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Interactive antimicrobial and toxicity profiles of conventional antimicrobials with Southern African medicinal plants

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ABSTRACT

Medicinal plant use plays an important role in the healthcare of many South Africans. Furthermore, in orthodox medicine, conventional antimicrobial agents are amongst the most commonly prescribed groups of drugs. Therefore, due to the prevalence of use of these two forms of healthcare, there is a high probability for their concurrent use. Thus, the aim of this study was to evaluate the interactive antimicrobial and toxicity profiles of six Southern African medicinal plants (*Agathosma betulina*, *Aloe ferox*, *Artemisia afra*, *Lippia javanica*, *Pelargonium sidoides* and *Sutherlandia frutescens*) when combined with seven conventional antimicrobials (ciprofloxacin, erythromycin, gentamicin, penicillin G, tetracycline, amphotericin B and nystatin). Antimicrobial activity was assessed using the minimum inhibitory concentration (MIC) assay against a range of pathogens and interactions were further classified using the sum of the fractional inhibitory concentration (∑FIC). Notable synergistic or antagonistic interactions were studied at various ratios (isobolograms). The toxicity of the individual samples, as well as the notable combinations, was assessed using the brine-shrimp lethality assay (BSLA) and the 3-(4,5 dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) assay on the HEK-293 human cell line. Of the 420 antimicrobial: plant combinations studied, 14.29% showed synergistic interactions, 7.56% antagonistic, 35.71% additive and 42.44% indifferent interactions. Some notable synergistic interactions (ciprofloxacin with *A. betulina* and *S. frutescens* against *Escherichia coli*) and antagonistic interactions (ciprofloxacin with *A. afra* organic extract against *Escherichia coli*) were identified. None of the notable combinations were found to show toxicity in the BSLA or MTT assay. In conclusion, the majority of combinations were found to have no notable interaction, alleviating some concern related to the concurrent use of these two forms of healthcare.

1. Introduction

Medicinal plants have been used for centuries as a source of medicine. The global importance of medicinal plants can be illustrated by the numerous conventional drugs that have been derived from plants and are currently used in clinical practice. Some examples of these drugs are quinine, atropine, opioids and taxol. In Africa, traditionally used medicinal plants play a vital role in the cultural heritage of the local people, with an estimated 60% of the population consulting traditional healers (Chinyama, 2009; Van Wyk et al., 2009). Approximately 3000 plants are used in traditional healing practices in South Africa by an estimated 200,000 traditional healers (Van Wyk et al., 2009). Popular Southern African medicinal plants, such as *Agathosma betulina*, *Aloe ferox*, *Artemisia afra*, *Lippia javanica*, *Pelargonium sidoides* and *Sutherlandia frutescens*, have been studied for their medicinal, antimicrobial and toxic properties (Table 1).

Interest in medicinal plant research has escalated, with the aim of identifying alternative antimicrobial therapies to overcome resistance (Aiyegoro and Okoh, 2009). There is, however, general consensus amongst the various studies, that plant derived antimicrobials possess a lower potency than conventional antimicrobials (Van Vuuren and Viljoen, 2011). Furthermore, antimicrobial resistance against conventional antimicrobials has been on the rise and has become a major public health concern. This has propelled research in the direction of combination therapies for enhanced efficacy. Many researchers have studied antimicrobial interactions between natural products, as well as combinations of natural products with conventional therapies. Websites now exist that are dedicated to herb–drug interactions (www.prescribeguide.com). Combinations of agents with antimicrobial properties that have already been investigated include combinations of various essential oils (Van Vuuren and Viljoen, 2006; Suliman et al., 2010) and conventional antimicrobial combinations with non-conventional antibiotics, such as anaesthetics (Gunics et al., 2000). Several studies investigating natural product combinations with conventional antimicrobials have already been conducted (Betoni et al., 2006; Rosato et al., 2007, 2008, 2009; D'Arrigo et al., 2010; Jarrar et al., 2010;

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Table 1
Medicinal plants investigated, with their traditional uses, evidence of toxicity and antimicrobial activity.

