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DEPARTMENT OF PUBLIC HEALTH

ASSESSMENT OF OXYTETRACYCLINE RESIDUE LEVELS IN BEEF CONSUMED IN DODOMA MUNICIPALITY CATCHMENT AREA, TANZANIA.

ENGELBERT BILASHOBOKA

MSC.EPIDEMIOLOGY

SUPERVISORS: **DR. B.MUDENDA** (BSc, MBChB, MSc, MD, FRCSEd)

MS NOSIKU MUNYINDA (BSc,MSc, PhD candidate)

PROF.DOMINIC M. KAMBARAGE (DVM, MVM, PhD.)

'Thesis submitted in partial fulfillment of the requirements towards the award of Master of Science Degree in Epidemiology'

DECLARATION
I Engelbert Bilashoboka hereby declare that this research report being presented for Masters
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Degree.
Signed: Date:
Engelbert Bilashoboka
(Candidte)
Supervisors:
I have read this dissertation and approved it for examination.
Dr Boyd Mudenda(Supervisor)
Signed: Date:
Department of Public Health, School of Medicine, University of Zambia
Co-supervisor:
I have read this dissertation and approved it
Mrs Nosiku Munyinda(Co-supervisor)
Signed: Date:
Department of Public Health, School of Medicine, University of Zambia

Mwalimu Julius K. Nyerere University of Agriculture and Technology, Musoma, Tanzania

Signed: Date:

Co-supervisor:

I have read this dissertation and approved it

 ${\bf Prof.D.M. Kambarage} ({\bf Co\text{-}supervisor})$

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Examiner:	Date:
Examiner:	Date:
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Head of Department:	
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DEDICATION

I dedicate this research to my loving wife, Fabiola Moshi and to our lovely sons Justus, Julian and Junior Engelbert. My attention to them was divided during my travel away to attend my studies. Their perseverance, love and prayers have made me accomplish this research.

ABSTRACT

Background: Worldwide, antimicrobials are widely used in food producing animals for treatment and/or prophylaxis of various bacterial infections because of continued high disease burdens as a result of poor animal health service delivery systems. For instance, in Tanzania, indiscriminate and irrational administration of antimicrobials, notably oxytetracycline (OTC) is not uncommon in rural areas because of under-developed animal health service delivery structure. The administration of OTC and other drugs is often done by animal keepers, without proper guidance of extension agents, thereby potentially leading to drug residues in meat and milk due to non-adherence to drug wash out periods. Despite this potential public health threat, to date there is no data about the extent of drug residue levels in beef consumed in places like Dodoma municipality where meat consumption is high.

Objectives: The main objective was to establish the level of oxytetracycline residue levels in beef consumed within Dodoma Municipality catchment area.

Methodolgoy: This was a cross-sectional study for which data on the knowledge, concerns and practices of animal keepers, traders, people involved in slaughtering animals and consumers was gathered using a semi-structured questionnaire. Quantification of drug residues in muscle, liver and kidney samples were collected and used in determining OTC levels using HPLC method. The quantitative data were analysed by Benferron's and Student-Newman Keuls' tests using Stata software version 13 for comparison of the residues between licensed and unlicensed premises and qualitative data were entered into Nvivo matrix for thematic analysis.

Results:The outcomes of the study were: (1) The level of OTC residues were 0.6 mg/Kg, 0.25 mg/Kg and 1.28 mg/Kg for muscle, liver and kidney tissues respectively. These levels are unacceptably high when compared to Maximum Residue Limit (MRL) of 0.2mg/Kg, 0.6mg/Kg and 1.2mg/Kg for muscle, liver and kidney respectively as set by Codex Alimentarius Commission (CAC). The quantities of OTC residues in samples from licensed and unlicensed food vending settings were not significantly different (p = 0.3676). Regardless of the licensure status and source of the sample, 53% of muscle, 65% of liver and 7.1% of kidney tissues had residue amounts which were above MRL. (2) Level of knowledge, concerns and practices of animal keepers, consumers and extension agents in relation to drug withdrawal requirements were assessed. Most of animal keepers interviewed were ignorant of drug residues and withdrawal periods. The majority of consumers were not aware of the drug residues in beef whereas the businessmen and law enforcers were aware.

Conclusion: The above results show how unacceptably high levels of drug residues have been introduced into the food chain and most of the consumers and farmers being ignorant of the impending public health threat of drug residues. The obtained results will be used in developing appropriate public health strategies in addressing drug residues which are now regarded as a global health threats and that must be tackled through a one health approach. It is envisaged that raising the awareness of beef value chain actors will be pivotal in reversing these negative trends.

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ABBREVIATIONS

μg/g Microgram per gram

A/B Antibiotic

ADI acceptable daily intake

CAC Codex Alimentarius Commission

CI Confidence Interval

EDTA Ethylene diamine tetraacetic acid

FAO Food and Agriculture Organization

HACCP Hazard Analysis Critical Control Programme

HPLC High Performance Liquid Chromatography

ml millilitre

mm milimeter

MRL Maximum Residue Limit

OTC Oxytetracycline

rpm revolutions per minute

Std Standard

TC Tetracycline

TFDA Tanzania Food and Drugs Authority

TFDC Act Tanzania Food Drugs and Cosmetic Act

TLC Thin Layer Chromatography

tRNA transfer Ribonucleic acid

UNZABREC University of Zambia Biomedical Research Committee

WHO World Health Organization

OPERATIONAL DEFINITIONS

Anaphylactic reactions: Severe and life-threatening allergic reactions which occur within seconds or minutes of exposure to something one is allergic to.

Drug residues: The very small amounts of veterinary medicines that can remain in animal products and could inadvertently make their way into the human food chain.

Drug resistance: WHO defined drug resistance as the resistance which occurs when microorganisms such as bacteria, viruses, fungi and parasites change in ways that render the medications used to cure the infections they cause ineffective.

Drug washout period: The WHO definition of washout period, as withdrawal time not to ingest the animal product, after a drug is administered to any food producing animal to allowable acceptable levels in a marketable food product such as meat, organ, eggs or other edible products.

Maximum Residue Limit: The maximum concentration of residues following administration of a veterinary medicine which is legally permitted or acceptable in food.

Toxic reactions: The reactions which occur after ingesting the food containing the antibiotic residues.

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CHAPTER ONE

1.0 Introduction

1.1 Background Information

Antibiotics are among the most widely used veterinary drugs in food-producing animals for therapeutic purposes and as dietary supplement. They may be administered orally as food additives like oxytetracycline (OTC) and sulfadimidine or directly by injection like OTC injection, Penistreptomycin, Gentamycin and Sulfadimidine to mention but a few (Abasi et al., 2009). Among the veterinary antibiotics, oxytetracycline is one of the most widely used antibiotics due to its broad spectrum activity and easy access by practitioners (Kaneene and Miller, 1997).

As pointed out, oxytetracycline works by interfering with the ability of bacteria to add new amino acids to the peptide chain to produce essential proteins. Without these proteins, the bacteria cannot grow, multiply and increase in numbers (Chopra and Roberts, 2001). Oxytetracycline therefore stops the spread of the infection and the remaining bacteria are killed by the immune system or eventually die. But resistance against this antibiotic is being built by strains of bacteria due to drug residues consumed in animal products.

In developing countries, due to scarcity of veterinary professionals and lack of enforcement of veterinary regulations, the drug is administered by lay persons whenever the demeanor of the animal is detected to be dull. In such a scenario, uncontrolled injection of animals with no treatment records makes identification of those treated very difficult. According to Bedada and Zewde (2012) the control of drugs from the government authorities and information on the actual rational drug use pertaining to veterinary drug is very limited. Some antibiotics are sold casually by street vendors like any other commodity with no regard for their use. Consequently there is a widespread and indiscriminate use of tetracyclines, the once effective broad spectrum antibiotic against both gram negative and positive organisms, has resulted in many bacteria acquiring resistance.

The Codex Alimentarius Commission/ Maximum Residue Limit stipulates that once the animal is injected with oxytetracycline, the antibiotic residues remain in the body of an

animal for 28 days and during this period, the animal should not be slaughtered for human consumption (Ahmadi et al., 2014). This however does not happen as many of these animals are sent for slaughter before the drug washout period has elapsed. Such a malpractice can have disastrous impact to the health of the consumers. The beef consumed with high levels of residue has cumulative effect of causing toxic and anaphylactic reactions to the consumers. This study aims at determining the levels of oxytetracycline residues in beef consumed in Dodoma.

1.2 Statement of the problem and justification

The oxytetracycline drug residues pose a real public health hazard to the consumer due to this exposure and can lead to various toxic effects including, anaphylactic reactions and development of drug-resistant strains of bacteria. According to Codex Alimentarius Commission of the FAO and WHO, the current standard maximum residue level (MRL) in beef muscles is 0.2 microgram per gram of beef (Ahmadi et al., 2014). However, due to the profit motive, farmers and business men do not follow the FAO and WHO guidelines. Once the animal falls ill and is treated with oxytetracycline, upon slight recovery the animal is sent for slaughter without taking into consideration of the drug washout period.

Even at the ranch setting where Hazard Analysis Critical Control Points (HACCP) is not applicable, some of the animals are sent for slaughter before the drug washout period of oxytetracycline is observed. Thus, the meat enters the market with oxytetracycline drug residues. This can lead to toxic reactions, anaphylactic reactions and development of antibiotic resistant strains of bacteria. In most of developing countries, slaughter houses/slaughter slabs do not have facilities for detecting drug residues.

As stipulated in the Animal Disease Act (Tanzania, 2003a) and Tanzania Food, Drugs and Cosmetic Act (Tanzania, 2003b), the washout periods of drugs are supposed to be observed before the animals are sent for slaughter. But due to absence of serious enforcement on these acts, animals have been slaughtered regardless of the washout periods. This poses a real threat to public health.

To date there is no study which has been conducted to establish levels of OTC residues in beef consumed at Dodoma municipality in Tanzania.

1.3 Significance of the study

The obtained results will work as a benchmark for advice to the law enforcers.

This will also create consumer awareness depending on the preference of the parts of beef. Being aware of the risks consumers can make an informed decision on the meat products they want to buy for consumption and awareness can empower the public to demand improved control to reduce the problem.

1.4 Research questions

- 1. What are the levels of oxytetracycline residues in beef consumed in Dodoma Municipality catchment area?
- 2. To what extent are the established levels between the licensed and unlicensed premises above the recommended MRL by Codex Alimentarius?
- 3. To what extent are the farmers, businessmen, consumers and law enforcers informed on drug washout period?

