# Etiology of acute febrile illness among out-patients seeking health care at kiteto district Hospital In Manyara Region, Tanzania.

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**Muhimbili University of Health and Allied Sciences**
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**Muhimbili University of Health and Allied Sciences**

**Department of Epidemiology and Biostatistics**

# ETIOLOGY OF ACUTE FEBRILE ILLNESS AMONG OUT-PATIENTS SEEKING HEALTH CARE AT KITETO DISTRICT HOSPITAL IN MANYARA REGION, TANZANIA

**By**

**Mercy Epafra Anga**

**A Dissertation Submitted in (Partial) Fulfillment of the Requirements for the Degree of Master of Science (Epidemiology and Laboratory Management) of**

**Muhimbili University of Health and Allied Sciences**
**October, 2019**

## CERTIFICATION

The undersigned certify that they have read and hereby recommend for accaptance by Muhimbili University of Health and Allied Sciences a dissertation entitled; **''Etiology of acute febrile illness among out-patients seeking health care at Kiteto District Hospital, Manyara Region, Tanzania'',** in (partial) fulfilment of the requirements for the degree of Master of Science (Epidemiology with Laboratory Management) of Muhimbili University of Health and Allied Science.

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**Prof. Mecky Matee**

(Supervisor)

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**Date**

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**Dr. Ahmed Abade**

(Co-supervisor)

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**Date**

## DECLARATION AND COPYRIGHT

I, **Mercy Epafra Anga,** 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.

**Signature**……. **Date**……

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## DEDICATION

I would like to dedicate this dissertation to my parents whose experience, resources and power encouraged me to do better in my studies.

I also, dedicate this dissertation to my lovely husband and my precious children Goodluck and Gladness for their patients during my absence for the entire period of my studies.

Lastly, to my adorable sisters whose love and encouragement helped me to completion of this work.

**ABSTRACT**

**Background:** Febrile illness continues to be an important public health problem especially in developing countries. This study was conducted at Kiteto District Hospital in Manyara Region to determine the etiology, associated risk factors, and management of acute febrile illness.

**Methods:** This was a hospital-based cross-sectional study conducted between January and February 2019 in Kiteto District Hospital in Northern Tanzania that involved out-patients presenting with temperature ≥38 $^{0}$C. A structured questionnaire was used to collect socio-demographic characteristics and associated risk factors *and* patients files were used to obtain clinical information and prescribed drugs. Blood was analyzed using AFI Taqman Array Cards (TAC) for detection of causative agents of febrile illness. Odds ratio and p-values were used to measure the strength of the association and statistical significant.

**Results:** A total of 426 febrile outpatients were enrolled. Their median age was 28 years, with an interquartile range of 20-38 years. A hundred and eighty four (43.2%) were detected with at least one pathogen and 40 (9.4%) had multiple infections. Non-malaria cases were 144, accounting for 34.5% of all febrile illnesses compared to malaria cases that accounted for less than 10% of the cases. Majority were detected with zoonotic bacteria; (61 (14.3%) *Brucella* spp, *Coxiella burnetii*, 17 (4.0%) *Leptospira* spp, and 51 (12.0%) dengue virus. Dengue virus and malaria were more in children aged below 14 years, whereas *Brucella* spp was more common among adults. The odds of being infected by any zoonotic bacteria were higher among livestock keepers [OR=1.64, 95%CI 1.02-2.65, p=0.042] and those using raw milk [OR=1.90, 95%CI 1.19-3.01, p= <0.000] and among people living in the same house with animals [OR=3.06, 95%CI 1.16-8.08, p =0.02]. The odds of developing malaria infection reported on those with mosquito breeding sites in the household compound [OR=2.30 95% CI 1.05-5.01, p =0.03]. Of 376 (38.0%) prescriptions given to febrile patients were inappropriately given antimalarial drugs despite negative results.

**Conclusion:** Non-malarial causes of acute febrile illness are highly prevalent and yet overlooked as compared with malaria, which is much less prevalent. The large diversity of etiologial agents underlines the need to devise a cheap and easy to use a multiplex assay.

**Keywords**: Acute febrile illness, etiology, diagnosis, management, Kiteto, Tanzania

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**LIST OF ACCRONOMY**

| | |
|---|---|
| AFI | Acute Febrile Illness |
| CDC | Center for Disease Control |
| CHIV | Chikungunya virus |
| DENV | Dengue virus |
| DNA | Deoxyribonucleic acid |
| ELISA | Enzyme Linked Immunoassay |
| ESR | Erythrocytes Sedimentation Rate |
| FUO | Fever of Unknown Origin |
| ITN | Insecticide Treated Net |
| KCRI | Kilimanjaro Clinical Research Institute |
| KDH | Kiteto District Hospital |
| LLIN | Long Lasting Insect Net |
| MRDT | Malaria Rapid Diagnostic Test |
| MOHCDGEC | Ministry of Health Community Development Gender Elderly and Children |
| NHLQATC | National Health Laboratory Quality Assuarence and training centre |
| OROV | Orthobunyavirus |
| PDC | Potentially Diagnostic Clues |
| QC | Quality control |

| | |
|---|---|
| RNA | Ribonucleic acid |
| RDT | Rapid Diagnostic test |
| RT-PCR | Real Time Polymerase Chain Reaction |
| RVF | Rift Valley Fever |
| TAC | TaqMan Array Card |
| WBC's | White Blood Cell |
| WHO | World Health Organization |

## DEFINITION OF TERMS

**Acute febrile illness** was defined as a patient with fever of 38°C or higher at presentation to health facility or history of fever that persisted for 2–7 days with no localizing cause.

**Arboviruses**: A class of viruses transmitted to humans by arthropods such as mosquitoes and ticks.

**Zoonotic diseases**: Infections that can be naturally transmissible from animals to humans and vice versa.

**Rapid Diagnostic Tests** are defined as any test yielding results within minutes and that can be performed in health centres with little infrastructure or trained personnel, preferably without electricity.

**Potentially diagnostic clues** are defined as all localizing signs, symptoms, and abnormalities potentially pointing toward a certain diagnosis

# CHAPTER ONE

## 1.0. INTRODUCTION

### 1.1. Background

Febrile illness, which has been associated with more than 200 pathogens, continues to be among the main causes of morbidity and mortality particularly in tropical and developing countries, (1). The poor state of the laboratory infrastructure of these countries, contributes to failure to determine the etiology of fever in many of the febrile cases. Studies have shown that febrile illness is caused by a wide range of pathogens including viruses, parasites, bacteria, and fungi (2), which are often ignored or under-diagnosed (3).

A large number of wide-ranging etiological agents pose major diagnostic challenges to most clinicians, especially in resource-limited countries (4). In malaria-endemic areas, most patients presenting with fever are considered to have malaria and are inappropriately given anti-malarial drugs (5). Pathogens such as *Rickettsia spp*, *Borrelia spp*, *Coxiella burnetii*, *Brucella* spp and *Leptospira spp* and arboviruses including dengue, chikungunya, and zika viruses have been associated with fever (6–8). Their occurrence varies with time and geography and therefore, necessitating continuous surveillance (9). Furthermore, new agents continue to be linked with febrile illness (10).

A group of tests including blood tests, cytology, histology, genetic testing, imaging studies and endoscopy that have been used to detect the etiology of fever (11). Even then, the etiology of a sizeable proportion of fever remains unclear (12,13).

In 2010, the World Health Organisation (WHO) replaced the strategy of antimalarial treatment for children with any non-specific febrile illness, with a policy of restricting antimalarial treatment to patients with a positive parasitological test for malaria (14). This policy has been adopted through out Africa, but its implementation face challenges include limited diagnostic capacity. The widespread introduction of antigen-based RDTs for malaria (mRDT) is now

showing clinicians that a large majority, of patients previously treated for malaria, have a non-malarial cause of febrile illness (15,16).

Despite the increase in non-malarial febrile patients in Tanzania (17–19), the actual cause remains unknown due to limited diagnostic capacity. Thus, there is a need to introduce more sensitive and specific molecular based assays that can detect simultaneously multiple aetiological agents in a timely manner. Multiplex RT-PCR has been used in the detection of emerging viral pathogens that cause febrile illnesses in sub-Sahara Africa (20), and can be used to detect other non-malarial causes of febrile illness. In this study TaqMan Array Card (TAC), which is a multplex RT PCR that can detect 26 agents, was used to determine etiology of febrile illness among patients with acute febrile illness seeking care at Kiteto District Hospital.

**1.2. Problem Statement**

In Tanzania, febrile illness is one of the leading causes of morbidity and mortality in all ages. Traditionally, most patients with febrile illnesses are as malaria cases and are treated with antimalarials. However studies have shown, a large proportion of these cases to be due to non-malarial pathogens (21–23). Hospital records obtained at Kiteto District Hospital shows that between January and August 2018 there were 24,018 fever cases, however, only 2624 (11%) of them were confirmed to be due to malaria. The etiology of the remaining 89% remained unknown due to the lack of appropriate diagnostic techniques.

Reports in Northern parts of Tanzania indicates a significant decline in the transmission of malaria in many regions, with increasing detection of leptospirosis, rickettsiosis, respiratory viruses, and arboviruses as etiologic agents of fevers (24–26). The challenge is the presentation of cases caused by the different agents is clinically indistinguishable. Overtreatment with antimalarial drugs is common, even in the setting of a negative test result, as is overtreatment with empiric antibacterial drugs. Non-malarial agents of acute febrile illness remain under-recognized and are poorly investigated.

### 1.3. Conceptual Framework

The conceptual framework in Figure 1 shows different etiologies and risk factors associated with febrile illness. Literature shows that a wide range of infectious agents such as viruses, bacteria, and parasites may cause of febrile illness, with similar clinical signs and symptoms (9,10,23,25). For proper management of febrile illness, the actual causative agent and risk factors associated with their acquisition need to be determined.

