



**EFFECT OF VITAMIN A-MINERAL SUPPLEMENTATION ON  
SERUM RETINOL AND OVERALL NUTRITIONAL STATUS OF  
SCHOOL CHILDREN AGED 6-10 YEARS IN WAKISO DISTRICT,  
UGANDA: A RANDOMIZED CONTROLLED TRIAL**

**BY  
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## Declaration

I, **GILBERT MANGUSHO** do declare that this is my own original work and it has not been submitted anywhere for any award whatsoever.

Signature & Date

.....

I have duly supervised the student in the research and writing of this thesis. I affirm that to the best of my knowledge, the above declaration is true.

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Signature ..... Date .....

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## **Dedication**

To my beloved mother Jane-Frances Kweyey, a constant inspiration in my life.

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## Acronyms

ACC/SCN	United Nations Administrative Committee on Coordination/Sub-Committee on Nutrition
CDC	Centers for Disease Control
CHD	Child Health Days
CIDA	Canadian International Development Agency
EPI	Expanded Program on Immunization
FAO	Food and Agricultural Organization
FFQ	Food Frequency Questionnaire
FMFH	Feeding Minds Fighting Hunger
FNB	Food and Nutrition Board
HAZ	Height for Age z-score
HIV/AIDS	Human Immunodeficiency Virus/ Acquired Immune Deficiency Syndrome
HKI	Helen Keller International
HPLC	High Performance Liquid Chromatography
IDA	Iron Deficiency Anaemia
IFPRI	International Food Policy Research Institute
IOM	Institute Of Medicine
IU	International Units
IVACG	International Vitamin A Consultative Group
MDG	Millennium Development Goals
MF	Micronutrient Forum
MoH	Ministry of Health
NAR	Net Attendance Ratio
PCD	Partnership for Child Development
RBP	Retinol Binding Protein
SAC	School Age Children
U5MR	Under 5 Mortality Rate
UBoS	Uganda Bureau of Statistics
UDHS	Uganda Demographic and Health Survey
UNCST	Uganda National Council for Science and Technology
UNICEF	United Nations International Children's Emergency Fund
UPE	Universal Primary Education
VA	Vitamin A
VAD	Vitamin A Deficiency
WAZ	Weight for Age z-score
WHO	World Health Organization
WHZ	Weight for Height z-score

## Definition of Terms

Biofortification	The process of breeding staple crops to enhance the content of essential micronutrients such as iron, zinc and provitamins A.
Fortification	Addition of calculated amounts of a micronutrient to an industrially processed food to address demonstrated deficiency of the nutrient or to raise dietary intake of the nutrient and prevent deficiency disorders.
Malnutrition	Having inadequate or excess intake or inappropriate kind of nutrients that manifests in growth (stunting, under-/overweight, wasting, etc), development and/or health and well-being.
Micronutrient	Nutrient required by the body in small quantities.
Nutritional Status	A measurement of the extent to which individuals' physiological needs for nutrients are met.
Placebo	A preparation/mixture without the ingredient/nutrient of interest.
Stunting	Being too short for one's age. "Stunted" is having height for age z-score more than 2 standard deviations below the mean height for age z-score for the reference population.
Supplementation	Adding a nutrient to the usual diet to complement a diet deficient in the nutrient or to meet a required level of intake for that nutrient.
Underweight	Having low weight for one's age. "Underweight" is having weight for age Z-score more than 2 standard deviations below the mean weight for age z-score for the reference population.
Vitamin A Deficiency	Inadequate vitamin A stores for body functions. Measured by Serum retinol levels below 20 micrograms per deciliter.
Wasting	Having low weight for one's height. "Wasted" is having weight for height Z-score more than 2 standard deviations below the mean weight for height z-score for the reference population.

## Abstract

**Introduction:** School aged children are faced by many nutritional problems including deficiencies of major micronutrients such as Vitamin A, Iron and Zinc. Vitamin A Deficiency is a public health problem in Uganda and efforts for its control have focused mainly on preschool children and lactating mothers but hardly on the school-age children. Supplementation is one way of improving the micronutrient status of children. Multiple-micronutrient supplementation is envisaged as one of the most cost-effective approaches to addressing the micronutrient problems in children. However, research in this area has been scanty.

**Overall Objective:** To assess the effect of Supplementation with vitamin A and other micronutrients (Iron and Zinc) on serum retinol and the general nutritional status of school children (6–10 years).

**Study design:** Primary schoolchildren (6-10 years) selected from three day primary Schools in Wakiso district were randomly allocated to two vitamin A-mineral supplementation groups: A (Vitamin A + Iron) and B (Vitamin A + iron + Zinc) in a randomized double-blind placebo-controlled study design. Serum retinol, weight and height were the parameters monitored in four school terms (16 months). Treatment groups were tested for significance at  $p < 0.05$ .

**Results:** Height for age and weight for age increased in all the groups. Body Mass Index for age increased only in the group that received vitamin A and iron without zinc (group A) but decreased in the others. These changes in nutritional status were, however, not significant. The level of stunting in group A (Vitamin A + Iron) remained at 9.5% while that in B (Vitamin A + Iron + Zinc) reduced from 9.8% to 0%. Underweight reduced by 50% in both groups A and B from 9.5% to 4.8% and 4.9% to 2.4% respectively, but increased by 33% in the control. Wasting on the other hand decreased from 4.8% to 2.4% in group A but increased in the other groups. There were significant increases in mean serum retinol in both groups A and B after supplementation but not in the control. Vitamin A Deficiency reduced

from 25% to 20.5% in group A, 22% to 18% in group B but increased in the control from 27.8% to 30.6%. The Vitamin A status was not associated with any of the anthropometric indices: stunting, underweight and wasting.

**Conclusion and recommendations:** Supplementation significantly reduced the prevalence of underweight but not stunting among the children. Serum retinol levels of the children were significantly increased in the supplemented groups and not in the control. The supplementation, irrespective of the regimen, produced significant reductions in the prevalence of Vitamin A Deficiency. Vitamin A status was not associated with weight-, height- or BMI for age in the school children. Supplementation with multiple micronutrients including vitamin A, Iron and Zinc was found to have beneficial effects in this cohort of school children. More studies are needed to better understand the contribution of schoolchild supplementation with micronutrients in areas where deficiencies of these nutrients are common, before a recommendation to supplement school children with multiple micronutrients can be considered.

# CHAPTER ONE

## INTRODUCTION

### 1.1 BACKGROUND

Evidence of high malnutrition and disease morbidity among school age children raises concern that the progress made in child survival and basic education programmes could be retarded. This group is faced by nutritional problems mainly stunting, underweight, anaemia, Iodine Deficiency and Vitamin A Deficiency. In countries experiencing the 'nutrition transition', however, overweight and obesity are increasing problems among the school-age children. Other health problems facing this age group include malaria, helminth infections, diarrhoeal diseases, respiratory infections and the direct and indirect effects of HIV/AIDS (Drake *et al.*, 2002).

#### 1.1.1 Nutritional Status

The nutritional status of a population is used to determine the magnitude of malnutrition that is prevailing at a particular time in the population. Malnutrition refers to disorders resulting from an inadequate (undernutrition) or excessive diet (overnutrition) or from failure to absorb or assimilate dietary elements. A deterioration of nutritional status is also a function of infections, poor sanitary conditions, repeated infections, diarrhoea and inadequate care.

Several parameters are used to assess the nutritional status of a population or an individual. Stunting, underweight and wasting are common anthropometric indicators of undernutrition. Stunting is a physical indicator of chronic or long-term malnutrition and is often linked to poor mental development. It is a cumulative process that is not easily reversed. Underweight is an indicator of both chronic and acute undernutrition while wasting is an indicator of acute undernutrition. On the other hand, overnutrition is measured by overweight and obesity. There are also clinical conditions that are associated with under- or overnutrition.

#### ***1.1.1.1 General Situation of Hunger and Malnutrition in the world***

There is large-scale hunger and malnutrition in the world. More than 850 million people today are chronically undernourished. Approximately 200 million children under five years of age suffer from acute or chronic symptoms of malnutrition. Majority (over 95%) of the undernourished are in developing countries: Asia and the Pacific –60% (India alone-27%); Sub-Saharan Africa –25%. Sub-Saharan Africa is also the region with the highest proportion of its population undernourished (FAO/FMFH, 2007).

#### ***1.1.1.2 Nutritional status of Preschool Children***

The prevalence of stunting, underweight and wasting varies by region and sub-region throughout low-income countries. The African region has the highest estimated prevalence of stunting worldwide (20.2-48.1%) and has the lowest rate of improvement. The prevalence of stunting in Asia (32.8-43.7%) is also high, particularly in South Central Asia and is significantly lower (9.3- 24%) in Latin America and the Caribbean (ACC/SCN, 2001).

The prevalence of underweight and wasting follow similar regional patterns to the prevalence of stunting. For Africa in the year 2000 underweight ranged from 14 to 36.5%; Asia 28.9 to 43.6% and Latin America and the Caribbean 3.2 to 15.4%. In 1995, estimated prevalence of wasting in Africa was 9.6%, Asia 10.4% and Latin America and the Caribbean 2.9% (Drake *et al.*, 2002).

#### ***1.1.1.3 Nutritional Status of School Children***

Few representative data are available on the levels of malnutrition in school-age children. However, the available data on school-age children follow the regional pattern of the more extensive representative data from surveys of preschool children. Children stunted at school age are likely to have been exposed to poor nutrition since early childhood and that the degree of stunting tends to increase throughout the school-age years. Low-birth-weight infants are more likely to become stunted children than those of normal birth-weight. Stunted children tend to become stunted adults and, stunted females of childbearing age tend to have



low-birth-weight infants (ACC/SCN, 2001; de Onis *et al.*, 2000) and the cycle of intergenerational malnutrition is perpetuated (UNICEF, 2006c; Setboonsarng, 2005).

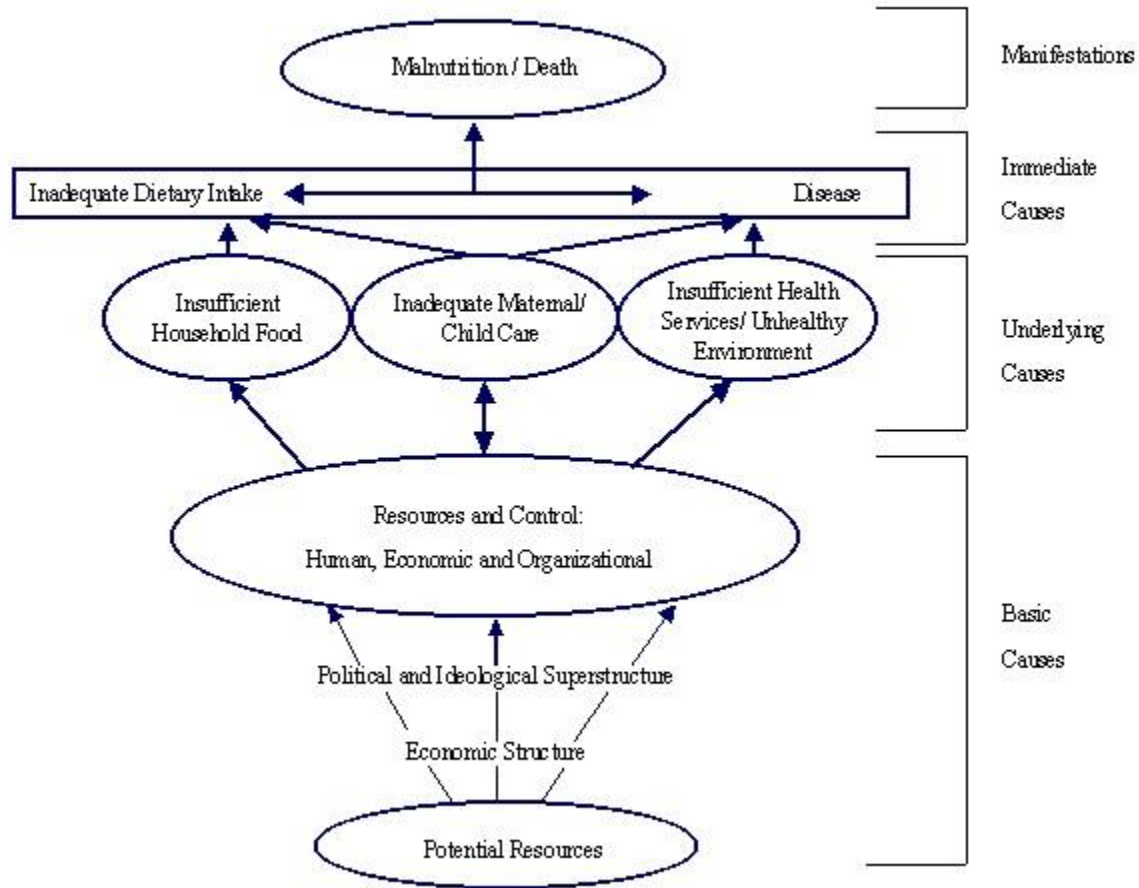
Underweight among school-age children, like stunting, can reflect a broad range of insults such as prenatal undernutrition, deficiencies of macro- and micronutrients, infection and, possibly, inadequate attention by caregivers. Wasting, which reflects acute malnutrition, is not as common as either stunting or underweight in school-age children.

Despite the absence of representative data, there is growing evidence of considerable burden of morbidity and mortality due to infectious diseases and undernutrition in school children. Fernando *et al.* (2000) found high rates of anaemia, stunting and wasting among Sri Lankan schoolchildren which were also comparable to those among Indian and African school children.

#### ***1.1.1.4 Causes of Malnutrition***

Due to the complexity and variety of malnutrition problems among and within countries and other geographical and social strata, the precise causes of malnutrition are accordingly specific. Nevertheless, UNICEF has adopted a model (Fig. 1.1) that can be used in various situations to explain the causes of malnutrition. In this model, malnutrition (and death) results from either inadequate nutrient intake or disease both of which are brought about by one or a combination of three interrelated conditions: inadequate household food security, inadequate maternal and child care and insufficient health services and an unhealthy environment. These underlying causes have a base in the social, political and economic environment that determines the utilization of potential resources (Maxwell & Smith, 1992).

Since the 1990s, there has been a renewed focus on the quality and/or composition of diet especially with respect to micronutrients. Increased understanding of the extent and far-reaching consequences of micronutrient deficiencies especially of iron, iodine and vitamin A, together with the existence of proven low-cost methods for preventing these deficiencies, have strengthened the focus on micronutrients.



**Figure 1.1 Conceptual framework of Malnutrition**

Source: [http://www.tulane.edu/~panda2/Analysis2/nutrdata/causes\\_of\\_malnutrition.htm](http://www.tulane.edu/~panda2/Analysis2/nutrdata/causes_of_malnutrition.htm)

#### ***1.1.1.5 Consequences of Malnutrition***

Poor nutrition and health result in a reduction in overall well-being and quality of life, and in the levels of development of human potential. Malnutrition can result in productivity and economic losses, as adults afflicted by nutritional and related disorders are unable to work; education losses, as children are too weakened or sickly to attend school or to learn properly; health care costs of caring for those suffering from nutrition-related illnesses; and costs to society of caring for those who are disabled and/or their families as well. Further, malnutrition is an important factor in the high Under-five Mortality Rate (U5MR) due to

preventable diseases and infections, such as measles, diarrhoea, malaria and pneumonia (Drake *et al.*, 2002; UNICEF 2006c).

#### ***1.1.1.6 Micronutrient Malnutrition***

Malnutrition in the form of deficiencies of essential vitamins and minerals continues to cause severe illness or death in millions of people worldwide. More than 3.5 billion people are affected by iron deficiency, 2 billion are at risk of iodine deficiency and 200 million pre-school children are affected by insufficient vitamin A. Even mild forms of these deficiencies can limit a child's development and learning capacity early in life, which can lead to cumulative deficits in school performance, resulting in higher school drop-out rates and a high burden of illiteracy in future populations (Drake *et al.*, 2002). Excessive intake of micronutrients such as Vitamin A, though not common, may also have undesirable effects on health, growth and development (WHO/FAO, 2004; Penniston & Tanumihardjo, 2006).

#### **1.1.2 Deficiencies of Vitamin A, Iron and Zinc**

Vitamin A (VA) plays an important role in vision, growth, reproduction and immunity (WHO/FAO, 2004). Deficiency of vitamin A in children has been referred to as a nutritionally acquired immune deficiency (Higdon, 2003), which puts them at increased risk of not only morbidity and mortality from infectious diseases but also decreased physical growth and blindness in early childhood (Drake *et al.*, 2002; FAO/FMFH, 2006; Olson, *et al.*, 2006; Smuts, 2005; UNICEF, 1998). The vicious cycle of Vitamin A Deficiency (VAD) and infections (Haselow *et al.*, 2004) in young children including those of school-age can have very devastating consequences. There is now extensive evidence that VAD is widespread in young children in many developing countries with a global estimate of over 250 million children under five years of age affected by VAD (WHO/FAO, 2004).

Over 1.2 billion people worldwide suffer from Iron Deficiency Anaemia (IDA) arising from a variety of causes including insufficient intake of iron-rich foods. Globally, about 53% of school age children suffer from IDA, of which, the prevalence is 58.4% in Asia and 49.8% in

Africa (Drake *et al.*, 2002). IDA, as well as VAD, can have significant negative impacts on the growth and development of children (FAO/FMFH, 2006). There are several other Vitamins and minerals that are essential for child growth and development, deficiency of which, also result in undesirable outcomes. It suffices to mention Zinc and Iodine.

The war against Iodine Deficiency Disorders (IDD) has to a large extent been sustained through the successful Universal Salt Iodization programme which ensures that commercially processed table salt (sodium chloride) is fortified with Iodine. These developments notwithstanding, more needs to be done to ensure sufficient intakes of Iodine in all populations (Drake *et al.*, 2002; FAO/FMFH, 2006).

