AIDS is a disease. It is an infection, a syndrome, an illness, a disorder, a condition threatening to human life. It is an epidemic- a social crisis, an economic catastrophe, a political challenge, a human disaster. AIDS is known. It has been analysed assessed assayed tested measured surveyed considered reflected documented depicted exhaustively described. Its virus is primal particular sub- cellular, mutant enveloped nitrogenous. Our knowledge of it is clear and precise. But the disease is also unknown. It is guessed estimated projected approximated sketched debated disputed controverted hidden obscured. Still, it is mere fact: an event, a circumstance, a happening, a reality as present as the ocean or the moon.

AIDS is mouth and tongue and scar and nerve and eye and brain and skin and tum and gut. AIDS is smell and feel- of sweat and grime and snot and breath and bowel and secretion, discharge, pus, putrescence, disintegration, excrement, waste. Human waste. AIDS is feeling- painful sharp tingling burning heavy dull weakening wasting enervating diminishing destroying bereaving. AIDS is fear. It is breathless and nameless.

AIDS is stigma disgrace discrimination hatred hardship abandonment isolation exclusion prohibition persecution poverty privation.

AIDS is a metaphor. It is a threat a tragedy a blight a blot a scar a stain a plague a scourge a pestilence a demon killer rampant rampaging murderer. It is made moral. It is condemnation deterrence retribution punishment, a sin a lesson a curse rebuke judgement. It is a disease.

By Edwin Cameron.

Danksagung

Für die Ermöglichung der Arbeit, möchte ich mich bei Herrn Prof. Ibrahim Elmadfa herzlich bedanken, der mir das Vertrauen schenkte, dieses Projekt durchführen zu können. Durch seine Kontakte nach Kenia wurde mir eine Tür geöffnet, die mir nicht nur diese Arbeit sondern auch die Erweiterung meiner Persönlichkeit durch einmalige Erfahrungen ermöglichte. Herzlichen Dank!

Einen sehr wichtigen Teil zu dieser Arbeit haben wohl meine Eltern, Peter und Inge Bründl, meine Schwestern, Petra, Sandra und Anita, und mein Schwager Matt, beigetragen, die immer an mich und meine Ideen geglaubt haben und mich in jeder Hinsicht unterstützt haben. Ob mit einer Umarmung, einem Telefonanruf oder dem Prüfen meiner Englischkenntnisse. Vielen Dank!

Ein Dankeschön auch an Maria Bründl für ihre finanzielle Unterstützung.

Ein ganz besonderes Dankeschön möchte ich nach Kenia schicken, an Dr. Elizabeth Kamau- Mbuthia, die mich bei der Organisation vor Ort außerordentlich unterstützt hat und mir meinen Aufenthalt in ihrem Land unvergesslich gemacht hat. Für die Zusammenarbeit und die Erfahrungen in Kenia, sowohl in der Projektdurchführung als auch in meiner Freizeit möchte ich mich bei meinen Kolleginnen Juddy, Anne, Dean, Lydia, Wycliff, Lydia und George bedanken. Des Weiteren möchte ich mich beim Institut für Biochemie der Egerton University und im speziellen bei Richard für seine Unterstützung bedanken.

Weiter möchte ich Dr. Elisabeth Fabian danken, die mir jederzeit Rede und Antwort stand, mich aus einer „chemischen“ Not rettete und meine Arbeit kritisch unter Augenschein nahm. Elisabeth, ich danke dir für deine Bemühungen und deine Geduld!

Für die finanzielle Unterstützung, welche die Projektdurchführung überhaupt erst ermöglicht hat, möchte ich mich bei Herrn Ludwig Bieringer und meiner Heimatgemeinde Wals- Siezenheim bedanken. Herzlichen Dank für Ihr entgegengebrachtes Vertrauen!

Abschließend möchte ich mich noch bei allen Probandinnen und ihren Kindern in Kenia bedanken und wünsche ihnen für ihre Zukunft das Allerbeste- Danke!

Für Mama und Papa

universität wien

DIPLOMARBEIT

Titel der Diplomarbeit

The Coherence between the maternal nutritional Status of Zinc and Selenium and the Transmission of HIV to the breast fed Child

angestrebter akademischer Grad

Magistra der Naturwissenschaften (Mag. rer.nat.)

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Wien, im Oktober 2008

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ABBREVIATION

AIDS	Aquired Immunodeficiency Syndrome
BMI	Body Mass Index
CD4+	Receptor molecule on the surface of the T- Helper Lymphocyte
CI	Confidence Intervall
Cu- Zn SOD	Coppe - Zinc Superoxiddismutase
DNA	Desoxyribonucleid acid
EDTA	Ethylenediaminetetraacetic acid
GSH- Px	Glutathione- Peroxidase
GSH	reduced Glutathione
GSSG	oxidized Glutathione
Hb	Haemoglobin
HIV	Human Immunodeficiency Virus
H2O2	Hydrogen peroxide
IF	Interferon
Ig	Immunglobuline
IL	Interleukin
MTCT	Mother to Child Transmission
NADPH	Nicotinamide adenine dinucleotide phosphate
NCp	Nucleocapsid protein
PGH	Provincial General Hospital
PCR	Polymerase Chain Reaction
RNA	Ribonucleic acid
ROS	Reactive Oxygen Species
Se	Selenium
SOD	Superoxide Dismutase
Tat	Transactivating protein
t- BHP	t- Butylhydroperoxide
TRx	Thioredoxin Reductase

U	Units
UNAIDS	United Oranisations for AIDS
UNICEF	United Nations
WHO	World Health Organisation
ZDV	Zidovudine (anti- retroviral drug)
Zn	Zinc

1. Background & Objective

Human immunodeficiency Virus (HIV)/ acquired immunodeficiency syndrome (AIDS) is now the single leading infectious cause of death in developing countries. According to data of the United Nations programme on HIV/ AIDS (UNAIDS) [UNAIDS, 2007] the estimated number of people living with HIV worldwide in 2007 was 33.2 million (30.6- 36.2 million people), 2.5 million (1.8- 4.1 million) became infected and 2.1 million (1.9- 2.4 million) people died with AIDS. 68% of all adults and 90% of all children suffering from AIDS globally live in the sub- Saharan Africa and 76% of all deaths due to AIDS in 2007 occurred there. Worldwide more adult women (15 years and older) than ever before are now living with HIV.

Two major challenges for developing countries are the prevention of heterosexual transmission of HIV and the prevention of mother to child transmission of the virus. HIV can be transferred from an infected mother to her infant before or during birth, or after birth through breastfeeding. Mother to infant transmission (MTCT) is also known as “perinatal transmission” or “vertical transmission”. Vertical transmission transfers the virus from one generation to the next. Worldwide it is the second most common form of HIV transmission after sexual transmission.
Various facts, like the viral load of the mother, the way of birth, the maternal immune and nutritional status and the breastfeeding duration and pattern have an important impact on vertical transmission.
Zn and Se are important immune- modulators in human body and thus may be critical in determining the outcome of MTCT via breastfeeding.

Therefore, the objective of this study was to get information about sociodemographic data (questionnaire), the breast feeding pattern, the general nutritional situation (24-h-recalls) and the status of Se and Zn assessed by the activities of erythrocyte glutathione peroxidase (GSH-Px) and superoxide dismutase (SOD) and its impact on MTCT in lactating Kenyan women.

2. Literature Review

2.1. HIV/ AIDS

Human immunodeficiency Virus (HIV)/ acquired immunodeficiency syndrome (AIDS) is now the single leading infectious cause of death in developing countries. The United Nations programme on HIV/ AIDS (UNAIDS) published in 2007 statistics about HIV infections, new infections and AIDS deaths [UNAIDS, 2007]. In 2007 the estimated number of people living with HIV worldwide was 33.2 million (30.6 - 36.2 million people), 2.5 million (1.8 - 4.1 million) became infected and 2.1 million (1.9 - 2.4 million) people died with AIDS. 68% of all adults and 90% of all children suffering from AIDS globally live in the sub-Saharan Africa and 76% of all deaths due to AIDS in 2007 occurred there. Globally more adult women (15 years and older) than ever before are now living with HIV. In sub- Saharan Africa for every ten adult men living with HIV, there are about 14 women living with HIV. Young women (15 - 24 years) are four times more likely to be HIV infected than young men.

The AIDS epidemic has a major demographic impact on many countries were the prevalence of HIV is high and this effects include an overall reduction in life expectancy, a decline in child survival and an increase in orphans [SEMBA and BLOEM, 2001].

Two major challenges for developing countries are the prevention of heterosexual transmission of HIV and the prevention of mother to child transmission of HIV. HIV can be transferred from an infected mother to her infant before or during birth, or after birth through breastfeeding. Mother to child transmission (MTCT) is also known as “perinatal transmission” or “vertical transmission”. Vertical transmission transfers the virus from one generation to the next. MTCT of HIV is thought to occur in 15% of births to HIV positive women. Worldwide it is the second most common form of HIV transmission after sexual transmission. A strong correlation was found between levels of free virus in the mother’s blood and vertical transmission. But there are some more factors, which can lead to MTCT:

- Maternal Immune Status. A low CD4+ count is associated with an increased risk of perinatal transmission
- **Maternal Nutritional Status**. Vitamin A deficiency. In Africa, vitamin A deficiency in pregnant women appears to increase the risk of perinatal transmission. However, it is not known yet whether supplements of vitamin A reduce this risk.
- Breastfeeding duration and pattern
- Maternal viral load
- Prolonged period between the time when the mother's water breaks and the time of delivery
- Presence of ulcerations in the mother caused by sexually transmitted infections
- Vaginal delivery. Some studies suggest that vaginal delivery increases the risk of transmission, but this has not been conclusively shown, and cesarian sections are not recommended as means of reducing the risk of HIV transmission.

In resource poor settings in Africa, breast milk is an important source of energy and protein for the first two years of life. It reduces the incidence and severity of infectious diseases, thereby lowering infant morbidity and mortality.

Guidelines on breastfeeding with HIV, prepared jointly by UNAIDS, WHO and UNICEF, recommend that breastfeeding should be protected, promoted and supported among HIV negative mothers and mothers of unknown infection status. These guidelines furthermore promote fully informed, free choice of infant feeding methods for HIV positive mothers, but introduce artificial feeding as a viable alternative to breastfeeding. This has led to a heated debate regarding the breastfeeding method in resource poor settings for infants whose mothers are infected with HIV. Mothers get confronted with contradictory messages:

Breast milk as the best form of infant feeding and breast milk as a source of a deadly virus.

A WHO conducted metaanalysis [UNAIDS, UNICEF, WHO, 2006] of data from developing countries showed that mortality from diarrhoea, acute respiratory infections and other infectious illness is five to six times higher in infants who are not breastfed than those who are breastfed for the first months of life. The infant will run a higher risk to die from infectious disease, than from HIV infected mothers during breastfeeding. Support for exclusive breastfeeding has also come from a study in Kisumu, Kenya. In this study, the incidence of HIV infection was greater for infants who started mixed breast feeding before 30 days than for infants who started mixed breast feeding after 30 days. The effects of viral load and severity of maternal illness were not factored into the results [THOMAS et al., 2005].

2.2. Micronutrients, Malnutrition and HIV

2.2.1. Nutrition in Developing Countries

In developing countries the main food are cereals such as maize, rice, wheat, sorghum, millet, tubers or legumes, which comprise 70% of the energy intake [CHANDRA, 1992]. Small amounts of vegetables and fruits are part of the diet as well as animal products. In general, the diet is low in energy and nutrient density. Severe micronutrient deficiencies often occur as multiple deficiencies and coexist with protein- energy malnutrition. In addition, the bioavailability of some vitamins and minerals are low too, e.g. cereals have a high content of phytate, which bind Zn, non- heme Fe and other minerals in the intestinal tract, thus preventing their absorption. To change the nutritional situation in developing countries unconditional commitment from not only health planners, but also from politicians is required to ensure multisectoral collaboration and the simultaneous use of several interventions such as plant breeding for higher micronutrient density, food diversification, food fortification, infectious disease control and micronutrient supplementation.

People living in developing countries suffer more often from diarrhoea, respiratory tract infections and harbour commonly several parasites. Most micronutrients such as vitamin A, beta-carotene, folic acid, vitamin B_{12}, vitamin C, riboflavin, Fe, **Zn** and **Se** are significant immune- modulators and thus critical in determining the outcome of host microbe interactions [BHASKARAM, 2002]. Most generalized infections lead to reduced food intake and absorption of nutrients and increased utilization and loss of nutrients. The nutritional status of an individual may affect exposure to infectious agents, susceptibility to infection and in particular the severity and duration of the infection. It may even affect the virulence of an infectious agent. Vitamin and mineral deficiencies may lead to oxidative stress and result in increased apoptosis of immune cells and increased morbidity.

2.2.2. Micronutrients and HIV/ AIDS

Several general factors such as micronutrients can contribute to the deficiency of trace elements in the HIV infection. The supply can be diminished through loss of appetite and dysphagia, but also malabsorption and diarrhoea contribute to the inadequate status of trace elements. Cu, Fe, Se (see 2.4) and Zn (see 2.5) are most frequent concerned trace elements. While the status of Fe, Se and Zn have been noticed to be lower in HIV positive than HIV negative people, plasma concentrations of Cu tend to be increased in presence of HIV infection [CAMPERA, 1999]. This was clearly shown in a study carried out with pregnant HIV positive women and a mixed group of HIV positive adults [OBI et al., 1997; MORENO et al., 1998].

During the infection malabsorption of Fe, folates and vitamin B_{12} can often be observed. Serum Fe, transferrin and haemoglobin have been noted to be decreased, while ferritine, which could be an independent marker of infection progression, has been increase.
A marginal status or deficiency of micronutrients can lead to more frequent infections with *cytomegalovirus* or *pneymocystis carinii* in HIV patients.

Furthermore, HIV negatively modulates the expression of transferrin receptors on lymphoid cell membranes which seems to be parallel to the cytopathogenicity of the virus [SAVARINO et al., 2000]. The metabolism of Fe plays an important role on disease progression and morbidity and the accumulation of Fe seems associated with shorter survival.

A prospective cohort study from Rwanda reported that HIV infected women with low serum vitamin A concentrations during pregnancy were more likely than were women with normal vitamin A concentrations to have an infant who was HIV positive [GRAHAM et al., 1993]. However, it has to be considered, that HIV infection could lead to vitamin A deficiency by adversely affecting nutrient absorption and metabolism. Furthermore, serum retinol is depressed during acute phase response to infection, even when liver stores are adequate [FILTEAU et al, 1993].

2.3. Breastfeeding

Breastfeeding is the natural way of feeding infants and young children. Exclusive breastfeeding for the first six months of life ensures optimal growth, development and health of the baby. After six months, breastfeeding with appropriate complementary food continues to contribute on the infants and young children growth, development and health. Low rates and early cessation of breastfeeding have important adverse effects on health, social and economic implications for women, children, the community and the environment [LEON-CAVER et al., 2002; BRYCE et al., 2003].

Newborns are high susceptible to infections during early life, because of the late development of the immune system. Neutrophile functions, macrophage activation by interferon (IF)-α, formation of T-cells, immunglobulin (Ig)G antibody to T-cell independent immunogens and complement components have all been noted to be at a low level during early infancy [GOLDMAN et al., 1997]. Newborns produce lower amounts of many cytokines, including granulocyte-/ macrophage colony stimulating factor, IF- α, interleukin (IL)-3, IL-4, IL-6, and tumor necrosis factor (TNF)-α than adults [LÖNNERDAL, 2000].

There is significant evidence for infants in developing countries, that breastfeeding prevents infections. Several investigations indicate that the incidence and prevalence of illness is considerably lower in breastfed infants than in infants fed other diets. Breast milk contains several components that directly participate in the immune function of the breastfed infant. Many components interact directly with immune factors in the breast milk or more indirectly affect its responsiveness and activity. This includes cells as well as soluble substances in breast milk. Furthermore it should be recognized that breast milk provides a very balanced supply of nutrients to the infant.

2.3.1. Breastfeeding, MTCT and Micronutrients

Breastfeeding, source of high quality nutrients and as a mean of promoting protection from common infections, is widely practiced during the first two years of life in sub- Saharan African countries. Nursing and HIV positive mothers are still a very controversial issue, but the WHO/ UNICEF expert consultation and others have concluded that in situations such as those prevailing in sub-Saharan Africa, where infectious diseases and malnutrition are the major causes of the extraordinary high infant mortality rate, breastfeeding should be encouraged regardless of maternal HIV status [WHO, 1992] [DUNN and NEWELL et al., 1992].

Immunological active breast milk components

There are many specific and non- specific immunologic factors in human breast milk (s.2.1.), which may protect against HIV infection in infants. Among 18 months old infants of HIV infected women in Rwanda, a lack of persistence of IgM in breast milk increased the risk of infant HIV infection [VAN DE PERRE, 1993].

Lactoferrin from breast milk showed anti- HIV effects in vitro by binding the V3 domain of the HIV envelope protein gp120 [SWART, 1998]. Also glykosaminoglycanes and chemokines contained in breast milk do have defence duties in the infant´s organism.

Many of these immunological properties of human breast milk and its nutritional quality are connected to the nutritional status of the mother.

Influence of micronutrients on MTCT

There is evidence that micronutrient deficiencies may be important co factors in HIV progression and transmission [FRIIS and MICHAELSEN, 1998; SEMBA and TANG, 1999]. An inadequate micronutrient status of HIV/ AIDS infected women can lead to numerous adverse biological mechanisms (**Fehler! Verweisquelle konnte nicht gefunden werden.**).

Table 1: Potential relations between micronutrient deficiency and MTCT

[Mod.: DREYFUSS and FAWZI, 2002]

Maternal factors	↑ Progression of HIV disease
	↓ Cellular and humoral immunity
	↑ Systemic viral load
	↑ Clinical progression and opportunistic infections
Breast	↓ Mucosal immunity
	↓ Epithelial integrity
	↑ Systemic viral load
Child factors	↓ Birth weight
	↓ Preterm birth
	↓ Gastrointestinal epithelial integrity
	↓Cellular and humoral immunity

↑... increased, ↓... decreased

In 1986 The University of Nairobi carried out a study on MTCT. The study was initiated to determine the frequency, timing and routes of HIV- 1 transmission, identify the risk factors resulting in increased transmission and to identify potential interventions to prevent it [EMBREE et al., 2000]. In the study neither mothers nor children received specific antiretroviral therapy. The occurrence of maternal mastitis and the presence of nipple lesions during breastfeeding were

each associated with an increased risk of postpartum transmission of HIV-1. Breast and nipple lesions were not more common among HIV-1 infected women than in a concomitantly enrolled cohort of HIV-1 uninfected mothers. In the Kenyan study breastfeeding for longer than 15 months was associated with an increased risk of HIV-1 postnatal transmission. Children were more likely to become infected if the mother seroconverted while breastfeeding (**Fehler! Verweisquelle konnte nicht gefunden werden.**). EMBREE et al., [2000] suggested to all women who were known to be HIV-1 seropositive and who were impelled to breastfeed, should be given advice to wean their infants as early as they can. The issue of the potential protective effect of exclusive breastfeeding without supplemental feedings during the period of breastfeeding remained to be confirmed [COUTSOUDIS et al., 1999].

Table 2: Ultimate HIV-1 status in children of HIV-1 infected women

[Mod.: EMBREE JE et al., 2000]

	Child HIV-1 uninfected	Child HIV-1 prenatal infected	Child HIV-1 postnatal infected
HIV-1 infected women at delivery	310	51	37
HIV-1 seroconverting women during follow up	7	0	5
Total number of children	317	51	42

In a randomized trial comparing breastfeeding and formula feeding among HIV-1 infected women, breastfed and formula fed infants had similar mortality rates after two years (frequent cause of death: pneumonia and diarrhoea). However, formula fed infants tended to have an increased risk of diarrhoea during the first three months of life while breastfed infants had better nutritional status during the first six months of life. The final conclusion was that infants, which were fed

with formula food, had a significant higher HIV-1 free survival at two years [NDUATI et al, 2001].

Comparable mortality rates between the breastfed and the formula fed infants of HIV positive women were also observed in another study [NDUATI et al., 2000]. Further, this investigation showed that in the first place the estimated rate of breast milk HIV-1 transmission was 16.2% during the first two years of life. Given an HIV-1 infection rate of 36.7% in the breastfeeding arm, breast milk transmission accounted for 44% of all infant infections among those exposed to breast milk. Moreover, the timing of breast milk and HIV-1 transmission was determined in this investigation. 76% of all breast milk transmissions occurred during the first six months of life indicating that substantial transmission takes mainly part during early breastfeeding.

Finally, this study found that HIV-1 free survival rates at two years were significantly lower in the breastfeeding group than in the formula one. Only 58% of the women who practised breastfeeding had an infant at two years, who was alive and HIV-1 infection free.

COUTSOUDIS et al. [2000] did a research about feeding patterns and HIV transmission in Durban, South Africa. Three different types of feeding practices: exclusive breastfeeding, mixed breastfeeding and never breastfed were investigated and compared. The three feeding groups did not differ significantly in any other risk factors (preterm delivery, mode of delivery, duration of membrane rupture, maternal CD4:CD8 cell ration, $CD4^+$ cell count or serum retinol level) identified to be associated with MTCT. There were no differences in diagnosis with mastitis or breast abscesses during follow up among 5 out of 118 exclusive breast feeders compared with 5 out of 276 mixed breast feeders. Mastitis or breast abscesses were not significantly higher among mothers of breastfed children who acquired HIV infection. Infants who were exclusively breastfed had no excess risk of MTCT of HIV-1 over six months when compared with infants who were not breastfed at all but given formula and other food [COUTSOUDIS et al., 2000]. Those at the highest risk were infants fed by HIV positive mothers on a mixture of breast milk and other foods and liquids. The exact mechanism is still under debate, but it may be associated with a

decrease below a critical threshold of protective factors in breast milk as a result of the consumption of less breast milk and replacement by formula and other liquids and solids. The scientists concluded that contaminated fluids and food introduced in mixed breastfed babies damage bowel and facilitate entry into the tissues of the HIV contained in breast milk.

An Indian study showed an increased risk of hospitalization, resulting from increased risk of morbidity, for formula fed Indian infants who were born to HIV infected mothers, compared with breast fed infants [PHADKE et al., 2003]. In the mentioned study all women received prenatal Zidovudine (ZDV) therapy for prevention of MTCT. Four infants died during the research, but it is worth to mention, that none of the infants was breastfed and HIV-1 PCR positive at 48 hours or two months postpartum. The scientists concluded that the use of prenatal ZDV or Nevirapine can significantly reduce the risk of MTCT and that the risks associated with commonly utilized alternative infant feedings and not with exclusive breastfeeding.

