

FINAL YEAR PROJECT REPORT

IN VITRO ANTIBACTERIAL EFFICACY OF *KIGELIA AFRICANA* EXTRACT ON PATHOGENIC *ESCHERICHIA COLI* ISOLATED FROM POULTRY IN SOROTI CITY

By

ALINOMUGASHA EMMANUEL

BU/UP/2021/2369

SUPERVISOR: MR ONGOM ISAAC

A RESEARCH DISSERTATION SUBMITTED TO THE FACULTY OF
AGRICULTURE AND ANIMAL SCIENCES IN PARTIAL FULFILLMENT OF
REQUIREMENTS FOR THE AWARD OF BACHELOR OF ANIMAL
PRODUCTION AND MANAGEMENT OF BUSITEMA UNIVERSITY

OCTOBER 2024

DEDICATION

I dedicate this work to JEHOVAH ALMIGHTY who has brought me this far and to those who supported me spiritually, academically, morally and financially. I will not list you all but JEHOVAH ALMIGHTY Reward you abundantly.

DECLARATION

DECLARATION	
I ALINOMUGASHA EMMANUEL declare that I wrote this final year research report and have never submitted it to any other institution of learning	
and have never submitted it to any vents assessed	
NAME: ALINOMUGASHA EMMANUEL	
REGISTRATION NUMBER: BU/UP/2021/2369	
SIGNITURE Emproy	
SIGNITURE CONTRACTOR	
DATE 19th October 2014	
п	
"	

APPROVAL

		-
	APPROVAL	
	This work was done by ALINOMUGASHA EMMANUEL under the supervision of MR	
	ONGOM ISAAC	
	MR ONGOM ISAAC	
	Research supervisor	
	Department of Animal Production and Management	
	Faculty of Agriculture and Animal Sciences – Arapai Campus	
	Busitema University	
	PO BOX, 236, Tororo, Uganda	
	DATE: 21/001/ 2024	
	SIGNITURE: WMORRIEL	
0	Nitel A18 m	

ACKNOWLEDGEMENT

I acknowledge the **Most High God** for the wisdom he has given to make me to complete the proposal successfully. Much thanks goes to my supervisor **Mr. Ongom Isaac** for the technical advice he gave to me. Lastly, I acknowledge my parents for the support they are giving to me.

TABLE OF CONTENTS

Contents

DEDICATION	I
DECLARATION	ІІ
APPROVAL	III
ACKNOWLEDGEMENT	IV
TABLE OF CONTENTS	V
LIST OF ABBREVIATIONS	VIII
LIST OF TABLES AND GRAPHS	IX
ABSTRACT	X
CHAPTER ONE	1
BACKGROUND OF THE STUDY	1
1.1 Introduction	1
1.2 Problem statement	2
1.3.1 General objective	2
1.3.2 Specific objectives	2
1.4 Research questions	2
1.5 Justification	2
1.6 significance	3
1.7 Scope	3
CHAPTER TWO	4
LITERATURE REVIEW	4
2.1Introduction	4
2.2.1 Distribution of kigelia africana	4
2.2.2 Diseases treated by Kigelia africana	5
2.2.3 Ethnopharmaclogical importance of kigelia africana	5

