

**Formulation of Herbal-Based Soap from *Rhoicissus tridentata* Root Extracts for
Management of Bacterial Skin Diseases**

By

Wabwiire Jacob

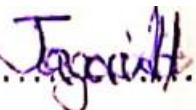
BU/UP/2018/3558

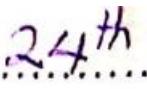
A research project report submitted to the Department of Chemistry for the partial fulfillment of
the requirement for the award of Bachelors of Science Education degree at Busitema University

MAY 2023

DECLARATION

I WABWIIRE JACOB declare that this research dissertation is my original work and has not been submitted elsewhere for the award of a degree. Where other people's work has been used, this has perhaps been recognized and cited according to the university policy.

Signature.....

Date..... /MAY/2023

WABWIIRE JACOB

BU/UP/2018/3558

APPROVAL

This research has been submitted for examination and has been approved by my supervisor.

Dr. Owor Richard Oriko

Signature ..

Date .. /MAY/2023

DEDICATION

I dedicate my work to my loving parents, Mr. Katebule Joseph and Mrs. Nabwire Jane for their infinite support. My brother Musana Joseph Katebule always held my hand when I lost hope to move forward.

ACKNOWLEDGEMENT

I want to express my special thanks of gratitude to my supervisor, Dr. Owor Richard for the guidance and support he gave to me. Appreciation also goes to the Busitema University Research and Innovation Fund (BURIF) for financial support towards this work.

Heartfelt thanks to my supportive parents, Mr. Katebule Joseph and Mrs. Nabwire Jane, for they sacrificed whatever they heard to enable me reach this stage in my education.

I also appreciate my brothers Musana Joseph, Musana Henry, Wandera Emmanuel, and Wandera Victor, and my sisters Achiengi Margaret and Sanyu Christine. I appreciate my supportive lecturer madam Namusana Hellen.

I appreciate my supportive friends Mwine Emmyson, Gomei Brian, Mwambu Junior, Dhikusooka Herman, Mwagale Flavia, Kampi Maria Gorretti, Chebijira Mercy, Kwemoi Robin, Opio Chrispin, Mugalagala Nicholus, Kiwuso Hassan, Namandare Seth, Watila Joshua, the entire natural products class and the 2019 chemistry class at large.

Table of Contents

DECLARATION	ii
APPROVAL.....	ii
ACKNOWLEDGEMENT.....	iv
ABSTRACT.....	1
ACRONYMS AND ABBREVIATION.....	1
List of Tables	3
List of Figures	4
CHAPTER ONE: INTRODUCTION.....	5
1.1 Background	5
1.2 Research Problem	7
1.3 Objectives.....	8
1.3.1 General Objective:.....	8
1.3.2 Specific Objectives:.....	8
1.4 Justification	8
CHAPTER TWO: LITERATURE REVIEW	10
2.1 <i>Rhoicissus Tridentata</i>	10
2.2 Ethnobotanical uses	11
2.3 Phytochemical compositions of <i>Rhoicissus tridentata</i>	11
2.3.1 Alkaloids	12
2.3.2 Flavonoids.....	13
2.3.3 Quinones	13
2.4 Phytochemical Screening.....	14
2.5 Pharmacological uses of <i>Rhoicissus tridentata</i>	15
2.5.1 Antimicrobial Activity	15
2.5.2 Anti-Inflammatory Activity	15
2.5.3 Antiproliferative Activity	15
2.5.4 Antioxidant Activity.....	16
2.5.5 Antidiabetic Activity	16
2.5.6 Ascaricidal Activity	16
2.6 Bacterial Skin Infections	16
2.6.1 Impetigo	17
2.6.2 Cellulitis	17