MTCT – the role of breast milk

Mastitis is an inflammation of the breast tissue characterized by an elevated concentration of leukocytes and sodium in breast milk caused by the opening of the paracellular pathways between mammary cells. It can result from different causes, such as a local mammary inflammation in response to either mechanical or infectious insult, milk stasis or mammary gland involution derived from reduced breast milk production as seen during weaning, micronutrient deficiencies or systemic infection [WILLUMSEN, 2002].

Breast milk sodium concentration is controlled around 5- 6 mmol/L, but in the case of mastitis it raises to 12- 16 mmol/L. Another indicator for breastfeeding problems and mastitis is the Na^+/K^+ ratio of the milk, which should be less than 0.6. Ratios greater than 1.0 indicate severe breastfeeding problems. There is evidence that HIV infected cells enter breast milk via this pathways, thereby increasing the HIV load and the risk of MTCT [DREYFUSS and FAWZI, 2002].

Infant thrush and mastitis also interrupts the mucosal barrier to transmission. In addition, mucosal infection with *candida* species results in migration to the site of infection of large numbers of activated $CD4^+$ cells and macrophages, which are the primary target cells for HIV-1. Thrush and mastitis both result in immune activation.

WILLUMSEN et al. [2001] found that the HIV-1 load in breast milk was below the level of detection (<200 copies/ml) in approximately one third of samples (90 participants) and was shed intermittently during the course of lactation over the first three months postpartum. The viral load of the breast milk was often below the detection limit in one breast, but could be determined in the other breast - no clear pattern could be observed in this investigation.

In another study by WILLUMSEN et al. [2003] the associations between subclinical mastitis, breast milk RNA viral load and infant feeding practices among breastfeeding HIV positive women in South Africa were investigated. In this survey a significant positive association between the Na^+/K^+ ratio as well as the IL-8 plasma concentration and the milk RNA viral load was found. There was no interaction between feeding mode and the effect of Na^+/K^+ ratio on RNA viral load, but there was a trend for higher viral load at all times in the mothers of the postnatal infected group compared with the uninfected group and a significantly higher Na^+ K^+ ratio in milk after 14 weeks. These children may have acquired infection through breast milk in early life, but available techniques cannot differ between this from of MTCT and delivery-acquired infection [WILLUMSEN et al., 2003].

MIOTTI et al. [1999] published following results of a study done in Malawi, where 47 of 672 breastfed infants became HIV positive: The cumulative risk of infection for infants continuing to breastfeed was 3.5%, at the end of five months, 7% at the end of 11 months, 8.9% at the end of 17 months and 10.3% at the end of 23 months (**Fehler! Verweisquelle konnte nicht gefunden werden.**). Still, symptomatic mothers are believed to transmit HIV more often to

their infants than asymptomatic women. Researchers could not find any connection or evidence for this assumption in the investigation. Compared to asymptomatic mothers, symptomatic mothers and mothers who died had a similar transmission rate. The finding of a higher postnatal transmission risk in women with lower parity was unexpected, as the women's younger age would likely be accompanied by lesser virologic and immunologic compromise. According to KUHN et al. [1997] this may be consistent with the hypothesis, that mothers who are less experienced with breast feeding are more likely to have subclinical mastitis and thereby a higher HIV transmission rate. This could be confirmed by the study of MIOTTI et al. [1999], where no association between clinical mastitis or cracked nipples and a higher MTCT rate was found.

Table 3: Risk Factors for HIV Transmission through Breastfeeding

[Mod.: MIOTTI et al., 1999]

	Comparison		**Infants**	
			HIV infected	**No infected**
Mother's age (y)	>25 (256)	<25 (413)	46	623
Breast Problems	No (354)	Yes (73)	28	399
Birth Weight (g)	<2500 (111)	>2500 (559)	47	623
Parity	1-3 (478)	>4 (194)	47	625

MTCT – the role of nutrients

SEMBA et al. [1999] found evidence, that oxidative stress and immune dysfunction are associated with increased risk for mastitis. Furthermore it is known, that micronutrient deficiencies may increase the susceptibility to mastitis by contributing this conditions. In a randomized dietary supplementation trial of pregnant women in Tanzania, not vitamin E rich sunflower oil but vitamin A rich red palm oil significantly reduced the risk of mastitis at three months postpartum

[FILTEAU, 1999]. The HIV status of the individuals was not assessed, so the potential beneficial effects of supplementation on HIV viral load in breast milk and MTCT could not be examined.
In a view studies it has been showed, that micronutrient supplementation during pregnancy reduced the risk of low birth weight and preterm birth which may be risk factors for vertical transmission during delivery or breast feeding. In several studies it was reported that HIV infected women were at increased risk of delivering low birth weight babies, of preterm deliveries and intra uterine growth retardation [OSMAN et al., 2001; COLEY et al., 2001; DREYFUSS et al., 2001]. However, coherence between low birth weight babies of HIV infected women and a poor nutritional status of the mothers is still discussed controversial.
A study carried out in South Africa did not find significant differences between birth weights of the babies of HIV infected and non infected women [BOBAT et al., 2001].

There is evidence, that MTCT in uteri or through breast feeding can be influenced by the epithelial integrity of the gastrointestinal mucosal lining of the foetus and infants. The risk can be reduced by the barrier function of the gastrointestinal mucosal epithelium and by the immune response in the gut. Vitamin A and Zn deficiencies are associated with epithelial impairment in the gastrointestinal tract.

2.4. Selenium

2.4.1. Function

Se is a very important component of glutathione peroxidase (GSH-Px), which is a reducing equivalent of glutathione. GSH-Px catabolises the reduction of organic and anorganic hydroperoxides and thereby exerts antioxidant effects in the human organism. Due to its protective effect against autotoxic processes GSH- Pxs can be found in high amounts in macrophages and erythrocytes. Up to now 4 Se dependent GSH-Pxs, all with different specifity have been identified [SHIKE et al., 2006] (**Fehler! Verweisquelle konnte nicht gefunden werden.**).

Table 4: Different Types of GSH- Px

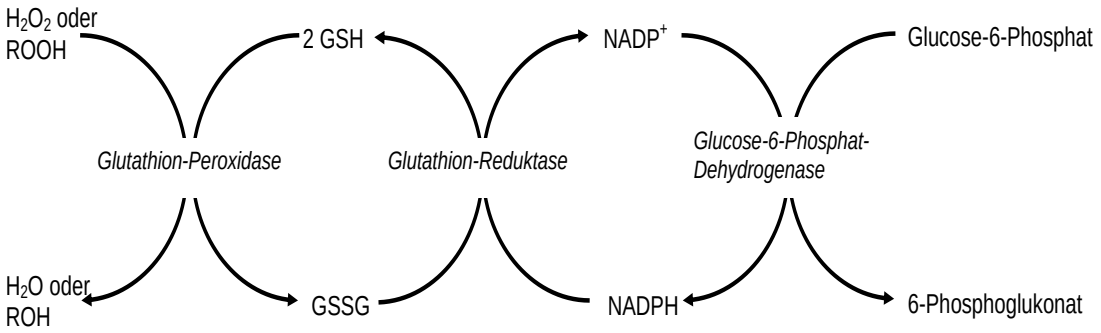
[Mod.: SHIKE et al., 2006]

GSH- Px 1	Most abundant member, present in all cells
GSH- Px 2	Cellular enzyme in the gastrointestinal tract
GSH- Px 3	Present in plasma and milk
GSH- Px 4	Phospholipide hydro peroxide, Catalyzes the reduction of fatty acid hydro peroxides that are esterified in phospholipids

The GSH-Pxs catabolise hydrogen peroxide and fatty acid derived hydroperoxides. They have generally been considered to protect cells from these oxidant molecules. The GSH-Pxs have regulatory roles in the cell by affecting the concentrations of oxidant molecules. Moreover, the GSH-Pxs have different localizations and substrate specificities, which could be part of a regulatory strategy.

In the enzymatic neutralization of reactive oxygen species (ROS), SOD catabolises the transformation of a superoxide radical into H_2O_2. GSH-Px detoxifies H_2O_2 and lipid peroxides (ROOH) using reduced glutathione (GSH). This reaction prevents the formation of an extremely toxic hydroxyl radical. During this reaction, GSH is transformed into oxidized glutathione (GSSG) which is regenerated again (to GSH) via glutathione reductase (GR) in presence of NADPH (Figure 1).

Figure 1: GSH- Px Circle



Furthermore Se is part of some more enzymes (**Fehler! Verweisquelle konnte nicht gefunden werden.**).

Table 5: Selenium dependent Enzymes

[Mod.: ELMADFA and LEITZMANN, 2004]

Iodithyronine Deiodinases Type 1- 3	Catalyze the deiodination of thyroxine, triiodthyronine and reverse triiodthyronine and thereby regulate the concentration of the active hormone triiodthyronine
Thioredoxin Reductase	A flavin containing selenoprotein dependent on reduced nicotinamide adenine dinucleotide phosphate that reduces the internal disulfide of thioredoxin. 3 isoforms: one in the cytosol, one in the mitochondrion and the third is expressed in the testis.
Selenoprotein P	An extracellular glycoprotein found in plasma and also associated with endothelial cells. One function of selenoprotein P is to supply Se to the brain to maintain normal neurologic function and to the testis for spermatogenesis. Selenoprotein P contains a large fraction of the plasma selenium. It has been associated with the oxidant defense properties of selenium.
Selenoprotein W	Is bound to GSH, for RedOx exchanges, Selenoprotein W concentration decreases in Se

	deficiency.

2.4.2. Deficiency

Se deficiency coexists with sever protein- energy malnutrition in humans. Deficiency of Se leads to marked changes in many biochemical systems. It decreases GSH-Px activities (used as index of the human Se status) in plasma and liver and is very sensitive to Se supplementation. In case of Se deficiency the phagocytic activity is restricted and the influence for infectious diseases is increased.

Se deficiency also reduces antibody production, cytokine synthesis, cell-mediated cytotoxicity and lymphocyte proliferation, is affecting the cytochrome P 450 system and responsible for the changes in thyroid hormone metabolism. The micronutrient deficiency in itself does not usually cause clinical symptoms in a free living human, but it can influence the course of various infections (e.g. *coxsackievirus infections*).

2.4.3. Assessment

Biochemical evaluations like measurement of selenoproteins, such as plasma selenoprotein P and GSH-Px, are useful in assessing the Se status in humans. Their assessments in blood or its fractions appear to be a satisfactory method to get information about the Se status in clinical practise and experimental investigations (**Fehler! Verweisquelle konnte nicht gefunden werden.**). Thus, adequate Se nutriture can be assumed if plasma GSH-Px activity and selenoprotein P concentration are normal or if plasma Se concentration is about 1 µmol/L or greater [ELMADFA and LEITZMANN, 2004; SHIKE et al., 2006].

Table 6: Assessment of Selenium in blood fractions

[Mod.: ELMADFA and LEITZMANN, 2004]

Plasma/ Serum level	*Short term* Acceptable Status: 7.9 µg/dl

	Young Healthy Women: 5.3 – 7.9 µg/dl Pregnant/ Nursing Women: 4 - 8 µg/dl
Blood cells	*Long term* *GSH-Px activity in erythrocytes and/or thrombocytes* Germany: 11.3 – 7.5 µg/dl
Urine	*Very seldom* Germany: 1.1 – 2.3 µg/dl

The most common functional index of Se status includes activity of GSH-Px in plasma, erythrocytes or whole blood. Based on data from large number studies, DIPLOCK [1993] concluded that blood or plasma Se correlated well with erythrocyte GSH-Px activity. With daily intakes up to 40 µg the enzyme activity reaches a plateau. Erythrocyte GSH-Px activity increases with intakes up to 60 to 80 µg/d, while platelet GSH-Px activity only plateaus with intakes around 100 - 110 µg/d [VAN DAEL and DEELSTRA, 1993].

Furthermore, Se urinary excreted as trimethylselenium can be used to assess the Se status of an individual.

Assessments of Se status by calculating the dietary intake from food composition tables is a risky procedure because of the wide variation in the Se content of foods. To make accurate statements a direct chemical analysis of the diet would be needed.

2.4.4. Source and Recommended Intake

In a Se rich soil, cereals may comprise about 75% of the total Se intake, but if the soil is poor, they may provide less than 10% of the intake. Liver, kidney, and seafood tend to have the highest amounts of Se (0.4 - 1.5 mg/kg wet weight). Dairy products provide <0.1 to 0.3 mg/kg, and fruits and vegetables show <0.1 mg/kg, only.

The daily recommended intake of Se for lactating women and infants is given in **Fehler! Verweisquelle konnte nicht gefunden werden.**:

Table 7: Recommended Intake of Selenium

[Mod.: D_A_CH, Referenzwerte für die Nährstoffzufuhr, 2001]

Lactating women	30- 70 µg/d
Infants 0- 4 months	5- 15 µg/d
Infants 4- 12 months	7- 30 µg/d

2.4.5. Selenium and HIV/ AIDS

In HIV infected people, the progression to AIDS and the decline of T-helper cell counts, are accompanied by a parallel decrease in blood Se levels. Se deficiency has been found to correlate with an increase of mortality among HIV infected individuals, even during asymptomatic stages.
The decrease of Se is even more important when there are opportunistic infections. The loss of $CD4^+$ lymphocytes and other immune cells causes a progressive weakening of the immune system. Microbes that are normally kept in check by the immune system do have the opportunity to flourish and cause a disease. Opportunistic infections like *cytomegalovirus disease, mycobacterium avium complex, tuberculosis* and *cancer* are directly responsible for up to 90 % of deaths associated with HIV disease.

Se supplementation is able to stimulate the proliferation of activated T-cells of the immune system. It elicitates an enhancement to response to antigen stimulation, to generate cytotoxic lymphocytes, to destroy tumor cells and to increased natural killer (NK) cell activity. Additionally, growth regulatory IL-2 receptors on the surface of activated lymphocytes and NK cells become unregulated in case of Se supplementation. IL-2 is very important for lymphocytes reproduction and their differentiation into cytotoxic cells. Also a lower content of IgM and IgG has been discussed.

HIV infection favours greater oxidative stress, which can lead to a higher viral replication [STEBENS, 2004]. GSH-Px and thioredoxin reductase (TRx), which include Se, play an important role in oxidative stress reduction (see 2.4.1) and

may inhibit the HI virus replication. The increase of free radicals leads to increased viral replication and an increase of host cell apoptosis, particularly $CD4^+$ cells. SANDSTRÖM et al. [1998] showed in a well exhibited study that cells over expressing GSH-Px accelerated viral replication and were associated with cytopathogenetic effects. A progressive depletion of GSH or plasmatic thiols has been observed through the course of the HIV infection. The decrease is directly correlated with the number of $CD4^+$ cells and inversely correlated with the viral load. There is a poor survival rate for HIV infected individuals with lower GSH levels and an improved survival rate when GSH is replenished and maintained [RODRIGUEZ et al., 2000].

In addition to these effects daily supplementation of 100 µg Se (as selenomethionine) for 12 months leads to a modulation of GSH-Px activity in HIV positive patients [DELMAS-BEAUVIEAUX et al., 2006]. In patients allocated to daily Se supplements, the erythrocyte GSH-Px increased from 47 to 67 U/g Hb, while it declined from 63 to 36 U/g Hb in HIV infected patients not receiving supplements. Allegedly there was a decline in oxidative stress markers in the Se group and an increase in the non- supplemented group, while there was no effect of Se on $CD4^+$ counts or the incidence of opportunistic infections [CONSTANS, 1999].

2.4.6. Selenium and MTCT

It is very important to mention, that in a pregnant AIDS patient a low Se concentration can lead to a higher risk of foetal mortality and a higher risk of intrapartal transmission [KUPKA et al., 2005].

2.5. Zinc

2.5.1. Function

In general the function of Zn in human body can be categorized in a catalytic, structural and regulatory one. Zn is part of many different enzymes e.g. metalloenzymes, which are important for the RNA and protein biosynthesis and

performs a catalytic role in various types of enzymes (there are about 300 enzymes, whose activity depends from Zn) [ELMADFA and LEITZMANN, 2004].
Cu/Zn- superoxide dismutase (SOD) is essential for the antioxidant defence system in humans (**Fehler! Verweisquelle konnte nicht gefunden werden.**).

Figure 2: Cu/Zn- SOD Circle

(A) $Cu^{2+} - SOD + O_2^{-} \rightarrow Cu^{1+} - SOD + O_2$

(B) $Cu^{1+} - SOD + O_2^{-} + 2H^{+} \rightarrow Cu^{2+} - SOD + H_2O_2$

Besides, Zn does have a catabolic effect on the quarter structure of holoenzymes, which makes them more resistant for heat and pH- changes.

Micronutrients are able to affect the function of the immune system and Zn is undoubtly playing a central role over here. Cells of the immune system do have a short life span, proliferate and differentiate instantly. Zn deficiency can lead easily to various and heavy immune dysfunctions in the humoral as well as in the cellular immune system (s.2.5.2). Zn is important for thymocyte development, T-cell function and thymic integrity. It is essential for the thymic functions by means of a Zn dependent thymic hormone called thymulin, which is indispensable for the differentiation and maturation of T cells.
The micronutrient is also necessary for the regulation of gene expressions. Especially in those enzymes, which are connected to bring up nucleic acid. It is also part of transcription factors in form of Zn- fingers (*Fehler! Verweisquelle konnte nicht gefunden werden.*). Zn- fingers do have a definite domain, which interact with the DNA. This is for a specific recognition of attaching DNA-sequences. Zn- fingers are also essential to the viral structure, proviral DNA

synthesis and production of infectious viruses and are required for the activity of reverse transcriptase [TANCHOU et al., 1998].

Classic examples of Zn- fingers are the retinoic acid and calcitriol receptors.
DNA arrays have recently shown that Zn deficient human monocytes and macrophages have gene clusters that are either positively or negatively responsive to Zn. This includes genes associated with apoptosis and receptor mediated immune regulation [SHILS et al., 2005].
Zn does play an important role in cell proliferation and in protein biosynthesis. Additionally it is supposed, that Zn does also have an influence on hormone metabolism e.g. insulin, growth hormones and sexual hormones.

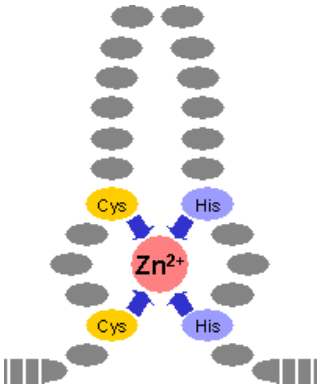


Figure 3: Zn- Fingers [www.wikipedia.org, 2008]

2.5.2. Deficiency

Zn deficiency belongs to the most common malnourishments worldwide. In accordance with age and physiological status some population groups have increased daily physiological requirement for absorbed Zn. The new synthesis of tissue during growth, pregnancy and lactation period requires higher daily intakes of zinc [SHILS et al., 2005]. Therefore, infants, pregnant and lactating women, young children and children recovering from malnutrition belong to those groups with higher Zn requirements and are thus at elevated risk for Zn deficiency.

Some evidence exists for the occurrence of deficiency among each named group in developing countries. Conclusions can be made as to consequences of Zn deficiency during pregnancy on maternal, foetal and infant health.
Zn deficiency results in a high number of adverse effects especially affecting the immune system (**Fehler! Verweisquelle konnte nicht gefunden werden.**).

Table 8: Adverse effects of Zn deficiency

[Mod.: SHILS et al., 2005]

Immune- system dependent	**Other**
▪ General deteriorated immunity	▪ Alleviated skinatrophy
▪ Affected humeral immunity	▪ Lower status of FTS (hormone of the thymus)
▪ Lymphopenie	▪ Stunting
▪ Affected maturation and proliferation of T- cells	▪ Alleviated wound healing
▪ Thymusatrophy	▪ Loss of appetite, loss of smell and taste
	▪ Complications during pregnancy
	▪ Disturbed metabolism of nucleic acids, proteins, carbohydrates, and fat

Zn deficiency can be attributed to five general causes occurring either in isolation or combination. These are inadequate intake, increased requirements, malabsorption, increased losses and impaired utilization [GERSHWIN et al, 2000].

2.5.3. Assessment

The efficient regulation of Zn homeostasis complicates the diagnosis in Zn status in humans - no accepted reliable indicators of individual Zn status do exist [ELMADFA and LEITZMANN, 2004]. Zn does not have a well established index that can be used in the clinical laboratory to evaluate the status. In healthy adults the normal plasma Zn concentration range varies from 12 - 18 µM (8 - 12 µg/dl). Assessing the Zn status on the basis of the activities of Zn containing enzymes (including plasma alkaline phosphatase, erythrocyte Cu/Zn-SOD and lymphocyte 5'-nucleotidase) is discussed controversial. Combinations of indicators may prove the value and give good information about Zn supply [ELMADFA and LEITZMANN, 2003] (Table 9).

Table 9: Assessment of Zinc

Plasma/ Serum level	0.6 – 1.2 mg/l
Blood cells	4 – 7.5 mg/l GSH-Px activity in erythrocytes*:* *1000 – 2000 U/g Hb*
Urine	0.25 – 0.85 mg/l

2.5.4. Source and Recommended Intake

The quality of dietary Zn sources depends on the amount and bioavailability of Zn from the foods. Organ and flesh of mammals, chicken, fish and crustaceans are the richest food sources and because they do not contain phytate, they are particularly good sources of absorbable Zn.

Foods of vegetable origin tend to be low in Zn, except for germs of grains and seeds, such as nuts and legumes. Availability of Zn from those foods is reduced (below 20 %) by the presence of phytic acid [GIBSON and FERGUSON, 1998].

Mixed animal and vegetable protein diets with a "phytic acid to Zn" molar ratio below 15 show a moderate Zn availability, cereal or legume based diets low in animal protein and with a molar ratio of "phytic acid to Zn" >15 result in a low

availability and have been associated with a suboptimal Zn status [GIBSON and FERGUSON, 1998].

Iron in pharmacological dose decrease Zn absorption, whilst Zn deficiency decreases iron absorption. High doses of Zn inhibit copper absorption. Alcohol abuse affects Zn absorption in a negative way.

Generally the absorption of Zn adapts to physiologic needs which are increasing during lactation and decreasing with aging.