1.5 Objectives

1.5.1 Main objective

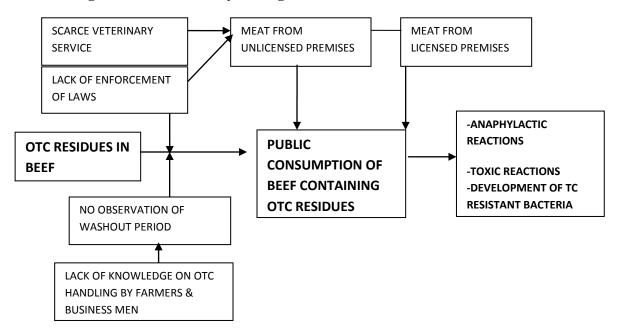
To establish the level of oxytetracycline residue levels in beef consumed within Dodoma Municipality catchment area.

1.5.2 Specific objectives

- 1. To analyse the levels of oxytetracycline residues from raw beef samples.
- 2. To compare the levels of OTC residues between the Licensed and Unlicensed premises with respect to MRLs.
- 3. To assess the knowledge, practice and attitudes of farmers, businessmen, consumers and law enforcers on drug washout periods.

1.6 Conceptual Framework

Figure 1. Problem analysis diagram



The scarcity of veterinary service and lack of enforcement of laws on control of use of veterinary antibiotic have prompted farmers to treat animals on their own and at times allowing treated animals to go for slaughter before the washout periods. Together with beef from unlicensed premises, oxytetracycline residue finds its way into the food chain for public consumption which poses the public health threat of toxic, anaphylactic reactions and development of oxytetracycline resistant bacteria. The quantification of OTC levels in beef will therefore be established from the beef from authorized and unlicensed premises and the findings will be the basis for advice to the relevant regulatory authorities for corrective action.

CHAPTER TWO

2.0 Literature Review

2.1 What is Oxytetracycline?

Oxytetracycline has been defined by Almaany English-English Dictionary as the yellowish crystalline antibiotic obtained from a soil actinomycete used to treat various bacterial and rickettsial infections. According to Chopra and Roberts (2001), OTC was discovered late 1940 from Streptomyces aureofaciens. The antibiotic works by inhibiting bacterial protein synthesis by preventing the association of aminoacycl-tRNA with the bacterial ribosome (Schnappinger and Hillen, 1996). The inhibition is done by Tetracycline through penetrating into the outer membrane of gram negative enteric bacteria as positively charged cation complexes. But studies have noted a remarkable resistance against this antibiotic 50 years after the discovery of this antibiotic. This resistance has been partly attributed to the use of OTC in food animals for treatment and prophylaxis purpose without proper observation of the withdrawal period (Singer et al., 2006).

2.2 Use of Oxytetracycline

2.2.1 Use of OTC in Developing Countries

Oxytetracycline is a broad spectrum antibiotic which is widely used against gram negative and gram positive bacteria including Rickettsia, Mycoplasm, Chlamydia and some protozoa. In most cases it is used to treat respiratory and gastrointestinal tract infections (Schnappinger and Hillen, 1996).

Developing countries have faced scarcity of veterinary professionals and paraprofessionals. There has been also lack of enforcement of veterinary regulations (Grasswitz et al., 2004). The drug is administered by lay persons whenever the demeanor of the animal is detected to be dull. Such a scenario of uncontrolled jabbing of animals leaves neither treatment record whatsoever nor caution on withdrawal periods. This practice has been branded as an indiscriminate use of antibiotics which could lead to residue deposition in edible tissues in a similar study which was done in Nigeria (Olatoye and Ehinmowo, 2010). The residues apart from toxic and anaphylactic reactions can also lead to alteration of the intestinal ecology leading to emergency of resistant microflora.

The resource-poor farmers in rural and peri-urban areas have little access to veterinary care from state and private veterinarian, veterinary paraprofessionals, animal health technicians and information about the prevention and treatment of livestock diseases as well as the drug washout periods of certain administered drugs (Cheneau et al., 2004).

Privatization was found to be the remedy but according to the study done in Tanzania and Kenya, it has resulted in the concentration of private practice veterinarians in urban, periurban and high potential farming areas, leaving more marginal farming areas without proper veterinary supervision (Cheneau et al., 2004). This has resulted in the farmers with no option but to treat the animals on their own without leaving any treatment record or observation of the drug washout periods.

A similar study conducted in Ethiopia (Bedada and Zewde, 2012) portrayed the lack of control of veterinary drugs and lack of rational use of drugs to be the major cause of introduction of drug residues in the food chain. It was further established that these antibiotics were sold on the market without prescription or along the road by informal vendors. Despite these gross violations, the regulatory authorities and producers in Ethiopia were not aware and were not prepared to deal with the risk of indiscriminate use of antimicrobials to the livestock and to the consumers.

2.2.2 Use of OTC in Developed Countries

In the United States the most commonly used tetracyclines in animals are chlortetracycline, tetracycline, oxytetracycline, doxycycline, and minocycline, with oxytetracycline being the most broadly used (Martin-Jimenez et al., 1997). Despite its popularity in the United States, this antibiotic is not officially approved for use in all domestic species or in all possible clinical situations. Thus, extra label use of oxytetracycline in food animals is common, and appropriate withdrawal times may not correspond with the ones established for label use.

2.3 Human Health Effects of OTC Residues

The human health effects of OTC residues are of two types namely, direct and short term hazards, and indirect and long term hazards (Muhammad, 2009).

Acute health effects

The direct and short term hazards can be experienced when edible animal tissues are consumed while containing maximum residual levels (Salehzadeh et al., 2006). This can result into direct toxic reactions to consumers who are allergic or hypersensitive to certain antibiotics (Horie et al., 2003). Apart from impact to human beings, the study done in Pakistan indicated diclofenac residue to have declining impact on the population of vultures (Oaks et al., 2004).

Chronic health effects

The indirect and long term hazards include microbiological effects, carcinogenicity, reproductive effects and teratogenic effects. The microbiological effects are one of the major health hazards in human beings (Nonga et al., 2009). Antibiotic residues consumed along with edible tissues like milk, meat and eggs can produce resistance in bacterial populations in the consumers. This contributes to therapeutic failures amongst such peoples (Gustafson and Bowen, 1997) because of the built up antibiotic resistance in the bacterial population due to continued exposure to antibiotic residues in the food chain.

It is also important to note that improper dosage of oxtetracycline especially at subtherapeutic levels can result in acute or chronic public health problems, which could be toxicological, microbiological or immunological including renal damage, drug resistance and allergic reactions respectively (Karimuribo, 2005).

2.4 Control of use of OTC

Due to the lagging behind of veterinary sectors in developing countries, the advisory services and treatment services to the sick animals have been left in the hands of the owners.

According to the study done in India (Rajput et al., 2012) Tetracyclines are most widely used antibiotics in veterinary medicine due to its broad spectrum of antimicrobial activity, availability and low cost. Unauthorized use of these antibiotics, the failure to follow label directions or inappropriate drug washout period before slaughtering of animals could lead to residues in food of animal origin, with potential adverse effects on human health.

From the study conducted in Nigeria (Olatoye and Ehinmowo, 2010), the greater proportion of cattle in Nigeria are reared by the nomadic herdsmen who administer chemotherapeutic

agents without veterinary prescription, a scenario which depicts to what extent the antibiotics are not under control of regulatory authorities. When such laymen use these drugs, correct dosage are unlikely to be administered and the drug washout periods are usually not observed.

This is attributed to the failure of regulatory authority to enforce the guidelines on rational use of veterinary drugs as depicted by the similar study in Ethiopia (Bedada and Zewde, 2012).

Tanzania like other developing countries is faced with acute shortage of veterinary practitioners, regulators and law enforcement personnel. The owners of the animals access the antibiotics without prescription and treat their animals with irrational doses and reasons. At times these animals are sent for slaughter without observing the washout period hence a high risk of meat entering the food chain with drug residues.

2.5 OTC washout period

This is the period during which the animal after being injected with oxytetracycline the animal is not allowed to be slaughtered for human consumption. The concentrations differ from one organ to the other. For oxytetracycline, the drug washout period is 28 days after the final treatment. Before this period the animal is not allowed to be slaughtered for human consumption (Capleton et al., 2006). The failure to follow recommended withdrawal times often results in residue problems.

2.6 OTC drug residues in beef

Oxytetracycline drug residues are the very small amounts of oxytetracycline that can remain in animal products and therefore make their way into the food chain. These residues include any degradation products, which are the result of the medicine breaking down. The antibiotic residues in food contributed 41.17% of the drug residues in food (Darwish et al., 2013). But this study blanketed the extent of antibiotic residues without specifically stating the types of antibiotics which contributed to that residue. A similar study conducted in Egypt tried to focus on oxytetracycline residues by examining 600 samples (made up of 200 samples from muscle, liver, and kidney) which were randomly obtained from bovine carcasses slaughtered at the Mansoura Abattoir (Salama et al., 2011). Two percent of samples tested positive for residues. Oxytetracycline residues exceeded the MRL.

Another similar study was done in Ethiopia. It was a cross-sectional study which was conducted from October 2006 to May 2007 to estimate the proportion of tetracycline levels in beef. The study focused on the Addis Ababa, Debre Zeit, and Nazareth slaughterhouses. Out of the total 384 samples analyzed for tetracycline residues, 71.3% had detectable oxytetracycline levels. Among the meat samples collected from the Addis Ababa, Debre Zeit, and Nazareth slaughter houses, 93.8%, 37.5%, and 82.1% tested positive for oxytetracycline. About 48% of the edible tissues had oxytetracycline levels above the recommended maximum limits (Bedada and Zewde, 2012). These results are comparatively higher in relation to Canada where there is zero tolerance of drug residues in food products. As pointed out (Salisbury et al., 1990) on an investigation on OTC residues in pork in Canada conducted on October 1987 up to March 1988, 2% of the products were found to have traces of residues which were below the MRL. But because of the law of zero tolerance all of the products were condemned.

Whereas, the study conducted in Tanzania, antimicrobial residues were detected in 36% of marketed milk samples from milk supply chains in and around Mwanza and Dar es Salaam during 1999 and 2000 (Kurwijila et al., 2006). But this study did not specify the type of antimicrobial residues. In another study, the occurrence of antibiotic residues in commercial chicken eggs was determined in the Morogoro municipality between January and February 2007. All eggs examined tested positive for antibiotic residues. The main residues detected were oxytetracycline, chlortetracyclines, chloramphenicol, doxycycline, and flumequine (Nonga et al., 2009).

From the literature review above, there is paucity of information with regard to the quantification of the OTC drug residue levels in beef in Tanzania more precisely in Dodoma municipality catchment area which is the likely scenario in other parts of the country. The obtained information will be the benchmark for gauging other parts in Tanzania.

2.7 Maximum Residue Limit (MRL)

The MRL is the measure currently used in the USA to determine safe residue levels in foods. According to Kaneene and Miller (1997), in the USA there is a proposal which recommends the use of toxicologically determined acceptable daily intake (ADI) levels for drugs in place of the MRL. The World Health Organization (WHO) and the Food and Agriculture

Organization (FAO) have set standards for acceptable daily intake (ADI) and maximum residue limits (Abbasi et al., 2007). European Union (EU), USA, Canada, and some other countries have set MRLs, too.

