MORBIDITY, RISK FACTORS, AND FLEA SPECIES RESPONSIBLE FOR TUNGIASIS IN SELECTED VILLAGES IN KISUMU COUNTY, KENYA

ABALLA ANDREW OKOTH (BSc. Biomedical Science)
P150/21572/2010
Department of Medical Laboratory Sciences

A research thesis submitted in partial fulfilment of the requirements for the award of the degree of Master of Science Infectious Diseases in the School of Medicine of Kenyatta University

April, 2015
DECLARATION

This thesis is my original work and has not been presented for a degree in any other University or any other award.

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

.................................................. ..................................................
Aballa Andrew Okoth P150/21572/2010

We confirm that the work reported in this thesis was carried out under our supervision as university supervisors:

Supervisors

Dr. Margaret Muturi PhD
Kenyatta University, Nairobi
Department of Medical Laboratory Science

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

Dr. Muhojo Ngethe PhD
Kenyatta University, Nairobi
Department of Pathology

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

Dr. Odada Peter Sumba PhD, MPH
Kenya Medical Research Institute (KEMRI), Kisumu

Signature..........................................
Date.............................................
DEDICATION

I dedicate this work to my late father, Samson Richard Aballa, and grandfather, Turfimo Aballa, who never got the opportunity to witness the work of my hands.
ACKNOWLEDGEMENT

I acknowledge the National Council for Science and Technology (NCST) for funding this study; Kenya Medical Research Institute (KEMRI) for approving this study; and the staff of the Zoonosis, Molecular Biology, and Serology laboratory of the Diagnostic Systems Laboratory Program (DLSP) at the Centres for Disease Control (CDC), Kisian station. I acknowledge the District Education Office (DEO) Kisumu West District, my supervisors Dr Margaret Muturi, Dr Ngethe Muhoho, and Dr Odada Peter Sumba for the invaluable advice, my family for their moral support, and residents of Otwenya for cooperating.
# TABLE OF CONTENTS

DECLARATION ............................................................................................................. i

DEDICATION ............................................................................................................... ii

ACKNOWLEDGEMENT ................................................................................................. iii

TABLE OF CONTENTS ................................................................................................. iv

LIST OF TABLES .......................................................................................................... viii

LIST OF FIGURES ......................................................................................................... ix

DEFINITION OF TERMS ............................................................................................... x

ABBREVIATIONS AND ACRONYMS ............................................................................... xi

ABSTRACT .................................................................................................................... xii

CHAPTER ONE ............................................................................................................. 1

1. INTRODUCTION ...................................................................................................... 1

1.1 Background of the Study ....................................................................................... 1

1.2 Statement of the Problem ..................................................................................... 2

1.3 Justification of the Study ..................................................................................... 2

1.4 Research Questions .............................................................................................. 2

1.5 Null Hypotheses ................................................................................................... 3

1.6 Objectives ............................................................................................................. 3

1.6.1 General Objective ............................................................................................ 3

1.6.2 Specific Objectives .......................................................................................... 3

1.7 Conceptual Framework ......................................................................................... 3

CHAPTER TWO ............................................................................................................. 4

2. LITERATURE REVIEW ............................................................................................ 4
2.1 Biology of Fleas ................................................................. 4
2.2 The Genus Tunga .............................................................. 4
2.2.1 Morphological Identification of *Tunga penetrans* and *Tunga trimamillata* .......... 5
2.2.2 Molecular Identification of *Tunga penetrans* and *Tunga trimamillata* ............... 5
2.3 Tungiasis Burden ............................................................. 6
2.3.1 Bacterial Superinfection .................................................. 6
2.4 Risk Factors for Tungiasis ................................................... 7
2.5 Diagnosis of Tungiasis ...................................................... 7
2.6 Treatment of Tungiasis ...................................................... 8

CHAPTER THREE ......................................................................... 9

3. MATERIALS AND METHODS .............................................. 9
3.1 Study Location .................................................................. 9
3.2 Research Design ............................................................. 10
3.3 Variables ....................................................................... 10
3.4 Target Population ......................................................... 10
3.5 Inclusion Criteria .......................................................... 10
3.6 Exclusion Criteria ......................................................... 10
3.7 Sample Size Calculation .................................................. 10
3.8 Sampling Techniques ...................................................... 11
3.8.1 Fishbowl Technique .................................................. 11
3.8.2 Probability Sampling Technique .................................. 11
3.9 Pilot Study .................................................................... 12
3.10 Reliability of Questionnaires ......................................... 12
3.11 Validity of Questionnaires ................................................................. 13
3.12 Data Collection Techniques .............................................................. 13
3.12.1 One on One Interviews ................................................................. 13
3.12.2 Identification of Tungiasis Cases ...................................................... 14
3.12.3 Collection of Neosomic and Free-Living Tunga Fleas ....................... 14
3.12.4 Morphological Characterisation of Collected Fleas ......................... 15
3.12.5 PCR Analysis of Fleas ................................................................. 15
3.13 Ethical Considerations ...................................................................... 16
3.14 Data Analysis ...................................................................................... 16

CHAPTER FOUR............................................................................................. 18

4. RESULTS................................................................................................. 18

4.1 Demographic Data ............................................................................... 18
4.1.1 Age of Participants ........................................................................... 19
4.1.2 Education of Household Heads ......................................................... 19
4.1.3 Occupation of Household Heads ....................................................... 19
4.1.4 History of Tungiasis ......................................................................... 20
4.1.5 Architecture of Study Houses ............................................................ 20
4.2 Prevalence of Tungiasis ....................................................................... 21
4.2.1 Prevalence by Gender ....................................................................... 21
4.2.2 Prevalence by Age ........................................................................... 21
4.3 Clinical Manifestations ....................................................................... 22
4.3.1 Parasite Load .................................................................................... 23
4.3.2 Topographical Location of Lesions .................................................... 23
4.3.3 Tungiasis Pathology ........................................................................................................... 23
4.4 Risk Factors for Tungiasis ...................................................................................................... 24
4.4.1 Population Attributable Fractions (PAFs) ................................................................. 26
4.5 Tungiasis Knowledge ......................................................................................................... 26
4.5.1 Etiological Agent ............................................................................................................ 27
4.5.2 Risk Factors ................................................................................................................... 27
4.5.3 Protective Factors .......................................................................................................... 27
4.5.4 Signs and Symptoms ..................................................................................................... 27
4.5.5 Tungiasis Knowledge Transfer ..................................................................................... 28
4.6 Tungiasis Perceptions ......................................................................................................... 29
4.6.1 Treatment Strategies .................................................................................................... 30
4.7 Vector for Tungiasis .......................................................................................................... 30

CHAPTER FIVE .......................................................................................................................... 32
5. DISCUSSION, CONCLUSIONS, AND RECOMMENDATIONS .................................. 32
5.1 Discussion .......................................................................................................................... 32
5.2 Conclusions ....................................................................................................................... 39
5.3 Recommendations ............................................................................................................. 39

REFERENCES ............................................................................................................................ 40

APPENDICES ............................................................................................................................. 45
LIST OF TABLES

Table 4.1: Demographic data: males were more than females. Nine out of 10 residents has a history of tungiasis ............................................................... 18

Table 4.2: Risk factors for tungiasis: gender, age, and flooring were major risk factors. 25

Table 4.3: Overcrowding, living in homes with sandy flooring sandy flooring, and presence of animal hosts in homes were major predictors for tungiasis ...... 26

Table 4.4: PAFs: modifying sandy floors was the best strategy for controlling tungiasis 26

Table 4.5: Tungiasis Knowledge: knowledge of tungiasis causation, risk factors, and protective factors was high. Signs and symptoms of tungiasis were poorly understood ................................................................................................................ 28

Table 4.6: Tungiasis perceptions: tungiasis negligence and stigma was evident ........... 30
LIST OF FIGURES

Figure 1.1: An integrated approach for control of tungiasis ........................................... 3

Figure 2.1: Morphological features of Siphonaptera species of fleas ................................. 4

Figure 3.1: A map of Kenya showing the location of Maseno Division and Otwenya Location ........................................................................................................... 9

Figure 4.1: Education level of household heads: 68% had primary level of education .... 19

Figure 4.2: Occupation of household heads: farming was the commonest economic activity among household heads ................................................................. 20

Figure 4.3: Architectural design of study houses: Iron sheets and mud were the commonest roofing and plastering materials ......................................................... 21

Figure 4.4: Prevalence of tungiasis: children (5-14) and the elderly (over 60 years) were high-risk groups for tungiasis ................................................................. 22

Figure 4.5: Clinical manifestations on cases: manipulated lesions was a common occurrence on cases ................................................................. 22

Figure 4.6: The 10-14 year age group was at risk of sever tungiasis infestation .............. 23

Figure 4.7: The heels of a twelve-year-old pupil form Urudi Rata village in Otwenya Location with tungiasis-induced ulcers and fissures .............................. 24

Figure 4.8: Tungiasis knowledge transfer: family members were the main source of information on tungiasis. Health professionals contributed marginally ...... 29

Figure 4.9: Flea diversity in homes: Ctenocephalid canis was the commonest flea species in homes followed by Tunga penetrans ................................................. 31
DEFINITION OF TERMS

Architecture: Conceptual structure or style in which a building is constructed with regard to place, period, or culture

Burden: Impact of a health problem in an area measured by the financial cost, morbidity, and mortality, among many other factors in a community

Causative Agent: Pathogen or biological agent that causes a disease

Dependency Ratio: Ratio of people not in the labour force (0-4 and over 60 years (dependent)) to those in the labour force, (15-59 years (productive))

Disease sequels: Abnormal body condition related to a given disease.

Morphology: Form, structure, and relationship between structures of an organism

Non-Virulent Lesions Lesions with inactive of dead female fleas. Lodged fleas do not induce serious or harmful effects

Population Attributable Fraction: Proportion of cases that will not occur, if exposure to an independent factor is reduced to a minimum

Sex Ratio: Proportional distribution of male to female residents of any give population aggregate, usually expressed as the total number of males per 100 females.

Virulent Lesions: Lesions with live female fleas. Lodged fleas induce serious or harmful effects on victims.
### ABBREVIATIONS AND ACRONYMS

<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>CDC</td>
<td>Centre for Disease Control</td>
</tr>
<tr>
<td>CNS</td>
<td>Central Nervous System</td>
</tr>
<tr>
<td>CSC</td>
<td>Centre Scientific Committee</td>
</tr>
<tr>
<td>DEO</td>
<td>District Education Officer</td>
</tr>
<tr>
<td>DC</td>
<td>District Commissioner</td>
</tr>
<tr>
<td>DLSP</td>
<td>Diagnostic Laboratory Systems Program</td>
</tr>
<tr>
<td>ERC</td>
<td>Ethics Review Committee</td>
</tr>
<tr>
<td>GoK</td>
<td>Government of Kenya</td>
</tr>
<tr>
<td>HIV</td>
<td>Human Immunodeficiency Virus</td>
</tr>
<tr>
<td>KEMRI</td>
<td>Kenya Medical Research Institute</td>
</tr>
<tr>
<td>KNBS</td>
<td>Kenya National Bureau of Statistics</td>
</tr>
<tr>
<td>PAF</td>
<td>Population Attributable Fraction</td>
</tr>
<tr>
<td>SSC</td>
<td>Scientific Steering Committee</td>
</tr>
</tbody>
</table>
ABSTRACT

Tungiasis is a neglected disease that afflicts four million Kenyans. To control its spread, there is a high demand for sustainable disease control strategies in rural endemic areas. The objective of this study was to determine the prevalence of tungiasis, associated risk factors, and the species of fleas causing it in rural western Kenya. A cross-sectional study was done in Otwenya Location, Maseno Division between August and September 2013. Seventy eight (78) randomly selected households were visited all household members (415) checked for tungiasis. Structured questionnaires were used to collect demographic and environmental data from household heads, neosomic and free-living fleas collected in houses and characterised using taxonomy and Polymerase Chain Reaction (PCR). Data analysis was done using the Statistical Package for Social Scientists (SPSS) version 17. Means and frequencies were calculated and the Fishers exact test and logistic regression with backward elimination used to identify risk factors for tungiasis. Tests for statistical significance were done at 95% confidence interval (CI). Participants were between 1 and 83 years old (females (52.8%) and males (47.2%)). In 39.7% of households, at least one case of tungiasis was found. The overall prevalence was 19.5%, peaking among 5-9 years olds (37.7%) and the elderly (60+ years) (43.8%). Tungiasis cases had between one and 53 lesions. The mean parasite load was 8.3 (males (9.6±1.6) and females (6.5±0.98), with pathological manifestations for tungiasis such as deformed toenails identified on 71.6% of cases. Approximately 19.8% had signs of bacterial infections. Living in houses with sandy flooring was a major risk factor for tungiasis (OR=11.1, p=0.01). Living in houses with animals such as dogs and cattle (OR=22.8, p=0.01) and overcrowding in houses (OR=3.5, p=0.04) were other risk factors for the disease. Stigma and negligence played a major role in the establishment and proliferation of tungiasis in the study area. Diverse species of fleas were trapped in study houses. However, *Tunga penetrans* was the only species causing tungiasis. Otwenya Location is a tungiasis endemic area. Sensitisation of residents on its control, developing outreach programs that dispel negligence and stigma, and eliminating modifiable risk factors in homes can help to lower its burden in the area.
CHAPTER ONE

1. INTRODUCTION

1.1 Background of the Study

Tungiasis is a cutaneous infestation by *Tunga penetrans* and *Tunga trimamillata*. Both sexes are hematophagous. However, only female fleas penetrate the host’s skin and feed on blood and exudates for egg maturation. Once they have penetrated the superficial layer of the skin, female fleas grow, fill with eggs, and undergo a five-stage developmental process, and induce pathological changes of the skin known as tungiasis (Franck *et al*., 2003). Eisele *et al* (2003) highlights these changes in her Fortaleza classification scheme.

