MICROALBUMINURIA, PROTEINURIA AND ASSOCIATED FACTORS AMONG HIV INFECTED CHILDREN ATTENDING HIV CARE AND TREATMENT CLINIC AT MUHIMBILI NATIONAL HOSPITAL, DAR ES SALAAM, TANZANIA

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A Dissertation report submitted for the Requirements of the Master of Science (Nephrology) Degree of the Muhimbili University of Health and Allied Sciences

Muhimbili University of Health and Allied Sciences

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CERTIFICATION

The undersigned certify that he has read and hereby recommend for submission to the Muhimbili University of Health and Allied Sciences a dissertation entitled: ***Microalbuminuria, proteinuria and associated factors among HIV infected children attending care and treatment clinic at Muhimbili National Hospital, Dar es Salaam, Tanzania,*** in partial fulfillment of the requirements for the degree of Master of Science (Nephrology) of the Muhimbili University of Health and Allied Sciences (MUHAS), Dar es Salaam, Tanzania.

Prof Eden E Maro (Supervisor)……..

Date.....

DECLARATION AND COPYRIGHT

I, **Francis Fredrick**, declare that this dissertation is my own original work and that it has not been presented and will not be presented to any other University for a similar or any other degree award.

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Date…….

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ABSTRACT

Background: Human immunodeficiency virus (HIV) infection is still a global challenge, and Sub-Sahara African countries including Tanzania contribute a great proportion in this epidemic disease**.** Children are among the most vulnerable group to HIV infection and acquire HIV infections mainly through mother to child transmission, which may occur in utero, intra-partum or through breastfeeding**.** HIV complications are attributed to either opportunistic infections or direct effects of virus to tissues. In the era of Highly Active Anti-retroviral Therapy (HAART), there has been a reduction in the incidence of opportunistic infections with reduction of both morbidity and mortality, which has paved way for manifestation of non-infectious complications including renal complications**.** The most common renal complication of HIV infection is HIV associated nephropathy whose earliest manifestation is microalbuminuria and proteinuria, and children of African descent are at risk of this complication. There is limited information on HIV associated nephropathy in children in Tanzania, although some studies have documented its occurrence in adults.

Objective: This study determined the prevalence of proteinuria and microalbuminuria and associated factors among HIV infected children attending HIV care and treatment clinic at Muhimbili National Hospital.

Methodology: This is a cross-sectional hospital based study which was conducted among HIV infected children aged between one year and 14 years. The children were consecutively recruited from paediatric HIV care and treatment clinic between February, 2011 and March, 2011. Standardized questionnaires were used to collect socio-demographic characteristics and clinical presentation of study participants. Spot urine was used to determine microalbuminuria and proteinuria using Microalbumin 2-1 combo test strips and dipsticks respectively. Serum creatinine, blood urea nitrogen, white blood cell count, CD4 count, haemoglobin level and renal ultrasound were performed.

Results: A total of 240 HIV infected children were recruited into this study. One hundred and seven (44.6%) were female. Mean age of participants was 7.6 ± 3.7 years. Two hundred and nineteen (91.2%) were using anti-retroviral drugs. Forty nine children (20.4%) had microalbuminuria and 17 (7.1%) had proteinuria. Prevalence of proteinuria was noted to be

significantly higher among children aged 10 years and above ($p<0.05$). Children with CD4+ percent < 25% were noted to have higher prevalence of microalbuminuria ($p<0.01$) and proteinuria ($p<0.01$) as compared to those with CD4+ above 25%. Children with microalbuminuria had significantly lower mean CD4+ count (937.4 ± 595.3 cell/µL) than those without microalbuminuria (1164.7 ± 664.3 cell/µL), $p<0.05$, similarly children with proteinuria had significantly lower mean CD4+ count (675.5 ± 352.3 cell/µL) as compared to those without proteinuria (1152 ± 662 cells/µL), $p<0.001$. Of the 153 children who had renal ultrasound performed 28 (11.7%) had increased cortical echogenicity, however no difference were noted on renal ultrasound findings between children with microalbuminuria/proteinuria and those without.

Conclusion: Microalbuminuria and proteinuria were prevalent in HIV infected children, and children with low CD4 counts and percent were more likely to have both microalbuminuria and proteinuria.

Recommendation: HIV infected children should be screened for proteinuria at initial visit and annually thereafter.

LIST OF ABBREVIATIONS

ACEI	Angiotensin Converting Enzyme Inhibitors
AIDS	Acquired Immuno-Deficiency Syndrome
ARV	Anti-Retroviral Therapy
ESRD	End Stage Renal Disease
CKD	Chronic Kidney Disease
CTC	Care and Treatment Clinic
DNA	Deoxy-ribonucleic Acid
FGF	Fibroblast Growth Factors
GFR	Glomerular Filtration Rate
HAART	Highly Active Anti-Retroviral Therapy
HIV	Human Immuno-Deficiency Virus
IDSA	Infectious Diseases Society of America
IL	Interleukin
INF	Interferon
KDOQI	Kidney Disease Outcome Quality Initiative
MNH	Muhimbili National Hospital
NKF	National Kidney Foundation
NNRTI	Non-nucleoside Reverse Transcriptase Inhibitors
NRTI	Nucleoside Reverse Transcriptase Inhibitors
PCR	Polymerase Chain Reaction
PGDF	Platelet Derived Growth Factors
TGF	Transforming Growth Factor
USA	United States of America
WHO	World Health Organization

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CHAPTER ONE: INTRODUCTION AND LITERATURE REVIEW

1.0 HIV associated Nephropathy

Human immunodeficiency virus (HIV) infection is still a global challenge, and Sub-Sahara African countries including Tanzania contribute a substantial proportion of this the epidemic.[1, 2] In the year 2001 it was estimated that 3 million children under the age of 15 years were infected with HIV globally, out of which 2.6 million were in Sub-Saharan Africa.[1] The number of newly infected children in Sub-Saharan Africa was estimated to be 390,000 in 2008.[3] In Tanzania it was estimated that 130,000 and 150,000 children between the age of 0-14 years were living with HIV in 2001 and 2008 respectively.[4]

Children acquire HIV infections mainly through mother to child transmission, and this may occur in utero, intra-partum or through breastfeeding. It is estimated that 30-40% of the infection in children occur in utero and 60-70% occur intra-partum.[5, 6] Other modes of HIV transmission have been reported to account for infection in children which include unsafe health care, traditional scarification, sexual abuse and early sexual debut.[7]