The same MRLs have been adopted by the Tanzania Food Drugs and Cosmetic Act (2003) and the Tanzania Animal and Disease Act (2003) with subsequent laws, regulations and guidelines. But enforcement of these laws is still questionable.

2.8 Different residues levels in different parts of beef

The levels of drug residues differ from one part of the animal to the other. The kidney contains the highest levels followed by liver then muscle contains the least.

The acceptable MRLs for tetracyclines as recommended by the Joint FAO/WHO Expert Committee on Food Additives is 200, 600, and 1200 µg/kg for muscles, liver, and kidney, respectively (Kaneene and Miller, 1997). The same MRLs have been adopted by the Tanzania Food Drugs and Cosmetic Act (2003) and the Tanzania Animal and Disease Act (2003).

2.9 Knowledge and altitude of farmers, businessmen and consumers on drug residues

According to the study conducted in North West Iran, the deficiency of consultation to the veterinary officer and lack of knowledge of washout requirements of drugs were the major causes of non-adherence to drug washout periods (Babapour, 2012). Other causes were ignorance of health risks associated with drugs, extensive farming practice and lack of previous training in animal farming in animal source foods which contributed to non-adherence to washout periods (Kurwijila et al., 2006).

Another source of knowledge to the farmers, businessmen and consumers is the veterinary sector. The veterinarians play an important role in avoiding violative residues (Knust, 2008). The Veterinarians have been implicated to be an important source of information about animal health products and food safety but due to their scarcity, their impact is not easily felt.

Due to profit motives behind, the businessmen/farmers turn the blind eye to the washout periods. This has highly contributed to the drug residues to find their way into the food chain.

In European countries the consumers play the decisive role in influencing the levels of drug residues in beef by their readiness to pay the premium price for beef without traces of drug residues. In spite of the entire legislative body, and all the European institutions associated with its enforcement, meat safety has been described to be a concern to many European consumers (AguIar, 2012).

The study conducted in Ghana revealed that in many developing countries, little is known about food safety in relation to antibiotic residues by consumers (Donkor et al., 2011). This has made the consumers to continue consuming unwholesome products without raising any concern.

CHAPTER THREE

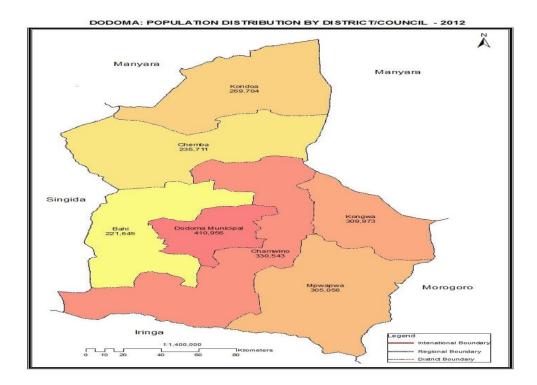
3.0 Methodology

3.1 Study Site and Setting

Dodoma is the capital city of Tanzania. On the western side it is bordered by Bahi district while on the south, east and northern parts it is bordered by Chamwino district. It is within the central zone and it has 4 universities and a parliamentary house, hence high demand of beef. Despite high demand of beef Dodoma has only one ultra-modern abattoir. The abattoir slaughters on average 20 animals a day, a number which was below the consumption capacity. The ante mortem and post mortem were done by the municipal meat inspector.

This study focused on the laboratory quantification of the levels of OTC residues in beef in Dodoma Municipality market by analyzing beef samples from 5 wards of Majengo, Viwandani, Makole, Makulu and Ntyuka. A total of 171 samples were collected. Of these, 128 samples were collected from licensed vending outlets (butchery shops, supermarket and abattoir) and another 43 from unlicensed vending outlets especially the bars where animals slaughtered under the tree were likely to sneak in. The comparison of the quantified levels between the two different vending points was made.

Figure 2. Map of Dodoma Municipality



3.2 Study Design

This was a Cross-sectional study using primary data. The Mixed Method Parallel Convergent design was employed. With this design, the qualitative and qualitative data were collected and analyzed at the same time but separately, after the analysis of both strands the decision was made on the dimensions to compare the results.

3.3 Quantitative Approach

3.3.1 Sampling and sample size consideration

3.3.1.1 Sampling Strategy

The stratified sampling was done for 5 supermarkets and 30 butchery shops to ensure generalization and validity of results. The stratification was done to ensure that samples were taken equally from the selected premises.

3.3.1.2 Sample Size

At CI 95% with prevalence of positive samples being 70% (Bedada and Zewde, 2012), the sample size was calculated as follows:

n=
$$(z/\Delta)^2$$
 p (1-p)
where z=1.96, p=0.7, Δ =0.08
= 0.3x0.7x(1.96/.08)² = 126.

3.3.2 Data collection

The samples were collected from butchery shops, supermarkets and abattoir.

The total of 171 samples were obtained i.e 34 samples from each ward. Each sample weighed 250gm. Three (3) cooler boxes with ice packs were used to collect and store the samples on daily basis.

The samples were collected using the sampling bags with identification marks and stored in the cooler box. The transportation of the samples was done under cold chain from the sampling site to the laboratory.

The data collection form was designed before the search for data took place. The form captured also the origin of the sample for traceability purpose if the need dictates so but in practice this was not possible because of the problem with the animal marketing system in Tanzania.

The quantitative data and qualitative data were collected and analyzed at the same time but separately in both strands. The results from both strands converged for comparison purposes.

Random selection of vending points in 5 wards was done from licensed and unlicensed premises. The sampling was done within 60 days i.e 1st October 2015 to 30th November 2015.

Laboratory results were used for quantitative data. The validity and reliability of these laboratory results were guaranteed by prior calibration and validation of analytical equipment and analytical reagents. The laboratory results also depended on the types of samples submitted in the laboratory for analysis.

3.3.2.1 Sample types

The sample types which were collected from the licensed and unlicensed premises were

Table 1. Types of Beef Samples

Sample	Number of	Number of Samples	Number of Samples	TOTAL
Type	sample per	from 5 wards	from 5 wards	NUMBER OF
	ward	(LICENSED) ¹	(UNLICENSED) ²	SAMPLES
Muscles	27	94	43	137
Liver	4	20	0	20
Kidney	3	13	1	14

¹Officially recognized beef vending outlets like butchery shops and supermarkets; ²Unofficially recognized beef vending outlets like bars and under the tree.

muscles, liver and kidneys as portrayed in Table 1.

3.3.2.2 Laboratory Analysis

3.3.2.2.1 Growth Inhibition Method

The laboratory analysis of 171 samples of beef started with growth inhibition technique using *Bacillus subtilis* whose results are portrayed by Table 4. The growth inhibition zone of the standard OTC was used to compare the inhibition zones in other samples which were inoculated in the *Bacillus subtilis* seeded nutrient agar incubated over night at 37 degrees

centigrade. With this method, the following samples inhibited bacterial growth: 60 muscle samples from licensed premises, 21 muscle samples from unlicensed premise, 14 kidney samples and 18 liver samples. But these results were inconclusive. At this stage, the samples containing antibiotics were established but this could not meet our objective of establishing the samples with OTC residues. The samples were subjected to the next method of high performance liquid chromatography (HPLC).

3.3.2.2.2 High Performance Liquid Chromatography (HPLC)

Sample preparation and extraction

The frozen beef samples were thawed and cut with scissors to remove fat and fascia before being weighed. The Ultra-Turrex T25 tissues homogenizer was used to homogenize muscle, liver and kidney tissues.

The homogenized 5g of muscle, liver or kidney products were mixed with 30ml of extraction solvent (5% trichloroacetic acid with 0.5% disodium ethylenediaminetetraacetate (EDTA) and acetonitrile-water (3:7) and then centrifuged for 25 min at 5300 rpm. The supernatant was filtered through a 0.45 µm filter paper into HPLC vial and injected 20 µl of filtered solution into HPLC for analysis. Due to the fact that OTC has tendency of forming complex with proteins, EDTA 0.5% was added because of its chelating properties to help in separating OTC from the proteins and facilitate extraction.

Hypersil BDC C18 column was used and OTC was separated at 24°C using a mobile phase of methanol-acetonitrile-0.2M oxalic acid (1:1:3.5) at pH=2.0 (with 28% aqueous ammonia) at a flow rate of 0.6 ml/min and wavelength detector set at 360 nm. Oxytetracycline residues were determined by using HPLC (model Shimadzu Class-VP Series, Kyoto, Japan) equipped with SIL-10 autoinjector with sample cooler and LC-10 on-line vacuum degassing solvent delivery unit. Chromatographic control, data collection and processing were carried out by using Shimadzu Class VP data software by aid of computer connected to the detector (Ueno, 1989). The quantities established were compared to the MRL set by CAC and categorized as being within, below or above the MRL.

The validity of the obtained OTC residues in beef consumed in Dodoma municipality catchment area by using HPLC was guaranteed by the prior calibration of the analytical equipments and validation of the method used.

The analytical equipments were calibrated by Tanzania Bureau of Standards (TBS) in November 2015. The calibration is normally done once a year.

The validation of the method was done for 2 consecutive days before running the samples to ascertain the recovery and repeatability, accuracy, precision, specificity/selectivity and linearity of the calibration curve.

The recovery and repeatability of the method were tested by analysing three samples of beef spiked with Standard oxytetracycline at 0.05, 0.25, 0.5, 0.65, 0.95, and 1.20 mg/Kg. The maximum detection limit of 1.20mg/Kg corresponded to 6 times the maximum residue limits (MRLs, 0.2mg/Kg) set by the CAC. The extraction recoveries from the spiked beef samples ranged from 85% to 99%.

The meat sample free of OTC was used a matrix in which standard OTC (0.05, 0.25, 0.5, 0.65, 0.95, and 1.20 mg/Kg) was injected to prepare the calibration curve whereby 20 μ L of each were injected into the HPLC system by the auto sampler injector pump. The peak and retention times were identified and compared to the standard. The results obtained for OTC residues in beef was linear ranging from 0.05 to 1.20mg/Kg.

The assessment of reproducibility and repeatability of the results was done by using 6 samples whereby 5 samples were spiked with working standard OTC at 0.05, 0.5 and 1.2mg/Kg while 1 sample was a reference blank. This exercise was done for 2 consecutive days. The results showed that the reproducibility and repeatability corresponded to the validation methods of TFDA with coefficients of variation of 3.4 to 7.8%.

The limit of detection (LOD) and limit of quantification (LOQ) of the assay method were determined whereby the LOD was at 0.05mg/Kg whereas the LOQ calculated from the spiked samples was 0.06mg/Kg indicating accuracy and precision of the method.