Tungiasis first was diagnosed in indigenous populations in Haiti in 1525 and described scientifically in 1758 in Brazil. In Africa, tungiasis was introduced inadvertently many times on the coast of West Africa between the 17th and 18th century, establishing on the Angolan coast in 1872. The disease then spread inwards throughout Sub Saharan Africa (SSA) via slave trade, the feet of Senegalese troops, and Kenyan workers (Heukelbach, 2005; Heukelbach and Ugboroiko, 2007; Ugboroiko *et al*., 2007). The endemic zones of tungiasis are now many in impoverished communities worldwide where it is neglected.

Tungiasis is a major problem in resource-poor settings in rural Kenya. The disease has been reported over a vast epidemiological range in Central province, Coast, Western, and Nyanza provinces where over four million people are infested and 10 million people are at risk of severe infestation and suffering (Ahadi Kenya Trust, 2010). On cases, tungiasis causes debilitating sequels such as pain and ulcers, induces psychological problems, and
heightens the economic burden of the severely infested by lowering their productivity (Njau et al., 2010; WHO, 2007). This maintains a cycle of poverty that is hard to break.

1.2 Statement of the Problem

There is a high demand for sustainable disease control strategies for tungiasis in Kenya. Effective strategies could be designed only if the burden of the disease, risk factors, and the biology of fleas causing tungiasis in a target population are well understood. Such data is scarce in Maseno Division. The epidemiological information on tungiasis in the area is also scarce. The available data is based mainly on the anecdotal observations of reporters, schoolteachers, and administrative personnel. This study filled these gaps.

1.3 Justification of the Study

Tungiasis affects negatively the lives of millions of Kenyans. By inducing pathological changes of the skin, the disease lowers the mobility of cases, increases the rate of primary school dropouts, and causes a high economic burden because of loss of productivity and high cost of long-term care. The results of this study will help health professionals, policy makers, and the scientific community in general to eradicate this ectoparasitic disease not only in Maseno Division, but also in other tungiasis endemic regions in Kenya as well.

1.4 Research Questions

i. What is the prevalence of tungiasis and severity of infestation of the affected?

ii. What are the risk factors for tungiasis?

iii. What is the level of Knowledge, Attitudes, and Practices (KAP) of residents?

iv. Which species of fleas causes tungiasis?
1.5 Null Hypotheses

$H_{01}$ Tungiasis is not a serious health problem.

$H_{02}$ *Tunga trimamillata* fleas are not responsible for tungiasis.

1.6 Objectives

1.6.1 General Objective

To determine the prevalence and species of fleas causing tungiasis.

1.6.2 Specific Objectives

i. To determine the prevalence of tungiasis and the severity of infestation on victims.

ii. To determine the risk factors for tungiasis.

iii. To determine the level of Knowledge, Attitudes, and Practices (KAP) of residents.

iv. To characterise the species of fleas causing tungiasis.

1.7 Conceptual Framework

To lower the burden of tungiasis, an integrated disease control approach involving health promotion and screening, early detection of tungiasis cases, primordial prevention of the risk factors, and sensitisation on residents to improve KAP is needed. Elimination of its causative agent in homes can also lower burden and contribute to its control (Figure 1.1)

**Figure 1.1:** An integrated approach for control of tungiasis
CHAPTER TWO

2. LITERATURE REVIEW

2.1 Biology of Fleas

Fleas are holometabolous blood sucking ectoparasites that infest a variety of hosts. Scientists and taxonomists worldwide have identified around 3,000 species of fleas that infest over 95% of mammals and approximately 6% of birds (Nagy et al., 2007). Their heads are shield or helmet shaped. Their bodies are laterally compressed, hard polished, and covered with short spines and fine hairs that are directed backwards as shown in Figure 2.1. Tunga fleas run with a velocity of one millimetre per second. They jump to a height of 20 centimetres by storing and releasing energy in their cuticular springs and applying force to the ground through the ends of their tibiae (Sutton and Burrows, 2011).

Figure 2.1: Morphological features of Siphonaptera species of fleas (Dryden, 2009)

2.2 The Genus Tunga

Originally, the genus Tunga consisted of nine species of fleas with only Tunga penetrans causing tungiasis. However, in 2003, Pampiglione et al discovered a new species, Tunga
trimamillata, that also causes tungiasis. The remaining Tunga species are parasites of wild animals, and are prevalent in Asia and the Americas (Pampiglione et al., 2005). To date, Tunga trimamillata has been isolated only in Peru and Ecuador in South America. However, because humans travel to and from South America and other parts of the world, colonisation of previously non-endemic areas is probable. This calls for studies to check for presence of T. trimamillata and its possible role in tungiasis in other endemic regions.

2.2.1 Morphological Identification of Tunga penetrans and Tunga trimamillata

Many structural features differentiate Tunga penetrans from Tunga trimamillata. The presence of three rounded protuberances on the anterior end of gravid female Tunga trimamillata, visible to the naked eye, but absent on Tunga penetrans differentiates the two species. These protuberances are from the first abdominal segment surrounding the head and thorax of Tunga trimamillata (Pampiglione et al., 2003). The shape and length of maxillary palp segments, pattern of spines on the antero-medial surface of the third leg tibia, shape of abdominal spiracles, the configuration of the hood of hilla of the spermatheca, the length of claspers and aedeagus, have also been used to differentiate Tunga penetrans and Tunga trimamillata (Luchetti et al., 2003; Luchetti et al., 2005).

2.2.2 Molecular Identification of Tunga penetrans and Tunga trimamillata

To overcome the challenge of diagnosing non-gravid fleas and body fragments (Luchetti et al., 2005) developed a low-cost and sustainable PCR-RFLP analysis protocol, utilising the Internal Transcribed Spacer 2 (ITS2) ribosomal domain. The intraspecies divergence of Tunga penetrans and Tunga trimamillata ITS2 is insignificant. Interspecies divergence is 4.9% with 21 nucleotide differences and two insertions or deletions (Yao et al., 2010;
Pampiglione et al., 2009). Amplifying this gene locus, digesting using RSA1 restriction enzyme and analysis of digested bands is suitable for speciation of these related fleas.

2.3 Tungiasis Burden

Flea borne infections are emerging globally. Tungiasis in particular causes considerable morbidity to residents of impoverished communities worldwide, with symptoms such as itching, swelling, soreness, and pain reported as the lodged flea grows (Feldmeier, et al., 2006; Rathe et al., 2009; Ariza et al., 2010; Kam et al., 2010). Over time, complications such as chronic lymphedema, tissue necrosis, fissures, and hyperkeratinosis develop as the flea evokes a peripheral inflammatory process involving leukocytes and eosinophils.

2.3.1 Bacterial Superinfection

Bacterial superinfection is a common clinical observation on severely infested cases. When bacteria colonise tungiasis lesions or the biofilm around the lodged fleas, they activate polymorphonuclear leukocytes, which release inflammation mediators, cytotoxic enzymes, and oxygen radicals that damage adjacent host tissues. Suppuration of lesions and pustules that rupture to form deep ulcers then develop (Kirketerp-møller et al., 2008; Ferreira et al., 2009; Swaminathan et al., 2005). Extraction of neosomic fleas using contaminated pins and sharp equipment such as razors and thorns can also be attributed to the spread of blood borne infectious parasites and viruses such as HIV and Hepatitis B.

Female Tunga fleas feed directly on blood vessels. Insidious attacks on individuals not only damage the affected vessels, but also cause severe discomfort and irritation, which is equally as important they are a disease threat. Impaired blood flow to severely infested areas causes gangrene, which necessitates amputation of fingers, toes, and sometimes the
foot or hand (Durden and Traub, 2002). Another concerning consequence of this dietary preference is that *Tunga* fleas as other species harbour harmful pathogens that they might disperse via contaminated faecal pellets or regurgitated blood meals (Bitam *et al*., 2010).

### 2.4 Risk Factors for Tungiasis

Many factors influence the prevalence of tungiasis in endemic areas. People living in poor dwellings such as houses with sandy floors are at a higher risk of tungiasis. Poverty, and behavioural attributes related to age and gender also combine with environmental risk factors to create epidemiological hotspots (Mazigo *et al*., 2010; Hotez *et al*., 2008). In many areas, misconceptions about tungiasis causation and occurrence if animal hosts such as dogs, cats, and peridomestic rodents such as rats in the home environment, have also been attributed with an increased risk of getting tungiasis (Heukelbach *et al*., 2005).

### 2.5 Diagnosis of Tungiasis

Tungiasis is diagnosed by identifying lodged fleas under the skin, usually as a dark and itching spot in the epidermis. The spot has a diameter of between 1-2 mm and is usually itchy and painful. Lesions with a central black dot and or brownish-black circular crust presenting as a white halo 3–10 mm in diameter are also diagnostic. When lodged fleas die, lesions present as circular or shrivelled black crusts with punched out residues, and necrosis of the skin (Feldmeier *et al*., 2004; Eisele *et al*., 2003). While presentation is typical in many cases, a majority often present with distorted or manipulated lesions.

Dermoscopy is a diagnostic tool for tungiasis. The presence of a pigmented brown ring with a central dark pore is diagnostic for the disease. Presence of ‘whitish chains’ prior to shaving a lesion’s papule has also been proposed (Bakos and Bakos, 2008). Even though
dermoscopy is effective for identifying single lesions, latrogenic rapturing of the flea’s abdomen makes it hard to extract. This induces an inflammatory reaction that not only takes long to heal, but also induces secondary cellulitis and septicaemia (Gibbs, 2008).

2.6 Treatment of Tungiasis

No drug is effective against tungiasis. Standard therapy involves total extraction of fleas using sterile instruments and disinfection of lesions. A treatment strategy involving sterile curettage of lesions and use of topical thiabendazole, ivermectin, or metrifonate has been found to be effective (Leung et al., 2007). However, this requires a skilled hand, hygiene, and light to illuminate infested hands and feet, many of which lack in rural areas. Victims inconsistently manipulate lesions leading to secondary infections. Topical disinfectants such as potassium permanganate a plant-based repellent called Zanzarin consisting of coconut oil also treat tungiasis. They repel free living fleas, suffocate neosomic ones, and over time, reverse tungiasis induced pathology permanently (Feldmeier et al., 2006).
CHAPTER THREE

3. MATERIALS AND METHODS

3.1 Study Location

This study was done in Otwenya Location in Maseno Division in Kenya. It lies between latitude 0° 20’ and 0° 50’ North and Longitude 34° 20’ and 35° 20’ East, has 15,759 residents, and a mean density of 387 people per square kilometre. Small-scale farming of maize and sorghum and rearing of cattle and goats are the main economic activities. The area is rural, mainly dry, and has red sandy soil that supports the development of *Tunga* fleas. Living conditions are poor with over 45% of its residents living under the national poverty line (National Coordinating Agency for Population and Development, 2010).

**Figure 3.1:** A map of Kenya showing the location of Maseno Division and Otwenya Location
3.2 Research Design

A descriptive research design was adopted. A crosssectional study was conducted.

3.3 Variables

The dependent variable was the presence of tungiasis. Independent variables were age, gender, and marital status of household heads. Family size and architectural design of study houses, behavioural attributes of respondents such as their level of knowledge on tungiasis, attitudes, and practices, and the presence of animal hosts in homes were also analysed. The flea species responsible for tungiasis in the study area was characterised.

3.4 Target Population

The target population for this study was the residents of Otwenya Location in Maseno Division. Approximately 15,759 children and adults were targeted.

3.5 Inclusion Criteria

Households with children below 15 years qualified for this study. Only family members who had lived in the area for at least a month prior to the start of the study were recruited.

3.6 Exclusion Criteria

Houses where household heads were absent during field visits were excluded from the study. Non-residents of Otwenya Location in homes were also excluded from the study.

3.7 Sample Size Calculation

A statistical formula for estimating the population proportion ($p$) in a disease prevalence study reported by Fosgate (2009) was used to calculate the sample size ($n$).
Standard normal deviation $Z$ score of 1.96 was used to calculate sample size ($n$).

An expected proportion ($p$) of 50%, and 5 degrees of accuracy ($e$) were used, at 95% CI.

$$n = \frac{(Z)^2 \times (p) \times (1-p)}{e^2}$$

$$n = \frac{1.96^2 \times (0.5) \times (1-0.5)}{0.05^2} = 384$$

The minimum sample size of 384 people was adjusted by a factor of 20% to cater for the possibility of having non-respondents. Therefore, a sample of 460 was targeted. Since the mean family size in the area was six (Place et al., 2004), 77 households were targeted.

### 3.8 Sampling Techniques

To lower bias and get a representative sample for the study a two-step random sampling technique utilising schools and schoolchildren adopted. A fishbowl draw technique was used to select 30% of schools in Otwenya and a probability random sampling technique used to select a fraction of pupils from recruited schools, each representing a household.

#### 3.8.1 Fishbowl Technique

A register of primary schools was obtained from the District Education Office in Kisumu West District and a unique number allocated to each school. Numbers were written on paper, folded, and placed in a fishbowl. The papers were mixed well and three drawn randomly. Mariwa primary, Urudi Rata, and Mbeka primary schools were selected.