HIV infection result in a multisystem disease, its complications can be attributed to either opportunistic infections as a result of weakened immune system or direct effects of virus to tissues.[6] In the era of Highly Active Anti-retroviral Therapy (HAART), there have been dramatic changes in the incidence of opportunistic infections with reduction of morbidity and mortality, which has paved way for manifestation of non-infectious complications including renal complications.[8-10]

Kidneys have been reported to be commonly involved in HIV infection. The spectrum of kidney involvement is broad ranging from exacerbation of common kidney diseases to acute and chronic conditions.[11] HIV associated nephropathy is a chronic disorder which has been described in children and adult with AIDS.[12 13] This condition is characterized by proteinuria and the rapid development of end stage renal disease (ESRD).[14]

1.1 Prevalence of microalbuminuria and proteinuria

Several factors including race, population under study and use of therapy for HIV influence the prevalence of HIV associated nephropathy. Although HIV associated nephropathy has been widely described among HIV infected adults with high preponderance among patients of African descent, children and adolescents of African descent are reported also to be at higher risk.[13] A prevalence of 10-15% was reported by Strauss et al in a study from a population of mainly African-American children,[12] while Steel-Duncan and colleagues reported a prevalence of 3.3% in a cohort study conducted among children in Jamaica.[15]

In Africa few studies have been conducted to document the prevalence of HIV associated nephropathy, however proteinuria which may be the first sign of this condition has been reported widely. Anochie et al analyzed 10 children with HIV associated nephropathy in a cohort study which was conducted among children at University of Harcourt Teaching Hospital, Nigeria, where 40% had proteinuria and 20% had microscopic haematuria.[16] Esozobor et al reported a prevalence of proteinuria of 20.5 % among HIV infected children as compared to 6% among their un-infected controls in a study which was conducted in Nigeria.[17] Mistry reported a prevalence of microalbuminuria of 25% among HIV infected children as compared to 1% in HIV un-infected controls in a study conducted at Chris Hani Baragwanath hospital in Soweto, South Africa.[18] Dimock et al in Bethesda, USA[19] reported a prevalence of microalbuminuria of 15% in adolescents and youth who acquired HIV infection in infancy or childhood.

Pathogenesis of HIV associated kidney disorders

The pathogenesis of HIV associated nephropathy is largely unknown. Nephropathy is reported to result from infection of renal epithelial and mesangial cells with HIV in a genetically susceptible host.[20] HIV is reported to enter epithelial cells by three routes which are binding of cell-free virus to specific receptors, fusion with infected mononuclear cells and direct transfer on intimate contact between infected mononuclear cells and epithelial cells.[21]

Renal epithelial cells do not express T-cell glycoproteins CD4, CCR5 and CXCR4 which are responsible for HIV-1 entry, instead HIV-1 uses other chemokine receptors for infection such as chemokine C-C motif ligand 3 (CCL3) or Duffy antigen receptor complex.[20] Infection of podocytes and mesangial cells with HIV induce an inflammatory response with expression of cytokines (TNFα, IL-2, and IFN-γ), growth factors (PDGF, TGF-β, and FGF-2), chemokines and adhesion molecules.[14, 18, 20]

Inflammatory mediators released following HIV-1 infection result in a combination of pathologic changes in the renal tissues including sclerosis and apoptosis. Infection of podocytes with HIV is postulated to induce these structures to de-differentiate and proliferate resulting in glomerular capillary collapse.[14, 20] Deposition of immune complexes in the renal tissues as a result of interaction between circulating immunoglobins with HIV are also reported to contribute in renal pathology.[22]

1.2 Clinical presentation

HIV associated nephropathy is usually a late event in the natural history of HIV disease, and usually occurs in poorly controlled HIV infection with elevated viral loads and low CD4 counts.[11, 23] However HIV associated nephropathy can be part of early manifestation of HIV infection and it has been reported to appear at seroconversion.[23, 24] Typical presentation of HIV associated nephropathy include proteinuria, which ranges from isolated proteinuria to nephrotic range, and significant renal dysfunction.[11] Other features of nephrotic syndrome including oedema and hypoalbuminemia may rarely be present, lack of other features of nephrotic syndrome are unclear but is postulated to result from salt wasting syndrome in HIV associated nephropathy or increased oncotic pressure from increased serum immunoglobulin.[23] Haematuria and hypertension are usually not present and when present one should think of an alternative cause for renal dysfunction.[24]

1.3 Diagnostic evaluation of HIV associated nephropathy

Diagnosis of HIV associated nephropathy is confirmed by renal biopsy. Histological features include a combination of glomerular and tubular lesions involving global or focal segmental glomerulosclerosis, microcystic transformation of renal tubules and interstitial inflammation followed by fibrosis with lymphocytic infiltration.[14, 15, 23] Glomerular collapse with podocytes hypertrophy of have been reported in adults with HIV associated nephropathy, however, many children have unique presentation of mesangial hyperplasia with no development of collapsing glomerulopathy.[14] Other histological features of HIV associated nephropathy include atrophied tubular epithelium and tubuloreticular inclusions on electron microscopy.[23]

Kidneys of patients with HIV associated nephropathy are enlarged and have hyperechogenic pattern on ultrasound.[23, 24] Renal dysfunction is not uncommon with raised serum creatinine which corresponds to reduced glomerular filtration rate. [15, 16, 24] Urine examination in these patients commonly show proteinuria, other findings on urinalysis include hyaline casts and rarely haematuria.[11, 24]

Clinical diagnosis of HIV associated nephropathy may be considered in the absence of renal biopsy, with the presence of the following features; HIV infected patient, African descent, proteinuria, raised serum creatinine and enlarged hyperechogenic kidneys on ultrasound, after exclusion of other causes of renal failure.[24]

Treatment of HIV associated nephropathy

The treatment of HIV associated nephropathy currently based on observational studies, because no randomized controlled trial results on its interventions has been published.[25] Anti-retroviral therapy has been reported in a number of observational studies to influence the course of HIV associated nephropathy and has been linked to reduced incidence of ESRD in HIV infected patients.[26, 27] Peters et al reported improvement of renal functions after two years of using HAART in a study conducted among HIV infected Ugandans in the Home- Based AIDS care clinical trial.[28]

Other interventions which are reported to be effective in treating HIV associated nephropathy include Angiotensin-converting enzyme inhibitors (ACEIs) and steroids.[23] Pathogenesis of HIV associated nephropathy is linked to increased cellular synthesis of transforming growth factor β (TGF-β) which is increased by angiotensin II making ACEIs logical therapy for HIV associated nephropathy.[22, 23]

Observational studies have reported corticosteroids to be beneficial in treating HIV infected patients with nephropathy.[22, 29] Use of corticosteroids in HIV associated nephropathy is associated with reduction of serum creatinine and proteinuria.[30] However the use of steroids have been reserved as second line therapy particularly for patients with deteriorating renal functions despite being on HAART.[23, 31]

Antiretroviral use and renal dysfunction

Use of highly active anti-retroviral drugs can result in a various toxic effects with damage to the kidneys, these effects may manifest as acute renal failure, kidney stones, tubular necrosis or chronic renal failure.[32] Three groups of HAART are in use in anti-retroviral therapy in Tanzania which includes nucleoside/nucleotide reverse transcriptase inhibitors (NRTIs), non-nucleoside reverse transcriptase inhibitors (NRTIs) and protease inhibitors (PIs).

