OCCURRENCE, DISTRIBUTION, SEROTYPES AND ANTIMICROBIAL RESISTANCE AMONG SALMONELLA ISOLATED FROM CATTLE AND ENVIRONMENTAL SAMPLES IN VHEMBE DISTRICT, SOUTH AFRICA

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Submitted in partial fulfilment of the academic requirements for the degree of

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College of Agriculture and Environmental sciences

Department of Agriculture and Animal Health

University of South Africa

Supervisor: Dr. E. MADOROBA

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Dedication

This dissertation is devoted to my lovely wife Madeleine Lubikamba Kapeta for your unconditional love that helps me to stay motivated and made me look forward to the realization of my dissertation.

To my beloved children: Jeriel Lumbala Kapeta and Asriel Ndemba Kapeta this is the inspiration for your future education accomplishment.

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Sincerely,

Daniel Kapeta

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Acronyms and abbreviations

AMR	Antimicrobial resistance
ARC	Agricultural Research Council
ATCC	American Type Culture Collection
BGA	Brilliant Green agar
BTA	Blood tryptose agar
Вр	Base pair(s)
BPW	Buffered peptone water
CDC	Centers for Disease Control
CLSI	Clinical and Laboratory Standards Institute
Cm	Centimeter
Diam	Diameter
DNA	Deoxyribonucleic acid
DT	Definitive phage type
EFSA	European Food Safety Authority
I	Intermediate
ISO	International Organization for Standardization
Kb	Kilo base
ml	Milliliter
mm	Millimeter
MAR	multiple antibiotic resistances
MDR	Multidrug resistance
MHA	Mueller-Hinton agar
NARMS	National Antimicrobial Resistance Monitoring System
NCCLS	National Committee for Clinical Laboratory Standards
OD	Optical Density
OVR	Ondersterpoort Veterinary Research
PCR	Polymerase Chain Reaction
R	Resistant

Rpm	Revolutions per minute
RMRDT	Red Meat Research and Development Trust
RVS	Rappaport–Vassiliadis soy broth
S	Susceptible
ST	Sequence type
U	Unity
UTIs	Urinary Tract Infections
TAE	Tris-Acetate-EDTA
WHO	World Health Organization
XLD	Xylose Lysine Desoxycholate agar
hð	Microgram
μΙ	Micro liter
°C	Degrees Celsius
%	Percentage
≤	Less than or equal to
≥	Greater than or equal to

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Declaration

The study has not been submitted in any form to another university and it represents the original work by the author. Information and ideas from other sources have been referenced and cited accordingly.

Student: Daniel Kapeta Djabintu

Date..... Day of 2017

Signature of the candidate.....

ABSTRACT

Background: *Salmonella* species is the etiologic agent of salmonellosis, which is a zoonotic infection that is characterized by diarrhea and systemic infection. Contaminated foods are usually the vehicles of *Salmonella* transmission along the food supply chain. Asymptomatic food production animals and effluents also contribute to contamination of meat. Antimicrobials have contributed significantly to treatment of salmonellosis. However, uncontrolled antimicrobial use is among the causes of antibiotic resistance, which results in treatment failure.

Aim and Objectives: The aim of this research study was to determine the extent of *Salmonella* spp contamination during the cattle slaughtering process in South African rural abattoirs (n = 23), water and cattle feaces. In addition, the aim was to determine antimicrobial resistance profiles of the *Salmonella* spp isolates. The specific objectives were: i) to establish the occurrence and distribution of *Salmonella* spp on cattle carcasses, hides, and intestinal contents and environmental samples using classical microbiology and molecular techniques; ii) to determine the *Salmonella* serovars using serotyping; and iii) to determine antimicrobial resistance patterns and multidrug resistance among the *Salmonella* isolates using the Kirby-Bauer disc diffusion method.

Materials and Methods: Classical microbiology techniques were used to analyze cattle faeces (n = 400), hides (n = 67), intestinal contents (n = 62), carcass sponges (n = 100), and water from the abattoirs (n = 75) for the presence of *Salmonella* spp. Further confirmation of the *Salmonella* isolates was done using Polymerase Chain Reaction whereby the *invA* gene was targeted. A total of 92 *Salmonella* spp isolates were recuperated. The 92 *Salmonella* were serotyped as described in the White-Kauffmann-Le Minor scheme. The 92 *Salmonella* spp isolates were further subjected to antimicrobial susceptibility examination towards the following antimicrobials: ampicillin (10µg), cefotaxine (30µg), kanamycin (30µg), oxytetracycline (30µg), and enrofloxacin (5µg) by using the Kirby-Bauer disk diffusion procedure.

Results and Discussion: All the isolates carried the *invA* genes. The average *Salmonella* spp occurrence on carcasses, hides, and intestinal contents was 35.37% (n = 81). Eleven of the faecal samples (2.75%) tested positive for *Salmonella* spp.

The *Salmonella* serovar that occurred more frequently was S. Enteritidis. Different serovars that were recognized on carcasses were not automatically found on the hides and intestinal contents. The incompatible frequency of the different *Salmonella* serovars on carcasses, intestinal contents and hides means that in addition to carriage on hides and in intestinal contents, new external causes that did not form part of this study also play a vital role concerning carcass contamination. Most *Salmonella* spp (n = 66; 71.7%) isolates were resistant to a minimum of one antimicrobial with main resistance detected towards oxytetracycline (51.90%). This emphasizes on the call for wise antimicrobial use at some stage in animal production and strict sanitation for the duration of slaughtering.

Conclusion and Recommendation: Briefly, cattle slaughtered in South African rural abattoirs harboured different types of *Salmonella* serovars that were resistant to antimicrobials, which could be a public health risk and danger. The outcome should support policymakers with determining the effectiveness of existing sanitary measures during cattle slaughtering in rural abattoirs, which is vital from socio-economic, public health, and epidemiological perspectives.

Key words: Salmonella, antimicrobial susceptibility and resistance, carcass contamination, polymerase chain reaction (PCR), rural abattoirs.

Publications arising from this thesis

- Evelyn Madoroba, Daniel Kapeta, Awoke K. Gelaw. 2016. Salmonella contamination, serovars and antimicrobial resistance profiles of cattle slaughtered in South Africa. Onderstepoort Journal of Veterinary Research; Vol 83, No 1 (2016), 8 pages. Doi: 10.4102/ojvr.v83i1.1109 (Refer to Appendix).
- Madoroba, E., Gelaw, A.K., Ramagoma, F., Kapeta, D. and Matloa, N. 2014. Zoonotic foodborne pathogens in rural cattle of Vhembe district, South Africa. Popular article. Red Meat/Rooivles, August Issue, 78-81.