however not different from the baseline value. This result was similar to that observed for MCHC where supplementation with moringa was observed to have raised the MCHC value above that of the baseline whereas the MCHC value of the pupils placed on tea not supplemented with moringa was not different from the baseline value.

A raised WBC value to 10.28±2.11 ($x10^9L^{-1}$) from the baseline value of 8.72±3.21($x10^9L^{-1}$) was observed with moringa supplementation. The observed value in pupils not placed on the supplemented tea was however not different from the baseline value but lowered than the value observed in subjects placed on supplemented diet. The RBC of the malnourished pupils placed on moringa supplemented diet was not different from the baseline value. The observed value was also not different from that of the subject placed on tea without supplementation.

DISCUSSION

BMI is a widely usedparameter and it is well admitted thatBMI<20 indicates mild or severe malnutrition (Campilo et al., 2003). Data from this study indicates that the malnourished pupils have lower BMI compared with the control group. The result also indicates that the level of education of the mothers may be a contributing factor to the nutritional status of the pupils. This is because the results obtained indicate that many of the mothers of the malnourished pupils do not have any formal education. This thus suggests that there is a positive correlation between the level of education of mothers and children nutritional status. Data from this study agrees with report from previous authors who reported a definite association between the literacy of the mothers and the nutritional status of their children (Eze et al., 2005; Vinod and Robert, 2010; Benta et al., 2012). Previous studies had reported that protein energy malnutrition is associated with haematological changes (Pooja et al., 2007; Saka et al., 2012; Urbain et al., 2013). Data from the present study agrees with thee reports. Malnourished pupils in this study (as different from the non malnourished children), presents low haemoglobin, Red blood cell and platelet count. The result thus indicates that malnutrition may result in anemia condition. This is in conformity with reports from previous authors (Warrier, 1990; Urbain et al., 2013).The reduction in the level of RBC and MCHC in the malnourished population) observed in this study) when compared with the control group may be suggestive of adaptation to lower metabolic oxygen requirements and decrease in lean body mass in malnourished pupils (Saka et al., 2012) or changes in the plasma volume as well as the intracellular body water in the body (Uner et al. 2001).All these are confirmation of anaemia in malnutrition. The anaemia of malnutrition may be attributable tovarious factors such as iron deficiency, and /or reduced red cell production in adaptation to a smaller lean bodymass (Olaf et al., 2005). Other factors that have been implicated include erythropoietin deficiency, deficiencies ofvitamins (folic acid, B12,) or trace elements (copper, zinc), infections and chronic diseases (Meffat et al., 1994; Olaf et al., 2005).The decrease in platelets count seen in malnourished pupils in this study can be attributed to a purported decrease in bonemarrow activities which indirectly affect mega karyocyte functions (Uner et al., 2001; Saka et al., 2012). An increased White Blood Cell Count (WBC) was also observed in the malnourished population in this study. The serum level of WBCs may indicate an organism's defensive capability against infections (Uner et al., 2001; Olaf et al., 2005). The increased WBC observed in the malnourished population may therefore be a confirmation of infection in malnutrition. The results of the investigation following intervention showed that supplementation with powdered *Moringa oleifera* leaves in malnourished children does not significantly raised the level of HB, RBC and PCV. The result is similar to the observation in malnourished children placed on tea without supplementation with moringa.

Finding from this observation is similar to the report of Urbain et al., (2013). A study by Buseri et al., (2010) reported the reference values for some haematological parameters in children aged 9-14 years to be 12.5-14.5g/dl, 35-45%, 4.8-4.9x10^{12}/L and 130-405x109/ L for HB, PCV, RBC and platelet count respectively. The values observed for these parameters in the present study indicates that although these parameters were not significantly elevated after intervention, both the PCV and platelet count were within the reference range in these two treated groups of malnourished pupils. The RBC and PCV were however below the reference range even after intervention with *Moringa oleifera.* It has been suggested that Improvementin the overall nutritional status does not necessarilysignify changes in biological parameters such as hemoglobin (Compaore et al., 2011; Urbain et al., 2013).Result from the present study agrees with this report. Iron supplementation during nutritional rehabilitation normally improves Hb levels significantly, but thisvariation is in turn affected by weight gain, as highweight gain is not conducive to significant changes in Hblevels (Georgieff et al., 2002; Thorsdoffir et al., 2003).