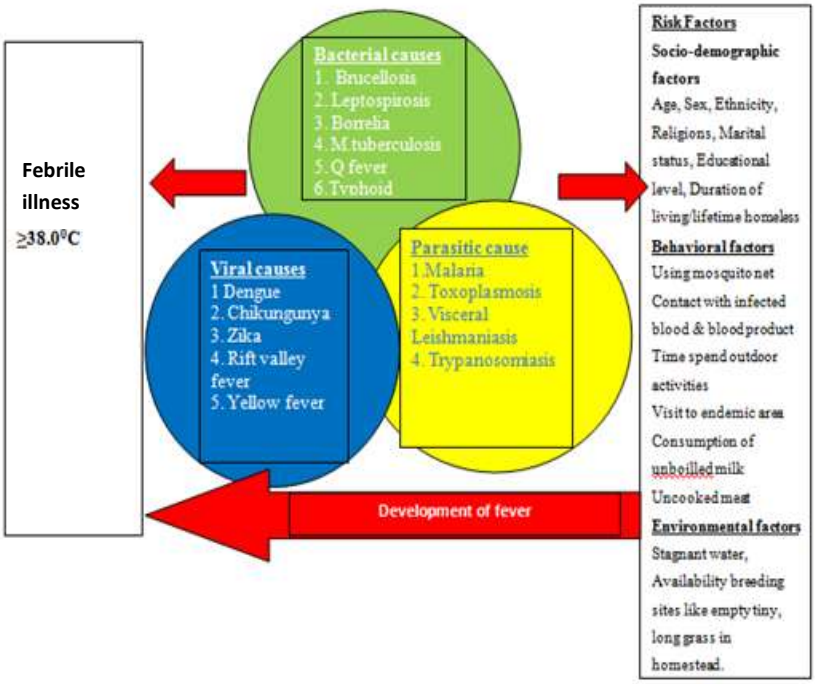



**Figure 1: Conceptual framework showing association etiology of fever (infectious agents) and their risk factors.**

### 1.4. Rationale of the study

The etiological agents associated with febrile illness are many and of varied nature, some of them new agents, which requires robust and innovative techniques that can detect a large repertoire of potential agents. This study employed a TaqMan Array Card that had a capacity to simultaneously detect 26 different pathogens, capable of comprehensively elucidate the cause of non-malarial febrile illness in the study area. This knowledge combined with the detailed investigation of the risk factors leads to the understanding of the epidemiology of febrile illness in the study area, with possible relevance to most parts of Tanzania. Therefore, the findings obtained from this study, combined with previous studies in neighboring northern Tanzania region, will provide the Ministry of Health Community Development Gender Elderly and Children (MOHCDGEC) with information necessary in planning management, prevention, and control measures. Determination of appropriateness of the medicines given to the patients is valuable for clinicians attending febrile cases, and should pave way for review policy and guidelines in management of febrile cases.

### 1.5. Research Questions

1. What is the etiology of febrile illness among out-patients seeking health care at Kiteto District Hospital in Manyara region?
2. What are the risk factors associated with the etiology of febrile illness among out-patients seeking health care at Kiteto District Hospital in Manyara region**?**
3. What is the appropriateness of prescriptions given among out-patients seeking health care at Kiteto District Hospital in Manyara region?

## 1.6. Objectives

### 1.6.1. Broad Objectives

To determine the etiology, risk factors and appropriateness on management of acute febrile illness among out-patients seeking health care at Kiteto District Hospital in Manyara Region.

### 1.6.2. Specific Objectives

1. To determine the etiology of acute febrile illness among out-patients seeking health care at Kiteto district Hospital in Manyara region.
2. To identify risk factors associated with etiology of febrile illness among out-patients seeking health care at Kiteto district Hospital in Manyara region.
3. To determine the appropriateness of prescriptions given among outpatients seeking health care at Kiteto district Hospital in Manyara region.

## 1.7. Study Hypothesis

Febrile illnesses among outpatients attending Kiteto District Hospital in Manyara region are caused by a large variety of organisms including bacteria, fungi, viruses and parasites that requires novel and robust techniques to detect them.

# CHAPTER TWO

## 2.0. LITERATURE REVIEW

### 2.1. Clinical characteristics of the patient presenting with febrile illness

Febrile illness shares clinical symptomatology characterized by fever, myalgia, headache, rash, nausea, muscle pain, abdominal pain (30,31). These symptoms are often non-specific, making it difficult to determine the causative agent (22,32,33). Resultantly, diagnosis and management of febrile illness become a challenge, especially in resource-deprived settings such as Tanzania.

### 2.2. Laboratory diagnosis

Laboratory diagnosis of acute febrile illness in developing countries is a great challenge, this is due to the unavailability of modern diagnostic equipment and technical expertise (34). Since there are more than 200 diseases that should be considered in the differential diagnosis. In many cases, the etiologies of acute febrile illness are often presented with similar clinical sign and symptom (17). Several testing algorithms approaches and protocol for diagnosis of acute febrile illness exist; however some of these approaches are not evidence-based but based on experience (34). Though applying of comprehensive and repeated history taking, physical examination and obligatory investigation give a clue in identifying etiology of fever. The availability of standardized diagnostic algorithms for the investigation of febrile illness is of paramount importance in management of febrile illness. In most African countries the exploration and investigation of acute febrile illness is limited to a blood smear and advanced to blood culture, chest radiograph, ELISA, agglutination rapid test (33,35). When the first-line tests are negative, practitioners are often unable to search for other causes of fever. However, diagnosis of acute febrile illness becomes even more challenging when the fever is due to a rare disease. Therefore, there is a need to introduce more sensitive and specific serological and molecular based assays that can detect aetiological agent of acute febrile illness even at low density (19). The use of molecular diagnostic methods for infectious diseases has improved

identification of the causative agents of febrile illness and minimizes the time for diagnosis (36). PCR reported to be used in the detection of emerging viral pathogens that cause febrile illnesses while most of the assays are time-consuming but multiplex RT-PCR is less time consuming (37). Laboratory detection of various pathogens simultaneously has been addressed by multiplex PCR whereby a number of pathogens can be detected in this method including bacteria, viruses and parasites in a single sample at the same time (37).

**2.3. The causative agents of febrile illness**

There are more than 200 infectious etiologies of fever that have been identified such as virus, bacteria, parasites, and fungi (17). Globally, malaria was the most known cause of febrile illness in tropical and subtropical regions. However, more than 125 countries have reported arboviruses such as dengue virus, chikungunya virus and zika virus to be endemic in their areas (27). A recent systematic review done in Latin America revealed that arboviruses were the main cause of febrile illness. Arboviruses had evolved and shown to have good environmental adaptability (7,31). In Africa and India studies have revealed several etiologies of febrile illness including *Salmonella typhimurium*, *Streptococcus pneumoniae*, *Staphylococcus aureus*, and *Haemophilus influenzae*, Enterobacteriaceae were bacterial infection cause a febrile illness in most of the hospitalized children (38–40). *Mycobacterium tuberculosis* has been reported to cause a febrile illness in HIV adult patients despite ART use in Uganda and Zambia (26). A study done in Kenya reported an increase of undifferentiated fever among febrile children caused by zoonotic infectious agents such as *Coxiella burnetii*, *Rickettsiae spp* and *Orientiae tshugamusi* (33), which have public important in human and animals (41,42). In Tanzania *Brucella melitensis* and *Brucella abortus* have been reported in pastoralist society being the main cause of hospitalized febrile illness patients (43). Other reported agents include *Leptospira spp*, *Coxiella burnetii*, arboviruses, *Rickettsia spp* and several other agents (37,38).

**2.4.Co-infections among patients with febrile illness**

Frequently patients with febrile illness could be infected with more than one agent (44), which may delay diagnosis and result in complications such as organ failure, bleeding, and shock (6,7). For example in dengue-endemic areas in Asia, dengue patients were found to be co-infected with leptospirosis, Q fever, and scrub typhus (45,46). In sub-Sahara Africa, Egypt reports that many febrile patients were co-infected with more than two pathogens such as *Salmonella enterica* serogroup typhi, *Leptospira* spp and *Brucella melitensis* (47). Co-infections have been found in studies conducted in Tanzania where a febrile patient with malaria could be co-infected with respiratory infection, gastroenteritis, urinary tract infection (22,48).

**2.5. Risk factors for malarial and non-malaria febrile illness**

The modes of transmission of the causative agents vary significantly and the risk of acquiring them depends on the environmental factor, behavior or socio-economical characteristics, which expose the susceptible host (6,7). Global warming, international trade, and travel have facilitated the spread of emerging and re-emerging worldwide like chikungunya, zika, and dengue, while arthropod-borne viruses have been associated with outdoor activities (49,50). Zoonotic diseases such as brucellosis have been associated with animal and animal products like blood, ingesting undercooked meat or unpasteurized milk increases the risk of contract infection (26). However, typhoid fever and rotavirus infection have been associated with poor sanitation (21,51). In a nutshell, a large range of risk factors is potentially associated with febrile illness, and knowledge on the risk factors associated with febrile illness in a particular locality is essential for control and prevention interventions.

**2.6. Prescription pattern of febrile illness**

In most malaria-endemic settings, many cases of febrile illness are regarded as malaria and are treated with antimalarials (52–54). For example, a study conducted in the Lake zone by Kazaura et al 2016 reported that 12% of under-five children with fever were wrongly given

antimalarial medication and only 14% were correctly prescribed with antibiotic based on laboratory-verified diagnosis (55). Similar observations have been cited in Kilimanjaro by Crump et al ( 2011) showing that limited diagnostic capacity leads to under-diagnosis of non-malarial agents such as *Leptospira spp, C. burnetii,* and *Rickettsia spp,* which ended up being treated empirically with antimalarials (56). The consequences of inappropriate prescriptions include treatment failure and the development of drug-resistant pathogens. The judicious use of antimicrobial agents, which requires determination of the causative infectious agents, is prudent for cost-effective management of patients.

# CHAPTER THREE

## 3.0. METHODOLOGY

### 3.1. Study design and duration

This study was a hospital-based cross-sectional study that was conducted in Kiteto District Hospital in Manyara region from January 2019 to February 2019.

### 3.2. Study Area

The study was conducted in Kiteto District, which is located in Manyara region, Tanzania. Kiteto District is one of the six districts of the Manyara Region of Tanzania. It is bordered to the north by the Simanjiro District, to the east by the Tanga Region and to the south and west by the Dodoma Region. The district headquarters are located in Kibaya. The area is semi-arid area and the main economic activities are farming and livestock keeping. The Kiteto District Hospital (KDH) has 120 beds and serves an estimated population of 244,669 (according to Census 2012). The facility also serves patients from neighboring regions such as Kondoa, Dodoma, Simanjiro, Arusha and Kilindi Tanga. The hospital has the following units; out-patient clinic, male ward, female ward, maternity ward and pediatric. The ten most prevalent diseases reported in this hospital are acute respiratory infections, urinary tract infections, typhoid fever, pneumonia, intestinal worms, malaria, diarrheal disease, anemia, diabetes, and hypertension.