On the other hand, less attention has been given to Zinc and the consequences of its deficiency yet it is an essential mineral in cell metabolism, immunity and many important aspects of growth and development (Zimmerman and Kraemer, 2007). Zinc deficiency is now well known to be widespread throughout the world with equally severe health consequences as iron or Vitamin A deficiency (Prasad, 1998). Zinc also interacts with vitamin A and Iron in many important ways (McLaren & Frigg, 2001).

Deficiencies of Vitamin A, Iron and Zinc seldom occur in isolation but rather tend to coexist. They are common in low-income countries because of reduced bioavailability of these micronutrients in foods of vegetable origin that form a large proportion of the diets of the poorest populations. These deficiencies are certainly a problem in school age children but the magnitude therein can only be inferred from data on preschool children which indicate that they are a public health problem in many developing countries (Drake *et al.*, 2002; HKI, 2006). Several studies have shown that multi-micronutrient supplementation or fortification can effectively alleviate deficiencies and improve the nutritional status of school children (Drake *et al.*, 2002; Muñoz *et al.*, 2000; van den Broek, 2003).

### **1.1.3 Vitamin A Supplementation**

The World Health Organization (WHO) defines Vitamin A supplementation as the periodic administration of high doses of the vitamin to Vitamin A deficient persons. The use of periodic large doses of vitamin A supplements is a highly cost-effective approach to preventing and treating vitamin A deficiency in young children. It has the advantage of immediately improving the vitamin A status (WHO/FAO, 2004; WHO/UNICEF, 1998). The theoretical/hypothetical relationships between the various factors involved in school-child supplementation with vitamin A are illustrated in Appendix (iii). Vitamin A supplementation in conjunction with optimal status of zinc and Iron improves both serum retinol and general nutritional status which are pivotal in achievement of long-term educational and livelihood goals.

In Uganda, the Ministry of Health has adopted a policy of vitamin A supplementation according to guidelines set by the World Health Organization (WHO) whereby, children 6 to 59 months of age (pre-school age) are supplemented twice yearly with vitamin A. This is incorporated into routine immunizations for measles and polio during Child Health Days (CHDs). The second part of this policy concerns post-partum supplementation for women whereby, they are given 200,000 IU of VA at any time within 2 months (8 weeks) after birth.

## **1.2 PROBLEM STATEMENT**

Nutrition and health is one of the priority issues identified for governments to achieve education targets (DFID, 2001). In Uganda, lower school enrolment/attendance, high drop out rates and absenteeism and poor academic performance in schools has been attributed to illness, which is also a function of malnutrition (UDHS, 2006). Because of its immense role, vitamin A is critical in the health and nutritional status of children. Therefore, its deficiency together with other micronutrients impacts heavily on the development of the school aged child.

Despite availability of vitamin A rich foods in Uganda, VAD is still widespread (UDHS,

2006). Moreover, most of the caretakers of the children affected by VAD are unaware of the problem. This means that children are highly vulnerable to both VAD and its disorders. Accordingly, WHO has developed Vitamin A repletion strategies, specifically Vitamin A supplementation for children 6 to 59 months of age and mothers post-partum but no such programme exists for school age children. There is a lack of sufficient data to show the potential impact of Vitamin A repletion strategies aimed at school-aged children.

Vitamin A supplementation, which has become common practice in many developing countries, leaves out the school age children who are actively growing and equally face a high risk of VAD due to high morbidity in this age group (Drake *et al.*, 2002). The 20% prevalence of VAD in Uganda (UDHS, 2006) and the inefficiency of vitamin A supplementation programs (UNICEF, 2001; 2006) shows that VAD is a problem of public health importance not only among the preschool but also the school age child and the general population.

### **1.3 SIGNIFICANCE/JUSTIFICATION**

It has been noted that malnourished children can exhibit catch-up growth if their environment improves, suggesting that interventions targeted at school-age children can supplement efforts in the preschool years to reduce levels of stunting and related effects on children's health and education (Drake *et al.*, 2002). Despite Uganda's favourable climate that potentially ensures food security for her population, both food and nutritional security still remain elusive especially among certain vulnerable groups. Undernutrition in the form of stunting, underweight and wasting is still widely prevalent (UDHS, 2006). This undernutrition coexists with a burden of disease, which is equally immense. Micronutrient malnutrition/deficiency or hidden hunger, which receives little attention in the face of overt hunger, is probably a major factor in the morbidity of preventable illnesses, physical and mental deformities, and loss in school days among children.

Vitamin A is one of the key micronutrients in human health. Its intake is crucial in determining not only the vitamin A but also the general nutritional status of children.

Effective control of VAD could make a large contribution towards the attainment of the Millenium Development Goal (MDG) for the reduction of child mortality by two thirds between 1990 and 2015 (Aguayo *et al.*, 2005) and other specific MDGs (Neidecker-Gonzales *et al.*, 2007).

Data are insufficient to show the effect of vitamin A supplementation on nutritional and vitamin A status among school-age children. Therefore, this study is important to assess the effect of supplementation on nutritional status among the school age children so as to build a basis for wider interventions. A deeper understanding of this relationship will strengthen efforts to improve the health of school age children and consequently, school attendance, enrolment and performance while at the same time reducing dropouts.

This research will help to:

- i. Document the effect of supplementation with vitamin A on the nutritional and vitamin A status of school-age children.
- ii. Provide useful data for further research in the health and nutrition of school age children in rural or peri-urban settings.

## **1.4 OBJECTIVES**

### **1.4.1 Overall Objective**

To assess the effect of supplementation with Vitamin A plus other micronutrients on serum retinol and the general nutritional status of primary school children (6–10 years) in a peri-urban setting in Uganda.

### **1.4.2 Specific Objectives**

- a. To assess the nutritional status of 6 to 10 year-old primary school children in Wakiso district, Central Uganda.
- b. To assess the Vitamin A status of the children

- c. To determine the effect of supplementation with vitamin A plus other micronutrients on the nutritional status of the school children.
- d. To determine the effect of supplementation with vitamin A plus other micronutrients on the Vitamin A status of the school children.
- e. To compare the effects of Vitamin A-mineral supplementation regimens on the nutritional and Vitamin A status of the children.

## **1.4 RESEARCH QUESTIONS**

- i. Can supplementation with Vitamin A, Iron and Zinc improve the nutritional status of peri-urban primary school children?
- ii. Can supplementation with Vitamin A, Iron and Zinc improve the Vitamin A status of peri-urban primary School children?
- iii. Is there any comparative advantage in supplementation with all the three micronutrients together compared to supplementation with Vitamin A and Iron alone without Zinc?

## **1.5 SCOPE OF THE STUDY**

The study was carried out in Wakiso district (Central Uganda) among school children aged 6 to 10 years from three non-boarding primary schools in Nabweru sub-county.



## CHAPTER TWO

### LITERATURE REVIEW

#### 2.1 NUTRITIONAL STATUS OF PRE-SCHOOL CHILDREN

Malnutrition is a major public health problem in many developing countries. Fawzi *et al.* (1997) reported that about 43% of children (or 230 million) under the age of five years in developing countries were stunted, while about 9% (or 50 million) were wasted. UNICEF's state of the World's children report (2008) shows a slight improvement of these indicators. It showed that worldwide, 25% of children under five years of age are severely or moderately malnourished but is even higher (28%) in Sub-Saharan Africa. For Uganda, underweight among children below five years is estimated at 16% (UDHS, 2006).

Stunting as well is still a big problem especially in developing and least developed countries (Table 2.1). A prevalence of 38% has been reported both in Sub-Saharan Africa (UNICEF, 2008) and Uganda (UDHS, 2006). de Onis *et al.* (2000) reported that the highest levels of stunting in Africa were prevalent in Eastern Africa and seemed to be increasing such that by 2005 there would be about 24.4 million children (53.2%) stunted. Strong evidence exists that poor growth is associated with delayed mental development. Growth retardation in early childhood is also associated with significant functional impairment in adult life (de Onis *et al.*, 2000).

**Table 2.1 Global Childhood Malnutrition**

Region	Uganda <sup>1</sup>	SSA	LDCs	Developing countries	World
Proportion underweight (%)	15.9(4.1)	28(8)	35(10)	26(10)	26
Proportion stunted (%)	38.1(15)	38	42	32	31
Proportion wasted (%)	6.1(2)	9	10	11	11

Values in parenthesis represent respective prevalence of severe malnutrition; <sup>1</sup>According to UDHS (2006); LDCs = Least Developed Countries, SSA = Sub-Saharan Africa. Source: UNICEF (2008)

Child nutritional status has been evaluated and recommended for use as a poverty indicator. While income is not the sum total of people's lives, health status as reflected by level of malnutrition is. Malnutrition is recognized as both cause and consequence of poverty. Investments in meeting the nutritional requirements and improving the nutritional status of children are fundamental to the achievement of sustainable development. High proportions of infants, children, adolescents, adults and elderly in the developing world suffer from one or more of the multiple forms of malnutrition. Poor preschool child nutritional status is known to have important long-term effects on the work capacity, intellectual performance and life-time earnings of adults (Setboonsarng *et al.*, 2005; Alderman *et al.*, 2003).

## **2.2 NUTRITIONAL STATUS OF SCHOOL AGE CHILDREN**

School-age children face health and nutritional problems that may affect their individual physical development, their capacity to attend school and their ability to learn and to take advantage of formal education. In 2000, the SCN working group emphasized the need to promote more research and operational work on the nutrition of school-age children (Drake *et al.*, 2002). World Food Programme (WFP) noted that school children are crucial to attainment of all MDGs and that a multisectoral approach of the School Feeding and Health Programme supports overall growth of children through: increased school attendance, school retention, improved school performance; health service support through health check ups and Deworming and enhanced Referral systems; Agriculture; Environment and AIDS prevention training (WFP, 2004).

Table 2.1 shows that malnutrition in the form of undernutrition is a big problem in the developing and the least developed world. The developed world, on the other hand is grappling with problems associated with overnutrition (Dehghan *et al.*, 2005). At least 155 million school-age children are overweight or obese (Sight & Life, 2007).

The Recommended Daily allowances (RDAs) for school age children (Table 2.2) have been set to meet the needs of all (97 to 98%) individuals in this age group.

**Table 2.2 Dietary Reference Values (DRVs) for boys and girls 4-18 years for selected micronutrients and energy**

Age (yrs)	4-6		7-10		11-14		15-18	
Gender	<i>Boys</i>	<i>Girls</i>	<i>Boys</i>	<i>Girls</i>	<i>Boys</i>	<i>Girls</i>	<i>Boys</i>	<i>Girls</i>
<b>Vitamin A (µg/day)</b>	<b>500</b>	<b>500</b>	<b>500</b>	<b>500</b>	600	600	700	600
<b>Iron (mg/day)</b>	6.1	6.1	8.7	8.7	11.3	14.8	11.3	14.8
<b>Zinc (mg/day)</b>	6.5	6.5	7	7	9	9	9.5	7
<b>Energy (kcal/day)</b>	1715	1545	1970	1940	2200	1845	2755	2110

(Source: Adapted from Holden and Macdonald, 2000)

## 2.3 SCHOOL AGE CHILD EDUCATION

### 2.3.1 School Enrollment and Attendance

School enrollment and attendance are measured by Net Attendance Ratio (NAR). The difference between the Net Attendance Ratio (NAR) for boys and that for girls in Uganda (Table 2.3) is insignificant. However, the low percentage of school entrants reaching grade 5 and the much lower NAR for secondary school both indicate a high level of dropouts. A more recent survey in Uganda (UDHS, 2006) reveals an improvement of NAR for primary school to 82%. It is also observed that despite the existence of Universal Primary Education (UPE) programme, about 30% of girls and boys have never attended school for various reasons. Another observation is that NAR is lower for rural than urban areas.

The rate of absenteeism among primary school pupils in Uganda is as high as 13%. Interestingly, Karamoja region of Uganda shows lower rates of absenteeism than all other regions except Kampala. This has been partly attributed to the free lunch offered to pupils in Karamoja as an incentive to increase school attendance. On the other hand, illness alone contributes over 30% of absenteeism in the country. UDHS (2006) further shows that 40% of boys and 45% of girls who live to reach primary school age will not attend school and over a third of those who enter primary school will not reach grade 5. Around 80% of all children of

secondary school age will not attend secondary school, further implying a high rate of dropouts and low achievement of educational goals and objectives.

**Table 2.3 Education and Gender parity across the world**

<b>Region</b>	<b>Uganda</b>	<b>SSA</b>	<b>LDCs</b>	<b>Developing countries</b>	<b>World</b>
NAR, Primary School, boys	82	64	65	80	80
NAR, Primary School, girls	81	60	63	78	78
NAR, Secondary School, boys	16	25	26	50	50
NAR, Secondary School, girls	16	22	24	46	47
% Primary School Entrants reaching Grade 5	48	70	67	77	78

Source: UNICEF (2008); NAR (Net Attendance Ratio) = Proportion of school age children actually attending school

### **2.3.2 Universal Primary Education**

A healthy and well-educated population is both a necessary condition for development and one of the central objectives for development. The Constitution of Uganda (Articles 30 and 34(2)) provide for the right to basic education for every Ugandan. A commitment to education has been identified as a development priority and universalizing Primary Education has been Government's chief education priority since 1997 and is central to the Education Sector Investment Plan (MFPED, 2004).

In the third Millennium Development Goal, all the 191 states pledge a commitment to "Achieve Universal Primary Education (UPE) by 2015." Since UPE was introduced in Uganda in 1996, enrolments have been increasing steadily at all levels of primary education (MFPED, 2004) and this would put Uganda on the road to achieving the MDG. But the problems of dropouts and absenteeism delay the achievement of the goal and undermine the quality of education achieved. Inadequate health including nutrition and sanitation facilities amongst other social and infrastructural factors, have been implicated in the perpetuation of dropouts and lower attendance.

As a result, the Government of Uganda has prioritized a number of key strategies, including a home based School Feeding programme aimed at enhancing retention of children in school while improving their health and nutritional status. In fact, one of the policy objectives of UPE is “Providing the minimum necessary facilities and resources to enable children enter and remain in school and complete the primary cycle of education” (Syngellakis & Arudo, 2006). In 2006, the Government introduced Universal Secondary Education (USE) so as to promote post-primary Education. Nevertheless, to ensure the success of UPE, the challenges of ill health/malnutrition and other factors that impair primary education remain to be addressed.

### 2.3.3 Child Survival and Education

Two MDGs indicators—U5MR (Table 2.4) and completion of Primary School Education are key indicators. Although under-fives in Least Developed Countries (LDCs) make up only 19% of the world’s U5s, they account for more than 40% of all U5 deaths. About 49% of the 10 million deaths among children less than 5 years old each year in the developing world are associated with malnutrition. In Uganda, the Infant and Under 5 Mortality Rates (IMR and U5MR) both declined by 16% and 13% respectively between 2001 and 2006 (UDHS, 2006). A significant and sustained improvement in the survival of children under five years of age will help promote early and increased school enrolment.

**Table 2.4 Child Survival in the world**

Mortality category	Region				
	Uganda	SSA	LDCs	Developing countries	World
U5MR (per 1000 live births), 2004	134	160	142	89	72
IMR (per 1000 live births) 2004	78	95	90	54	49

Source: UNICEF (2008)

## 2.4 VITAMIN A

Vitamin A is an alcohol, occurring in nature predominantly in the form of fatty acid esters (de Man, 1999). It is a fat-soluble vitamin, which the human body cannot make and is stored principally in the liver (Haselow *et al.*, 2004). Synthetic vitamin A is made as acetate or palmitate and marketed commercially in the form of oil solutions, stabilized powders, or aqueous emulsions.

Vitamin A levels are frequently expressed in International Units (IU), although this unit is no longer officially accepted. One IU is equal to 0.344ug of crystalline vitamin A acetate, or 0.300ug vitamin A alcohol, or 0.600ug B-carotene (de Man, 1999). The conversion of B-carotene to vitamin A has however, been revised upwards (Sommer & Davidson, 2002) as shown in the next section (2.3.1).

Vitamin A is unstable in the presence of light, oxygen, lipid peroxides or conditions that favour lipid oxidation. These conditions can cause breakdown of vitamin A by oxidation (de Man, 1999).

### 2.4.1 Carotenoids, Provitamins A and Vitamin A activity

Preformed vitamin A (retinol) does not exist in plants and fungi. Rather, their vitamin A activity is associated with some carotenoids. Among the over 600 carotenoids known,  $\beta$ -Carotene exhibits the highest vitamin A activity and is present in virtually all carotenogenic foods in varying quantities (Rodriguez-Amaya, 1997; Rodriguez-Amaya and Kimura, (2004). Other carotenoids with vitamin A activity are  $\alpha$ -carotene and  $\beta$ -cryptoxanthine.

Considering the structure of  $\beta$ -carotene (Fig.2.1), two molecules of retinol (vitamin A) are potentially obtainable from one molecule. But, the conversion of provitamins A in carotenoids to vitamin A is so inefficient that  $\beta$ -carotene exhibits only about 50% of the

vitamin A activity exhibited by retinol on a mass basis. Among carotenoids, however,  $\beta$ -carotene is assigned 100% provitamin A activity.  $\alpha$ -Carotene and  $\beta$ -Cryptoxanthine exhibit only about 50% of the vitamin A activity of  $\beta$ -carotene.

According to FAO/WHO (2004) a bioconversion factor of 1:6 for Retinol Equivalents (RE) to  $\beta$ -carotene should be retained until studies confirm otherwise. Studies from developing countries, however, suggest that it takes 21  $\mu\text{g}$  of  $\beta$ -carotene from a typical mixed plant diet of vegetables and fruits and 27  $\mu\text{g}$  of  $\beta$ -carotene from green leafy vegetables to yield 1  $\mu\text{g}$  of RE (Sommer & Davidson, 2002). The Food and Nutrition Board (FNB), Institute of Medicine (IOM), USA, however, revised the ratio to 1:12 for  $\beta$ -carotene and 1:24 for other carotenoids (Micronutrient Forum, 2007).