Table 1. The socio-demographic characteristics of the study population.

Character	Non malnourished (n= 62)	Malnourished (n= 62)
Age (year)		
Range	9-12	8-13
Mean	10.3±2.1	10.7±1.1
Gender		
Female	36 (53.39%)	41 (63.07%)
Male	29 (44.61%)	24 (36.92%)
BMI		
Range	19.34-23.26	15.13- 17.21
Mean	20.51±3.21	16.46±2.11
Maternal education		
None	15(23.07%)	34 (52.31%)
Primary	27 (41.53%)	18 (27.69%)
Secondary	23 (35.38%)	13 (20.00%)
Post secondary	none	none

Table 2. Haematological values of the study population.

Prameters	Treatment	
	Non malnourished	Malnourished
PCV (%)	37.90±2.57^{a}	33.24±1.44^{a}
Hb (g/dl)	14.35±1.90^{a}	10.01±1.23^{b}
WBC (x$10^{9}L^{-1}$)	5.89±2.42^{a}	8.72±3.21^{b}
RBC (x10^{9} cell/mm^{3})	4.21±0.34^{a}	4.05±0.22^{b}
Platelet count (x$10^{9}L^{-1}$)	244.90±46.12^{a}	221.46±33.67^{b}
MCHC (gHb/dlRBC)	35.40±4.79^{a}	24.92±4.23^{b}

- PMOL= Pupils placed on tea + powdered *Moringa oleifera* leaves.
- WPMOL= Pupils placed on tea without powdered *Moringa oleifera* leaves.
- Values represent mean± standard deviation (n= 62).
- Values with different superscript in the same row are significantly (P<0.05) different.

Table 3. Haematological values of malnourished pupils after intervention

Parameters	Treatment		
	Baseline	PMOL	WPMOL
PCV (%)	33.24 ± 3.44^{a}	37.93 ± 2.42^{a}	33.82 ± 1.12^{a}
HB (g/dl)	11.01 ± 1.23^{a}	10.09 ± 1.31^{a}	12.7 ± 1.01^{a}
WBC ($x10^{9}L\text{-}^{1}$)	8.72 ± 3.21^{a}	10.28 ± 2.11^{a}	9.8 ± 0.11^{a}
RBC ($x10^{12}L\text{-}^{1}$)	4.21 ± 0.34^{a}	4.46 ± 0.12^{a}	4.12 ± 0.18^{a}
Platelet count ($x10^{9}L\text{-}^{1}$)	224.46 ± 33.67^{a}	302.4 ± 45.13^{b}	227 ± 10.12^{a}
MCHC (g/dL)	24.92 ± 4.23^{a}	32.41 ± 2.20^{b}	22.78 ± 2.11^{a}

- PMOL= Pupils placed on tea + powdered *Moringa oleifera* leaves.
- WPMOL= Pupils placed on tea without powdered *Moringa oleifera* leaves.
- Values represent mean± standard deviation (n=31).
- Values with different superscript in the same row are significantly (P<0.05) different.

The observed increase in platelet count with supplementation suggests the stimulatory effect of *Moringa oleifera* on thrombopoietin. This may implies that the powdered leaves can promote thrombopoiesis, repair the minute vascular damage and considerably manage thrombocytopenia in malnutrition (Compaore, 2011). This was not observed in children whose tea was not supplemented with moringa leaves suggesting that moringa leaves intake works better than just tea in managing thrombocytopenia in cases of malnutrition.
The study also revealed that supplementation with Powdered *Moringa oleifera* leaves elicits significant increase in thelevel of white blood cell counts. The observed increase in the levels of thisparametersuggest that the principal function of phagocytes, which is to defend against invading microorganisms by ingesting and destroying them, thereby contributing to cellular inflammatory processes, will be enhanced during intake(Paul, 1993; Swenson and Reece, 1993; Adedapo et al., 2005).

CONCLUSION

This study affirms that malnutrition results in haematological alterations. It also demonstrated that *Moringa oleifera* powdered leaves when taking along with tea could be beneficial in malnutrition than taking tea alone. Work is on-going in our laboratory to investigate variations in some other nutritional indices of malnutrition in order to corroborate results from this study.

Conflict of interest

The authors declare that there are no conflicts of interest and that the authors of this manuscript have no financial or personal relationship with any organization which could influence the work.

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