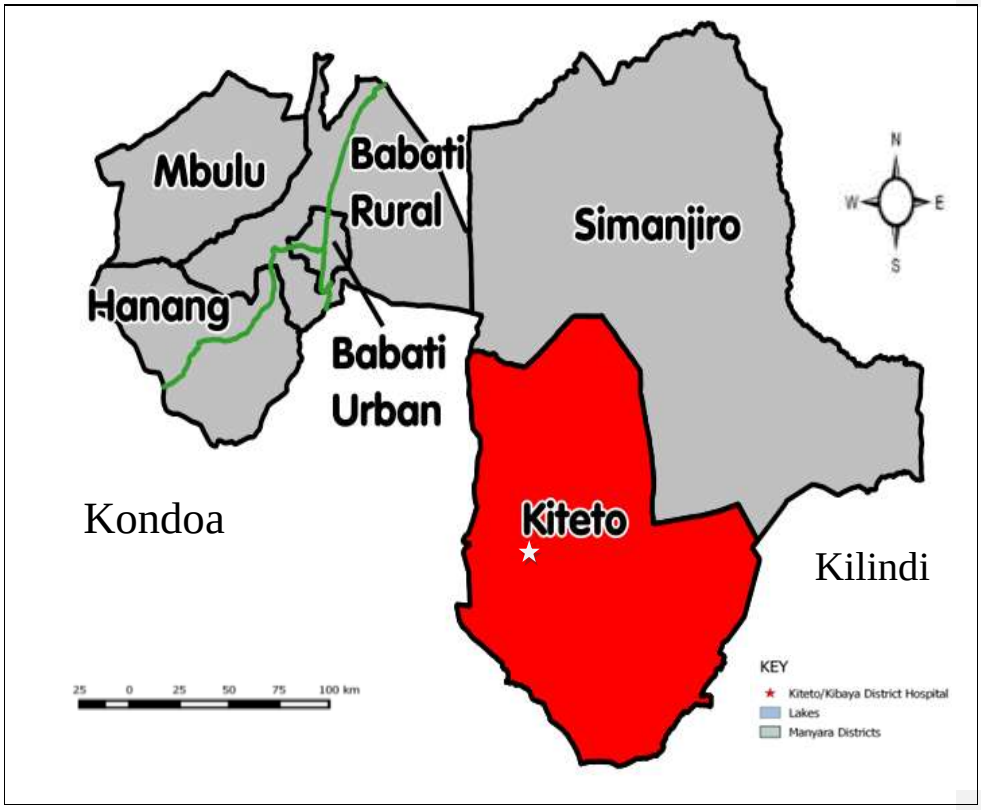



**Figure 2: A sketch map of the Manyara region showing its districts and among them is Kiteto district**

### 3.3. Study Population

Participants were patients of all ages seen at the KDH outpatient clinic with fever (≥38.0 $^{0}$C) that had persisted for 2-7 days. The study excluded patients who were severely ill and could not talk or provide informed consent.

### 3.4.1. Inclusion Criteria

Inclusion into the study was subject to obtaining informed consent from adults and assent from parents or guardians of children aged less than 15 years.

### 3.4.2 Exclusion Criteria

Patients were excluded if they either refused to consent or were too sick to consent. Children were excluded if assent was not obtained from the parent of the guardian.

### 3.5. Sample Size Calculation

The study sample size was estimated using the Kish Leslie formula (Kish, 1967) formula using the following assumption.

$$N = \frac{Z^2 P(100-P)}{\varepsilon^2}$$

Where: N= Sample size

- P= Propotion of outpatients with acute febrile illness was assumed to be 50%.
- ε= Margin of error = 5%
- Z= Standard Normal deviate (1.96 for 95% confidence level)
- N= $1.96^2$*50(100-50)/25= 384

Assume a non-response rate of 10% then the required participant was

Formular: Adjusting for non-response rate

$$n = \frac{N}{1 - 0 \cdot 1}$$

Where,

n = adjusted sample size

N= Initial sample size=384 /1- 0.1 = 426 participants

The sample size for this study was 426 people.

### 3.6. Sampling Technique

The study participants were selected by conveniently sampling strategy whereby all out-patients meeting the operational definition and that consented to participate were recruited until the required sample size was reached.

### 3.7. Variables

#### 3.7.1. Dependent Variable

Detection of etiological agents of acute febrile illness including viruses, bacteria, and parasites.

#### 3.7.2. Independent Variables

The independent variables were sociodemographic factors, which included age, sex, marital status, education level, occupation, and residence. Clinical features included fever, headache, joint pain, malaise, back pain, nausea, sore throat, abdominal pain, loss of appetite, chill, resto-ocular pain, dizziness, and diarrhea. Social and behavioral factors included using a treated mosquito net, contact with infected blood/body product or body fluid, time spend outdoors activities, visit/live endemic area, ingesting uncooked/undercooked dairy products such as milk or meat and animal husbandry. Environmental factors included house surrounded by shrub and stagnant water.

### 3.8. Data Collection

Patients were interviewed using a semistructured questionnaire to obtain information regarding socio-demographic characteristics, clinical history, associated risk factors, and retrieval of patients files were used to obtain prescribed drugs after being seen by clinicians.

#### 3.8.1. Blood collection and processing

A trained phlebotomist aseptically collected 2.5 mL of venous blood in EDTA tube. The collected blood samples were aliquoted at KDH and stored at -20 $^{0}$C until the day of processing. The blood samples were transported in a frozen state in a cool box to the

Kilimanjaro Clinical Research Institute (KCRI) and those which turn positive were sent to the National Health Laboratory Quality Assurance and Training Centre (NHLQATC) for confirmation.

**3.8.2. Laboratory Procedures**

TaqMan Array Card (TAC), which is a real-time PCR test, was used for simultaneous detection of 26 pathogens including 15 viruses, 8 bacteria, and 3 protozoa of particular relevance for Sub-Sahara Africa counties (Table 1).

**3.8.3. Malaria Rapid Diagnostic Test (MRDT)**

A drop of the patient's blood was collected from a venous blood specimen, placed onto the cassette, and allowed to flow through an absorbent material of the cassette. Results were read after 5 minutes and were presented by the presence or absence of red bands on the test position, indicating positive and negative tests, respectively. The first red band is for control of a test and the second red band is for the test. (Control band should appear in either positive or negative test for ensuring test validity).

**3.8.4. Microscopy**

A drop of the patient's blood was spread on a glass slide to make a thick blood smear. The smear was left to dry and stained using 5% Giemsa stain for 30 minutes then washed gently in tap water and air-dried. The stained smears were then examined under a microscope at 1000-fold magnification. Malaria parasites are recognizable by their physical features (trophozoites, schizonts, and gametocytes) on the red blood cells that they have infected.

**3.8.5. TaqMan Array Cards**

TaqMan Array Card (TAC) a real-time PCR test was used for simultaneous detection of 26 pathogens comprising 15 viruses, 8 bacteria, and 3 protozoa of particular relevance to Sub-Sahara Africa (37) as shown in Appendix 5.

**3.8.5.1. Quality control for TaqMan Array Card**

Quality assurance involved checking the reagents or kits, performing investigations according to standard operating procedures and use of positive control and negative controls. The TAC positive control were synthetic constructs containing all the targets (plasmids for DNA targets and in vitro transcript for RNA viruses), which serve as standard curve material for later quantification analyses. Extraction blank was included in each batch of extraction and assayed on the card to monitor and rule out laboratory contamination. The negative control contained a master mix with nuclease-free water was run once every 10 cards.

**3.8.5.2. DNA extraction**

DNA was extracted from blood specimens using high pure viral nucleic acid large volume kit. About 2.5 ml of blood sample underwent a lysate preparation process and was then purified. Elution of nucleic acid was performed using spin columns, including a high pure extender assembly. Two extrinsic controls (Phocine Herpesvirus 1 and bacteriophage MS2) were included to monitor the extraction and amplification efficiency and assure the validity of testing results. Nucleic acid was processed on TAC using the TaqMan Fast Virus 1-Step kit (Life Technologies, Carlsbad, CA), which is a real-time system. The pathogen-specific assays were validated on analytical specimens for linearity, repeatability, reproducibility, sensitivity, and specificity. The lower limit of detection was determined for each target, and matrix inhibition was assessed. The efficiencies of the TAC assays ranged from 85 to 98%.

**3.8.5.3. TaqMan Array Card reaction**

Forty-six microliter of total nucleic acid extracts containing 0.1% blue dextran 2000 were mixed with 50µL 2x reaction mix 2µL of Taq Mix and 2µL ROX reference dye (AgPath one-step RT-PCR reagents) to make the reaction mixture final volume of 100 µL. The primer and the probe were placed on the TAC in advance, and the TAC was manufactured by life Technologies. The working concentrations for probes and primers and in the TAC assay were 250nM and 900nM, respectively. After thorough mixing, the reaction mixture was loaded into each port of the card, and the cards were

centrifuged at 1200 rpm for 1 minute twice to allow even distribution of the fluid in the reaction wells. Then the cards were sealed before being placed in the ViiA $_{TM}$ 7 real-time PCR system (Life Technologies). The cycling conditions were as follows: 50 $^0$C for 15 minutes, 95 $^0$C for 20 seconds followed by 45 two-step cycles of 3 sec at 95 $^0$C and 30 sec at 60 $^0$C. Interpretation of Laboratory results by TaqMan Based Real-Time Polymerase Chain Reaction (RT- PCR) was based on duplicate reaction, where yield amplification (Quantification cycle [Cq ]< 45) interpreted as positive (37). The lower limit of detection was determined for each target, and matrix inhibition was assessed. The efficiencies of the TAC assays ranged from 85 to 98%.

**3.9. Data management and analysis plan**

Data were coded, entered, cleaned and analyzed using Epi info version 3.5 Statistical software and STATA 13.1. Descriptive analysis was done by frequencies, means, standard deviations, and quartiles of study variables. The odds ratio was used to measure the strength of the association between outcome and exposure. Factors with p-value <0.05 were considered as having a significant association with the etiology of febrile illness. All factors with p-value ≤0.2 at bivariate analysis were entered into the unconditional logistic regression model to generate independent factors associated with the development of particular etiology of febrile illness. Any factor with a P-value of ≤0.05 at multivariate analysis was regarded as independently associated with febrile illness.

**3.10. Ethical Approval**

Ethical clearance for conducting this study was obtained from the MUHAS Senate Research and Publications Committee (DA.287/298/01A). Written informed consent was obtained from each patients agreed to participate; after been informed about the study potential benefits and harm, where participation was voluntary. For children aged less than 15 years, assent was sought from parents or accompanying guardians. Participant's information was securely stored.

# CHAPTER FOUR

## 4.0. RESULTS

### 4.1. Socio-demographic distribution of patients attended out-patient clinic seeking health care at Kiteto district Hospital in Manyara Region

A total of 426 febrile out-patients, 165 (38.7%) males and 261 (61.3%) females were recruited during the study period. Their median age was 28 years, with an interquartile range of 20-38 years (Table 1). Of them, 216 (50.7%) had no education, 238 (55.9%) were married and 342 (80.3%) had informal employment by occupation.