	2.2.4 Antibacterial activity of kigelia africana extract	5
	2.3 History of oxytetracycline	5
	2.3.1Mode of action of Oxytetracycline	6
Cŀ	HAPTER THREE	7
M	ATERIALS AND METHODS	7
	3.1 Study area	7
	3.2 Research Approach	7
	3.3 Sampling design	7
	3.5 Experimental design	7
	3.5.1 Identifying Kigelia africana tree	7
	3.5.2 Collecting of leaves	7
	3.5.3 Cleaning and washing of the Kigelia africana leaves	7
	3.5.4 Drying of the plant	8
	3.5.5 Grinding of the leaves	8
	3.5.6 Soxhlet and Water Bath Extraction and Mass Determination of K. africana	
	Extract	8
	3.5.7 Reconstituting the k. Africana extract	8
	3.6.0 Laboratory Preparation of MacConkey and MHA Agars	8
	3.7.0 Procurement and Culturing of E. coli from Colibacillosis-Affected Poultry	9
	3.7.1 Source of the bacteria	9
	3.7.2 Culturing of e.coli bacteria	9
	3.8 Preparing discs for use in the kirby-bauer disk diffusion method	9
	3.9.0 Serial dilutions for OTC and K. africana	9
	3.9.1Making serial dilutions for OTC	9
	3.9.2 Making serial dilutions of k. Africana extract	10
	3.10 Kirby-Bauer disc diffusion test method of K.africana	10
	3.11 Kirby-Bauer disc diffusion test method of Oxytetracycline	10

3.12 Observational design	11
3.13 Statistical design	11
3.14 Data presentation	11
3.15 Ethical consideration and clearance	11
3.16 Environmental considerations	11
3.17 Limitations of the study	12
CHAPTER FOUR	13
RESULTS AND DISCUSSION	13
4.2 Discussion Of Results	20
CHAPTER FIVE	22
CONCLUSION AND RECOMMENDATIONS	22
5.1 Conclusion	22
5.2 Recommendation	22
APPENDIX 1	I
LIST OF REFERENCES	I
ADDENDIY 2	V

LIST OF ABBREVIATIONS

BUAC - Busitema University Arapai Campus

MIC - Minimum Inhibitory Concentration

MBC - Minimum Bactericidal Concentration

FAAS - Faculty of Agriculture and Animal Sciences

OTC - Oxytetracycline

E.coli – Escherichia coli

K. africana – Kigelia africana

g- grams

mg – milligrams

UV – Ultraviolet

LIST OF TABLES AND GRAPHS

Table 4-1 Showing the in vitro antibacterial efficacy of K.africana on E.coli
harvested from poultrypage 13
Graph 4 -1 Showing in vitro antibacterial efficacy of K.africana on E.coli harvested
from poultrypage 14
Table 4-2 Showing in vitro antibacterial efficacy of OTC on <i>E.coli</i> harvested from
poultrypage 15
Graph 4-2 Showing in vitro antibacterial efficacy of OTC on <i>E.coli</i> harvested from
poultrypage 16
Table 4-3 Showing the comparative in vitro efficacy of both K. africana and OTC on
E.coli harvested from poultrypage 17
Graph 4-3 Showing the comparative in vitro efficacy of both K. africana and OTC or
E.coli harvested from poultrypage 18
Table 4-4 S showing the paired t test values analysis on the comparison of the in vitro
antibacterial activity of K. africana and OTC on E.coli harvested from
poultrypage 19

ABSTRACT

Kigelia africana, commonly referred to as the sausage tree or African sausage tree, is a deciduous tree native to tropical Africa. It belongs to the Bignoniaceae family and is renowned for its large, sausage-shaped fruits.. It is also native to the Soroti City, and the locals call it **edodoi**.. Escherichia coli infections in poultry present a significant public health concern, resulting in heightened morbidity and mortality rates. This is primarily due to the rapid transmission of the bacteria through contaminated feed, water, and inadequate sanitation conditions.. While farmers in Soroti recognised the potential of medicinal plants for treating livestock diseases, most research has primarily focused on E. coli infections in humans. This has resulted in a notable gap in understanding the antibacterial efficacy of Kigelia africana against E. coli in poultry.

An in vitro study was conducted on this plant (*K. africana*) to check for its in vitro antibacterial efficacy on pathogenic E. coli harvested from poultry in Soroti City. Only the healthy, disease-free leaves were picked, air-dried in the shade, and ground using an electric grinder.

