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P.O. Box 236, Tororo, Uganda
Gen: +256 - 45 444 8838
Fax: +256 - 45 4436517
Email: info@adm.busitema.ac.ug

www.busitema.ac.ug

**ANTIBIOTIC RESISTANCE OF BACTERIA ISOLATED FROM MASTITIC MILK
IN BUGANGAIZI EAST COUNTY, KIBAALE DISTRICT**

BY



TURYASINGURA VICTOR

BU/UP/2013/185

Victorturyasingura@gmail.com

**A DISSERTATION SUBMITTED TO THE FACULTY OF AGRICULTURE AND ANIMAL
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DEGREE OF BACHELOR OF ANIMAL PRODUCTION AND MANAGEMENT OF
BUSITEMA UNIVERSITY**

MAY, 2016

DECLARATION

I **TURYASINGURA VICTOR**, declare that this dissertation is my original work and has not been submitted to any institution for any academic award.

Signature 

Date 16/08/2016

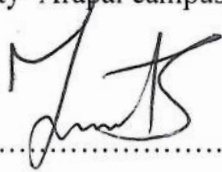
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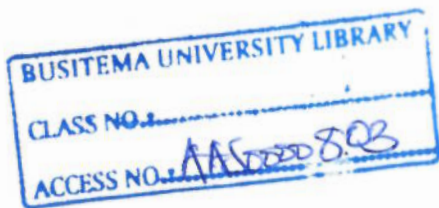
Mr. MBOGUA JOSEPH

Department of Animal Production and Management

Faculty of Agriculture and Animal Sciences

Busitema University- Arapai campus

Signature  Date 06/10/2016



DEDICATION

I dedicate this work to my parents Mr. Twebaze Felix & Mrs. Tugumisirize Rosemary, TOFTA Educational Trust, Br Deodat Aganyira, Mr. John Kirkwood relatives and friends who endeavored to sacrifice the little they had to enable me study up to this level.

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LIST OF ABBRIVIATIONS ACRONYMS AND SYMBOLS

% - Percentage

Col - Coliform

CONS - Coagulase Negative Staphylococci

Cory - Corynebacteria

CPS - Coagulase Positive Staphylococci

DNA - Deoxyribonucleic Acid

E.coli- Escherichia coli

e.g - For Example

e.tc - etcetra

MIC - Minimum Inhibitory Concentration

NCCLS - National Committee for Clinical Laboratory Standards

°C - Degrees Celcius

PBPS - Penicilin Binding Protein

rRNA - Ribosomal Ribonucleic Acid

Spp - species

Staph - Staphylococcus

Strep -- Streptococcius

β - Beta

μm -Micrometre

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ABSTRACT

Drug resistant mastitis is on the rise in Uganda especially in the dairy sector and is becoming a subject of great concern. This study was undertaken to establish the antibiotic resistance of mastitis bacteria pathogen isolated from selected farms in Bugangaizi East Kibaale District

Across sectional study was carried out in the lactating local and exotic breeds in the study area, culture and sensitivity tests were done in the laboratory following the standard laboratory methods using antibiotics which were found to be commonly used in the area.

The study revealed that staphylococcus (64.7 %) were the most frequently isolated organisms, followed by coliform (25 %) , streptococcus (7.3 %) and the least common was corynebacteria (3 %). Penicillin antibiotics were the most common resisted (70.2 %), followed by Ampicillin (45.6%), Cloxacilin (31.6%), Tetracycline (22.8%) and Chloramphenicol (5.3%), Erythromycin (3.5%) and Gentamicin (1.8) were the least resisted.

The resistance patterns within selected farms showed the same distribution with penicillin having the highest resistance in almost all the farms except one. Gentamicin, Erythromycin and chloramphenicol had least resistance in all farms.

The results of the ANOVA test gave a p-value of 0.014 this means that prior exposure of animals to antibiotics had an influence on antibiotic resistance since the null hypothesis of No difference between exposed and Non-exposed was rejected.

In conclusion, this study found out that prior exposure of animals to antibiotics had an influence on antibiotic resistance, poor milking hygiene ,poor environmental hygiene accompanied by poor treatment regime were the most predisposing factors of animals to mastitis.