Some conditions of stress, like infectious diseases, may alert absorption efficiency. Losses of Zn include menstruation (0.1 - 0.5 mg total) and parturition (100 mg/foetus, 100 mg/placenta).

Lactation produces Zn losses of 2.2 mg/d at four weeks and 0.9 mg/d at 35 weeks. Therefore the recommended daily intake of Zn is higher for lactating than non-lactating young women (**Fehler! Verweisquelle konnte nicht gefunden werden.**10).

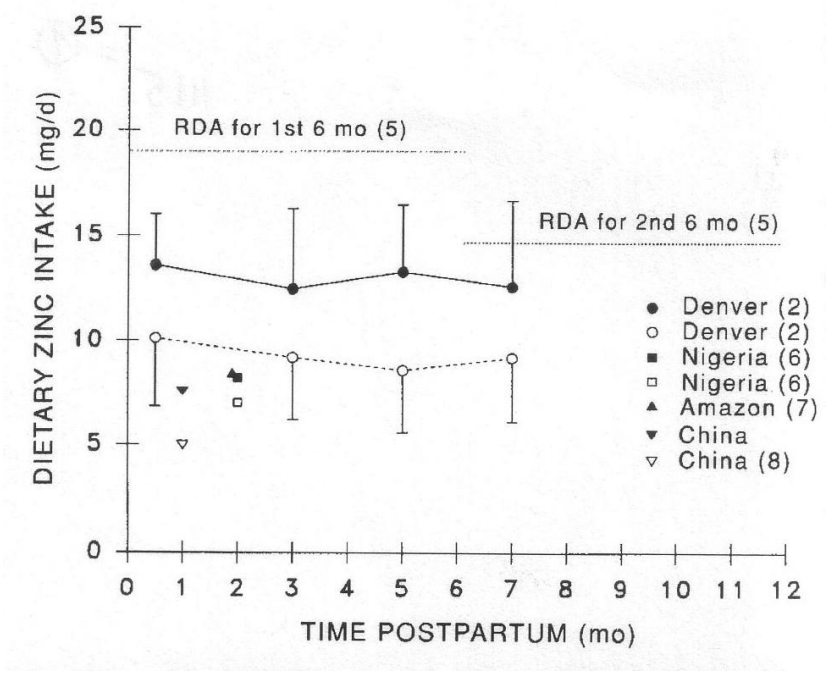
Some women do not produce milk with normal amounts of Zn (134 µg/ 100 g human milk), but this defect has not been found to be due to an altered expression of the transporter Zn-T4 in the mammary gland [SHILS et al., 2005]. The availability of Zn from human breast milk is much better than the availability from cow milk. An exclusive breastfed infant takes up to 1 mg Zn/day and so it is well supplied (**Fehler! Verweisquelle konnte nicht gefunden werden.**10). In formula food the concentration of Zn is higher, but breast milk includes a ligand, which provides an ideal uptake of Zn by the infant.

Table 10: Recommended Intake of Zinc

[Mod.: D_A_CH, Referenzwerte für die Nährstoffzufuhr, 2001]

Lactating women	11 mg/d
Infants 0- 4 months	1 mg/d
Infants 4- 12 months	2 mg/d

During lactation the requirements for Zn are quantitatively greater than those during pregnancy. In the early weeks postpartum excretion in the milk is ~30 - 40 µmol Zn/d; this declines to ~15 µmol Zn by two to three months postpartum [KREBS, 1998]. As shown in **Fehler! Verweisquelle konnte nicht gefunden werden.** lactating women do have a higher recommended intake than non lactating women.



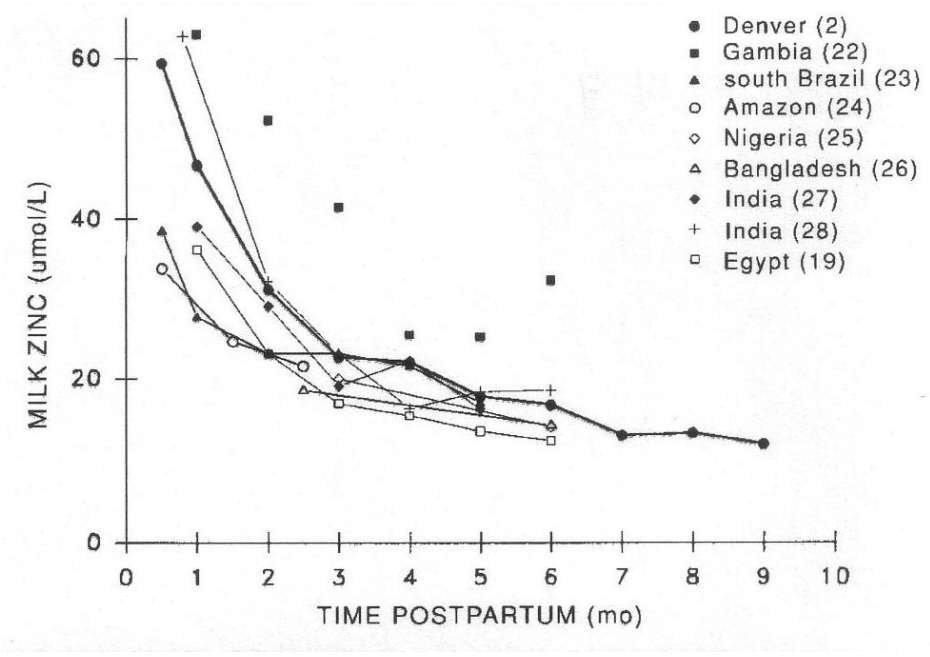
Solid symbols = lactating women

Open Symbols = non lactating women

Figure 4: Comparison of dietary Zn intakes for lactating and non lactating women in the United States and in developing countries **[Mod.: KREBS, 1998]**

Numerous cross- sectional and limited longitudinal studies showed no consistent correlation between maternal dietary Zn intakes and milk Zn concentrations when maternal intakes are relatively high. Data on milk Zn concentrations from women in developing countries were obtained primarily by using cross- sectional design methods and often with wide ranges of time over which sampling had been undertaken. Data from several developing countries

and longitudinal data from well- nourished US women are compared in *Fehler! Verweisquelle konnte nicht gefunden werden.*.



Connected symbols represent longitudinal
or semi longitudinal data.

***Figure 5: Comparison of milk Zn concentrations from lactating US women and women in developing countries** [Mod.: KREBS, 1998]*

Although the outcome of various studies dealing with the relationship between maternal Zn status and milk Zn concentration is inhomogeneous, the majority of preceded investigations support the hypothesis that low dietary Zn intake is associated with lower milk Zn concentrations.

2.5.5. Zinc and HIV/ AIDS

Zinc plays an important role in HIV infection. Its concentration in the human body can influence the transcription of numerous genes implicated in the immune system. HIV infected people are very often delicate to Zn deficiency because of malabsorption, loss of nutrient intake or diarrhoea. Patients with

AIDS exhibit clinical symptoms similar to those associated with Zn deficiency, including immune deficiencies, impaired taste and appetite, decreased food intake, gastro- intestinal malfunction with diarrhoea, alopecia, epithelial lesions and hypogonadism.

30 to 50 % of adult HIV infected patients in developed and nearly 90 % in developing countries suffer from diarrhoea, regularly. The high incidence of diarrhoea among HIV patients in developing countries may not only be attributable to the higher exposure to pathogens due to the lack of sanitary facilities and clean water, but also to a cereal based diet with a lower amount of absorbable Zn.

In Descriptive and analytic epidemiologic study designs have been used to examine the role of Zn status in HIV/ AIDS in humans. Low plasma Zn was observed in up to 96 % of AIDS/ AIDS related complex patients [BEACH et al., 1992]. BODGEN et al. [2000] and MOCCHEGIANI et al. [1999] reported of a significant correlation between plasma Zn levels and immune parameters such as CD4+ T- cell counts and HIV RNA levels when data from HIV positive subjects were evaluated cross sectional.

Zambia, the effects of Zn, Se, and vitamins A, C, E on health parameters in HIV patients were assessed in a randomized, placebo–controlled trial among 106 subjects with persistent diarrhoea [KELLY and MUSONDA et al., 1999]. The findings of this study indicated no effect of the micronutrient supplement containing 200 mg of Zn given daily for 2 weeks (in total the patients were observed for 12 weeks) on diarrhoea, morbidity and mortality.

LAI et al. [2001] found a connection between a low serum Zn level and the progression of clinical symptoms and an increased mortality among HIV patients. Further clinical trials investigating the effect of supplemented Zn on the health status of HIV patients also noticed a decreased number of opportunistic infections, especially in *pnemocystis carinii* and *candida* infections [MOCCHEGIANI et al., 2000].

In many different tissues, Zn constitutes are part of the defence system against oxidative stress because it binds to thiol groups and prevents their oxidation. Zn also induces the synthesis of metallothioneins, which are important ROS scavengers in human body.

Cu/Zn-SOD is essential for the antioxidant defence system. Zn deficiency in HIV-1 infection may compromise the production of Cu/Zn-SOD and adversely affect the antioxidant response of the overproduction of free radicals and lipid peroxides observed in the early disease.

In vitro Cu/Zn-SOD has been demonstrated to reduce HIV-1 replication whereas oxidative stress stimulates the replication of HIV-1 and increases the individual's viral load [FAVIER et al., 1994]. However, it should be noted that the HI virus needs Zn for its replication and therefore a long term supplementation can cause negative effects (Zn enhances enzymatic activity of viral integrase and its zinc fingers) [LEE et al., 1997].

The main reason for not suggesting Zn supplementation is the discovery that HIV- Tat protein and HIV nucleocapsid protein NCp7 are strongly Zn dependent with high binding affinity and both proteins are relevant for HIV replication [MELLY et al., 1996]. Thus, major Zn bioavailability by supplementation with Zn might induce a major activation of Tat and NCp7 proteins resulting in consequent quick viral HIV replication and rapid HIV progression.

Zn metabolism is also altered in the late stages of AIDS. Some articles have reported a beneficial effect of supplementation with Zn on the recovery of immune efficiency in stage IV of HIV infection [ZAZZO et al., 1989]. Most of the studies have been carried out in small cohorts of concerned individuals; they are suggestive to give a dominant role to Zn in HIV infection to thwart the appearance of opportunistic infections [ISA et al., 1992; BAUM et al., 1995].

HIV-1 infected individuals are particularly susceptible to Zn deficiency, but on the other hand, excessive Zn may stimulate HIV-1.

2.5.6. Zinc and MTCT

Zn deficiency may have detrimental effects on important predictors of vertical transmission, such as clinical, immunological or viral stage of HIV among pregnant and lactating women. Zn may influence the maintenance of the child's membrane integrity, which could lower a women's risk of transmitting the virus [O'DELL, 2000]. By contributing to intestinal mucosal integrity and natural immunity, an adequate maternal Zn status may protect the breastfed child from contracting the virus from the mother. Additionally, Zn may prevent transmission by decreasing the risk for prematurity and low birth weight, which is for sure an intermediate risk factor for MTCT.

2.6 Situation in Kenya

Kenya has an estimated population of 34 million people - about 58.2% of them live with less than US$ 2 a day. Although Kenya has been demonstrated a clear trend of decreasing HIV prevalence over the past several years, there are still 26% of the population who are living with HIV/ AIDS. During the last three years, critical HIV services have been scaled up and as a result general awareness and knowledge of HIV transmission are nearly universal. The downward trend was especially profound in the urban sites of Busia, Meru, Nakuru and Thika, where the prevalence declined from 28% in 1999 to 9% in 2003 among 15-49 year old women attending antenatal clinics [HALLETT, 2006].

Up to 40 % of pregnant women attending antenatal care clinics in 2004 benefit from prevention of MTCT services. In a study by KIARIE et al. [2004] carried out in the Kenyatta National Hospital in Nairobi the feeding practice of HIV positive women was determined. 79 % of HIV-1 infected women were practicing exclusive replacement feeding or exclusive breastfeeding at the first week. Many of the exclusive breastfeeding women started mixed feeding afterwards. Exclusive breastfeeding was associated with younger age and better knowledge about prevention of MTCT. This led to the conclusion that younger women may be more open receiving education and advice from clinical staff [KIARIE et al., 2004].

3. Research Methodology

3.1. Study Design and Data Collection

The data collecting took place in the Provincial General Hospital, Nakuru, Kenya. Over a period of three weeks 81 lactating women (between 16 and 38 years old) were interviewed face- to- face by trained, Kiswahili speaking staff about their sociodemographic status, medical condition and eating habits. For more detailed information about their eating habits a 24- Hour Recall was used. Medical staff took 5 ml venous blood from each respondent for the determination of the Zn and Se status in the laboratory. The blood samples were transferred into heparin coated tubes.

A total of 79 subjects took part in this study. 40.5% (n= 32) of all recruited women were HIV positive and 59.5% (n= 47) were HIV negative (**Fehler! Verweisquelle konnte nicht gefunden werden.**).

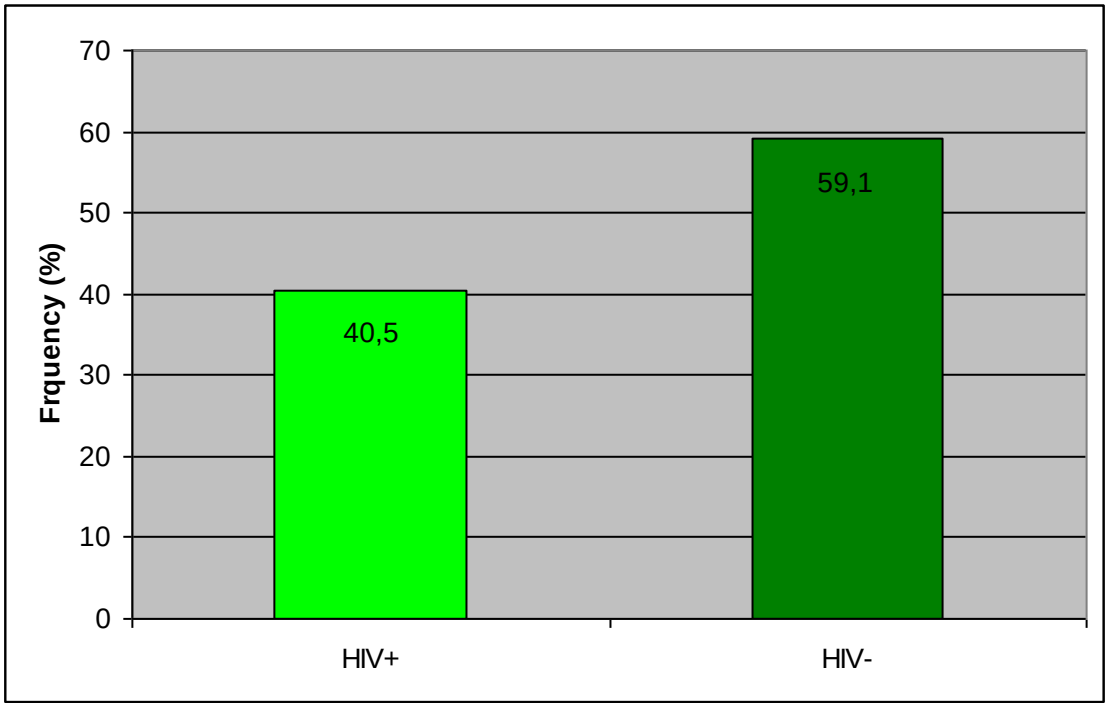


Figure 6: Respondents (total=79; HIV+= 32; HIV-: 47) and their HIV Status

All the infected respondents were asymptomatic at the time of data collecting. The negative group acted as control. The respondents had to prove their HIV status with the Antenatal Cards from the Ministry of Health of Kenya. Infant HIV status could not be determined under the age of 18 months, because the available tests could only determine the status onwards this age. The children had to have a Child Health Card from the Ministry of Kenya (Kenya Expanded Programme on Immunization).

Pilot testing of the questionnaire was done at a community eastern of Nakuru, where women had similar characteristics as those attending the Provincial General Hospital.

The study was approved by the Ministry of Science and Technology, Kenya, the Ethics Committee of the Nakuru General Hospital, Kenya and the Ethics Committee of the Medical University of Vienna and the Vienna General Hospital- AKH, Austria.

3.2. Anthropometry

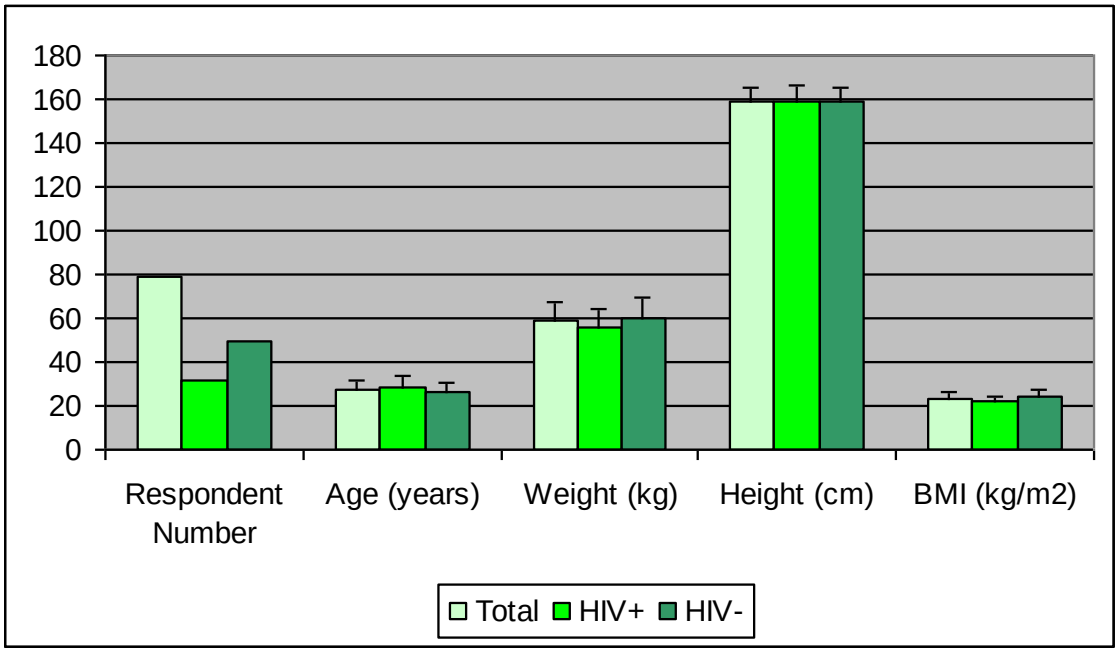
The mothers weight was measured with a Camry scale (0 kg - 120 kg) and their lengths were taken with a stadiometer (0 cm - 207 cm). The women wore light clothes. The lengths was taken without shoes and the women were asked to stand feet facing the front, shoulders and heels touching the back of the stadiometer bar (**Fehler! Verweisquelle konnte nicht gefunden werden.**).

For calculating the mothers Body Mass Index (BMI) the following formula was used:

$$\text{BMI } [kg/m^2] = \text{weight (kg)} / \text{height}^{\,2}\ (m^2)$$

The body weights of the subjects were classified in under-, normal, overweight and obesity (**Fehler! Verweisquelle konnte nicht gefunden werden.**11).

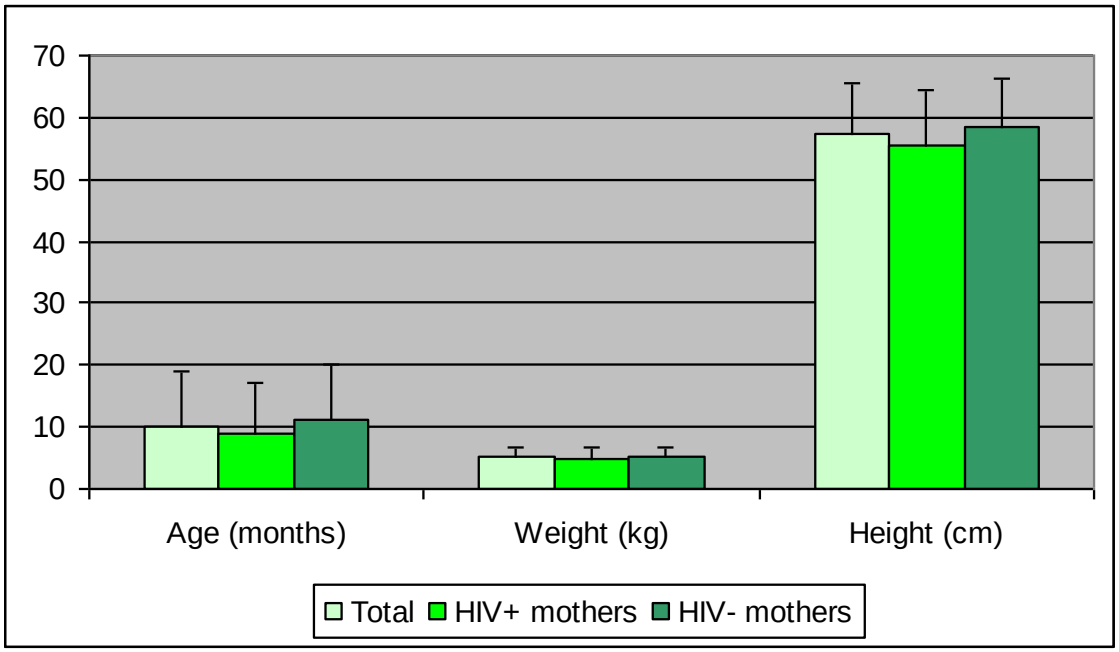
Table 11: BMI classification	
<18.5	Underweight
18.5-24	Normal Weight
24-30	Overweight
>30	Obesity



	Respondent Number	**Age (years)**	**Weight (kg)**	**Height (cm)**	**BMI (kg/m^2)**
Total	79	27±5	59±8	159±6	23±3
HIV+	32	28±6	56±8	159±7	22±2
HIV-	47	26±5	60±9	159±9	24±3

Figure 7: Characteristics of the Respondents

The infants and children's weight was measured with a Salter England scale (Model 2356S; 0 kg - 25 kg) or a Seca 725 1021009 (0.5 kg- 16 kg) and their lengths was measured with a Shorr Board (0 cm- 130 cm). They wore light clothes or a light blanket. For lengths measurement they wore no socks and shoes.