2.6.3 Erysipelas.....	17
2.6.4 Folliculitis.....	17
2.7 <i>Staphylococcus aureus</i>.....	18
2.8 Diagnosis of Bacterial Skin Infections.....	19
CHAPTER THREE: MATERIALS AND METHODOLOGY	21
3.1 Study Site	21
3.2 Plant material collection and identification.....	21
3.3 Sample preparation	21
3.4. Aqueous extract.....	21
3.5. Extraction with Organic solvents.....	22
3.6 Preliminary Phytochemical analysis	22
3.6.1 Test for Flavonoids.....	22
3.6.2 Test for steroids	23
3.6.3 Test for Terpenoids	23
3.6.4 Test for Tannins	23
3.6.5 Test for glycosides	24
3.6.6 Test for saponins	24
3.6.7 Test for alkaloids	24
3.6.8 Tests for Phenols	24
3.6.9 Test for quinones	25
3.7 Total phytochemical content.....	25
3.7.1 Total alkaloid content	25
3.7.2 Total flavonoid content.....	25
3.7.3 Total phenolic content	26
3.7.4 Total tannins content	27
3.8 Formulation of herbal soap (Rhoitiderm).....	28
3.8.1 Materials required and their proportions.....	28
CHAPTER FOUR: RESULTS AND DISCUSSION	33
4.1. Phytochemical Screening.....	33
4.2. Formulation.....	35
CHAPTER FIVE: CONCLUSION AND RECOMMENDATION.....	38
5.1 Conclusion	38
5.2 Recommendation.....	38

REFERENCES.....	39
-----------------	----

ABSTRACT

Bacterial skin infections are a major health issue worldwide. The major bacterial skin infections are cellulitis, folliculitis, impetigo, erysipelas, Staphylococcal scalded skin syndrome, and many others. These are majorly caused by gram-positive bacterial strains mainly *Staphylococcus aureus*. Recent literature shows that various forms of treatment have been developed to curb bacterial skin infections. Among these is the use of medicinal plants majorly in traditional African Societies. In this report, the major objective was to carry out phytochemical screening of root extracts of *R. tridentata* and to formulate a herbal soap product based on *Rhoicissus tridentata* root extracts for management of bacterial skin infections. The method of analysis described by Harbone, 1998 was employed during phytochemical screening and this showed the presence of flavonoids, alkaloids, and phenols as main phytochemicals. The formulated herbal soap was named Rhoitiderm and was found to have good physical-chemical properties such as lemon smell, pH, color, cleansing, and lathering power. Anti-bacterial assay of this herbal soap needs to be done to evaluate its pharmacological uses.

Keywords

Rhoicissus tridentata, *Staphylococcus aureus*, phytochemicals, Rhoitiderm, *Pentas decora*, anti-bacterial.

List of Tables

List of Figures

ACRONYMS AND ABBREVIATION

SA	<i>Staphylococcus aureus</i>
WHO	World Health Organisation
DALYs	Disability-Adjusted Life Years
HIV	Human Immunodeficiency Virus
AIDS	Acquired Immune Deficiency Syndrome
NTDs	Neglected Tropical Diseases
GDB	Global Disease Burden

UV	Ultra Violet
SSSS	Staphylococcal Scalded Skin Syndrome
TSS	Toxic Shock Syndrome
GABHS	Group A Beta-Hemolytic Streptococcus pyogenes
ETOAc	Ethyl Acetate
ETOAcE	Ethyl Acetate Extract
<i>P. decora</i>	<i>Pentas decora</i>

List of Tables

Table 1: Biochemical tests of Identifying <i>Staphylococcus aureus</i> from other <i>Staphylococci</i> Strains	19
Table 2: showing the ingredients used in the formulation of soap and their respective quantities	30
Table 3: Table showing lathering test for formulated soap	31
Table 4: Results of Phytochemical Screening of ethanolic root extract of <i>R. tridentata</i>	33

