

Original Research Article

Evaluation of the antibacterial activity of commonly used plants for wound treatment in southwestern Nigeria

ABSTRACT

Aims: Infection of wounds by microorganisms can prolong wound healing process and result in wound associated complications. Therefore, wound treatment entails the use of antimicrobial agents usually administered directly on the wound where possible to prevent microbial colonization. Traditionally, various plants have been used in wound treatment in different regions of the world. This study evaluated the contribution of the antibacterial activity of four plants commonly use in the treatment of wound in southwestern Nigeria to their ethnobotanically acclaimed wound healing property.

Methodology: The antibacterial activity of aqueous and ethanolic extracts of the selected plants (*Chromolaena odorata*, *Sida acuta*, *Ageratum conyzoides* and *Carica papaya*) was evaluated using the agar well diffusion assay. Wound isolates of *Pseudomonas aeruginosa* and *Staphylococcus aureus*, two commonly isolated Gram negative and Gram-positive bacteria from wounds were used for this study. Antibacterial activity was inferred for plant extracts that achieved zone of inhibition ≥ 7 mm in diameter (size of the well inclusive).

Results: Generally, the ethanolic extracts of the selected plants showed better extraction yield and antibacterial activity compared to the aqueous extracts. The ethanolic extracts of the four selected plants demonstrated antibacterial activity against the test organisms used while only the aqueous extracts of *Chromolaena odorata* and *Sida acuta* showed activity against both test organisms. The aqueous extracts of *Ageratum conyzoides* and *Carica papaya* only showed antibacterial activity against *S. aureus*.

Conclusion: Results from this study demonstrated that the antibacterial activity of the selected plants contributes to their acclaimed wound healing property. Although there is need to investigate the role of other non-antibacterial properties of the plants that may be associated with wound healing to fully understand the usefulness of the plants in wound treatment.

Keywords: Wound infection, *Chromolaena odorata*, *Sida acuta*, *Ageratum conyzoides*, *Carica papaya*, antibacterial activity

1. INTRODUCTION

Microbial colonization of wound can result in the complication of the wound healing process (Guo and DiPietro, 2010; Maheswary *et al.*, 2021). Following a break in the cutaneous layer resulting from a wound, there is an exposure of subcutaneous tissue. The exposed subcutaneous tissue provides favourable conditions such as moisture and nutrients required for the colonization and rapid growth of microorganisms (Chiller *et al.*, 2001). Thus, wounds

are very prone to microbial colonization. Microorganisms including opportunistic microbial species that are normal microfloral of the intact skin are common wound colonizers (Chiller *et al.*, 2001; Bowler *et al.*, 2001). Commonly isolated bacteria species that colonize wounds include *Pseudomonas species*, *Staphylococcus aureus*, *Klebsiella species*, *Escherichia coli* and *Streptococcus species* (Bessa *et al.*, 2015).

The prevention of microbial contamination of wound through the correct application of appropriate antimicrobial is crucial to wound healing. Wound management usually include the application of antiseptics and antibiotics to control microbial contamination (Daeschlein, 2013). Plants are generally source of cheap pharmaceuticals that have been employed in various medical interventions in different part of the world for ages (Petrovska, 2012). Traditionally, plants have been employed in the treatment of wounds (Shalu *et al.*, 2016; Shedoeva *et al.*, 2019). This study investigated the antibacterial activity of *Chromolaena odorata*, *Sida acuta*, *Ageratum conyzoides* and *Carica papaya*, four plants commonly used in the treatment of wounds in the southwestern part of Nigeria. *Chromolaena odorata* is a widely distributed weed in tropical Africa, North America, and South and Southeast Asia (Kriticos *et al.*, 2005). It's locally known as *Ewe Akintola* in southwestern part of Nigeria. *Sida acuta* and *Ageratum conyzoides* likewise are commonly growing weed in tropical Africa and are locally known in southwestern Nigeria as *Isekotu* and *Imi esu* respectively (Okunade, 2002; Shittu and Alagbe, 2020). *Carica papaya* locally known as *Ibepe* in southwest Nigeria is a fruit tree of tropical and subtropical regions (Yogiraj *et al.*, 2004). The leaves of the selected plants are commonly employed in the southwestern part of Nigeria in wound treatments. In this study, the contribution of their possible antibacterial activity to their acclaimed wound healing property was investigated.