	Age (months)	Weight (kg)	Height (cm)
Total	10±9	5.2±1.6	57.5±8.2
HIV+ mothers	9±8	5±1.8	55.3±9.3
HIV- mothers	11±9	5.3±1.4	58±7.5

Figure 8: Infants (n=79) characteristic

3.4. Analysis of Selenium and Zinc Status

3.4.1. Blood Preparation

The venous blood samples were stored cool and transported to the Biochemistry Laboratory at Egerton University within four hours after being taken. The samples were transferred into Pyrex centrifugal tubes and centrifugated for 10 minutes at 3000 RPM in a bench top centrifuge (Model: Hettich EBA 3S). Afterwards the plasma was taken off and the red blood cells were left in the tubes. The erythrocytes were washed for three times with an isotonic phosphorus buffer. Afterwards the erythrocyte suspensions were frozen (-20°C) in 500 µl Eppendorff tubes.

3.4.2. Analysis of Glutathione Peroxidase (GSH-Px) EC 1.11.1.9 Activity in Erythrocytes

Method

In a first reaction of this method [BEUTLER, 1984] glutathione peroxidase (GSH-Px) catalyzes with hydroperoxides the oxidation of reduced (GSH) to oxidized glutathione (GSSG). In a second reaction the GSSG changes into GSH in the presence of $NADPH_2$ and glutathione reductase (GR). The decrease of $NADPH_2$ is proportional to GSH-Px activity and can be measured photometrical.

$$2\ GSH + ROOH \xrightarrow{GSH\text{-}Px} GSSG + ROH + H_2O$$

$$GSSG + NADPH_2 \xrightarrow{GR} 2\ GSH + NADP^+$$

Material and Equipment

Reagents:		
Glutathione (reduced)	Roche	Nr.127736

Glutathione Reductase (GR) fr. yeast	Roche	Nr.105678
NADPH Tetrasodiumsalt	Roche	Nr.107824
t-Butylhydroperoxide, 70% (BHP)	Fluka	Nr.199955
Tris(hydroxymethyl)-aminomethan-Hydrochloride	Fluka	Nr.93350
Sodium-EDTA	Riedel de Haën	Nr.27285
Potassiumhexacyanoferrat	Merck	Nr.4973
Potassiumcyanid	Aldrich	Nr.20781-0
aqua dest.		

Solutions:

GSH-Solution [1 M]: 61.46 mg Glutathione in 2 ml aqua dest.

GR-Solution [10 U/ml]: 50 µl GR solution in 2950 µl aqua dest.

NADPH-Solution [2 mM]: 3.48 g NADPH in 2 ml aqua dest.

BHP-Solution [7 mM]: 20 µl t-BHP in 20 ml aqua dest.

Tris-Buffer [1 M]: 15,76 g Tris-HCl + 186 g Sodium-EDTA in 100 ml aqua dest., pH 8.0

Haemoglobine Reagent: 198 mg Potassiumhexacyanoferrat + 49 mg Potassiumcyanid in 1l aqua dest.

Equipment:

pH-Meter (Model: EZDO, PL- 500)

Water bath (Model: ub5- nüve)

Scale (Model: Citizen, CY220; 0.01 g- 220 g)

UV/ VIS Photometer (Model: CE 2021 Cecil, 2000 Series)

Analysis

For the analysis of GSH-Px activity 100 µl of an erythrocyte suspension are haemolyzed by adding 900 µl aqua dest. Following pipette pattern was used:

Reagents	*Blank [µl]*	*Sample [µl]*
aqua dest.	670	660
Tris-Buffer	100	100
GSH	20	20
GR	100	100
NADPH	100	100
Haemolysat	-	10
mix well and incubate 10 Minutes at 37°C		
t-BHP	10	10

After adding t-BHP the decrease of the extinction (340 nm) was measured over a time period of 90 seconds.

The haemoglobin concentration was determined photometrical (546 nm) after mixing 100 µl erythrocyte suspension and 5 ml haemoglobin reagent.

Evaluation

The calculation of the GSH-PX activity is based on the Lambert-Beer´s Law:

$$E = \varepsilon * c * d$$

E = extinction, ε = molar extinction coefficient [$1mol^{-1}.cm^{-1}.l$], c = concentration/ activity, d = width of the cuvette [cm] =1

Following formula was used for calculating the activity of the erythrocyte GSH-Px:

Activity of GSH-Px: $A = \Delta E * V_c / (6.22 * V_H)$

A = activity of GSH-Px, ΔE = decrease of the extinction/min, V_c = total volume, V_H = volume of the haemolysate, 6,22 = molar extinction coefficient [$lmmol^{-1}cm^{-1}$] of NADPH at 340 nm

Activity of GSH-Px: A * 100/Hb [g/dl] = U/g Hb

A = activity of GSH-Px, Hb = concentration of haemoglobin, U = units

The coefficient of variation of this method was 1.86 %.

3.2.1. Analysis of the Superoxide Dismutase Activity (SOD) EC 1.15.11 in Erythrocytes

Method

The activity of SOD was measured using the method of MARKLUND and MARKLUND [1974], modified by BEUTLER [1984]. SOD is able to stop the autoxidation of pyrogallol. Pyrogallol autoxidates in watery solution very fast and gives a yellowish colour which concentration is measured photometrical.

$$2\ O_2^{\bullet -} + 2\ H \xrightarrow{SOD} O_2 + H_2O_2$$

Material and Equipment

Reagents:		
Pyrogallol	Riedel de Haën	Nr.16040
Ethanol p.a.	Riedel de Haën	Nr.32221
Methanol p.a.	Merck	Nr.106009.2500
Chloroform	Riedel de Haën	Nr.32286
HCl 37%	Riedel de Haën	Nr.30721
Tris(hydroxymethyl)-aminomethan-hydrochlorid	Fluka	Nr.93350
Sodium-EDTA	Riedel de Haën	Nr.27285
Potassiumhexacyanoferrat	Merck	Nr.4973
Potassiumcyanid	Aldrich	Nr.20781-0

Solutions:

Pyrogallol-Solution: 126.11 mg Pyrogallol + 82 µl HCl in 100 ml aqua dest.

Tris-Buffer [1 M]: 15.76 g Tris-HCl + 186 g Sodium-EDTA in 100 ml aqua dest.; pH 8.0

Haemoglobin Reagent: 198 mg Potassiumhexacyanoferrat + 49 mg Potassiumcyanid in 1l aqua dest.

Equipment:

pH-Meter (Model: EZDO, PL- 500)

Centrifuge, 3000 RPM (Model: Hettich EBA 3S)

Water bath (Model: ub5- nüve)

Scale (Model: Citizen, CY220; 0.01 g- 220 g)

UV/ VIS Photometer (Model: CE 2021 Cecil, 2000 Series)

Analysis

300 µl of the erythrocyte suspension are haemolyzed by adding 450 µl aqua dest.. 500 µl of this haemolysate are mixed with 3.5 ml aqua dest., 1 ml ethanol and 600 µl chloroform, vortexed for 60 seconds and centrifuged for 2 minutes (3000 U/min). The upper phase of the conditioned sample is used for this pipette pattern:

Reagent	BL [µl]	A_1 [µl]	A_2 [µl]	A_3 [µl]	A_4 [µl]	A_5 [µl]
Tris-Buffer	100	100	100	100	100	100
aqua dest.	880	840	820	780	730	680
Sample	0	40	60	100	150	200
mix and incubate 10 minutes at 25 °C						
Pyrogallol-Solution	20	20	20	20	20	20

BL…blank; A…sample

After addition of pyrogallol the extinction (320 nm) is measured over a time period of 3 minutes.

Determination of haemoglobin s. 0.

Evaluation

ΔE/min is calculated for all six concentration steps of each sample using a linear regression.

The concentration of the sample (x) at the 50% inhibition of the autoxidation of pyrogallol is calculated by using this formula:

$$y = k * x + d \qquad x = (y - d)/k$$

x = concentration of the sample at 50% inhibition of the pyrogallol autoxidation
y = (ΔE/min)/2 of the sample blank
k = gradient of the curve
d = point of intersection of the curve with the y-axis

The activity of the erythrocyte SOD has been calculated as followed:

$$A = GV/(PV * x)$$

A = enzyme activity, GV = total volume = 5.6 ml, PV = volume of the sample = 0.5 ml, x = concentration of the sample at 50% inhibition of pyrogallol autoxidation

Activity of SOD: A * 100/Hb [g/dl] = U/g Hb

A = activity of SOD, Hb = concentration of haemoglobin, U = Units

The coefficient of variation of this method was 1.9%.

3.5. Evaluation and Statistical Analysis

Statistical Package for Social Sciences (SPSS) for Windows (Version 15.0) was used for all statistical analysis. Distribution of the data was tested by implementing the *Kolmogorov-Smirnov Test*. Summary statistics (frequencies) were used to describe the data and trends, independent sample t-tests were used to describe differences between the HIV positive and HIV negative women for the various factors. Partial and bivariate correlations were used to describe coherences between different factors.
For assessing the mothers' nutrient intake, 24-Hour Recalls were evaluated by the Nutrisurvey program Kenya (www.Nutrisurvey.de).

4. Results and Discussion

4.1. Sociodemographic data

Demographic Data

Demographic data showed that 65% of all women participating in this study lived out of town, 10% in an urban area, 10% in a peri- urban area and 15% in a rural area of Nakuru.

Within the HIV positive women 37% lived out of town and 3% lived in urban areas. 16% lived in peri- urban and only 9% in rural areas.

In the HIV negative group 65% lived out of town, 10% lived in urban areas, 15% in rural areas, and 10% in peri- urban areas (**Fehler! Verweisquelle konnte nicht gefunden werden.** 9).

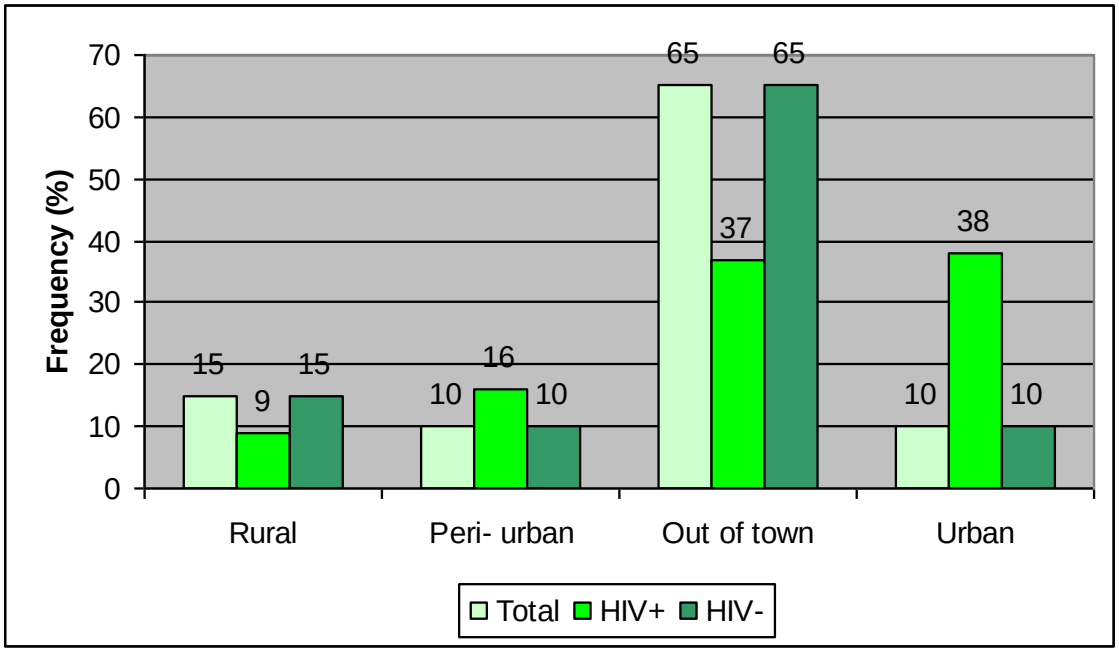


Figure 9: Origin of the Respondents

Refuge/Migration

35% (HIV+: 36%, HIV-: 64%) of the study participants had to leave their homes during the post election violence in January and February 2008. This fact was

also recorded during this study because living in a place of refuge is known to have a great influence on the nutritional status of humans. There was no statistical significance between the infected and uninfected women concerning this situation.

Marital Status and Age

More than three quarter (78%) of all respondents were married, 14% were single living mothers, 4% lived divorced or separated from their husbands and another 4% were widowed.

In the HIV positive group 56% of the women were married, 26% lived in single households, 9% were separated or divorced from their partners and 9% were widowed.

The marital status of HIV negative women was quite different from that of HIV positive participants. In contrast to the HIV positive group, 94% of all HIV negative respondents were married and just 6% were single living mothers (Figure 10).

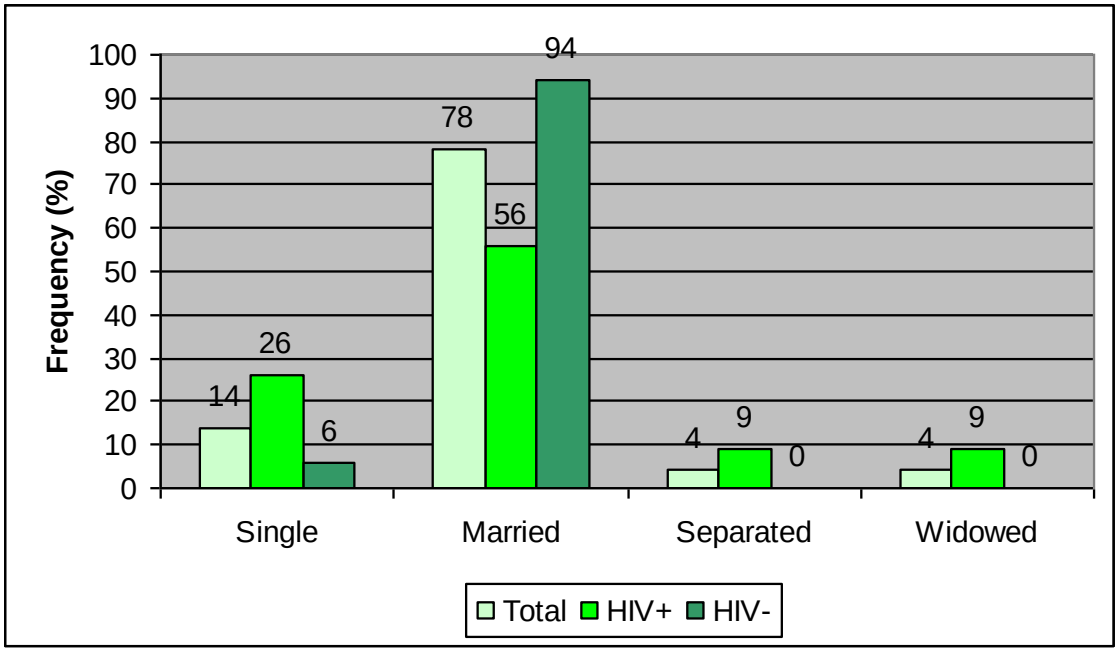


Figure 10: Marital Status of the Respondents

There are big differences in the marital status of HIV positive and HIV negative women, but there is no statistical significance (p< 0.106).

The evaluation of these sociodemographic parameters shows, that more than half of the investigated women were living out of town, were middle aged (total: 27±5 y; HIV+: 28±6 y; HIV-: 26±5 y) and married (Figure 11).

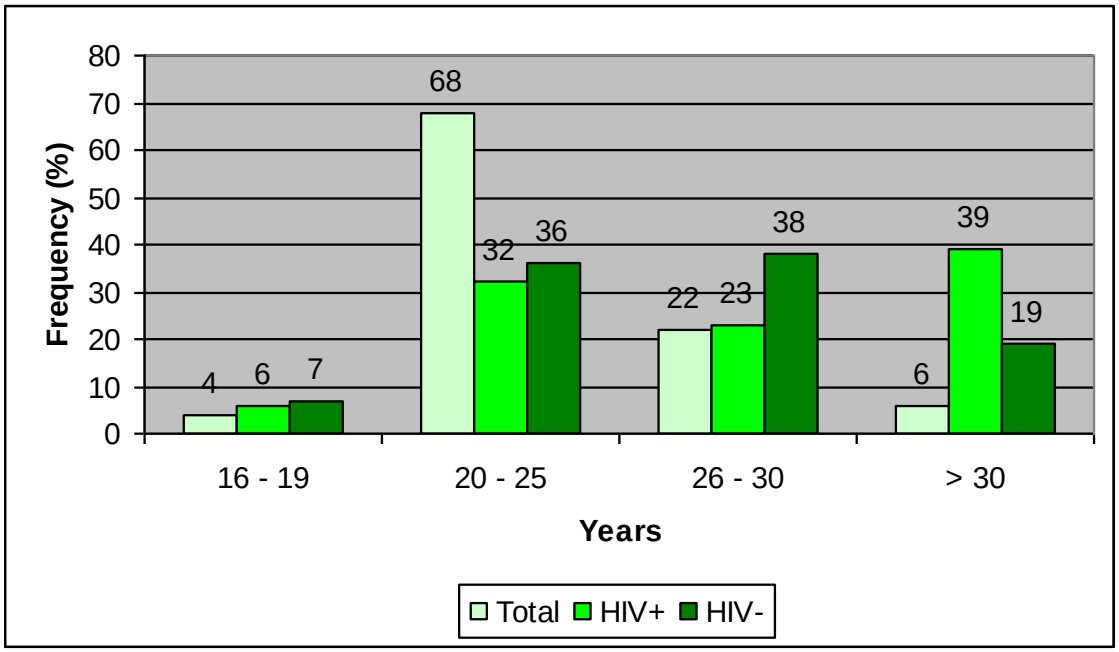


Figure 11: Age (years) of the Respondents

Education

96% (HIV+: 38%; HIV-: 62%) of all investigated women participating in the study had at least basic education (primary school), 47% (HIV+: 22%; HIV-: 78%) had a higher (secondary or tertiary) graduation and only 4% (HIV+: 4%; HIV-: 0%) did not have any school leaving certificate.

This means that 49% of all interviewed women had a school leaving certificate of primary school, 37% of secondary school, 10% had a tertiary education (university degree) and only 4% had not got any education.

However, data pointed out that there was a big gap between the HIV positive and HIV negative group in terms of education. HIV negative women had significant ($p < 0.05$) higher school leaving certificates than HIV positive women. When the education factor was broken down according to HIV status following outcome could be observed:

In the positive group 9% had no education at all, 66% attended to primary school, 16% to secondary school and 9% to tertiary school. Among the HIV negative participants the majority had a graduation of secondary school (51%). 38% had attended primary school and even 11% had a tertiary education (Figure 12).

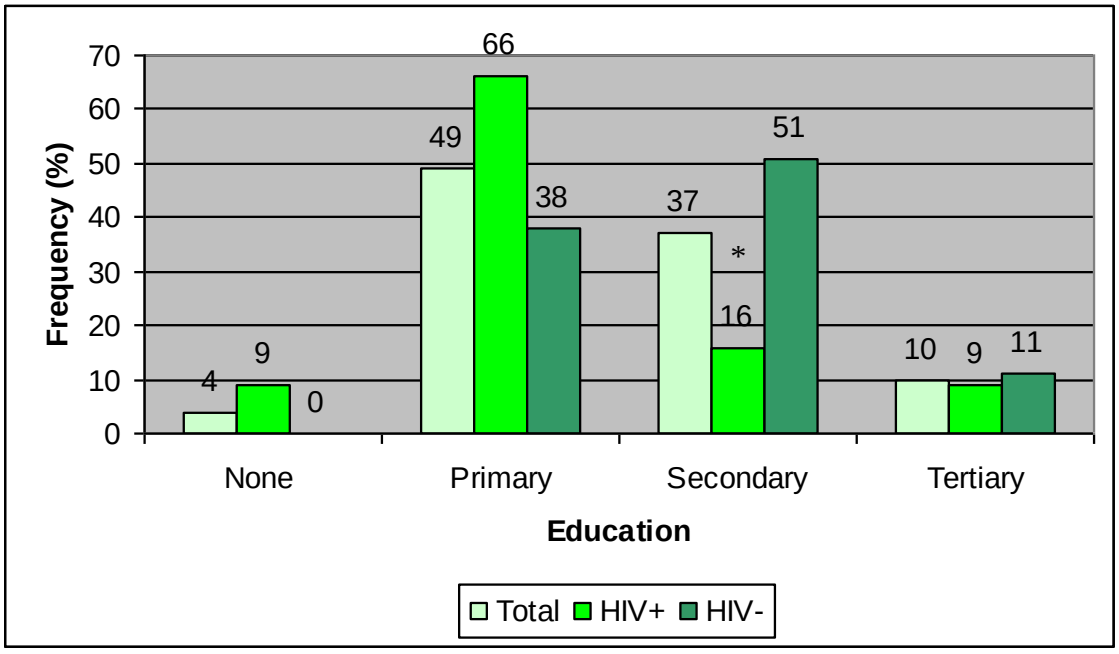


Figure 12: Educational Status of the Respondents (*: HIV+< HIV-; p>0.05)

Statements and Discussion: Sociodemographic Data

The impact of demographic data is very important for an adequate food supply. Women, who are married, do have more money to buy food and have more time to farm their fields, than women who are single living, divorced or widowed and have to do both, earning money and farming.

In a large epidemiologic study DARMON and DREWNOWSKI [2008] showed that diet quality followed a socioeconomic gradient. Higher-quality diets were associated with greater affluence energy-dense diets that are nutrient-poor were preferentially consumed by persons of lower socioeconomic status and of more limited economic means. They demonstrated that whole grains, lean meats, fish, low-fat dairy products, fresh vegetables and fruit were more likely to be consumed by groups of higher socioeconomic status. In contrast, the consumption of refined grains and added fats had been associated with lower socioeconomic status. Although micronutrient intake and diet quality are affected by education and origin, little evidence indicated that mentioned factors affected either total energy intakes or the macronutrient composition of the diet.

Also education is a very important factor influencing the nutritional status of humans. Well educated women do have a better knowledge about adequate nutritional supply, HIV/ AIDS and its prevention of transmission. In order to inform about general health, MTCT, HIV/ AIDS or nutrition it is very useful, if the women are able to read and write. So, the educational standard of a pregnant/ lactating woman is very important for the implementation of possible interventions, training programs and further care.