**Table 1: Socio-demographic characteristic of patients attended out-patient clinic seeking health care at Kiteto district Hospital in Manyara Region**

| Variable | Young Children <5 years N=23, n(%) | Older children 5-14 years N=45, n(%) | Adult ≥15 years N=358, n(%) | Total N=426 |
|---|---|---|---|---|
| **Age (years)** | | | | |
| Median | 3 | 10 | 30 | 28 |
| **Gender** | | | | |
| Male | 10 (43.5) | 34 (75.6) | 121 (33.8) | 165 (38.7) |
| Female | 13 (56.5) | 11 (24.4) | 237 (66.2) | 261 (61.3) |
| **Education level** | | | | |
| None | 22 (95.7) | 30 (66.7) | 164 (45.8) | 216 (50.7) |
| Primary | 1 (4.3) | 12 (26.7) | 131 (36.6) | 144 (33.8) |
| Secondary | NA | 3 (6.7) | 42 (11.7) | 45 (10.6) |
| College | NA | NA | 21 (5.9) | 21 (4.9) |
| **Marital status** | | | | |
| Single | 23 (100.0) | 45 (100.0) | 31 (8.7) | 99 (23.2) |
| Married | NA | NA | 238 (66.5) | 238 (55.9) |
| Cohabit | NA | NA | 67 (18.7) | 67 (15.7) |
| Widow | NA | NA | 22 (6.1) | 22 (5.2) |
| **Occupation** | | | | |
| Informalemployment | 8 (34.8) | 15 (33.3) | 319 (89.1) | 342 (80.3) |
| Formal employment | NA | NA | 28 (7.8) | 28 (6.6) |
| Children/Student | 15 (65.2) | 30 (66.7) | 11 (3.1) | 56 (13.2) |

NA= Not applicable

**4.2. Clinical characteristics of patients presenting with acute febrile illness in Kiteto District Hospital**

The chief complaints were general body malaise and fatigue 195 (45.8%), joint pain 176 (41.3%) and back pain 169 (39.7%). The mean axillary temperature was 38.4°C (range 38-40°C) (Figure 3).

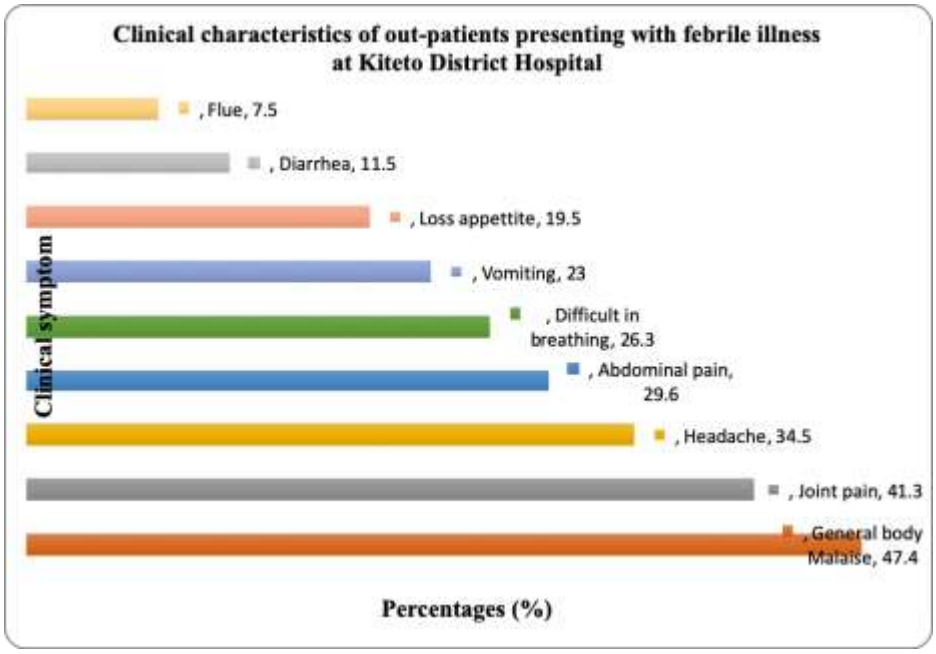



**Figure 3: Clinical characteristics of out-patients presenting with acute febrile illness at Kiteto District Hospital in Manyara region**

**4.3.The distribution of causative agents by age among out-patients seeking health care at Kiteto district hospital in Manyara Region**

Of the 426 febrile outpatients who attended Kiteto district hospital with febrile illness underwent TaqMan Array Card, of which we detected at least one pathogen in 184 (43.2%). Dengue virus and malaria were more common in children below 14 years, whereas *Brucella spp* was more common among adults. Of the 426 samples tested for malaria, only 24 (5.6%) tested positive for microscopy (Table 2).

**Table 2: Age-specific causative agent of febrile illness among out-patients seeking health care at Kiteto District Hospital in Manyara Region**

| Micro-organism | Young children <5 years, N=23, n(%) | Older children 5-14years N=45, n(%) | Adult ≥15 years N=358, n(%) | All age N=426,n(%) |
|---|---|---|---|---|
| *Brucella* spp | 2 (8.7) | 7 (15.6) | 52 (14.5) | 61(14.3) |
| Dengue virus | 5 (21.7) | 8 (17.8) | 38 (10.6) | 51 (12.0) |
| *Coxiella burnetii* | 0 (0.0) | 1(2.2) | 22 (6.1) | 23 (5.4) |
| *Leptospira* spp | 0 (0.0) | 1 (2.2) | 16 (4.5) | 17 (4.0) |
| *P.falciparum(PCR)* | 3 (13.0) | 6 (13.3) | 28 (7.8) | 37 (8.7) |
| *P. falciparum (Microscopy)* | 5 (21.7) | 8 (17.8) | 15 (4.2) | 24 (5.6) |
| *Salmonella typhi* | 2 (8.7) | 4 (8.9) | 33 (9.0) | 39 (9.2) |

**4.4. Multiple infection detected in patients among patients presenting with febrile illness at Kiteto District Hospital in Manyara region**

One hundred and forty-four (33.8%) patients were infected with one pathogen, 37 (8.7%) by two pathogens and (0.7%) by three pathogens. The majority of patients found to be infected by bacterial diasease were co-infected by viral disease by 4.7% (Figure 4).

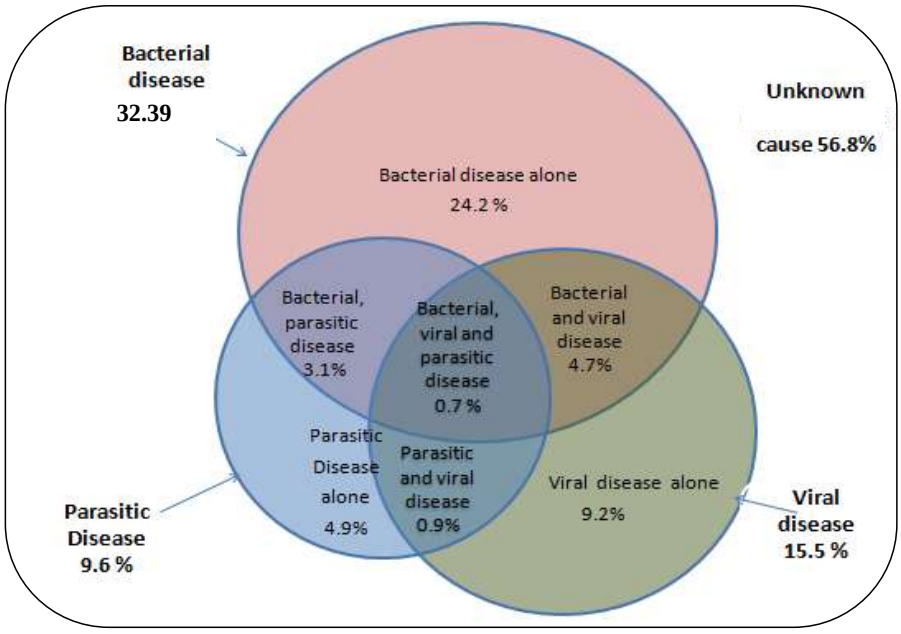



**Figure 4: Occurrence of multiple infections among out-patients presenting with febrile illness at Kiteto District Hospital in Manyara region**

**4.5. Factors that were associated with malaria**

The odds of having malaria was 2.3 times higher among those reported their house compound was surrounded by grasses as mosquito breeding site [OR=2.30 95% CI 1.05-5.01, p value=0.03]. Other factors were not statistically significant (Table 3).

**Table 3: The risk factors associated with detection of parasites causing malaria among out-patients presenting with febrile illness at Kiteto District Hospital in Manyara region**

| | Malaria parasites detection | | | |
|---|---|---|---|---|
| **Risk factors** | Positive,n(%) N=37 | Negative,n(%) N=389 | **COR (95%CI)** | **P-Value** |
| **Age group (yrs)** | | | | |
| <5 | 3 (13.0) | 20 (87.0) | Reference | |
| 5-14 | 6 (13.3) | 39 (86.7) | 1.02 (0.21-4.52) | 0.97 |
| ≥15 | 28 (7.8) | 330 (92.3) | 0.57 (0.15-2.02) | 0.38 |
| **Sex** | | | | |
| Male | 18 (10.9) | 147 (89.1) | 1.56 (0.79- 3.07) | 0.195 |
| Female | 19 (7.3) | 242 (92.7) | Reference | |
| **Education level** | | | | |
| None | 20 (9.3) | 196 (90.7) | 2.04 (0.26-16.02) | 0.50 |
| Primary | 15 (10.4) | 129 (89.6) | 2.32 (0.29-18.58) | 0.43 |
| Secondary | 1 (2.2) | 44 (97.8) | 0.45 (0.02-7.64) | 0.58 |
| Colledge | 1 (4.8) | 20 (95.2) | Reference | |
| **Occupation** | | | | |
| Informal employment | 27 (7.9) | 315 (92.1) | 1.11 (0.25-4.95) | 0.887 |
| Formal employment | 2 (7.14) | 26 (92.9) | Reference | |
| Children/Student | 8 (14.29) | 48 (85.7) | 2.16 (0.43-10.96) | 0.350 |
| **Distance from forest to compound** | | | | |
| <1km | 27 (6.9) | 362 (93.1) | Reference | |
| >1km | 10 (27.0) | 27 (73.0) | 0.20(0.09-046) | 0.000 |
| **Use mosquito repellent** | | | | |
| Yes | 7(10.8) | 58(89.23) | Reference | |
| No | 30(8.3) | 331 (91.69) | 0.75(0.32-1.79) | 0.52 |
| **Compound surrounded by stagnant water body** | | | | |
| Yes | 33 (9.09) | 330 (90.91) | 1.5 (0.5-4.3) | 0.48 |
| No | 4 (6.35) | 59 (93.65) | Reference | |
| **Use of Insecticide treated net** | | | | |
| Yes | 10 (8.1) | 113 (91.9) | Reference | |
| No | 27 (8.9) | 276 (91.1) | 1.11 (0.51-2.36) | 0.93 |
| **Window with wire mesh** | | | | |
| Yes | 10 (10.20) | 88 (89.80) | Reference | |
| No | 27 (8.23) | 301 (91.77) | 1.53 (0.74-3.16) | 0.25 |
| **House sorrounded by grasses** | | | | |
| Yes | 27 (7.5) | 335 (92.5) | 1.43 (0.19-2.95) | 0.03 |
| No | 10 (15.6) | 54 (84.4) | Reference | |

**4.6. Factors that were associated by dengue fever**

Dengue infection was 1.25 time more likely to have had history of travel to high risk area (Dar es Salaam and Tanga [OR=1.25, 95%CI 1.06-2.54, p value=0.025]. Males were more likely to test positive than females, however the association was not statistically significant (Table 4).