#### 3.8.2 Probability Sampling Technique

A probability sampling technique was used to select 150 pupils from the selected schools. Infested and non-infested pupils had an equal chance of selection. In each school, a
register with names of pupils was generated. Each pupil was allocated a unique number and the Research Randomizer V4.0 (http://www.randomizer.org/) software used to select 50 pupils. In total, 150 pupils representing the households targeted were selected.

3.9 Pilot Study

A pilot study was carried out to access the feasibility and acceptability of this study and test the reliability and validity of data collection tools. A systematic random sampling technique was used to select 10 household heads and similar questionnaires used to collect piloting data on two sampling durations over a two-week period.

3.10 Reliability of Questionnaires

The reliability of research instruments was determined using the test-retest method. Data was extracted from pilot questionnaires and entered into a database. Descriptive statistics such as frequencies and means were explored and similarity between responses between continuous variables determined by computing their Intra-Class Correlation Coefficients (ICCs). To determine whether the responses were significantly similar or not, ICC’s were interpreted as being modest (0.30 to 49), moderate (0.50 to 0.69), and high (0.70 to 1.00). Similarity between responses for categorical variables was determined by computing Chi square statistics and analysis of the Kappa (K) statistic. The K statistic was interpreted as poor (0.0 to 0.40), moderate (0.41 to 0.60), substantial (0.61–0.80), and high (0.81–1.0).

After data analysis, ICC coefficients for many continuous variables (age and household size) were high (over 0.70). K statistic for categorical variables such as gender, marital status, and educational level of respondents was also high (0.81–1.0). As expected, the K
statistic for multi-answer questions, especially those assessing the knowledge, attitude, and practices regarding tungiasis were moderate (0.41 to 0.60) to substantial (0.61–0.80).

3.11 Validity of Questionnaires

The validity of research instruments was determined using the face validity technique. Data collection instruments were submitted to independent researchers at KEMRI-CGHR Kisumu and KEMRI-CCR in Nairobi for review. They checked the structure of research instruments and their suitability for accomplishing the study’s objectives. All corrections the reviewers proposed were done in the two research instruments before data collection.

3.12 Data Collection Techniques

Three techniques were used to collect data during the study. One-on-one interviews were used to collect household and environmental data from household heads. Observational checklists were used to collect personal data from participants and various laboratory techniques used to characterise the species of fleas responsible for tungiasis in the area.

3.12.1 One on One Interviews

After obtaining informed consent, a trained interviewer collected socioeconomic, marital, and demographic data from household heads. Data on major environmental factors such as the characteristics of a dwelling unit (the type of floor, wall, and roofing materials) and ownership of domestic animals such as dogs, cattle, pigs, and cats households were also collected during at this stage. Interviews were structured using pre-tested questionnaires.
3.12.2 Identification of Tungiasis Cases

Relevant demographic data was recorded on observational checklists and hands and feet of participants cleaned using Dettol antiseptic solution (1:100 volume per volume) to kill germs. Hands and feet were dried using paper towels and a magnifying lens used to check for tungiasis. Presence of single and or clustered nodules with central black dots was diagnostic for the disease. Presence of pale whitish to brownish papules between 1-2 mm in diameter, presence of white patches approximately 3-10 mm wide and dark lesions with shrivelled crusts, dead fleas, or punched residues were also diagnostic for tungiasis.

3.12.3 Collection of Neosomic and Free-Living *Tunga* Fleas

To identify the vector for tungiasis, neosomic and free-living *Tunga* fleas were collected on cases and in homes and characterized using morphological and molecular techniques. After physical examination, an intact lesion was identified on willing cases and the skin around it cleaned using soap and water. A cotton swab soaked in 70% alcohol was used. The lesion was anaesthised using ethyl chloride spray and the neosomic flea extracted via sterile cutterage. A sterile hypodermic needle fitted on a syringe was used to break the skin around the lesion and a sterile forceps used to extract the lodged flea. Samples were stored in 70% ethanol in 1mL micro centrifuge tubes, and stored in a cool box with ice.

Free-living fleas were trapped at night inside houses with cases. A trap made of a kerosene lantern and shallow plastic troughs was placed in bedrooms to collect fleas. A drop of Dettol antiseptic was added to water in the troughs to break surface tension and prevent fleas from escaping. In the morning, trapped fleas were aspirated using pipettes, stored in 1mL tubes with 100% ethanol, and stored on the bench at room temperature.
3.12.4 Morphological Characterisation of Collected Fleas

To differentiate *Tunga* species of fleas from other species collected in the field, samples were mounted on clean glass slides with distilled water. Slides were mounted on a light microscope and specimens visualised at x40 magnification. During the process, the flea identification scheme in Appendix 10 was used to separate *Tunga* fleas from other flea species. Checking for the presence or absence of a spermatheca and claspers, free-living *Tunga* fleas were identified as male or female. The structure and arrangement of spines on maxillary palp segments and third leg tibiae were used to distinguish the two species.

3.12.5 PCR Analysis of Fleas

PCR laboratory protocol was used to confirm the results of morphological analysis. Genomic DNA (gDNA) was extracted from whole fleas using the QIAamp DNeasy Blood and Tissue Extraction Kit (Qiagen) following manufacturer’s directions. The 5.8S ITS2 gene loci of DNA positive samples were amplified using PCR in 25-µL reaction mixtures. To check for *Tunga penetrans*, a 25-µL reaction mixture with 12-µL PCR buffer, 0.75-µL forward ITS-2A-F (GCGTCAACATGTGAACTGCA) and reverse ITS-2A-R (TTAGCGCACGACTAGACGAC) primers, 6.5 µL of water, and 5-µL DNA was used. For *Tunga trimamillata*, a 25-µL mixture containing 12-µL PCR buffer, 0.75-µL forward ITS-2A-F (GCGTCAACATGTGAACTGCA) and reverse ITS-2B-R (TAGCGCTCGACAATACGACG) primer, 6.5µL water, and 5 µL DNA was used.

A standard cycling protocol was used for the two reactions. An initial denaturation step at 95 °C was included for five minutes and 35 PCR cycling steps of 1) denaturation at 95 °C for 30 seconds, 2) annealing at 53 °C for 30 seconds, and 3) elongation at 72 °C for one
minute done. A final elongation step was done at 72 °C for seven minutes and the PCR reaction held at 4 °C indefinitely. To check the success of PCR and identify the species of *Tunga* fleas collected, 10-µL of PCR amplicons were electrophoresed on 2% agarose gels with ethidium bromide and the results visualised under ultraviolet (UV) light.

3.13 Ethical Considerations

The Scientific Steering Committee (SSC) and Ethical Review Committee (ERC) at the Kenya Medical Research Institute (KEMRI) approved this study (SSC 2511). During the study, informed consent was sought from participants before data collection. The identity of participants also remained anonymous to maintain their confidentiality. To lower the risk of stigmatisation, health education pamphlets on tungiasis were distributed in schools and the community before conducting the definitive study. Even though participants did not benefit financially, they were educated on tungiasis and all cases treated free.

3.14 Data Analysis

The Statistical Package for Social Scientists (SPSS) version 17 and the GraphPad Prism software version 5.02 for Windows, San Diego California USA (www.graphpad.com) were used for data analysis. Data was extracted from questionnaires and observation checklists, entered into SPSS, and frequencies and means of demographic data explored. A contingency analysis using Fishers test and analysis of odds ratios was done, a logistic regression model with backward elimination used to identify modifiable predictors for tungiasis in the study, and their Population Attributable Fractions (PAFs) calculated. To generate knowledge scores, a score of “1” was assigned for correct answers and “0” for wrong answers. Scores were summed to generate an overall knowledge score for each
category, knowledge scores converted to percentages, and re-categorised as either low (<50%), moderate (50-74%), and high (≥75%). For attitude scores, a score of “1” was assigned for good attitudes and “0” for bad ones. Scores were summed, means calculated, and overall attitude scores re-categorised as bad (<mean) and good (≥mean). All tests for statistical significance were calculated at the 95% level of confidence (p<0.05).
CHAPTER FOUR

4. RESULTS

4.1 Demographic Data

Three villages, Urudi Rata, Mbeka, and Mariwa in Otwenya location were selected and 78 households recruited. Approximately 39.7% of study households were in Urudi Rata village, 35.6% in Mbeka village, and 24.4% in Mariwa village. Four hundred and fifteen (415) children and adults were recruited. Females were 52.8%. The 20-39 year old age group was 23.6%. Over 60 year olds were only 3.6%. Approximately 71.8% of household heads were female, 57.7% were married, and 67.9% had primary education (Table 4.1).

Table 4.1: Demographic data: males were more than females. Approximately, nine out of 10 residents has a history of tungiasis

<table>
<thead>
<tr>
<th>Variables</th>
<th>Frequency (n)</th>
<th>Percentage (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Study village</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Urudi Rata</td>
<td>31</td>
<td>39.7</td>
</tr>
<tr>
<td>Mbeka</td>
<td>28</td>
<td>35.9</td>
</tr>
<tr>
<td>Mariwa</td>
<td>19</td>
<td>24.4</td>
</tr>
<tr>
<td>Gender profile</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>196</td>
<td>47.2</td>
</tr>
<tr>
<td>Female</td>
<td>219</td>
<td>52.8</td>
</tr>
<tr>
<td>Age profile</td>
<td></td>
<td></td>
</tr>
<tr>
<td>&lt;4</td>
<td>45</td>
<td>10.8</td>
</tr>
<tr>
<td>5-9</td>
<td>61</td>
<td>14.7</td>
</tr>
<tr>
<td>10-14</td>
<td>82</td>
<td>19.8</td>
</tr>
<tr>
<td>15-19</td>
<td>51</td>
<td>12.3</td>
</tr>
<tr>
<td>20-39</td>
<td>98</td>
<td>23.6</td>
</tr>
<tr>
<td>40-59</td>
<td>62</td>
<td>14.9</td>
</tr>
<tr>
<td>60+</td>
<td>16</td>
<td>3.9</td>
</tr>
<tr>
<td>Family size</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Crowded (6+)</td>
<td>38</td>
<td>48.7</td>
</tr>
<tr>
<td>Non-crowded (&lt;6)</td>
<td>40</td>
<td>51.3</td>
</tr>
<tr>
<td>History of tungiasis</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>364</td>
<td>87.7</td>
</tr>
<tr>
<td>No</td>
<td>51</td>
<td>12.3</td>
</tr>
<tr>
<td>Household Architecture</td>
<td></td>
<td></td>
</tr>
<tr>
<td>1. Flooring</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sandy</td>
<td>26</td>
<td>33.3</td>
</tr>
<tr>
<td>Dung Plastered</td>
<td>47</td>
<td>60.3</td>
</tr>
<tr>
<td>Cemented</td>
<td>5</td>
<td>6.4</td>
</tr>
<tr>
<td>2. Roofing</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Grass or Makuti</td>
<td>5</td>
<td>6.4</td>
</tr>
<tr>
<td>Iron Sheets</td>
<td>73</td>
<td>93.6</td>
</tr>
<tr>
<td>3. Walls</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mud</td>
<td>43</td>
<td>55.1</td>
</tr>
<tr>
<td>Cemented</td>
<td>5</td>
<td>6.4</td>
</tr>
<tr>
<td>Dung Plastered</td>
<td>30</td>
<td>38.5</td>
</tr>
</tbody>
</table>
4.1.1 Age of Participants

Participants were between 1 and 83 years old. The mean age was 22 years. Median age was 16 years. The population structure was wider at the base, with infants (<5) (10.8%) and 5-14 year olds (34.5%) forming 45.3% of the study population. 20-30 year olds were 23.6%, 40-59 year olds 14.9%, and the elderly the least forming 3.9% of the population.

4.1.2 Education of Household Heads

Approximately 68% of household heads had primary level education, 27% had secondary level education, while 1.3% had tertiary education (polytechnic, university, or college). The remaining 4% lacked formal education (Figure 4.1).

![Figure 4.1: Education level of household heads: 68% had primary level of education](image)

4.1.3 Occupation of Household Heads

Approximately 41.0% of household heads were farmers. Businesspersons were 26.9%. Carpenters were the least at 2%. Approximately 26.9% were unemployed (Figure 4.2).
Figure 4.2: Occupation of household heads: farming was the commonest economic activity among household heads

4.1.4 History of Tungiasis

Of the 415 participants sampled, 87.7% had a history of tungiasis, with approximately 82% having their first exposure by age nine. By age 14, 92% had had their first exposure to tungiasis. More males (88.8%) than females (86.8%) had a history of tungiasis. Males also had their first exposure earlier than women did. While 100% of males with a history of tungiasis had their first exposure by age 14, only 83.7% of females did. However, relationship between gender and history of tungiasis was insignificant ($X^2=0.4$, $P=0.5$).

4.1.5 Architecture of Study Houses

Approximately 93.6% of houses had iron sheet roofs. Makuti and grass thatched roofs were on 6.4% of houses. Floors plastered with cow dung were found on 60.3% of study houses. Approximately, 6.4% had cemented floors, while 33.3% had loose earth or sandy floors. As shown in Figure 4.3, 55.1% of houses had mud or earth walls. Walls plastered using cow dung and cement was found on 38.5% and 6.4% of study houses respectively.
4.2 Prevalence of Tungiasis

In 39.7% of households visited, at least one person had intact or manipulated tungiasis lesions. Of the 39.7% household with cases, 54.8% were in Urudi Rata, 32.3% in Mbeka, and 12.9% in Mariwa village. The prevalence of tungiasis in the study area was 19.5%.

4.2.1 Prevalence by Gender

Prevalence of tungiasis varied by gender of participants: approximately 25% of males and 14.6% of female had intact and or manipulated tungiasis lesions.