HIV Status

Among both, urban and rural residents the seroprevalence was found to be rising significantly ($p<0.01$) with increasing educational attainment. Further, it also shows the likelihood of being infected is slightly higher among unmarried women than married. Within the HIV positive group 26% were single living mothers whereas in the HIV negative group only 6% were single living women. FYLKESNES K et al. [1997] confirmed this result in a Zambian study investigating this impact on the HIV status of childbearing women.

Infants and HIV status

Within the HIV positive group 28% of the children were known to be HIV positive. Because of the difficulty of HIV determination in infants, it could be

possible, that the named number is higher and the Number of HIV negative infants is lower than 72%.

44% of the HIV positive mothers within the HIV positive infant group chose the exclusive breastfeeding option and 56% the mixed feeding option.

State of Health

The HIV respondents were asked to talk about their complaints and pains like headache, dizziness and tiredness (Table 12). In total 39% (HIV+: 26%; HIV-:74%) of the study participants recorded to feel free of pain, more than a quarter (27%; HIV+: 33%; HIV-: 67%) of all respondents complained about one disease, 16% (HIV+:61%; HIV-: 39%) did have two, 10% (HIV+: 75%;HIV-: 25%) three and 4% (HIV+: 67%; HIV-: 33%) did have four illnesses. Only 3% (HIV+: 50%; HIV-:50 %) complained about five and 1% (HIV+: 0%; HIV-: 1%) about more than five health problems (Figure 13).

Table 12: Frequency of Complaints

	Total (%)	**HIV+ (%)**	**HIV- (%)**
Tiredness	20	44	9
Rash	5	9	2
Dizziness	13	9	15
Nausea	10	19	4
Fever	5	3	6
Diarrhoea	2	3	2
Vomiting	2	6	0
Swelling	2	3	2
Weakness	13	22	6
Allergy	11	9	13
Cold	24	19	28
Other	17	22	15

If HIV positive and HIV negative women were broken down to separate groups, a quarter (25%) of all HIV positive subjects did not have any pain. 22% did have one, 25% two, 19% three, 6% more than and 3% recorded to have more than five complaints. Tiredness (44%), weakness (22%) and colds (19%) were the most common illnesses within the HIV positive group.

Among the HIV negative group almost half (49%) of the subjects were free of complaints, 30% of these participants were affected by one sickness, 11% complained about two, 4% about three, 2% about four and 4% about five or over five health problems. The most common health problems within the HIV negative group were coughs and sneezes (27.7%).

Compared to the HIV negative group, the incidence of health problems like nausea, tiredness or fever etc. was significantly ($p < 0.05$) higher HIV positive study participants.

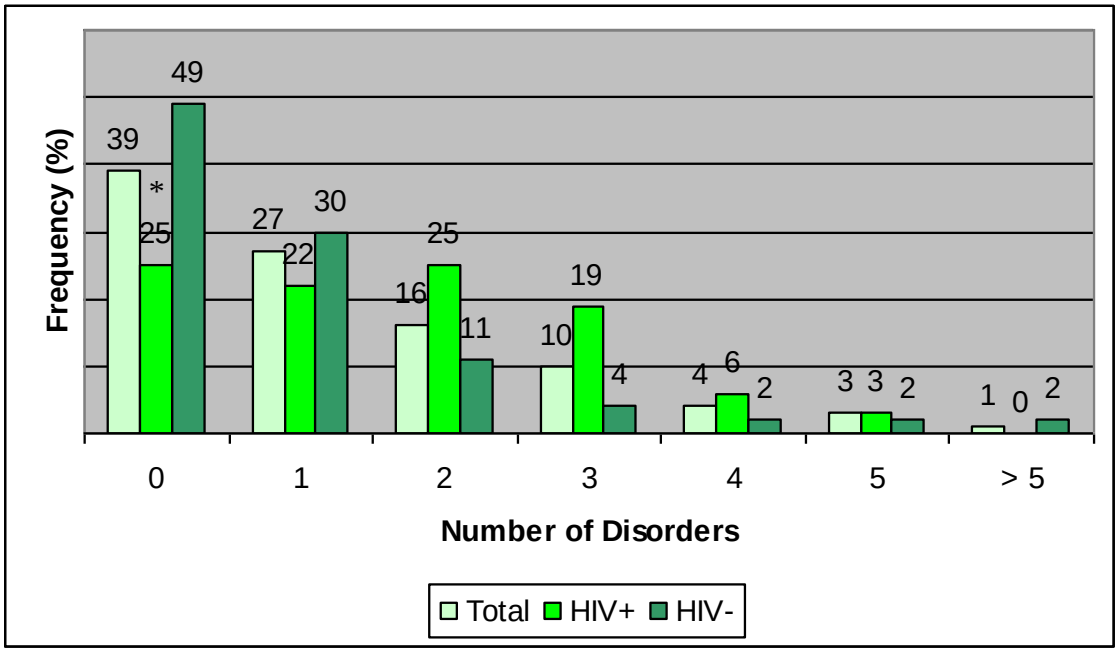


Figure 13: Respondents State of Health ***(*: HIV+ < HIV-; p<0.05)***

Further, HIV positive women recorded significantly ($p<0.05$) more often problems with their breast health and with breast feeding than HIV negative

women however, no significant correlations between the frequency of breast problems, the problems of breast feeding and their HIV status could be found.

Offspring

The results of this study showed that 59% of all participating women had one to two children, 28% three to four, 10% five to six children and 3% had seven to nine children (**Fehler! Verweisquelle konnte nicht gefunden werden.**14).

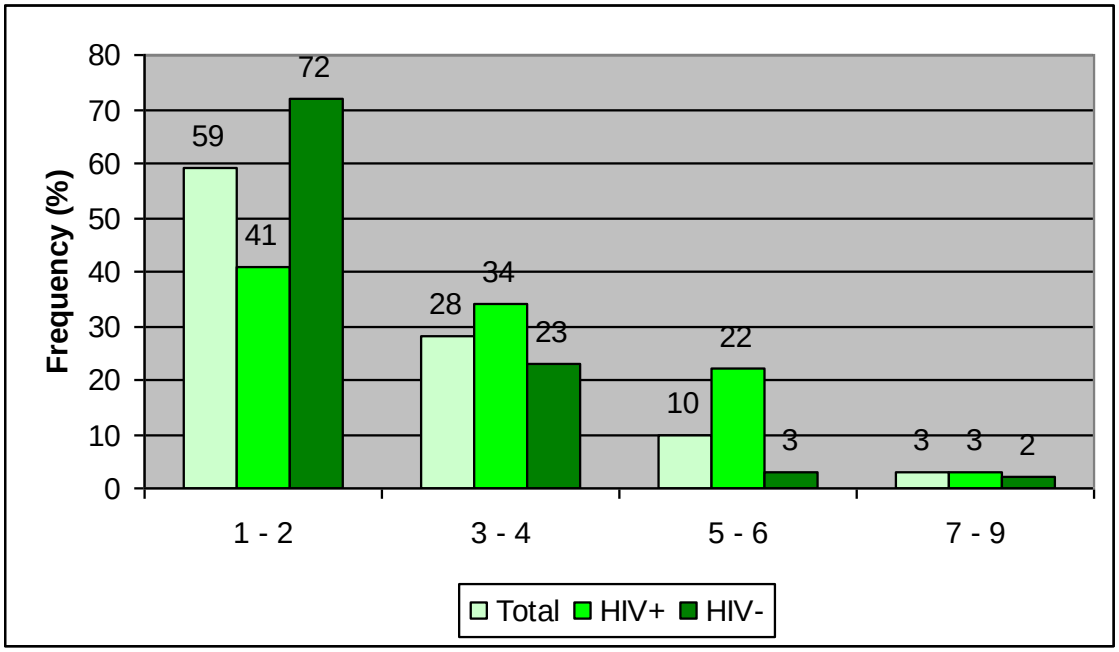


Figure 14: Number of born Children

The results showed that HIV infected women had slightly but not significant more children than HIV uninfected women (HIV+: 3- 4 children = 34%; HIV-: 1-2 children = 72%).

4.2. Anthropometric and Nutritional Data

4.2.1. Anthropometric Data

The average BMI of all recruited women was 23.2±3 kg/m^2 (min.: 18.1 kg/m^2; max.: 36.6 kg/m^2). In the HIV positive group the mean BMI was calculated at 22±2 kg/m^2. The minimum BMI in the HIV positive group was 18.7 kg/m^2 and the highest BMI at 28.3 kg/m^2. In the HIV negative group the mean BMI was calculated at 24±3 kg/m^2, the lowest BMI at 18.1 kg/m^2 and the highest at 36.6 kg/m^2 (Figure 15).

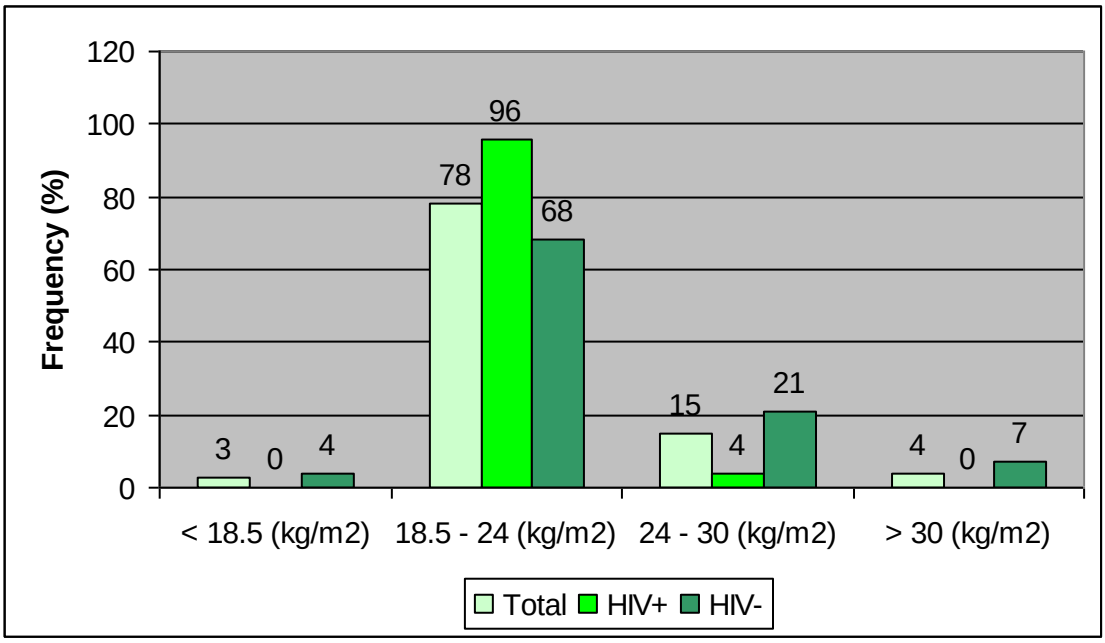


Figure 15: BMI (kg/m2) of the Respondents

Among the HIV positive women the mean BMI was slightly lower (22.3±2.1 kg/ m^2) than the mean BMI (23.7±3.5 kg/ m^2) of the HIV negative control group. There was a statistical significance between the infected and uninfected group ($p < 0.05$; CI 95%).

A differentiation of the BMIs in under-, normal-, overweight and obesity indicated following:

13% (HIV+: 0%; HIV-: 13%) of all recruited women were underweight (had a BMI lower than 18.5 kg/m^2), 78% (HIV+: 48%; HIV-: 52%) were normal weight with a BMI between 18.5 kg/m^2 and 24 kg/m^2. 15% (HIV+: 17%; HIV-: 83%)

were overweight and had a BMI between 24 kg/m^2 and 30 kg/m^2 and 4% of the participants (HIV+: 0%; HIV-: 4%) were obese with a BMI over 30 kg/m^2.
Within the HIV positive group women were either in the normal weight group (96%) or in the overweight group (4%) – none of the subjects could be classified as underweight or obese.
68% of the HIV negative women were in the normal weight group, 21% in the overweight group, 7% were obese and 4% were underweight.

4.1.1. Nutritional Data

24- Hour Recalls

For calculating the nutrients intake the 24-h-Recall method was used. This method reflects just an impression of the nutritional intake of an average day – therefore, the results have to be interpreted with caution. Several factors have been suggested to underlie the problem of inaccurate reports, for example: psychosocial, behavioural and cultural factors, memory disturbance and wrong reporting of food portions. MENNEN et al. [2000] assumed that people from rural Cameroon know better than urban occupants, what they ate, since the food diversity may be lower and most of them are farmers, growing their own food and thus have less chance of underreporting their energy intake by lack of memory.

Energy

Within the respondent group the mean energy intake per day was 2020±609 kcal (min.: 175 kcal; max.: 2897 kcal). The average energy intake in the HIV positive group was 966±496 kcal, but ranged from 175 kcal to 2020 kcal. The mean energy intake within the seronegative control group was 1363±627 kcal, the lowest daily intake among these subjects was 534 kcal and the highest 2897 kcal (Table 12). The statistical analysis indicated a significant ($p<0.05$) difference of the total energy intake between the HIV positive and HIV negative group.

According to the Kenyan Ministry of Health [MINISTRY OF HEALTH, KENYA, 2007] the energy intake of an asymptomatic HIV positive, lactating women should be 2850 kcal/d and for HIV negative, lactating mothers 2640 kcal/d. The evaluation of the data gained in this study clearly showed a great discrepancy (in both, the HIV positive and negative group) between the recommended and the indeed daily energy intake of Kenyan lactating women.

Table 13: Macronutrient intake assessed by a 24-h-Recalls

Nutrient	**Total** **(mean±sd)**	**HIV+** **(mean±sd)**	**HIV-** **(mean±sd)**
Energy (kcal)	2020 ± 609	966 ± 496*	1363 ± 627
Protein (g)	47 ± 33	33 ± 9	55 ± 37
Fat (g)	33 ± 30	24 ± 15	39 ± 33
Carbohydrates (g)	176 ± 80	65 ± 81	190 ± 77
Fibre (g)	22 ± 21	19 ± 13	24 ± 11
Water (g)	1571 ± 691	1141 ± 464	1826 ± 679

*: HIV+ < HIV-; $p<0.05$

Protein

The daily recommended intake of protein for lactating women is 63 g/day [D- A-CH; 2001]. Within the total group investigated in this study the average protein intake was 47±33 g/d (min.:6 g/d; max.: 181 g/d) (Tab. 12) or 16±7.4 E% (Fig. 15). In the HIV positive group the intake was 33±9 g/d (min.: 6 g/d; max.: 46 g/d) - so 15±9 E% of the daily energy intake was derived from protein. The HIV negative group had a protein intake of 55±37 g/d (min: 14 g/d; max.: 181 g/d) resulting in a total of 17±7 E%.

The results of this study showed that the HIV negative group had a (not significant) better protein supply than the HIV positive group. However, the participants of both groups had an average protein intake below the recommended level.

Meat and egg consumption

Meat was consumed by 63% (HIV+: 29%; HIV-: 71%) of all investigated subjects once a week. 28% (HIV+: 68%; HIV-: 32%) of all women ate meat once a month, 8% (HIV+: 34%; HIV-: 66%) every day and 1% (HIV+:0%; HIV-: 1%) never had meat.

The meat consumption within the HIV positive and HIV negative group was quite different, but no statistical significant differences could be found. In the HIV positive group 48% of the women ate meat at least once a month, 45% ate meat once a week and 7% had meat every day.

Within the HIV negative control group almost three quarter (74%) ate meat once a week, 15% ate meat once a month, 9% had it every day and 2% never ate meat at all, because of the cost (Figure 16).

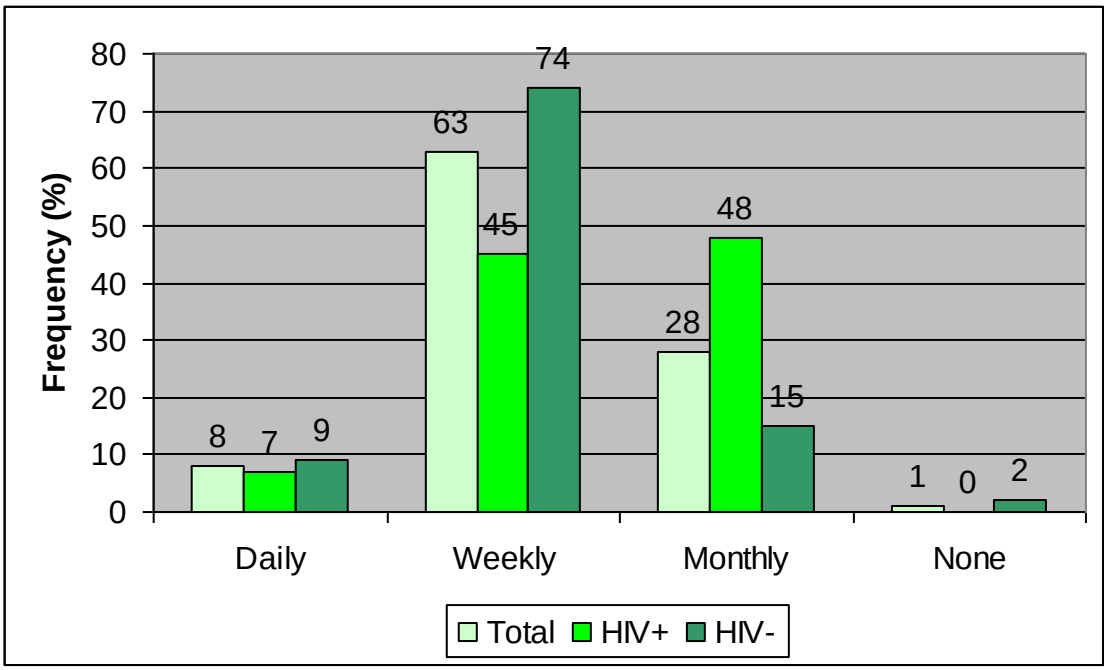


Figure 16: Frequency of Meat Consumption

In the total group egg consumption could be categorized as followed: 55% of all women (HIV+: 36%, HIV-: 64%) stated to eat eggs weekly, 18% (HIV+: 62%, HIV-: 38%) monthly, 6% (HIV+: 25%, HIV-: 75%) daily, 1% (HIV+: 25%, HIV-: 75%) once a year and 20% (HIV+: 44%, HIV-: 56%) of all respondents never

eat eggs. In both, the HIV positive and the HIV negative group this pattern was quite comparable and no significant differences were observed.

Fat

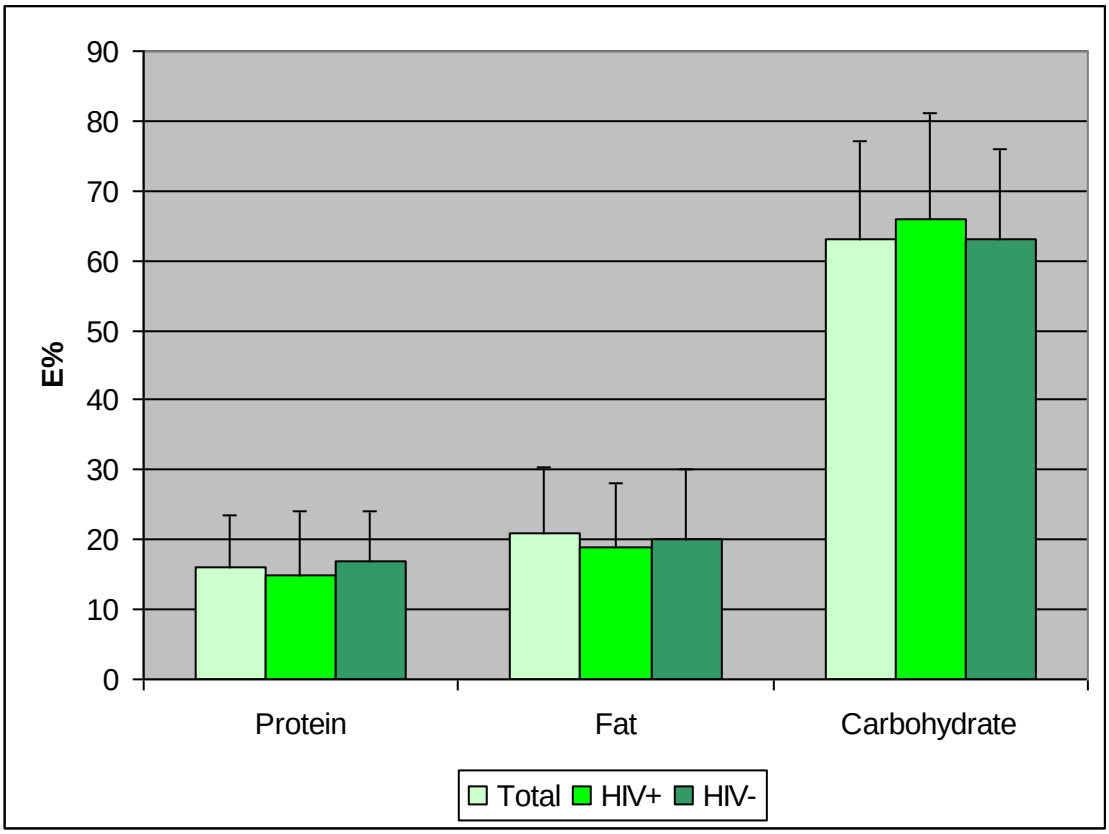
In lactating women the fat intake should be 30-35% percent of the daily energy intake [D- A- CH; 2001]. Within the total group of this study the daily fat intake was 33±30 g/d (Table 12) or 21±9.5 E% (Figure 17), ranging from 2- 151 g/d and 5-53 E%, respectively.

Within the HIV positive group the fat intake was 24±15 g/d or 19±9 E%. In the HIV negative group the mean fat consumption per day was 39±33 g/d (min.: 4 g/d; max.: 151 g/d) or 20±10 E%.

Carbohydrates

The daily recommended intake of carbohydrates for lactating women is set at 50 E% [D- A- CH, 2000].

The intake of carbohydrates observed in this study was quite high (Table 12) – on average 176±80 g/day, resulting in a total of 63±14 E% of daily energy intake. In the HIV positive group the mean carbohydrate intake was 65±81 g/day or 66±15 E%, respectively. Especially in the HIV negative respondent group the average intake (190±77 g/day or 63±13 E%) of carbohydrates was remarkable high and well above the recommendation of 50 E%.



	Protein (E%)	Fat (E%)	Carbohydrates (E%)
Total	16±7.4	20±9.5	62±14
HIV+	15±9	19±9	66±15
HIV-	16±7	21±10	61±13

Figure 17: Energy % of Macronutrients

Dietary Fibre

In lactating women the dietary fibre intake should be 30 g/d [D- A- CH; 2001]. Within the total group of this study the daily fibre intake was 22±21 g/d (Table 12), ranging from 2- 54 g/d.