**Table 4: The risk factors of dengue fever among out-patients presenting with febrile illness at Kiteto District Hospital in Manyara region.**

| | Dengue | | | |
|---|---|---|---|---|
| **Risk factors** | Positive n (%)N=51 | Negative, n (%) N=375 | COR (95%CI) | P-Value |
| **Age-group (yrs)** | | | | |
| <5 | 5 (21.7) | 18 (78.3) | Reference | |
| 5-14 | 8 (17.8) | 37 (82.2) | 0.43 (0.15 -1.74) | 0.69 |
| ≥15 | 38 (10.6) | 320 (89.4) | 0.42 (0.15-1.20) | 0.08 |
| **Sex** | | | | |
| Male | 25 (15.2) | 140 (84.9) | 1.61 (0.90 - 2.91) | 0.11 |
| Female | 26 (10.0) | 235 (90.0) | Reference | |
| **Education level** | | | | |
| None | 31 (14.4) | 185 (85.7) | 3.35 (0.43-25.88) | 0.25 |
| Primary | 14 (9.7) | 130 (90.3) | 2.15 (0.26-17.29) | 0.47 |
| Secondary | 5 (11.1) | 40 (89.0) | 2.50 (0.27-22.86) | 0.42 |
| College | 1 (4.8) | 20 (95.2) | Reference | |
| **Occupation** | | | | |
| Informal employment | 41(11.99) | 301(88.01) | 1.4(0.33-3.93) | 0.84 |
| Formal employment | 3 (10.71) | 25 ( 89.29) | Reference | |
| Children/Student | 7 (12.50) | 49 (87.50 ) | 1.2(0.28-5.00) | 0.81 |
| **House surrounded by grasses** | | | | |
| Yes | 45 ( 12.40) | 318 (87.60) | 1.3(0.54-3.3) | 0.51 |
| No | 6 (9.52 ) | 57( 90.48 ) | Reference | |
| **Distance from forest to household compound** | | | | |
| < 1 km | 36 (9.3) | 353(90.8) | 1.14(1.01-3.13) | 0.000 |
| > 1km | 15 (40.5) | 22 (59.5) | Reference | |
| **Use of mosquito repellent** | | | | |
| Yes | 7 (10.77) | 58 ( 89.23) | Reference | |
| No | 44( 12.19) | 317 (87.81) | 1.15(0.49-2.78) | 0.75 |
| **Household compound surrounded with stagnant water body** | | | | |
| Yes | 44 (12.12 ) | 319 (87.88) | Reference | |
| No | 7 ( 11.11) | 56 (88.89) | 1.1(0.47-2.57) | 0.82 |
| **History of travel to risk area** | | | | |
| Yes | 40 (78.43) | 279 (74.4) | 1.25 (0.62-2.54) | 0.025 |
| No | 11 (21.57) | 96 (25.6) | Reference | |

**4.7. Factors that were associated by Salmonellosis**

The odds of being infected by *Salmonella typhi* was 2.2 times higher among those who reported not using toilet [OR=2.2, 95%CI 1.1-4.47, p value=0.01].

**Table 5: The risk factors associated with *Salmonella typhi* among febrile casesat Kiteto District Hospital in Manyara region.**

| | Salmonellosis | | | |
|---|---|---|---|---|
| **Risk factors** | **Positive n(%) N=39** | **Negative n(%), N=387** | **COR (95%CI)** | **P-Value** |
| **Age-group(yrs)** | | | | |
| <5 | 2 (8.70) | 21 (91.30) | Reference | |
| 5-14 | 4 (8.9) | 41(91.1) | 1.02 (0.17-6.06) | 0.979 |
| ≥15 | 33 (9.2) | 325 (90.8) | 0.81 (0.23-2.84) | 0.933 |
| **Sex** | | | | |
| Male | 14 (35.9) | 151(39.02) | 0.88 (0.44-1.74) | 0.703 |
| Female | 25 (64.1) | 236(60.98) | Reference | |
| **Water treatment** | | | | |
| Yes | 11 (10.6) | 93 (24.03) | Reference | |
| No | 28 (8.7) | 294 (91.3) | 1.29 (0.59-2.59) | 0.001 |
| **Water source** | | | | |
| Yes | 8 (9.0) | 81(91.0) | Reference | |
| No | 3 (20.0) | 12 (80.0) | 1.3 (0.67-2.48) | 0.447 |
| **Hand washing after toilet** | | | | |
| Yes | 7 (5.9) | 112 (94.1) | Reference | |
| No | 23 (11.2) | 183 (88.8) | 0.32 (0.17 -0.63) | 0.001 |
| **Hand washing before eating** | | | | |
| Yes | 17 (17.0) | 83 (83.0) | Reference | |
| No | 22 (6.8) | 304 (93.3) | 0.35 (0.18 - 0.70) | 0.003 |
| **Use toilet** | | | | |
| **Yes** | 9 (23.1) | 295 (76.23) | Reference | |
| **No** | 30 (76.92) | 95 (23.77) | 2.2 (1.1- 4.23) | 0.01 |
| **Use soap during hand washing** | | | | |
| **Yes** | 14 (9.8) | 111 (90.2) | Reference | |
| **No** | 25 (8.9) | 276 (91.1) | 1.11 (0.54-2.26) | 0.78 |
| **Milk treatment** | | | | |
| **Yes** | 20 (9.3) | 195 (90.7) | Reference | |
| **No** | 19(9.0) | 192 (91.0) | 1.0(0.5-1.9) | 0.92 |

**4.8.Factors associated with febrile illness caused by bacterial zoonosis**

The odds of being infected by any zoonotic bacteria were 1.64 times higher among livestock keepers [OR=1.64, 95%CI 1.02-2.65, p value=0.042] and those using raw milk [OR=1.90, 95%CI 1.19-3.01, p-value= 0.000] and 3.06 times higher among people living in the same house with animals [OR=3.06, 95%CI 1.16-8.08, p value=0.02].

**Table 6: The risk factors associated with zoonotic bacteria infection among out-patients presenting with febrile illness at Kiteto District Hospital in Manyara region.**

| | Zoonotic bacteria | | | |
|---|---|---|---|---|
| **Risk factors** | **Positive N=94** | **Negative N=332** | **Crude OR(95%CI** | **P-value** |
| **Age-group** | | | | |
| <5 | 2 (8.7)) | 21 (91.3) | Reference | |
| 5-14 | 9 (20.0) | 36 (80.0) | 2.6 (0.52-13.32) | 0.251 |
| ≥15 | 83 (23.2) | 275 (76.8) | 3.17 (0.73-13.80) | 0.10 |
| **Sex** | | | | |
| Male | 36 (21.8) | 129 (78.2) | 0.98 (0.61-1.56) | 0.922 |
| Female | 58(22.2) | 203 (77.8) | Reference | |
| **Livestock keeper** | | | | |
| No | 37 (28.2) | 94 (71.8) | Reference | |
| Yes | 57 (19.3) | 238 (80.7) | 1.64 (1.02-2.65) | **0.042** |
| **Animal vacinated, n=295** | | | | |
| Yes | 36 (29.3) | 87(70.7) | Reference | |
| No | 58(33.7) | 114(66.3) | 1.72(1.04-3.87) | **0.04** |
| **Animal house separated, n= 295** | | | | |
| Yes | 32 (36.4) | 56 (63.6) | Reference | |
| No | 62 (30.0) | 145 (70.0) | 9.8(2.97-32.4) | **0.000** |
| **Milk treated** | | | | |
| Yes | 38 (16.9) | 187 (83.1) | Reference | |
| No | 56 (27.9) | 145 (72.1) | 3.04 (1.86-4.97) | **0.000** |
| **Using unpasteurized/ sour milk** | | | | |
| Yes | 56 (19.2) | 235 (80.8) | 1.0 (0.5-1.44) | **0.61** |
| No | 38 (28.2) | 97 (71.8) | Reference | |
| **Consume sick animal /dead** | | | | |
| Yes | 45 (29.2) | 109 (70.8) | 4.8 (2.9-7.93) | **0.000** |
| No | 49 (18.0) | 223 (82.0) | Reference | |

Zoonotic infection= *Brucella*, *Coxiella burnetii* and *Leptospira*

**4.9. Multivariate logistic regression model on risk factors associated with the etiology of febrile illness**

In multivariate logistic regression presence of grasses act as mosquito breeding sites at household compound was associated with malaria [OR=2.3, 95%CI 1.05-5.01, p-value= 0.037], using sour milk [OR= 1.50, 95%CI 1.15-3.70, p-value= 0.05 ], raw milk [OR=1.2, 95%CI 1.07-2.56, p-value- 0.001] and not habit use toilet [OR=3.1, 95% CI 2.3-6.8, p-value=0.000] were independent factors associated with febrile illness (Table 7).