This study also found out that staphylococci were the major causes of mastitis in Bugangaizi East Kibaale District and they are highly resistant to penicillin.

CHAPTER ONE

1.0 INTRODUCTION

1.1 Back ground

Milk production is an important component of Uganda's live stock sector which is carried out by the traditional sector, specialized dairy farms and Governmental parastatal farms (MAAIF 1984, 2011). However, milk production in the traditional sector is still poor and there are many constraints that limit maximal production. Bovine mastitis has been described as one of the major constraints and most costly disease in the dairy production that limit enhanced milk production, efficiency in many dairy herds despite the amount of knowledge available on the subject. (Hogeveen, 2005). The prevalence of mastitis disease in Uganda has been ranging from 60-70% and has not changed over the years due to poor management and poor hygiene (Byarugaba *et al.*, 1998). Most estimates have shown a 30% reduction in production per cow per lactation, making the disease one of the most costly and serious problem affecting the dairy industry worldwide. (Hunderra *et al.*, 2005)

Bovine mastitis is known to cause a great deal of economic losses such as reduction in milk yield due to disturbances in the udder tissue in clinical and sub-clinical mastitis, discarding of large quantities of milk which cannot be consumed either due to lowered quality or drug residue in it, culling of previously high milk producing cows with chronic mastitis. Such cows may pose danger of transmission of resistant bacteria to other animals through close contact, overcrowding or poor milking hygiene, the latter being mainly by the use of same piece of cloth for drying the udders of all milking cows.

There are also high treatment costs due to repeated treatments, a trophy of quarter or more of the udder quarters due to chronic mastitis. Resistant genes can be passed from one group of organisms to another. e.g from less pathogenic organisms to more pathogenic ones thus causing disease leading to serious losses (Sabath, 1982). Transmission of resistant bacteria from animals to humans through animal products pose health hazards in humans either by transmitting resistance or may cause disease out breaks if the resistant strains get established in humans (Williams, 1982).

Antibiotics are commonly used in food producing and animals for treatment, growth promotion (Sabath, 1982) or prophylactically for prevention of occurrence of disease such as dry cow therapy for prevention of occurrence of clinical mastitis (Tranert, 1983). Although Antibiotics are restricted drugs can only be

REFERENCES

- Aarestrup F.M., N.E Jensen N.E., (1998). Development of penicillin resistance among staphylococcus aureus isolated from bovine mastitis in Denmark and other countries .Microb Drug resistant 4-247-256.
- Anakolo S, Gathoni A, Tura, Galgalz, Milch and Mwangi (2004). "prevalence of bovine mastitis among small holder dairy herds in Kenya" Israel Journal of veterinary medicine, 59(1-2).
- Anonymous, (2002) Milk and dairy beef residue prevention protocol.
- Byarugaba D.K (1993). Across sectional study of mastitis on dairy farms in North Kyadondo county of Kampala district. BVM special research project, Makerere University ,Kampala.
- Byarugaba D.K ,(2005) . Antimicrobial resistance and its containment in developing countries,pg 617-646.
- Byarugaba D.K, Khaitisa M.L., Opuda Asibo J.,(1998)"Bovine mastitis in North Kyadondo county of Kampala District". Ugandan veterinary journal 4:139-146.
- Chambers H.F., (2003). Solving staphylococcal resistance of beta-lactams. Trends microbiology 11-145-148.
- Coles, E.H., (1998). Microbiology Veterinary clinical pathology by coles, E.H (4th .edn) .saundersco.philadelphia, USA pg . 359-367.
- Dricuru M., (1995). Antibiotic resistance patterns of mastitis bacterial pathogens from isolated farms served by the Ambulatory clinic of the faculty of veterinary medicine Makerere university, Kampala -pg 38.
- Faye and Alary (2001). Livestock to 2020.The next food revolution .Publ .IFPRI, Washington,U.S A.
- Gitta ,S.P., (2003). Antibiotic resistance of selected enteric bacteria isolates from Gorillas, domesticated ruminants and humans in and around Bwindi impenetrable forest national park. Project report, July 2003. Kampala: Makerere University page 16-23.
- Hogeveen H.(2005). Mastitis in Dairy production: current knowledge and future solutions, wageningen. Academic publishers, Netherlands.