Worldwide, about 60% of dietary vitamin A is estimated to come from provitamins A, this rises to 82% in developing countries. The all-trans-isomers of carotenoids exhibit greater Vitamin A activity than the cis-isomers, although many plants contain a mixture of both all-trans and cis-isomers of retinoids and carotenoids. Thermal processing of carotenoids can cause a trans-to-cis-isomer conversion leading to loss of vitamin A activity (Fennema, 1996). Apart from isomerization, loss of vitamin A activity in carotenogenic foods to a greater extent occurs due to oxidation. Both isomerization and oxidation can also occur together. Processing therefore needs to be optimized in order to minimize loss and enhance bioavailability (Rodriguez-Amaya and Kimura, 2004).

#### **2.4.2 Importance of Vitamin A**

Vitamin A is a group of compounds that play an important role in vision, bone growth, reproduction, cell division, and cell differentiation. There are two categories of vitamin A: preformed vitamin A from animal sources and pro-vitamin A (carotenoids) from plant sources. In humans, either form of vitamin A when ingested becomes available as retinol (active form). Retinol can be converted into retinal and by oxidation and further still, to retinoic acid (WHO/FAO, 2004).

Vitamin A has become known as an anti-infective vitamin because of its role in the immune system. Retinol and its metabolites are required for maintaining the integrity and functioning of the skin and mucosal cells (cells lining the surfaces of the respiratory, urinary, and intestinal tracts) which form the body's primary line of defense. Vitamin A also plays a central role in the making and activation of lymphocytes. Besides its role in the immune system, vitamin A appears to be important for the regulation of gene expression (especially for growth hormone) and making of red blood cells (Higdon, 2003). Generally, vitamin A is key in the development and function of virtually all cells, not just those of epithelium and endothelium (Jason *et al.*, 2002)

Vitamin A has been used in the prevention of some types of cancer and treatment of some skin diseases (Higdon, 2003), measles and xerophthalmia (WHO/UNICEF, 1998). Vitamin A interacts significantly with other micronutrients especially Zinc and Iron. Much as Zinc is important for Vitamin A metabolism, the synergistic interrelationships between vitamin A and both Iron and Zinc are of public health importance.

Aside from the provitamin A activity, carotenoids have many other health promoting functions attributed to their antioxidant property through deactivation of free radicals and quenching of singlet oxygen. The developed world is now focusing on these benefits other than vitamin A activity. Meanwhile, because VAD remains a serious public health problem in developing countries and animal products are largely unaffordable, dietary sources of provitamins A and their adequacy continue to be the major concern (Rodriquez-Amaya, 1997; FAO/WHO, 2004).

### **2.4.3 Structure and Function of Vitamin A**

Preformed vitamin A is referred to as retinol (an alcohol). This can be reversibly oxidized to form retinal (retinaldehyde) which in turn can be irreversibly oxidized to retinoic acid. Retinol, retinal, retinoic acid, and related compounds are known as Retinoids. Beta-carotene and other carotenoids that can be converted by the body into retinol are referred to as Provitamin A carotenoids (Higdon, 2003). Provitamin A carotenoids are hydrolyzed to form

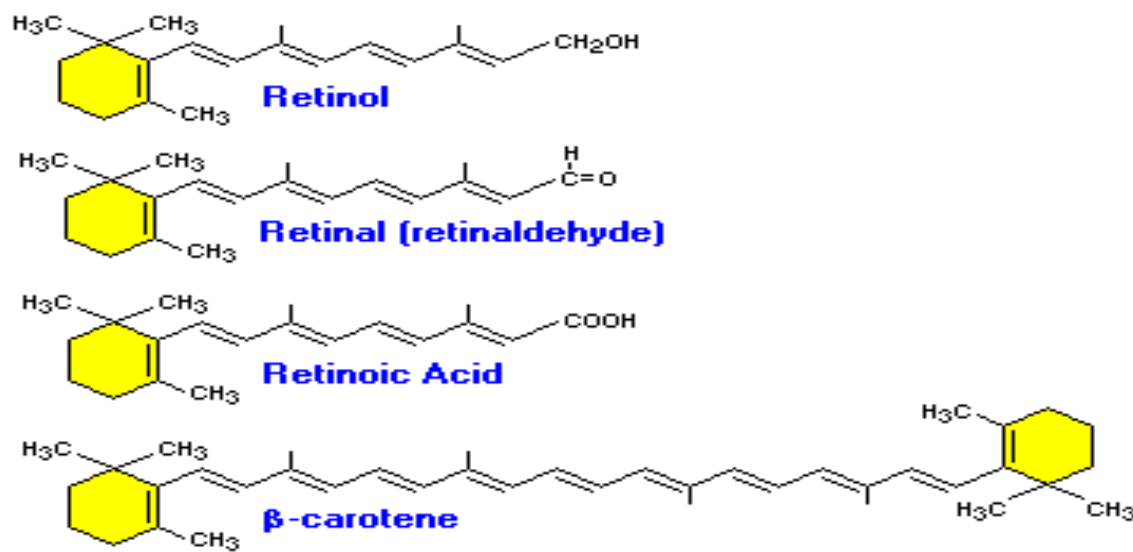


retinol. Many different geometric isomers of retinoids are possible as a result of either a *trans* or *cis* configuration of four of the five double bonds found in the polyene chain. Although some *cis* isomers occur naturally and carry out essential functions for example, the 11-*cis*-retinal isomer in vertebrate photoreception, they are less stable and can readily convert to the *all-trans* configuration.

**Table 2.5 Structural properties of Retinol**

Chemical formula	$C_{20}H_{30}O$
Molar mass	286.456 g/mol

Adapted from: Schnepf (2002)



**Figure 2.1 Chemical structures of Retinol, Retinal, Retinoic acid and B-carotene**

Adapted from: [http://www.vivo.colostate.edu/hbooks/pathphys/misc\\_topics/vitamina.html](http://www.vivo.colostate.edu/hbooks/pathphys/misc_topics/vitamina.html)

## 2.4.4 Sources of Vitamin A

### 2.4.4.1 Preformed Vitamin A/Retinol

Foods of animal origin provide mainly preformed vitamin A or retinol. Animal sources include egg yolk, fish (WHO/FAO, 2004), mammalian liver, whole milk, and some fortified food products (Olson *et al.*, 2006), milk products and meat (Rodriguez-Amaya, 1997).

Highest levels of vitamin A are found in certain fish liver oils, such as cod and tuna. Retinoids in plants or animals can occur in the all-trans or all-cis isomeric forms but the all-trans form is the most predominant and active biologically (de Man, 1999).

#### ***2.4.4.2 Sources of Provitamins A***

Good sources of provitamin A in vegetable products are green leafy vegetables, yellow vegetables (e.g. carrots, pumpkins, squash), sweet potatoes (orange fleshed), tomatoes and broccoli. One of the highest known concentrations of carotenoids occurs in crude or red palm oil, containing about 15 to 300 times more Retinol Equivalent (RE) than carrots, green leafy vegetables and tomatoes (de Man, 1999; WHO, 2004).

### **2.4.5 Metabolism of Vitamin A**

#### ***2.4.5.1 Occurrence in nature***

Vitamin A exists in the diet primarily as long-chain fatty acid retinyl esters along with small amounts of retinol and retinoic acid. Provitamins A in foods of vegetable origin are also associated with cellular lipids embedded in chloroplasts and chromoplasts. Normal digestive processes free vitamin A and carotenoids from food matrices. Retinyl esters are hydrolyzed in the intestinal lumen by pancreatic and intestinal brush border enzymes. The retinol and freed carotenoids are incorporated into micellar solutions (chylomicrons). Retinol and some carotenoids enter the intestinal mucosal brush border by diffusion in accord with the concentration gradient between the micelle and plasma membrane of enterocytes (WHO/FAO, 2004).

Some of the provitamin A carotenoids in the enterocytes are converted to retinol by a cleavage enzyme. Retinol is trapped intracellularly by re-esterification and binding to intracellular proteins while retinyl esters and unconverted carotenoids enter blood through the thoracic duct. If not immediately needed, retinol is re-esterified and retained in the fat-storing cells of the liver (hepatocytes) and carotenoids are deposited elsewhere in fatty tissues throughout the body (WHO/FAO, 2004).

Following hydrolysis of stored retinyl esters, retinol combines with a plasma-specific transport protein, Retinol Binding Protein (RBP), mainly within the liver cells. The RBP-retinol complex (holo-RBP) is secreted into the blood where it forms a complex with transthyretin and circulates in the blood delivering the lipophilic retinol to tissues. Holo-RBP is filtered into the glomerulus but is recovered at the kidney tubule and recycled (WHO/FAO, 2004).

Normally, vitamin A leaves the body in urine as inactive metabolites and in bile secretions as potentially recyclable glucuronide conjugates of retinol. Urine, however, is not yet a useful biological fluid for assessment of vitamin A nutriture (WHO/FAO, 2004).

#### ***2.4.5.2 Bioavailability***

Retinoids are absorbed effectively except under conditions in which malabsorption of fat occurs. Retinyl acetate and palmitate are as effectively utilized as non-esterified retinol (Fennema, 1996). On the other hand, Carotenoids in many foods undergo markedly less intestinal absorption than retinol. Other factors which influence absorption and utilization of provitamin A include amount; type and physical form of the carotenoids in the diet; intake of fat, vitamin E, and fibre; protein and zinc status; existence of certain diseases and parasitic infection.

## **2.5 UNDERNUTRITION AND CHILDREN'S EDUCATION**

### **2.5.1 Effects of Undernutrition on children's Education**

The studies of the effect of under-nutrition on cognitive ability, though not entirely conclusive, indicate that chronic under-nutrition is associated with lower achievement levels among school children. Recent studies have found that: severe stunting in the first two years of life lowers test scores in School age (8-11 years); chronic malnutrition lowers language and mathematics test scores (Vietnam) and that short stature may lead to late enrolment for Primary school children (Drake *et al.*, 2002; World Bank, 2006).

Both hunger and malnutrition impact negatively on the education of school age children. Incidences of absenteeism such as in Uganda have been blamed partly on inadequate or sheer lack of nutritious food. Children especially in rural areas of developing countries would rather stay home than go to school and endure hunger (Bundy *et al.*, 2009). The consequences of deficient or inadequate diets manifesting as illness or some forms of physical incapacity often have negative implications on educational progress.

The cyclical relationship between hunger and malnutrition on one side and child education on the other (FAO, 2005; World Bank, 2006) is interesting. Nutrient deficiencies contribute to a vicious cycle of malnutrition, underdevelopment and poverty affecting already underprivileged populations since nutritionally deficient adults are less productive, less innovative and more susceptible to diseases. While education is viewed as a tool to improve livelihoods and fight poverty, disease, hunger and malnutrition; in the face of the latter, education itself is hampered (Campose-Browers & Wittenmyer, 2007). Thus, it is beneficial to strategically allocate resources to the fight against hunger and malnutrition while investing in education so as to break the vicious cycle of illiteracy, poverty, disease, malnutrition and hunger as spelled out in the first MDG: “*Eradicate extreme poverty and hunger.*”

## **2.5.2 The role of School feeding and Gardening in Child Education**

### ***2.5.2.1 School Feeding***

Offering nutritious meals to children in African schools can make a difference to their health and well-being, and can also provide an incentive to families to send them to school. School feeding programs in Ethiopia, Mali, Mozambique, Senegal and Tanzania helped to ensure that over one million children had access to nutritious meals and a primary education (CDC, 1996; CIDA, 2007).

School feeding could play a central role in the New Partnership for African Development (NEPAD) by providing a direct link to agricultural development through enhancing local production, school gardens and incorporation of agriculture into school curricula (WFP,

2004). School feeding is also seen as a vehicle for fighting disease and promoting local agriculture and eliminating abject poverty. Hence, the need to expand school feeding programs (IFPRI, 2004). Some experts believe school-feeding programs could be key to achieving the Millennium Development Goals (MDGs) of reducing hunger by half and achieving Universal Primary Education (UPE) by 2015.

Studies in Bangladesh have attested to the benefits of school feeding, including the potential to address community health problems. The impact of school feeding can be improved by offering, in addition, food for schooling and a package of health and nutrition interventions including, among others, micronutrient supplementation and nutrition education to school children (CDC, 1996; IFPRI, 2004).

Gardening in the context of school feeding programmes enables pupils to develop valuable agricultural knowledge and skills by learning how to grow micronutrient-rich vegetables and fruits. Children have the chance to test fresh foods and supplement rations handed out as part of the School Feeding Programme (IFPRI, 2004).

Even as many governments in developing countries are making massive efforts to improve basic education, a core component for building development capacity, serious considerations must be made about the impact of the health and nutrition on the school-aged population and the role diet plays in the cognitive development and school performance of children in these countries. Studies in countries such as Sri Lanka, Zimbabwe, Thailand have found levels of malnutrition among school children as high as over thirty percent and yet some of these countries are large producers and exporters of food. There is agreement from these studies also, that children who would have to walk long distances to school would not do so on empty stomachs. These scenarios present net losses in educational attainment and underline the need for school feeding programmes and practical focus on the nutrition of the school child.

#### ***2.5.2.2 School Gardens***

Across the international educational landscape, Garden-based learning programs have gained popularity. Proponents of school garden programs outline multiple developmental benefits

that school gardens can have on children—namely, emotional, aesthetic, and even spiritual in addition to social and intellectual benefits, in a variety of contexts. Research has identified many positive effects to pupils of working in the garden. This include moral development, enhanced daily academic curriculum, pleasure from watching the flourishing garden products and chance to increase interactions with parents and other adults. In addition, the children learn the value of living things and acquire a sense of responsibility.

Use of gardens as a teaching method is associated with a number of merits. Children retain more skills and knowledge in such practical environments than in a conventional classroom; they are motivated by the connection to the earth; concepts can be made more meaningful in a natural laboratory; children are introduced to teamwork. But, one of the core purposes of a school garden is to improve nutrition, diet and health.

School gardens serve as an ideal context for nutritional programs. Children who plant and harvest their own vegetables are more willing to taste and like them. The gardens have been used to teach children about nutrition and how to make healthier food choices including healthy eating habits and the importance of fruits and vegetables. Gardening activities can also have far-reaching benefits such as bridging schools and communities, promoting intergenerational knowledge (including nutrition knowledge) transfer and environmental awareness; providing opportunities for cultural exchange and building life skills.

## **2.6 MICRONUTRIENT DEFICIENCIES**

Nutritional anaemia, particularly Iron Deficiency Anaemia (IDA), and deficiencies of iodine and vitamin A are major problems for School Age Children (SAC) in low-income countries. It has been shown that such deficiencies can negatively impact on growth, increase susceptibility to infection and also impair the mental development and learning ability of school children.

### **2.6.1 Vitamin A Deficiency (VAD)**

WHO defines VAD as tissue concentrations of vitamin A low enough to have adverse health consequences even if there is no clinical evidence of Xerophthalmia (WHO/FAO, 2004). Sommer & Davidson (2002) define VAD as liver stores below 20 µg and VAD disorders as any health or physiologic consequences attributable to VAD whether clinically evident or not. According to Haselow *et al.* (2004), VAD is a systemic disease that affects cells and systems throughout the body. It is considered a public health problem if 15% or more of children sampled have serum retinol levels of < 20 µg/dl (0.7 µmol/l).

Symptoms of VAD may include, but are not limited to, loss of appetite, frequent infections (especially respiratory), hair loss, rashes, dry skin and eyes, visual difficulties including night blindness, poor growth and development, and fatigue (Higdon, 2003,). These symptoms are, however, non-specific (WHO/FAO, 2004).

#### ***2.6.1.1 Prevalence of Vitamin A Deficiency***

There is now extensive evidence that vitamin A deficiency (VAD) is widespread in young children in many developing countries. WHO/FAO (2004) estimates that globally, over 250 million children under five years of age are affected by VAD. According to HKI (2006), 127 million preschool children and 7 million pregnant mothers are Vitamin A deficient and a total of 800,000 children die every year due to lack of vitamin A. The prevalence of both clinical and sub-clinical VAD is highest in Africa and South-East Asia (WHO/FAO, 2004; UNICEF, 1998; WHO/UNICEF, 1998). In Sub-Saharan Africa, it is estimated that 42% (43.2 million) children under five years of age are at risk of VAD (Haselow *et al.*, 2004). Table 2.6 shows the prevalence of VAD across the world.

**Table 2.6 World prevalence of VAD in U5s**

Region	Serum retinol <0.7µmol/l		Xerophthalmia	
	Percent	No. (millions)	Percent	No. (millions)
Africa	32	33.4	1.5	1.6
Eastern Mediterranean	21	12.7	0.9	0.5
South/South East Asia	23	55.8	1.2	2.2
Western Pacific	14	17.1	0.2	0.2
Americas	17	8.2	0.2	0.8
Total	25	127.3	0.9	4.4

Source: Micronutrient Forum (2007)

Serum retinol and xerophthalmia are indicators of VAD. Serum retinol < 0.35 µmol/l indicate clinical deficiency while levels from 0.35 to 0.7µmol/l indicate sub-clinical deficiency. But, sub-clinical deficiency may be present at levels between 0.7 and 1.05µmol/l and occasionally above 1.05µmol/l. However; only levels below 0.35 are associated with xerophthalmia (WHO, 2004). Worldwide, it is estimated that over three million children under five are affected by clinical VAD (HKI, 2006). Nonetheless, Africa has the highest prevalence of clinical VAD (MF, 2007).

The Uganda Demographic and Health Survey conducted by Uganda Bureau of Statistics (UBOS) in 2001/02 revealed VAD prevalence as 28% among children 6-59 months and 52% among women 15-49 years (Mason *et al.*, 2001). A latter survey (UDHS, 2006) showed the prevalence among children to be 20% and among women 15-49 years of age to be 19%. There, therefore appears to have been an improvement in the vitamin A status but the veracity of this is not clear because different methods were employed in assessing vitamin A status in the two surveys and caution is advised in comparing the prevalence rates (UDHS, 2006).