4.2.2 Prevalence by Age

By age of participants, prevalence of tungiasis followed and S-shaped curve, with 4.4% of infants infested. Prevalence rose between five and nine year olds (37.7%), dropped among 20 to 39 year olds (7.1%), and peaked among the elderly (43.8%) (Figure 4.4).
Figure 4.4: Prevalence of tungiasis: children (5-14) and the elderly (over 60 years) were high-risk groups for tungiasis

4.3 Clinical Manifestations

After examination, 677 tungiasis lesions were counted on 81 cases. 51.3% of lesions were altered either partially or wholly leaving a crater-like depression. Virulent lesions formed 45% of the total number counted, while 3.7% were non-virulent lesions (Figure 4.5).

Figure 4.5: Clinical manifestations on cases: manipulated lesions was a common occurrence on cases
4.3.1 Parasite Load

Tungiasis cases had between one and 53 lesions. Approximately 46.9% were lightly infested (1-4 lesions), while 38.3% and 14.8% were moderate (5-12 lesions) and heavily infested (13+ lesions) respectively. The mean parasite load on cases was 8.3. By gender, males (9.6 ± 1.62) were highly infested than females (6.5 ± 0.98) (U=669.5, P=0.27). By age, 10 to 14 years olds were severely infested (13.73 ± 2.91). 20 to 39 years olds were moderately infested (8.0 ± 1.56) and infants lightly infested (2.0 ± 0.00) (Figure 4.6).

![Figure 4.6](image)

**Figure 4.6:** The 10-14 year age group was at risk of sever tungiasis infestation

4.3.2 Topographical Location of Lesions

The feet (99.1%) were invariably infested with jiggers. Only 0.8% of lesions were on the hands. Approximately 34.1% of lesions were on the nail bed of toes. Area between toes heels, heel, and the lateral rim of the feet had 24.4%, 22.6%, and 11.7% of the diagnosed lesions respectively. Fingers (0.7%) and palms of cases (0.1%) were the least infested.

4.3.3 Tungiasis Pathology

Deformed toenails especially on the big toes were identified on 71.6% of cases. Oedema (32.1%) and loss of toenails (30.9%) were also common, while major signs of bacterial
superinfection were reported on 19.8% of tungiasis cases. Many infested cases (17.3%) had suppurrated lesions. Micro abscesses and pustules on lesions were reported on 2.5% of cases. Finally, signs of prolonged infestation were found on 55.6% of cases. Many cases (34.6%) had deep fissures. The remaining 21.0% had ulcerated lesions (Figure 4.7).

![Figure 4.7: The heels of a twelve-year-old pupil from Urudi Rata village in Otwenya Location with tungiasis-induced ulcers and fissures (Aballa A. O., 2013)](image)

### 4.4 Risk Factors for Tungiasis

After univariate analysis, several risk factors for tungiasis were identified in the study area. The odds of tungiasis infestation was higher in Urudi Rata (OR=3.2, P=0.02). A significant difference in prevalence biased towards males (OR=1.9, P<0.01), under 15’s (OR=2.3, P<0.01), and the elderly (OR=3.4, P=0.02) was also evident. Living in houses with sandy floors (OR=9.0, P<0.01), mud walls (OR=6.7, P<0.01), and animals that fleas parasitise (OR=10.3, P=0.01); overcrowding (OR=1.3, P<0.1); and living in male-headed
houses (OR=4.0, P=0.01) or houses headed by widows or widowers (OR=9.0, P<0.01) also increased the odds of infestation. Marriage (OR=0.3, P<0.01), and dung plastering of floors in homes (OR=0.1, P<0.01) were protective factors for tungiasis (Table 4.2).

Table 4.2: Risk factors for tungiasis: gender, age, and flooring were major risk factors

<table>
<thead>
<tr>
<th>Presence of Tungiasis</th>
<th>n</th>
<th>(%)</th>
<th>OR (95% CI)</th>
<th>P Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Village</td>
<td>31</td>
<td></td>
<td></td>
<td>0.03</td>
</tr>
<tr>
<td>Urudi Rata</td>
<td>18</td>
<td>58.1</td>
<td>3.2 (2.3 to 8.4)</td>
<td>0.02</td>
</tr>
<tr>
<td>Mbeka</td>
<td>10</td>
<td>35.7</td>
<td>0.7 (0.3 to 1.9)</td>
<td>0.63</td>
</tr>
<tr>
<td>Mariwa</td>
<td>4</td>
<td>21.1</td>
<td>0.3 (0.1 to 1.1)</td>
<td>0.06</td>
</tr>
<tr>
<td>Gender of Participants</td>
<td>81</td>
<td></td>
<td></td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Male</td>
<td>49</td>
<td>25.0</td>
<td>1.9 (1.2 to 3.2)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Female</td>
<td>32</td>
<td>14.6</td>
<td>0.5 (0.3 to 0.8)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Age of Participants</td>
<td>81</td>
<td></td>
<td></td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>&lt;15</td>
<td>50</td>
<td>26.6</td>
<td>2.3 (1.4 to 3.8)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>15-19</td>
<td>6</td>
<td>11.8</td>
<td>0.5 (0.2 to 1.3)</td>
<td>0.19</td>
</tr>
<tr>
<td>20-39</td>
<td>7</td>
<td>7.1</td>
<td>0.3 (0.1 to 0.6)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>40-59</td>
<td>11</td>
<td>17.7</td>
<td>0.8 (0.4 to 1.7)</td>
<td>0.73</td>
</tr>
<tr>
<td>60+</td>
<td>7</td>
<td>43.8</td>
<td>3.4 (1.2 to 9.5)</td>
<td>0.02</td>
</tr>
<tr>
<td>Gender of Household Heads</td>
<td>31</td>
<td></td>
<td></td>
<td>0.01</td>
</tr>
<tr>
<td>Male</td>
<td>14</td>
<td>63.6</td>
<td>4.0 (1.1 to 11.4)</td>
<td>0.01</td>
</tr>
<tr>
<td>Female</td>
<td>17</td>
<td>30.4</td>
<td>0.2 (0.1 to 0.7)</td>
<td>0.01</td>
</tr>
<tr>
<td>Marital Status of Heads</td>
<td>31</td>
<td></td>
<td></td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Single</td>
<td>3</td>
<td>25.0</td>
<td>0.5 (0.1 to 1.8)</td>
<td>0.34</td>
</tr>
<tr>
<td>Married</td>
<td>12</td>
<td>26.7</td>
<td>0.3 (0.1 to 0.7)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Widowed</td>
<td>16</td>
<td>76.2</td>
<td>9.0 (2.8 to 28.7)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Occupation of Heads</td>
<td>31</td>
<td></td>
<td></td>
<td>0.08</td>
</tr>
<tr>
<td>Education of Heads</td>
<td>31</td>
<td></td>
<td></td>
<td>0.36</td>
</tr>
<tr>
<td>Family Size</td>
<td>31</td>
<td></td>
<td></td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Less than 6 (Normal)</td>
<td>9</td>
<td>33.7</td>
<td>0.3 (0.1 to 0.6)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>6 or more (Overcrowded)</td>
<td>22</td>
<td>41.5</td>
<td>1.3 (1.5 to 10.4)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Type of Walls</td>
<td>31</td>
<td></td>
<td></td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Mud</td>
<td>25</td>
<td>58.1</td>
<td>6.7 (2.3 to 19.5)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Cemented</td>
<td>2</td>
<td>40.0</td>
<td>1.0 (0.2 to 6.4)</td>
<td>1.00</td>
</tr>
<tr>
<td>Dung Plastered</td>
<td>4</td>
<td>13.3</td>
<td>0.1 (0.0 to 0.4)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Type of Floor</td>
<td>31</td>
<td></td>
<td></td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Earth or Sandy</td>
<td>19</td>
<td>73.1</td>
<td>9.0 (3.1 to 26.7)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Cemented</td>
<td>2</td>
<td>40.0</td>
<td>1.0 (0.2 to 6.4)</td>
<td>1.00</td>
</tr>
<tr>
<td>Dung Plastered</td>
<td>10</td>
<td>21.3</td>
<td>0.1 (0.1 to 0.4)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Type of Roof</td>
<td>31</td>
<td></td>
<td></td>
<td>1.00</td>
</tr>
<tr>
<td>Animal hosts on compound</td>
<td>30</td>
<td>46.2</td>
<td>10.3 (1.3 to 83.8)</td>
<td>0.01</td>
</tr>
</tbody>
</table>
4.4.1 Population Attributable Fractions (PAFs)

Overcrowding (OR=3.5, P=0.04), living in houses with sandy floors (OR=11.1, P<0.01), and animals in homes (OR=22.8, P=0.01) were the predictors for tungiasis (Table 4.3).

Table 4.3: Overcrowding, living in homes with sandy flooring, and presence of animal hosts in homes were major predictors for tungiasis

<table>
<thead>
<tr>
<th>Risk Factors</th>
<th>OR</th>
<th>Std. Err.</th>
<th>z</th>
<th>P(z)</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Overcrowding</td>
<td>3.49</td>
<td>2.09</td>
<td>2.09</td>
<td>0.04</td>
<td>1.18 to 11.27</td>
</tr>
<tr>
<td>Animal Hosts</td>
<td>22.76</td>
<td>28.06</td>
<td>2.54</td>
<td>0.01</td>
<td>2.03 to 254.95</td>
</tr>
<tr>
<td>Sandy Floors</td>
<td>11.15</td>
<td>7.38</td>
<td>3.65</td>
<td>0.00</td>
<td>3.05 to 40.77</td>
</tr>
<tr>
<td>_cons</td>
<td>0.009</td>
<td>0.01</td>
<td>-3.49</td>
<td>0.00</td>
<td>0.00 to 0.13</td>
</tr>
</tbody>
</table>

Plastering floors, non-crowding in homes and good animal husbandry practices in homes can lower prevalence of tungiasis by 66.5%, 29.5%, and 43.9% respectively (Table 4.4).

Table 4.4: PAFs: modifying sandy floors was the best strategy for controlling tungiasis

<table>
<thead>
<tr>
<th>Predictor Factor</th>
<th>OR</th>
<th>AR</th>
<th>% exposed</th>
<th>PAF</th>
</tr>
</thead>
<tbody>
<tr>
<td>Overcrowding</td>
<td>3.5</td>
<td>0.71</td>
<td>41.5</td>
<td>29.5</td>
</tr>
<tr>
<td>Sandy Floors</td>
<td>11.2</td>
<td>0.91</td>
<td>73.1</td>
<td>66.5</td>
</tr>
<tr>
<td>Presence of Animal Hosts</td>
<td>22.8</td>
<td>0.95</td>
<td>46.2</td>
<td>43.9</td>
</tr>
</tbody>
</table>

Where:
OR = Odds ratio
AR = Attributable Risk, calculated as (OR−1)/OR,
PAF = Population Attributable Fraction calculated as % exposed × AR

4.5 Tungiasis Knowledge

Approximately 75.7% of respondents had a moderate knowledge of tungiasis. 19.2% had a poor understanding of tungiasis, while 5.1% were highly knowledgeable. Knowledge of tungiasis did not vary by the gender (p=0.16). However, respondents with no formal
education had a poor knowledge of tungiasis than those with formal education OR=35.56 (1.7 to 732.6). Knowledge did not influence occurrence of tungiasis in homes (p=0.49).

4.5.1 Etiological Agent
Approximately 69.2%) had high knowledge of tungiasis causation. When asked to state the causative agent(s) for tungiasis 80.9% mentioned fleas. Dust, witchcraft, bad hygiene practices, and germs featured in 41.2%, 20.6%, 14.6%, and 14% of responses.

4.5.2 Risk Factors
During interviews, 70.5% of respondents identified four to five risk factors for tungiasis correctly. Approximately 18.0% knew less than four factors, while only 11.5% identified more than five risk factors for tungiasis correctly. Of all risk factors stated, poor hygiene featured in 84.9% of responses. Approximately, 41.1%, 20.5%, and 13.7% talked of the architecture of a house, walking barefoot on the ground, and presence of animal hosts.

4.5.3 Protective Factors
Approximately 90.1% and 87.3% of respondents knew that good personal hygiene and cleaning the environment could lower the prevalence of tungiasis in the home. Dusting of floors and plastering of sandy floors were talked of by 55% and 43.7% of respondents, while 18.3% and 16.7% thought of wearing closed shoes, and inspecting the feet daily.

4.5.4 Signs and Symptoms
Only 6.4% of respondents identified four of six commonest signs of tungiasis correctly. Approximately 14.1% of respondents knew three signs and or symptoms of tungiasis. A majority (79.5%) did not know or knew less than three signs or symptoms of tungiasis.
An itching sensation featured in 91.9% of responses. Approximately, 70.3%, 21.6%, and 16.2% talked of pain, a watery discharge, and presence of a black spot (Table 4.5).

### Table 4.5: Tungiasis Knowledge

Knowledge of tungiasis causation, risk factors, and protective factors was high. Signs and symptoms of tungiasis were poorly understood.