Nucleoside analogs include zidovudine, lamivudine, emtricitabine, abacavir, didanosine, stavudine and zalcitabine. Tenofovir is the only nucleotide analog approved for treatment of HIV. Nucleoside analogs have a favorable renal safety profile, with only rare reports of direct toxicity to the renal tubules.[33] Lamivudine and Abacavir have been implicated in Fanconi syndrome[33] and Abacavir have been reported to cause immuno-allergic interstitial nephritis.[32-34]

Tenofovir is an acyclic phosphonate that has been associated with development of renal tubular damage. Tenofovir toxicity result in proximal tubulopathy which may present with proteinuria, hypokalemia, glycosuria, hypophosphatemia, phosphaturia and aminoaciduria.[33, 32, 34] Mild elevation in serum creatinine was reported after starting

tenofovir with no clinically significant nephrotoxicity in retrospective study conducted in Toronto, Canada between January 2002 and December 2003 by Antoniou et al.[35] In a multicentre prospective study which was conducted in HIV infected children in Spain, Soler-Palacin et al reported abnormal urine osmolality, decreased tubular phosphate absorption and proteinuria, no significant changes in creatinine clearance was reported.[36] Efavirenz and nevirapine are two commonly used non-nucleoside analogs, NNRTIs as a group has minimal nephrotoxic potential. These drugs are primarily metabolized by hepatic cytochrome P450 and are not actively secreted in the kidney.[33]

Protease inhibitors do not exhibit profound nephrotoxic effects as they are metabolized by hepatic cytochrome P450.[33] Indinavir is the most frequently PI linked with renal and urologic side effects, which manifest with reversible acute renal failure, chronic renal failure, leukocyturia, microhaematuria, mild proteinuria, nephrolithiasis, papillary necrosis and crystalluria.[33 34] Andiman et al reported a twofold increased risk of renal dysfunction among HIV infected children and youth, exposed to tenofovir and/or indinavir as compared to participants exposed to other ARVs drugs in a Paediatric AIDS Clinical Trial Group multi centre study in United states.[37]

Chronic kidney disease and HIV infection

Chronic kidney disease (CKD) is defined as presence of kidney damage or glomerular filtration rate (GFR) of less than 60mL/min/1.73 m^2 for 3 months or more, irrespective of the diagnosis.[38] Classification of CKD in children is similar to adult classification which is based on National Kidney Foundation's Kidney Disease Outcomes Quality Initiative (NKF-K/DOQI) classification scheme.[39] According to K/DOQI classification CKD is characterized by stage 1 which is mild disease through stage 5 which is end stage renal disease.

In children glomerular filtration rate is estimated using a modified formula which was established and modified by Schwartz et al which utilizes serum creatinine and height of the child.[40, 41] Normal levels of GFR in children varies with age, gender and height, and increases with age reaching approximately adult level at the age of 2 years, therefore the

GFR ranges used to define the 5 CKD stages applies only to children aged 2 years and above.[38, 39]

Table 1 National Kidney Foundation's Kidney Disease Outcomes Quality Initiative (NKF-K/DOQI) stages of chronic kidney disease[42]

Stage	Description	GFR (mL/min/1.73 m^2)
1	Kidney damage with normal or increased GFR	>90
2	Kidney damage with mild decrease in GFR	60-89
3	Moderate decrease in GFR	30-59
4	Severe decrease in GFR	15-29
5	Kidney failure	<15 or dialysis

Chronic kidney disease affects HIV infected persons and is one of the leading non-infectious complication.[43] HIV associated nephropathy is the most common cause of CKD in HIV infected patients.[22, 44] Chronic kidney disease causes increased progression to AIDS and death in HIV infected patients even in those who are on HAART.[45, 46] Kidney disease should be detected early in HIV infected patients in order to slow its progression and to improve outcome of HIV infection.

National Kidney Foundation (NKF) defines CKD by abnormal urinalysis in the form of proteinuria and estimated GFR < 60 mL/min/1.73 m^2 of at least 3 months duration.[42] This makes screening for CKD in HIV infected patients possible by evaluating for proteinuria and renal dysfunction as recommended by Association of the Infectious Diseases Society of America (IDSA).[31]

Urinary protein excretion in children is <4mg/m^2/hour or <100mg/m^2/day for both boys and girls.[38] Albumin accounts for 30-40% of urinary excreted protein, other components of urinary protein include Tamm-Horsfall protein (50%), immunoglobulins (5-10%) and light

chains (5%).[47] Proteinuria is an excretion of protein exceeding the normal values and it is a feature of chronic kidney disease and has also been reported as a predictor of ESRD.[42, 48] Albumin excretion in urine between 30 to 300 mg/day is referred to as microalbuminuria,[49] and cannot be detected by tests used for testing proteinuria. Microalbuminuria is an early marker of nephropathy in both diabetic and non-diabetic patients,[50, 51] therefore excessive albumin excretion may signify renal glomerular disease.[52] Microalbuminuria has been reported as a predictor of proteinuria in HIV infected individuals,[53] and is associated with poorly controlled HIV infection with low CD4 count and high viral load.[54] Eighty six percent (86%) of patients who presented with microalbuminuria had HIV associated nephropathy proven by biopsy in a study which was conducted among HIV infected adults in south Africa by Han et al.[50]

CHAPTER TWO: PROBLEM STATEMENT

HIV is a significant cause of morbidity and mortality in paediatric population and most of HIV infections in children and adolescents are acquired vertically.[5, 6] The burden of HIV infection in children in Tanzania is enormous, although morbidity attributable to opportunistic infections is reported to have gone down following the use of HAART, these children are susceptible to complications not related to opportunistic infections as a result of improved survival and the fact that these children are growing with HIV infections.