**Table 7: Multivariate logistic regression model on risk factors associated with the etiology of febrile illness.**

| Variable | COR (95%CI) | P-Value | AOR(95%CI) | p-value |
|---|---|---|---|---|
| **Malaria** | | | | |
| **Presence of grasses act as mosquito breeding site** | | | | |
| Yes | 1.43 (0.19-2.95) | 0.035 | 2.3 (1.05-5.01) | 0.037 |
| No | Reference | | Reference | |
| **Distance from forest to compound** | | | | |
| <1km | Reference | | Reference | |
| >1km | 0.20 (0.09-0.46) | 0.000 | 0.7 (0.13-1.6 | 0.010 |
| **Dengue** | | | | |
| **History of travel to risk area** | | | | |
| Yes | 1.25 (0.62-2.54) | 0.025 | 1.25 (0.62-2.54) | 0.025 |
| No | Reference | | Reference | |
| **Distance from forest to compound** | | | | |
| < 1 km | 1.14 (1.01-3.13) | 0.000 | 1.02 (0.01-3.13) | 0.020 |
| > 1km | Reference | | Reference | |
| **Bacterial Zoonotic infection** | | | | |
| **Animal house separate** | | | | |
| Yes | Reference | | Reference | |
| No | 3.06 (1.16 - 8.08) | 0.023 | 1.67 (0.51-5.43) | 0.040 |
| **Animal vacinated, n=295** | | | | |
| Yes | Reference | | Reference | |
| No | 1.72 (1.04-3.87) | 0.040 | 0.65 (0.03-1.36) | 0.140 |
| **Milk treated by boiling** | | | | |
| Yes | Reference | | Reference | |
| No | 1.90 (1.19 - 3.03) | 0.007 | 5.26 (1.89-14.64) | 0.001 |
| **Using sour milk** | | | | |
| Yes | 1.64 (1.05 - 5.01) | 0.040 | 1.50 (1.15-3.70) | 0.005 |
| No | Reference | | Reference | |
| **Eating sick /dead animal** | | | | |
| Yes | 1.88 (1.18 - 2.99) | 0.008 | 0.52 (0.09-3.00) | 0.464 |
| No | Reference | | Reference | |
| **Salmonellosis** | | | | |
| **Hand washing after visiting the toilet** | | | | |
| No | 1.04 (0.21 - 1.19) | 0.11 | 2.2 (0.89-5.07) | 0.089 |
| Yes | Reference | | Reference | |
| **Habit of use toilet** | | | | |
| **Yes** | Reference | | Reference | |
| **No** | 2.2 (1.1- 4.47) | 0.01 | 3.1 (2.3-6.8) | 0.000 |

**4.10.The prescription pattern and management of febrile patient among out-patients presenting with fever at Kiteto District Hospital in Manyara region**

A total of 376 prescriptions were given to patients, of whom 156 (41.5%) were antimalarials, 97 (25.8%) antibiotics and 123 (32.7%) were a combination of antimalarial and antibiotics. Of these, 143 mRDT negatives were wrongly given antimalarials, and 97 cases were given antibiotics without having a bacterial pathogen detected (Table 8).

**Table 8: Appropriateness antimicrobial agents given to febrile patients at Kiteto District Hospital, Manyara**

| | **Antimicrobial agents** | | |
|---|---|---|---|
| **Variables** | **Antimalarial n (%)** | **Antibiotics n (%)** | **Antimalaria and Antibiotic n (%)** |
| **MRDT positive** | 13 (8.3) | 1 (1.0) | 10 (8.1) |
| **MRDT negative** | 143 (91.7) | 96 (99.0) | 113 (91.9) |
| **Total** | **156** | **97** | **123** |

# CHAPTER FIVE

## 5.0. DISCUSSION

This study aimed to identify etiology of febrile illness, risk factors and assess the management of febrile patients attending Kiteto District Hospital. With the aid of TaqMan Array Card, pathogens were identified in 184 (43.2%) patients. Surprisingly, about 60% of the patients had no pathogen detected despite not having a history of recent medication. A few of the missed cases could be related with the performance of the TAC assay, which has a sensitivity of between of 82 to 94% and specifity of 98% to 99%. However it is more likely that the responsible etiological agents were different from the screened ones, or the cause of fever was not due to microbial pathogens, warranting further investigations.

Interestingly the non-malaria cases were four times more frequent than malaria cases, accounting for about one third (34.5%) of all febrile illnesses compared to malaria cases, accounted for less than 10% of the cases. In another study in neighboring Kilimanjaro region malaria accounted for only 1.6% of febrile illness (35). Overall, the most common cause of febrile illness found this study was zoonotic bacterial that included *Brucella spp*, *C.burnetii*, and *Leptospira* spp, which collectively accounted for one out of five cases (22.1%) and arboviruses which were detected in 12% of the cases. This study found out that dengue, an arboviruses which has been reported in neighbouring Kilombero and Moshi (36,35) and more recently in Dar es Salaam and Tanga (24,57). These findings are similar to those found in a study done in Northern Tanzania that found 26.2% of febrile illness was due to zoonotic bacterial infection and 7.9% due to arboviruses (37). The predominance of non-malarial causes of febrile illness has also been reported in studies conducted in Tanga and Dar es Salaam, where bacteria such *as Rickettsia*, *Leptospira, C. burnetii* and arboviruses such as *Chikungunya* isolated from cases (23,22). Collectively these studies seem to indicate that in the North-eastern part of Tanzania, malaria is less common compared with non-malarial microbial causes of fever, which are highly prevalent and yet overlooked and underappreciated.

Some age differences were found in the occurrence of the pathogens *Dengue, Brucella* spp and *P. falciparum* were most frequent in children aged less than 14 years, while in adults were zoonotic bacteria were more frequent among adults. The latter could probably due to animal husbandry activities and consumption of unpasteurized milk (58). The predominance of Brucella spp has also been reported in previous studies in Northern Tanzania (35,59). Other studies reported *Staphylococcus aureus, Escherichia coli, Klebsiella species, Streptococcus spp, Chikungunya, Rickettsia spp, Shigella flexneri, Pseudomonas aeruginosa in children and adult* (28,35,36,48).

In this study, forty (9.4%) patients were found to be co-infected by more than one pathogen(s), with 8.7% co-infected with two pathogens and 0.7% with three pathogens. Most of the coinfections were *Brucella spp* with either *C. burnetii,* Dengue virus or *P. falciparum*. These results are relatively similar to another study done in Kilombero that reported co-infections in 0.8% of febrile patients (36) and the other two studies were done in Tanzania also reported multiple co-infections were 25.5% and 39.5% among febrile illness patients (22,60). Multiple co-infections apart from complicating diagnosis could to poor prognosis due to the inability to detect the involved pathogens (6,7).

Expectedly, the risk of acquiring zoonotic bacterial infection was live stock-keeping [OR=1.64, 95%CI 1.02-2.65, p-value= 0.042], living with animals in the same house [OR=3.06, 95%CI 1.16-8.08, p-value= 0.02], eating sick and dead animals [OR=1.88, 95%CI 1.18-2.99, p-value=0.007] and using raw milk those using raw milk [OR=1.90, 95%CI 1.19-3.01, p-value= 0.000]. These factors are known to be associated with zoonotic bacterial infections by other studies (26,58,32).

The risk of acquiring the dengue virus was to travel to risk area [OR=1.25, 95% CI 0.62-2.54, p-value =0.04], an area where cases of dengue have been reported. This finding is similar to the finding reported by other studies that found traveling to risk area is the most important risk for acquiring dengue (24,61).

In this study, malaria was associated with presence grasses which act as mosquito breeding sites at household compound [OR=2.3, 95%CI 1.05-5.01, P-value= 0.03]. These findings were in line with other studies found that house surrounded by grasses which act as breeding mosquito site, which favor transmission of malaria in the community (5). Interestingly using insecticide-treated net (ITN) [OR=1.11, 95%CI 0.51-2.36, P-value=0.79] and having windows with wire mesh [OR=1.53, 95%CI 0.74-3.16, p-value= 0.25] were not associated with decreased risk for malaria. These could be due to the outdoor acquisition of malaria and changing feedings of mosquitoes, including feeding during the day (62).

Also found out that, the risk factor associated with salmonellosis was the habit of not using toilet [OR=3.1, 95% CI 2.3-6.8, p-value=0.000]. This is keeping with another study which reported that the use of the toilet was protective for acquiring salmonellosis. Other risk factors such as hand washing after visiting the toilet, hand washing before eating and water treatment were found not significant in this study but significant to other studies (63) which is not surprising given its mode of transmission.

Concerning the appropriateness of prescriptions given to febrile patients, we found about one-third (38.0%) of the antimalarial prescriptions were given to patients with MRDT negative results, while 10 (2.7%) patients were prescribed with both antimalarial and antibiotics despite being MRDT positive. And ninety-seven 25.8% were given antibiotics with no evidence of bacterial infection. The over-prescription of antimalarials has also reported in Manyara, in which 99.6% of the individuals for malaria without having parasitemia (52).

Results of this study are similar to those among under-fives with febrile in the Lake Zone of Tanzania, which showed over-prescription of both antimalarials and antibiotics of as high as 14% (55). The consequences of inappropriate medication may include the development of drug resistance, worsening of patient outcomes and misuse of resources. Training of clinicians and providing regular supportive supervision to improve prescription tendencies, clinical outcomes and minimize the risk of drug resistance could reduce the irregular prescription of antimalarials and antibiotics.

### 5.1 The strength of the study

The study employed a robust PCR technology that could detect a wide range of pathogens relevant to febrile illness in sub-Saharan Africa. The protocol used in the study has been validated previously validate and found to be useful in endemic settings (36,37). The technique was able to detect a wide range of pathogens and revealed the dominancy of non-malarial microbial causes of illness, most of them being less regarded and overlooked compared with malaria.

### 5.2 Study limitations

The main limitation of the study was the TaqMan Array Card (TAC) results could not be used for the management of patients due to prolonged turn around time. Samples had to be transported to a distant laboratory for analysis and interpretation. Also, the etiology of febrile illness in 60% of the cases could not be found, probably due to causes other than those were screened in this study. This warrants further investigation to find the possible causes.

# CHAPTER SIX

## 6.0. CONCLUSION AND RECOMMENDATIONS

### 6.1. Conclusion

The majority of febrile illnesses in Kiteto District Hospital were due to non-malaria agents, which were detected in about one third (34.5%) of the cases compared to malaria, which was detected in less than 10% of the cases. Comparing the prescriptions given to the patients with laboratory results it was found 38.0% were inappropriately treated. The wide range of pathogens found in this study calls for a revision of guidelines and case management of patients presenting with febrile illness, especially those who will be found to have no malaria parasites.

### 6.2. Recommendations

- There is a need to devise a cheap and easy to use a multiplex assay that could detect a wide range of potential agents given the wide range of pathogens causing febrile illness.
- There is a need to find etiology of febrile illness, which could not be established in 60% of the febrile patients.
- Training of clinicians and providing regular supportive supervision is required to improve prescription tendencies, clinical outcomes and minimize the risk of drug resistance could reduce the irregular prescription of antimalarials and antibiotics.
- The community need to be sensitized on the presence of multiple pathogens causing a febrile illness in the area and risk of acquiring for them to take necessary precaution

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## APPENDICES

**Appendix 1: Informed Consent (English Version)**

**MUHIMBILI UNIVERSITY OF HEALTH AND ALLIED SCIENCES**

**DIRECTORATE OF RESEARCH AND PUBLICATION, MUHAS**

**INFORMED CONSENT FORM**

ID.NO | | | | | | |

Consent to Participate in ……..