3.3.3 Variables and Measurements

Table 2. Variables

TYPE	VARIABLE	INDICATOR	MEASUREMENT
Dependent Variables	a) Quantity of OTC residues	Levels of residues with respect to Std	μg/g
	a) Number of samples above the MRL	Proportion of samples above MRL	Percentage
Independent Variables	a)Beef type (chuck, striploin, fillet, kidney, liver)	Levels of residues with respect to MRL	μg/g
	b)Irrational/Uncontrolled use of A/B	No traceability of animal and Treatment record	Concentration of A/B
	c)Adherence to drug withdrawal periods	No slaughter before 28 days after treatment	days
	d)Knowledge of Drug withdrawal period and Law enforcement		

3.3.4 Data Analysis

The laboratory data were analysed as follows: Continuous variables (quantities of OTC residues in mg/Kg) slightly failed to be symmetrical, but right skewed. I made an assumption of normal distribution based on the smallness of sample size (171 samples). The test for normality by the Shapiro-Wilk W test (quantile-quantile plot) was done. Since there was no evidence of gross deviation from normal distribution of the continuous variables, the descriptive statistics for continuous variables was hinged on pairwise comparison of the means between the Licensed and Unlicensed premises.

The pairwise comparison of the means was done using Benferron's and Student-Newman Keuls' methods. The generalized linear model was used to select the best predictors of OTC residues in our samples after subjecting the predictor variable to univariate and multivariate analyses. All analyses were done by using Stata software version 13.

3.4 Qualitative Approach

3.4.1 Sampling and sample size consideration

3.4.1.1 Sampling Strategy

The qualitative strand employed the cross-sectional surveys design which is very useful in assessing practices, attitudes, knowledge and beliefs of a population in relation to a particular health related event more precisely the drug residues. Purposive sampling method was used to establish the sample size for businessmen, farmers and law enforcers while convenient sampling method was employed for consumers as indicated in Table 3.

The results from these surveys did not only give an indication of knowledge of people on beef drug residues but also provided a basis for designing appropriate public health measures (e.g., health education campaigns).

The qualitative strand aimed at obtaining information on the knowledge, practice and attitude of farmers, businessmen, law enforcers and consumers on drug washout periods and drug residues.

3.4.1.2 Sample Size

Table 3.Sample size consideration (qualitative)

Interviewee (sample)	Type of Sampling		
Farmers (20 or more till saturation)	Purposive Sampling		
Business men (20 or more till saturation)	Purposive Sampling		
Consumers(10)	Convenient Sampling		
Law enforcers (5 or more till saturation)	Purposive Sampling		

3.4.2 Data Collection

The data collection tool was the Semi-Structured interview guide with open ended questions.

There were 4 interview guides: interview guides for farmers, businessmen, consumers and law enforcers.

To ensure credibility and dependability, the interview guides were pretested to farmers, businessmen, consumers and law enforcers from Madukani ward, a ward different from the 5 wards which were interviewed finally for data collection. Revision of the tool was done. The age of the interviewee were not below eighteen (18) years old.

3.4.3 Data Analysis

The qualitative data were entered into Nvivo matrix first. Then thematic analysis was used to analyze qualitative data.

3.5 Ethical Considerations

The ethical clearance was sought from the University of Zambia Biomedical Research Ethics Committee (UNZABREC) before embarking upon this research.

In Tanzania, the permission was sought from TFDA and the Municipal Council, Livestock Development Department.

The result of the study was likely to have negative impact on the business. That being the case I was buying the samples like any other customer without self-disclosure.

3.6 Dissemination Plan

The obtained information will be disseminated through the University of Zambia accredited journals. The Relevant Authorities like TFDA and Ministry of Livestock Development will receive the findings for further action.

The Tanzania Beef Consumer Association and the Tanzania Meat Board will be notified of the findings so as to create awareness among the consumers of the danger they are exposed to

CHAPTER FOUR

4.0 Quantitative Results

In this study, one hundred seventy one (171) beef samples were collected from five (5) wards within Dodoma municipality catchment area. The concerned wards were Majengo, Viwandani, Makole, Makulu and Ntyuka. The samples were collected from licensed premise and unlicensed premised as portrayed in Table 4. The licensed premises were butchers, supermarkets and abattoirs whereas unlicensed premises for sale of beef were considered to be bars and emergency slaughters (under the tree). The samples were muscles (137 samples), liver (20 samples) and kidneys (14 samples).

The samples were subjected to Growth Inhibition Method by using Nutrient agar seeded with *Bacillus subtilis* to note the extent of inhibition of growth as an indicator of presence of antibiotic residue (Table 4). The samples were finally run by HPLC to quantify the levels of OTC residues (Table 5).

Table 4: Growth Inhibition Method Results (Bacillus subtilis)

	SAMPLE TYPE	SAMPLE SOURCE	NO GROWTH (%)	GROWTH (%)	TOTAL SAMPLES
A.	Muscles	Licensed Premise	60(64%)	34(36%)	94
		Unlicensed Premise	21(49%)	22(51%)	43
В.	Livers	Licensed Premise	18(90%)	2(10%)	20
		Unlicensed Premise	0	0	0
C.	Kidneys	Licensed Premise	14(100%)	0	14
		Unlicensed Premise	0	0	0
	TOTAL				171

Table 4.0 shows the results of the samples which were subjected to Growth Inhibition Technique using nutrient agar seeded with *Bacillus subtilis*. For muscles from licensed premise, there were

60 samples, 21 samples from unlicensed premise, 18 samples of liver and 14 samples of kidneys which inhibited growth but at this stage it was not possible to know the type of antibiotic and quantities till the next method of high performance liquid chromatography (HPLC).

Table 5: OTC Residue Quantification in beef by HPLC

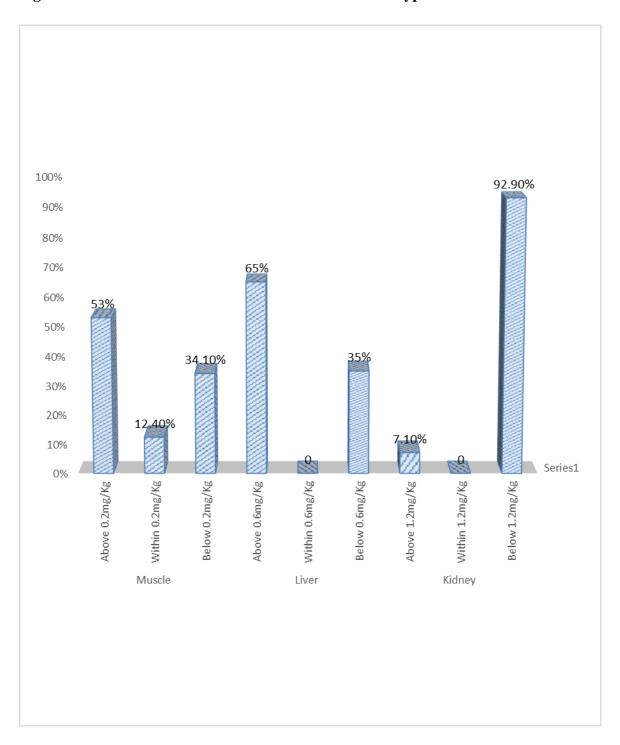
	SAMPLE			TOTAL SAMPLE		
	TYPE		LICENSED PREMISE (%)		UNLICENSED PREMISE(%)	TYPES
A .	Muscle	0.2mg/Kg	i. Below limit (<0.2mg/Kg)	27(57%)	20(43%)	47
			ii. Within limit(0.2mg/Kg)	12(71%)	5(29%)	17
			iii.Above limit (>0.2mg/Kg)	55(75%)	18(25%)	73
B.	Liver	0.6mg/Kg	i. Below limit (<0.6mg/Kg)	7(100%)	0	7
			ii.Above limit (>0.6mg/Kg)	13(100%)	0	13
C.	Kidneys	1.2mg/Kg	i. Below limit (<1.2mg/Kg)	13(100%)	0	13
			ii.Above limit (>1.2mg/Kg)	1(100%)	0	1

¹Maximum Residue Limit as set by Codex Alimentarius Commission according to different tissue types.

Table 5: This table shows the OTC residue results for muscle, liver and kidney samples by HPLC whereby 73 muscle samples, 13 liver samples and 1 kidney sample were above the set residue limit.

The assessment of OTC residue levels in muscles, livers and kidneys from both licensed and unlicensed premises was done by using HPLC. The analysis percentagewise based on tissue type is described by Figure 3.

Figure 3: Assessment of OTC Residue based on Tissue Type



Adjusting for the licensure status and source of the samples, 65% (13/20) of the liver tissues were found to have OTC residue levels above the MRL of 0.6mg/Kg while 35% (7/20) had residues below the limit (Table 6). The kidney tissues were found to have 7.1% (1/14) of the samples above the MRL of 1.2mg/Kg whereas 92.9% (13/14) of the samples were below the MRL.

Table 6: Mean Levels of OTC Residues according to Tissue Type

Number of samples	Mean (SD) ¹	MRL^2
137	0.60(1.19)	0.20
14	0.25(0.43)	0.60
20	1.28(1.03)	1.20
	samples 137 14	samples 137 0.60(1.19) 14 0.25(0.43)

¹ Mean residues levels (Standard Deviation) in mg/Kg; ² Maximum Residue Limit as set by CAC in mg/Kg

Table 6: The mean OTC residues are above the maximum residue limit (MRL) in muscles and liver while below the limit in the kidney. The anomaly of the kidney to have residues below the limit is explained in the subsequent discussion.

Analysis of OTC residues was further done based on tissue type, licensure status of the premise and the source of the samples as portrayed by Table 7, 53% (73/137) were found to have levels of OTC residues above the maximum residue limit, MRL (0.2mg/Kg). Only 34.1% (47/137) of the muscle tissue had residues below the maximum residue limit whereas 12.4% (17/137) had OTC residues within the limit.

When considered regardless of the tissue type, 61 % (104/171) of the edible parts had detectable OTC residues while 51% (87/171) of edible parts had OTC residues above MRL.

Viewed from the source of the sample without taking into account of tissue types and licensure status, the butcheries, supermarkets and slaughterhouses had mean residues of 0.64mg/Kg, 0.77mg/Kg and 0.39mg/Kg respectively (Table 7).

Table 7: OTC residue quantification by Tissue, Licensure and Sample Source

Variable		MRL(MAR) ²
Muscle	0.60(1.19)	0.20(10.72)
Liver	0.25(0.43)	0.60(3.00)
Kidney	1.28(1.03)	1.20(1.67)
Licensed	0.63(0.89)	(15.94)
Unlicensed	0.73(1.70)	(10.72)
Butchery	0.64(0.83)	(5.34)
Supermarket	0.77(1.12)	(5.94)
Slaughterhouse	0.39(0.80)	(2.95)
Bars	0.37(0.38)	(1.35)
Emergency slaughters	2.29(3.64)	(10.72)
	Muscle Liver Kidney Licensed Unlicensed Butchery Supermarket Slaughterhouse Bars	Muscle 0.60(1.19) Liver 0.25(0.43) Kidney 1.28(1.03) Licensed 0.63(0.89) Unlicensed 0.73(1.70) Butchery 0.64(0.83) Supermarket 0.77(1.12) Slaughterhouse 0.39(0.80) Bars 0.37(0.38)

¹ Mean residues levels (Standard Deviation) in mg/Kg; ² Maximum Residue Limit (MRL) as set by CAC in mg/Kg against the Maximum Actual Residues (MAR) as found in the samples in mg/Kg based on tissue, licensure and sample source.