Within the HIV positive group the dietary fibre intake was 19±13 g/d (min.: 2 g/d; max.: 54 g/d). In the HIV negative group the mean fibre consumption per day was 24±11 g/d (min.: 9 g/d; max.: 54 g/d).

Vitamins and Minerals

It should be noted, that HIV positive women had lower micronutrient intakes than HIV negative women (Table 13). The mean intake of total folic acid was not even half of the recommended (600 µg/d) level (HIV+: 149±97 µg/d; HIV-: 249±169 µg/d). Although it is a policy of the Kenyan government for women to be given folic acid supplement, only 46.7% received at least a combination of supplements with folate or folate on its own [KAMAU- MBUTHIA, 2006].

Also the iron intake was low (HIV+: 8±5 mg/d; HIV-: 13±6 mg/d). Like in Zn intake, the iron bioavailability was limited by the high plant food sources. On the one hand fiber and phytate hinder the iron uptake and on the other hand the bioavailability was limited by the consumption of non-heme iron from plants.

Table 14: Micronutrient intake assessed in a 24 Hour Recall

Nutrient	**Total** (mean±sd)	**HIV+** (mean±sd)	**HIV-** (mean±sd)	**Recommend** D- A- CH
Total folic acid (µg)	212 ± 154	149 ± 97	249 ± 169	600
Vitamin C (mg)	86 ± 74	149 ± 58	102 ± 79	150
Vitamin A (mg)*	1.6 ± 1.1	0.6 ± 0.5	1.6 ± 1.1	1.5
Vitamin E (mg)**	10 ± 8	8 ± 7	11 ± 8	17
Iron (mg)	11 ± 6	8 ± 5	13 ± 6	20
Zinc (mg)	9 ± 5.6	7±4.5	10.3±6	11

*1mg Retinol- aequivalent = 1 mg Retinol

**1 mg RRR -α- Tocopherol- aequivalent = 1 mg RRR- α -Tocopherol = 1.49 IE

In both, the HIV positive and HIV negative group, the intake of Vitamin C (HIV+: 149±58 mg/d; HIV-: 102±79 mg/d) and Vitamin E (HIV+: 8±7 mg/d; HIV-: 11±8 mg/d) could not reach the recommended level.

The Vitamin A intake was quite in the recommended range. In the HIV positive group the intake was 591±485 µg/d and within the HIV negative respondent group 1602±1141 µg/d.

The nutrient intake of the total respondents did not reach the recommended amounts in any nutrient. The HIV positive group's nutrient intake was observed to be lower in all macronutrients and evaluated micronutrients. However, the differences between the investigated groups could not be assessed as significant.

Zinc

The mean Zn intake of all respondent women was 9±5.6 mg/d (min.: 1.4 mg/d; max.: 31.2 mg/d). The mean value of Zn intake within the positive group was 7±4.5 mg/d (min.: 1.4 mg/d, max.: 17.1 mg/d) and within the HIV negative control group 10.3±6 mg/d (min.: 2.7 mg/d; max.: 31.2 mg/d) so the Zn intake of the HIV positive women was significant ($p<0.01$) lower than of negative lactating women. However, the recommended Zn intake of 11 mg/d could not be reached in both groups.

The mean Zn intake in the HIV positive group was half as much than recommended. Within the HIV negative group the average Zn intake was slightly below, but close to the recommended value.

A study of dietary patterns of pregnant women in an impoverished part of Lima in Peru reported about the same inadequate Zn intake like in this study [SACCO et al., 2003]. If it is taken into account, that HIV negative women do have a higher educational standard and a higher income, they are more likely to be able to effort better-balanced nutrition.

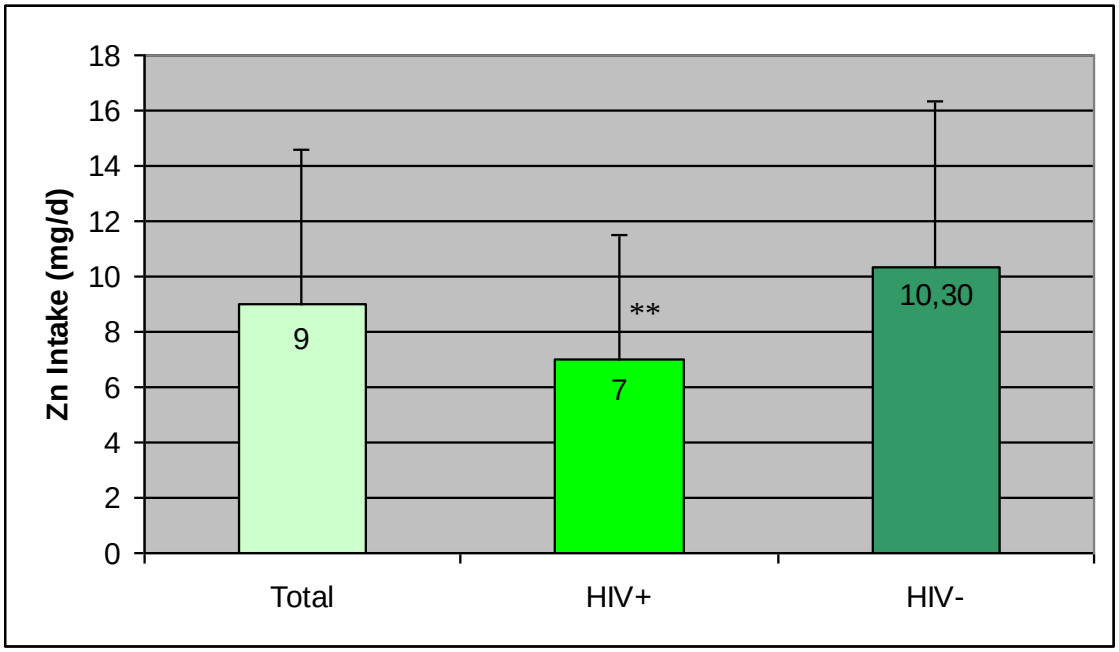


Figure 18: Zn Intake of Respondents (**: HIV+ <HIV-; p<0.01)

The nutritional adequacy of dietary Zn depends on both, its amounts and bioavailability in the diet. In many developing countries, the content of flesh foods in rural diets is often low so that their contribution to total dietary Zn intake is small. Plant based diets often contain high levels of phytic acid and dietary fiber. Those components are known to inhibit the absorption of dietary Zn. The bioavailability of dietary Zn can be predicted from ratio of phytic acid to Zn in diets. The critical Phy: Zn molar ratios associated with risk of Zn deficiency are equivocal; ratios above 15 have been associated with biochemical and in some cases clinical signs of Zn deficiency in humans. Typical African dishes do have elevated Phy: Zn molar ratios (Table 15).

Table 15: Zn and Food

[Mod.:Ferguson et al., 1993]

Food	Zn [mg]	Phy [mg]	Phy: Zn [ratio]
Rice and stew	0.6	118	21
Rice and beans	0.5	107	18
Gari and beans	0.9	178	22

Groundnut soup	0.8	81	10

All dishes plus tomatoes, red peppers, salt, onion, palm oil

Water and liquid consumption

Already 75% (HIV+: 43%; HIV-: 57%) of the study participants got their drinking water out of pipes, 9% (HIV+: 43%; HIV-: 57%) of all respondents obtained their water from boreholes, 6% (HIV+: 20%; HIV-: 80%) bought their water in shops, 1% (HIV+: 100 %; HIV-: 0 %) got their water from a spring and 1% (HIV+: 0 %; HIV-: 100 %) from a river. 8% (HIV+: 33%; HIV-: 67%) stated other water supplies.

The recommendation for water/liquid intake for lactating women is 2300 ml/d (equivalent g/d). Most of the women preferred drinking black tea with milk than pure water.

Within the total respondent group the mean water/liquid intake (includes drinking water/liquid and water content in food) was 1571±691 g/d (min.: 483 g/d; max.: 3102 g/d). The water/liquid intake in the HIV positive group was lower than in HIV negative group, but there is no statistical significance. The mean intake of HIV positive women was 1141±464 g/d (min.: 483 g/d; max.: 1914 g/d) and in the HIV negative group 1826±679 g/d (min.: 505 g/d; max.: 3102 g/d).

None of the groups did reach the recommendation.

Statements and Discussion: Nutritional Data

In this study the daily energy intake of the participating women was lower (HIV+: 965.64±496 kcal < HIV-: 1363.09±627kcal; $p<0.05$) than the recommendation of 2850 kcal/d. The same situation was observed concerning the intake of protein (recommended: 63g/d; HIV+: 33±9 g/d; HIV- 55±37 g/d), fat (recommended: 30–35 E%; HIV+: 19±9E%; HIV-: 20±10) and dietary fibre (recommended: 30 g/d; HIV+: 19±13 g/d; HIV-: 24±11 g/d). The intake of carbohydrates was higher than the recommended amount (recommended: 50 E%; HIV+: 65±15 E%; HIV-: 63±13 E%). It has to be stated that HIV negative

control group perform better or even has higher intakes (e.g. carbohydrates) than the recommended amount.

In a study done in South Africa the energy intake of HIV positive women was significantly higher ($p < 0.05$; CI 95%) than in the HIV negative group. The mean intake of the total group exceeded the recommended dietary intakes. Fat was even higher than the recommendend 30 – 30% of the daily energy intake, dietary fiber was very low [HATTINGH et al., 2006]

Data evaluated in this study indicated a very low intake of animal products and animal protein. Zinc and iron intakes were also quite low and additionally the bioavailability of these micronutrients is poor because of the high phytate, fiber and tea content of the typical diet.

Further, vitamin B_{12} intake is extremely low, and at least mild-to-moderate iodine deficiency is reported to be present in Kenya.

Results from the Collaborative Research Support Program (CRSP) reported of the highest predicted prevalence of inadequacy in developing countries for pregnant/ lactating women were iron, zinc, calcium, riboflavin, vitamin B-12, retinol and vitamin E. Animal source food are the main and most bioavailable source of these micronutrients in most diets and the inadequate intakes in the Nutrition CRSP were due to the low intakes of animal products. The percent of total energy from animal source food was only 18% in Egypt, 12% in Mexico and 8% in Kenya [NYAMBOSE et al., 2002].

4.2. Breastfeeding

65% (HIV+: 39%; HIV-: 61%) of the recruited women chose exclusive breastfeeding and 35% (HIV+: 43%; HIV-: 57%) chose the mixed feeding (breastfeeding combined with formula feeding) option.

In this study no significant differences between the frequency of chosen feeding options and the HIV status could be found.

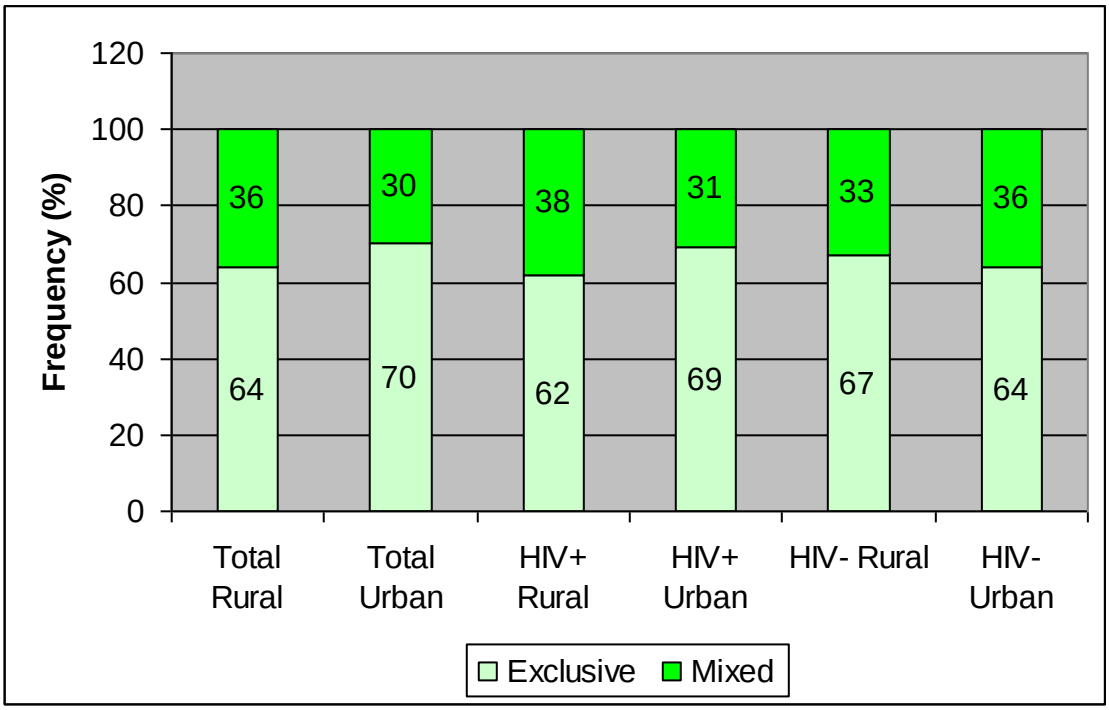


Figure 19: Breastfeeding Option and Origin

Statements and Discussion: Breastfeeding

The pattern of adherence to exclusive infant feeding was similar to what has been found in other studies. Among women who maintained breastfeeding, a strong belief in the benefits of this feeding option and a supportive home environment was important [DOHERTY et al., 2006; OMARI et al., 2003]. Socioeconomic and infrastructural benefits like the availability of electricity, a kettle, a flask and formula food makes exclusive formula feeding interesting for HIV positive mothers. So women living in urban areas would have a profit because of the easier accessibility of the utensils for formula feeding (Figure19; Figure 20).

It has to be noted, that with clearing up about HIV/ AIDS and the falling of stigmatization HIV positive women would be more encouraged to attend PMTCT (prevention of mother to child transmission) programs and to organize formula food for their babies. For these mothers formula feeding could get a properly alternative and probably contribute to minimize MTCT of the virus via breastfeeding.

4.4 Selenium – GSH-Px Activity

For assessing the Se status in HIV positive and HIV negative women the erythrocyte GSH-Px activity was analyzed in the laboratory. The mean value within all women was at 24.01±7.21 U/g Hb (min.: 10.00 U/g Hb; max: 44.27 U/g Hb). Within the HIV positive women the average activity of GSH-Px was 25.21±8.30 U/g Hb (min.: 13.04 U/g Hb; max.: 44.27 U/g Hb), among the HIV negative control group the mean level was 22.85±6.53 U/g Hb (min.: 10.00 U/g Hb; max.: 38.00 U/g Hb) (Figure 21).

There were no statistically significant differences between the seropositive and seronegative group and their assessed GSH-Px values.

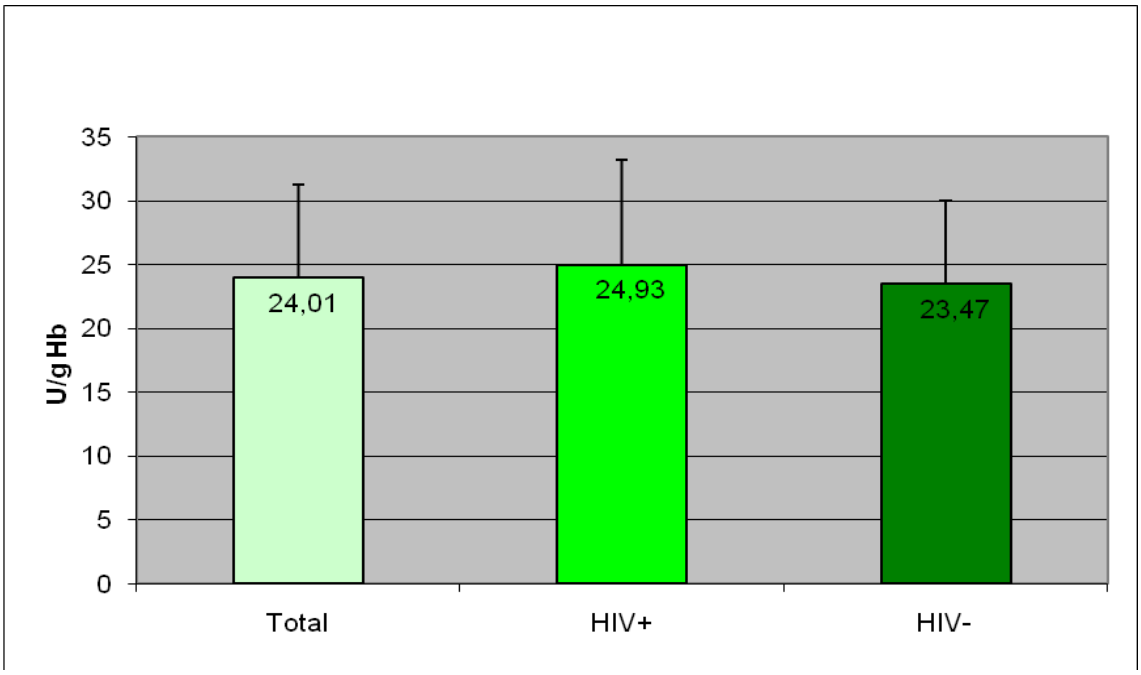


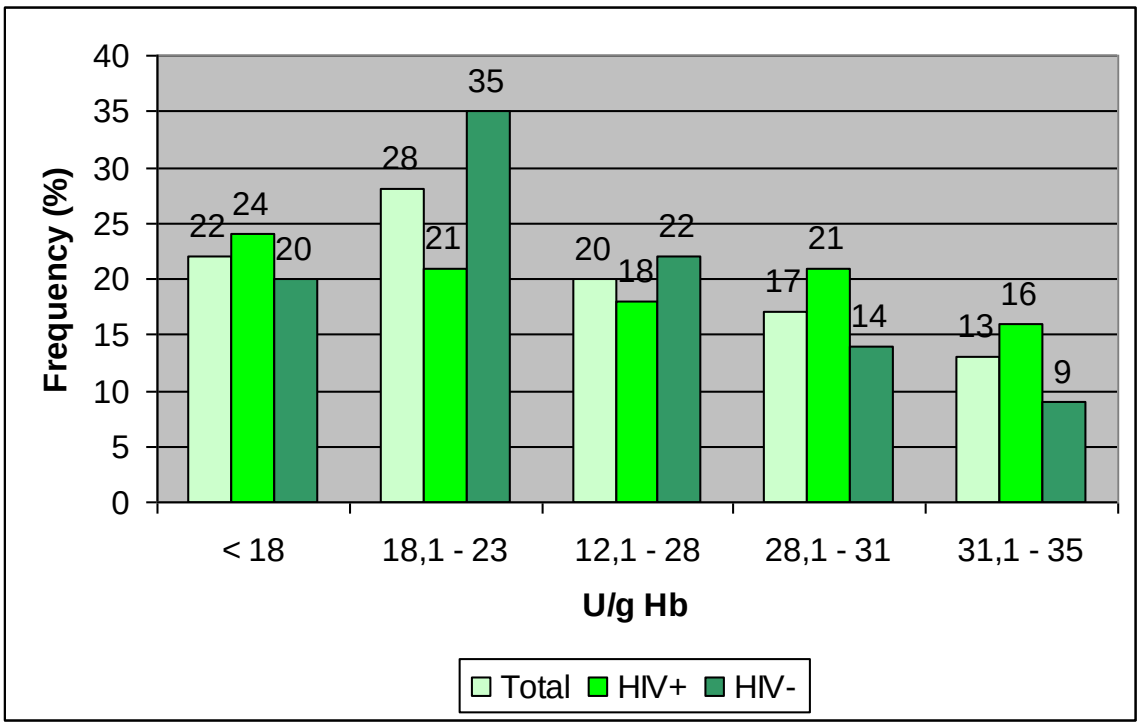
Figure 20: GSH- Px Activity of the Respondents

22% (HIV+: 43%; HIV-: 57%) of all recruited women had a value lower than 18 U/g Hb, 28% (HIV+: 25%; HIV-: 75%) were between 18.10- 23 U/g Hb, 20% (HIV+: 31%; HIV-: 69%) between 23.10-28 U/g Hb, 17% (HIV+: 50%; HIV-: 50%) between 28.10- 30 U/g Hb and 13% (HIV+: 57%; HIV-: 43%) between 30.10 and 35 U/g Hb.

Within the HIV positive almost a quarter (24%) had an GSH-Px activity under 18.00 U/g Hb, 21% in the range of 18.10- 23 U/g Hb, 18% between 23.10- 28 U/g Hb. 21% of the HIV positive subjects showed activities between 28.10- 31 U/g Hb and 16% in the range of 31.10- 35 U/g Hb.

In the HIV negative control 20% had lower enzyme activity levels than 18 U/g Hb, 35% ranged between 18.10- 23 U/g Hb, 22% between 23.10- 28 U/g Hb, 14% had values between 28.10- 31 U/g Hb and 9% had GSH-Px activity levels between 31.10- 35 U/g Hb (Figure 22).

The statistical distribution of the GSH-Px activities among the HIV positive participants did not significantly differ from that of the HIV negative subjects.