HIV infected children of African descent have been reported to have similar risk as adults for renal complications of HIV/AIDS. The most common renal complication of HIV infection is HIV associated nephropathy which manifest as proteinuria, ranging from mild to nephrotic range proteinuria, as an early presentation,[13] followed later with other features of renal dysfunction.

Several studies have been conducted on renal complications of HIV infection in sub-Saharan Africa; most of these studies were conducted among adults with few focusing on children. In Tanzania proteinuria has been reported to be common among HIV infected adults. There is limited information regarding HIV renal complications in children in Sub-Saharan Africa, and no study has been conducted in Tanzania on renal complications of HIV among children, although it is known that risk of these complications in children is similar to adults as reported by studies from other settings, and children contribute significantly to the burden of HIV infection.

CHAPTER THREE: RATIONALE OF THE STUDY

This study was conducted among HIV infected children attending HIV care and treatment clinic at Muhimbili National Hospital. Renal complications of HIV infection which have been reported among HIV infected adult in Tanzania, are expected to be defined in HIV infected children by the results of this study. The information provided by this study will be useful for justifying evaluation of HIV infected children for complications which are not related to opportunistic infections.

Although no definitive treatment of HIV related nephropathy and other renal complications have been established, HAART have been reported to be effective in HIV associated nephropathy for HIV naive patients, and for those on HAART, steroids and angiotensin converting enzyme inhibitors have been found to be effective. Therefore the results of this study will be important in whether there is a need to consider planning for follow up and eventually provision of treatment for these children.

This study also provided important baseline information in planning for screening of renal complications of HIV among HIV infected children, which can be cost-effectively done in our setting.

CHAPTER FOUR: OBJECTIVES

4.1 Broad objective

To determine the prevalence of microalbuminuria, proteinuria and associated factors among HIV infected children attending care and treatment clinic (CTC) at Muhimbili National Hospital (MNH).

4.2 Specific objectives

4.2.1 To determine the prevalence of microalbuminuria and proteinuria in HIV infected children attending CTC at Muhimbili National Hospital.

4.2.2 To describe renal ultrasonographic findings of HIV infected children attending CTC at Muhimbili National Hospital.

4.2.3 To determine factors associated with microalbuminuria and proteinuria among HIV infected children attending CTC at Muhimbili National Hospital.

CHAPTER FIVE: METHODOLOGY

5.1 Study design

The present investigation is a hospital based descriptive (cross-sectional) study to determine the magnitude of microalbuminuria and associated factors among HIV infected children attending CTC at MNH.

5.2 Study duration

Recruitment of participant in this research was carried out for three months from January, 2011 to March, 2011.

5.3 Study area

This investigation was carried out in paediatric HIV care and treatment clinic of Muhimbili National Hospital (MNH). Paediatric HIV care and treatment services are offered in clinics located in the out-patient clinic building. The clinic provides services on working form Monday to Friday. The clinic has two nurses, one permanent medical officer and one paediatrician. The clinic receives an average of fifteen patients per day.

5.4 Study population

All HIV infected children aged between one year and 14 years, with documented HIV positive results (HIV DNA PCR for those aged <18 months and Antibody test for those aged >18 months) attending CTC at Muhimbili National Hospital were eligible.

5.5 Inclusion criteria

The following were inclusion criteria into this study:

5.5.1 HIV infected children aged between one year and 14 years

5.5.2 Children who assented and their parents/ guardian provided written informed consent

5.6 Exclusion criteria

The exclusion criteria included the following:

5.6.1 HIV infected children with known co-existing chronic kidney disease

5.6.2 HIV infected children with urinary tract infection

5.6.3 HIV infected children with known co-existing Diabetes mellitus

5.6.4 HIV infected children with Tuberculosis

5.7 Sample size

Sample size was calculated using:

$$N = \frac{Z^2 P(1-P)}{E^2}$$

Where;

Z is percentage point corresponding to significance level. For a significance level of 5%, **Z** is 1.96

P is the prevalence of microalbuminuria in a cohort of adolescents and youth who acquired HIV infection in infancy or childhood, in Bethesda, USA which is 15%.[19]

E corresponds to maximum likely error and is 0.05

Therefore **N** is 195 and the minimum sample size was 215.

5.8 Sampling procedure

Consecutive recruitment sampling was used in this study. Recruited participants were interviewed on their socio-demographic details such as age, sex, etc. Physical examination was carried out for each participant and information regarding use of HAART was sought from their case notes.

Venipuncture was performed to all participants at the time of recruitment and blood specimens were sent to the Central Pathology Laboratory (CPL) and were analyzed for serum creatinine, blood urea nitrogen, total white blood cell count, CD4 count, and haemoglobin level.

Urine specimens were obtained by midstream clean catch and were tested for microalbuminuria, proteinuria, nitrite, leucocytes esterase and haematuria. Microalbumin 2-1 combo test were used to test urine for microalbuminuria, this test determine albumin to creatinine ratio. Proteinuria, nitrite, leukocyte esterase and haematuria were tested using URiSCAN® strips. Participants who tested positive for nitrite and leukocyte esterase tests were excluded from the study as were presumed to have urinary tract infection.

Renal ultrasound appointments were given to all participants, and parents/guardians were reminded by telephone of their appointments, however only 153 participants attended for renal ultrasound examination. Renal ultrasounds were performed by sonographer and were verified by certified radiologist. Results of renal ultrasound were communicated to the participants' parent/guardian on the day of renal ultrasound. Cortical echogenicity of the sonograms were graded relative to the liver or spleen. When echogenicity of the cortex of the kidney was less than that of liver/spleen grade 0, cortical echogenicity equal to that of liver/spleen grade 1, renal cortex more echogenic than liver/spleen grade 2 and grade 3 when renal sinus and renal cortex have equal echogenicity.

Participants were weighed using a standard beam balance (SECA®) with the participant putting on light clothing and no shoes. Older participants' weights were taken while standing on a weighing scale (SECA). Calibration of weighing scale to zero was performed on each day of recruitment**.**

Length or height of participants was measured using either a length board or stadiometer. Parents/guardians assisted in removing shoes and gently laying the child in supine position on the board, with their heads placed at 90^{o} to the fixed head piece. The investigator straightened the legs of the child at the knees and ensured that feet were at right angle to the sliding foot piece which was to be brought into contact with the child's heels.