### ***2.6.1.2 Vitamin A Deficiency among School Age Children***

Vitamin A Deficiency can occur in individuals of any age, not excepting the school-age children. It is estimated that 85m SAC are at increased risk of acute respiratory and other infections because they are deficient in vitamin A. Although little is known about VAD in SAC (Singh and West, 2004), recent studies conducted in Bangladesh, South Africa, Mexico,



Tanzania, Vietnam, Ghana, Indonesia and Northern Ethiopia, suggest that VAD is a public health problem in SAC (Drake *et al.*, 2002). According to WHO/FAO, (2004), the prevalence of Bitot's spots may be highest among school-age children. Moreover, VAD may have a greater impact on mortality of older than of younger children (Sommer, 1995).

### **2.6.1.3 Causes of Vitamin A Deficiency**

VAD occurs when insufficient vitamin A is consumed in the diet, too little is absorbed from the food eaten, or too much is lost due to illness (Haselow *et al.*, 2004; Allen and Gillespie, 2001). In the developing world, multiple micronutrient deficiencies mostly iron, vitamin A, and iodine are common because the cereal-based diets commonly consumed predispose people to insufficient absorption of both iron and zinc, and low intakes of several vitamins including VA because of their high phytate content (Smuts, 2005). Dietary inadequacy is thought to be the major cause for vitamin A and iron deficiency (van den Broek, 2003).

During infections, both demand and loss of vitamin A increase (UNICEF, 2006; Stephensen, 2001). Kikafunda *et al.* (1998) found that despite the wide immunization coverage in Uganda among children below 69 months of age, the prevalence of diarrhea and other infections was still high. VAD can result from rapid utilization of vitamin A during illness (particularly measles, diarrhea and fevers); pregnancy and lactation; and during phases of rapid growth in children (Haselow *et al.*, 2004). Several authors also agree that HIV/AIDS increases demand for vitamin A and may cause VAD in people infected with HIV/AIDS (Semba *et al.*, 1994; McLaren & Frigg, 2001; Oguntibeju *et al.*, 2004).

Malnutrition, including Protein–Energy Malnutrition is one of the risk factors for VAD. Other risk factors include alcoholism, type 1 diabetes mellitus (Higdon, 2003), worm infections (UNICEF/WHO, 2004), malaria, being under five years of age, fat malabsorption, liver cirrhosis and a variety of socioeconomic factors (Yamamura *et al.*, 2004). A cyclic relationship between infections and VAD has been suggested, whereby, each seems to increase the risk of the other. Studies done in Kenya (Nabakwe *et al.*, 2005) showed a spiral relationship between malaria and VAD. Whereas VAD can weaken resistance to malaria,

malaria can lead to VAD by impeding VA transport from liver. Generally though, the relationships between morbidity and VAD are complex (McLaren & Frigg, 2001). Berger *et al.* (2007) showed a similar kind of relationship between VAD and diarrhoeal diseases but further revealed that children missed by periodical vitamin A supplementation were more likely to suffer diarrhoeal disease than those who received the supplements.

#### ***2.6.1.4 Effects of Vitamin A Deficiency***

Mild or clinical VAD causes impaired immune function, increased severity of some infections, increased risk of mortality and/or slowed recovery from infectious diseases (especially measles and diarrhea), blindness in children, and contributes to decreased physical growth (Drake *et al.*, 2002; FAO/EMPH, 2006; Olson *et al.*, 2006; Smuts, 2005; Jason *et al.*, 2002). VAD has been identified as the leading cause of preventable childhood blindness (UNICEF, 1998), and has been considered as a nutritionally acquired immune deficiency (Higdon, 2003). VAD also affects iron metabolism and may exacerbate iron deficiency anemia (IDA). Survivors of VAD are left crippled, chronically vulnerable to illness and intellectually disabled (UNICEF, 1998).

#### ***2.6.1.5 Assessment of VAD***

##### **i) Biochemical Assessment**

Several methods are available for assessing VAD. So far the most widely used method is that of measuring serum retinol using High Performance Liquid Chromatography (HPLC) (Sommer & Davidson, 2002). This has been used extensively for assessing VAD among children under five years and women of reproductive age. Yet the method said to be the most accurate is that of assessing liver stores of vitamin A by biopsy. This method, however, is impractical.

Measurement of retinol binding protein (RBP) and other biologically active molecules are novel methods that are not yet widely used. It has been used for example in determining

prevalence of VAD among pre-school age children in Uganda in 2006 (UDHS, 2006). Other proxy methods can be used to determine liver stores. These methods are not only expensive, but also technically complicated and are rarely used (Mclaren & Frigg, 2001).

## **ii) Clinical Assessment**

Another method of assessing VAD is clinical assessment for overt signs of vitamin A deficiency. These are mainly eye (ocular) changes (Xerophthalmia) including night blindness and cases of measles and sometimes, diarrhoea. Clinical assessment is effective only for clinical VAD, which affects only a small proportion of the population globally. Sub-clinical VAD on the other hand, is the cause of widespread morbidity, mortality and disorders associated with vitamin A deficiency (WHO / FAO, 2004).

## **iii) Dietary Assessment**

This method is less a measure of vitamin A status than levels of dietary intake. It provides complementary information not available otherwise and has the advantages of being less invasive, less expensive, being able to be used for large populations and provides insights for dietary interventions (Mclaren & Frigg, 2001; WHO/FAO, 2004). Rapid and simplified Food Frequency Questionnaires (FFQs) can be used to identify groups at risk for inadequate intake of vitamin A and thus at risk for Vitamin A Deficiency (Gibson & Ferguson, 1999).

## **2.6.2 Other micronutrient Deficiencies**

### ***i) Iron Deficiency and Anaemia***

Of the 2-3.5 billion people suffering from anaemia worldwide, over 1.2 billion suffer from Iron Deficiency Anaemia (IDA). IDA is caused by: insufficient intake of iron rich foods; parasitic infections (hookworm and malaria); deficiencies of other nutrients. An estimated 53% of the 210 million SAC suffer from IDA, of which, the prevalence is 58.4% in Asia and 49.8% in Africa (Drake *et al.*, 2002). IDA in children is associated with decreased physical growth and development, impaired immune function, increased fatigue, long-term

impairment in mental and motor development and impaired reproductive functions. IDA also affects cognitive function and school achievement (Drake *et al.*, 2002; FAO/FMFH, 2006).

Several studies have been carried out to determine the impact of supplementation with iron and other micronutrients on the nutritional status of children. There have been conflicting findings on impact of daily iron supplementation on growth. While some found increases in growth and appetite, others found decreased weight gains and yet others did not find any beneficial response. However, there is general agreement that iron supplementation has significant impact on linear growth in anaemic infants, pre-SAC and SAC and prevents decreases in haemoglobin concentration of non-anaemic school age children. Studies done among Indonesian and Indian school children showed that iron supplementation improved exam performance, concentration, memory and visual/motor co-ordination (Drake *et al.*, 2002).

## ***ii) Zinc Deficiency***

Zinc is an essential mineral that is involved in numerous aspects of cellular metabolism. It is required for the catalytic activity of approximately 100 enzymes and it plays a role in immune function, protein synthesis, wound healing, DNA synthesis, and cell division. Zinc also supports normal growth and development during pregnancy, childhood, and adolescence and is required for proper sense of taste and smell. A daily intake of zinc is required to maintain a steady state because the body has no specialized zinc storage system. It is ubiquitous within cells but total body content is 1.5 to 2.5g (Zimmerman and Kraemer, 2007; Hess *et al.*, 2007).

It is now clear that the deficiency of zinc is very widespread throughout the world and may even be as prevalent as iron deficiency anemia, affecting nearly one billion people. Unfortunately, however, little has been done to correct this deficiency. The consequences of zinc deficiency are several and they impact on human health severely. Growth retardation, male hypogonadism, neuro-sensory changes (abnormal dark adaptation and changes in taste acuity), delayed wound healing, abnormal immune functions, and impaired cognitive

functions are some of the major effects of human zinc deficiency which are reversible with zinc supplementation. A mild deficiency of zinc in pregnant women is associated with increased maternal morbidity, abnormal taste sensation, prolonged gestation, inefficient labor, atonic bleeding, and increased risks to the fetus (Prasad, 1998).

Zinc deficiency places children in many low-income countries at increased risk of illness and death from infectious diseases (Black, 2003) such as diarrhea and pneumonia. Dietary Zinc deficiency is common because of reduced zinc bioavailability in foods of vegetable origin that form a large proportion of the diets of the poorest populations. Animal products such as meat, fish and offal are the richest sources of easily assimilated zinc. In addition to low intake and malabsorption, other causes of Zinc deficiency include increased requirements, increased losses, and impaired utilization depending on life stage or pathological conditions (Zimmerman and Kraemer, 2007).

### ***iii) Multiple Micronutrient Deficiencies***

Single micronutrient deficiencies seldom occur in isolation, but instead, interact and tend to cluster. For example, iron deficiency and VAD often coexist in the same population. Providing vitamin A supplementation/fortification can both improve Vitamin A status as well as Iron metabolism in deficient populations. SAC, like most populations in low-income countries suffer from multiple micronutrient deficiencies, although the extent of the problem in this age Group cannot be ascertained. Inference can, however, be made from data on pre-SAC. Thirteen to 27 percent of pre-SAC are estimated to have two or more micronutrient deficiencies suggesting that 100 million pre-school children are affected (Drake *et al.*, 2002; HKI, 2006; Graham *et al.*, 2007; Miller *et al.*, 2006).

Given the frequent overlap and clustering of micronutrient deficiencies, multiple micronutrient supplementation or fortified foods may be a cost effective strategy to address nutrient deficiencies in SAC and enable synergistic effects between certain micronutrients. Different studies in Africa and Asia have shown beneficial effects of multiple over single

micronutrient supplementation/fortification on growth and nutritional status (Drake *et al.*, 2002).

## **2.7 MICRONUTRIENT SUPPLEMENTATION**

### **2.7.1 Vitamin A Supplementation**

The administration of high-dose vitamin A supplements is one means of improving vitamin A status in children (MOST project, 2001). In Uganda, Vitamin A supplementation has been integrated into measles and polio immunizations during Child Health Days (CHDs). This strategy is tailored to benefit from popular child health days and immunization campaigns so that many at-risk children can be reached rather inexpensively. In fact, WHO recommends Vitamin A supplementation for improving vitamin A status of young children (Benn *et al.*, 2009)

In many countries in Asia and Africa, WHO/UNICEF recommend two vitamin A supplementation regimes to effectively reach at-risk groups: (a) 4-6 monthly (high dose vitamin A to children 6-59 months in affected areas. This is considered safe, cost effective and efficient, and; (b) Post-partum Vitamin A supplementation, whereby a high dose (200,000 IU) of vitamin A is administered to a mother within eight weeks after birth. This is aimed at protecting the children at early life and replenishing the mothers' vitamin A stores.

Given the frequent overlap and clustering of micronutrient deficiencies, multiple micronutrient supplementation or fortified foods may be a cost-effective strategy to address nutrient deficiencies in SAC in addition to synergistic effects between certain micronutrients (Drake *et al.*, 2002; Wieringa *et al.*, 2003). In addition, some studies have cautioned that Vitamin A alone is unlikely to have an important effect on linear growth (Bhandari *et al.*, 2001).

Earlier studies on children 6-48 months done in Indonesia had shown that vitamin A supplementation selectively improved linear growth among VA deficient children and that

the effect becomes more remarkable with increasing age (Hadi, *et al.*, 2000). Other studies however, failed to show any significant effect of vitamin A supplementation on growth. Rahman *et al.* (1997) showed that combined short-term Zinc supplementation and a single dose of vitamin A had no significant effects on weight and height increments in 12-35 month-old children over a six-month period.

UNICEF recommends that before the phase-out of supplementation, priority countries should continue supplementation at effective coverage levels in order to realize international goals for child survival and VAD control. While food fortification is a long-term strategy of reducing VAD, vitamin A/multiple micronutrient supplementation may be effective for eradicating VAD in SAC (Drake *et al.*, 2002).

Apart from improving vitamin A status of children and measles case management, Vitamin A plays several other important roles in humans. These are linked to reduction of morbidity and mortality in children. Vitamin A supplementation reduces morbidity and mortality in children living in areas endemic for vitamin A deficiency (Idindili *et al.*, 2007). Improvement in vitamin A status did appear to reduce severe morbidity, particularly in children with measles (Beaton *et al.*, 1994).

Other findings indicated that vitamin A supplementation does not reduce common morbidity in children with mild-to-moderate vitamin A deficiency in areas where access to health care and immunization are good (Ramakrishnan *et al.*, 1995). Further, studies carried out among young children 6-35 months in a peri-urban area of Mexico city found that Vitamin A supplementation increased diarrheal disease and respiratory tract infections but the effects were heterogeneous in different subgroups of children (Long *et al.*, 2006). Earlier, Beaton *et al.* (1994) in a meta-analysis of several studies, found no consistent effects of vitamin A supplementation on frequency or prevalence of diarrhoeal and respiratory infections.

### ***Supplementation Coverage***

For populations where VAD is a public health problem, UNICEF/WHO considers 70% supplementation coverage for children 6-59 months as “effective coverage” which puts such

countries on the road to attainment of international development goals (UNICEF). According to UDHS (2006), only 36% of all children 6-59 months of age in Uganda received high dose vitamin A supplements in the six months period before the survey. A somewhat similar result was reported in the earlier (UDHS, 2000/01) survey. This reveals Uganda's supplementation coverage levels to be way below the recommended levels, hence hampering progress to achieving international development goals.

Efforts were (and still are) underway to achieve and sustain high coverage in countries with high mortality rates. While on track in many countries, the realization of the vitamin A targets is slower than it could have been. Perhaps the big difference lies with advocacy around vitamin A supplementation, which has been plagued with difficulties including a belief among policy makers that the public health importance of Vitamin A supplementation is to reduce blindness (ACC/SCN, 2001).

### **2.7.2 Multiple Micronutrient Supplementation**

Multiple micronutrient deficiencies are common in children in developing countries and it is possible that more than one micronutrient may limit growth. Studies have suggested that it is not a single nutrient that limits a child's growth potential but rather a combination of several macro- and micronutrients that act as limiting factors for growth (Ramakrishnan *et al.*, 2009a; Rahman *et al.*, 2002). The coexistence of multiple micronutrient deficiencies is increasingly recognized as a widespread public health problem in developing countries (Munoz *et al.*, 2000). Therefore, the correction of a single deficiency may not be enough to improve growth substantially (Rivera *et al.*, 2001).

A meta-analysis of micronutrient supplementation interventions concluded that whereas iron and vitamin A interventions do not improve child growth, there was a suggestion of benefit for multiple micronutrient interventions (Ramakrishnan *et al.*, 2009b). Joint supplementation (of Iron and Zinc) generally does not negatively affect the biochemical outcomes expected from individual supplementation. Even in the presence of zinc, the benefit of iron supplementation on iron indicators was significant and important (Walker *et al.*, 2005).



According to Bhandari *et al.* (2001), although Vitamin A is unlikely to have an important effect on linear growth, zinc and iron seem to have a modest effect in deficient populations. However, limited available evidence did not allow conclusion on whether a combination of micronutrients, with or without additional food, would have a greater impact than that seen with zinc alone.

### ***Interaction between Vitamin A, Zinc and Iron***

Zinc influences many aspects of Vitamin A metabolism including absorption, transport and utilization. Many enzymes are zinc-dependent and among them is Retinol dehydrogenase involved in rod function. Zinc deficiency is commonly associated with low plasma concentrations of vitamin A. With zinc deficiency, there is impaired synthesis of important proteins such as retinol binding protein (RBP). This impairment affects retinol transport from the liver to the circulation and other tissues (Munoz *et al.*, 2000).

Zinc supplementation has an anti-diarrhea effect, similar to that of Vitamin A. In a different study, it was found that supplementation with both vitamin A and zinc improved lymphocyte responsiveness (McLaren & Frigg, 2001). Dijkhuizen *et al.* (2004) found that Zinc supplementation during pregnancy improved the vitamin A status of mothers and infants post-partum and suggested that addition of zinc and  $\beta$ -carotene to Iron supplements during pregnancy could be effective in improving the vitamin A status of mothers and infants.

Reports have indicated beneficial effects of zinc supplementation on vitamin A metabolism in malnourished children, preterm infants, and adults with alcoholic cirrhosis and have also shown that supplementation with Iron and/or Zinc, independent of vitamin A, improves serum retinol levels (Muñoz *et al.*, 2000). The evidence of Vitamin A-Zinc synergies in humans and their public health outcomes is, however, inconclusive and requires further studies (Christian and West, 1998).

Vitamin A and Iron appear to complement each other in reducing iron deficiency anemia (IDA) (McLaren & Frigg, 2001). Several studies have shown that vitamin A supplementation

significantly increases haemoglobin levels (Drake *et al.*, 2002; van den Broek, 2003). Vitamin A is essential for haematopoiesis, hence; vitamin A supplementation could improve haemoglobin and iron status -vitamin A is required for the mobilization and utilization of iron for haemoglobin synthesis (van den Broek, 2003). Other studies show that co-supplementation with iron and vitamin A in the treatment of anaemia reduces the incidence of co-infection which may be exacerbated when Iron alone is used. Longitudinal studies where the effect of iron supplementation on vitamin A status has been evaluated are rare (Munoz *et al.*, 2000).

### **2.7.3 Other interventions to control VAD**

#### ***i) Food Fortification***

Rivera *et al.* (2001) suggest that interventions to improve infant growth in communities with VAD problem should include improvement in micronutrient intakes. Micronutrient food fortification may be effective in growth promotion and reducing morbidity (Sarma *et al.*, 2006) and according to Dary & Mora (2002), this may replace vitamin A supplementation in mothers and children.