The method used for extraction was Soxhlet extraction, and then the mixture with extract was transferred to a water bath to remove methanol. The extract was then reconstituted with 0.2% DMSO, and antimicrobial screening was conducted using the Kirby-Bauer disc diffusion method. All the experiments were conducted in the laboratory of Busitema University, Faculty of Agriculture and Animal Sciences.

K. africana showed a considerable zone of inhibition of 16.6667 mm on the pathogenic E. coli, meaning that it had strong antibacterial properties against the bacteria. Oxytetracycline, a known commercial antibiotic on the market, was used in the study as a comparative drug, and it was effective as expected, with the maximum zone of inhibition measured was 26.67 mm against the pathogenic Escherichia coli.

In conclusion, Kigelia africana leaf extracts possess a range of antibacterial properties that effectively treat colibacillosis in poultry. Furthermore, additional research is needed to assess the efficacy of the fruit, roots, and bark against E. coli derived from poultry.

CHAPTER ONE

BACKGROUND OF THE STUDY

1.1 Introduction

Kigelia africana commonly known as sausage tree or African sausage tree is a deciduous tree native to tropical africa. It belongs to the Bignoniaceae family and is characterised by its large, sausage-shaped fruits.(Nabatanzi et al., 2020b). The tree can attain a height of up to 20 metres and possesses a broad, spreading crown. Its leaves are compound and sizeable, consisting of multiple leaflets. The flowers, which are pollinated by birds and bats, are conspicuous and exhibit a reddish-orange colour.(Henry, 2014). The fruits of K. africana are notable for their large sausage shaped like appearance, measuring up to 60 cm in length and weighing as much as 10 kg. When unripe, they are green and turn to brown or black when ripe, (Nabatanzi et al., 2020a).

The antibacterial activity of *K africana* has been extensively studied, and multiple studies have highlighted its potential in this area. Extracts from various parts of the plant, including the leaves, fruit, and bark, have been shown to possess antibacterial effects against a wide range of organisms. For instance, ethanol extracts of *K.africana* leaves have exhibited maximum antibacterial activity against *Escherichia. coli*, while aqueous extracts have shown moderate activity against *S. aureus* and *P. aeruginosa*.

Moreover, the traditional use of roasted seeds, bark, and fruit extracts of *K. africana* in the treatment of bacterial infections further supports its potential antibacterial properties. The presence of specific compounds, such as alkaloids, glycosides, terpenoids, flavonoids, tannins, reducing sugars, saponins anthocyanosides in *K africana* extracts has also been associated with their antibacterial activity.

These findings emphasize the potential of *K. africana* as a source of antibacterial agents and justify further investigation into its antibacterial properties for potential therapeutic applications, (P. K. Chauhan, Rahul Thory, 2019). Therefore, this study aimed at investigating the antibacterial effect of *K. africana* extract on escherichia coli bacteria isolated from poultry in Soroti city, Uganda.