List of Figures

Figure 1: Rhoicissus tridentata leaves.....	11
Figure 2: Skin infected with impetigo ((a)-noon-bullous impetigo, (b)-bullous impetigo)	17
Figure 3: Gram positive bacteria.....	18
Figure 4: Staphylococcus aureus. S. aureus occurs in ‘grape-like’ clusters.	18
Figure 5: Powdered rootstocks of R. tridentata.....	21
Figure 6: A calibration curve for total flavonoid using standard quercetin solution.	26
Figure 7: A calibration curve for determination of total phenolic content of R. tridentata using standard ascorbic acid.....	27
Figure 8: A calibration curve for determination of total tannins using standard ascorbic acid	28
Figure 9: Making soap. (a)- Preparation of lye solution, (b) - mixing lye solution and coconut oil.....	29
Figure 10: Formulated herbal soap (RHOITRDERM); (a)- Soap after 24 hours, (b)- Reshaping the formulated soap, (c)- Soap cut into small shaped bars.....	30
Figure 11: Total phytochemical content of major phytochemicals in R. tridentata root extracts.	34
Figure 12: Herbal soap formulated from aqueous extract of R. tridentata.....	35
Figure 13: Results of pH test; (a) - the universal pH paper turned green indicating a pH above 8. While (b) the pH meter measured 12.24.	35
Figure 14: lather formation by sample soap (a – distilled water and b- hard water)	36
Figure 15: The cleansing ability of Rhoitiderm. In 15(b), the clean cotton wool was soaked in soap solution of distilled water as in 14(a) while the dirty cotton wool was soaked in solution in Figure 14(b).	36

CHAPTER ONE: INTRODUCTION

1.1 Background

There is widespread skin disease in rural areas in Sub-Saharan Africa. This is attributed to hot humid climates, overcrowding, poverty, and poor infrastructure (Bissek et al., 2012).

Skin conditions are usually the mirror of more severe illnesses such as HIV/AIDs (Njoroge & Bussmann, 2007) and neglected tropical diseases (NTDs), such as elephantiasis (R. J. Hay et al., 2014). Statistics indicate that 92% of individuals suffering from HIV have cutaneous and mucosal complications (Njoroge & Bussmann, 2007). As a result, skin diseases are among the most common infections seen in primary healthcare settings in tropical areas.

Of the new attendances at outpatient departments in developing countries, 11% of the patients have skin problems. Astonishingly, skin and subcutaneous conditions were the 18th global disease burden in the global Disability-Adjusted Life Years (DALYs) disease burden ranking in 2013 (GDB2013) and were the 4th leading cause of disability worldwide between 2010 and 2013 (Xue, Zhou, Xu, Li, Bao, Cheng, He, Xu, Ren, Zheng, et al., 2022).

The highest rates in the general population in the whole world are due to pyoderma, scabies, and superficial mycoses (Mahé et al., 2005). In Uganda, the most reported skin conditions are allergies, autoimmune reactions, bacteria, viruses, cancers, hemangioma, pellagra, and fungus. Bacterial skin infections rank the 28th diagnosis among hospitalized patients (Stulberg, Penrod, & Blatny, 2002). The most common bacterial skin infections include impetigo, boils and carbuncles, folliculitis, cellulitis, and erysipelas.

Bacterial skin diseases appear to lack much attention globally despite their intense effect on the global disease burden (Xue, Zhou, Xu, Li, Bao, Cheng, He, Xu, Ren, & rong Zheng, 2022). The burden caused by bacterial skin diseases is due to skin bacterial pathogens developing strong resistance to multiple drugs. These multi-drug resistant bacterial strains include; the gram-positive *Staphylococcus aureus* and Group A *Streptococcus pyogenes* and the gram-negative *Pseudomonas*, *Escherichia*, and *Klebsiella* (R. Hay et al., 2006)

Methicillin-resistant *Staphylococcus aureus* accounts for 50% of skin and soft tissue infections (Pfalzgraff, Brandenburg, & Weindl, 2018). However, *Streptococcus pyogenes* have been found to cause a worthy number of skin infections. Various antibiotics such as erythromycin,