Many commercially produced foods such as cooking oil, wheat flour, sugar, have been fortified at different levels in different countries with vitamin A. In Uganda, there is a national food fortification programme which seeks to have staple foods in the country fortified with a range of micronutrients. The main objective is to increase micronutrient intake in the entire population. Under this programme, some commercial industries produce cooking oil fortified with vitamin A; maize flour fortified with a mix of several micronutrients and sugar fortified with vitamin A. Plans are underway to fortify wheat flour with vitamin A, iron, Zinc and the B-vitamins (MOH, 2008).

However, it has been observed that fortification may not be effective in the short term as most at-risk poor groups may not access foods fortified with vitamin A (Kapinga, *et al.*, 2000; WHO/UNICEF, 1998) and identifying the most suitable staple foods to reach the most

affected communities still remain as challenges. Nevertheless, at the first meeting of the Micronutrient Forum (MF), it was noted that there was good progress in food fortification in developing countries (Micronutrient Forum, 2007).

## ***ii) Bio-fortification***

Biofortification is the process of breeding staple crops to enhance the content of essential micronutrients such as iron, zinc and provitamins A using conventional breeding practices. The purpose of biofortifying staple crops is to improve human health and well-being by reducing the burden of disease caused by micronutrient deficiency. Biofortified crops such as orange/yellow maize, rice, orange-fleshed sweet potato could play a big role in the control of vitamin A deficiency (Campos-Browers & Wittenmyer, 2007; Jaarsveld *et al.*, 2005; Howe & Tanumihardjo, 2006).

Based on micronutrient deficiency rates, there is compelling evidence that biofortification, can be a key objective among others, for plant breeders. This has been successfully demonstrated for crops such as sweet potato. To date, orange-flesh sweet potato lines with high levels of  $\beta$ -carotene (over 200  $\mu\text{g/g}$ ) have been identified, and beans with improved agronomic traits and grain type and 50–70% more iron have been bred through conventional means (Nestel *et al.*, 2006).

Biofortification has the advantages of: reaching nearly all family members especially in low-income households and undernourished populations in relatively remote rural areas; low recurrent costs making it cost effective; becoming highly sustainable once in place; and the possibility of the process positively affecting crop survival and yields (Nestel *et al.*, 2006). A Global Challenge Program on Biofortification was approved in 2003 by the Consultative Group on International Agricultural Research (CGIAR) with the objective of enriching staple crops among resource-poor consumers such as rice, wheat, maize, beans, cassava and sweet potato with iron, zinc, vitamin A, selenium, and iodine, as needed (Graham, 2003).

Biofortification, however, is not free of challenges. Because of regulatory and political restrictions on the use of transgenic approaches, much of the resources of plant breeders like

*HarvestPlus* have been devoted to conventional breeding (Nestel *et al.*, 2006). Pray and Huang (2007) revealed that in contrast to biofortification involving genetic modification (GM) or transgenic biofortification, non-GM biofortification did not have the same problems of going through the biosafety regulations and facing political concerns of the government, stating that, “Cases of political opposition to non-GM biofortification or industrial fortification in China have been rare or non-existent.” Transgenic approaches, nonetheless, are in some cases necessary, for example, Golden Rice containing 37  $\mu\text{g/g}$  carotenoid, of which 31  $\mu\text{g/g}$  is  $\beta$ -carotene, is now available thanks to these approaches.

In addition to restrictive policies, another challenge for biofortification is to get consumer acceptance for biofortified crops, thereby increasing the intake of the target nutrients (Nestel *et al.*, 2006). Although biofortification is thought of as being a complement to industrial fortification, especially to be able to reach hilly or mountainous areas which are not well integrated into the market economy and having major Vitamin A, Vitamin B, iron, and zinc deficiencies (Pray and Huang, 2007), it is a long term investment which may not be promptly or easily accessible to communities most at risk of Vitamin A Deficiency or other deficiencies.

### ***iii) Dietary Diversification***

Promotion of vitamin A/ $\beta$ -carotene-rich staple foods especially in poor communities has been considered the most feasible and sustainable intervention to prevent VAD among all age groups (Kapinga *et al.*, 2004). Increased consumption of inexpensive vegetables and fruits is highly likely to reduce significantly the risks of VAD, including nutritional blindness in developing countries (Fawzi *et al.*, 1993). However, Sommer & Davidson (2002) have argued that increased consumption of plant  $\beta$ -carotene alone especially in developing countries is insufficient to control the problem of VAD in young children. Nevertheless, studies recommend that dietary improvement approaches need to be an integral part of a sustainable strategy to control VAD (Aguayo *et al.*, 2005; Yamamura *et al.*, 2004)

In poor countries, where consumption of foods of plant origin predominates, dietary interventions should achieve a 10-fold increase in consumption of foods of non-animal sources which are rich in  $\beta$ -carotene in order to meet Vitamin A requirements of the population and prevent vitamin A deficiency. The problem, however, is the lack of well-designed assessments to attest to the efficiency and effectiveness of dietary interventions in reducing the burden of deficiency (WHO/UNICEF, 1998).

Many of the most severe health consequences of the leading micronutrient deficiencies could be greatly alleviated by ensuring adequate food supplies and varied diets that provide essential vitamins and minerals. And yet, access to sufficient supplies of a variety of safe, good-quality foods remains a serious problem in many countries, even where food supplies are adequate at the national level. In every country, some form of hunger and malnutrition continues to exist (FAO/FMFH, 2006). There is need to promote horticultural programs to increase the availability of vitamin A foods when they cannot be obtained from animal sources (Haselow *et al.*, 2004).

It has been observed that the diet of school age children does not differ much from that of the whole family and therefore, promotion of vitamin A/ $\beta$ -carotene-rich foods among School Age Children has a synergistic effect of improving intakes in families (Drake *et al.*, 2002; Holden and Macdonald, 2000). In addition use of primary school infrastructure can make it possible to reach large numbers of people cost-effectively. This could result in better lives for millions of people around the world (IFPRI, 2004).

#### ***iv) Other broader interventions***

##### ***a) Nutrition Education***

Nutrition education programmes are needed to encourage the use of inexpensive readily available  $\beta$ -carotene-vegetable sources for home consumption as well as in feeding programmes for school children (Wadhwa *et al.*, 1994). These programmes are also needed to diversify diets and promote dietary change in order to ensure an adequate intake of vitamin A and other micronutrients (Haselow *et al.*, 2004). Improved nutrition, such as an adequate

intake of vitamin A and iodine, can bring profound benefits to entire populations (UNICEF, 1998). This also contributes to the well being of children and families, including better school attendance (MOST project, 2001).

#### ***b) Exclusive Breastfeeding***

Recent evidence has shown that, on a population basis, exclusive breastfeeding for six months is the optimal way of feeding infants. Thereafter infants should receive complementary foods with continued breastfeeding up to two years of age or beyond (WHO, 2008). Exclusive Breastfeeding confers to the child both short and long-term benefits with regard to growth, health and development. It promotes sensory and cognitive development, and protects the infant against infectious and chronic diseases. Exclusive breastfeeding of infants, including giving colostrum, to ensure adequate nutritional intake, and providing post partum mothers with a high dose of vitamin A to increase the vitamin A content of their breast milk should be promoted and protected (Haselow *et al.*, 2004).

### **2.7.4 Benefits and Goals of VAD Control/Elimination**

#### ***a) Benefits of VAD control/Elimination***

According to UNICEF (2006b), the benefits of controlling VAD include enhancing child's chances of survival, reducing severity of childhood illnesses, easing strain on health systems and hospitals and contributing to well being of children, their families and communities. In communities where VAD exists, improving vitamin A status of young children can, on average reduce the risk of: all-cause mortality by 23%; death from measles by 50% and death from diarrhea by 33% (WHO/UNICEF, 1998; Haselow *et al.*, 2004).

Studies by Semba *et al.* (1994) suggested that improved vitamin A status in HIV-positive pregnant women could reduce mother-to-child transmission of HIV, but according to McLaren & Frigg, (2001), this suggestion was not confirmed by latter studies. Regardless of HIV/AIDS status, pregnant and lactating mothers plus their infants (born and unborn) benefit immensely from good vitamin A nutriture (Haselow *et al.*, 2004). Improving vitamin A status

of vulnerable people especially in developing countries contributes to overall efforts to improve nutrition, which is critical for the attainment of not only the fourth MDG but indeed all the other MDGs (UNICEF, 2006c; WFP, 2004).

### ***b) Goals of VAD Control/Elimination***

Goals/targets are extremely important for advocacy; they spur policy makers to action. However, negative repercussions are not infrequent in the interpretation and implementation of goals, for example, in the case of short-term goals for vitamin A: overburdening of health infrastructure and causing donor fatigue; therein lies the bigger challenge. Effective planning thus requires short, medium and long-term goals (ACC/SCN, 2001). There is a widespread lack of appreciation of the extent of malnutrition and the damage it does to health, survival, human capacity and thus development. Therefore, as a means of quantifying the potential benefits of nutrition improvements in terms that policy-makers will appreciate, and as a guide for action, as well as a means of measuring success, targets/goals have an important role in nutrition policy analysis and advocacy (ACC/SCN, 2001).

To achieve the benefits of reducing VAD and prevent the effects of VAD, a number of goals have been set for control and eventual elimination of VAD and its effects:

#### ***i) Virtual elimination of VAD and its consequences including blindness by 2000***

This goal set by the World Summit for Children in 1990 was endorsed by: Policy conference on ending hidden hunger; International Conference on Nutrition (ICN) and the World Health Assembly in 1991, 1992 and 1993 respectively (WHO, 1996). The UN General Assembly Session on Children (2002) underscored the importance of VAD control to child survival and attainment of the fourth Millennium Development Goal (MDG): two thirds reduction in U5M by 2015.

In 1998, the UN standing committee on nutrition noted, “While some progress is being made, there is irrefutable evidence that VAD is widespread among children in the world.” To fast

track the progress, the committee identified reaching policy makers globally as a key strategy involving USAID, UNICEF, CIDA and MI. Benchmarks were set for countries unable to reach that goal requiring them to have: a detailed plan and budget by the end of 2000; process indicators by mid 2002 and done assessments of national VAD surveys by end of 2005.

*ii) World Fit for Children Goal: Achieve sustainable elimination of Iodine Deficiency Disorders by 2005 and VAD by 2010 (UNICEF, 2006b)*

The UN General Assembly set this goal in May 2002, in a session dedicated exclusively to children. Latter reports (UNICEF, 2008) show that while some countries are on track to achieving some of the goals, others, especially in Sub-Saharan Africa are not showing sufficient progress. The report recommends the prioritization of nutrition in national development programmes in order to realize all the other goals for children. Nevertheless, in achieving the goals, effective and proven interventions should be combined with strategies that will empower communities and strengthen health services delivery (ACC/SCN, 1998).



## **CHAPTER THREE**

### **METHODOLOGY**

#### **3.1 STUDY DESIGN**

This study, concerning vitamin A supplementation, was part of a larger study in which micronutrients: Zinc and Iron were also investigated. In this study, a randomized, double-blind, placebo-controlled design was used. This design, which tested the effect of Vitamin A supplementation, also allowed testing for potential interaction between vitamin A and mineral supplements (Iron and Zinc). The study took a total period of 16 months within which were four months of effective supplementation.

#### **3.2 STUDY AREA AND SUBJECTS**

The subjects for this study were school children aged 6-10 years. The study was carried out in three primary schools in Wakiso district in Central Uganda. The three peri-urban government day primary schools with children from areas with similar socio-economic characteristics (St. Theresa, Kazo Mixed and Kazo Church of Uganda, C/U) are found in Nabweru sub-county (6 km Northwest of Kampala. GPS: N00° 21' 08.1" x E32° 32' 30.3").

The schools were purposively selected because, apart from having similar socio-economic characteristics which were different from those of a typical urban setting, they were nearest to Kampala where analytical laboratories were available and spoilage of samples could be minimized. Secondly, these schools had large populations of children from diverse ethnic or cultural backgrounds.

#### **3.3 INCLUSION AND EXCLUSION CRITERIA**

##### **3.3.1 Inclusion criteria**

- Children falling in the range of 6-10 years

- Children not taking dietary/prescription supplements especially multivitamins and minerals
- Children with no health complications (as determined by the paediatrician on the research team).
- Children whose parents/guardians gave permission to participate in the study.

### **3.3.2 Exclusion criteria**

- Children whose parents/guardians did not give permission to participate in the study.
- Severely wasted children (these children were referred for medical attention)
- Children taking dietary/prescription supplements especially multivitamins and minerals.
- Children with chronic illnesses (as identified by the paediatrician on the team). These children were also referred for medical attention.

## **3.4 SAMPLE SIZE DETERMINATION**

The sample size for this study was determined using the following equation,  $n = Z^2pq/d^2$  (Israel, 2003) at 95% confidence interval

Where:

$n$  = sample size

$P$  = prevalence of VAD in the affected population = prevalence of VAD in under-fives in Uganda = 20% (UDHS, 2006)

$q = 1-p$  = proportion not malnourished (without VAD) = 80%

$d$  = error of assumption = 5%

$Z$  = normal distribution at 95% confidence interval (1.96)

Using the above formula, a sample size of 246 (Two hundred forty-six) children was determined. However, due to financial constraints especially in analyzing blood samples outside Uganda, a sample size of 156 children was recruited for the study. A random sampling procedure was adopted with the sampling frame composed of all the children in the

three schools. The inclusion and exclusion criteria were then applied to select 156 children to participate in the study.

### 3.5 MEASURABLE VARIABLES

#### 3.5.1 Supplementation Regimen

Using random numbers, the children were assigned to three treatment groups as shown in Table 3.1 below.

**Table 3.1 Supplementation Regimens applied to groups of children**

<b>Treatment Group</b>	<b>Description of treatment</b>	<b>N</b>
A	Vitamin A and Iron at RDAs	52
B	Vitamin A and Iron plus Zinc at RDAs	52
C	Placebo alone	52

These supplements were mixed in ready-to-drink fruit juice (*Sun sip*, Britannia Allied Industries, Uganda Limited) and given to the children at break time, on school days; Monday through Friday. The treatment period lasted four school terms (10-15 weeks each) excluding weekends, term and public holidays. On every supplementation day, each child was given 300 ml of juice. A checklist was developed by which the presence and absence of the children was detected.

#### 3.5.2 Biochemical Analysis of Serum Retinol

##### *3.5.2.1 Handling of Serum samples*

Biochemical data (Serum retinol) for all children in the three groups were collected twice: at baseline and at the end of the supplementation trial. Venous blood samples were drawn from the children into 4 ml sterile vacuuttes (EDTA K3) and freeze-dried prior to centrifuging (500g for five minutes) at the end of each day. The serum samples were kept frozen at -80°C

until analysis. The samples were then shipped to Kenya Medical Research Institute (KEMRI) in Nairobi (Kenya) in frozen state where analysis of serum retinol was carried out using High Performance Liquid Chromatography (HPLC).

#### **3.5.2.2 Retinol analysis**

Serum was denatured with ethanol containing an internal standard (retinyl acetate) and serum retinol was measured by reversed phase HPLC as described by Kaplan *et al.* (1987). The list of chemicals and apparatus used is shown in Appendix (ii).

##### *Extraction*

The 250 µL serum was denatured with equal volume of absolute ethanol containing retinyl acetate as internal standard then Vortexed for 1 minute. Extraction was carried out twice with 1.5 ml hexane each time, plus centrifugation, saving and combining the two extracts. This was followed by evaporating the combined extract with a gentle stream of white spot nitrogen gas in a water bath at 37°C. The residue was dissolved in 100 µL mobile phase and vortexed, followed by injecting 30 µL into High Performance Liquid Chromatograph.

##### *HPLC Conditions*

The C<sub>18</sub> Column: µBondaPak<sup>™</sup> 3.9 x 300 mm with 10 µm silica particles and a waters Guard column (Waters, Inc. Milford, MA, USA) was used as stationary phase. The Mobile phase: Isocratic methanol:HPLC water 95:10 v/v. was degassed by sonication before use. The Column temperature was set at room temperature, 25°C, with a flow rate: 2 ml/minute and a UV variable detector set at 325 nm. The Chart speed was 10 cm/min and Calculation (Peak area, internal standard) was done by linear regression.

#### **3.5.3 Anthropometry**

Calibrated equipment and standardized techniques according to Lohman *et al.* (1993) were employed to take a number of anthropometric measurements on the children. The

measurements were taken in triplicate with the children wearing light clothing and no shoes. Each measurement was taken by the same anthropometricist to eliminate inter-examiner error. Height was measured to the nearest 0.1 cm using a Height Measuring Board (Short Productions, Woonsocket, RI). Weight was recorded to the nearest 0.1 kg using a 136 kg digital Scale (Tanita Corporation Tokyo Japan, THD-305, China).

The age of the children, to the nearest month, was calculated from the children's birth records, where available, or determined using a local events calendar. Z-scores for height-for-age (HAZ), weight-for-age (WAZ) and weight-for-height (WHZ) were calculated using EPIINFO (Version 6.0 USD Inc., Stone Mountain, GA, USA, CDC 2002) and interpreted using WHO (2007) reference standards.

### **3.6 DATA COMPUTATION AND STATISTICAL ANALYSES**

Epi Info 2002, version 3.4 (Centres for Disease Control and Prevention, Atlanta, GA, USA) was used for calculation of anthropometric indices. All the data were computed and analyzed using SPSS (version 12). Chi-square tests were carried out to establish associations between categorical data while the Students' T-tests and correlation analyses were performed for continuous data. Analysis of variance (ANOVA) was used to determine the difference between means of different treatment groups at baseline. Statistical significance for all tests was set at  $p < 0.05$ .

### **3.7 QUALITY CONTROL**

During the study, the following steps were ensured to obtain quality data;

- Designing of “good” data-recording forms to ensure accurate and proper recording of data. The questionnaires were pre-tested to ensure validity.
- Enumerators were adequately trained and equipment was standardized to minimize inter-observer and intra-observer errors.
- Cross checking of the data-recording forms to make sure errors made were corrected in time.

- Statistical checks for errors were also made.