<table>
<thead>
<tr>
<th>Knowledge of tungiasis</th>
<th>Frequency (n)</th>
<th>Percentage (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>What causes tungiasis?</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fleas</td>
<td>55</td>
<td>80.9</td>
</tr>
<tr>
<td>Dust</td>
<td>28</td>
<td>41.2</td>
</tr>
<tr>
<td>Witchcraft</td>
<td>14</td>
<td>20.6</td>
</tr>
<tr>
<td>Poor hygiene</td>
<td>10</td>
<td>14.7</td>
</tr>
<tr>
<td>Sharing clothes</td>
<td>5</td>
<td>7.4</td>
</tr>
<tr>
<td>Poverty</td>
<td>1</td>
<td>1.5</td>
</tr>
<tr>
<td>What are the symptoms of tungiasis?</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Itching</td>
<td>68</td>
<td>91.9</td>
</tr>
<tr>
<td>Pain</td>
<td>52</td>
<td>70.3</td>
</tr>
<tr>
<td>Watery discharge</td>
<td>16</td>
<td>21.6</td>
</tr>
<tr>
<td>Swelling</td>
<td>12</td>
<td>16.2</td>
</tr>
<tr>
<td>Black spot</td>
<td>7</td>
<td>9.5</td>
</tr>
<tr>
<td>What are the risk factors for tungiasis?</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Poor hygiene</td>
<td>62</td>
<td>84.9</td>
</tr>
<tr>
<td>Dust</td>
<td>47</td>
<td>64.4</td>
</tr>
<tr>
<td>House architecture</td>
<td>30</td>
<td>41.1</td>
</tr>
<tr>
<td>Soil type</td>
<td>21</td>
<td>28.8</td>
</tr>
<tr>
<td>Walking barefoot</td>
<td>15</td>
<td>20.5</td>
</tr>
<tr>
<td>Animal hosts</td>
<td>10</td>
<td>13.7</td>
</tr>
<tr>
<td>Migration</td>
<td>2</td>
<td>2.7</td>
</tr>
<tr>
<td>What are the preventive factors for tungiasis?</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Good hygiene</td>
<td>64</td>
<td>90.1</td>
</tr>
<tr>
<td>Plastering homes</td>
<td>31</td>
<td>43.7</td>
</tr>
<tr>
<td>Clean environment</td>
<td>62</td>
<td>87.3</td>
</tr>
<tr>
<td>Wearing shoes</td>
<td>13</td>
<td>18.3</td>
</tr>
<tr>
<td>Daily inspection</td>
<td>12</td>
<td>16.9</td>
</tr>
<tr>
<td>Avoid endemic areas</td>
<td>1</td>
<td>1.4</td>
</tr>
</tbody>
</table>

### 4.5.5 Tungiasis Knowledge Transfer

Approximately 75.6% of respondents gained their knowledge of tungiasis from other family members. School and personal studies informed 20.6% of respondents. Nurses and doctors contributed marginally (2.9%) to the spread of tungiasis knowledge (Figure 4.8).
Figure 4.8: Tungiasis knowledge transfer: family members were the main source of information on tungiasis. Health professionals contributed marginally.

4.6 Tungiasis Perceptions

Approximately 82.1% of respondents had a negative attitude towards tungiasis, with both negligence and stigma evident. When asked how serious tungiasis is in the area 64.1% thought the disease is not serious. When infested, only 5.4% and 6.8% have or would consider visiting a doctor or seeking professional treatment respectively. 89.7% were ashamed of tungiasis, while 92.3% talked of having strained relationships with family, friends, and neighbors. To avoid ridicule, approximately 71.8% would try to hide the presence of tungiasis in their homes by avoiding social events such as church (Table 4.6).

Even though tungiasis perceptions did not vary significantly by education (p=0.43), age (p=0.12), and gender (p=0.09) of participants, respondents with a poor knowledge had negative attitudes (OR=9.1 (0.5 to 161.3), p=0.04). The Odds of tungiasis infestation was high in homes headed by people with negative perceptions (OR=4.9 (1.0-24.0), p=0.03).
Table 4.6: Tungiasis perceptions: tungiasis negligence and stigma was evident

<table>
<thead>
<tr>
<th>Tungiasis perception</th>
<th>(n)</th>
<th>(%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>How serious is tungiasis in this area?</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Serious</td>
<td>28</td>
<td>35.9</td>
</tr>
<tr>
<td>Not serious</td>
<td>50</td>
<td>64.1</td>
</tr>
<tr>
<td>If infested:</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Would you visit a physician?</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>4</td>
<td>5.4</td>
</tr>
<tr>
<td>No</td>
<td>74</td>
<td>94.6</td>
</tr>
<tr>
<td>Would you seek professional treatment?</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>5</td>
<td>6.8</td>
</tr>
<tr>
<td>No</td>
<td>73</td>
<td>93.2</td>
</tr>
<tr>
<td>Are you ashamed of tungiasis?</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>70</td>
<td>89.7</td>
</tr>
<tr>
<td>No</td>
<td>8</td>
<td>10.3</td>
</tr>
<tr>
<td>Has tungiasis affected your relationships of that of your relatives?</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>72</td>
<td>92.3</td>
</tr>
<tr>
<td>No</td>
<td>6</td>
<td>7.7</td>
</tr>
<tr>
<td>Would you hide the presence of tungiasis in your home?</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>56</td>
<td>71.8</td>
</tr>
<tr>
<td>No</td>
<td>22</td>
<td>28.2</td>
</tr>
</tbody>
</table>

4.6.1 Treatment Strategies

The treatment strategies for tungiasis were diverse, with 91.1% of participants extracting lesions to relieve tungiasis. 29.5% of respondents used an Insecticidal concentrates called DIP (29.5%), 15.5% used grease, while 9.0% and 6.4% used petroleum jelly and Vics vaporub respectively. Disinfectants such as Dettol and medicinal plants such as Mexican marigold were used by 5.1% and 1.3% of respondents respectively to treat tungiasis.

4.7 Vector for Tungiasis

Over the duration of the study, 36 neosomic and 49 free-living fleas were collected and characterised using taxonomical keys. After laboratory analysis, all neosomic fleas were identified to be *Tunga penetrans*. Approximately 67% of collected free-living fleas were *Ctenocephalid canis*. *Tunga* species formed 21% of the total, while *Ctenocephalid felis*,
Echidnophaga galicea (chicken flea), and Xenopsylla cheopis (Oriental rat flea) formed 6.0%, 4.0%, and 2.0% of the total fleas collected. Characterised Tunga fleas were Tunga penetrans, with a sex ratio distortion biased towards female fleas evident (Figure 4.9).

Figure 4.9: Flea diversity in homes: Ctenocephalid canis was the commonest flea species in homes followed by Tunga penetrans
CHAPTER FIVE

5. DISCUSSION, CONCLUSIONS, AND RECOMMENDATIONS

5.1 Discussion

Our data show that in Otwenya Location, the prevalence of tungiasis was approximately 19.5%. While this value is lower than the 34% reported in Northeast Brazil (Wilcke et al., 2002) and 57% reported in Central Kenya (Njau et al., 2012), the disease is a major threat to health in the area, considering that this study was done in the rainy season when tungiasis prevalence is supposed to be at its lowest. Moreover, unlike the study in Central Kenya that sampled only the 5-14 year old sentinel group to determine the prevalence of tungiasis, this study covered a wider geographical area and sampled residents of all ages. Overall, one in every nine residents of Otwenya Location had a history of tungiasis, with approximately 82% of cases having their first exposure by age nine. This is testament of the endemicity of the disease in this rural community. Children have their first exposure at a young age and grapple with the burden of tungiasis through adulthood and old age.

Males had a higher prevalence of tungiasis. Even though (Bourée et al., 2012) found a similar result in rural Cameroon, Muehlen et al., (2003) found no significant difference in Brazil. This inconsistency may be a result of varied environmental conditions and varied behavioural differences in different populations, and not differences in susceptibility of the two genders. For instance, as in many developing countries, male children in Kenya spend most of their free time outdoors playing on dusty grounds. Females on the other hand spend most of their free time doing household chores. This lowers exposure to adult fleas and therefore, their risk of contracting tungiasis. Females by nature are also health conscious than males. They are cleaner than males, good at detecting diseases early, and
unlike males, often ask for help when they have a problem. This not only lowers their overall risk of contracting tungiasis, but the risk of severe tungiasis infestation as well. In household headed by males, the prevalence of tungiasis was also higher than in those headed by women, with odds of infestation peaking in households headed by widowers.

As reported by (Muehlen et al., 2006) in Brazil, the distribution of tungiasis prevalence followed an S-shape curve that peaked among children and the elderly. During infancy, children are immobile. They are under the watchful eyes of the parents and rarely get in contact with sand where Tunga penetrans replicates. However, as they grow older, enrol for Early Childhood Education (ECD) and primary school, they start exploring the world. Children spend most of their free time outdoors playing on dusty ground, which increases their exposure to adult Tunga penetrans. ECD and primary schools in Otwenya Location are also overcrowded and have dilapidated dusty floors and mud walls that support the development of adult fleas. When a child presents with tungiasis, others who do not have shoes or walk with slippers or torn shoes that do not cover their feet sufficiently contract tungiasis easily, spreading the disease to their homes and their villages. This explains the low prevalence of tungiasis among infants and its steep rise among school going children.

The prevalence of tungiasis dropped steeply among the 15 to 19 year old age group. This, as proposed by (Kimani et al., 2012), may be a result of the physical changes on children and the good behavioural attributes that people adopt as they grow older. For instance, as children approach puberty, many of them become conscious about their health. They stop playing on dusty environments where Tunga penetrans develops, become cleaner, and learn how to detect, prevent, and treat tungiasis when infested before infestation becomes
debilitating. This not only lowers the risk of tungiasis, but of severe infestation among the group as well. However, as inequities such as poor eyesight and hygiene set in at old age, exposure to *Tunga penetrans* increases, which increases the risk of tungiasis infestation.

The observation that the level of education of household heads did not influence the Odds of tungiasis infestation in homes contradicted the results of (Solomon *et al*., 2008) and (Dartigues *et al*., 2013). In their studies in Ecuador and in Brazil, the risk of contracting infectious diseases was lower in homes headed by literate household heads than in homes headed by illiterate household heads. This was not the case in Otwenya Location. Both illiterate and educated household heads knew about tungiasis. Both illiterate and educated household heads considered tungiasis a normal occurrence and ignored professional help when infested with the disease. Therefore, occurrence of tungiasis in homes in Otwenya Location might be a result of negligence and poor attitudes towards tungiasis, and not the education level of household heads. The varied exposure levels in homes and differences of behaviour of people in different environments may have also contributed to this result. Developing sensitisation and or outreach programs that dispel negligence and tungiasis stigma can therefore improve the health seeking behaviour of residents, enhance adoption of modern tungiasis treatments and controls, and lower the burden of this disease further.

Morbidity associated with tungiasis was considerable. Itching, pain, and erythema were common. Clinical manifestations were diverse, with 99.1% of cases having lesions on the feet. The mean parasite load was eight, with males having a higher mean parasite load (9) than females (6). This result relates with that of (Muehlen *et al*., 2003). The behaviour of males such as playing on dusty ground, low health oriented behaviour, and negligence of
diseases not only increases their overall risk of tungiasis, but of severe infestation as well. Deformation and loss of toenails was common. These pathological alterations might be attributed to *Tunga penetrans*’s predilection for the nail bed of toes. The site is soft and poorly keratinises. Once fleas aggregate, they deform nails and ultimately uproot them from their bed as they undergo neosomy. Sequels of prolonged tungiasis infestation such as fissures and ulcers were also common, with severely infested cases having trouble walking and working. If left unaddressed therefore, tungiasis might increase the dropout rate of severely infested pupils in Otwenya Location. It might also lower the economic productivity of infested adults, hinder personal growth and development, and maintain a cycle of poverty and suffering that is hard to break. This is worrying, considering that approximately 45% of residents of the area are currently living under the poverty line.

Superinfection of tungiasis lesions was a common clinical observation in the study area. Even though bacteriological laboratory cultures were not done to support this empirical observation, signs of secondary infections such as suppurated lesions and presence of micro abscesses on lesions were identified on 19.8% of cases. Considering that 91.1% extracted lesions to relieve the effects of tungiasis, people who use non-sterilised sharps such as sewing pins, thorns, and wood splinters often inoculate harmful bacteria under the skin. Resultant ulcers and wounds left on the skin after manipulating lesions might also be a source of secondary infections. Approximately 94.9% did not use disinfectants such as Dettol to clean lesions once manipulated. As they walk barefoot in home and or on dirty surfaces outdoors, bacteria in the environment might colonise ulcers and wounds, causing severe morbidity and suffering. *Streptococcus* species and anaerobic bacteria such as *Clostridium tetani* have been isolated from such lesions (Kehr et al., 2007).
The occurrence of tungiasis in homes was associated with a few modifiable factors that can be targeted in controls. As reported in Cameroon (Bourée et al., 2012) and Nigeria (Muehlen et al., 2006), the conditions related to housing were important risk factors for tungiasis. Type of flooring, for example, influenced the odds of tungiasis infestation, with people living in houses with sandy flooring having nine times the odds of infestation than those living in houses with plastered floors. This is not surprising considering that the life cycle of *Tunga penetrans* takes place in sandy soil. As reported by (Pampiglione et al., 2009), the important role that animal reservoirs play in transmission of tungiasis in homes was evident in the study area. In houses where domesticated animals such as dogs, cats, cattle, and goats roamed freely or slept indoors, those infested with *Tunga penetrans* fleas spread eggs on the environment as gravid fleas develop, maintaining a cycle of tungiasis infestation on human beings. This explains the high prevalence of tungiasis in households where animal hosts were present than in households without alternative hosts for fleas. Educating household heads on these risk factors and developing sustainable interventions for eliminating them in homes can help to manage and ultimately eliminate tungiasis.