Hello! My name is Mr/Miss … ……working on this research project with the objective of investigating the Blood infection among febrile illness patients at Kiteto District Hospital, Manyara.

**Purpose of the study**

We are determining the causes of febrile illness among non-malaria patients using a method called Multiplex TaqMan RT-PCR. Participating in this research will bring the patient to know which microorganism cause complication after laboratory investigation and the appropriate management of the case.

**What Participation Involves**

If you agree to join the study you will be required to give some blood specimen, answer questions, be examined, and interviewed, or in case a child; guardian may be asked and sample collected from child.

**Confidentiality**

All information we collect on forms will be entered into computers with only the study identification number.

**Risks**

We do not expect that any harm will happen to you because of joining this study, sometimes minor bruise and pain may occur during withdrawal of blood sample.

**Rights to withdraw and Alternatives**

Taking part in this study is completely your choice. If you choice not to participate in the study or you decide to stop participating in the study you will continue to receive all services you would normally get from this hospital. You can stop participating in this study at any time even if you have already given your consent. Refusal to participate or withdrawal from the study will not involve penalty or loss of any benefits to which you are otherwise entitled.

**Benefits**

If you agree to participate in this study, your child may benefit from knowing the causes of his/her fever. However, like all participants you will benefit from seeing a physician accordingly and being followed closely by our nurses to make sure you are getting the best available treatment.

**In case of Injury**

We do not anticipate that any injury will occur to you or your child as a result of participation in this study. However, if any physical injury resulting from participation in this research occurs, we will provide you or your child with medical treatment according to the current standard of care in Tanzania. There will be no additional compensations to you or your child.

Who to Contact

If there is any question about this study please don't hesitate to communicate to the Director of Research and Publication Dr. Bruno Sunguya, the Muhimbili University of Health and Allied Science (MUHAS), P. O. Box 65001, Dar es Salaam, Tel. no 2150302-6.

Proffessor. M.Matee,The study Supervisor (Mobile: 0713081162)

Dr. Ahmed Abade,The study Supervisor (Mobile: 0683498787)

Ms. Mercy Epafra Anga –Principal Investigator (Mobile 0653620780).

Signature:

Do you agree?

Participant agree…… Participant not agree…….

I, …….have read the contents in this form. My questions have been answered. I agree to participate in this study/ my child.

Signature of participants…….

Signature of research assistant……..... Date of signed consent…….....

**Appendix 2: Idhini Ya Muhusika (Swahili Version)**

**CHUO KIKUU CHA AFYA YA TIBA MUHIMBILI**

**KITIVO CHA SAYANSI YA AFYA YA JAMII**
**KARATASI YA USHIRIKI**

**Nambari ya utambuzi** | | | | | | |

Uthibitisho wa kushiriki ……..

Habari! Jina langu naitwa Bw/Bibi … ……nafanya kazi katika utafiti huu ambao lengo lake ni kuangalia Maambukizi katika damu miongoni mwa wagonjwa wa homa ambayo si malaria katika hospital ya wilaya Kiteto, Manyara.

**Lengo la utafiti huu**

Utafiti huu utaangalia Maambukizi katika damu miongoni mwa wagonjwa wa homa ambayo si malaria kwa kutumia njia mpya na ya kisasa ijulikanayo kama TaqMan RT-PCR. Ushiriki wako katika utafiti huu utapelekea kujua ni aina gani ya vimalea vya magonjwa ambayo imo ndani ya mzunguko wako wa damu baada ya kuchunguza damu yako Maabara, na pia itasaidia ni aina gani ya utabibu utumike katika kukuondoshea matatizo yatokanayo na mdudu husika.

**Mchango tarajiwa kutoka kwa mshiriki**

Iwapo umekubaliana na ushiriki katika utafiti huu utatakiwa usaidie kiwango kidogo cha damu, uchunguzwe afya yako na utusaidie katika kujibu maswali machache ambayo tutakuuliza. Kwa watoto mzazi/mlezi ndie atakua na jukumu la kuturuhusu kuchukua damu kwa mtoto na kujibu maswali kwa niaba ya mtoto.

**Usiri wa taarifa katika utafiti huu**

Taarifa zote zitakazokusanywa zitaingizwa katika computer pamoja na nambari ambayo mshiriki atapatiwa, majina itabakia kuwa katika makaratasi ambayo yatahifadhiwa kwa usiri mkubwa na mtafiti mkuu.

**Mazingira hatarishi**

Hatutarajii kupatwa na tatizo lolote kutokana na ushiriki wako, kama itatokea ni maumivu hafifu wakati wa kuchukua damu kutoka kwa mshiriki.

**Haki ya kujiondoa katika utafiti huu**

Kushiriki katika utafiti huu ni jambo la hiari. Kama utaamua kutoshiriki katika utafiti huu au kujiondoa katika utafiti bado utaendelea kupata haki zote za msingi amabazo kikawaida ulipaswa kupatiwa. Unaweza kujiondoa mda wowote hata kama ulishatia saini katika hati ya makubaliano. Kukataa kushiriki au hutotozwa gharama yoyote na haki zako zote utapata kama inavyostahiki.

**Faida**

Kama utachagua kushiriki katika utafiti huu, mtot wako atapata kueleweka ni tatizo gani lilipelekea homa yake. Cha Zaidi na muhimu ni kupata fursa ya kuonekana na daktari na kufatiliwa kwa ukaribu na muuguzi wetu ili kuhakikisha unapata tiba sahihi.

**Kama itatokezea ajali**

Kama tulivyosema mwanzoni kwamba hatutarajii kupatwa na tatizo lolote kutokana na ushiriki wako. Lakini iwapo ajali itatokeazea kwako au kwa mtoto wako kutokana na kushiriki kwako katika utafiti huu, uongozi unakuhakikishia kukupatia tiba kwa kuzingatia viwango vya tiba vitolewavyo Tanzania. Hakutokua na marejesho ya ziada kutokana na kupata ajali baada ya kutibiwa.

**Kwa mawasiliano**

Kama kuna swali kuhusianan na utafiti huu itakubidi kuwasiliana na
Mkurugenzi wa Utafiti na Uchapishaji Dr Bruno Sunguya - Chuo Kikuu cha Afya na Sayansi ya Tiba Muhimbili, S.L.P. 65001 DSM. Simu namba 2150302-6. Kama una maswali zaidi unaweza kuwasiliana:
Proffessor. M.Matee – Msimamizi wa Utafiti (Simu: 0713081162)
Dr. Ahmed Abade - Msimamizi wa Utafiti (Simu: 0683498787)

Ms. Mercy Epafra Anga –Mtafiti Mkuu (Simu 0653620780)

"Nimesoma na nimeelewa ombi lako, kwa hiari yangu, bila ya nguvu wala ahadi zozote nakubali kushiriki katika utafiti huu".

"Samahani, sipo tayari kushiriki ☐ Nipo tayari Kushiriki ☐

Sahihi ya mshiriki/Alama ya dole gumba……..... Tarehe.....

Sahihi ya mshiriki……Sahihi ya mwakilishi wake (Mama/Mlezi)……

Sahihi ya msaidizi mtafiti……..Tarehe ya makubaliano……

Sahihi ya muulizaji..... Tarehe .....

**Appendix 3: Clinical Information**

**1. Name of the patient.....**

2. History of fever in the preceding 24 hours; .....Yes .....No

3. Date of onset of fever ...../...../.....

4. Axillary temperature: .....$^{0}$C

5. Laboratory results

(a) MRDT result: …….Pos.....No.....

(b) Blood slide results...../200WBC

6. Prescribed drugs

(a)..... (b).....

**Symptoms information (Should be done by clinician)**

| | In relation to your current illness, when did you start to feel unwell? ...../..../..... | | | | |
|---|---|---|---|---|---|
| | Did you experience any of the following symptoms? | | | | |
| | Symptoms | Yes | No | Unknown | Comments/Specify |
| 1. | Fever | | | | Measure |
| 2. | Headache | | | | |
| 3. | Chill | | | | |
| 4. | Dizziness | | | | |
| 5. | Sore throat | | | | |
| 6. | Retro-ocular pain | | | | |
| 7. | Back pain | | | | |
| 8. | Thoracic pain | | | | |
| 9. | Malaise | | | | |
| 10. | Myalgia | | | | |
| 11. | Joint pain/Arthralgia | | | | |
| 12. | Petechial | | | | |

| | | | | | |
|---|---|---|---|---|---|
| 13. | Stiff neck | | | | |
| 14. | Abdominal pain | | | | |
| 15. | Loss of appetite | | | | |
| 16. | Nausea | | | | |
| 17. | Vomiting (non bloody) | | | | |
| 18. | Diarrhea (non bloody) | | | | |
| 19. | Bloody vomiting | | | | |
| 20. | Bloody diarrhea | | | | |
| 21. | Nasal bleeding | | | | |
| 22. | Gum bleeding | | | | |
| 23. | Cough unproductive | | | | |
| 24. | Sweating | | | | |

**Appendix 4: Questionnaire (English version)**

**PART 1: DEMOGRAPHIC CHARACTERISTIC**

1. Identification/study number----------
2. Hospital identification number----------
3. Address ----------
4. Study clinician's initials:----------
6. Sex: .....Male .....Female
7. Occupation (1) Farmer (2) Pastoralist (3) Businessman (4) Watchman/Guard (5)Employed (6) None
8. Date of birth ...../...../.....Age: .....Years.....Months
9. Date of enrolment...../...../.....
10. Name of guardian:.....
11. Phone number and contact address of patient's guardian.....

