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2 **ANTIMICROBIAL AND WOUND HEALING PROPERTIES**
3 **OF LEAF EXTRACTS OF *SPERMACOCE VERTICILLATA***
4 ***LINN***

Comment [PP1]: Please check the correct form of nomenclature.

6
7 **Abstract**

8 The Rubiaceae family is one of the most important medicinal families that are found in the
9 tropical region of the world. There are many species under the Rubiaceae family such as
10 *Coffea arabica*, *Cinchona Officinalis*, *Spermacoce verticillata*, and many more. There are
11 several parts used to extract the metabolites such as the leaf, bark, root, seed, and many more.
12 The leaf of *Spermacoce verticillata* has many medicinal values as it contains metabolites that
13 help to reduce microbial activity and help to heal the wound. The microbial activity can lead
14 to many skin infections, internal disease, and so on. The metabolites act on some of the
15 factors such as inhibition of cell wall formation, inhibition of protein synthesis, and many
16 more.

Comment [PP2]: Scientific names should be in Italics format.

17 *Wound healing is referred to as the cuts and infection on the human skin*, leaf extract helps to
18 increase the fibrin formation that helps to clot the blood, except fibrin formation metabolic
19 compounds helps to reduce the microbial load from the wound itself to reduce the chance of
20 infection. Plant leaves contain many metabolites that act as antimicrobial substances for
21 humans such as minimoidiones A, Cytochalasin D, multiforicin I, xylarenones E, and so on.
22 Some factors can disrupt the sequence of wound healing such as local factors containing
23 blood sugar that seriously affect the wound healing and external factors such as
24 contamination of microbes in the wound. The wound healing property of leaf extract helps
25 the body's immune system to fight against the microbes that cause a delay in the wound
26 healing process by releasing harmful metabolic activity.

Comment [PP3]: Statement is not clear.

27 **Keywords:** *Wound healing*, *Rubiaceae family*, *Spermacoce verticillata*, *human body*.

Comment [PP4]: It should be in Italics format.

Comment [PP5]: All keywords are not in correct format.

33 Introduction

34 Traditional medicine has wide acceptance in all countries irrespective of its sophistication
35 and development. [This type of medicine is popular in rural areas as it has easy access, low
36 cost, fewer side effects, and greater efficiency than any other medicine. *Spermacoce*
37 *verticillata* Linn, also known as the *Borrelia verticillate* Linn, is a semi-woody or perennial
38 shrub, it belongs to the family of Rubiaceae. According to the name, this shrub has
39 verticillaster inflorescence. This plant is a type of weed that has many medicinal values.
40 Namukobe *et al.* (2021) stated that western tropical African is the finding region of the plant.
41 The local name of the plant is 'DIOLA eribulin', the leaf of this plant has more medicinal
42 value than any other part of the plant. Antimicrobial properties can be defined as the
43 substances that can kill harmful microbes in the human body. [_____
44 The wound healing property can be defined as the acceleration of the wound healing process
45 of humans. The wound healing procedure requires fast cell regeneration that can replace the
46 damaged cell. This plant can also be found in the tropical regions of Asia and America.
47 This herb has the property that can propel the fibrin formation that helps the blood to clot in a
48 wound. The leaf extract also has antimicrobial properties that inhibit harmful bacteria to grow
49 in the wound by inhibiting the cell wall formation, inhibition of protein synthesis,
50 depolarization of cell membrane, inhibition of nucleic acid synthesis, inhibition of the
51 metabolic pathways of microorganisms. Plants that have been used in African society have
52 various antimicrobial properties against many harmful microorganisms. Ouattara *et al.* (2020)
53 argued that this plant has antimicrobial properties against the *Staphylococcus aureus*,
54 *Salmonella typhi*, *Escherichia coli*, *Enterobacter sp.*, *Klebsiella sp.*, and so on. Western
55 countries are now depending upon plant-based medicine for their modern medicine, reducing
56 the number of chemicals helping people from side effects of drugs. [_____
57

Comment [PP6]: Add reference.

Comment [PP7]: Add reference.

Comment [PP8]: Name of scientist should not be in italics.

Comment [PP9]: Name of scientist should not be in italics.

Comment [PP10]: Add reference.

Comment [PP11]: Add reference.

Comment [PP12]: Add references.