Plant, family and common names	Part used/mode of administration	Traditional medicinal uses	Known toxicity	Known antimicrobial activity[a]	References
Agathosma betulina (Berg.) Pillans, Rutaceae, buchu (Khoi, English), boegoe (Afrikaans), ibuchu (Xhosa).	Decoction or alcoholic tincture from leaves for gastrointestinal complaints. Infusions prepared from leaves ingested for kidney troubles. Buchu vinegar applied topically.	Kidney and urinary tract infections (UTI's), wounds, boils, rash, burns, gastrointestinal complaints, antibiotic protection of corpses.	No toxic effect on kidney cells (IC_{50} > 100 μg/ml). Allergic reactions have occurred.	Very weak activity against *E. coli, S. aureus, B. cereus, E. faecalis, K. pneumoniae, P. aeruginosa* and *C. neoformans.*	Watt and Breyer-Brandwijk (1962), Hutchings et al. (1996), Lis-Balchin et al. (2001), Moolla (2005), Moolla and Viljoen (2008), Van Wyk et al. (2009), Suliman et al. (2010), Van Wyk (2011).
Aloe ferox Mill., Asphodelaceae, bitteraalwyn, Kaapse aalwyn (Afrikaans), bitter aloe (English), umhlaba (Xhosa, Zulu, Sotho).	Fresh juice from leaves or decoctions and powders from leaves or roots applied topically or sniffed.	Ophthalmic inflammation, sexually transmitted infections, wounds, burns, sinusitis, conjunctivitis.	Joint weakness, partial paralysis, effects similar to curare poisoning, overdoses lead to nephritis, gastritis and pelvic congestion. No cytotoxicity at low doses.	Moderate to very weak activity against *C. albicans, Neisseria gonorrhoeae* and *Herpes simplex.*	Watt and Breyer-Brandwijk, 1962, Hutchings et al. (1996), Kambizi et al. (2007), Kambizi and Afolayan (2008), Van Wyk et al. (2009), Van Wyk (2011), Wintola et al. (2011).
Artemisia afra Jacq. ex. Willd., Asteraceae, umhlonyane (Xhosa, Zulu), lengana (Sotho, Tswana), als, alsem, wildeals (Afrikaans), african wormwood (English).	Infusion or decoction from leaves or roots for ingestion, poultice of leaves for topical application. Fumes from boiled leaves for inhalation.	Respiratory infections (coughs, colds, pneumonia, croup, whooping cough), gastrointestinal complaints, malaria, intestinal worms, boils.	Pulmonary oedema, haemorrhagic nephritis, degenerative liver changes, central nervous system effects due to thujone (hallucinations, confusion).	Moderate to very weak activity against *B. cereus, E. faecalis, S. aureus, E. coli, K. pneumoniae, C. albicans P. aeruginosa*, and *C. neoformans.*	Watt and Breyer-Brandwijk (1962), Hutchings et al. (1996), Huffman et al. (2002), Van Vuuren and Viljoen (2006), Mukinda and Syce (2007), Van Wyk et al. (2009), Suliman et al. (2010), Van Wyk (2011).
Lippia javanica (Burm. F.) Spreng., Verbenaceae, musukudu, bokhukhwane (Tswana), inzinziniba (Xhosa), umsuzwane (Zulu), mumara (Shona), fever tea (English), koorsbossie (Afrikaans).	Weak infusions prepared from leaves, twigs and roots made with milk or water, smoke inhalation or the direct application of leaves.	Respiratory infections (coughs, colds, bronchitis, influenza), skin infections, gastrointestinal complaints, malaria, measles, rashes, disinfecting anthrax-infected meat.	Photosensitivity but no other evidence of toxicity.	Moderate to very weak activity against *S. aureus, B. cereus, E. faecalis, E. coli, K. pneumoniae, C. albicans, P. aeruginosa* and *C. neoformans.*	Watt and Breyer-Brandwijk (1962), Hutchings et al. (1996), Huffman et al. (2002), Van Vuuren and Viljoen (2006), Van Wyk et al. (2009), Van Wyk (2011).
Pelargonium sidoides DC., Geraniaceae, umckaloabo (Zulu), silverleaf geranium (English), kalwerbossie (Afrikaans).	Root decoction or infusion made with milk or water for ingestion and topical application. Root can be chewed or powdered for ingestion with food.	Respiratory infections (bronchitis, sinusitis, influenza, pneumonia), sexually transmitted infections, gastrointestinal complaints, wounds.	Hepatotoxicity reports caused by *P. sidoides* ruled out.	Moderate to very weak activity against *Mycobacterium tuberculosis, S. aureus, S. pneumoniae, E. coli, K. pneumoniae, P. aeruginosa* and *Haemophilus influenzae.*	Watt and Breyer-Brandwijk (1962), Hutchings et al. (1996), Mukinda and Syce (2007), Van Wyk et al. (2009), Kolodziej (2011), Van Wyk (2011), Teschke et al. (2012).
Sutherlandia frutescens (L.) R. Br., Fabaceae, kankerbos (Afrikaans), cancer bush (English).	Strong decoctions or alcoholic tinctures made from leaves for internal or external use.	Respiratory infections (chronic bronchitis, colds, influenza), UTI's, wounds, gastrointestinal complaints, internal cancer and septicaemia.	No toxic effects on liver, kidney, muscles, lungs, bone and biochemical parameters found in mice given enormous doses. Considered safe due to long history of use in South Africa without reports of any toxicity. No toxic effects in healthy adults.	Moderate to very weak activity against *S. aureus* and other Staphylococcal spp., *E. faecalis* and *E. coli.*	Watt and Breyer-Brandwijk (1962), Hutchings et al. (1996), Seier et al. (2002), Katerere and Eloff (2005), Fu et al. (2008), Van Wyk et al. (2009), Van Wyk (2011).

[a] Moderate antimicrobial activity = MIC of 1.00–3.00 mg/ml; very weak antimicrobial activity = MIC of ≥8.00 mg/ml.