### **3.8 ETHICAL CONSIDERATIONS**

Ethical approval for this study was sought and obtained from the Uganda National Council of Science and Technology (UNCST). Approval to conduct the study was also granted by the Ministry of Education and Sports, the school administration, Wakiso district administration and Nabweru Sub-county office in addition to Local council leaders. The study objectives and methodologies were explained to the parents and guardians who gave their written consent.

## CHAPTER FOUR

### RESULTS

#### 4.1 SOCIO-DEMOGRAPHIC CHARACTERISTICS OF THE SAMPLED CHILDREN

Six to ten year-old schoolchildren from three selected non-boarding primary schools in a peri-urban area of Uganda were sampled for this study. A total of 156 children were recruited. These were allocated to three groups tagged to different supplementation regimens as shown in Table 3.1. Children in the first group (A) were given Vitamin A and Iron. Those in group B were given Vitamin A, Iron plus Zinc. The third group (C) which served as a control received a placebo. The size of each treatment group (n) which completed the study and for which valid results, anthropometric and biochemical were obtained is shown in Table 4.1. The dropout rates were 17% for the biochemical assessment and 24% for anthropometry.

**Table 4.1 Gender, Size and Age characteristics of the study sample**

Variable	Treatment Group		
	A (Vitamin A + Iron)	B (Vitamin A + Iron + Zinc)	C (Control)
Male (baseline)	50%	51%	37%
Female (baseline)	50%	49%	63%
Age, years (baseline)	8.06 ± 1.55	7.94 ± 0.81	7.86 ± 1.06
Size, n (Anthropometry)	42	41	36
Size, n (Biochemical)	44	50	36

The proportions of children male and female in all the groups A (Vitamin A and Iron), B (Vitamin A, Iron plus Zinc) and C (Control) are also shown in Table 4.1. There was an almost equal allocation of both male and female children to the treatment groups.

The mean age across the treatment groups ranged from  $7.86 \pm 1.06$  to  $8.06 \pm 1.55$  years as shown in Table 4.1. This age range falls within the desired range of six to ten years.

## 4.2 NUTRITIONAL STATUS OF THE SAMPLED CHILDREN

A number of indicators were used to classify the nutritional status of the school children. These include weight for age, height for age and BMI for age. Z-scores for these indices (HAZ, WAZ and BMIZ respectively) were used in the classification according to the WHO standard growth references (WHO, 2007). The z-scores were computed using Epi-info software and were used to describe the nutritional status.

### 4.2.1 Nutritional status at baseline

A summary of the mean and standard deviation (SD) of the BMIZ, HAZ and WAZ for the treatment groups A, B and the control, C at baseline is presented in Table 4.2. Overall, the mean BMIZ, HAZ and WAZ at baseline were  $-0.31 \pm 0.86$ ,  $-0.77 \pm 1.14$  and  $-0.68 \pm 0.99$  respectively. There were no significant differences between the treatment groups in the mean values for all the indicators: HAZ, WAZ and BMIZ at baseline ( $p > 0.05$ ). Having no significant differences between the treatment groups is very important because it means that any effects seen can be attributed to the treatment, in this case, supplementation.

**Table 4.2 Mean BMI for age (BMIZ), Height for age (HAZ) and Weight for age (WAZ) z-scores of the children at baseline**

Nutritional status index	Treatment Group				P value*
	Vitamin A + Iron (Group A)	Vitamin A + Iron + Zinc (Group B)	Control (Group C)	Overall	
BMIZ	$-0.26 \pm 0.99$	$-0.31 \pm 0.77$	$-0.37 \pm 0.83$	$-0.31 \pm 0.86$	0.854
HAZ	$-0.63 \pm 1.06$	$-0.62 \pm 0.99$	$-1.11 \pm 1.34$	$-0.77 \pm 1.14$	0.100
WAZ	$-0.55 \pm 1.0$	$-0.56 \pm 0.72$	$-0.97 \pm 1.17$	$-0.68 \pm 0.99$	0.111

\*p value (ANOVA) for difference in mean measure of nutritional status index between groups A, B and C



The children were classified as stunted or underweight if their height for age or weight for age z-scores respectively were below -2. At baseline, overall, the prevalence of stunting, underweight and wasting were, respectively 15.1%, 10.2% and 3.3%.

#### 4.2.2 Nutritional status after supplementation

At the end of the supplementation, the anthropometric parameters: age, height and weight were again measured as at baseline. Z-scores for height for age, Weight for age and BMI for age were calculated and are presented in Table 4.3.

**Table 4.3 Mean BMI for age (BMIZ), Height for age (HAZ) and Weight for age (WAZ) of the children after supplementation**

Nutritional status index	Treatment group			P*
	A (Vitamin A + Iron only)	B (Vitamin A + Iron + Zinc)	C (Control)	
BMIZ	-0.13 ± 1.00	-0.42 ± 0.8	-0.54 ± 0.9	0.184
HAZ	-0.63 ± 0.93	-0.45 ± 0.92	-0.87 ± 1.32	0.230
WAZ	-0.44 ± 0.83	-0.54 ± 0.65	-0.95 ± 1.17	0.058

\*p-value for difference in mean measure of nutritional status index between groups after supplementation

There were changes in the proportion of children stunted, underweight or wasted after the supplementation. Table 4.4 shows the respective prevalence of stunting, underweight and wasting among children in all the study groups after supplementation.

**Table 4.4 Proportions of children stunted (HAZ < -2), Underweight (WAZ < -2) and Wasted (BMIZ < -2) after supplementation**

Form of malnutrition	Treatment group		
	A (Vitamin A + Iron only)	B (Vitamin A + Iron + Zinc)	C (Control)
Stunting (%)	9.5	0	22.2
Underweight (%)	4.8	2.4	22.2
Wasting (%)	2.4	4.9	5.6

### 4.2.3 Change in nutritional status of the children

There were changes in the measures of the various nutritional indices after supplementation as shown in Table 4.5. The values have been obtained by subtracting the baseline values (Table 4.2) from the respective post-supplementation values shown in Table 4.3 and computing the mean and standard deviation thereof. The mean Values for some indices decreased while others increased.

The Body Mass Index (BMI) z-scores slightly decreased in Group B (Vitamin A + Iron + Zinc) and the control. The decrease in BMIZ in the control was statistically significant ( $p < 0.05$ ) but that in B was not. In Group A (Vitamin A + Iron), there was a slight but non-significant rise ( $p > 0.05$ ) in BMIZ.

Both HAZ and WAZ increased in all the groups. The increases in HAZ among the children in group B and the control were both significant ( $p < 0.05$ ) but that in group A was not ( $p > 0.05$ ). The increases in WAZ in all the groups were not significant.

**Table 4.5 Mean Changes in BMIZ, HAZ and WAZ of the children after supplementation**

Nutritional status	Treatment group		
	A (Vitamin A + Iron <sup>a</sup> )	B (Vitamin A + Iron + Zinc <sup>b</sup> )	C (Control <sup>c</sup> )
BMIZ	0.13 ± 0.63	-0.11 ± 0.43	-0.16 ± 0.36
<i>P value</i> **	0.352	0.108	0.009*
HAZ	0.28 ± 0.69	0.17 ± 0.28	0.24 ± 0.62
<i>P value</i> **	0.074	0.000*	0.025*
WAZ	0.28 ± 0.67	0.02 ± 0.34	0.02 ± 0.43
<i>P value</i> **	0.074	0.730	0.784

<sup>a</sup>n = 42; <sup>b</sup>n = 41; <sup>c</sup>n = 36

\*Change is significant at 95% confidence level

\*\*p-value associated with the change of the nutritional index between baseline and post supplementation

The proportions of children stunted, underweight or wasted also changed from the baseline levels after supplementation. Table 4.6 shows the nature and magnitudes of the changes in the proportions of children stunted, underweight and wasted after supplementation.

**Table 4.6 Changes in the levels of stunting and underweight in the groups after supplementation**

Form of malnutrition	Treatment group		
	A (n=42)	B (n=41)	C (n=36)
<b>Stunting</b>	0	9.8	5.5
<i>% change in stunting</i>	0%	100%	20%
<b>Underweight</b>	4.7	2.5	-5.5
<i>% change in underweight</i>	49%	51%	33%
<b>Wasting</b>	2.4	-2.5	-2.8
<i>% change in wasting</i>	50%	104%	50%

Treatment groups A: Vitamin A plus Iron, B: Vitamin A, Iron plus Zinc, C: Control

Minus sign indicates an increase in the prevalence of the form of malnutrition

The proportion of children stunted in group B reduced by 100%; that in A did not change at all while that for children in the control reduced by 20%. Stunting, therefore, reduced in group B and the control.

The prevalence of Underweight as well decreased in both groups A and B by about 50% but increased by 33% among the children in the control group. Wasting on the other hand increased in both groups B and the control but reduced in group A.

#### **4.2.4 Interrelationships between different Anthropometric Indicators**

##### **a) Stunting (Height for Age) and Underweight (Weight for Age)**

At baseline, there was a significant association between stunting and underweight in group B as well as the control (C) ( $P < 0.05$ ) but not in A ( $p > 0.05$ ). But, after supplementation, there was no longer any association between the two indicators in group B (Vitamin A, Iron plus Zinc) while that among the control remained significant ( $p = 0.000$ ). Thus, there was a change in the relationship between the two indicators in only group B and not in A (Vitamin A plus Iron) or C (control).

### **b) Underweight (Weight for Age) and Wasting (BMI for age)**

Both Weight for Age and BMI for Age can be used as indicators for underweight or overweight in children. However, because BMI for age is a composite indicator combining weight and height, it is as good an indicator for wasting as Weight for Height. There was a significant positive correlation ( $p < 0.05$ ) between the two indicators for all groups at both baseline and post-supplementation.

### **c) Underweight, Stunting, Wasting and Gender**

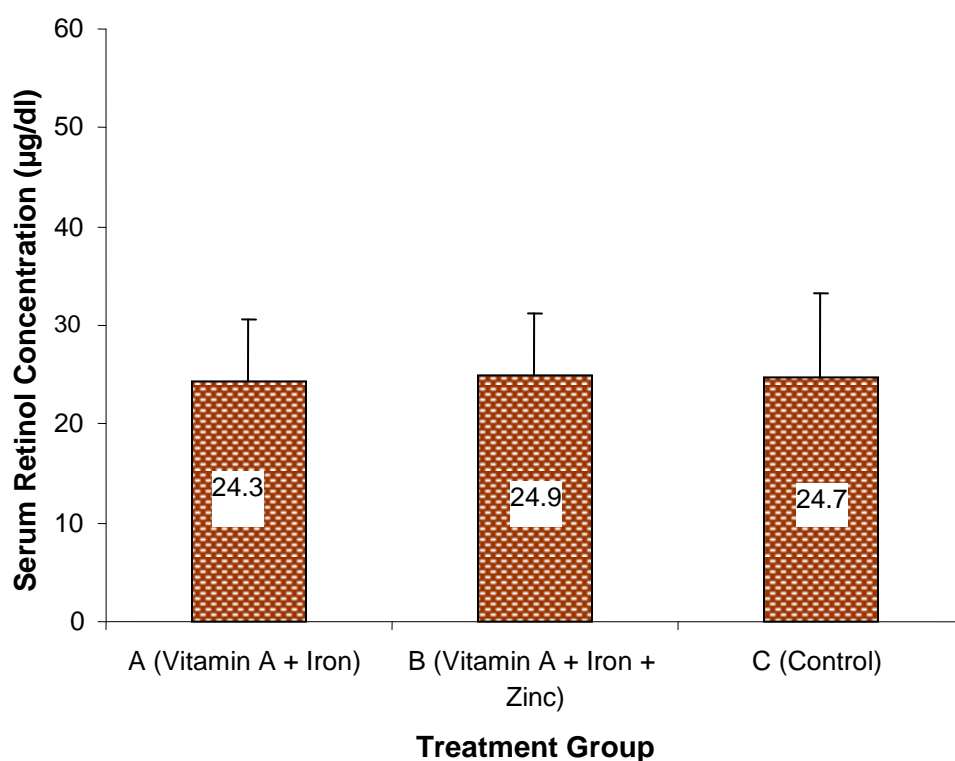
All the nutritional status indicators: underweight, stunting and wasting did not have any significant association with the gender of the children ( $p > 0.05$ ).

## **4.3 VITAMIN A STATUS OF THE CHILDREN**

### **4.3.1 Vitamin A status of the children at baseline**

The vitamin A status of the children was determined by measurement of serum retinol concentration using High Performance Liquid Chromatography (HPLC). The serum retinol concentration was also used to classify the children as Vitamin A Deficient or normal.

The mean serum retinol concentrations of the children in the three groups at baseline are shown in Fig. 4.1. It is apparent that the serum retinol concentration was similar across all the groups. Statistical analysis also revealed no significant differences in the levels of serum retinol between the groups ( $p = 0.904$ ). The mean retinol concentration among school children was  $24.26 \pm 6.35$  µg/dl in group A (Vitamin A plus Iron),  $24.89 \pm 6.37$  µg/dl in group B (Vitamin A, Iron plus Zinc), and  $24.73 \pm 8.52$  µg/dl in group C (control).



**Fig. 4.1 Mean serum retinol concentration of the children at baseline**  
(n for groups A, B and C were 44, 50 and 36 respectively)

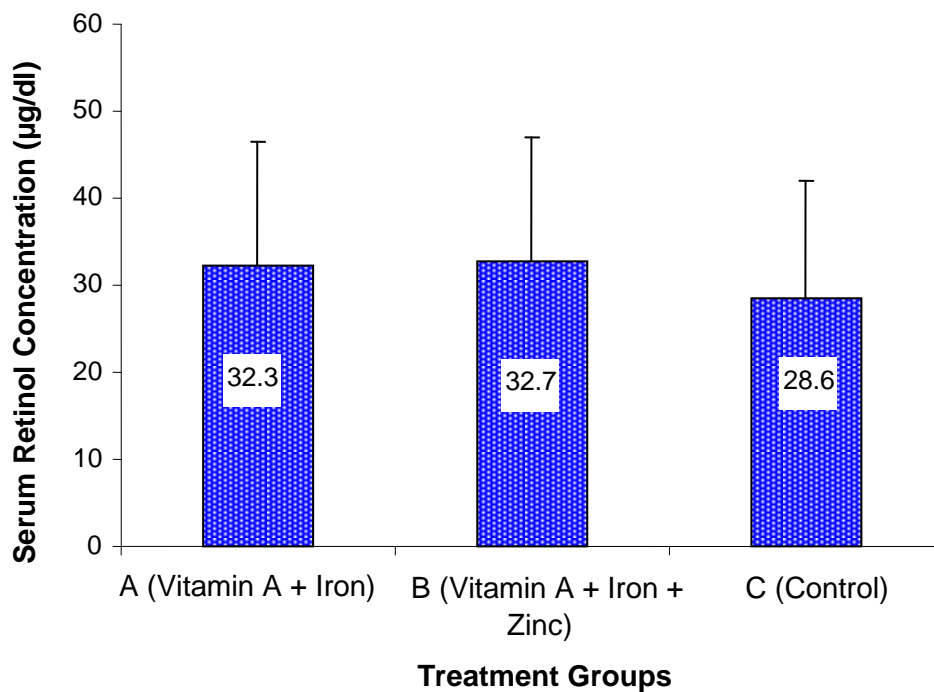
The prevalence of Vitamin A Deficiency among the school age children based on serum retinol levels at baseline is shown in Table 4.7. Children who had serum retinol less than 20 µg/dl were considered Vitamin A Deficient while those with serum retinol 20 µg/dl or more were regarded normal. On average, approximately 25% of the school children in the study were vitamin A Deficient.

**Table 4.7 Prevalence of Vitamin A Deficiency (VAD) (serum retinol < 20 µg/dl) at baseline**

Treatment group	VAD (%)	n
A (Vitamin A + Iron)	25	44
B (Vitamin A + Iron + Zinc)	22	50
C (Control)	27.6	36

### 4.3.2 Vitamin A Status after Supplementation

The mean serum retinol concentration of the children in the three groups after supplementation is shown Fig. 4.2. The levels were  $32.29 \pm 14.12$   $\mu\text{g/dl}$  in group A (Vitamin A plus Iron),  $32.72 \pm 14.18$   $\mu\text{g/dl}$  in group B (Vitamin A, Iron plus Zinc) and  $28.57 \pm 13.54$   $\mu\text{g/dl}$  in group C (control). After supplementation, as well as at baseline, there was no significant difference in the mean serum retinol concentration among the children in the groups ( $p = 0.353$ ).



**Fig. 4.2 Mean serum retinol concentration of the children at post-supplementation**

(n for groups A, B and C were 44, 50 and 36 respectively)

There were, however, some differences among the groups in the changes in serum retinol concentration that occurred after supplementation. These changes are shown in Table 4.8. There were increases in the mean serum retinol concentration in all the groups. A significant increase ( $8.03$   $\mu\text{g/dl}$ ) occurred in group A between baseline and post-supplementation ( $p = 0.000$ ). There was also a significant increase ( $p = 0.021$ ) of  $7.83$   $\mu\text{g/dl}$  in Group B (from  $24.89$  at baseline). On the other hand, there was no significant change in the mean serum retinol in the control group ( $p = 0.117$ ).