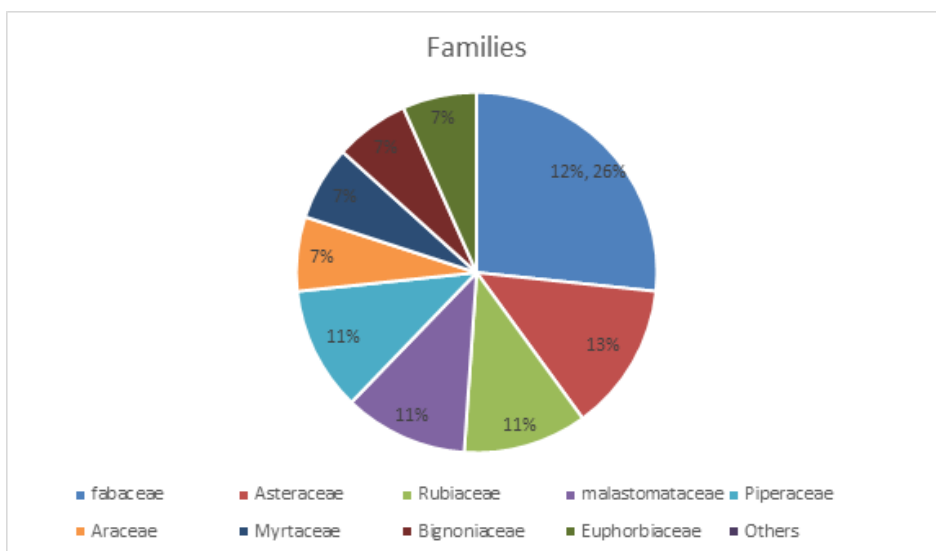


Figure 1: Importance of plant families in herbal medicine

(Source: Tinde van Ande, 2019)

Comment [PP13]: The first letter of legend should be upper case letter.

Literature review

Antimicrobial properties of *Spermacoce verticillata* Linn.

Comment [PP14]: Name should be Italics.

The human body is the ideal place for all different microorganisms, some of the microorganisms are useful for the human body, and the rest of the microorganisms are harmful to the human body. *Spermacoce verticillata* is an herb that is related to the family of Rubiaceae, this family has many antimicrobial activities that prevent the microbes from growing in the human body. Microorganisms can be various types such as bacteria, viruses, protozoa, fungi, and many more. ZERROUG *et al.* (2018) opined those human immune systems are the primary barrier between infection and microorganisms. The Rubiaceae family has an excellent number of antimicrobial activities that help to reduce the microbes load from humans. The *Spermacoce verticillata* have antimicrobial properties such as antifungal, antiprotozoal, antibacterial activities, and so on. Plant leaves contain many metabolites that act as antimicrobial substances for humans such as minimoidiones A, Cytochalasin D, multiforicin I, xylarenones E, and so on.

Comment [PP15]: Add references.

The Rubiaceae family has a wide range of herbs that all contain medicinal properties, especially in the leaf section. Microbes are found everywhere, the effect of mycobacterial activity can be beneficial and can be harmful, and this is decided by the metabolites produced by the microorganism. The production of the harmful metabolism can be reduced or ceased

80 by the activity of metabolites produced by the *Spermacoce verticillata*. Riondato *et al.* (2019)
81 stated that many ways can reduce the production of microbial metabolites such as reducing
82 the effect of metabolites, dissolving the outer membrane of the microbe, dissolving the
83 nucleic acid, and sealing the protein synthesis, and so on. Some of the diseases caused by
84 fungus are candidiasis, aspergillosis, blastomycosis, and so on. An example of such a
85 metabolite that is found from *Spermacoce verticillata* is 'Cytochalasin D', this metabolite acts
86 as the antifungal drug that inhibits the metabolites of a fungus and hence reduces the effect of
87 the fungus.

88 **Wound healing properties of *Spermacoce verticillata* Linn**

Comment [PP16]: Correct the format.

89 The wound can be defined as the break or damage of the skin surface of the human body.
90 Wounds can be minor cuts, abrasions, bites, surgical wounds, and so on. Stages including
91 wound healing are clotting, the proliferative stage, angiogenesis, fibroplasia, epithelialization,
92 and remodeling phase. Tanvir *et al.* (2019) argued that the leaf extract of the *Spermacoce*
93 *verticillata* helps the proliferative stage of wound healing that overall propel the wound
94 healing procedure. Studies have been found that 70% of wound healing medicines are made
95 from the metabolites of plants. The Rubiaceae family has more medicinal valuable plants
96 available than most of the family, *Spermacoce verticillata* is one of the important species that
97 have a comprehensive role in wound healing properties. For successful wound healing, the
98 stages of wound healing should be carried out properly. Some factors can disrupt the
99 sequence of wound healing such as local factors containing blood sugar that seriously affect
100 the wound healing and external factors such as contamination of microbes in the wound.