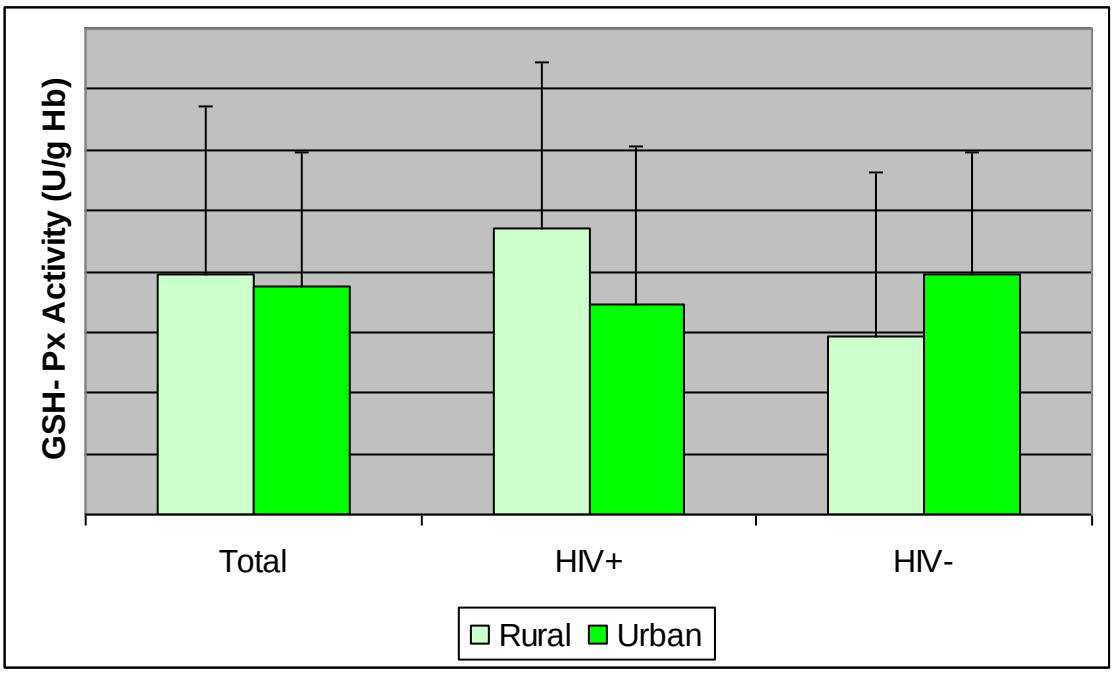


	<18 (U/g Hb)	18.1 – 23 (U/g Hb)	23.1 – 28 (U/g Hb)	28.1 – 31 (U/g Hb)	31.1 – 35 (U/g Hb)
Total (%)	22	28	20	17	13
HIV+ (%)	24	21	18	21	16
HIV- (%)	20	35	22	14	9

Figure 21: Erythrocyte GSH-Px activity (U/g Hb)

The normal range of GSH-Px activity should be between 18 and 25 U/g Hb [LOOK et al, 1997]. It has to be noted, that 20% of the HIV negative women had GSH-Px activities lower than 18 U/g Hb and in the HIV positive group the range of 18.1- 23 U/g Hb were the biggest group with 21%.

Further evaluation of the data showed that the demographic origin (urban/rural) of a woman did not affect GSH-Px activity. The mean value in the total urban origin group was 18.84±11 U/g Hb (HIV+: 17.35±13 U/g Hb; HIV-: 19.76±10 U/g Hb) and within the rural origin group 19.77±13.83 U/g Hb (HIV+: 23.6±13.6 U/g Hb; HIV-: 14.6±13.5 U/g Hb) - no statistical significant differences could be observed between the HIV positive and HIV negative groups (Figure 23).



	Total (U/g Hb)	HIV+ (U/g Hb)	HIV- (U/g Hb)
Rural	19.77±13.83	23.6±13.6	14.6±13.5
Urban	18.84±11	17.35±13	19.76±10

Figure 22: GSH-Px Activity of the Respondents living in different Areas

In contrast to other investigations no correlations between age and GSH-Px activity could be identified in this study. However, there are studies indicating that HIV positive lactating women with a higher age, lower education and higher

parity have a higher plasma Se values, causing a higher GSH-Px activity levels in this group [DELMAS- BEAUVIEUX et al., 1996].

In a study by KUPKA et al. [2004] among HIV infected pregnant women in sub-Saharan Africa, lower plasma selenium levels were associated with increased mortality and weakly with decreased CD^+ 4 cell counts.

SNOOK et al. [1987] reported neither of no correlation of Se and the mothers BMI nor with the infants' birth weights.
Some studies reported a significant dependence of the Se status from the area (urban/rural) where people are living. The Se contained in various plant derived foods depends on the Se soil content. In the region of Nakuru, Kenya, where this study was carried out soils are not poor of Se. So it can be assumed that crops and other plant foods contain an acceptable amount of this micronutrient. However it has to be noted that the food content is not equal to the bioavailability of Se (s. 2.4.4). Among different foods the highest Se content is found within intestines (0.4- 1.5 µg/g) and meat (0.1- 0.4 µg/g) but these foods are not primarily consumed by Kenyan women. Finally, there was no urban/rural influence on the Se status identified in this investigation.

The mothers' intake of Se is very important and has a big influence on the breast milk concentration. Coherence between demographic data and breast milk Se content is discussed controversial.
VERTAINEN and KANTOLA [2001] reported a significant difference in breast milk Se concentrations between rural and urban Finnish mothers. Like in this investigation SNOOK et al. [1987] did not observe a significant difference in milk Se between rural and urban mothers.

No significant correlation was found between the state of health and GSH-Px activity. There was no significant correlation found between the positive status of the baby, the origin of the HIV positive mother, her income and her GSH-Px

activity. Within the HIV positive group was no significant correlation found between GSH- Px and the HIV status of the child.

4.2. Zinc – SOD Activity

For Zn assessment, laboratory analyses of erythrocyte SOD activity were done. The mean value within all women was at 1005.2±253.5 U/g Hb (min.: 593.4 U/g Hb; max: 1969.5 U/g Hb). The mean value within the HIV positive women was 939.5±217.3U/g Hb (min.: 593.4 U/g Hb; max.: 1677.7 U/g Hb). The mean value in the HIV negative control group was 1075.4±256.1 U/g Hb (min.: 686.3 U/g Hb; max.: 1969.5 U/g Hb) (Figure 24).
There was no statistical significant difference of the erythrocyte SOD activity between HIV positive and negative respondents.

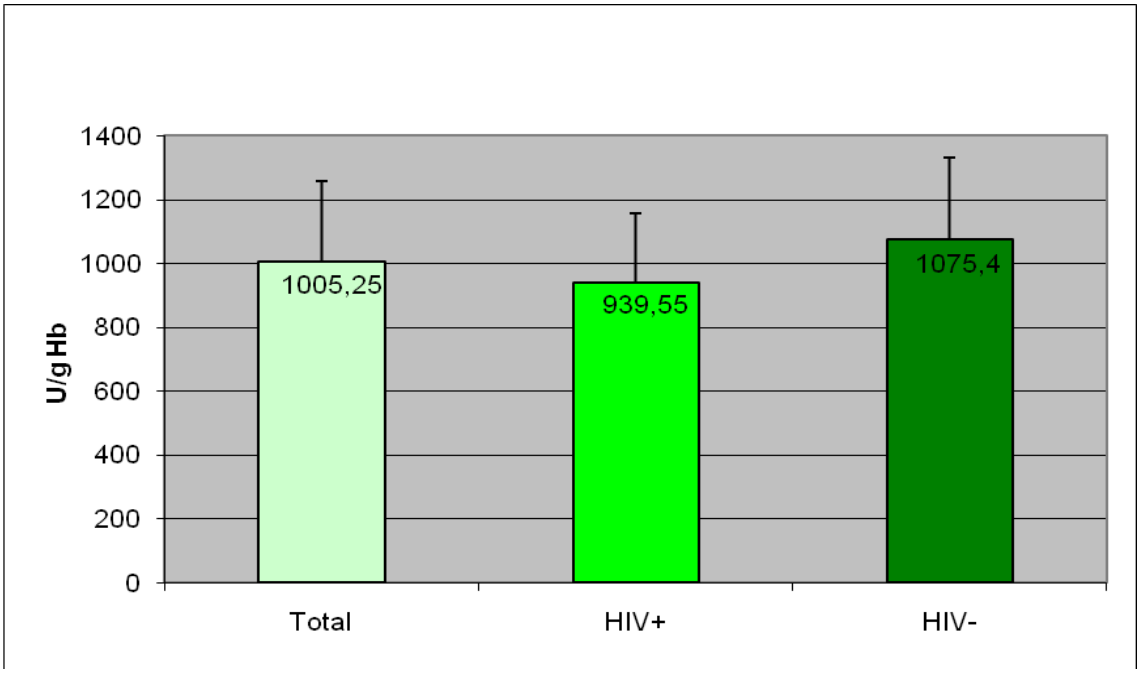


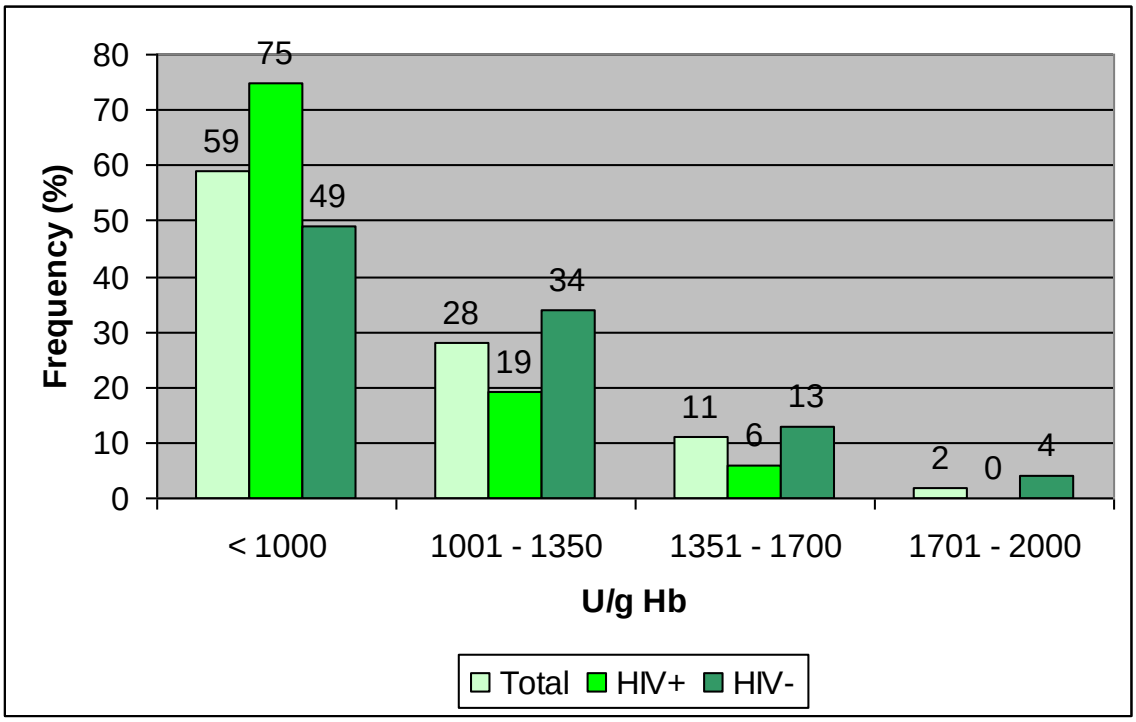
Figure 23: SOD Activity of the Respondents

59% (HIV+: 52%; HIV-: 48%) of all recruited women had a SOD activity lower than 1000 U/g Hb, 28% (HIV+: 29%; HIV-: 71%) between 1001- 1350 U/g Hb,

11% (HIV+: 29%; HIV-: 71%) between 1351- 1700 U/g Hb and 2% (HIV+: 0 %; HIV-: 100 %) between 1701- 2000 U/g Hb.

Within the HIV positive three quarter (75%) of the respondents had an enzyme activity under 1000 U/g Hb, 19% in the range of 1001- 1350 U/g Hb, and 6% between 1351- 1700 U/g Hb. In none of the HIV positive women SOD activity reached the level of 1700 U/g Hb.

More than half (59%) of the HIV negative recruited women had levels below 1000 U/g Hb, 34% had levels between 1001- 1350 U/g Hb, 13% were between 1351- 1700 U/g Hb, and 4% had values over 1700 U/g Hb (Figure 25).



	<1000 (U/g Hb)	1001–1350 (U/g Hb)	1351–1700 (U/g Hb)	1701-2000 (U/g Hb)
Total (%)	59	28	11	2
HIV+ (%)	75	19	6	0
HIV- (%)	49	34	13	4

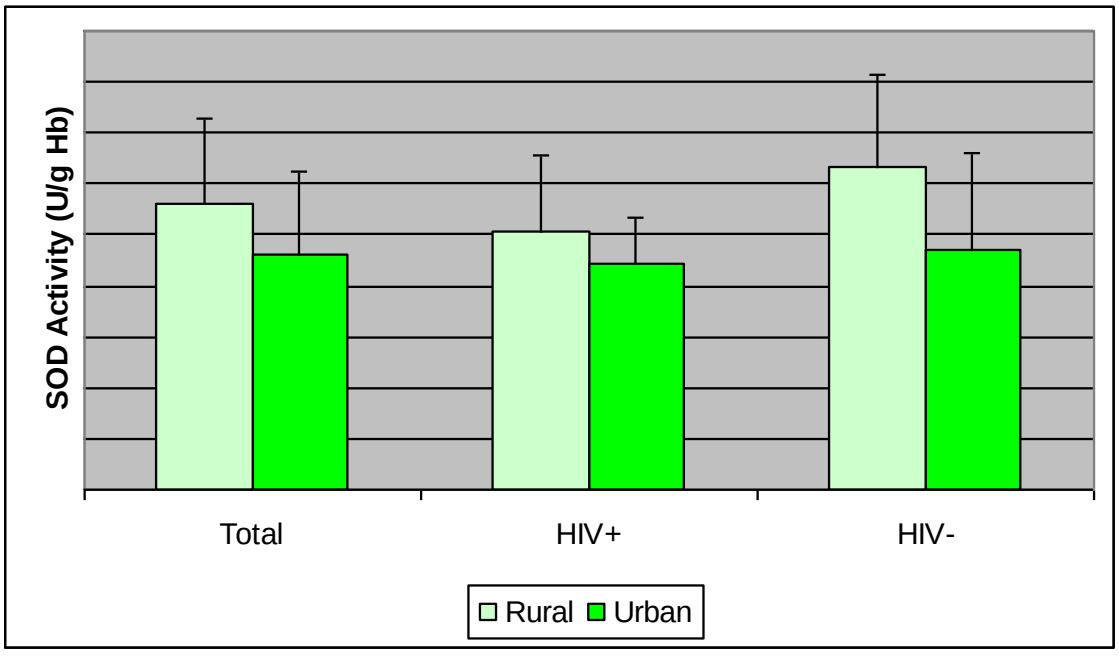
Figure 24: Erythrocyte SOD activity (U/g Hb)

GUEMOURI L et al [1991] fixed the normal range for erythrocyte SOD activity between 1000-1700 U/g Hb.
In this study, 59% of all women, 75 % of the HIV positive group and almost half (49%) of the HIV negative subjects had enzyme activity levels below the value of 1000 U/g Hb.

There was no significant correlation between Zn intake and erythrocyte SOD activity.

In reference to age, a significant positive correlation ($r= 0.099$, $p< 0.05$) between the HIV status and SOD activity could be found in this investigation. Among the HIV negative control group no significant correlation of these parameters were observed.

Moreover the evaluation of the data showed a statistical significant correlation ($r=0.018$, $p< 0.05$) between SOD activity and origin (urban, rural, out of town, peri- urban). Women living in rural areas (1121±339.6 U/g H) had a significant ($p<0.05$) higher SOD activity than women living in an urban area (918.6±322.2 U/g Hb) (Figure. 26).



	Total (U/g Hb)	HIV+ (U/g Hb)	HIV- (U/g Hb)
Rural	1121±339.6	1012.4±299	1265.8±361.5
Urban	918.6±322.2	886.4±178.7	938.67±386

Figure 25: SOD Activity of the Respondents living in different Areas

Most of the women living in rural areas have to plant their own vegetables and keep animals, like cows and chicken for feeding the whole family. Women in urban areas need more money to get a well balanced diet and most of the interviewed women did not have enough money to provide adequate nutrition for themselves. However, no significant correlation between income and SOD activity could be found.

There was no significant correlation between the state of health and SOD activity and no significant relationship between SOD activity of the HIV positive mothers and the HIV status of the breast fed children. Further, no significant correlation between SOD activity, origin of the HIV positive mother and the risk of MTCT could be observed. Worth mentioning is, that HIV positive mothers with HIV positive infants do have SOD activities lower than 1000 U/g Hb.

5. Conclusion and Recommendation

The study shows, that HIV/ AIDS exists irrespective of age, education, social class and demographic origin. There are differences between the infected and uninfected women for sociodemographic characteristics, anthropometrical and nutritional data, SOD and GSH-Px activity. HIV positive women do not live in matrimony than HIV negative women, are not that well educated and are not their food supply is not that good- although HIV negative lactating mothers in Nakuru are also not very well supplied with micro- and macronutrients.

The evaluation of nutrient intake data (according to 24-h-recalls) clearly indicated that a majority of the study participants were malnourished. The required energy intake did not reach the recommended daily amount. Lactating and HIV positive women do need extra energy- on the one hand they use energy for producing milk and on the other hand their body needs energy to strengthen their immune system and to fight against the HI virus. Moreover, the micronutrient intake of the investigated subjects was generally too low. The recommended daily intake neither of vitamin C, iron and total folic acid nor of vitamin E or Zn were reached- in the HIV positive and HIV negative group.
Interventions for improving the women's nutrient intake and their health status should be an overall strategy, regardless whether the women are HIV negative or HIV positive. Due to the low educational status, especially within the HIV positive group, just an advice to eat more wouldn't be useful. A follow- up teaching programme would be helpful, where childbearing women learn how to optimize their nutritional status by choosing the right food composition e.g. Se and Zn uptake will be supported by eating proteins (meat) and lowered by high phytate concentrations (green leafs).

Consumption of more high quality foods, including animal source food, has the benefit of increasing the intake of many nutrients simultaneously. Some of these nutrients, as well as other bioactive constituents of foods, would not be provided

in either supplementation or fortification programs. All household members could benefit from improved household dietary quality, unlike supplementation programs, which are usually confined to women and young children, and fortification programs that may not reach the most needy population groups.

Low intakes of macro- and micronutrients and low BMI values of HIV positive mothers are a problem and an additional risk factor for morbidity, mortality and MTCT. In Tanzania, maternal multivitamin (vitamins B, C, and E) supplementation reduced the risk of HIV transmission via breastfeeding among women who were nutritionally and immunologically compromised, and had additional health benefits such as a decreased incidence of fetal loss and a reduced rate of maternal disease progression [FAWZI et al., 2001]. Especially in HIV positive women malnourishment can lead to an inactive immune system and opportunistic infections are more common. It would be very useful to teach and show the women how to enrich their daily diet with “extra energy” without spending more money.

Most of the study participants choose exclusive breastfeeding, because the women in countries like Kenya do not have clean water out of the tap, the utensils and the money for infant formula food. Breastfeeding is also the “easier” way, because most of them have to go back to work e.g. farm their fields or leave the infants with their housemaids.
In this study it could be shown that the decision for an exclusive feeding option whether breast feeding or formula feeding depends on the fully informed parents and has to be promoted. For the infants development it is not very useful to mix different feeding options because of its proposed adverse effects on the mucosal integrity and its increased risk of MTCT.
The adoption of formula feeding is not widespread in developing regions such as sub-Saharan Africa, partly due to its high cost and partly due to the social stigma associated with not breastfeeding. Further, in settings where access to adequate hygienic conditions is limited, formula feeding may lead to increased infant morbidity and mortality.

No clear coherence between MTCT and Se and Zn could be found in this study. Thus the HIV negative group was slightly better provided with Zn than the HIV negative group. The amount of Se and Zn intake depends, as already stated before, on the Se and Zn soil concentration. In the area of Nakuru, Rift Valley no Se/Zn poor soil is known by local agrarian scientist. However, it has to be taken into account, that Se/Zn intake is not the same as Se/Zn bioavailability. Finally it has to be noted, that a lack of Zn and Se supply respectively, is not just a HIV/AIDS problem in Kenya it is a problem of the whole female population.

Although the results of this study could not identify a clear coherence between MTCT and the status of Se and Zn, other studies [BAUM MK et al., 1995; NDUATI RW et al., 1995; SEMBA RD et al., 1999] showed connections within these three parameters. An intervention study with a higher respondent number and carried out over a longer period of time would give more and concrete information about the coherence between MTCT, Se and Zn. Much more factors (e.g. CD^+4 count, viral load of the mother, HIV status of the infant after delivery) which depend on Se and Zn have to be assessed and discussed.
Further research is warranted to investigate the role of other nutrients in vertical HIV transmission via breastfeeding and overall maternal and infant health among HIV-infected breastfeeding women and their children.
Finally it can be concluded that in Kenya a lot of information work about the necessity of nutrition has to be done in the future, whether the persons concerned are HIV positive or HIV negative.

6. Summary

In developing countries an estimated 1.2 billion people live with less than 2 € a day. In the last years another threat joined famine and thirst: AIDS. In the previous ten years the acquired immunodeficiency syndrome became a deadly hazard for the people and their countries economic situation.

In May and June 2008 a study was done in the Provincial General Hospital Nakuru, Rift Valley. The coherence between the maternal nutritional status of Zn/ Se and the MTCT of HIV was assessed. 79 lactating women (HIV+: 32; HIV-: 47) took part. The respondents had to answer questions about nutrition and health, 24 hour Recalls were done and the women had to give 5 ml venal blood for laboratory analysis.

The women nutritional status is not satifactory. Both in macronutrients and micronutrients, they do not reach the recommended intakes. Especially the energy intake is alarmingly (recommended: 2850 kcal; mean intake: 2020.15±609 kcal.

By assessing SOD and GSH-Px activity, conclusions about the functional status of Zn and Se could be done. In general all women are quite low in Zn and Se, no matter if the respondent was HIV positive or HIV negative. The mean value of SOD activity within the HIV positive group was 939.5±217.3 U/g Hb, among the HIV negative respondents 1075.4±256.1 U/g Hb (recommendation: 1000 - 1700 U/ g Hb). Within the HIV positive group the mean GSH-Px activity value was 25.21±8.3 U/g Hb and within the HIV negative group 22.85±6.53 U/g Hb (recommendation: 25 – 32 U/g Hb).
No significant differences were observed between the HIV positive and negative groups. Further, it could be found that origin (urban/ rural), education and maritial status did not have an influence on the activity levels of both enzymes and the Se and Zn status, respectively.

No reference to coherence between MTCT of HIV and Se/ Zn could be found.
It has to be noted, that follow up nutritional advices – for both, HIV positive and HIV negative women – would be necessary, to provide adequate nutritional supply.

7. Zusammenfassung

In den Ländern der 3. Welt leben schätzungsweise 1,2 Milliarden Menschen mit weniger als 2 € pro Tag. In den vergangene Jahren gesellte sich zu Hunger und Durst eine weitere Gefahr: AIDS. Das erworbene Immundefizienz Syndrom bekam in den letzten 10 Jahren eine tödliche Gefahr für den Menschen und bedeutet gleichzeitig einen wirtschaftlichen Rückschlag.