Estimated GFR (eGFR) for all participants was calculated from modified Bedside Schwartz equation for children which utilizes two variables (i.e. serum creatinine and height), using an eGFR calculator downloadable from the web (http:www.nkepd.nih.gov/professionals/gfr_calculator/idmsa_schartz.htm) with creatinine methods traceable to Integrated Database Management System (IDMS).The validated formula is expressed as;

eGFR (mL/min/1.73 m^2) = (**0.413 x Height**) / **Serum creatinine**

DEFINITION OF TERMS

Microalbuminuria:

Urinary albumin/creatinine ratio of ≥30 mg/g was defined as abnormal excretion of albumin meeting criteria for microalbuminuria in this study.

Proteinuria:

Positive dipstick test of +1 and above for protein, corresponding to ≥ 30 mg/dL was defined as proteinuria in this study.

Renal dysfunction:

Renal dysfunction was defined by presence of proteinuria and reduced glomerular filtration rate corresponding to <60 mL/min/1.73m^2.

CHAPTER SIX: DATA MANAGEMENT AND ETHICAL CONSIDERATION

6.1 Data entry and analysis

All filled questionnaires were coded before entering into the computer using Statistical Package for Social Sciences (SPSS) version 16. Data cleaning was carried out by consistence checks. Analysis was performed using the same SPSS package. Frequency distribution and two way tables were used to summarize the data and association between independent and dependent categorical variables were determined by Chi-square X^2 and Fisher's exact tests. Student's t-test was used to determine association between means. P value of < 0.05 was considered statistically significant.

6.2 Ethical consideration

Ethical clearance and permission to conduct this study were sought from MUHAS ethical committee and MNH administration respectively. Parents/caretakers of participants were requested to sign informed consent forms prior to recruitment; assent was also sought from children aged 10 years and above. Consent and assent were sought after providing information to parents/guardians and participants regarding this study particularly benefits, risks involved and the importance of doing this study, also they were informed the procedures involved when if they chose to let their children to participate in this study.

Excluded participants received treatment in the clinic according to the existing protocols.

Results of investigations performed in this study were communicated to the parents/guardians of participants. Participants who were noted to have renal dysfunction received treatment as well as close follow up in the clinic as their results were communicated to their attending physicians.

CHAPTER SEVEN: RESULTS

Demographic and baseline characteristics of study population

During the study period two hundred and forty children were recruited into the study while 11 children were excluded from the study. Out of the excluded participants five were presumed to have urinary tract infection, two were on anti-tuberculosis drugs and urine specimen could be obtained for three participants. Of the 240 recruited participants 133 (55.4%) were males. The age of participants ranged from one year to 14 years with mean age of 7.6±3.7 years. Two hundred and nineteen (91.2%) participants were using anti-retroviral drugs out of which 129 (53%) were using stavudine, lamivudine and nevirapine combination.

Table2a Demographic and baseline characteristics of study participants

Variable	N (%)
Age	
1- < 5 years	66 (27.5)
5 - <10 years	94 (39.2)
≥10 years	80 (33.3)
Sex	
Male	133 (55.4)
Female	107 (44.6)
Using anti-retroviral (ARV)	
Yes	219 (91.2)
No	21 (8.8)
Laboratory values	
Mean serum creatinine	51.3 ± 6.8 µmol/L
Mean blood urea nitrogen	2.8 ± 0.9 mmol/L
Mean eGFR	84±14.7 mL/min/1.73 m^2
Mean white blood cell count	6.7 ± 2.8 x 10^3 cells/µL
Mean CD4 percent	31.1 ± 10%
Mean haemoglobin level	10.9 ± 1.4 g/dL

eGFR; estimated glomerular filtration rate.

Table 2b Type of Anti-retroviral used by participants

Antiretroviral combination	Number (%)
Stavudine, lamivudine, Nevirapine	129 (53.8%)
Stavudine, Lamivudine, Efavirenz	33 (13.8%)
Zidovudine, Lamivudine, Nevirapine	28 (11.7%)
Zidovudine, Lamivudine, Efavirenz	24 (10%)
Abacavir, Didanosine, Kaletra	5 (2.1%)

Prevalence of microalbuminuria and proteinuria

Forty nine children (20.4%) out of 240 had microalbuminuria. No statistically difference was noted in the prevalence of microalbuminuria between different age groups (p=0.669) and between males and females (p=0.359), table 3. The prevalence of proteinuria in this study was 7.1% (17/240) and it was noted to be higher among HIV infected children aged 120 months and above, $p<0.05$ (Figure 1). No difference was noted when proteinuria was compared according to sex, p=0.831, (table 3). There was an overlap of microalbuminuria and proteinuria whereby all participants who had proteinuria had also microalbuminuria.

Table 3 Prevalence of microalbuminuria and proteinuria

	N	Microalbuminuria N (%)	p value	Proteinuria N (%)	p value
Age					
<5 years	66	12 (18.2)		4 (6.1)	
5-10 years	94	18 (19.1)		2 (2.1)	
≥10 years	80	19 (23.8)	0.669	11 (13.8)	0.012*
Sex					
Male	133	30 (22.6)		9 (6.8)	
Female	107	19 (17.8)	0.359	8 (7.5)	0.831

*Fisher's exact test

Factors affecting prevalence of microalbuminuria and proteinuria

The percentage of CD4+ was noted to influence occurrence of microalbuminuria and proteinuria in this study. A higher proportion of children (35.9%) with microalbuminuria was noted among participants with CD4+ percent <25% as compared to those who had CD4+ percent above 25% (14.8). This difference was statistically significant, p<0.01.Proportion of children with proteinuria was also noted to be higher among those with CD4 percent below 25% (15.6%) as compared to those with CD4 percent above 25% (4.0%). This difference was also statistically significant p<0.01.

No difference was noted in the proportions of both microalbuminuria and proteinuria among children who were using anti-retroviral drugs as compared to those who were not using them.

Table 4 Factors affecting prevalence of proteinuria and microalbuminuria

	Microalbuminuria		**Proteinuria**	
Variables	N (%)	p value	N (%)	p value
CD 4+ percent				
<25%	23 (35.9)		10 (15.6)	
≥25%	26 (14.8)	0.001	7 (4.0)	0.004
Using ARV				
Yes	45 (20.5)		16 (7.3)	
No	4 (19.0)	1.00*	1 (4.8)	1.00*

*Fisher's exact test

Children with microalbuminuria had significantly lower CD4+ count (*M* = 937.4, *SD* = 595.3) than those without microalbuminuria (*M* = 1164.7, *SD* = 664.3), *t* (238) = -2.2, *p*=0.03. Similarly children with proteinuria had significantly lower CD4+ count (*M* = 675.5, *SD* = 352.3) as compared to children without proteinuria (*M* = 1152, *SD* = 662), *t* (25.6) = -5.0, p< 0.001.