Table 8: Comparison of the Mean OTC Residues by Licensure

Licensure	Mean Residues ¹	SD^2	Maximum ³	95% Confidence
Status				Interval (CI)
Licensed	0.63	0.89	5.94	30 .49
Unlicensed	0.73	1.70	10.72	
¹ Mean residues in mg/Kg; ² Standard Deviation; ³ Maximum residue in mg/Kg				

Table 8: This table shows the mean residues of the samples from the two groups based on licensure status whereby the samples from unlicensed premise had slightly higher OTC residues (0.73mg/Kg) compared to those from licensed premises(0.63). But the comparison of the mean residues from the two groups by Benferron's and Student-Newman Keuls' methods revealed no significant difference between the two groups at 95% Confidence Interval (-.30, .49).

Table 9: Determination of OTC residues

Predictors	Univariate Analysis		Multivariate Analysis	
	P Value	Confidence Interval	P Value	Confidence Interval
Tissue Type	0.05	00 .51	0.02	0.03 0.56
Licensure	0.63	29 .49	0.85	-0.88 0.72
Source	0.42	08 .18	0.39	-0.15 0.37

Table 9: The table shows the predictor variables for OTC residues after subjection to univariate and multivariate analysis whereby the tissue type was found to be a significant predictor variable (p value = 0.028).

CHAPTER FIVE

5.0 Qualitative Results

The qualitative results established the knowledge, concerns, attitudes and practice of farmers, businessmen, consumers and law enforcers from the five wards. This was measured by using the semi structured questionnaires. The semi structured questionnaires were administered to the beef stakeholders. The interviewees were stratified into farmers, businessmen, law enforcers and consumers. There were 10 farmers (two from each ward), 10 businessmen (two from each ward), five law enforcers and five consumers.

5.1 Concerns, Knowledge and Practice on antibiotics

Qualitatively, the interviewed farmers, businessmen, consumers and law enforcers had different levels of knowledge on the use of antibiotics, washout periods, antibiotic residues and negative impacts of consuming beef containing antibiotic residues.

Of the interviewed farmers, majority admitted to be giving antibiotics to animals to promote weight gain and protect their animals against diseases. But they claimed not to be aware of the washout periods and antibiotic residues. At Ntyuka ward, one of the farmer emphasized:

... I believe once I have given my animals the antibiotics they will gain weight quickly and will not get diseases easily because they are protected by the medicine. When trekking my animals to the auction I give them enough dose of oxytetracycline with red cap. (Respondent number 3).

Oxytetracycline with a *red cap* was a long acting antibiotic (20%) which was implicated to be in use whenever the farmer speaks of *red cap*. This implied that most of the animals from the farmers who keep animals for sale for slaughter were injected with mega doses of OTC to the animals and were introducing drug residues in the food chain. This might have been one of the reasons of finding one beef sample with the highest level of OTC residues up to 10.71mg/Kg.

Majority of the business were aware of the rational use of antibiotics but they used antibiotics under profit motives to ensure the animals arrived at the auction while protected from transit fever. The law enforcement officers were quite knowledgeable about rational use of antibiotics and lamented on the abuse of antibiotics but most of the consumers were not aware on the rational use of antibiotics.

5.2 Public health implications of drug residues

Minority of the consumers were ignorant of the public implications of the drug residues. They believed whatever comes from the butchery shops is safe for their consumption. But only one consumer admitted to have heard that drug residues could cause health problems to the consumers but did not know what exact health problems. The consumer admitted:

I once heard that drug residues in beef, milk and eggs can cause health problems but as long as I am not aware of the exact health problems I continue to consume beef as usual. (Respondent 6).

On the awareness of illegal source of beef, majority of consumers were aware of illegal source of beef though they did not admit to be sourcing beef from the illegal premises. The consumers were bitter at the practice of slaughtering dead animals for human consumption and considered this practice to be inhuman. One of the consumers lamented on the practice:

Such animals should not be slaughtered for human consumption. They should be slaughtered for dogs or should be buried. It is dangerous. But people are not aware of the danger of eating such like an animal. There is need of educating them. (Respondent number 2).

5.3 Enforcement of laws on drugs

All of the law enforcers who were contacted were quite knowledgeable on drug washout periods and drug residues though they admitted the adherence to the washout periods by cattle owners to be the problem. The drugs were found to be sold without any control. The owners treat the animals on their own without leaving any treatment record whatsoever. One of the law enforcer remarked:

Not only in my council but also in other parts of Tanzania veterinary drugs are sold without any control. I mean without prescription drugs are accessed by lay persons. This is underrating the profession by continuing with illegal possession of veterinary drugs. At time these drugs are sold at market place in open places without adherence to temperature and light storage conditions of these drugs. Sometimes the farmers do not use the advice from law enforcers but instead help each other to decide on the type of medicine and dosage to administer once the animal falls sick. (Respondent number 1).

5.4 Adherence to washout periods

On the awareness of drug washout periods and drug residues, majority of the businessmen were aware though they could not tell the exact washout periods. But being prompted by profit motive behind, the observation of these periods was not possible. One of the businessmen at Majengo ward confessed:

... at times we inject the animals with drugs to prevent disease and gain weight quickly or buy animals for slaughter without knowledge whether the animals were treated or not and take them for slaughter to get profit quickly. (Respondent number 7).

The few farmers were aware of the wash out requirements but none of them observed the wash out requirement for milk or beef products.

Knowledge was found to be the major driver of the attitude of administering antibiotics to the animals intended for slaughter with the purpose of quick weight gain and protection against transit fever.

CHAPTER SIX

6.0 Discussion

This study established 51% of edible tissues to have residues which are unacceptably above the MRLs set by CAC as adapted by TFDC Act (2003) which are 0.2mg/Kg, 0.6mg/Kg and 1.2mg/Kg for muscle, liver and kidney tissues respectively. This shows to what extent people have been exposed to unacceptably high levels of drug residues.

These results were similar to the results of the study which was conducted in Ethiopia where out of the total 384 samples analyzed for tetracycline residues, 71.3% had detectable oxytetracycline levels. About 48% of the edible tissues had oxytetracycline levels above the recommended maximum limits (Bedada and Zewde, 2012). These results are comparatively higher in relation to Canada where there is zero tolerance of drug residues in food products. As pointed out (Salisbury et al., 1990) on an investigation on OTC residues in pork in Canada conducted on October 1987 up to March 1988, 2% of the products were found to have traces of residues which were below the MRL. But because of the law of zero tolerance all of the products were condemned.

With this study, OTC residue levels in beef consumed in Dodoma Municipality catchment area was made. Of all the samples, the muscle tissue had higher levels of OTC residues. But when considering the mean OTC residues, the muscle tissue and the liver tissue had mean residues above the MRLs (Table 7). The mean residues levels in muscle tissue was 0.60mg/Kg ranging from 0 to 10.72mg/Kg, whereas the liver tissue had mean residue of 1.28mg/Kg ranging from 0.19mg/Kg to 3.0mg/Kg. The kidney had low mean residue of 0.26mg/Kg ranging from 0.01mg/Kg to 1.67mg/Kg. There was one muscle sample with unexpectedly high levels of OTC residues up to 10.72mg/Kg. The sample was taken from unlicensed premise (emergency slaughter) at Makulu ward. This is explained by confession of the interviewed farmers who treated the animals but when death was eminent they resorted to slaughtering of the animal. Given the pharmacokinetics of OTC, the sick animal has high concentration of OTC due to alteration of haemodynamics especially drop in blood pressure, cardiac output, glomerular filtration, renal blood flow and decreased hepatic drug metabolism (Nazawi, 2014), all of which bottleneck to lower clearance rate and longer half-life. The kidney tissues which were expected to have comparatively higher levels of OTC residues were found to have low levels of OTC due to the similar reason of pharmacokinetics of the drug. Another reason behind high levels of residues in that sample is the injection sites through which the drugs are administered into the animals which are neck and buttocks muscles. The sample from the injection site of an animal which has been injected recently is expected to have high levels of residues compared to muscle away from the injection site. Given the current animal marketing system even at the slaughterhouse, the traceability of the slaughtered animal was difficult to underscore with the aim of feedback to the culprits for corrective action.

Based on the licensure status of the premise, the mean residues of the samples from the two sources, the samples from unlicensed premise had slightly higher OTC residues (0.73mg/Kg) compared to those from licensed premises (0.63) as shown in Table 8. But the comparison of the residues among the two groups by Benferron's and Student-Newman Keuls' methods revealed no significant difference between the two groups at 95% Confidence Interval (-.30, .49).

From the licensed premises the residues ranged from 0 to 5.94mg/Kg whereas from unlicensed premises the residues ranged from 0 to 10.72mg/Kg. The results are indicating how people who are sourcing beef from licensed and unlicensed premises are all exposed to consumption of drug residues.

The samples from the bars and emergency slaughters (slaughter under the tree) had OTC residues of 0.37mg/Kg and 2.30mg/Kg respectively. From the above findings, the entire source of beef is not safe in terms of drug residues because the samples from the entire source are all above the MRLs. The samples from the emergency slaughter had the highest mean of OTC residues. Remarkably, the emergency slaughter source was the one with the sample with highest levels of OTC residues of 10.72mg/Kg which is unacceptably high.

The generalized linear model was run through the variables to detect the best OTC residue predictor variable between tissue type, licensure and source. The univariate analysis of the variables revealed the tissue type to be significant predictor at p value 0.05 while licensure and source were not significant at p values 0.63 and 0.42 respectively.

When considering the predictor variables of OTC residues holistically (tissue type, licensure and source) by using the generalized linear model (Table 9), it was established that the tissue type was a significant predictor at p value = 0.028 at 95% CI (0.03 0.56) at multivariate analysis.

Backed up by the survey on the knowledge, concerns and practice of farmers, businessmen, consumers and law enforcers, the comparison was made between quantitative results and qualitative results. The credibility and dependability of the qualitative results was guaranteed by

prior testing of the research tool to the above mentioned groups of respondents with subsequent modification of the tool.

As portrayed by the result, all of the farmers admitted to be giving antibiotics to animals to promote weight gain and protect their animals against diseases. But they claimed not to be aware of the washout periods and antibiotic residues as emphasized by respondent number 3 at Ntyuka village.

Oxytetracycline with a *red cap* is a long acting antibiotic (20%) which is implicated to be in use whenever the farmer speaks of *red cap*. This implies that most of the animals from the farmers who keep animals for sale for slaughter are injecting mega doses of OTC to the animals and selling them without considering the washout periods. Indeed this poses the real public health threat to the consumers. This is similar to the findings of the study conducted in North West Iran where it was established that the deficiency of consultation to the veterinary officer and lack of knowledge of washout requirements of drugs were the major causes of non-adherence to drug washout periods (Babapour, 2012).