REFERENCES

- Anand, U., Tudu, C. K., Nandy, S., Sunita, K., Tripathi, V., Loake, G. J., . . . Proćkow, J. (2022). Ethnodermatological use of medicinal plants in India: from ayurvedic formulations to clinical perspectives—a review. *Journal of ethnopharmacology*, 284, 114744.
- Bayisa, T. H. (2018). Phytochemical investigation and antimicrobial activities of leaf extracts of *Rhoicissus tridentata*.
- Bernard, P. (2008). Management of common bacterial infections of the skin. *Current opinion in infectious diseases*, 21(2), 122-128.
- Bissek, A.-C. Z.-K., Tabah, E. N., Kouotou, E., Sini, V., Yepnjo, F. N., Nditancho, R., . . . Fonsah, J. Y. (2012). The spectrum of skin diseases in a rural setting in Cameroon (sub-Saharan Africa). *BMC dermatology*, 12(1), 1-10.
- Brookes, K., & Katsoulis, L. (2006). Bioactive components of *Rhoicissus tridentate*: A pregnancy-related traditional medicine. *South African journal of science*, 102(5), 267-272.
- Chang, A. Y., Scheel, A., Dewyer, A., Hovis, I. W., Sarnacki, R., Aliku, T., . . . Maurer, T. A. (2019). Prevalence, clinical features and antibiotic susceptibility of group a streptococcal skin infections in school children in urban Western and northern Uganda. *The Pediatric Infectious Disease Journal*, 38(12), 1183-1188.
- Deurenberg, R. H., & Stobberingh, E. E. (2008). The evolution of *Staphylococcus aureus*. *Infection, genetics and evolution*, 8(6), 747-763.
- Dube, N. P., Siwe-Noundou, X., Krause, R. W., Kemboi, D., Tembu, V. J., & Manicum, A.-L. (2021). Review of the Traditional Uses, Phytochemistry, and Pharmacological Activities of *Rhoicissus* Species (Vitaceae). *Molecules*, 26(8), 2306.
- Edward, O., Bazira, J., & Grace, M. (2015). Clinical Description, Bacterial Causes and the Association of HIV with Pyodermas Presenting at a Skin Clinic of a Tertiary Hospital in Rural South Western Uganda.
- Foster, T. J. (2002). *Staphylococcus aureus*. *Molecular Medical Microbiology*, 839-888.
- Havsteen, B. H. (2002). The biochemistry and medical significance of the flavonoids. *Pharmacology & therapeutics*, 96(2-3), 67-202.
- Hay, R., Bendeck, S. E., Chen, S., Estrada, R., Haddix, A., McLeod, T., & Mahé, A. (2006). Skin diseases. *Disease Control Priorities in Developing Countries*. 2nd edition.