Renal ultrasound features of study participants

Among 240 study participants 153 had renal ultrasound performed. Out of the 153 preformed ultrasounds 28 (11.7%) had increased cortical echogenicity. All performed renal ultrasound examination showed normal kidney size.

The renal ultrasound features did not show any difference in terms of increased echogenicity between children with and without microalbuminuria and proteinuria. Three children (10.7%) with microalbuminuria had increased echogenicity while 31 (24.8%) without microalbuminuria had increased echogenicity on renal ultrasound, p= 0.134. Two children (7.1%) of children with proteinuria had increased echogenicity as compared to 13 (10.4%) children without proteinuria, p= 0.740.

Table 5 Relationship between increased renal ultrasonographic echogenicity and presence of microalbuminuria/ proteinuria

		Proteinuria			**Microalbuminuria**	
Echogenicity	**Yes**	**No**	**p-value**	**Yes**	**No**	**p-value**
	n (%)	**n (%)**		**n (%)**	**n (%)**	
Increased	2 (7.1)	26 (92.6)		3 (10.7)	25 (89.3)	
Normal	13 (10.4)	112 (89.6)	0.740*	31 (24.8)	94 (75.2)	0.134*
		N = 153				

*Fisher's exact test

Renal function of study participants

Fourteen (5.8%) of children had moderate reduction in renal reduction in renal function as determined by calculated glomerular filtration rate eGFR (Table 6).

Table 6: Classification of participants' renal function

Category	**eGFR (ml/min/1.73 m^2)**	**n (%)**
Normal	≥ 90	83 (34.6)
Mild reduction	60-89	143 (59.6)
Moderate reduction	30-59	14 (5.8)

Participants were further classified according to National Kidney Foundation Chronic Kidney Diseases classification, 30 (12.5%) participants met criteria for CKD out of which 10 (4.2%) had stage one, 6 (2.5%) had stage 2 and 14 (5.8%) had stage 3**.**

CHAPTER EIGHT: DISCUSSION, STUDY LIMITATIONS, CONCLUSION AND RECOMMENDATIONS

8.1 Discussion

This study was conducted in a cohort of HIV infected children receiving care and treatment at Muhimbili National Hospital, participants in this study were aged between one year and 14 years. The aim was to determine the prevalence of microalbuminuria and proteinuria which are early markers of kidney damage. Prevalence of microalbuminuria of 20.4% was noted in this study, this prevalence was not seen to vary with age. The observed prevalence is comparable to 25% reported among HIV infected children in South Africa[18] and 15% in a cohort of adolescent and young adults who acquired HIV infection in utero and infancy in Bethesda, USA.[19]

Microalbuminuria has been reported to be an important early marker of kidney damage in various systemic diseases including diabetes mellitus and hypertension.[51, 55, 56] Microalbuminuria may indicate manifestation of renal complication in HIV infected patients and biopsy proven HIV associated nephropathy have been documented among HIV infected patients presenting with microalbuminuria. Han et al reported HIV nephropathy in majority of HIV infected patients who presented with microalbuminuria.[50] Finding of microalbuminuria in this study may indicate presence of early stages of kidney damage in these children which may include HIV associated nephropathy.

Proteinuria was present in 7.1% of the study participants, this is lower than the one reported by Strauss et al among African American children[57] and it is higher than 3.3% reported by Duncan-Steel et al in Jamaican children.[15] Older children (≥ 10 years) in this study had significantly higher prevalence of proteinuria as compared to younger children. This is in contrast to microalbuminuria which was present equally in all age groups and may point towards continuum of kidney damage starting with microalbuminuria and progressing to proteinuria. Szczech et al reported microalbuminuria to be an important predictor of proteinuria in HIV infected adults, in that study HIV infected patients with microalbuminuria were more likely to have proteinuria.[53] It may be logical to expect full blown HIVAN to develop in accordance to this continuum; however biopsy proven

HIVAN have been reported in Microalbuminuric patient as has been the case for proteinuria.[16, 58]

In this study microalbuminuria and proteinuria correlated inversely with CD4 percent. With low CD4 percent viral load is expected to be high and this increases likelihood of renal complications of HIV which result from infection of renal epithelial and mesangial cells with HIV.[20] In children with HIV infection CD4 percent is utilized as an indicator of immunosuppression because of age variation in CD4 count which is utilized in adults.[59] Mean CD4 count was noted to be significantly lower among children with both microalbuminuria and proteinuria this is similar to reported negative correlation between CD4 count and proteinuria in HIV infected adults.[50, 60, 61] Although the recommended marker for level of immunity in HIV infected children is CD4 percent, it is evident from this study that CD4 count may also be used to indicate likely presence of microalbuminuria and proteinuria in HIV infected children despite the existing age variation.

Prevalence of microalbuminuria and proteinuria were not different between children using ARV and those not using them. This is in contrast to finding by Esozobor et al who reported lower prevalence of proteinuria among HIV infected children using ARV as compared to those who were not on ARV in Nigeria.[17] This is an interesting finding and one may attribute this to contribution of ARV renal adverse effects which are known to present with proteinuria,[32-34] however in this study none of the participants was using tenofovir which is the most nephrotoxic ARV drug.

Similar prevalence of microalbuminuria/proteinuria between HIV infected children on ARV and those who were not using ARV indicate the existing possibility of developing renal complications with infection of renal cells with HIV even at low viral loads. Renal cells are among the reservoirs of HIV in the body, even under successful suppression of viral replication possibility of these cells being infected with HIV may still exist.[62] Loss of kidney function can occur despite use of HAART, as Choi et al reported in HIV infected adults in a study conducted in San Francisco, USA whereby HIV infected patients who had achieved durable viral suppression were observed to lose kidney function.[63] It is therefore

important for HIV care providers to be vigilant in identifying renal complication even with well controlled viral load under ARV therapy.

Abdominal ultrasound which is one method of evaluating patient with kidney diseases is not routinely performed in the care of HIV infected patients. In this study 153 out of 240 had abdominal ultrasound performed, increased renal cortical echogenicity was noted in 28 (11.7%) participants, none of the performed ultrasound revealed enlarged kidneys. Increased cortical echogenicity was equally present in proportion among participants with microalbuminuria/proteinuria as in those without.