2. MATERIAL AND METHODS

Collection and preparation of plant material

Chromolaena odorata, *Sida acuta*, and *Ageratum conyzoides* wildy growing in Akungba-Akoko were collected during the raining season, the leaves were plucked and washed severally in clean water. The leaves of *Carica papaya* tree were also plucked and washed as in the other plants. All plants used were identified based on their morphological characters. The washed leaves were spread out on clean flat surfaces and kept to air dry in a well aerated room under shed. The dried leaves of the plants were ground to fine powder using mechanical blender (Silver crest, Germany) and subsequently stored in airtight container until when needed.

Preparation of plant extract

Plant extracts were prepared according to the method of Akinbobola and Dada (2014) with slight modification. The powdered plant materials (100 g) were suspended into 1000 mL of the extraction solvents used (distilled water and ethanol) for 72 hours. The suspensions were wrist shaken vigorously at intervals throughout the duration of the extraction. The suspensions were subsequently filtered through a muslin cloth after the extraction period to obtain the plant filtrates. Filtrates from the ethanol extraction solvent were air dried at room temperature while the filtrate from the aqueous extraction were dried through lyophilisation under vacuum in a freeze dryer (LSBC50 benchtop freeze dryer). The recovered extracts were weighed to compare the percentage recovery of each extract before the extracts were reconstituted in 30% Dimethyl sulphoxide (DMSO) by dissolving 1 g of the extracts in 10 mL of DMSO to obtain a concentration of 100 mg/mL of the extracts. The reconstituted extracts were vigorously shaken severally and allowed to properly dissolve in DMSO for 24 hours before being sterilised by filtration through 0.22 µm filter (Millipore, Germany).

Evaluation of antibacterial activity of plant extract

The antibacterial activity of the plant extracts against wound isolates of *P. aeruginosa* and *S. aureus* was evaluated using the agar well diffusion method according to the method of Pai *et al.* (2011). Inoculum of the test organism were prepared from overnight fresh cultures and

adjusted to obtain the 0.5 McFarland standard. Wells (6 mm in diameter) bored in freshly prepare Mueller-Hinton agar (Lab M, UK) seeded with the standardised inoculum of the test organisms were filled with 50 µL of the plant extracts. Sterile 30% DMSO was used as the solvent control while tetracycline, a commonly used antibiotics for topical wound treatment in the region was used as the standard for the antibacterial activity assay. Antibacterial activity of the plant extract was evaluated by measuring the diameter of the zone of inhibition.

3. RESULTS AND DISCUSSION

The percentage recovery yield of the different solvents used for the plant extraction is shown in Table 1. Higher extracts yields were generally recovered from the ethanolic extraction compared to the aqueous extraction. *Sida acuta* extracted with ethanol gave the highest yield overall (11%) while the lowest yield (1.5%) was recorded for *Sida acuta* aqueous extract. Evaluation of the activity of the different plant extracts against *P. aeruginosa* and *S. aureus* is shown in Table 2. Plant extracts achieving diameter of zone of inhibition ≥ 7 mm (inclusive of bored well) were inferred as having antibacterial activity against the test isolates. Most of the plant extracts tested showed activity against the clinical isolates of *P. aeruginosa* and *S. aureus* used for the study. However, the aqueous extracts of *Ageratum conyzoides* and *Carica papaya* showed no activity against the *P. aeruginosa* isolate.

The infection of wound by microorganisms can result in a delay in the wound healing process and other complications like formation of abscesses and invasion of adjacent tissues or even the blood stream by microorganisms or their products (Bowler, 2002). Infection of wound with antibiotics resistant bacteria strains and formation of biofilms in wounds by common wound colonizers like *P. aeruginosa* complicate the control of wound infections (Schaber *et al.*, 2007; Pirvanescu *et al.*, 2014)). Therefore, prevention of wound infection through the use of antibiotics and antiseptics is a priority in wound management. Various plants have been traditionally used in the treatment of wound in various parts of the world, this study evaluated the contribution of the antibacterial activity of four plants commonly used in the treatment of wound in southwestern part of Nigeria to their acclaimed wound healing property.