APPENDIX 1

LIST OF REFERENCES

- A.B. Katende, A. B. and B. T. (2000). USEFUL TREES AND SHRUBS FOR UGANDA. In S. Editing: Bo Tengnas Natural Resource Management Consultant ltd. S-310 38 Simlangsdalen & N.-K. Copy-editing, design and typesetting by: Caroline Agola P. O. Box 21582 (Eds.), Regional Land Management Unit (RELMA), Swedish International Development Cooperation Agency (Sida). Regional Land Management Unit, RELMA/Sida, ICRAF House, Gigiri P. O. Box 63403, Nairobi, Kenya. http://books.google.co.jp/books?id=sZYfAQAAIAAJ
- Allen, M. E. (2005). *MacConkey Agar Plates*. 1–2. http://www.asmscience.org/content/education/imagegallery/image.2850
- Gabriel, O. A., & Olubunmi, A. (2009). Comprehensive scientific demystification of Kigelia africana: A review. *African Journal of Pure and Applied Chemistry*, 3(9), 158–164.
- Giovanardi, D., Campagnari, E., Sperati Ruffoni, L., Pesente, P., Ortali, G., & Furlattini, V. (2005). Avian pathogenic Escherichia coli transmission from broiler breeders to their progeny in an integrated poultry production chain. *Avian Pathology*, *34*(4), 313–318. https://doi.org/10.1080/03079450500179046
- Henry, D. M. (2014). Training Manual for R2.A1, R2.A2 and R2.A3 by Busitema University Arapai Campus. In *Busitema University* (first edit, Vol. 1).
- Horne, J. H., Woolley, H. W., Roy, A., Schluchter, W. M., & Howard, J. J. (2022).
 Teaching Basic Calculations in an Introductory Biology Lab. *Journal of Microbiology & Biology Education*, 23(1). https://doi.org/10.1128/jmbe.00199-21
- Hudzicki, J. (2012). Kirby-Bauer Disk Diffusion Susceptibility Test Protocol Author Information. American Society For Microbiology, December 2009, 1–13. https://www.asm.org/Protocols/Kirby-Bauer-Disk-Diffusion-Susceptibility-Test-Pro
- Hussain, T., Fatima, I., Rafay, M., Shabir, S., Akram, M., & Bano, S. (2016). Evaluation of antibacterial and antioxidant activity of leaves, fruit and bark of kigelia Africana. *Pakistan Journal of Botany*, 48(1), 277–283.
- Institute, C. and L. S. (2024). Quality System Regulation for Laboratory-Developed Tests

- A Practical Guide for the Laboratory.
- KGaA, M. (2018). Mueller Hinton Agar (M-HAgar). www.sigmaaldrich.com
- Lin, C.-C., Chuang, L.-T., Wang, Y.-C., Tang, Y.-H., Liao, J.-S., Amoako-Atta, B., Glew, R. H., & Glew, R. S. (2012). Polyphenol and antioxidant content of Kigelia africana leaves from Ghana. *Food*, *6*(1), 1–5.
- Nabatanzi, A., Nkadimeng, S. M., Lall, N., Kabasa, J. D., & McGaw, L. J. (2020a).

 Antioxidant and Anti-Inflammatory Activities of Kigelia africana (Lam.) Benth.

 Evidence-Based Complementary and Alternative Medicine, 2020.

 https://doi.org/10.1155/2020/4352084
- Nabatanzi, A., Nkadimeng, S. M., Lall, N., Kabasa, J. D., & McGaw, L. J. (2020b). Ethnobotany, phytochemistry and pharmacological activity of kigelia africana (Lam.) benth. (bignoniaceae). *Plants*, *9*(6), 1–29. https://doi.org/10.3390/plants9060753
- Olufemi, A., Omotayo, O., David, A., Monjeed, I., Adebola, O., Bilikis, S., & Temitope, A. (2017). Kigelia africana Stem Bark, Fruit and Leaf Extracts Alleviate Benzene-induced Leukaemia in Rats. *Journal of Pharmaceutical Research International*, 18(2), 1–10. https://doi.org/10.9734/jpri/2017/34625
- P. K. Chauhan, Rahul Thory, V. S. and I. P. P. (2019). *Phytochemical, Antioxidant and Antibacterial Activity of Chloroform and Methanolic Fruit Extracts of Kigelia africana*. *II*(23), 72. www.ujpah.in
- Plumb, D. C. (2008). Veterinary Drug Handbook Sixth Edition. In O. D. and C. T. D. C. A. tony buffington, dVM, Phd, dACVN Professor of Veterinary Clinical sciences the Ohio state university Veterinary Hospital Columbus, T. Andrea G. Cannon, dVM, dACVd Animal dermatology & Allergy rocklin, CA; Modesto, CA; boise, id Dermatological Agents, T. P. of C. O. P. Gigi davidson, diCVP North Carolina state university raleigh, NC Ophthalmology Products, A. A. P. C. C. D. for D. M. Camille deClementi, VMd, dAbt AsPCA Animal Poison Control Center urbana, iL Overdose and Toxin Exposure Decontamination Guidelines, O. D. and C. T. D. Michelle duMond, rVt Nutrition support specialist the Ohio state university Veterinary Hospital Columbus, & M. M. dinah Jordan, Pharmd, diCVP Mississippi state university Mississippi state, Ms Insulin, Maropitant, T. sandra Koch, dVM, Ms,