**Table 4.8 Mean changes in serum retinol concentration after supplementation**

Change in Serum retinol	Treatment group		
	<sup>1</sup> Vitamin A + Iron	<sup>2</sup> Vitamin A + Iron + Zinc	<sup>3</sup> Control
Mean (µg/dl)	8.03	7.83	3.84
Standard Deviation	13.89	13.52	14.32
♠ <i>P</i> value	0.000*	0.021*	0.117
♠♠ <i>P</i> value	0.236	0.176	

<sup>1</sup>n=44; <sup>2</sup>n=50; <sup>3</sup>n=36

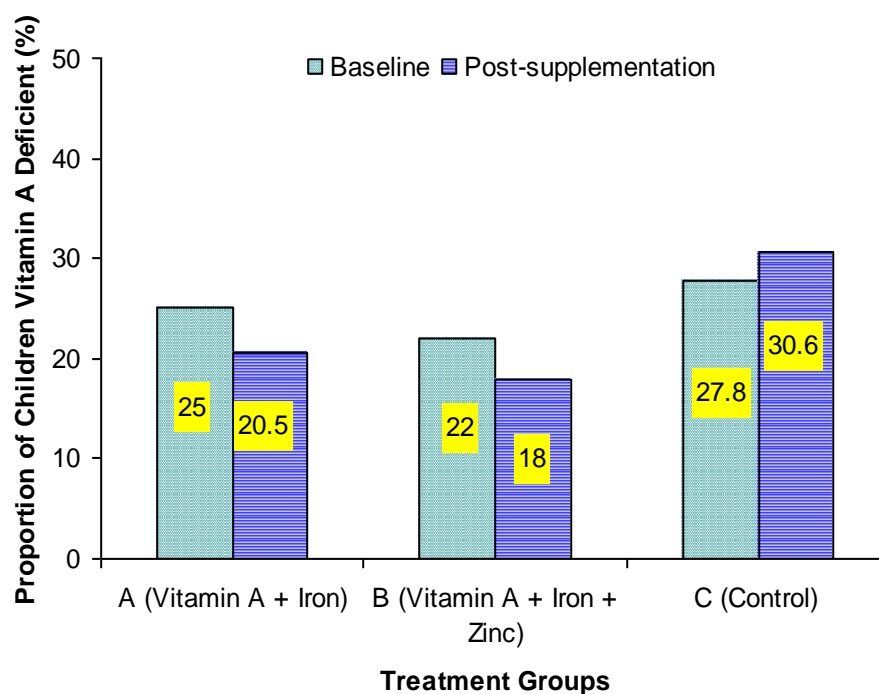
♠*p* values for changes in serum retinol of children between baseline and post-supplementation within a group

♠♠*p* values for changes in serum retinol for children in the group compared to those for children in the control

\*Change is significant at 95% confidence level.

The public health significance of the changes in serum retinol can be ascertained by critically examining the relative changes in each group compared to the control. Although there were significant increases in the mean serum retinol concentration within groups A (Vitamin A + Iron) and B (vitamin A + Iron + Zinc), neither was significantly different from the increase that took place in the control ( $p > 0.05$ ) as shown in Table 4.8.

The prevalence of Vitamin A deficiency in all the groups was determined both at baseline and post-supplementation. Vitamin A deficiency classification was based on serum retinol concentration below 20 µg/dl. Fig. 4.3 shows the respective VAD prevalence after supplementation compared to the baseline levels. A reduction in VAD prevalence occurred among children in all groups but C (control). Vitamin A Deficiency prevalence dropped by 18% in both groups A (Vitamin A + Iron) and B (Vitamin A + Iron + Zinc) while in the control, prevalence increased by 10%.



**Figure 4.3 Proportion (%) of children Vitamin A Deficient (serum retinol less than 20 µg/dl) at baseline and post-supplementation**  
(n = 44, 50 and 36 for groups A, B and C respectively)

#### 4.3.3 Relationship between Vitamin A Status and Stunting, Underweight

Vitamin A status did not affect stunting among the children since the relationship between the two variables was the same at baseline and after the supplementation, for the supplemented groups as well as the control. Vitamin A status did not as well significantly affect the underweight status of the children. There was, however, an increase in underweight in the control group among whom vitamin A Deficiency also increased. This possibly suggests that the supplementation prevented a possible deterioration in the children's weight for age since such a change did not take place in the supplemented groups. This kind of role had been suggested from similar studies done earlier elsewhere (Drake et al., 2002).



#### **4.3.4 Relationship between Vitamin A status and other socio-demographic factors**

##### ***4.3.4.1 Gender and Vitamin A status***

There was no significant relationship between vitamin A status and the gender of the children at baseline. This relationship was not altered after supplementation. Therefore, the effect of the supplementation was not affected by the gender of the children.

##### ***4.3.4.2 Age and Vitamin A Status***

There was a weak ( $p > 0.05$ ) correlation between the serum retinol and age of the children for all groups both before ( $r = 0.047$ ) and after supplementation ( $r$  for groups A, B and C were 0.329, -0.096 and -0.364 respectively).

## **CHAPTER FIVE**

### **DISCUSSION**

#### **5.1 THE STUDY POPULATION**

The study population was composed of primary school children selected from day schools. Absenteeism among the pupils and their inconsistency in school attendance led to some of them dropping out of the study. The consistent sample size was also affected by the long study period due to breaks during holidays and weekends. To maintain the effective period of four months, the study had to cover several actual months making the total period 16 months. Paired biochemical results (baseline and post-supplementation) for 130 children (83%) were obtained, of whom, 119 (76%) had valid paired results for anthropometry.

##### **5.1.1 Gender characteristics of the children**

The children were not stratified according to sex at recruitment but analyses were carried out to determine the effect of gender on the outcomes of the supplementation trial. The description covers only the children who participated to the end of the study and does not include those who dropped out during the study. As seen in Table 4.1, the gender proportions in the different groups were nearly the same and therefore, gender would not be expected to affect the results of the trial.

##### **5.1.2 Age of the study children**

Child age was one of the inclusion and exclusion criteria used at the beginning of this study. Although primary school children officially lie in the range of 6 to 12 years, 6 to 10 year-olds were selected for this study so as to reduce the effect of confounding factors associated with age such as adolescence (Kumar & Rajagopalan, 2006; Benefice *et al.*, 2003) and also to provide a range that would be easier to manage during the study.

## 5.2 NUTRITIONAL STATUS OF THE CHILDREN

### 5.2.1 Nutritional status of the children at baseline

A number of indicators were used to classify the nutritional status of the school children. These include Weight for age, Height for age and BMI for age. Z-scores for these indices were used in the classification into stunted, underweight or wasted categories according to the WHO standard growth references (WHO, 2007).

All the anthropometric indices for the study population lay within two standard deviations from the means of their standard references (Tables 4.2 and 4.3) showing that the children were generally well nourished both before and after the intervention (supplementation). They, however, showed a general tendency towards undernutrition, that is, stunting and underweight as the mean indices (HAZ, WAZ and BMIZ) lay below the standard means. It can be deduced that the general nutritional status of the children was below the optimal levels as defined by WHO (2007) criteria.

Mwaniki *et al.* (2002), in a similar study in Kenya but among much older (mean age 13 years) school children also found the nutritional status of the children to be below optimal. Studies elsewhere have also shown higher levels of malnutrition among school children (Drake *et al.*, 2002). This also differs from the nutritional status of children under five years of age in Uganda (UDHS, 2006). Therefore, while focus is directed at the emerging problem of childhood overweight and obesity and the double burden of malnutrition in developing countries (FAO, 2006; Benefice *et al.*, 2003), the less than optimal nutritional status among school children in semi-urban or rural areas who are not necessarily malnourished should be an area of concern.

## 5.2.2 Effect of Supplementation on the nutritional status of the children

### 5.2.2.1 Effect of supplementation on BMI for age (Wasting)

BMI for age is a composite measure of nutritional status. It is calculated as a ratio of the weight in kilogrammes to the square of the height in metres. There are cutoffs used to determine if a person is overweight ( $\text{BMI} \geq 25$ ), obese ( $\text{BMI} \geq 30$ ) or underweight ( $\text{BMI} < 18.5$ ) (Benefice *et al.*, 2003). For adults, BMI may be used as a proxy indicator of the levels of body fat (Banik *et al.*, 2007). But, for children of age 5 to 19 years, the World Health Organization recommends the use of BMI for age z-scores (BMIZ) to determine wasting (WHO, 2007; Cole *et al.*, 2007).

There was a significant decrease in BMIZ among the children in the control. No significant changes in the BMI z-scores occurred in the supplemented groups after supplementation (Table 4.5). The decrease in BMIZ in the control was due to a decrease in weight gain that occurred in this group relative to height. These results therefore suggest that both regimens A (Vitamin A plus Iron without Zinc) and B (Vitamin A, Iron plus Zinc) prevented a possible reduction in weight gain among the children. Failure to gain sufficient weight relative to height (wasting) can be attributed to a number of factors such as poor dietary intake and illness/disease.

Levels of wasting increased in group B and the control (Table 4.6), unlike in group A where there was a decrease in wasting. In this case, the regimen containing only Vitamin A and Iron (A) seemed to be more effective in reducing wasting than that containing all the three micronutrients (Group B). In addition, basing on these findings, it is important to note that the school children were generally in good nutritional status both at baseline and after supplementation. Studies among younger children who are either malnourished or having an underlying health condition have shown such children to gain weight after vitamin A supplementation (Hadi *et al.*, 2000).

#### ***5.2.2.2 Effect of supplementation on the children's Height for age and the prevalence of Stunting***

Children whose height-for-age Z-scores are below minus two standard deviations (-2 SD) are considered short for their age (stunted) and are chronically malnourished and those of whom they lie below minus three standard deviations (-3 SD) are considered severely stunted. Stunting reflects failure to receive adequate nutrition over a long period of time and is also affected by recurrent and chronic illness. Height-for-age, therefore, represents the long-term effects of malnutrition in a population and is not sensitive to recent, short-term changes in dietary intake.

As shown in Table 4.5, Height for Age Z-scores (HAZ) increased significantly in the group that received Vitamin A, Iron plus Zinc (B) and also in the control (C). Among children in the group which received Vitamin A and Iron only without Zinc (Group A), the increase in HAZ was not statistically significant. Also, when the increases in HAZ within the groups A and B were compared with those within the control (C), no significant differences were found ( $p = 0.812$  and  $0.527$  for groups A and B respectively). Thus, the observed improvements in Height for age of the children were not attributable to the intervention (supplementation). Kirkwood *et al.* (1996) and Rahman *et al.* (2002) did not as well find any significant effect of supplementation on height.

These findings agree with many other studies demonstrating no effect on height of vitamin A supplementation trials. However, it is at odds with findings by Hadi *et al.* (2000) which showed an increase in height of children after supplementation. It is worth noting that Hadi *et al.* (2000) used 4-monthly supplementation among 6-48 month-old children while this study involved daily supplementation of 6-10 year-old school children. Furthermore, the positive change among children in Hadi *et al.* (2000) was observed only among children with very low serum retinol while the baseline assessment in this study shows that children had normal serum retinol levels (Fig. 4.1).

At the end of the intervention, the mean HAZ was similar across all treatment groups ( $p = 0.232$ ). This indicates that the changes in height for age that occurred within the groups A and B were not caused by the differential treatment applied to them. Perhaps the reason why HAZ increased at all across all the groups is that young children of primary school age including 6 to 10 year-olds are intrinsically gaining height. In the absence of severe illnesses, hunger, stress and other socioeconomic and physiological conditions that retard growth (UNICEF, 1998), these children would be expected to gain height regardless of the vitamin A supplementation.

An increase in HAZ is, however, possible only if the children are gaining height at a rate higher than the average for reference children of the same age and sex. In this study, it was perhaps by chance that conditions other than those introduced by the study (supplements) were favourable for child growth in height. Secondly, children above five years can make basic choices about feeding, hygiene and general health unlike their minors, the preschoolers, who are largely under the care of mothers or other care-givers. It is also possible that the presence of this intervention inadvertently raised awareness among the children about their nutrition and health thus stimulating their conscience to make decisions that positively affected their height for age status.

A significant gain in height has also been demonstrated among anaemic school children after vitamin A supplementation (Mwanri *et al.*, 2000). Our study was carried out among healthy school children and therefore, a comparison of the outcomes of the two studies in as far as height or height for age is concerned is difficult. The role of the baseline status in respect of other underlying health conditions in this regard cannot be ascertained. Other studies have, however, suggested that baseline conditions significantly affect outcomes in such studies (Munoz *et al.*, 2000). In another study only remotely similar to the present, Kumar and Rajagopalan (2006), using multivitamin food supplements on residential school children for nine months showed significant gains in height, not necessarily height for age.

Even though the mean height for age at baseline was not significantly different for all treatment groups ( $p = 0.159$ ), levels of stunting dropped significantly in group B but not in

group A or the control (Table 4.6). This implies that catch-up growth during the supplementation period occurred among children in group B who were stunted. This indicates that supplementation had a role in reducing the prevalence of stunting among the children who received all the three micronutrients: Vitamin A, Iron and Zinc. This is consistent with the increase in mean height for age z-scores observed particularly in this group. But, given that stunting is a chronic problem and that supplementations with Vitamin A and Iron (group A) and placebo (Control) produced minimal changes in stunting, the 100% change in group B can be considered unique and perhaps this effect is not attributable to the supplemental regimes administered.

#### ***5.2.2.3 Effect of supplementation on the children's Weight for Age and the prevalence of Underweight***

Weight-for-age is a composite index that takes into account both acute and chronic malnutrition. Children whose weight-for-age measures are below minus two standard deviations ( $< -2$  SD) are classified as underweight and those below minus three standard deviations ( $< -3$  SD) as severely underweight.

The World Health Organization recommends the use of revised standards for assessing underweight among children aged five to ten years (WHO, 2007). At the end of this study, however, there were children slightly above 10 years for which these standards would not apply. Nevertheless, weight for age Z-scores were calculated and assessed using CDC (2000) growth reference criteria embedded in Epi info nutrition software.

Despite the increases in weight for age z-scores in all the groups after supplementation, analysis showed that there was no significant difference ( $p > 0.05$ ) between baseline and post-supplementation levels. Since the changes were similar in both the intervention and control groups, it is implicit that the intervention did not have any significant effect on the children's weight for age status. The observed increases are expected of children who are actively growing. It is important to note that although children in this age category experience a slower growth rate than children below five years of age, they are nonetheless, still growing.

Further, a change in the proportion of children who were underweight could indicate a role for the intervention. Results showed an increase in the overall proportion of children underweight in the control group while in the intervention groups A and B, there was a decrease (Table 4.6). As shown, levels of underweight in all the groups were generally below that of public health significance (15%) according to WHO criteria. Therefore, our assessment of the baseline results showed that underweight was not a public health problem among the studied school children 6 to 10 years of age. However, since weight-for-age is an indicator of both chronic and acute malnutrition, seasonality can play a big role in the magnitude of the problem. For instance, work-loads and feeding patterns at both school and homes vary at different times of the year and these affect the anthropometric characteristics of the children.

Since the two supplementation regimens produced identical reductions in the prevalence of underweight (Table 4.6) while nearly the opposite (increase in underweight) occurred in the control, the role of the supplementation, irrespective of the regimens, cannot be ruled out. The increase in underweight among the controls was strange given that there was a net increase in mean weight for age Z-scores. This means that many children in the control had their Weight for Age Z-scores falling below the cut-off of -2.

There are varying results from studies on the effect of vitamin A supplementation on weight. Mwanri *et al.* (2000) found an increase in weight, not necessarily weight for age, when anaemic school children in Tanzania were supplemented with Vitamin A and Iron only without Zinc. Another study (Villamor *et al.*, 2002) found that multivitamin supplements including vitamin A improved weight gain among HIV-infected pregnant women. These studies, which demonstrated positive outcomes, were carried out on subjects with an underlying health condition. Among healthy but preschool children (Fawzi *et al.*, 1997; Ramakrishnan *et al.*, 1995; Kirkwood *et al.*, 1996) no significant effect of supplementation with vitamin A on growth has been demonstrated. Our results confer with findings that supplementation does not have any significant effect on weight among school children.

There are many causes of underweight among children. According to the UNICEF conceptual framework of malnutrition (Fig.1.1), it can be caused by inadequate dietary intake



or disease. School children do not usually have adequate discipline and hygiene associated with a healthy diet (Biro *et al.*, 2007). Therefore, they are often likely to have low or excess intake of energy and other nutrients.

In addition, the study schools, like many primary schools in developing countries do not provide lunch for their pupils (MOES, 2009; LCD, 2007). As a result, the children have to move between home and school at lunch breaks or else carry packed lunch and yet few can afford to do either and would rather remain hungry until they go home after school. Thus, the children endure periods of starvation and reduced dietary intake.

Infections with disease and/or helminthes may also cause weight loss or reduced weight gain. It is possible that the relative effect of these factors was mediated by the supplementation since underweight increased in the controls but decreased among the supplemented groups (Table 4.6). Long *et al.* (2007) has shown that supplementation with vitamin A alone or with Zinc is associated with reduced incidences of infection with some of the common parasitic helminthes among young children. There are many factors that predispose school children to disease. While in school, the risk of acquiring an acute respiratory or other contagious disease is high. These also affect their nutritional status given the spiral relationship between infection and malnutrition (UNICEF, 1998; Allen and Gillespie, 2001). Moreover, communally owned water sources are less cared for, thus more likely to be contaminated and be a source of diarrhoeal disease. The ultimate effect of both disease and low dietary intake is to cause the child to lose weight or the capacity to gain sufficient weight for its age.

Generally, the children in this study were in good nutritional status. But, considering that all the indicators of malnutrition were below optimal, and that similar studies done elsewhere have reported higher rates of malnutrition among school children of this age group (Chowdhury *et al.*, 2007), it is imperative that school age children are not neglected in interventions that address malnutrition in childhood. The nutritional status of this demographic category of the population is particularly important because progress and achievement in education and long-term socioeconomic development hinge on the physical and mental growth and development within this group (de Onis *et al.* 2000; Mwaniki *et al.*,

2002). Further, to prevent, an intergenerational cycle of malnutrition and poverty (UNICEF, 2006c; Setboonsarng, 2005; UNICEF, 2000; MFPED, 2004), the school children ought to be specifically targeted.

### **5.2.3 Interrelationships between different Anthropometric Indicators**

#### **a) Stunting (Height for Age) and Underweight (Weight for Age)**

A significant association implies that a stunted child was more likely to be underweight than one who was not stunted. This situation was present among the children at baseline. Therefore, the kind of malnutrition present in this population was largely of a chronic nature and both stunting and underweight were likely to co-occur.