The antibacterial activities of aqueous and ethanolic extracts of *Chromolaena odorata*, *Sida acuta*, *Ageratum conyzoides* and *Carica papaya* against wound isolates of *P. aeruginosa* and *S. aureus*, was evaluated in this study. Two extraction solvents of different polarity were used in extracting the phytochemicals present in the plants studied. This is to ensure that the different phytochemicals present in the selected plants were extracted during the extraction process for antibacterial analysis. The products of the different extraction solvents used in this study differs in percentage yield. Generally, higher yields of the extracts were recorded for the ethanolic extracts compared to the aqueous extract. Similar observation was reported by Thophon *et al.* (2016) and Stanley *et al.* (2014) in the extraction of *Chromolaena odorata*. However, Truong *et al.* (2019) reported a different observation while evaluating the extraction potential of different solvents in the extraction of the phytochemical constituents of *Severinia buxifolia*. The difference in result from these studies can be attributed to the difference in the polarity of the phytochemical constituents of the plants studied.

The aqueous and ethanolic extracts of the different plants studied showed antibacterial activity against the clinical isolates of *S. aureus*. Similar observation was recorded for *P. aeruginosa* except for the aqueous extract of *Ageratum conyzoides* and *Carica papaya* which showed no antibacterial activity against *P. aeruginosa* isolate. Similar antibacterial activities of the studied plants against other bacteria isolates but not wound isolates have been reported in literature. Thophon *et al.* (2016) reported similar antibacterial activity of aqueous and ethanolic extracts of *Chromolaena odorata* against typed cultures of common bacteria causative agents of skin Infections. Fadehan *et al.* (2015) and Senthilkumar *et al.* (2018) likewise demonstrated the antibacterial activity of aqueous extract of *Ageratum*

conyzoides and *Sida acuta* against *Klebsiella pneumoniae* and *S. aureus* respectively. Similar to the observation in this study, Nirosha and Mangalanayaki (2013) demonstrated that the ethanolic extracts of *Carica papaya* has better antibacterial activity compared to the aqueous extract.

Table 1: The percentage yield of the crude plant extracts

Plant species	Extracts	Percentage yield
<i>Chromolaena odorata</i>	Water	1.8%
	Ethanol	2.1%
<i>Sida acuta</i>	Water	1.5%
	Ethanol	11%
<i>Ageratum conyzoides</i>	Water	1.7%
	Ethanol	2.2%
<i>Carica papaya</i>	Water	1.6%
	Ethanol	2.0%

Table 2: Evaluation of the antibacterial activity of plant extracts against wound isolates of *P. aeruginosa* and *S. aureus*

Plant species	Extracts	<i>P. aeruginosa</i>	<i>S. aureus</i>
<i>Chromolaena odorata</i>	Water	+	+
	Ethanol	+	+
<i>Sida acuta</i>	Water	+	+
	Ethanol	+	+
<i>Ageratum conyzoides</i>	Water	-	+
	Ethanol	+	+
<i>Carica papaya</i>	Water	-	+
	Ethanol	+	+
	30% DMSO	-	-
	Tetracycline	+	+
(-) no apparent zone of inhibition (no antibacterial activity) (+) zone of inhibition $\geq$ 7mm (antibacterial activity)			

4. CONCLUSION

Observations from this study demonstrated that extracts from the plant studied largely have antibacterial activity against common wound colonisers. Therefore, the antibacterial activity of the studied plant contributes to their acclaimed wound healing property. Although, the antimicrobial activity of the studied extract was lower than the antimicrobial activity of commercially available antibiotics that is commonly used in the region of interest in the treatment of wound, the plants extract however provide a cheap source of antimicrobial agent. Also, plant extracts have not been linked with the emergence of antibiotics resistant bacteria strains, thus the use of purified plant extract in wound treatment may help in reducing the indiscriminate of antibiotics.

COMPETING INTERESTS DISCLAIMER:

Authors have declared that no competing interests exist. The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

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