Renal sonographic findings in HIV associated nephropathy are reported to consist of enlarged kidneys with increased echogenicity.[24] However in children normal or small kidney sizes have been reported as presentation of HIV associated nephropathy. In a study which was conducted among HIV infected children in Nigeria, Anochie et al reported ten children who had biopsy proven HIV associated nephropathy, out of which three had grossly enlarged and echogenic renal ultrasound scans, while seven scans showed normal sized kidneys with loss of cortico-medullary differentiation.[16] Similar findings were reported by Steel-Duncan et al of small and normal sized kidneys with echogenicity among HIV infected Jamaican children with biopsy proven HIV associated nephropathy.[15]

Thirty participants (12.5%) were noted to have chronic kidney disease (CKD) as defined by proteinuria and decreased glomerular filtration rate in this study, out of these participants 14 were in CKD stage three. These children were all asymptomatic, which is consistent with reported asymptomatic CKD presentation with normal blood pressure and without oedema in HIV infection even at the end stage renal failure stage.[16, 46] This silent presentation poses a challenge to clinicians who need to identify these children early enough and plan for their appropriate management and follow up because CKD result in rapid progression to AIDS in patients with HIV infection.[45, 46]

Dipstick based methods were utilized in evaluating for microalbuminuria and proteinuria. These tests can be performed at bedside and provide test results within few minutes, they do not require special skills in carrying them out and are therefore cost effective enough to

be used as screening tools in identifying HIV infected children with proteinuria. National Kidney Foundation has recommended dipstick as a screening tool for patients who are at risk of developing kidney diseases in particular patients with Hypertension and Diabetes mellitus.[42] These tests can be utilized cost effectively in screening for renal complications among HIV infected patients.

Tanzania's guideline for HIV/AIDS treatment recommends periodic screening for adverse effects of nephrotoxic ARV by measuring serum creatinine and blood urea nitrogen.[64] It was noted in this study that microalbuminuria and proteinuria which are recognized manifestations of renal damage may be present in the absence of elevated serum creatinine or BUN, these two renal function tests are usually raised with significant loss of renal function, therefore they are unreliable in early detection of kidney damage resulting from HIV infection or accompanying the use of ARV. It is worth therefore to include testing for proteinuria in HIV infected patients during first visit to the clinic and annually. Infectious Disease Society of America in its guideline for management of HIV/AIDS recommend screening for proteinuria at initial visit, and close follow up for patients with proteinuria, with further evaluation with renal ultrasound and renal biopsy for those with persistent proteinuria.[65]

8.2 Study limitations

Proteinuria and microalbuminuria were assessed only once in this study. Glomerular filtration rate was calculated using modified Schwartz formula which was validated for children with chronic diseases aged between 2 and 18 years this may have affected the calculated eGFR in this study as our participants did not have CKD and some were below the age of two years. In this study other causes of microalbuminuria and proteinuria were not looked for.

8.3 Coclusion

Microalbuminuria and proteinuria are noted to be present in HIV infected children. Dipstick urinalysis and microalbumin 2-1 combo strips which are bed side tests can be used to detect microalbuminuria which may be an earliest stage of HIV kidney

complications. Microalbuminuria and proteinuria assessment using dipstick method may be utilized in our setting as cost effective way screening for renal complications in HIV infected children. Proteinuria was more likely to be present in older children as opposed to microalbuminuria which was not age dependant. Children with low CD4 counts and percent were more likely to have both microalbuminuria and proteinuria and therefore CD4 percent and count may indicate likelihood of having kidney complications.

8.4 Recommendations

1. HIV infected children attending care and treatment clinics should be screened for microalbuminuria at initial visit and annually thereafter.

2. HIV infected children with low CD4 count should be screened for microalbuminuria and proteinuria and with subsequent follow up for those with microalbuminuria and proteinuria.

3. A large prospective study is recommended to evaluate microalbuminuria and proteinuria with a long term follow up.

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APPENDICES

10.1 QUESTIONAIRE

Title; Microalbuminuria, proteinuria and associated factors among HIV infected children attending care and treatment clinic at Muhimbili National Hospital

1. Serial number……. Date of attendance..…/……/……

2. Name of patient ……..Hosp. Reg. number……..

3. Date of birth……. /..…../……... Sex Male…….. Female……

4. Participant is on HAART; Yes…….. No……..

If yes i) Date of starting HAART……../……/……..

ii) Drugs the participant is receiving now…….

iii) Drugs which the participant started on at initiation of HAART……..

……..

4. Symptoms on attendance:

i)……..

ii)…….....

iii)……

iv)……

v)…….

5. Physical examination

Weight……. Height……

Wt/Ht (age below 5 yrs)……

Body mass index (age above 5 yrs)…….BP……

Other physical findings:

i)……

ii)……..

6. Laboratory tests results

6.1. Dipstick urinalysis:

Protein: +1..... +2..... ≥ +3.....

Red blood cells (blood):.....

Nitrite test:.....

Leukocyte esterase test:.....

6.2. Microalbuminuria strip test: Positive..... Negative.....

6.3. Serum biochemistry:

Serum creatinine:.....

Blood urea nitrogen (BUN):.....

6.4. Haematology:

CD4 count:.....

CD4 percent:.....

Haemoglobin count:.....

6.5. Viral load:

Current viral load:……..

Viral load within the past 6 months:…….

6.6 Renal ultrasound findings:

……..……...

10.2 CONSENT FORM

Informed consent form

Consent to participate in the study on microalbuminuria, proteinuria and associated factors among HIV infected children attending HIV care and treatment clinic at Muhimbili National Hospital.

My name is Francis Fredrick; I am a postgraduate student in the department of Internal Medicine at Muhimbili University of Health and Allied Sciences. As part of the training I am conducting a research on microalbuminuria and proteinuria among HIV infected children at Muhimbili National Hospital.

Purpose of study: To describe prevalence of microalbuminuria and proteinuria and their associated factors among HIV infected children at Muhimbili National Hospital paediatric care and treatment clinic.

What participation involve: If you allow your child to participate in this study, you will be interviewed for information regarding your child's illness, your child will be physically examined. Urine specimen will be requested from your child and blood will be drawn form your child for laboratory investigations.

Confidentiality: Information regarding your/your child's illness will be handled with utmost secrecy in order to maintain confidentiality.

Risk: Risks involved in this study are minimal and are mainly minor discomfort on drawing blood specimen.

Right to withdraw and alternatives: Participating in this study is completely by voluntary choice. If you choose not to participate in this study your child will continue to receive all the services that are normally provided in this hospital.