The PAFs calculated for tungiasis predictors indicated that sustainable controls strategies could be developed in Otwenya Location. Modifying sandy floors in homes, for instance, can lower the prevalence of tungiasis by 66.5%. Residents who cannot afford cement or plaster can use locally available material such as cow dung to eliminate the favourable environment that *Tunga penetrans* needs to replicate. Cow dung is also a good repellent and cheap to repair when used in houses (Ng’ang’a et al., 2008; Torr et al., 2011). Even though it is impossible to eliminate domestic animals from homes, educating household heads in the study area on the benefits of animal containment and good animal husbandry
practices can lower the prevalence of tungiasis by 43.9%. Daily inspection of lower limbs and body parts that come in contact with the ground, extraction of fleas, and disinfection of resultant ulcers, for instance, has been found to be effective (Pampiglione et al., 2009).

As in Italy (Veraldi and Valsecchi 2007), treatment strategies for tungiasis were diverse, with manipulation of lesions being the commonest. Even though unorthodox, early extraction is effective and touted as one of the most sustainable remedies for tungiasis in resource poor settings. Unfortunately, because of lack of light to illuminate body parts and lack of sterilised instruments to treat lesions safely, the risk of physical trauma and creation of ulcers that secondary bacteria can colonise and transmission of blood borne infections such as HIV between cases is too great to be ignored. Apart from clawing lesions using thorns and splinters, petroleum jelly and petroleum hydrocarbons such as grease and paraffin were common remedies for tungiasis. Even though 15% and 9% of respondents considered them effective, these by products are residual, requiring weeks of frequent use to be effective. They are also costly, ineffective against off host flea stages such as eggs and pupa, and therefore, are unsuitable for routine treatment of tungiasis. To lower the burden of tungiasis in the area, the potential of using safe modern insecticide and indigenous plants such as Mexican marigold to manage it should also be determined.

The distribution of fleas in the genus Tungidae suggests that there is a link between flea taxa of Africa and South America. However, since the discovery of Tunga trimamillata is South America in 2003, this is the first study that has attempted to determine its presence and possible role in tungiasis in Kenya. Diverse flea species were trapped, with dog fleas (Ctenocephalid canis) forming 67% of the flea population trapped. The presence of
*Ctenocephalid felis, Tunga penetrans, Echidnophaga galicea, and Xenopsylla cheopis* in the same environment hinted to a low inter-species competition between fleas, even in the presence of a few hosts. *Tunga trimamillata* was not isolated in the study area. However, because of the small number of fleas characterised and no scientific studies to compare this result with, further studies should be conducted in Kenya to corroborate this result.
5.2 Conclusions

1. The 19.5% prevalence reported was lower than the 57% reported in Central Kenya.
2. Children (5-14 years) and the elderly (over 60 years) were high-risk groups.
3. Sandy flooring, overcrowding in homes, and presence of animal hosts in homes were predictors for tungiasis.
4. Retrogressive cultural beliefs regarding tungiasis were common.
5. Extraction of lesions, insecticidal concentrates, and petroleum hydrocarbons such as grease were the commonest remedies for tungiasis.
6. *Tunga penetrans* was the only flea species responsible for tungiasis: *Tunga trimamillata* was not isolated in the study area.

5.3 Recommendations

1. The government and Non-Governmental Organisations (NGO’s) combating tungiasis should conduct health education campaigns and outreach campaigns that dispel stigma and negligence of tungiasis.
2. The ministry of health and the country government of Kisumu should improve the condition of local health institutions and roll out outreach programs that encourage tungiasis victims to seek professional help when infested.
3. Studies should be done to determine the potential of using indigenous plants such as Mexican Marigold to treat tungiasis.
4. Further studies should be done to determine the presence of *Tunga trimamillata* and its possible role in tungiasis in other endemic areas in Kenya.
REFERENCES


APPENDICES

Appendix 1: Questionnaire

MORBIDITY ASSESSMENT AND CHARACTERISATION OF FLEA SPECIES RESPONSIBLE FOR TUNGIASIS IN KISUMU COUNTY, KENYA

INTERVIEW QUESTIONNAIRE

Personal Identification
Study ID: .................. Date of Birth: .................. Sex: 1. (Male) 2. (Female)
Household number: .............................................. Interview Date: ..................
Village: ........................................ Interviewers’ name: ........................................

Section 1: Household Information
Q1. How many are you in your family? ................................
Q2. What type of accommodation do you have?
   1. Rented  2. Own house  3. Other ......................

Section 2: Personal and Socio-economic Information
Q3. Which year were you born? .................................. 
Q4. What is your marital status?
   0. Single  1. Married  2. Widowed  3. Divorced
Q5. Which religion do you belong?
Q6. What is the highest level of school you have ever attended?
Q7. What is your current occupation? (If more than one, circle all that apply)
   0. Unemployed  1. Student  2. Farmer  3. Daily wager

Section 3: Knowledge, Attitude, and Practices
Q8. Do you know what tungiasis disease is? 1. (Yes) 2. (No)
   If “No” proceed to Q27.
Q9. What do you think causes tungiasis in humans?
   0. Don’t Know  1. Fleas/ Organisms  2. Witchcraft
Q10. How did you learn about tungiasis? (Circle all that apply)
   1. Family member (Specify)  2. School/ Personal studies
   3. Health professional  4. Neighbour  5. Advertisement (Specify) .... 6. Other (Specify)...
   7. This study
Q11. How serious do you think tungiasis is in this area?
   0. Do not Know  1. Very serious  2. Somewhat serious  3. Not very serious
Q12. Have you ever suffered from tungiasis? 1. (Yes) 2. (No)
   If no, proceed to Q19.
Q13. Can you remember when tungiasis first occurred?
0. Don’t Know 1. Know
If answer is (Know) at the age of…………………… / in the year……………………

Q14. When did tungiasis last occur?
0. Don’t Know 1. Last 4 months 2. Less than 12 months 3. More than a year

Q15. Was tungiasis diagnosed by a physician?
1. (Yes) 2. (No)

Q16. Did you take any medication or undergo professional treatment?
1. (Yes) 2. (No)

Q17. Are/ were you limited in your daily abilities because of tungiasis?
If (1-4), how were you limited…………………………………………………………

Q18. How often do you check yourself for presence of tungiasis?

Q19. What do you think are the signs and symptoms of tungiasis?
0. Don’t know 1. Pain 2. Itching 3. Swelling 4. Other …………………

Q20. What do you think predisposes an individual to tungiasis? (List all stated)
..........................................................................................
..........................................................................................
..........................................................................................

Q21. What do you do to protect yourself from getting tungiasis?
..........................................................................................
..........................................................................................

Q22. If infested with tungiasis, which means can be used to treat it?
..........................................................................................
..........................................................................................

Q23. Who would you talk to if you have a health problem?
0. No one 1. Spouse 2. Parent 3. Other family member(Specify)…
4. Neighbour 5. Other (Specify)……….. 6. Don’t know

Q24. Can tungiasis affect your relationship with others (friends, family, etc.)?
0. Do not know 1. Yes 2. No

Q25. In case you had tungiasis, would you be ashamed of having the disease?
0. Do not know 1. Yes 2. No

Q26. Would you want to hide that you have tungiasis?
0. Do not know 1. Yes 2. No

Q27. Household/ Housing architecture:
Walls? 1. Earthen/ Mud 2. Cemented 3. Other………………

Presence of Alternative Hosts

Q28. Do you rear any animals in your home? 1. (Yes) 2. (No)
If “Yes,” which/ how many animals do you have at home? (Circle all that apply)

Q29. Where do you keep the animals?
1. Animal pen 2. Indoors 3. Outside 4. Other (Specify)………..

Q30. How often do you clean your animals?
0. Do not clean 1. Once a month 2. Once a year 3. Other (Specify)………..

Q31. What do you use to clean them?…………………………………………………………
Appendix 2: Questionnaire (Luo)

TIMO NONRO NI TUO MAR MINEME E GWENG MA NI KISUMU COUNTY, KENYA

DWARO PARO

Nonro Mar N’gato
Number mar jaduoko……… Higa mar nyuol………Kidieny 1. (Dichuo) 2. (Dhako)
Kwan ot…………………………………………………Tarik mar penj…………………………
Gweng………………………………………………Nying ja penjo…………………………

Nonro Mar Jo Ot

Q1. Un ji adi e odu?……………………………………
Q2. I dak kama nade?
   1. Adak e oda  2. A ja pango  3. Ma moko(Dimb wach)……

Wach E Wi Ngato Kod Chal Yuto Mare
Q3. Ne o nywoli e higa mane? (Higa)…………………………...
Q4. Kend mari chal nade? (Luor achiel kende)
   0.Pok o kenda/Akendo 1.Okenda/Akendo 2.Chi/Chuar liel 3.Weruok e kend
Q5. In ja din mane?
Q6. I somo nyaka e okang mane?
   0. Ne ok a dhi e school  1.Klas aboro 2.Klas a par gi ariyo 3.Kar tegrwok
      4.Mbala riany
Q7. I timo tich mane? (Ka ngeny ne achiel to lour duto mani kare)
   0. Ok ati  1.A nyathi skul  2.A japur  3. A goyo otong/tich lwedo

Ngeyo, Kaka Ikawe Kod Tim
Q8. Bende I ngeyo gima mineme en?  1. (Angeyo)  2. (Akia)
   Ka en “Akia” dhi e penjo mar 13
Q9. Ango mi paro ni kel mineme kuom dhano?
Q10. Ne ingeyo nade e wi mineme? (Luor mago ma kare)
         tudruok (Dimb wach)……………… 6. Mamoko (Dimb wach) 7. Nonro ni
Q11. Iparo ni mineme ngeny manade e gweng ni?
   0. Akia  1. Ngeny ahinya  2. Ngeny moromo  3. Ok ngeny
Q12. Be mineme ose maki e ngima ni?  1. (Kamano)  2. (Ooyo)
    Ka ooyo dhi e number19.
Q13. Be inyalo paro chieng mane mineme omaki mokwongo?  0. Akia  1. Angeyo
    Ka dwoko en (Angeyo) ne in gi higni adi………………/ ne en e higa mane……
Q14. Mineme ne omaki mogik chieng mane?
    1. Dwche angwen mokalo  2. Matin ne deche angwen  3. Osekalo higa achiel
Q15. Ne oyude gi ja thieth koso ne ingeyo kendi? 1. (Kamano) 2. (Ooyo)
Q16. Be ne imuonyo yat mora moro kata ne othiethi gi ja thieth? 1. (Yes) 2. (No)
Ne idhi thieth kanye?
Ango momiyo ne ok idhi e thieth?(Kik I penj ka 22 dwoko en “Kamano”)
Q17. Bende tuo ni moni timo yore gi ma pile?
Ka duoko en (1-4), omoni timo yore gi nade?........................................
Q18. Ikao kinde maromo nade mond ingi ka in gi mineme?
Q19. Ranyisi mineme gin mage?
Q20. Ango ma iparo ni miyo ji yudo mineme?
........................................................................................................
Q21. Ere kak inyalo gengo yudo mineme?
........................................................................................................
Q22. Ka mineme omaki, ere kaka ngato nyalu thiethore?
........................................................................................................
Q23. Ngano ema inyalo ngiso ka itwo?
0. Onge 2. Jaoda 3. Janyuol na 4. Jomoko e od wa (Dimb wach)…
Q24. Mineme nyalu ketho hera kindi gi jomamoko? (Osiepe, joodu, jogwenge)
Q25. Ka dipo ni mineme omaki, wiyi nyalu kuot gi tuo no?
Q26. De iher pondo nikech in gi mineme (Achang)?
Q27. Kaka ot chalo (Gik gedo) ingiyo kod ja neno
Wi tado 1. Lum/Makuti 2. Mabati 3. Mamoko (Dimb wach)………
Kor ot 1. Chuodho/ Loo 2. Omwon gi smiti 3. Mamoko (Dimb wach)……

Bedo mar gik moko

Q28. Bene I pidho jamni ma moko e dala ni? 1. (Kamano) 2. (Ooyo)
Ka dwoko en “Kamano” jamni mage ma in godo dala ka? (Luor te ma kare)
Q29. I kano gi kanye
Q30. I lwoko jamni bang ndalo adi?
Q31. Itiyo kod ango sama I lwoko jamni go?..............................................
Appendix 3: Observation Checklist

MORBIDITY ASSESSMENT AND CHARACTERISATION OF FLEA SPECIES RESPONSIBLE FOR TUNGIASIS IN KISUMU COUNTY, KENYA

OBSERVATION CHECKLIST

Personal Identification
ID of respondent: ......................... Date of Birth: ....................... Sex: 1. (Male) 2. (Female)
Household number: ............................................. Observation Date: ...................................
Village: .................................................Observers Name: ....................................................

Prevalence and Intensity of Infestation
Q1. The presence of tungiasis? 1. (Yes) 2. (No)
Q2. Location and number of lesions (fill all that apply)

<table>
<thead>
<tr>
<th>Topographical location</th>
<th>Number of lesions</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Singly</td>
</tr>
<tr>
<td>Feet</td>
<td></td>
</tr>
<tr>
<td>1. Periungal</td>
<td></td>
</tr>
<tr>
<td>2. Between toes</td>
<td></td>
</tr>
<tr>
<td>3. Heel</td>
<td></td>
</tr>
<tr>
<td>4. Sole of feet</td>
<td></td>
</tr>
<tr>
<td>5. Instep/ lateral rim</td>
<td></td>
</tr>
<tr>
<td>6. Other (Specify)</td>
<td></td>
</tr>
<tr>
<td>Hands</td>
<td></td>
</tr>
<tr>
<td>7. Palms</td>
<td></td>
</tr>
<tr>
<td>8. Elbow</td>
<td></td>
</tr>
<tr>
<td>9. Fingers</td>
<td></td>
</tr>
<tr>
<td>10. Wrist</td>
<td></td>
</tr>
<tr>
<td>8. Other (specify)</td>
<td></td>
</tr>
</tbody>
</table>

Q3. Condition of lesion. 1. (Manipulated) 2. (Un-manipulated)
Q4. Stage of lesions (Fortaleza classification) (fill all that apply)

<table>
<thead>
<tr>
<th>Stage of lesion</th>
<th>Number of lesions</th>
</tr>
</thead>
<tbody>
<tr>
<td>Virulent stages</td>
<td></td>
</tr>
<tr>
<td>1. Stage 1 (Penetration)</td>
<td></td>
</tr>
<tr>
<td>2. Stage 2 (Erythemia)</td>
<td></td>
</tr>
<tr>
<td>3. Stage 3 (White halo)</td>
<td></td>
</tr>
<tr>
<td>Avirulent stages</td>
<td></td>
</tr>
<tr>
<td>4. Stage 4 (Inviable)</td>
<td></td>
</tr>
<tr>
<td>5. Stage 5 (Dead)</td>
<td></td>
</tr>
</tbody>
</table>

Tungiasis’ Pathology
Q5. Observed clinical pathology (circle all that apply)
1. Erythema 2. Suppuration 3. Deformation of toenails
7. Ulcer 8. Fissures 9. Other (Specify).............