Im Provincial General Hospital Nakuru, Rift Valley, wurden im Mai und Juni 2008 eine Studie durchgeführt, bei der die HIV Übertragung von der Mutter auf das gestillte Kind in Abhängigkeit von deren Zink- und Selenversorgung untersucht wurde. 79 stillende Frauen (HIV+: 32; HIV-: 47) nahmen Teil. Die Probandinnen hatten Fragen zum Thema Ernährung und Gesundheit zu beantworten, ein 24 Stunden Ernährungsprotokoll wurde erstellt und je 5 ml venöses Blut wurden abgenommen.

Der Ernährungszustand der Frauen kann im Gesamten als nicht zufriedenstellend beurteilt werden. Sowohl im Makronährstoffbereich, als auch im Mikronährstoffbereich wurde die empfohlene tägliche Zufuhr nicht erreicht. Alarmierend ist die geringe Energieaufnahme- die Empfehlung liegt bei 2800 kcal, die Aufnahme bei 2020.15±609 kcal.

Mithilfe der Messung der SOD Aktivität und der GSH- Px Aktivität konnten Rückschlüsse auf die funktionelle Versorgung von Zn und Se getroffen werden. Prinzipiell kann gesagt werden, dass sowohl die Gruppe der HIV positiven als auch die Gruppe der HIV negativen Frauen ungenügend mit Zink und Selen versorgt sind. Die durchschnittliche Aktivität von SOD lag innerhalb der HIV positiven Frauen bei 939.5±217.3U/g Hb und innerhalb der HIV negativen Frauen bei 1075.4±256.1 U/g Hb (Soll: 1000- 1700 U/ g Hb). Die Se Versorgung unterscheidet sich nicht wesentlich von der der Zn Versorgung. Innerhalb der HIV positiven Gruppe liegen die Werte bei 25,21±8.30 U/g Hb und in der HIV negativen Gruppe bei 22,85±6.53 U/g Hb (Soll: 25 – 32 U/g Hb).

In dieser Studie gibt es keine Hinweise auf einen Zusammenhang zwischen MTCT von HIV und Selen/ Zink. Grundsätzlich muss gesagt werden, dass begleitenden Ernährungsaufklärung, sowohl für HIV positive als auch HIV negative Frauen nötig ist, um den nicht ausreichenden Ernährungszustand sowohl im Makro- als auch im Mikronährstoffbereich zu verbessern.

8. Literature

ARNAUD J, PRUAL A, PRESIOSI P. Effect of iron supplementation during pregnancy on trace element (Cu, Se, Zn) concentrations in serum and breast milk from Nigerian women. Ann Nutr Metab 1993; 37:262-71

BAUM MK, SHOR- POSNER G, LU Y, ROSNER B, SAUBELICH HE, FLETCHER MA, SZAPOCZNIK J, EISDORFER C, BURING JE, HENNENKES CH. Micronutrients and HIV- 1 disease progression. AIDS 1995; 9:1051-1056

BAUM MK, et al. HIV- 1 infection in women is associated with severe nutritional deficiencies. J AIDS Hum Retrovirol 1997; 16, 272

BEACH RS, MANTERO- ATIENZA E, SHOR- POSNER G. Specific nutrient abnormalities in asymptomatic HIV- 1 infection. AIDS 1992; 6:701-8

BHASKARAM P. Micronutrient Malnutrition, Infection, and Integrity: An Overview. Nutrition Reviews 2002; Vol 60, No. 5, 40-45

BOBAT R, COOVADIA H, MOODLEY D, COUTSOUDIS A, GOUWS E. Growth in early childhood in a cohort of children born to HIV- 1 infected women from Durban, South Africa. Annuals of Tropical Paediatrics 2001; 3:203-210

BOGDEN JD, KEMP FW, HAN S. Status of selected nutrients and progression of human immunodeficiency virus type 1 infection. Am J Clin Nutr 2000; 72:809-15

BROWN KH, RIVERA JA, BHUTTA Z, GIBSON RS, KING JC, RUEL M, SANDSTRÖM B, WASANTWISUT E, HOTZ C, LÖNNERDAL B, LOPEZ DE ROMANA D, PEERSOON J. Assessment of the risk of Zinc deficiency in populations and options for its control. Food and Nutrition Bulletin 2004; 25:19-202

BURBANO X, MIGUEZ- BURBANO MJ, MC COLLISTER K, ZHANG G, RODRUGUEZ A, RUIZ P, LECUSAY R, SHOR- POSNER G. Impact of selenium chemoprevention clinical trial on hospital admission of HIV- infected participants. HIV Clin Trials 2002; 3: 483- 491

BRYCE J, EL ARIFEEN S, PARIYO G, LANATA C, GWATKIN D, HABICHT JP. Reducing child mortality: Can public health deliver? Lancet 2003; 362:159-64

CAMERON E. Witness to AIDS. I. B. Tauris & Co Ltd. 2005; 42

CAMPA A, et al. Mortality risk in selenium deficiency HIV positive children. J AIDS Hum Retrovirol 1999; 20, 508

CHANDRA RK. Effect of vitamin and trace- element supplementation on immune responses and infection in elderly subjects. Lancet 1992; 340:1124

COLEY L, MSAMANGA I, FAWZI M, KAAYA S, HERTZMARK E, KAPIGA S, SPIEGELMANN D, HUNTER D, FAWZI W. The association between maternal HIV- 1 infection and pregnancy outcomes in Dar es Salaam, Tanzania. Br J Obstetr and Gyn 2001; 108(11):1125-1133

CONSTANS J. Selenium and AIDS, in Watson RR, Ed. Nutrients and Foods in AIDS. Boca Raton, Fl, CRC Press, 1999

COUTSOUDIS A, PILLAY K, SPOONER E, KUHN L, COOVADIA H. Influence of infant- feeding patterns on early mother- to- child transmission of HIV- 1 in Durban, South Africa: a prospective cohort. Lancet 1999; 354:471-476

COUTSOUDIS A, PILLAY K, KUHN L, SPOONER E, TSAI WY, COOVADIA HM. Method of feeding and transmission of HIV- 1 from mothers to children by 15 months of age: prospective cohort study from Durban, South Africa. AIDS 2001; 15:379-387

DARMON N, DREWNOWSKI A. Does social class predict diet quality? Am J Clin Nutr 2008. 87: 5, 1107-1117

German Nutrition Society, Austrian Nutrition Society, Swiss Society for Nutrition Research, Swiss Nutrition Association: Reference Values for Nutrient Intake (D-A-CH). Frankfurt am Main, Umschau/Braus, 2000

DELMAS- BEAUVIEAUX MC, PEUCHANT E, COUCHOURON A, CONSTANS J, SIMONOFF M, PELLEGRIN JL, LENG B, CONRI C, CLERC M. The enzymatic antioxidant system in blood and glutathione status in human immunodeficiency virus (HIV)- infected patient: effects of supplementation with selenium or beta- carotene. Am J Clin Nutr 1996; 64: 101- 107

DELMAS- BEAUVIEAUX MC, PEUCHANT E, COUCHOURON A, CONSTANS J, SERGEANT C, SIMONOFF M, PELLEGRIN JL, LENG B, CONRI C, CLERC M. The enzymatic antioxidant system in blood and glutathione status in human immunodeficiency virus- infected patients: Effects of supplementation with selenium or beta- carotene. Am J Clin Nutr 2006; 64(1), 101

DIPLOCK AT. Indexes of selenium status in human populations. Am J Clin Nutr 1993; 57(2 Suppl.), 256S

DOHERTY T, CHOPRA M, NKONKI L, JACKSON D, PERSSON LA. A longitudinal qualitative study of infant feeding decision making and practices among HIV positive women in South Africa. J Nutr 2006; 2421- 2426

DREYFUSS M, MSAMANGA G, SPIEGELMANN D, HUNTER D, URASSA J, HERTZMARK E, FAWZI W. Determinants of low birth weight among HIV-infected pregnant women in Tanzania. Am J Clin Nutr 2001; 74(6):814-826

DUNN DT, NEWELL ML, ADES AE, PECKHAM CS. Risk of human immunodeficiency virus type 1 transmission through breast feeding. Lancet 1992; 340:585-588

ELMADFA I, LEITZMANN C. Ernährung des Menschen. Ulmer Verlag. 2004, 627

EMBREE JE, NJENGA S, DATTA P, NAGELKERKE NJD, NDINYA- ACHOLA JO, MOHAMMED Z, RAMDAHIN S, BWAYO JJ, PLUMMER FA. Risk factors for postnatal mother- child transmission of HIV-1. AIDS 2000; 14:2535- 2541

FAVIER A, SAPPEY C, LECLERC P, FAURE P, MICOUD M. Antioxidant status and lipid peroxidation in patients infected with HIV. Chem Biol Interact 1994; 91:165-180

FERGUSON EL, GIBSON RS, THOMPSON LU, OUNPUU S, BERRY M. Phytate zinc and calcium contents of 30 East African foods and their calculated phytate: zinc, Ca: phytate and [Ca][phytate]/ [Zn] molar ratios. J Clin Nutr 1989; 50: 1450- 1456

FILTEAU SM, MORRIS SS, ABBOTT RA. Influence of morbidity on serum retinol of children in a community- based study in northern Ghana. Am J Clin Nutr 1993; 58:192-7

FILTEAU SM, LIETZ G, MULOKOZI G, BILOTTA S, HENRY CJ, TOMKINS AM. Milk cytokines and subclinical breast inflammation in Tanzanian women: effects of dietary red palm oil or sunflower oil supplementation. Immunology 1999; 97:595-600

FYLKESNES K, MUBAGA MUSONDA R, KASUMBA K, NDHLOVU Z, MLUANDA F, KAETANO L, CHIPAILA CC. The HIV epidemic in Zambia: socio-demographic prevalence patterns and indications of trends among childbearing women. AIDS 1997; 11:339- 345

FRANKEL AD, BREDT DS, PABO CO. Tat protein- from human immunodeficiency virus forms a metal- linked dimer. Science 1988; 240:70-73

GERSHWIN ME, GERMAN JB, KEEN CL. Nutrition and Immunology: Principles and Practice (Gershwin M. E.). Humana Press Inc., 2000; 505

GIBSON RS, FERGUSON EL. Assessment of dietary zinc in a population. Am J Clin Nutr 1998; 68(suppl):430S-4S

GOLDMAN AS, CHEDA S, GAROFALO R. Spectrum of immunomodulating agents in human milk. Int J Pediatr Hematol/ Oncol 1997; 4:491-7

GRAHAM N, BULTERYS M, CHAO A, HOPKINS J. Effects of maternal vitamin A deficiency on infant mortality and perinatal HIV transmission. National Conference on Human Retroviruses and Related Infection. Baltimore: John Hopkins University, 1993

GUEMOURI L, ARTUR Y, HERBETH B, JEANDEL C, CUNY G, SIEST G. Biological Variability of Superoxide Dismutase, Glutathione Peroxidase and Catalase in Blood. Clin Chem 1991; 37: 11, S1932- 1937

HALLET TB. Declines in HIV prevalence can be associated with changing sexual behaviour in Uganda, urban Kenya, Zimbabwe, and urban Haiti. Sexually Transmitted Infections 2006; 82(Suppl. I): i1-i8

HATTINGH Z., WASLH C.M., VELDMANN F.J., BESTE C.J. Macronutrient intake of HIV- serpositive women in Mangaung, South Africa. Nutr Research 2006; 26:2; S53- 58

HUDDLE JM, GIBSON RS, CULLINAN TR. Is zinc a limiting nutrient in the diets of rural pregnant Malawian women? Br J Nutr 1998; 79: 257- 265

ISA I, LUCCHINI A, LODI S, GIACCHETTI M. Blood zinc status and zinc treatment in human immunodeficiency virus– infected patients. Int J Clin Lab Res 1992; 22:45-47

KAMAU- MBUTHIA E, MWONYA R, ELMADFA I. The Impact of maternal HIV status on Infant Feeding Patterns in Nakuru, Kenya. J Hum Lact 2008; 24:34-41

KANTOLA M, VARTIAINEN T. Changes in selenium, zinc, copper and cadmium contents in human milk during the time when selenium has been supplemented to fertilizers in Finland. J Trace Elements in Med and Bio 2001; 15, 11-17.

KIARIE JN, RICHARDSON BA, MBORI- NGACHA D, NDUATI RW, JOHN-STEWART GC. Infant feeding Practices of Women in a Perinatal HIV- 1 Prevention Study in Nairobi, Kenya. J Acquir Defic Syndr 2004; 35:75-81

KEBS NF, REIDINGER CJ, HARTLEY S, ROBERTSON AD, HAMBIDGE KM. Zinc supplementation during lactation: effects on maternal status and milk zinc concentrations. Am J Clin Nutr 1995; 61:1030-6

KELLY P, MUSONDA R, KAFWEMBE E, KAETANO L, KEANE E and FARTHING M. Micronutrient supplementation in the AIDS diarrhoea- wasting syndrome in Zambia: a randomized controlled trial. AIDS 1999; 13(4), 495

KUHN L, STEIN Z. Infant survival, HIV infection, and feeding alternatives in less- developed countries. Am J Public Health 1997; 87:926-931

KUPKA R, MSAMANGA GI, SPIEGELMANN D, MORRIS S, MUGUSI F, HUNTER DJ, FAWZI WW. Selenium status is associated with accelerated HIV disease progression among HIV- 1 infected pregnant omen in Tanzania. J Nutr 2004. 134: 2556- 2560

KUPKA R,GARLAND M, MSAMANGA GI, SPIEGELAMNN D, HUNTER DJ, FAWZI WW. Selenium status, pregnancy outcomes and mother to child transmission of HIV- 1. J Acquir Immune Def Syndr 2005 Oct; 40(2):219-25

LACASSE M, FORTIER C, CHARKIR J, COTE L, DESTAURIERS N. Acquired resistance and persistence of Candida albicans following oral candidiasis in the mouse: a model of the carrier state in humans. Oral Microbiol Immunol 1993; 8:313-318

LEHTI KK. Breast milk folic acid and zinc concentrations of lactating, low socio-economic, Amazonian women and the effect of age and parity on the same two nutrients. Eur J Clin Nutr 1990; 44:675-80

LEON- CAVER N, LUTTER C, ROSS J, MARTIN L. Quantifying the benefits of breastfeeding: a summary of the evidence. Pan American Health Organization, Washington DC, 2002

LÖNNERDAL B. Nutrition and Immunology- Principles and Practice. Edited by GERSHWIN ME, KEEN C, GERMAN JB. Humana Press Inc., 2000; 171-179

MELLY Y, DE ROQUIGNY H, MORELLET M, ROQUES BP, GERARD D. Zinc binding to the HIV- nucleocapsid protein: a thermodynamic investigation by fluorescence spectroscopy. Biochemistry 1996; 35:5175- 5182

MINISTRY OF HEALTH, REPUBLIC OF KENYA. Kenyan National Guidelines on Nutrition and HIV and HIV/ AIDS. 2007

MIOTTI PG, TAHA TET, KUMWENDA NI, BROADHEAD R, LABAN AR, VAN DER HOEVEN L, CHIPHANGWI JD, LIOMBA G, BIGGAR RJ. HIV through Breastfeeding. A study in Malawi. JAMA 1999; Vol 282, No 8, 744-749

MOCCHEGIANI E, VECCIA S, ANCARANI F, SCALISE G, FABRIS N. Benefit of oral zinc supplementation as an adjunct to zidovudine (AZT) therapy against opportunistic infections in AIDS. Int J Immunopharm 1995; Vol 17 No 9:719-727

MOCCHEGIANI E, MUZZIOLI M, GAETTI R. Contribution of zinc to reduce CD4+ risk factor for severe infection relapse in aging: parallelism with HIV. Int J Immunopharmacol 1999; 21:271-81

MOCCHEGIANI E, MUZZIOLI M. Therapeutic Application of Zinc in Human Immunodeficiency Virus against Opportunistic Infections. J Nutr 2000 May; 130:1424S–31S

MORENO T. Serum copper concentration in HIV infection patients and relationships with other biochemical indices. Sci Total Enviro. 1998; 217, 21

NDUATI RW, JOHN GC, RICHARDSON BA. Human immunodeficiency virus type 1- infected cells in breast milk: association with immunosuppression and vitamin A deficiency. J Infect Dis 1995; 172:1461-8

NDUATI R, JOHN G, MBORI – NGACHA D, RICHARDSON BA, OVERBAUGH J, MWATHA A, NDINYA- ACHOLA J, BWAYO J, ONYANGO FE, HUGHES J, KREISS J. Effect of Breastfeeding and Formula Feeding on Transmission of HIV-1. A Randomized Trial. JAMA 2000; 283:9

NDUATI R, MBORI- NGACHA D, JOHN G. Morbidity and mortality in breastfed and formula- fed infants of HIV- 1 infected women: a randomized clinical trial. JAMA 2001; 286:2413-2420

NEWBURG DS, LINHARDT RJ, AMPOFO SA, YOLKEN RH. Human milk glycosaminoglycans inhibit HIV glycoprotein gp120 binding to its host cell CD4 receptor. J Nutr 1995; 125:419-24

NYAMBOSE J, KOSKI K.G., TUCKER K.L.High Intra/Interindividual Variance Ratios for Energy and Nutrient Intakes of Pregnant Women in Rural Malawi Show That Many Days Are Required to Estimate Usual Intake. J Nutr 2002; 132:1313-1318

OBI CL. Subtypes of HIV- 1 and the impact of dual infections of HIV- 1 and measles virus on micronutrient levels of pregnant women in Harare, Zimbabwe. Centr Afr J Med 1997; 43, 165

O'DELL BL. Role of zinc in plasma membrane function. J Nutr 2000; 130:1432S-6S

OLSON DM, MIJOVIC JE, SADOWSKY DW. Control of human parturition. Semin Perinatol 1995; 19:52-63

OMARI AA, LUO C, KANKASA C, BHAT GJ, BUNN J. Infant feeding practices of mothers of known HIV status in Lusaka, Zambia. Health Policy Plan 2003; 18: 156-62

OSMAN B, CHALLIS K, COTIRO M, NORDAHL G, BERGSTROM S. Perinatal outcome in an obstetric cohort of Mozambican women. Journal of Tropical paediatrics 2001; 47(1):30-38

PHADKE MA, GADGIL B, BHARUCHA KE, SHROTRI AN, SASTRY J, GUPTE NA, BROOKMEYER R, PARANJAPE RS, BULAKH PM, PISAL H, SURYAVANSHI N, SHANKAR AV, PROPPER L, JOSHI PL, BOLLINGER RC. Replacement- fed infants born to HIV- infected mothers in India have a high early postpartum rate of hospitalization. J Nutr 2003; 133:3153-3157

RODRIGUEZ JF. Plasma glutathione concentrations in children infected with human immunodeficiency virus. Pediatr Inf Dis J 2000; 17, 236

SANDSTROM PA. Antioxidant defenses influence HIV- 1 replication and associated cytopathic effects. Free Radic Biol Med 1999; 17, 279

SAVARINO A. Modulation surface transferrin receptors in lymphoid cells de novo infected with HIV type- 1. Cell Biochem Function 1999; 17,47

STEBENS WE. Oxidative stress in viral hepatitis and AIDS. Exp Mol Pathol 2004 Oct; 77(2):121-32

SEMBA RD, NEVILLE MC. Breast- feeding, mastitis, and HIV transmission: nutritional implications. Nutr Rev 1999; 57:146-53

SEMBA RD, TANG AM. Micronutrients and the pathogenesis of human immunodeficiency virus infection. Br J Nutr 1999; 81(3):181

SEMBA RD., BLOEM M. Nutrition and Health in Developing Countries. Humana Press Inc., Totowa- New Jersey, 2001, 592

SHILS ME, SHIKE M, ROSS AC, CABALLERO B, COUSINS RJ. Modern Nutrition in health and disease. Lippincott Williams and Wilkins. 2005; 2146

SIMMA I, AHMED S, CARLSSON L, THOMPSON RHP. Breast milk zinc and copper concentrations in Bangladesh. Br J Nutr 1990; 63:91-6

SNOOK JT, PALMQUIST DL, MOXON AL, CANTOR AH, VIVIAN VM. Selenium content of foods purchased or produced in Ohio. Journal of the American Dietetic Association 1987; 87, 744- 749

SWART PJ, KUIPERS EM, SMIT C, VAN DER STRATE BW, HARMSEN MC, MEIJER DK. Lactoferrin. Antiviral Activity of lactoferrin. Adv Exp Med Biol 1998; 443: 205-213

THOMAS T, AMOMKUL P, MBORI- NGACHA D. The Kisumu Breastfeeding Study. Kisumu, Kenya, 2005

TANCHOU V, DECIMO D, PECHOUX C, LENER D, ROGEMOND V, BERTHOUX L, OTTMAN M, DARLIX JL. Role of N- terminal Zinc- finger of

human immunodeficiency virus type-1 nucleocapsid protein in virus structure and replication. J Virol 1999; 72(5), 4442

UNAIDS/ UNICEF/ WHO. AIDS Epidemic Update 2007. http://www.unaids.org/eu/KnowledgeCentre/HIVData/EpiUpdate/EpiUpdArchieev/2007/Default.asp Hit: January 2008

VAN DE PERRE P, SIMONON A, HITIMANA DG, et al. Infective and anti-infective properties of breastmilk from HIV- 1 infected women. Lancet 1993; 341:914-8

VAN DAEL P, DEELSTRA H. Selenium. Int J Vitam Nutr. Res. 1993; 63(4), 312

WHO- World Health Organization. Breast Feeding and HIV. Progress in Human Reproduction Research. Geneva, Switzerland. WHO Press Release No. 20, May 4, 1992

WILLUMSEN JF, NEWELL ML, FILTEAU SM, COUTSOUDIS A, DWARIKA S, YORK D, TOMKINS AM, COOVADIA HM. Variation in breastmilk HIV- 1 viral load in left and right breasts during the first 3 months of lactation. AIDS 2001; Vol 15 No 14 1896- 1897

WILLUMSEN JF, FILTEAU SM, COUTSOUDIS A, NEWELL ML, ROLLINS NC, COOVADIA HM, TOMKINS AM. Breastmilk RNA viral load in HIV- infected South African women: effects of subclinical mastitis and infant feeding. AIDS 2003, 17:407-414

ZAZZO JF, ROUVEIX B, RAJAGOPALON P, LEVACHER M, GIRARD PM. Effect of zinc on the immune status of zinc- depleted AIDS related complex patients. Clin Nutr 1989; 8:259-261