All the children in Group B who were stunted at baseline were no longer stunted at the end of the supplementation making it difficult to compare the two indicators in this group at end of supplementation. For group A, there was no significant relationship between stunting and underweight at both baseline and after supplementation. This implies that the supplementation did not cause any change in the relationship between the two forms of malnutrition, demonstrating a similar effect or none at all on both indicators.

The change in the relationship between stunting and underweight in Group B can be attributed to the role of the supplementation regime (Vitamin A + Iron + Zinc) in modifying the two variables although unequally or differently.

#### **b) Underweight (Weight for Age) and BMI for age z-score**

Both weight for age and BMI for age can be used as indicators for underweight in children. However, many authors have preferred using thinness or wasting to underweight in describing the BMI-for-age status of children. Indeed BMI-for-age has been described as a better indicator of wasting than is weight-for height in a given group of children (Cole *et al.*, 2007). It should be expected that WAZ and BMIZ have a significant positive correlation

since both indices are functions of weight. Unsurprisingly, results show a significant positive correlation ( $p < 0.05$ ) between weight for age and BMI for age Z-scores for all groups at both baseline and post-supplementation.

### **c) Underweight, Stunting and Gender**

Both underweight and stunting were not significantly associated with the gender of the children. Therefore, the male children were as likely as the female ones to be underweight and/or stunted. Unlike this one, several studies have reported boys to be more likely than girls to be stunted or underweight (UDHS, 2006; Schemann *et al.*, 2002; Fernando *et al.*, 2000). The UDHS (2006) was carried out among children below five years but the others were among children above five years of age. We did not stratify the children according to gender during selection of subjects in this study and this would constrain conclusions about the relationship with gender of stunting and underweight and the effect of supplementation thereof. But, results have shown that across the groups, there was no significant difference in gender distribution. Hence, it can be concluded that stunting and underweight were independent of the gender of the children.

## **5.3 VITAMIN A STATUS OF THE CHILDREN**

### **5.3.1 Assessment of Vitamin A status**

Serum retinol analysis by HPLC is the most commonly used indicator of vitamin A status of a population (Bowman & Russel, 2001; de Pee and Dary, 2002). Several other methods, however, exist that are equally or more sensitive and reliable. These methods include measurements of the Retinol Binding Protein (RBP), which has been used in many studies and believed to correlate very closely with serum retinol (Baingana *et al.*, 2008; WHO/FAO, 2004; UDHS, 2006). RBP measurement was not used in this study because it was unavailable in local laboratories and very expensive where it was available. Other methods include measurement of serum Retinol-Binding Protein/Transthyrein (RBP/TTR) molar ratio and Retinoyl  $\beta$ -glucuronide (RAG) hydrolysis test (WHO/FAO, 2004).

Serum retinol concentration by HPLC was used in this study for many reasons. It is an accurate indicator of vitamin A status and has been used in many related studies (Idindili *et al.*, 2007; UDHS, 2001; Munoz *et al.*, 2000). Estimations of prevalence of vitamin A deficiency have been carried out in as many studies using a classification of serum retinol concentration (Mwaniki *et al.*, 2002) and the method is recommended for tracking progress towards elimination of VAD (Baingana *et al.*, 2008). It is also reasonable to use serum retinol by HPLC for sample surveys like this rather than in population-based surveys where it may not be feasible due to operational difficulties and its requirement for highly technical expertise (Baingana *et al.*, 2008).

### **5.3.2 Serum retinol concentration at baseline**

The average mean serum retinol concentration among school children was computed as  $24.63 \pm 7.08$  µg/dl. This represents the average serum retinol concentration of the three groups at baseline. There was no significant difference between the groups ( $p = 0.904$ ). Therefore, the mean serum retinol concentration for all the children was within the normal range for vitamin A status ( $\geq 20$  µg/dl) (WHO/FAO, 2004; Sommer & Davidson, 2002). However, the criterion that classified as deficiency a serum retinol concentration below 20 µg/dl is used for all age categories including children under five years of age. Considering school children of mean age between eight and nine years, average serum retinol concentration of 24.63 µg/dl would indicate a tendency towards inadequacy of vitamin A among these children.

Normal levels of serum retinol can be as high as over 85 µg/dl or 3 µmol/l (Penniston & Tanumihardjo, 2006). The Serum retinol concentration among these children is considerably marginal; hence, they stand a risk of deficiency. In as much as the children are vulnerable to infection by respiratory, diarrhoeal or other diseases by virtue of their low vitamin A levels, any such infection can further reduce their vitamin A levels to clinically deficient levels (Stephenson, 2001; Haselow *et al.*, 2004; Allen & Gillespie, 2001; McLaren & Frigg, 2001; Oguntibeju *et al.*, 2004).

### **5.3.3 Prevalence of Vitamin A Deficiency at baseline**

Approximately 25% of the school children aged 6 to 10 years were found to be Vitamin A Deficient, indicating that vitamin A Deficiency among school age children is a public health problem in this area just like it is for the preschool children (UDHS, 2006). This could also indicate a similar problem among children in similar circumstances throughout the country. Indeed, this level is way above the limit for public health importance of 15% (de Pee & Dary, 2002) and should call for practical interventions targeting the school children. This finding is at variance with that by others (Holden and Macdonald, 2000) that show that dietary deficiency states are rare in school-age children and adolescents, but, supports the concern that vitamin A deficiency is a public health problem among school age children (Drake *et al*, 2002; Singh and West Jr., 2004).

Other studies in Africa have shown much higher levels of vitamin A Deficiency (VAD) among school age children. Berhe *et al.* (2007) revealed rates of VAD among Ethiopian school children as high as 57% in a study that, however, included children from malaria- and schistosoma-endemic areas. The prevalence of VAD (25%) among school children in peri-urban areas of Uganda, according to this study, is very similar to that of their counterparts in south-eastern Asia (23.4%) (Singh and West Jr., 2004). These levels among school children mirror those among preschoolers in respective regions (Table 2.6). This reflects not only a similarity in region-specific causative factors among preschool and school children but also suggests the need to focus on school children as well, in VAD control interventions where VAD is endemic.

### **5.3.4 Effect of supplementation on Vitamin A status**

#### ***5.3.4.1 Effect on Serum retinol Concentration***

The mean serum retinol concentration of the children in all groups after supplementation were higher than those at baseline (Figs. 4.1 and 4.2), illustrating the fact that the children's serum retinol concentrations increased after supplementation. Many studies have reported

increases in serum retinol concentration in various Vitamin A supplementation trials (Mwaniki *et al.*, 2002; Bahl *et al.*, 2002; Zimmermann *et al.*, 2006). Table 4.8 shows the differences between the baseline and post-supplementation levels of serum retinol. There were increases in all the groups. The observation that significant increases ( $p < 0.05$ ) occurred in Groups A and B and not in the control ( $p > 0.05$ ) imply that vitamin A-mineral supplementation regimes indeed contributed to increases in serum retinol concentration.

At baseline, the means of serum retinol concentration of children in all the groups: A (Vitamin A + Iron), B (Vitamin A + Iron + Zinc) and C (Control) were similar ( $p > 0.05$ ). At the end of the supplementation, the concentration in both groups A and B were also not significantly different. However, the difference in mean serum retinol concentration after supplementation between the two groups A and B (0.4 µg/dl) was negligible compared to the difference between either of the groups and the control (3.7 and 4.1 µg/dl for A and B respectively). This shows that slightly greater changes in serum retinol occurred in groups A and B than in the control.

It can therefore be inferred as well, that increases in serum retinol concentration can be achieved with vitamin A-mineral supplementation among school age children irrespective of the regimental combination. This observation suggests that supplementation with vitamin A and Iron alone is as effective in improving serum retinol concentration as with the two micronutrients plus Zinc. However, although there were raises in the mean serum retinol among the supplemented groups, these were not significantly different from those in the control ( $p > 0.05$ ).

This particular finding did not corroborate exactly that of a study among similarly healthy, but slightly older school children in Kenya (Mwaniki *et al.*, 2002) in which multiple-micronutrient supplementation produced significant increases in serum retinol concentration. The difference could be explained by the fact that older children are more likely to consume vitamin A rich foods than younger ones and therefore, differences in levels of intake could have caused the substantial differences in the outcomes of these two trials. Secondly, the study by Mwaniki *et al.* (2002) drew subjects from an area with known malaria and

Schistosoma endemicity. It has been shown that worm infections and malaria are risk factors for VAD. It has also been demonstrated that children with low vitamin A status at baseline can show greater improvements with vitamin A and/or Iron/Zinc supplementation than those with adequate vitamin A levels.

Another study (Dijkhuizen *et al.*, 2004) carried out among pregnant women showed improvements in vitamin A status of mothers and their infants when Zinc and  $\beta$ -carotene were added to Iron/folate supplements. Our study differs from this in the nature of subjects and also the regimental composition, thus, differences in outcome could have arisen.

#### ***5.3.4.2 Effect on prevalence of Vitamin A Deficiency***

The changes in levels of serum retinol corresponded to those in Vitamin A Deficiency in all groups but the control (Table 4.8 and Fig. 4.3). Reduction in Vitamin A Deficiency among children in groups A (Vitamin A + Iron) and B (Vitamin A + Iron + Zinc) accompanied increases in their serum retinol concentration. On the contrary, there was apparently an abnormal relationship between serum retinol and prevalence of vitamin A Deficiency in the control as both increased, yet the inverse relationship would have been expected.

The differences in changes in serum retinol concentration (Table 4.8) and in proportions of children vitamin A Deficient between the supplemented and control groups (Fig 4.3) demonstrate the role of the two supplementation regimens in changing the vitamin A status of the children. Since the two regimens (Vitamin A + Iron and Vitamin A + Iron + Zinc) were associated with nearly identical outcomes in improving serum retinol and reducing vitamin A Deficiency, there seems to be no comparative advantage in supplementing school children with all the three micronutrients together.

The observation that Zinc does not appear to make any difference is disturbing given the important role attributed to it in vitamin A metabolism (McLaren & Frigg, 2001; Christian *et al.*, 2001; Dijkhuizen *et al.*, 2004). It should be marked, however, that these studies were carried out among younger children than those in this study. Perhaps Zinc is a limiting

nutrient in younger children but not in older ones and therefore, its effect may be more marked in younger than older children.

### **5.3.5 The role of Zinc and Iron in Vitamin A status of children**

The metabolism of vitamin A requires the presence of Zinc because many enzymes involved are dependent on Zinc (McLaren & Frigg, 2001; Dijkhuizen *et al.*, 2004; Christian *et al.*, 2001). It would, therefore, be expected that in the children that were given only Vitamin A and Iron without Zinc (Group A), the metabolism of vitamin A would not be as efficient as in those who received all the three micronutrients together (Group B). The results from this study, however, stopped short of demonstrating this scenario but instead showed the two regimens having a similar effect. Although we did not include a single micronutrient regimen in our study, we know from other studies (Ramakrishnan *et al.*, 2004) that multi-micronutrient interventions are more efficient than single micronutrients in improving child growth.

There is substantial evidence that metabolism of vitamin A is not independent of that of iron. Many studies have demonstrated a co-occurrence of vitamin A deficiency and Iron Deficiency Anaemia (Miller *et al.*, 2006; Smith *et al.*, 1999; Ramakrishnan *et al.*, 1995; Graham *et al.*, 2007). Vitamin A status may not be dependent on plasma haemoglobin levels but the reverse is true (Smith *et al.*, 1999). Therefore, although inclusion of iron in the supplementation would not be expected to cause increased elevation of serum retinol levels, Muñoz *et al.* (2000) showed that supplementation with Iron and/or Zinc, independent of vitamin A, improved serum retinol levels. Our observation is that vitamin A and Iron alone do not improve serum retinol levels any more than the two plus Zinc.

### **5.3.6 Relationship between Vitamin A status, Stunting and Underweight**

Vitamin A status did not affect stunting among the children since the relationship between the two variables was the same at baseline and after the supplementation for the supplemented group, A, as well as the control. Vitamin A status did not as well significantly



affect the underweight or wasting status of the children. There was, however, an increase in underweight and wasting in the control group among whom vitamin A Deficiency also increased. This possibly suggests that the supplementation prevented a possible deterioration in the children's weight for age since such a change did not take place in the supplemented groups. This kind of role had been mooted by Drake et al. (2002) from a similar study.

### **5.3.7 Relationship between Vitamin A status and other socio-demographic factors**

#### ***5.3.7.1 Gender and Vitamin A status***

The effect of vitamin A supplementation was independent of gender. The female children were as likely as the male ones to become vitamin A deficient or to attain normal vitamin A status at both baseline and post-supplementation. This differs from many observations that boys were more likely to be vitamin A deficient than girls (UDHS, 2006) but similar to observation by Mwaniki *et al.* (2002) that the effect of vitamin A supplementation did not depend on the gender of the children.

#### **5.3.7.2 Age and Vitamin A Status**

A weak non-significant correlation between the serum retinol and age of the children was found for all groups both before and after supplementation. A positive but significant relationship between age and serum retinol was found among older subjects (Faure *et al.*, 2006) and also among female, but not male children (Mwaniki *et al.*, 2002). But, for this study, since the relationship between age and serum retinol was the same at the beginning as at the end, the effects of the supplementation trial were equally distributed across the age range of the children. Supplementation therefore, did not affect the relationship between age and vitamin A status of the children.

## CHAPTER SIX

### CONCLUSION AND RECOMMENDATIONS

#### 6.1 CONCLUSIONS

This study was set out to achieve the main objective of establishing a relationship between supplementation with Vitamin A, Zinc and Iron in varying regimental combinations on both the overall nutritional and vitamin A status of school children. Accordingly, a number of conclusions have been drawn including deductions on the role vitamin A supplementation can play in controlling vitamin A deficiency in school children.

##### *Nutritional Status of school children and effect of supplementation*

There are high levels of malnutrition among school children. Chronic malnutrition in form of stunting and underweight is prevalent in this age group. The average stunting rate stood high at about 15% while that of underweight was about 10%. Stunting among these children is clearly a public health problem while underweight approaches the level of public health significance.

While the supplementation appeared to improve indicators of nutritional status, the effect was not significant. Moreover, the vitamin A status was not associated significantly with other indicators of nutritional status including height for age, weight for age and BMI for age Z-scores.

##### *Vitamin A status of school children and the effect of supplementation*

School children in rural areas have marginally sufficient vitamin A levels in their blood. The average serum retinol concentration for the school children was about 25 µg/dl. This indicates low vitamin A stores hence a high risk for vitamin A deficiency. Supplementation with vitamin A, Iron plus Zinc improved the serum retinol concentration of the children

receiving them by nearly eight µg/dl. Therefore, supplementation with all the three or probably more micronutrients can have a positive role in improving serum retinol concentrations in school children.

Consistent with the low mean serum concentrations in school children is the high level of vitamin A deficiency among them. On average, about 19% of the school children were vitamin A deficient. Thus, vitamin A deficiency is a public health problem even among the school children especially in rural or peri-urban areas.

Although there was no significant increase in serum retinol concentrations of the children, supplementation with all the three micronutrients: Vitamin A, Iron plus Zinc as well as with Vitamin A and Iron only significantly reduced the levels of Vitamin A Deficiency among the children. This kind of intervention among school children is, therefore, relevant in the control of vitamin A deficiency especially in developing countries where risk and prevalence of Vitamin A Deficiency are high.

#### *Vitamin A Supplementation regimens*

Supplementation regimen that contained Vitamin A at RDA, Iron plus Zinc did not show significantly different improvements compared to that which contained only Vitamin A and Iron as far as increasing retinol concentrations and reducing Vitamin A Deficiency were concerned.

## **6.2 RECOMMENDATIONS**

The vitamin A supplementation policy in Uganda should provide for interventions targeting the school children. This study has demonstrated that significant reductions in vitamin A deficiency can be achieved through the use of vitamin A, Iron and Zinc supplements together. More studies are needed in as far as micronutrient supplementation of school children is concerned, especially in rural communities where dietary vitamin A intake is inadequate, before a recommendation to this effect can be considered.

Given the common occurrence of multiple micronutrient deficiencies and the positive synergistic action of multiple micronutrient supplements, that is, Vitamin A, Zinc and Iron in reducing Vitamin A Deficiency, it may be more beneficial if regimens containing all the three or more micronutrients are included in supplementation interventions rather than just one or two of them.

It is also recommended that, as more studies on multi-micronutrient supplementation of school children are undertaken, other strategies including dietary interventions, food fortification and bio-fortification, together with other public health measures should continue to be encouraged. School children should be enabled to have adequate dietary intake on a daily basis and provisions for balanced school lunches are particularly recommended. Whereas the benefit of saving a life or improving the quality of life outweighs any cost, it is always important to carry out a cost-benefit analysis to determine the most appropriate package of interventions for specific situations.

Generally, in order to fully understand the contribution of multi-micronutrient supplementation among school children, further research especially in the school age group is encouraged.

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## APPENDICES

### i. Data-recording form for Anthropometric measurements

Name of child/index of child \_\_\_\_\_

Sex (M/F) \_\_\_\_\_ DOB (dd/mm/yyyy) \_\_\_\_\_

Age (months) \_\_\_\_\_

Weight (Kg)		Height (cm)	

### ii. List of Chemicals and Equipment

#### a) Chemicals

The following chemicals were used during the analysis of retinal;

- Ethanol Absolute AR (Unilab)
- Methanol, HPLC (Prolabo)
- Hexane. HPLC (Avonchem Limited)
- White spot Nitrogen gas (BOC, Kenya)
- All-trans Retinol, (Synthetic, Sigma)
- Retinyl acetate (Sigma)

#### b) Apparatus / equipment

The apparatuses will include:

- Liquid chromatograph
- UV variable detector L4000H (Hitachi)
- Manual injector: Rheodyne (cotati)
- D 2520 GPC Integrator (Hitachi)
- L6000 pump (Hitachi)
- 50µL Hamilton syringe and needle
- 20 µL loop

Anthropometric equipment

- Stadiometers
- Digital Weighing scale

### iii) Theoretical/Conceptual framework for vitamin A supplementation