Q6. Any other observation
................................................................................................................................................
................................................................................................................................................
................................................................................................................................................
................................................................................................................................................
................................................................................................................................................
................................................................................................................................................
Appendix 4: Consent Form

MORBIDITY ASSESSMENT AND CHARACTERISATION OF FLEA SPECIES RESPONSIBLE FOR TUNGIASIS IN KISUMU COUNTY, KENYA

PARTICIPANT INFORMED CONSENT FORM

This form is for all individuals invited to participate in our study. The title of our research project is “Morbidity assessment and characterisation of flea species responsible for tungiasis in Kisumu County, Kenya.”

Investigators and Institutional Affiliations

Aballa Andrew Okoth-PI\textsuperscript{1}, Odada Peter Sumba\textsuperscript{2}, Margaret Muturi\textsuperscript{1}, and Muhoho D. Ngethe\textsuperscript{1}

\textsuperscript{1}Kenyatta University, School of Health Science, P.O.Box 43844–00100, Nairobi, Kenya
\textsuperscript{2}KEMRI–CGHR, P.O.Box 1578–40100, Kisumu, Kenya

PART I: Information Sheet

Introduction

We are researching on tungiasis. You are invited to participate in this research. Take your time to read this form. You can also discuss it with your family members or friends before you make a decision. If you find words that that you do not know, you are free to ask me and I will explain.

Purpose of Research

We want understand how big a problem tungiasis is in this area. We want to know the number of people who are affected by tungiasis in your home and village. We also want to know the type of insects that cause tungiasis in this area. Such information will help us to know how to protect your family members and you, from the effects of this disease.

Participant Selection

We are inviting people have been living in this area for the past one month. It does not matter whether you have tungiasis or not.

Voluntary Participation

Before you accept, you should understand that participation in this study is voluntary. You can stop being a participant at any time, without any penalties.

Description of the Process
If you agree to be in this study, I will ask you a few questions about tungiasis. I will then check your hands and feet for signs of the disease. If you have tungiasis, I will clean the affected area using a disinfectant that will kill the tungiasis. This will take a few hours.

Risks

This study will not harm you in any way. Your neighbours and friends who do not know about tungiasis might however, talk bad about you.

Benefits

We will not pay you for participating in this study. We will however, train you on how to treat tungiasis and how to protect all your family members and you against the disease.

Confidentiality

Any information we collect about you during this study will be stored in safe location under lock and key. No friend or neighbour of yours will know that you participated in this study. Instead of writing your name, we will numbers that only researchers in this study will know about. I assure you that your name and other details you give me will remain between you and me.

Sharing the Results

We will share the information we get with other people who are learning about tungiasis and people living in this area. We will announce when these small meetings will take place in your village and invite you to attend. After the meetings, we will share the results with other people from all over the world.

Whom to Contact

If you have any questions you are free to ask them now, or later, after we have started the study. If you have questions, use the following contacts: 1) Andrew Aballa, P.O.Box 6031–40100 Kisumu, Tel. 0722289431, E-mail andrew.aballa@gmail.com, 2) Kenyatta University, P.O.Box, 43844–00100, Nairobi, Kenya, or 3) Secretary, KEMRI/ERC P.O. Box 54840–00200 Tel. 2722541 ext 3317, 0722 205 901, 0733 400003 Nairobi, Kenya.

PART II: Certificate of Consent

I have read this information, or it has been read to me. I have also had the opportunity to ask questions about it and all questions that I have asked have been answered to my satisfaction. I consent voluntarily to participate as a participant in this research.

Name of participant:........................Signature of participant:.....................Date:.....................
If Illiterate

*A literate witness must sign (selected by the participant and should have no connection to the research team). Illiterate participants should include their thumbprint as well.*

I have witnessed accurate reading this consent form to the participant. He or she has had the opportunity to ask questions. I confirm that he or she has given consent willingly.

Name of witness........................Signature of witness........................Date...................

Statement by the Researcher/Person Taking Consent

I have accurately read out the information sheet to the participant. I have ensured that he or she understands that we will do the following: 1) Interview him or her using a questionnaire, 2) check for tungiasis, and 3) trap fleas inside the house.

I confirm that the participant got an opportunity to ask questions about the study. I answered all questions correctly and to the best of my ability. I also confirm that the participant has not been coerced to giving consent. He or she has consented freely and voluntarily. The participant has retained a copy of the informed consent form.

Name of Researcher:....................Signature of Researcher:.....................Date:............................
Appendix 5: Consent Form (Luo)

TIMO NONRO KOD NGIYO NI KUTE MAGE MAKELO TUO MAR MINEME E GWENGJE MA NI KISUMU COUNTY, KENYA

YIE MAR JO DUOKO

Nying N’gama Otelo Ne Nonro

Aballa Andrew Okoth\(^1\), Odada Peter Sumba\(^2\), Margaret Muturi\(^1\), Muhoho D. Ngethe\(^1\)

1. Mbalaryany ma Kenyatta, 2. KEMRI–CVBCR

I: Weche ma nyaka jo duoko nge

Chakruok

Wa timo nonro mar tuo mar mineme e gweng u ka. Wa rawki mondo ibed achiel kwom joma wabiro tiyo godo e nonro ni. Kau seche mangeny mondo isom barua ni to kendo ibed thuolo some ji jo odu ma moko kata osiepe gi ka pok i miyo wa dwoko. Ka iyudo weche moko ma ikia kata ma ok wa lero maber to inayalo penja mondo amed lero ni.

Geno Mar Nonro Ni

Nonro wa ni dwaro ngiyo ni tu mar mineme chendo e okang manade e gweng ni. Dwoko ma wabiro yudo biro miyo wanyal tiego yore manyalo duoko tuo mar mineme e gwengje kaka ma. Mondo wanyal timo mano, onego watim nonro ma kama: (1) Wangi ni tuo mar mineme chendo e okang maromo nade e gweng ni (2) Wange ni jomo mineme omako chandore e okang maromo nade (3) Wange ni kute mage makelo mineme e gweng ni. Mondo wanyal ngeyo magi te, wadwaro ngeyo paro mar joma odak e gweng ni kaka in.

Kaka Ibiro Tim Nonro

Mokuongo, wabiro penji penjo to wabiro ndiko duoko te ma ibiro miyo wa. Ka wase timo mano, wabiro ngiyo dendi ka ni gi kute mag mineme.

Yiero Joma Ibiro Ti Godo E Nonro Ni

Wadwaro tiyo gi ji te ma odak e County ma Kisumo

Yie Mar Ja Duoko

Ka pok i yie bedo achiel ma wabro tiyo godo e nonro wa ni, nyaka inge ni bedo achiel kuom joma wabiro tiyo godo e nonro en dwaro mari. Wanyalo goli e nonro wa ni sa mora mora mora kata inyalo weyo bedo achiel kuom joma watiyo godo sa mora mora ma idwaro.
Kaka Ibiro Tim Nonro

Wabiro chako nonro ni gi penji penjo matin. Ka wasiete ko penj, wabiro ngiyo lweti gi tiendi ni gi kute mag mineme. Ka oyud ni in gi mineme to wabirothiethi kaka dwarore

Ndalo Ma Wabiro Kao Ka Watimo Nonro Ni

Nonro ni biro kao mana ndalo achiel kende lakini wanyalo duogo sa asaya ka ochuno.

Gima Nyal0 Hinyi

Onge gimora mora ma biro hinyi sama watimo nonro ni.

Gima Ibiro Yudo

Ka iyie bedo e nonro ni, ibiro yudo puonj ma kama: Wabiro puonji e yore ma inyalo gengori godo kuom kute makelo mineme. Ok wabi miyi pesa lakini yie mari mar bedo e nonro ni biro konyo wa tiego yore mag gengruok kod thietho mineme ma biro konyo jo gwengu duto kod nyithind nyitindu mabiro nyuol e higni ma biro.

Siri Na Kodi

Weche te ma wabiro wacho kodl biro bedo kinda kodl. Onge kama wabiro ndiko nyingi. Onge ngama biro ngiyo nyingi nikech wabiro tiyo gi number. E ndalo duto ma wabiro timo nonro ni, onge ngama biro ngiyo ngama in mita mana joma timo nonro ni.

Duoko


Ngama Onego Itudri Godo

Ka in gi penjo mora mora, inyalo penjo sani kata bange. Ka idwaro penjo bange to inyalo tudri Kod: 1) Andrew Aballa, P.O.Box 6031–40100, Kisumu, 0722289431, kata ndiko na barua e mbui, andrew.aballa@gmail.com, 2) 2) Kenyatta University, Medical Lab Sciences, P.O.Box, 43844–00100, kata 3) Secretary, KEMRI/ERC P.O. Box 54840–00200 Tel. 2722541 ext 3317, 0722 205 901, 0733 400003 Nairobi, Kenya.

II: Barua mar yie

A somo weche te ma ondik e barua ni, kata osom na weche tee. Ayudo thuolo mar penjo penjo ma an go, to oduoka ma kare. Ayie bedo achiel kuom jama ibiro ti godo e nonro ni.

Nying ja duoko:........................Sei mar ja duoko:........................Tarik:..........................
Ka Jadooko Ok Nyal Somo

Ja neno manyalo somo nyaka go sei (ka nyalore, jadooko onego oyier ngama odwaro ni obed ja neno ne. Ja neno ok onego obed achiel kuom joma timo nonro ni). Jo duoko ma ok nyal somo kata goyo sei nyaka ket alama mar Iwedo.

Awinjo ka osom ne jadooko ni weche ma ni e bara ni e yo ma kare. Ja duoko oyudo thuolo mar penjo penjo ma en godo. Jadooko oyie owuon bedo achiel kuom jama ibiro ti godo e nonro ni.

Nying ja neno..........................Sei mar ja neno..........................Tarik..............................

Wach Ja Tim Nonro


Nying ja nonro:..........................Sei mar ja nonro:..........................Tarik:..............................
Appendix 6: Parent Agreement Form (English)

MORBIDITY ASSESSMENT AND CHARACTERIZATION OF FLEAS SPECIES RESPONSIBLE FOR TUNGIASIS IN KISUMU COUNTY, KENYA

Purpose of Research
This study evaluates the degree of the problem that tungiasis causes in this area. Results that we get from this study will help us formulate an effective disease control strategy.

Voluntary Participation
Before you accept, know that consent to participate in this study is voluntary. You may withdraw your child from this study whenever you feel like, without any penalties.

Risks/Benefits
You may face the risk of stigma from community members who have a bad perception about people who have tungiasis. We will do our level best to lower such risks. Although we will not pay you for enrolling your child or children in this study, you will enjoy the following benefits: First, we will train you on how to manage jigger infestation. Finally, you will also learn how to take care of your home to protect yourself against tungiasis.

Confidentiality
Any personal information collected during this research will remain between you and me. No community member will know that your child participated in this study. Instead of writing a name, your child will have a reference number that only researchers will know. If you have any questions about your child’s rights as a participant, you can contact me via my mobile (0722289431), or the Secretary, KEMRI/ERC P.O. Box 54840–00200 Tel. 2722541 ext 3317, 0722 205 901, 0733 400003 Nairobi, Kenya.

Statement of Consent
I read the above consent form. Andrew Okoth Aballa has explained the nature, demands, risk, and benefits of the project to me. I am aware that I have the opportunity to ask questions about this research. I also understand that I may withdraw my consent and discontinue my child’s or children’s participation at any time without penalty.

…………………………………..……………………………………………………………………………
Child’s Name Signature of Legal Guardian Date

I have explained to the above named individual the nature and purpose, the benefits and possible risks associated with participation in this study. I have answered all questions raised by the guardian. I have also given the guardian a copy of this signed consent form.

…………………………………..……………………………………………………………………………
Researcher Date
Appendix 7: Parent Agreement Form (Luo)

TIMO NONRO KOD NGIYO NI KUTE MAGE MAKELO TUO MAR MINEME E GWENGE MA NI KISUMU COUNTY, KENYA

Geno Mar Nonro Ni
Wa timo nonro mar tuo mar mineme e gweng u ka. Gik ma wabiro ngeyo ka wasetieko nonro ni biro konyo wa tiego yore mag tieko tuo mar mineme e gwengu ka.

Yie Mar Ja Duoko

Ka pok i yie bedo achiel ma wabro tiyo godo e nonro wa ni, nyaka inge ni bedo achiel kuom joma wabiro tiyo godo e nonro en dwaro mari. In thuolo golo nyathini e nonro ni saa asaya ma idwaro.