Most of businessmen were aware of the washout periods and drug residues though they could not tell the exact washout periods of OTC. Despite that knowledge, being prompted by profit motive behind, they confessed not to be ready to wait for the washout period or risk the animal to death before slaughter.

This reveals the fact that some businessmen out of profit motive they are ready to introduce into the market something which they know it is prohibited by the law. This way the drug residues have been introduced into the food chain. Apart from profit motive, other causes were ignorance of health risks associated with drugs, extensive farming practice and lack of previous training in animal farming in animal source foods which contributed to non-adherence to washout periods (Kurwijila et al., 2006).

Viewed from the consumer side, majority of the consumers were not aware of the drug residues but all of them were aware of illegal source of beef though they did not admit to be sourcing beef from the illegal premises. The consumers were bitter at the practice of slaughtering dead animals for human consumption and considered this practice to be inhuman. This malpractice was lamented upon by respondent number 2.

It is quite evident that consumers were not informed on drug residues and the public health implication of consuming beef containing drug residues. This calls the need for educating the consumers, farmers and businessmen on the public health concern (toxic or anaphylactic reactions and drug resistance) of consuming beef with drug residues. Educational approach to some extent can conscientize them to be watchdog over one another. From the consumers testimony, it was established that there was illegal slaughter of the animals. Some of the beef was coming from unlicensed premise. The study conducted in Ghana revealed that in many developing countries, little is known about food safety in relation to antibiotic residues by consumers(Donkor et al., 2011). This has made the consumers to continue consuming unwholesome products without raising any concern.

All of the law enforcers who were contacted were quite knowledgeable on drug washout periods and drug residues though they admitted the adherence to the washout periods by cattle owners to be the problem. The drugs were found to be sold without any control. The owners treat the animals on their own without leaving any treatment record whatsoever.

The above remark shows that veterinary drugs throughout the country are sold and administered without any control by law enforcers. As pointed out by the respondent, the farmers help each other to treat their animals because of easy access to antibiotics without any demand of prescription. This malpractice of treating animals leaves no record whatsoever. This results into treated animals to be allowed for slaughter without observing the washout periods. Hence, the drug residues find their way into the food chain. Given the current practice, even law enforcers are not in position to remedy the situation unless they use the police force to make several raids at the market places to seize the street vended veterinary medicine. This is in line with a similar study conducted in Ethiopia (Bedada and Zewde, 2012) which portrayed the lack of control of veterinary drugs and lack of rational use of drugs which were attributed to failure of enforcement of the already stipulated laws governing veterinary drugs. At times these antibiotics are found sold in the market without prescription or along the road by informal vendors.

Such unacceptably high levels of OTC residues have public health implications of drug resistance (Nonga et al., 2009), toxic and anaphylactic reaction (Horie et al., 2003), (Karimuribo, 2005), teratogenic effects to pregnant mothers (Czeizel et al., 1998), teeth disclouration and hypoplasic development of the teeth (Czeizel and Bauer, 2000) when such residues are consumed by infant.

Limitations and Strength of the study

Few cattle owners living in the outskirt of the municipality practice fattening program to the animals. Traceability of the sample back to the owner of the animal for corrective action was a problem due to the animal marketing system in Tanzania. It was also difficulty to get equal samples from the licensed and unlicensed premises. Due to the nature of illegal business, it was difficulty to get enough samples from unlicensed premises especially samples from emergency slaughters or slaughter under the tree. However the study unveiled the truth about residues and knowledge of beef value chain actors. Due to Dodoma Capital city being centrally located coupled with big population, most of the animals from the other parts of Tanzania are brought into Dodoma, so the results portray the true picture of the other parts of Tanzania. This study used both qualitative and quantitative strands complementing each other to gather enough information.

Conclusion

The major aims of this cross sectional study were to establish and assess the OTC residue levels of beef consumed in Dodoma Municipality catchment area in Tanzania. The total number of 171 beef samples were analyzed by HPLC with calibrated equipments and validated methods to guarantee validity and reliability of the results. These results will work as the bench of advice to the law enforcers. The results of these samples from licensed and unlicensed premises indicated 51% (87/171) of the samples to contain residues above the MRLs of 0.2mg/Kg, 0.6mg/Kg and 1.2mg/Kg for muscles, liver and kidney respectively. This is unacceptably high and needs immediate corrective action by law enforcers and other stakeholders in the beef sector.

These results confirm that the food chain in Dodoma is contaminated by OTC residues at the high levels. This has in turn posed a real public health threat to the consumers who are likely to succumb to toxic/anaphylactic reactions and drug resistance (Senyuva et al., 2000).

If such high levels of OTC residues are consumed by the pregnant mother during the second month of pregnancy there are higher chances of teratogenic risks to the foetus (Czeizel et al., 1998) and can cause discolouration of primary and secondary teeth as well as hypoplasic development of teeth when consumed by infants or pregnant mothers during the last 2 trimesters (Czeizel and Bauer, 2000).

Recommendations

It is highly recommended for the Ministry of Livestock Development and Fisheries to intervene in screening the animals at the market and enhance traceability of the animals by improving the animal marketing system in Tanzania. The law enforcers should actively seek, find and take action against the practice of slaughter under the tree. It is also recommended to have effective control of veterinary drugs such that some of the drugs like antibiotics should be bought from licensed shops with prescription. The businessmen dealing with fattening of the animals for slaughter should be identified and monitored on their daily practice to ensure the washout periods are observed.

Last but not least, education should be availed to the farmers, consumers and businessmen on the public health threat posed by consumption of beef containing drug residues. With education, joint efforts of these groups can bring tangible results to the fight against drug residues.

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8.0 Appendices:

STUDY TITLE: ASSESSMENT OF OXYTETRACYCLINE RESIDUE LEVELS IN BEEF CONSUMED IN DODOMA MUNICIPALITY CATCHMENT AREA, TANZANIA.

8.1 Information Sheet

THE UNIVERSITY OF ZAMBIA SCHOOL OF MEDICINE DEPARTMENT OF PUBLIC HEALTH



PRINCIPAL INVESTIGATOR: Engelbert Bilashoboka IRB NO....

INTRODUCTION

My name is Engelbert Bilashoboka. I am a student with the University of Zambia School of Medicine Department of Public health. I am conducting this study in partial fulfillment of completion of my Masters of Science program in Epidemiology. I am the investigator on this study. The study is being done in abattoir, supermarkets and beef vending points by using TFDA laboratory in Tanzania

PURPOSE OF THE RESEARCH

Antibiotics are among the most widely used veterinary drugs in food-producing animals for therapeutic purposes and as dietary supplement without observing the washout period.

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The Oxytetracycline drug residues pose a real public health hazard to the consumer due to this exposure and can lead to various toxic effects including, anaphytlactic reactions and development of drug-resistant strains of bacteria. According to Codex Alimentarius Commission of the FAO and WHO, the current standard maximum residue level (MRL) in beef muscles is 0.2 microgram per gram of beef attainable after 28 days from the last treatment with oxytetracycline (CAC/MRL, 2012). However, due to the profit motive, farmers and business men do not follow the FAO and WHO guidelines. Once the animal falls ill and is treated with oxytetracycline; upon slight recovery the animal is sent for slaughter without taking into consideration of the drug washout period.

The obtained results will work as a benchmark for advice to the law enforcers.

This will also create consumer awareness depending on the preference of the parts of beef. Being aware of the impending danger the combined efforts to fight the problem can be possible.

WHY ARE YOU BEING ASKED TO PARTICIPATE?

I am asking you to participate in this study because of your role and the knowledge you have on the drug residues in beef consumed in Dodoma Municipality. Your knowledge will provide valuable insights to the study in the fight against drug residues in beef.

PROCEDURES

If you agree to participate in this study, you will be asked questions on your knowledge on drug washout period, drug residues and human health risks of consuming beef containing residues. Your suggestions on how to fight against the drug residues will be documented.

During the study you will be asked on the factors that have aggravated the drug residue problem. You will also be asked on your suggestions and experience on implementation of the fight against drug residues.

RISKS/DISCOMFORTS TO PARTICIPANT OR OTHERS

If you agree to take part in the study, the information required in this study is your personal knowledge on drug washout period, drug residues and human health risks of residues. There may be some discomfort in disclosing the knowledge you have. Your honest responses to these questions are very important for future studies and also in informing policy makers and stake

holders on challenges and success of food safety. Your responses will be treated with

confidentiality. If you experience any discomforts and would like to discontinue, you are free to

do so.

BENEFITS

If you agree to participate in this study there are no direct benefits to you but you will be

contributing to the understanding of drug washout period, drug residues and human health risks of

consuming beef containing drug residues. The information collected will be useful to inform

policy makers and stakeholders on drug residues in beef consumed in Dodoma Municipality.

CONFIDENTIALITY

Data collected from you regarding your role and the knowledge you have on the drug residues in

beef consumed in Dodoma Municipality will be kept strictly confidential and can only be shared

with your permission and anything you say will be kept completely confidential during the

interviews. Your name will not be used to identify you and the information collected. I would

greatly appreciate your honest response during the interview.

VOLUNTARY PARTICIPATION

Participation in this study is out of your free will without any coercion. You are free not to answer

any question deemed personal or otherwise. However, you are at liberty to withdraw from the

study at any moment without any penalty.

WHO DO I CALL IF I HAVE QUESTIONS OR PROBLEMS?

If you have any questions or concerns about the research please contact:

1. Contact, Principal Investigator

Engelbert Bilashoboka: +260974307049/+255766729416

University of Zambia School of Medicine

Department of Public health

Box 50110 Lusaka Zambia; E-mail: bilashoboka.engelbert@gmail.com

2. Chair, University of Zambia Biomedical Research Committee (UNZABREC)

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Ridgeway Campus Box 50110 Lusaka Zambia

Telephone +260-1-256067 Telegram UNZA, LUSAKA, Fax+260-1-250753, Telex UNZALU ZA 44370, Email: unzarec@unza.zm

3. Dr Boyd Mudenda (Supervisor)

University of Zambia School of Medicine

Department of Public health

Box 50110 Lusaka Zambia; Email: boyd@boydmudenda.com

4. Ms Nosiku Munyinda (Co-Supervisor)

University of Zambia School of Medicine

Department of Public health

Box 50110 Lusaka Zambia; Email: ssiku@hotmail.com

5. Professor Dominic Mukama Kambarage

Sokoine University of Agriculture

Faculty of Veterinary Medicine

Box 3000 Morogoro Tanzania. Email: dkambarage@yahoo.co.uk

8.2 Consent Form

WHAT DOES YOUR SIGNATURE AND THUMBPRINT ON THIS CONSENT MEAN?