101 The leaf is one of the important organs in plants, here most of the chemical reaction takes
102 place such as photosynthesis, glycolysis, Krebs cycle, and hence a lot of metabolites are
103 produced. These metabolites have various properties. Singh *et al.* (2019) opined that the
104 wound healing property of leaf extract helps the body's immune system to fight against the
105 microbes that cause a delay in the wound healing process by releasing harmful metabolic
106 activity. Metabolites found in *Spermacoce verticillata* have a multidirectional activity to
107 propel the wound healing activity such as accelerating the fibrin production with the help of
108 blood cells. The leaf extract can be dissolved in methanol, chloroform, and water for their
109 optimum use. Fibrin is a type of polymer made out of protein that blocks the exit point of
110 blood and fluid. The other property is the reduction of toxins produced by the microbes that
111 delay fibrin production.

Comment [PP17]: Add more references.

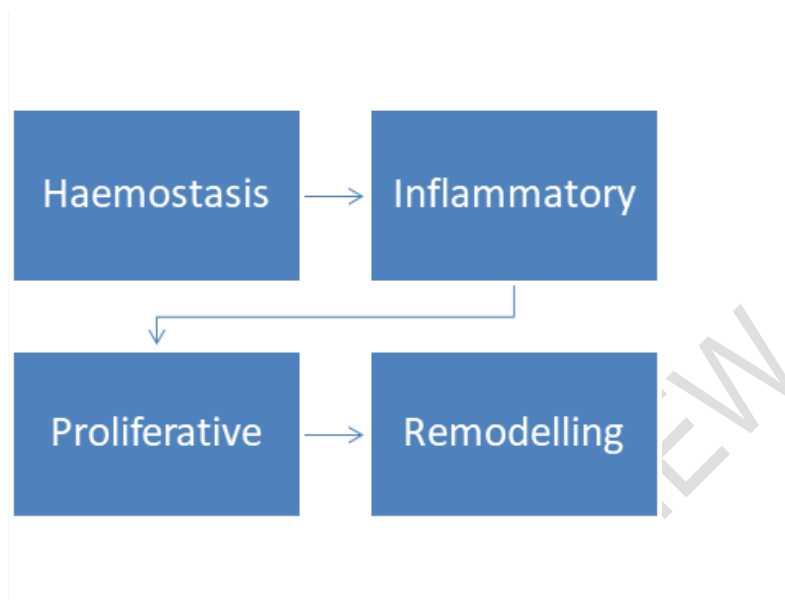


Figure 2: Steps for wound healing by *Spermacoce verticillata*

(Source: Singh, 2019)

Comment [PP18]: Diagram is not clear; it should be more informative.

Methodology

The research method is a wide term that includes data collection and analysis. The research philosophy can be defined as a collection of data, analysis of data, and use of data (Mohajan, 2018). This literature is based on the *secondary qualitative research method* as there is the use of a journal, science articles to collect the data about the metabolites that are found in *Spermacoce verticillata*. Analysis of the result has found that 70% of the antimicrobial and wound healing drugs come from plant-based metabolites. These metabolites can act as antimicrobial agents as well as wound healing agents.

This literature is also based on the *deductive approach* that describes the scientific investigation about the topic and collection of data from appropriate scientific journals. The steps including wound healing by the usage of leaf extract obtained indicate that it is based on a deductive approach (Mohajan, 2018). This is scientific literature hence it is based on the correct facts that have been collected to justify the literature therefore it can be defined as a *positivism* research approach. The research article is based on the *descriptive approach* that is by this research the question 'what diseases can be cured by the leaf extract' and 'why plant-based metabolites are effective against most of the microbes' can be answered.

Comment [PP19]: Add materials and methods you have used but not what it means.

Comment [PP20]: Is this related to your research. No need to say that.

132 **Discussion**

133 **Usage of leaf extract of *Spermacoce Verticillata* for curing diseases**

134 Microbes are everywhere, the disease caused by the microbes are of various types. There are
135 different kinds of microbes present in the human body, by the exposed wound or sensitive
136 skin the metabolites produced by them can harm the human body severely. Aamir *et al.*
137 (2020) stated that the disease can be formed by certain types of microbes such as fungus,
138 protozoa, bacteria, and they cause superficial infection such as skin disease. The leaf extract
139 of *Spermacoce verticillata* works upon the external infection caused by the microbes such as
140 athlete's foot, ringworm, cellulitis, erysipelas, and so on.

141 Suksungworn *et al.* (2021) argued that *Spermacoce verticillata* has various compounds that
142 act as antifungal, antibacterial, and antiprotozoal. Some of the compounds are *cytochalasin*
143 *D*, *11-bromo roquefortine*, *4-hydroxy-mellein*, *8-methyl-mellein*, *guignardone I* ,
144 *guignardone B*, *mycoleptones A*, *mycoleptones B* and many more. These compounds are
145 classified as **alkaloids**, **coumarin**, **meroterpene**, **polyketide**, and so on. These compounds
146 are found in the leaf extract of almost every species of the Rubiaceae family and act as
147 antimicrobial properties that also help to propel the wound healing properties by reducing the
148 microbial load from the wound. Leaf extract of *Spermacoce verticillata* has been used by the
149 tribal people for many years. Currently, western countries are investing in this species to
150 collect the base compound for their medicine.