- dACVd College of Veterinary Medicine, university of Minnesota st. Paul, MN Dermatological Agents, & T. M. elaine Lust, Pharmd, diCVP Creighton university Omaha, Ne Ceftiofur (Free Acid, Sodium and HCl), Danofloxacin (Eds.), *PharmaVet Inc.* (Sixth edit). PharmaVet Inc.
- Saini, A. K., Chauhan, P. K., Singh, V., & Sharma, P. (2013a). Phytochemical, Antioxidant and in vitro Antibacterial Activity of Aqueous and Ethanolic Fruit Extracts of Kigelia Africana. *Indian Journal of Pharmaceutical and Biological Research*, *I*(02), 46–52. https://doi.org/10.30750/ijpbr.1.2.12
- Saini, A. K., Chauhan, P. K., Singh, V., & Sharma, P. (2013b). Phytochemical, Antioxidant and in vitro Antibacterial Activity of Aqueous and Ethanolic Fruit Extracts of Kigelia Africana. *Indian Journal of Pharmaceutical and Biological Research*, *1*(02), 46–52. https://doi.org/10.30750/ijpbr.1.2.12
- Semwal, D. K., Semwal, R. B., Ahmad, A., Kamatou, G. P., & Viljoen, A. M. (2014). Inhibitory Effects of Extracts and Isolates from Kigelia africana Fruits against Pathogenic Bacteria and Yeasts. 8(12), 1192–1197.
- Sidjui, L. S., Toghueo, R. M. K., Menkem Zeu, E., Mbouna, C. D. J., Leddet, V. M.-, Herbette, G., Fekam, F. B., Ollivier, E., & Folefoc, G. N. (2016). Antibacterial Activity of the Crude Extracts, Fractions and Compounds from the Stem Barks of Jacaranda mimosifolia and Kigelia africana (Bignoniaceae). *Pharmacologia*, 7(1), 22–31. https://doi.org/10.5567/pharmacologia.2016.22.31
- SS, S., SM, D., & MA, A. (2022). Antibacterial and Toxicity Evaluation of Stem Bark Extract of Kigelia Africana (Lam.) Benth. *Global Academic Journal of Agriculture and Biosciences*, 4(6), 86–91. https://doi.org/10.36348/gajab.2022.v04i06.001
- Susan E. Aiello, M. A. M. (2016). [PDF] The Merck Veterinary Manual. In D. Dana G. Allen, DVM, MSc, D. Peter D. Constable, BVSc (Hons), MS, PhD, D. Andrew Dart, BVSc, PhD, DACVS, P. Peter R. Davies, BVSc, D. (Avian P. Katherine E. Quesenberry, DVM, Facvs. Philip T. Reeves, BVSc, PhD, & P. Jagdev M. Sharma, BVSc, MS (Eds.), MERCK & CO., INC. KENILWORTH, NJ, USA (ELEVENTH E).
- wofford college. (n.d.). *Imagine*, 1–6. https://webs.wofford.edu/mossre/pdf/lab_solutions.pdf

Zirintunda, G., Kateregga, J., Nalule, S., Vudriko, P., Biryomumaisho, S., & Acai, J. O. (2024). An inventory of ethnoveterinary knowledge for chicken disease control in Soroti district, Uganda. *Journal of Medicinal Plants for Economic Development*, 8(1), 1–12. https://doi.org/10.4102/jomped.v8i1.248