**PART 2: EPIDEMIOLOGICAL QUESTIONS**

1. Did you go to the forest for work or other reasons?
   (a) No (b) Yes (c) Don't know(DK)
2. Reasons of going to the forest **Multiple responses possible**
   Picking forest products Hunting Paddy farming GardeningSoldier/ranger
   Specify_____
3. How far away from home is the forest? Distance _____ km
4. When was the last time you went to the forest?
   a) Last night < b)1 week c) 1 to < 4 weeks d) ≥ 4 weeks e)Not sure
5. How many nights/hours did you stay in the forest, farm or garden on their last visit?
   a. Nights: b. Hours:

6. Have you travelled away from home or overseas in the 2 weeks prior to illness?(Dar or Tanga)

a.Yes b.No

7. Is there water source which is stagnant near the compound?

a.Yes b.No

8. What primary action do you take to prevent being bitten by mosquitoes?

MULTIPLE ANSWERS POSSIBLE

a. Mosquito coil b. Repellent c. Indoor spraying d. Mosquito nets e. Mosquito wire screen

9. What steps do you take to prevent dengue/malaria fever transmission?

10. Does your house has window wire mesh to prevent mosquito and other insect to enter the house? a.Yes b.No

11. Does your household have any mosquito net or hammock net that can be used for sleeping?

a. Yes b. No

12. Do you use the mosquito net?

a. Yes b. Don't know

13. What is the source of water used by family?

a. Rive r b. stream water c. Pipe water/ water vender d. Public water

14. How many minutes do you take to travel to this source of water?

a. ≤30 minute b. ≥30 minutes c. ≥1 hour

15. Do you treat water before drinking?

a. No b. Yes c. Don’t know

16. What is the mode of water of treatment are you using?

a. Boiling b. Filtration c. Chlorination d. None

17. Do you know the importance of hand washing?

a. Yes b. No

18. Do you have a toilet in your home?

a.Yes b. No

19. What kind of the toilet do you have?

a. Latrine b. Flash toilet c. On the ground d. Flying toilet

20. Do you wash hands after coming from toilet?

a. Yes b. No

21. Do you wash hands before eating any thing?

a. Yes b. No

22. How do you treat milk you produce before you consume it at home?

a. Boiling b. Chilling c. FiltrationFiltering d. No treatment

23. How frequent do you take fermented milk in the home

a. Yes b. No

24. Do you get your animals vaccinated?

a.Yes b. No

**Appendix 5: Swahili Dodoso**

**SEHEMU YA KWANZA (1): UTAMBULISHO BINAFSI**

Namba ya dodoso .....tarehe ya Mahojiano ..... ../ ..... / .....

Namba ya utambulisho hospital/ Namba ya kadi .....

Namba ya simu .....Mkoa ....., Wilaya .....,

Kijiji.....

**SEHEMU YA PILI (2). TAARIFA BINAFSI**

1. Je, wewe una umri gani (umri katika miaka kamili) .....

2. Jinsia ya mshiriki wa utafiti?

a. Kiume b. Kike

3. Ni kiwango gani cha elimu ulihudhuria?

a. Hakuna b. Msingi c. Sekondari d. Elimu ya vyuo

4. Unajishughulisha na shughuli gani kwa ajili ya kipato/ kujikimu?

a.Mfugaji b. Mlinzi c. Mfanyabiashara d. Mvuvi e. Mwajiriwa

f. Mkulima g. Nyingine (kutaja) .....

5. Je, we ni kabila gani?

a. Masai b. Mmang'ati c. Mnguu d. Mburuge e. Wengine (kutaja)

6. Hali ya ndoa ya mshiriki wa utafiti

a. aliyeolewa/Aliyeoa b. Hajaolewa/sijaolewa c. Talaka d. Mjane

e. Kukaa pamoja bila kufunga ndoa

7. Umewahi kusafiri hivi karibuni mbali na nyumbani kwako wiki mbili kabla ya kuugua?

a. Hapana b. Ndio c. Sijui

8. Je, una kawaida ya kwenda msituni kufanya kazi au matembezi?

a. Hapana b. Ndio c. Sijui

9. Zitaje sababu zinazokufanya kwenda msituni?

a. Kusanya kuni b. Kuwinda c. Kulima

d. Kusalimia ndugu waishio huko e. Kazi f. Kuchimba madini g. Kurina asali

h. Mengineyo, yataje.....

10. Je, kuna umbali gani kutoka mahali unapoishi na msitu mkubwa ulipo?

:.....km

11. Je, unakumbuka mara ya mwisho kwenda msituni?

a. Jana b. < 1 wiki c. 1 hadi < 4 wiki d. ≥ 4 wiki e. Sina uhakika

12. Je, ulikaa kwa muda gani huko msituni?

a. Usiku b. Masaa c. Sijui

13. Ulipokuwa msituni ulitumia njia yoyote kuzuia using'atwe na mbu au wadudu?

a.Ndio b.Hapana

14. Sababu za kutokutumia dawa ya kufukuza/kuua mbu au chandarua kwa kipindi ukiwa porini nini?

a. Sipendi kutumia b. Nilisahau kuchukua nyumbani d. Hakuna pakuning'iniza chandarua e. Sina pesa ya kununua chandarua g. Sijui

15. Je wewe una tumia neti unapolala?

a. Hapana b. Ndio c. Sijui

16. Uliipata wapi neti ya kuzuia mbu?

a. Hospitali/ serikali zamitaa b. Mashirika binafsi

c. Dukani/ Sokoni d. Sehemu nyingine, itaje.....

17. Neti ya kuzuia mbu ulipoipata ilikuwa imekwisha wekewa dawa tayari?

a. Hapana b. Ndio c. Sijui/ sifahamu

18. Je, ulipoipata neti ulikuta dawa ya kuweka kwenye neti ili kuua mbu?

a. Hapana b. Ndio c. Sijui/Sikumbuki/ Sifahamu

19. Mambo gani unafanya kujizuia kung'atwa na mbu?

a. Kulala ndani ya neti iliyowekwa dawa b. Dawa ya kujipaka kufukuza mbu

c. Dawa ya kupulizia ndani ninapoishi d. Wavu wa kuzuia mbu kwenye madirisha ya nyumba. e. Kuvaa nguo yenye mikono mirefu

20. Nyumba yako inamadirisha yenye wavu unaozuia mbu?

a. Hapana b. Ndio c. Sijui

21. Je mazingira yako yamezungukwa na nyasi au misitu?

a. Hapana b. Ndio

22. Nyumba yako imezungukwa na madimbwi au maji yaliyotuama?

a. Hapana b.Ndio

23. Maji ya kunywa unayotumia yanatoka wapi?

a.Mtoni b. Ziwani c. Chemichemi d. Bombani

24. Kuna maandalizi yoyote kwa maji yako ya kunywa kuhakikisha usalama?

a. Hapana b. Ndio c. Sijui

25. Je maji hayo ya kunywa unayaandaa aje kuyafanya kuwa salama?

a. Chemsha b. water guard (dawa) c. Chuja d. Sijui

26. Kabla ya kula unatabia ya kunawa mikono na maji na sabuni?

a. Hapana b. Ndio c. Sijui

27. Unapotoka chooni una nawa na maji na sabuni?

a. Hapana b. Ndio c. Sijui

28. Je nyumbani kwako umejenga choo?

a. Hapana b. Ndio

29. Choo ulichonacho ni cha aina gani?

a. shimo b.cha maji c. Porini

30. Una mifugo?

a. Hapana b. Ndio

31. Je mifugo yako inachanjo?

a. Hapana b. Ndio c. Sijui

32. Wewe mwenyewe unahusika kuhudumia mifugo yako? ( Kama sio wewe nenda swali linalofuata sivyo liruke). a. Hapana b. Ndio

33. Nani anahusika kuhudumia mifugo yako?

a. Kijana wa kazi b. Watoto c. Mume/Mke

d. Mengineyo, yataje.....

34. Umewahi kushika damu au maji yatokayo kwa mifugo, wiki mbili zilizopita kabla ya kuugua? a. Hapana b. Ndio c. Sijui

35. Nyumba ya mifugo na yako imetofautishwa?

a. Hapana b. Ndio c. Sijui

36. Je, maziwa yanaandaliwaje kabla ya kutumika kwa matumizi ya nyumbani?

a. Kuchemshwa b. Kuchujwa c. Chilling d. Hakuna

37. Je unatumia maziwa mgando?

a. Hapana b. Ndio

**Appendix 6: Table X: The range of pathogens that could be detected by the TaqMan Array Card (TAC) RT-PCR assay**

| **Viruses** | **Bacteria** | **Parasites** |
|---|---|---|
| *Chikungunya, Crimean-Congo hemorrhagic fever virus, Dengue, Ebola virus, Bundibugyo virus, Sudan virus, Hantaviruses (HTN and SEO), Hepatitis E, Marburg, Nipah virus, O'nyong virus, Rift Valley fever virus, West Nile virus, Yellow fever virus*) | *Bartonella spp., Brucella spp., Coxiella burnetii, Leptospira spp., Rickettsia spp., Salmonella enterica, and Salmonella enterica serovar Typhi, Yersinia pestis* | *Leishmania spp., Plasmodium spp., Trypanosoma brucei* |

**Appendix 7: Introduction Letter**



**MUHIMBILI UNIVERSITY OF HEALTH AND ALLIED SCIENCES**

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Ref. No. HD/MUH/T.348/2017

21st January, 2019.

Regional Administrative Secretary,
P.O.Box 310,
Babati.

**RE: INTRODUCTION LETTER**

The bearer of this letter Ms. Mercy Epafra Anga is a Postgraduate student at Muhimbili University of Heath and Allied Sciences (MUHAS) who is pursuing MSc. Epidemiology and Laboratory Management.

She is conducting a research titled ***"Assesment of Etiology of Febrile illness among outpatients seeking health in Kiteto District Hospital, Manyara Region, Tanzania"***

Kindly provide her the necessary assistance to facilitate the conduct of her study.

Thank you for your cooperation,

Sincerely,

Ms. Sharifa Kambi,
**For. Director of Postgraduate Studies**
c.c Dean, School of Public Health and Social Science.

**Appendix 8: Approval of ethical clearance**

**MUHIMBILI UNIVERSITY OF HEALTH AND ALLIED SCIENCES**

**OFFICE OF THE DIRECTOR OF POSTGRADUATE STUDIES**

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Ref. No. DA.287/298/01A/ 19th January, 2019

Ms. Mercy Epafra Anga
MSc. Epidemiology and Laboratory Management
**MUHAS**

**RE: APPROVAL OF ETHICAL CLEARANCE FOR A STUDY TITLED: "ASSESSMENT OF THE ETIOLOGY OF FEBRILE ILLNESS AMONG OUT-PATIENTS SEEKING HEALTH CARE IN KITETO DISTRICT HOSPITAL, MANYARA REGION, TANZANIA "**

Reference is made to the above heading.

I am pleased to inform you that, the Chairman has, on behalf of the Senate, approved ethical clearance for the above-mentioned study. Hence you may proceed with the planned study.

The ethical clearance is valid for one year only, from 18th January, 2019 to 17th January, 2020. In case you do not complete data analysis and dissertation report writing by 17th January, 2020, you will have to apply for renewal of ethical clearance prior to the expiry date.

Dr. Emmanuel Balandya
**ACTING: DIRECTOR OF POSTGRADUATE STUDIES**

cc: Director of Research and Publications
cc: Dean, School of Public Health and Social Sciences, MUHAS