Benefits: If you agree to participate in this study, your child will benefit by knowing the status of his/her kidney function, and were necessary appropriate treatment will be provided.

In case of problem who to contact: If you have any question regarding this study you should contact Francis Fredrick on +255 718 630 160. If you have questions about your rights as a participant, you may contact Prof M. Aboud, **Chairman of Muhimbili University of Health and Allied Sciences, Research and Publication Committee, P. O. Box 65001, Dar es Salaam, Telephone number 2150302-6**

Have you understood what it means to participate in this study? Yes……. No……..
Do you agree to let your child to participate in this study? Yes……. No……

Signature of parent/guardian……
Signature of witness……... Date ……

10.3 ASSENT FORM

Informed assent form

Assent to participate in the study on microalbuminuria, proteinuria and associated factors among HIV infected children attending HIV care and treatment clinic at Muhimbili National Hospital.

My name is Francis Fredrick; I am a postgraduate student in the department of Internal Medicine at Muhimbili University of Health and Allied Sciences. As part of the training I am conducting a research in children attending care and treatment clinic at MNH looking at their kidney functions.

What participation involve: If you agree to participate in this study, I will interview you parent/guardian to get information about your illness, then I will examine you. I will need your blood and urine specimen for testing.

Confidentiality: Information regarding your illness will be handled with utmost secrecy.

Risk: Risks involved in this study are minimal and are mainly minor discomfort on drawing blood specimen.

Right not to participate: Participating in this study is completely by voluntary choice. If you choose not to participate in this study you will continue to receive all the services that are normally provided in this hospital.

Benefits: If you agree to participate in this study, you will benefit by knowing the status of your kidney function, and were necessary appropriate treatment will be provided.

Have you understood what it means to participate in this study? Yes……. No……..
Do you agree to participate in this study? Yes……. No……

Signature of participant……
Signature of witness……... Date ……

10.4 CONSENT FORM-SWAHILI VERSION

Fomu ya mzazi/mlezi

Jina langu ni Francis Fredrick, mimi ni mwanafunzi wa masomo ya uzamivu katika fani ya magonjwa ya figo katika Chuo Kikuu cha Afya Muhimbili. Ninafanya utafiti wa kuangalia matatizo ya figo kwa Watoto wenye maambukizi ya virusi vya ukimwi (VVU) wanaopata matibabu katika Hospitali ya Taifa Muhimbili.

Dhumuni la utafiti: kutambua matatizo ya figo yanayopelekea uwepo wa protini katika mkojo miongoni mwa Watoto wenye maambukizi ya VVU wanaopata huduma katika Hospitali ya Taifa Muhimbili.

Ushiriki katika utafiti: ukimruhusu mtoto wako ashiriki katika utafiti huu, utaulizwa maswali juu ya hali ya mwanao, mwanao atapimwa na utaombwa utupatie mkojo wa mtoto wako pamoja na damu kwa ajili ya vipimo.

Siri na faragha: Taarifa utakazo tupatia juu ya hali ya ugonjwa wa mwanao zitatunzwa kwa siri, na hakuna mtu yeyote atapatiwa taarifa hizi zaidi ya wahusika wa utafiti huu.

Madhara: Ushiriki wa mwanao katika utafiti huu hautegemewi kuwa na madhara yeyote zaidi ya maumivu kidogo wakati wa kuchukua damu kwa ajili ya vipimo.

Haki ya kutoshiriki: Ushiriki katika utafiti huu ni wa hiari, kama hautapenda kushiriki katika utafiti huu mtoto wako atapatiwa huduma katika hospitali kama kawaida.

Faida: Endapo utakubali mtoto wako ashiriki katika utafiti huu, atapata nafasi ya kuchunguzwa hali ya figo zake na pale itakapohitajika atapatiwa matibabu/ushauri kulingana na matatizo yatakayoonekana.

Taarifa kukiwa na tatizo: Kama una swali au unahitaji taarifa kuhusiana na utafiti huu, unaweza kuwasiliana na Mtafiti Francis Fredrick kupitia simu namba +255 718 630 160. Kama una swali juu ya haki zako kama mshiriki wa utafiti, wasiliana na Prof M. Aboud, **Mwenyekiti, Kamati ya Utafiti ya Chuo Kikuu cha Afya Muhimbili, S.L.P. 65001, Dar es Salaam, simu namba 2150302-6**

Nimepata taarifa juu ya utafiti huu, ninakubali mtoto wangu ashiriki katika utafiti.

Sahihi ya mzazi/mlezi…….

Sahihi ya shuhuda……

Tarehe……./……/2011.

10.5 ASSENT FORM-SWAHILI VERSION

Fomu ya mtoto (umri wa miaka 10 na zaidi)

Jina langu ni Francis Fredrick, mimi ni mwanafunzi wa masomo ya uzamivu katika fani ya magonjwa ya figo katika Chuo Kikuu cha Afya Muhimbili. Ninafanya utafiti wa kuangalia matatizo ya figo kwa Watoto wenye maambukizi ya virusi vya UKIMWI (VVU) wanaopata matibabu katika Hospitali ya Taifa Muhimbili.

Madhumuni ya utafiti: kutambua matatizo ya figo yanayopelekea kwa watoto wanaohudhuria kliniki hapa Muhimbili.

Ushiriki katika utafiti: ukikubali kushiriki katika huu utafiti nitamwuliza maswali mzazi/mlezi wako juu ya ugonjwa wako na kasha nitakupima. Nitahitaji damu na mkojo wako kwa ajili ya vipimo..

Siri na faragha: Taarifa za ugonjwa wako zitatunzwa kwa siri.

Madhara: Ushiriki wako katika utafiti huu hautegemewi kuwa na madhara yeyote zaidi ya maumivu kidogo wakati wa kuchukua damu kwa ajili ya vipimo.

Haki ya kutoshiriki: Ushiriki katika utafiti huu ni wa hiari, kama hautapenda kushiriki katika utafiti huu utapatiwa huduma katika hospitali kama kawaida.

Faida: Endapo utakubali kushiriki katika utafiti huu, utapata nafasi ya kuchunguzwa hali ya figo zako na pale itakapohitajika utapatiwa matibabu/ushauri kulingana na matatizo yatakayoonekana.

Nimepata taarifa juu ya utafiti huu, ninakubali kushiriki katika utafiti.

Sahihi ya mshiriki…….

Sahihi ya shuhuda……Tarehe……./……./2011.