Your signature (or thumbprint/mark) on this form means:

- You have been informed about the research purpose, procedures, possible benefits and risks.
- You have been given the chance to ask questions before you sign.
- You have voluntarily agreed to be in this study

rint name of Adult Participant	Signature of Adult Participant	Date
Print name of Person Obtaining Si	ignature of Person Obtaining Conse	ent Date consent

Ask the participant to mark a "left thumb impression" in this box if the participant is unable to provide a signature above.

8.0 Appendices

MADA: TATHIMINI JUU YA KIWANGO CHA OXYTETRACYCLINE ILIYOKO KATIKA NYAMA INAYOLIWA KATIKA MANISPAA YA DODOMA, TANZANIA.

8.1 Information Sheet

THE UNIVERSITY OF ZAMBIA SCHOOL OF MEDICINE DEPARTMENT OF PUBLIC HEALTH



MTAFITI MKUU: Engelbert Bilashoboka

IRB NO....

UTANGULIZI

Naitwa Engelbert Bilashoboka. Ni mwanafunzi katika Chuo Kikuu cha Zambia katika Kitivo cha Tiba idara ya Afya ya Jamii.Ninafnya utafiti katika kutimiza matakwa ya kupata Shahada ya Uzamili katika Afya ya Jamii. Mimi ni mtafiti katika utafiti huu. Utafiti utafanyika katika machinjio na soko la nyama kwa kutumia maabara ya TFA nchini Tanzania.

LENGO LA UTAFITI:

Vijiuavijasumu(antibiotic) ni mojawapo kati ya dawa za mifugo zinazotumika kwa ajili ya tiba na kinga kwa kuongezwa katika vyakula vya mifugo bila kujali muda wa kusitisha uchinjaji wa mnyama aliyetumia dawa hizo.

Mabaki ya dawa ya oxytetracycline ni hatarishi kwa afya ya mlaji kwani husababisha mzio na kujengeka kwa vimelea sugu. Maagizo ya Shirika la Chakula na Shirika la Afya Duniani kuwa mabaki ya oxytetracycline yanayoruhusiwa katika nyama si zaidi ya 0.2microgramu kwa kila gramu ya nyama. Pamoja na maagizo hayo, kwa msukumo wa kutengeneza faida, wakulima pamoja na wafanyabiashara wamekuwa wakiruhusu wanyama kuchinjwa kabla ya muda huo.

Taarifa zitakazopatikana kutokana na utafiti huu zitasaidia wafuatiliaji wa utekelezaji wa sheria kukaza uzi pale walipolegeza.

Pia kwa ujulisha umma juu ya hatari iliyopo katika kula nyama yenye mabaki ya dawa, mapambano ya pamoja dhidi ya tatizo hili yatafanikiwa.

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KWA NINI UNAOMBWA KUSHIRIKI KATIKA UTAFITI HUU?

Ninakuomba ushiriki katika utafiti huu kwa kuwa nafasi yako na uelewa wako juu ya mabaki ya dawa katika nyama ni muhimu katika utafiti huu. Uelewa wako utatoa msaada mkubwa katika mapambano dhidi ya mabaki ya dawa katika nyama.

TARATIBU

Kama umekubali kushiriki katika utafiti huu, utaulizwa maswali juu ya uelewa wako juu ya muda wa kuzuia uchinjaji wa mnyama aliyetibiwa, mabaki ya dawa katika nyama na athari za mabaki ya dawa kwa afya ya binadamu. Mapendekezo yako juu ya mapambano dhidi ya tatizo hili yataheshimiwa na kuandikwa.

Katika utafiti huu utaulizwa juu ya mambo yanayochangia tatizo hili. Utaulizwa pia juu ya mapendekezo yako katika mapambano dhidi ya tatizo hili.

ATHARI

Kama umekubali kushiriki katika utafiti huu, taarifa utazozitoa ni uelewa wako juu ya muda wa mnyama kuruhusiwa kuchinjwa, mabaki ya dawa mwilini na athari za mabaki ya dawa kwa afya ya binadamu. Huenda usijisikie vizuri wakati wa kueleza ukweli wa kinachoendelea. Lakini tambua kuwa kufunua kwako ukweli juu ya suala kutasaidia hata watunga sera na wadau wa sekta ya chakula kuja na mkakati wa kuhakikisha usalama wa vyakula. Majibu yako yatatunzwa kwa usiri wa hali ya juu. Pindi utakapojisikia vibaya wakati wa mahojiano na ukataka kuachana na utafiti huu, una uhuru wote wa kufanya hivyo.

FAIDA

Ukikubali kushiriki katika utafiti huu hakuna faida za moja kwa moja utakazopata ipokuwa utakuwa umefanikwa kuchangia katika uelewa wa muda wa zuio kabla kuchinja, mabaki ya dawa na athari kwa afya ya binadamu itokanayo na nyama yenye mabaki ya dawa. Taarifa zitakazopatikana zitasaidia kuwahabarisha watunzi wa sera na wadau wa nyama juu ya mabaki ya dawa katika nyama inayoliwa Dodoma.

USIRI

Taarifa zitakazopatikana juu ya nafasi yako na uelewa wako juu ya mabaki ya dawa katika nyama

inayoliwa katika Manispaa ya Dodoma zitatunzwa kwa usiri wa hali ya juu na kila kitu

utakachokisema wakati wa mahojiano kitaendelea kubaki siri. Kamwe jina lako halitatumika

katika taarifa zilizopatikana kutoka kwako.

UTASHI

Ushiriki wako katika utafiti huu ni wa hiari bila shuruti. Uko huru kuamua kutojibu swali ambalo

hulipendi na kuendelea na swali jingine. Unaweza ukaamua kujiondoa katika utafiti muda wowote

bila kulipishwa gharama yoyote ile.

NAWEZA KUWASILIANA NA NANI NIKIWA NA SWALI AU SHIDA?

Ukiwa na swali au shida inayohusiana na utafiti huu unaweza kuwasiliana na:

1. Mawasiliano, Mtafiti Mkuu

Engelbert Bilashoboka: +260974307049/+255766729416

University of Zambia School of Medicine

Department of Public health

Box 50110 Lusaka Zambia; E-mail: bilashoboka.engelbert@gmail.com

2. Mwenyekiti, University of Zambia Biomedical Research Committee (UNZABREC)

Ridgeway Campus Box 50110 Lusaka Zambia

Telephone +260-1-256067 Telegram UNZA, LUSAKA, Fax+260-1-250753, Telex

UNZALU ZA 44370, Email: unzarec@unza.zm

3. Dr Boyd Mudenda (Supervisor)

University of Zambia School of Medicine

Department of Public health

Box 50110 Lusaka Zambia; Email: boyd@boydmudenda.com

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4. Ms Nosiku Munyinda (Co-Supervisor)

University of Zambia School of Medicine

Department of Public health

Box 50110 Lusaka Zambia; Email: ssiku@hotmail.com

5. Professor Dominic Mukama Kambarage

Sokoine University of Agriculture

Faculty of Veterinary Medicine

Box 3000 Morogoro Tanzania

Email: <u>dkambarage@yahoo.co.uk</u> Report writing, Premise Registration and

Licensing

8.2 Consent Form

NINI MAANA YA SAHIHI YAKO NA DOLE GUMBA

Sahihi yako au alama ya dole gumba katika fomu hii inamaanisha yafuatayo:

- Umeelezwa lengo, taratibu, faida na athari za utafiti huu
- Umepewa nafasi ya kuuliza maswali kabla ya kusaini
- Kwa hiari yako umekubali kushiriki katika utafiti huu

Jina la Mshiriki	Saini ya Mshiriki	Tarehe

Muombe mshiriki aweke alama ya dole gumba kama hawezi kuweka saini.

8.3 Questionnaire

QUESTIONNAIRE TO DETERMINE KNOWLEDGE OF FARMERS, BUSINESSMEN AND CONSUMERS ON DRUG RESIDUES AND DRUG WASHOUT PERIODS.

Questi	onnaire number Date:
Initials	of Researcher
Type o	f facility
	DEMOGRAPHIC CHARACTERISTICS OF INTERVIEWEE
	Category of interviewee 1.Farmer 2. Businessman 3. Consumer
	Age Sex:Male/Female
	A. PRACTICE&KNOWLEDGE OF BUSINESS MEN
Q101	What is your education level?
	Elimu yako ni kiwango gani?
Q102	There has been a practice of adding antibiotic in animal feed to promote growth.
	What are your views about this practice?
	Kumekuwepo tabia ya uongezaji wa vijiuavijasumu katika vyakula vya mifugo ili
	kuboresha ukuaji. Naomba kujua mawazo yako kuhusu hili.
Q103	There has been a practice of adding antibiotic in animal drinking water to prevent
	disease. What are your views about this practice?
	Kumekuwepo tabia ya uongezaji wa vijiuavijasumu katika maji ya mifugo ili
	kuboresha ukuaji. Naomba kujua mawazo yako kuhusu hili.
Q104	It is a common practice to add antibiotic in animal feed and drinking water to
	promote growth. What do you think about this practice?
	Kumekuwepo tabia ya uongezaji wa vijiuavijasumu katika vyakula na maji ya
	mifugo ili kuboresha ukuaji. Naomba kujua mawazo yako kuhusu hili.

Q105	What do you think if supervision of administration of these antibiotics could be
	done by a veterinarian?
	Naomba kupata mawazo yako kama usimamizi wa matumizi ya dawa hizi
	ungefanywa na maafisa mifugo.
Q106	Briefly tell when your animal gets sick what do you do?
	Kwa ufupi nieleze mnyama wako akiugua huwa unafanya nini?
Q107	Kindly tell me how many different bottles or sachets of antibiotics do you have in
	your yard for treating animals when they are sick?
	Tafadhali niambie ni aina ngapi za dawa za chupa au vipaketi ulizo nazo katika
	shamba lako kwako ajili ya kutibu mifugo endapo itaugua?
Q108	After treating the animal with antibiotic, where do some of the small amounts of
	remnants of drug go?
	Unajua baada ya matibabu ya mnyama dawa ndani ya mwili wa mnyama huelekea
	wapi?
Q109	How many days after the last treatment can the animal be allowed to go for
	slaughter?
	Unajua ni baada ya siku ngapi tangu mnyama atibiwe anaruhusiwa kuchinjwa kwa
	ajili ya matumizi ya binadamu?
Q110	It is expensive to keep the animals. What do you do if you treat the animal and it
	fails to recover?
	Ni gharama kutunza mnyama. Ukitibu mnyama akashindikana kupona unafanya
	nini?
	B. PRACTICE&KNOWLEDGE OF FARMERS
Q201	What is your education level?
\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\	Elimu yako ni kiwango gani?
	Lumu yako ni kiwango gani:

Q202	There has been a practice of adding antibiotic in animal feed to promote growth.
	What are your views about this practice?
	Kumekuwepo tabia ya uongezaji wa vijiuavijasumu katika vyakula vya mifugo ili
	kuboresha ukuaji. Naomba kujua mawazo yako kuhusu hili.
Q203	There has been a practice of adding antibiotic in animal drinking water to prevent
	disease. What are your views about this practice?
	Kumekuwepo tabia ya uongezaji wa vijiuavijasumu katika maji ya mifugo ili
	kuboresha ukuaji. Naomba kujua mawazo yako kuhusu hili.
Q204	It is a common practice to add antibiotic in animal feed and drinking water to
	promote growth. What do you think about this practice?
	Kumekuwepo tabia ya uongezaji wa vijiuavijasumu katika vyakula na maji ya
	mifugo ili kuboresha ukuaji. Naomba kujua mawazo yako kuhusu hili.
Q205	What do you think if supervision of administration of these antibiotics could be
	done by a veterinarian?
	Naomba kupata mawazo yako kama usimamizi wa matumizi ya dawa hizi
	ungefanywa na maafisa mifugo.
Q206	Briefly tell when your animal gets sick what do you do?
	Kwa ufupi nieleze mnyama wako akiugua huwa unafanya nini?
Q207	Kindly tell me how many different bottles or sachets of antibiotics do you have in
	your yard for treating animals when they are sick?
	Tafadhali niambie ni aina ngapi za dawa za chupa au vipaketi ulizo nazo katika
	shamba lako kwako ajili ya kutibu mifugo endapo itaugua?
0200	After treating the enimal with antihiotic where do some of the small amounts of
Q208	After treating the animal with antibiotic, where do some of the small amounts of
	remnants of drug go?