- Hunderra.(2005).” Dairy cattle mastitis in and around sebeta, Ethiopia” intern JAPPI Res vet med,3(4).
- Linda I. Likofsky, John W., Barlow, Carlos Santisteban, Ynte H. Schukken (2003). A comparison of antimicrobial susceptibility patterns for staphylococcus aureus in organic and convectional dairy herds. Microbial drug resistance Vol.9,supplement 1. Marry annLiebert, Inc.
- Livemore D. can better prescription turn the tide of resistance ? Nat. Rev. microbial(2003),36;11-23. Craven ,et al.,(1986) .
- Livemore D.M (2003). Bacterial resistance : origins, epidemiology and impact. Clinical infectious diseases 36-511-523.
- Massida O., MontanariMontanari M.P, Mingola M.(1996). Borderline methicillin-susceptible staphylococcudaureus strains have more in common than susceptibility to penicillinase-resistant penicillin. Antimicrobial Agents chemotherapy.40-2769-2774.
- Merck, (1991).Chemotherapeutics .the Merck vet. Manual.(7th .Edn). pg 1414-1419, 1420-1449. Merck and co., inc .Rahway, NJ., USA.
- Ministry of Agriculture ,Animal Industries and Fisheries (1992). White paper on government Dairy sector policy .confidentialRepor, Kampala;pg 18.
- Ministry of agriculture ,Animal Industries and Fisheries(1984). Livestock development marketing study in Uganda, Final report. Winrock International, Morrilton, Arkansas 22110:ppg 61-74.
- Ministry of Agriculture Animal Industry and Fisheries (1998). The animal production sector .annual Report, Entebe, Uganda:pg.30-35, 50-51.
- Nakavuma J., Byarugaba , D.K., Kitombo, F.X. and Musisi, L.N,(1994). Microbiological Diagnosis and Drug resistance patterns of infectious causes of mastitis in cattle Uganda J.ofAgr. Sci.2:1.pg.22-28.
- Odongo ,M.O and Ambani, A.I.A,(1989). Microganisms isolated from bovine milk samples .bulletin of Animal Health production Africa.37:195-196.
- Okello –Uma.(1974). A study of the etiology and means of control of bovine mastitis in exotic and indigenous dairy cattle in Uganda.PHD. Thesis, MakerereUniversity ,Kampala.

- Rege ,J.E.O., Mutwiri , G.K. and Mulei, C.M.,(1990). Antibiotic Resistance patterns of bacteria implicated in bovine mastitis and some canine infections, bulletin of Animal Health and production Africa.37:4, 319-326.
- Sabath , L.D., (1982). Emergency of bacterial resistance to antibiotics occurring during therapy. Academic press ,New York and London.pg 17-25.
- Smithcor J.F, Catcott E.J. (1974). Mastitis progress in cattle and sheep practices. Infectious , mastitis, reproduction, American veterinary publication Inc,USAp.429-509.
- Susan A.(1997). Mastitis in large animals . The merck veterinary manual , 8th edition, merck and co., Inc. whitehouse USA :1009-1010.
- The merck vet. Manual.(7thEdn). Pg. 686-691. Merck and co., Inc. Rahway, USA. Martin, S.W, Meek, A.H and willeberg.p., (1988). Veterinary epidemiology,(2ndedn) . Iowa state University press. AMES. Ch. 2pg. 22-47.
- Tranter ,W.P.,(1983). Herd epidemiological studies. Bovine mastitis .in :Campell, R.S.F ., Copeman, D.B;Goddard; M.E.; Johnson, S.J. and Tranter, W.P.A. A course manual in veterinary Epidemiology.Edited by Campell, R.S.F. Australian Universities' International Developmentbprogram, canbera, Australia (AUIP).60-66.
- Williams , JD.,(1982). Epidemiological Trends in Antibiotic Resistance and concerns for the future .academic press, New York and London. Pg .35-47.
- Williamson G. and payne W.J.A (1984). An introduction to Animal Husbandry in the Tropics 1st edition Longman , London, New York USA: pg 62-365.
- Wise j.M (1999). A comparison of Antibiotic susceptibility patterns for staphylococcus aureus in Organic and convetional Dairy Herds. J.Danish veterinary Laboratory, corpenhagen, Denmark.Pg .20