- Hay, R. J., & Asiedu, K. (2018). Skin-related neglected tropical diseases (skin NTDs)—a new challenge. In (Vol. 4, pp. 4): MDPI.
- Hay, R. J., Johns, N. E., Williams, H. C., Bolliger, I. W., Dellavalle, R. P., Margolis, D. J., . . . Wulf, S. K. (2014). The global burden of skin disease in 2010: an analysis of the prevalence and impact of skin conditions. *Journal of Investigative Dermatology*, 134(6), 1527-1534.
- Ibrahim, F., Khan, T., & Pujalte, G. G. (2015). Bacterial skin infections. *Primary Care: Clinics in Office Practice*, 42(4), 485-499.
- Karak, P. (2019). Biological activities of flavonoids: an overview. *Int. J. Pharm. Sci. Res.*, 10(4), 1567-1574.
- Mahé, A., Faye, O., N'Diaye, H. T., Ly, F., Konare, H., Keita, S., . . . Hay, R. (2005). Definition of an algorithm for the management of common skin diseases at primary health care level in sub-Saharan Africa. *Transactions of the Royal Society of Tropical Medicine and Hygiene*, 99(1), 39-47.
- Mdletshe, N. W. (2018). *Comparison of pharmalogical activity of rhoicissus tomentosa and Rhoicissus tridentata for the treatment of elephantiasis in South Africa*. University of the Free State,
- Naidoo, V., Chikoto, H., Bekker, L., & Eloff, J. (2006). Antioxidant compounds in *Rhoicissus tridentata* extracts may explain their antibabesial activity: research in action. *South African journal of science*, 102(5), 198-200.
- Nalule, A., Mbaria, J., Kimenju, J., & Olila, D. (2012). Ascaricidal activity of *Rhoicissus tridentata* root-tuber ethanolic and water extracts. *Livest. Res. Rural. Dev*, 24.
- Njoroge, G. N., & Bussmann, R. W. (2007). Ethnotherapeutic management of skin diseases among the Kikuyus of Central Kenya. *Journal of ethnopharmacology*, 111(2), 303-307.
- Palit, A., & Inamadar, A. C. (2010). Current concepts in the management of bacterial skin infections in children. *Indian Journal of Dermatology, Venereology and Leprology*, 76, 476.
- Panche, A. N., Diwan, A. D., & Chandra, S. R. (2016). Flavonoids: an overview. *Journal of nutritional science*, 5, e47.

- Pfaltzgraff, A., Brandenburg, K., & Weindl, G. (2018). Antimicrobial Peptides and Their Therapeutic Potential for Bacterial Skin Infections and Wounds. *Frontiers in Pharmacology*, 9. doi:10.3389/fphar.2018.00281
- Roberts, M. F. (2013). *Alkaloids: biochemistry, ecology, and medicinal applications*: Springer Science & Business Media.
- Shamsudin, N. F., Ahmed, Q. U., Mahmood, S., Ali Shah, S. A., Khatib, A., Mukhtar, S., . . . Zakaria, Z. A. (2022). Antibacterial Effects of Flavonoids and Their Structure-Activity Relationship Study: A Comparative Interpretation. *Molecules*, 27(4), 1149.
- Sofowora, A. (1996). Research on medicinal plants and traditional medicine in Africa. *The Journal of Alternative and Complementary Medicine*, 2(3), 365-372.
- Stulberg, D. L., Penrod, M. A., & Blatny, R. A. (2002). Common bacterial skin infections. *American family physician*, 66(1), 119.
- Sukumaran, V., & Senanayake, S. (2016). Bacterial skin and soft tissue infections. *Australian prescriber*, 39(5), 159.
- Taylor, T. A., & Unakal, C. G. (2022). *Staphylococcus aureus*. In *StatPearls [Internet]*: StatPearls Publishing.
- Tushabe, F., Mwebaze, E., & Kiwanuka, F. (2011). *An image-based diagnosis of virus and bacterial skin infections*. Paper presented at the The International Conference on Complications in Interventional Radiology.
- Urton, N. R., Olivier, M. C., & Robertson, B. (1986). The taxonomy of the *Rhoicissus tridentata* (Vitaceae) complex in southern Africa. *South African Journal of Botany*, 52(5), 389-396.
- Xue, Y., Zhou, J., Xu, B.-N., Li, Y., Bao, W., Cheng, X. L., . . . rong Zheng, Y. (2022). Global Burden of Bacterial Skin Diseases: A Systematic Analysis Combined With Sociodemographic Index, 1990–2019. *Frontiers in medicine*, 9.
- Xue, Y., Zhou, J., Xu, B.-N., Li, Y., Bao, W., Cheng, X. L., . . . Jia, C. Y. (2022). Global Burden of Bacterial Skin Diseases: A Systematic Analysis Combined With Sociodemographic Index, 1990–2019. *Frontiers in Medicine*, 9. doi:10.3389/fmed.2022.861115