	Unajua baada ya matibabu ya mnyama dawa ndani ya mwili wa mnyama huelekea
	wapi?
Q209	How many days after the last treatment can the animal be allowed to go for
	slaughter?
	Unajua ni baada ya siku ngapi tangu mnyama atibiwe anaruhusiwa kuchinjwa kwa
	ajili ya matumizi ya binadamu?
Q210	It is expensive to keep the animals. What do you do if you treat the animal and it
	fails to recover?
	Ni gharama kutunza mnyama. Ukitibu mnyama akashindikana kupona unafanya
	nini?
	C. PRACTICE AND KNOWLEDGE OF CONSUMERS
Q301	What is your education level?
	Elimu yako ni kiwango gani?
Q302	What is the source of beef consumed by your household?
	Ukitaka kitoweo cha nyama kwa matumizi ya nyumbani huwa unapata wapi?
Q303	What are unofficial sources of beef consumed to your household?
	Unajua maeneo yasiyo rasmi ambako kitoweo cha nyama kinapatikana?
Q304	What are your views on the animals which fail to recover from a disease but are
	slaughtered for sale?
	Nini mawazo yako kuhusu wanyama wanaotibiwa bila mafanikio na mwishowe
	kuchinjwa kwa ajili ya matumizi ya binadamu?
Q305	Would you tell me where do remnants of a drug after treatment go?
	Unajua baada ya matibabu ya mnyama dawa ndani ya mwili wa mnyama huelekea
	wapi?
Q306	What are health problems caused by consumption of beef containing remnants of

	drugs?
	Unajua ni madhara gani yanaweza kumpata mtu baada ya kula nyama yenye
	mabaki ya dawa?
	D. PRACTICE AND KNOWLEDGE OF LAW ENFORCERS
Q301	What is your role as an inspector?
	Wewe kama mkaguzi kazi yako ni nini?
Q302	How often do you visit farmers/livestock keepers for advice?
	Ni kwa kiasi gani unawatembelea wakulima/wafugaji kwa ajili ya ushauri?
Q303	How are veterinary drugs sold/dispensed from the outlets?
	Unajua dawa za mifugo huuzwa kwa mtindo gani kutoka katika maduka ya dawa za
	mifugo?
Q304	What are your views on the animals which fail to recover from a disease but are
	slaughtered for sale?
	Nini mawazo yako kuhusu wanyama wanaotibiwa bila mafanikio na mwishowe
	kuchinjwa kwa ajili ya matumizi ya binadamu?
Q305	Would you tell me where do remnants of a drug after treatment go?
	Unajua baada ya matibabu ya mnyama dawa ndani ya mwili wa mnyama huelekea
0.00	wapi?
Q306	What are health problems caused by consumption of beef containing remnants of
	drugs?
	Unajua ni madhara gani yanaweza kumpata mtu baada ya kula nyama yenye
	mabaki ya dawa?

8	4	Ethical	Clearance	and	Permission	Letters
v.	т.	Luncai	Cicarance	anu	T CI IIIISSIUII	Lucius

THE UNITED REPUBLIC OF TANZANIA DODOMA MUNICIPAL COUNCIL

(All correspondence to be addressed to Municipal Director)

DODOMA REGION

Tel.: 2354817/2321550 Fax: 026 - 2321550



Office of Municipal Director P.O. Box 1249

Dodoma

E-mail: dodomamunicipality@yahoo.co.uk

In reply please quote:

Ref.No.HMD/E.10/4/15

Date: 20th December, 2015

MRS. C. JACOBS
UNIVERSITY OF ZAMBIA
SCHOOL OF MEDICINE
DEPARTMENT OF PUBLIC HEALTH
P.O BOX 50110
LUSAKA, ZAMBIA

REF. PERMIT FOR STUDENT TO CONDUCT RESEARCH AT DODOMA MUNICIPAL

Reference is made to your letter of 18/06/2015 with no reference number

I am very glad to let you know that the permit is granted for your student Mr. **ENGELBERT BILASHOBOKA MBEKENGA** to collect data / information on various issues related to his study on "Assessment of Oxytetracycline Residue Levels in Beef Consumed in Dodoma Municipality Catchment Area, Tanzania". The student can be free to visit all areas of his needs in various Wards within the Municipality as per his choice.

The students should respect all laws and regulations especially the "The National Security Act - 1970" while conducting his research in all the Wards that he will be working as well as where he will be guided by the Ward's authority. A Swahili version letter to be presented to anyone concerned in the Wards and other areas is attached.

Wish you all the best

Innocent O Kessy MANISP A DODGMA For: Municipal Director

DODOMA.

Copy to:

Municipal Livestock & Fisheries Development Officer

JAMHURI YA MUUNGANO WA TANZANIA HALMASHAURI YA MANISPAA DODOMA

(Barua zote zipelekwe kwa Mkurugenzi wa Manispaa)

	MKOA WA DODOMA Tel.: 2354817/2321550 Fax: 2321550 Ofisi ya Mkurugenzi wa Manispaa, S.L.P.1249 Dodoma E-mail: dodomamunicipality@yahob.co.uk
4	Unapojibu tafadhali taja:
	Mkuu wa Chuo, Chuo cha, Kilkwu CHA ZAMBIA
•	YAH: KUFANYA MAZOEZI KWA VITENDO/UTAFITI
	Tafadhali rejea somo hapo juu. Napenda kukujulisha kuwa ombi lako la mwanachuo/wanachuo wako ENGEURIERI B.: mr. EUCHGA limekubalika.
	Mtajwa afike Ofisi ya ZINAZOHWSIKA MANISARA M Dubama kwa ajili ya kuanza mazoezi-hayo/kufanya utafiti huo kwa tarehe husika. Aidha Manispaa haitakuwa na fungu lolote la kumlipa/kuwalipa mwanachuo/wanachuo huyo/hao.
	Nakutakia kazi njema.
	Kny: MKURUGENZI WA MANISPAA, DODOMA
	Nakala: Mkuu wa Idara/Kitengo MIJUGO NA MAENDILIO YA UVUVI Mwanachuo Ndugu: ENGELBEKT B MBEICON SA Mtendaji wa Kata/Kijiji/Mtaa ya YOYOTE I TAKAYOHUSUCA

TANZANIA FOOD AND DRUGS AUTHORITY

E-mail: Telephone: info@tfda.or.tz

+255 22 2450512, 2450751 +255 22 2452108

Fax No. Website: +255 22 2452108 +255 22 2450793 www.tfda.or.tz

All letters should be addressed to the Director General In reply please quote Our Ref No: TFDA

Nelson Mandela Road Mabibo External, P.O. Box 77150, DAR ES SALAAM, TANZANIA.

Ref. No.:

TFDA/PF.255/25

August 19, 2015

The University of Zambia, Biomedical Research Committee (UNZABREC), P.O. Box 50110, LUSAKA – ZAMBIA.

RE: PERMISSION LETTER TO USE TFDA LABORATORY

- 1. Reference is made to the above subject matter.
- 2. Dr. Engelbert Bilashoboka is an employee of TFDA and he is conducting research entitled "Assessment of Oxytetracycline (OTC) residue levels in beef consumed in Dodoma region of Tanzania. He will use TFDA Laboratory facilities to carry out OTC residue analysis in beef.
- 3. In view of the above TFDA grants permission to Dr. Bilashoboka to conduct his research at its laboratory.

Thank you for your cooperation.

Yours sincerely,

M.A. Fimbo

AG. DIRECTOR GENERAL

MAF/cnu/rm/jm

MISSION

To protect and promote public health by ensuring quality, safety and effectiveness of food, drugs, cosmetics and medical devices.



THE UNIVERSITY OF ZAMBIA

BIOMEDICAL RESEARCH ETHICS COMMITTEE

Telephone: 260-1-256067 Telegrams: UNZA, LUSAKA Telex: UNZALU ZA 44370 Fax: + 260-1-250753

E-mail: unzarec@unza.zm

Assurance No. FWA00000338 IRB00001131 of IORG0000774

25th November, 2015.

Our Ref: 011-06-15.

Dr. Engelbert Bilashoboka, TFDA Central Zone, P.O Box 1253, Dodoma, Tanzania.

Dear Dr. Bilashoboka,

RE: RESUBMITTED RESEARCH PROPOSAL: "ASSESSMENT OF OXYTETRACYCLINE RESIDUE LEVELS IN BEEF CONSUMED IN DODOMA MUNICIPALITY CATCHMENT AREA, TANZANIA" (REF. No. 011-06-15)

The above-mentioned research proposal was presented to the Biomedical Research Ethics Committee on 30^{th} October, 2015. The proposal is approved.

CONDITIONS:

- This approval is based strictly on your submitted proposal. Should there be need for you to modify or change the study design or methodology, you will need to seek clearance from the Research Ethics Committee.
- If you have need for further clarification please consult this office. Please note that it is mandatory that you
 submit a detailed progress report of your study to this Committee every six months and a final copy of your
 report at the end of the study.
- Any serious adverse events must be reported at once to this Committee.
- Please note that when your approval expires you may need to request for renewal. The request should be
 accompanied by a Progress Report (Progress Report Forms can be obtained from the Secretariat).
- Ensure that a final copy of the results is submitted to this Committee.

Yours sincerely,

Dr. S. H. Nzala VICE-CHAIRPERSON

Date of approval:

25th November, 2015.

Date of expiry: 24th November, 2016.

Ridgeway Campus

P.O. Box 50110

Lusaka, Zambia