Gima Nyalu Hinyi/ Ma Ibiro Yudo

Onge gima biro hinyo nyathini sama watimo nonro ni. Moloyo, ka iyie ni nyathini obed a chiel kuom joma wabiro tiyo godo, ubiro yudo puonj ma kama: ok wabi chuli pesa mondo nyathini obed a chiel kuom joma wabiro tiyo godo. Kata kamano, wabiro puonji e yore ma inyalo gengori godo kuom kute makelo mineme. Yie mari mondo nyathini obed achiel kuom joma wabiro tiyo godo e nonro ni biro konyo wa tiego yore mag gengruok kod thietho mineme ma biro konyo jo gwengu duto kod nyithind nyitindu.

Siri Na Kodi


Barua Mar Yie

Asomo barua mar yie ni maber. Andrew Okoth Aballa o nono na gik ma odwaro timo gi nonro ni, gik ma nyalu hinya, kod gik ma abiyo youdo ka nyathini obedo achiel kuom joma gibirotiyo godo. Penjo te mane an godo jo nonoro tiego na e yo ma kare. Mogik, jo nonro ni olero na e you ma kare ni anyalo golo nyathini e nonro saa asaya ma adwaro.

…………………………………………………………………………………………………………………………
Nying Nyathi Sei mar Ja Nyuol Tarik

Alero ne janyuol nyathini gima nonro ni en, gima gibro yudo, kod gima nyalu hinyo gi. Aduoko peno tee mane engodo e you makare. Aweyo ne copy mar barua mar yie ni.

…………………………………………………………………………………………………………………………
Ja Nonro Tarik
Appendix 8:  Child Assent Form (English)

MORBIDITY ASSESSMENT AND CHARACTERISATION OF FLEA SPECIES RESPONSIBLE FOR TUNGIASIS IN KISUMU COUNTY KENYA

My name is Andrew Okoth Aballa. I am trying to stop tungiasis from affecting children and adults in your village. To do this, I must learn how you and other family members live in at home. I must also know how many people are currently suffering from tungiasis in your village, and the kind of insects that are causing the disease.

If you decide to help me collect this information, all I will do is check your hands and feet to see if you are suffering from tungiasis. If you have any tungiasis wounds on your hands and or feet, I will clean and then treat them well, without causing you pain.

Even though I will not pay you for helping me to learn about tungiasis, you will enjoy other good things. First, I will teach you how to clean your hands and feet, to prevent tungiasis from getting into them. You will also learn how to clean your surroundings, to prevent tungiasis from growing in numbers, and causing problems in your home.

After I have finished learning about tungiasis in your home and those of other children like you, I will write a report of the information I got, and share it with other people in Kenya. This does not mean that I will tell them your name or details, when talking about what I learn. Your name will appear nowhere on the report.

Your parents have said that it is okay for you to help me learn about tungiasis in your village. However, the decision to help me collect information is yours and no one else’s. It is okay if you do not want to be a part of this study. No one will be mad at you. If you want to be in the study now and change your mind later, that is also okay. You can stop helping collect information on jigger at any time that you so please.

If you have any questions on what we have discussed, feel free to ask them now. If you experience any problems in future, my telephone number is 0722289431. You can also contact the Secretary, KEMRI/ERC P.O. Box 54840–00200 Tel. 2722541 ext 3317, 0722 205 901, 0733 400003 Nairobi, Kenya.

Agreement

I have decided to be in the study even though I know that I do not have to do it. Andrew Okoth Aballa has answered all my questions.

_________________________________________  _____________
Signature of Study Participant                      Date

_________________________________________  _____________
Signature of Researcher                              Date
Appendix 9:  Child Assent Form (Luo)

TIMO NONRO KOD NGIYO NI KUTE MAGE MAKELO TUO MAR MINEME E GWENGE MA NI E COUNTY MA KISUMO, E KENYA


Ka iyie bedo anchel kuom joma wabiro tiyo godo e nonoro ni, wabiro chako nonro ni gi penji penjo matin. Ka wasietieko gi penjo, wabiro ngiyo lweti gi tiendi ni mondo wangi ka ni gi kute mag mineme. Ka oyud ni in gi mineme to wabirothiethi kaka dwarore. Onge lit ma ibiro winjo sama watimo magi

Ok wabi miyi pesa lakini yie mari mar bedo e nonro ni biro konyo wa tiego yore mag gengruok kod thietho mineme ma biro konyo jo gwengu duto kod nyithind nyitindu mabiro nyuol e higni ma biro. Ka iyie bedo e nonro ni, ibiro yudo puonj ma kama: Wabiro puonji e yore ma inyalo gengori godo kuom kute makeno mineme.

Ka asetieko nono weche mag mineme e gwengu, abiro ndiko ripot manyiso gima ayoudo. Kata kamano, weche te ma wabiro wacho biro bedo kinda kodi. Onge kama wabiro ndiko nyingi. Onge ngama biro ngeyo nyingi mita mana joma timo nonro ni.

Jo nyuol ni ose yie ni Inyalo konyo nono weche mag mineme e gwengu. Kata kamano, onge ngama biro goyi, kata nyisi weche ma richo, ka itamori tiyo koda. Mogik, kata ka I yie tiyo koda sani to I loko pachi bange, onge ngama biro dhao ni, kata tami weyo nono ni. Oyie ni weyo nonro mar mineme ni sa mora mora ma idwaro.

Ka in gi penjo mora mora e weche ma wase wacho ka, in thuolo penjo gi sani. Ka iyudo chandruok mora mora e ndalo mabiro, namba na mar simo en 0722289431. Inyalo ywa na toll saa asaya. Ka ok iyuda ndik ne ja goro mar KEMRI ERC e: KEMRI/ ERC P.O. Box 54840–00200 Simu. 2722541 ext 3317, 0722 205 901, 0733 400003 Nairobi, Kenya

Winjruok

Ayie bedo anchel kuom joma ibiro ti godo ka inono weche mage mineme e gweng wa. Andrew Okoth Aballa oduoko penjo te mane an godo e yo ma kare.

…………………………………

Sei Mari  Tarik

…………………………………

Sei Mar ja Nonro  Tarik
Appendix 10: Flea Identification Scheme

**GENAL AND PRONTAL COMBS PRESENT**

- Genal combs of five or more spines present and eyes present
  - Genal combs horizontal with pointed spines
    - Head less than twice body height. First spine of Genal comb shorter than the second
      - Dog flea (*Ctenocephalides canis*)
    - Head length twice body height. Spine 1 and spine 2 of Genal comb approximately equal
      - Cat flea (*Ctenocephalides felis*)
  - Genal combs four in number and eyes missing
    - Mouse Flea (*Leptospylla seginis*)

- Genal combs vertical with blunt spines
  - Rabbit Flea (*Cadiopsylla symplex*)

**GENAL AND PRONTAL COMBS ABSENT**

- Thorax normal, front margin of head rounded.
  - One row of bristles on abdominal segments
    - Mesopleuron not divided, ocular bristles below the eye
      - Pulex irritans
  - Thorax contracted, front margin of head angular
    - Two rows of bristles on abdomen
      - Polygenis gwyni

- Head shield or helmet shaped. Spines quarter way on the third leg tibia
  - Tunga spp.
Appendix 11: Flea Trap

A locally developed trap was used to collect free-living fleas overnight in study houses where cases were diagnosed. All traps utilised kerosene lanterns as flea attractants. Lit lanterns were placed next to four shallow troughs, half filled with water. A non-ionic detergent (Savlon) was used to break surface tension, while maintaining the samples in an excellent state, for molecular analyses. Tunga fleas are poor jumpers. At full flight, they run with a velocity of up to one centimetre per second, and can leap to a height of up to 20 centimetres. To maximise the number of fleas trapped per session, troughs were around three centimetres deep, and over 25 centimetres wide. As light and heat from the lantern attracted fleas, they landed on the water, and drowned.
Appendix 12: Kenyatta University Approval Letter

KENYATTA UNIVERSITY
GRADUATE SCHOOL

P.O. Box 43844, 00100
NAIROBI, KENYA
Tel. 810901 Ext. 57530

Internal Memo

FROM: Dean, Graduate School
TO: Aballa Andrew Okoth
    C/o Medical Lab Sciences Dept.

DATE: 16th July, 2012
REF: F150/21572/10

SUBJECT: APPROVAL OF RESEARCH PROPOSAL

This is to inform you that Graduate School Board, at its meeting of 9th July, 2012, approved your Research Proposal for the M.Sc Degree entitled “Morbidity Assessment and Characterisation of Flea Species Responsible for Tungiasis in Kisumu County, Kenya.”

You may now proceed with your Data Collection.

Thank you.

JOSEPHINE NJAGI
FOR: DEAN, GRADUATE SCHOOL

c.c. Chairman, Medical Lab Sciences Department

Supervisors:
1. Dr. Margaret Muturi
   C/o Dept of Medical Lab Sciences

2. Dr. Muheho Ng’ethe
   School of Health Sciences

3. Dr. Odada Peter Sumba PhD,MPH
   Kenya Medical Research Institute (KEMRI)
   P.O. Box 1578 – 40100, KISUMU
   C/o Dept of Medical Lab Sciences
Appendix 13: KEMRI SSC Approval Letter

KENYA MEDICAL RESEARCH INSTITUTE

P.O. Box 54840-00200, NAIROBI, Kenya
Tel (254) (020) 2722541, 2713349, 0722-205901, 0733-400003; Fax: (254) (020) 2720030
E-mail: director@kemri.org  info@kemri.org  Website: www.kemri.org

ESACIPAC/SSC/101326

Andrew Abulla

16th January, 2013

Thro’
Director, CGHR
KISUMU

KISUMU

REF: SSC No. 2511 (New)–Morbidity assessment and characterization of flea species responsible for jiggers in Kisumu County, Kenya

I am pleased to inform you that the above mentioned proposal, in which you are the PI, was discussed by the KEMRI Scientific Steering Committee (SSC), during its 198th meeting held on 15th January, 2013 and has since been approved for implementation by the SSC.

Kindly submit 4 copies of the new protocol to SSC within 2 weeks from the date of this letter i.e, 30th January 2013.

We advise that work on this project can only start when ERC approval is received.

Sammy Njenga, PhD
SECRETARY, SSC

In Search of Better Health
Appendix 14: KEMRI ERC Approval Letter

KENYA MEDICAL RESEARCH INSTITUTE

P.O. Box 1578, KISUMU

KEMRI/RES/73/1

August 13, 2013

TO: ANDREW ABALLA
PRINCIPAL INVESTIGATOR

THROUGH: DR. STEPHEN MUNGA,
ACTING DIRECTOR, CGHR
KISUMU

Dear Sir,

RE: SSC PROTOCOL NO. 2511 – REVISED (RE-SUBMISSION): MORBIDITY ASSESSMENT AND CHARACTERISATION OF FLEA SPECIES RESPONSIBLE FOR JIGGERS IN KISUMU COUNTY, KENYA

Reference is made to your letter dated August 2, 2013. The ERC Secretariat acknowledges receipt of the revised proposal on August 8, 2013.

This is to inform you that the Committee determines that the issues raised at the 213th meeting held on 19th March, 2013 have been adequately addressed. The study is granted approval for implementation effective this 13th day of August 2013. Please note that authorization to conduct this study will automatically expire on August 12, 2014. If you plan to continue with data collection or analysis beyond this date, please submit an application for continuing approval to the ERC Secretariat by July 1, 2014.

Any unanticipated problems resulting from the implementation of this protocol should be brought to the attention of the ERC. You are also required to submit any proposed changes to this protocol to the ERC prior to initiation and advise the ERC when the study is completed or discontinued.

You may embark on the study.

Yours faithfully,

DR. ELIZABETH BUKUSI,
ACTING SECRETARY,
KEMRI ETHICS REVIEW COMMITTEE
Appendix 15: Ministry of Education Approval Letter

MINISTRY OF EDUCATION

Telegram:
Telephone: Kisumu (057) 2022626
When replying please quote

DISTRICT EDUCATION OFFICE
KISUMU WEST DISTRICT
P.O. BOX 19
PAW-AKUCHE

REF: KWD/GA/23/8/VOL.1/75

TO ALL HEADTEACHERS
OTWENYA ZONE


The above mentioned officer is authorized to carry out research in Otwenya zone in Kisumu West District. "Morbidity Assessment and Characterization of Flea Species Responsible for Tungiasis in Kisumu County, Kenya."

Kindly accord him the necessary assistance.

GEORGE OUMA
FOR: DISTRICT EDUCATION OFFICER
KISUMU WEST.
Appendix 16: District Commissioner Approval Letter

OFFICE OF THE PRESIDENT

Telegram: DISTRICTER, HOLO
Telephone: 0202674771
Email: dckisumuwest@yahoo.com
When replying please quote:

OFFICE OF THE DISTRICT COMMISSIONER
KISUMU WEST DISTRICT
P. O. BOX 4
PAW· AKUCHE

REF: ADM/3 VOL.1/127

12th March, 2013

TO WHOM IT MAY CONCERN

RE: RESEARCH AUTHORIZATION

This is to confirm that Mr. Abdalla Andrew Okoth, a Postgraduate Student in the Department of Medical Lab Sciences at Kenyatta University, has been authorized to conduct a research proposal on “Assessment and Characterization of Flea Species Responsible for Tungiasis in Kisumu County,” within Kisumu West District, for period ending 31st July, 2013.

Kindly accord him the necessary assistance.

G. B. KEITU
DISTRICT COMMISSIONER
KISUMU WEST DISTRICT

cc

The Medical Officer of Health
KISUMU WEST.