Classification	Compound	Biological activities
Alkaloid	Cytochalasin D	Antifungal
Alkaloid	11-bromo roquefortine	Antibacterial
Coumarin	4-hydroxy-mellein	Antifungal
Coumarin	8-methyl-mellein	Antifungal
Meroterpene	guignardone I	Antibacterial
Meroterpene	guignardone B	Antibacterial
Polyketide	mycoleptones A	Antiprotozoal
Polyketide	mycoleptones B	Antiprotozoal

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152 **Table 1: Compounds found in Rubiaceae family and their biological activities**

153

(Source: Influenced by Suksungworn, 2021)

154

155 **Analysis of properties of leaf extracts acting as an antimicrobial and wound**
156 **healing agent**

157 The Rubiaceae family has one of the most medicinal valued plants available. *Spermacoce*
158 *verticillata* has excellent antimicrobial activity and wound healing properties that are found in
159 leaves. Ajaib *et al.* (2018) opined that the extraction of leaves can be done on various
160 solvents such as alcohol, methanol, chloroform, water, and so on. The activity of the
161 compounds is dependent on the solvent hence choosing the correct solvent for each
162 compound is important. Methanol as a solvent works well, many bacteria such as *Bacillus*
163 *subtilis*, *Klebsiella pneumonia*, *Pseudomonas aeruginosa*, and many more show less activity
164 in the presence of methanol mixed compound. The basic principle is the same: the leaf extract
165 has various compounds that have several activities regarding antibacterial, antiprotozoal,
166 antifungal. These compounds have several properties that help to decrease the population of
167 the microbes, such as inhibiting the cell wall formation, inhibiting the protein synthesis,
168 degeneration of the genetic materials, depolarizing the cell membrane, and many more.

169 Inhibition of the cell wall formation leads to the destruction of the protoplasm of the
170 particular microorganism, inhibiting protein synthesis can lead to cease of metabolite
171 production of the microbes, degenerating the genetic material results from overall death of
172 the particular microorganism, the cell membrane is a polarized structure that repels the
173 charged particles to enter or exit from the cell of microorganism, depolarizing leads to the
174 destruction of the cell membrane and hence death of the microbe. Patel *et al.* (2020) stated
175 that the wound healing property is directly related to the antimicrobial properties of the leaf
176 extract. The wound can be defined as damaged skin tissue, without proper treatment this
177 damaged tissue can lead to infection and increase the microbial activities at the wound. The
178 leaf extract promotes fibrin protection that acts as a barrier between blood and the outer
179 surface of the skin, also it acts on microbes that are present on the wound and can cause
180 infection.

181 **Importance of Rubiaceae family in medicinal value**

182 The Rubiaceae family is one of the largest tropical families that are found all over the world.
183 Several plant species are found in the Rubiaceae family such as *Coffea arabica*, *Cinchona*
184 *Officinalis*, *Hamelia patens*, *Rubia sp.*, *Gardenia sp.*, *Ixora sp.*, and so on. These plants can be
185 used in various diseases such as ulcers, jaundice, asthma, leprosy, cough, fever, hyperacidity,
186 and so on. AUTHINARAYANAN *et al.* (2019) argued that these plants have several

187 metabolic compounds that work on those diseases, some of the compounds are **decahedron-**
188 **2-methoxy, β sitosterol-glucoside, isovanillin**, and so on. The compounds are derived from
189 the various parts of the plants such as a leaf, bark, root, fruit, seed, whole plant, and so on.
190

Plant part used	Number of species
Leaf	17
Bark	6
Root	5
Fruit	3
Seed	2
Whole plant	1
Aerial part	1
Stem bark	1

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192 **Table 2: Table showing plant parts used to extract metabolites that have medicinal**
193 **value (Rubiaceae family)**
194 (Source: Influenced by Patel, 2020)

195 Conclusion

196 The use of plant-based medicine has been occurring since ancient times. The medicinal value
197 of the plant is extraordinary; it is also used by many medical organizations and tribal people
198 that live in tropical areas on earth. The invention of modern chemicals that has a quick
199 response in any kind of disease but also has their side effects, but plant-based drugs have very
200 few side effects or no side effects. *Spermacoce verticillata* Linn is one of the medicinally
201 valued plants of the family Rubiaceae. The leaf extract of this species has various medicinal
202 values regarding antimicrobial activities and wound healing. More use of these kinds of
203 shrubs can comprehensively reduce the chance of side effects that can cause other difficulties.

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UNDER PEER REVIEW

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219 **Reference**

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