American Journal of Microbiological Research, 2017, Vol. 5, No. 4, 91-93
Available online at http://pubs.sciepub.com/ajmr/5/4/4

DOI:10.12691/ajmr-5-4-4

Antimicrobial Properties of Phenolic Extracts against Nosocomial Drug Resistant Organisms

Sambasivarao Yaragalla[1,*], Surya k krishnanbabu[2], Jaisankar L Gopinath[2], Buvan G Gengadharan[2], Thanagam Brightline MK[2], Harnell Madhurin[3]

[1]Microbiology, Spartan Health Sciences University, Community Health and Research Centre of Spartan, St. Lucia
[2]Basic Science-5, Spartan Health Sciences University, St. Lucia
[3]Regional Plant Institute, V-fot, St. Lucia
*Corresponding author: siva@spartanmed.org

Abstract Nosocomial Multi Drug resistance microbial infection is a global concern nowadays. Due to its multi-drug resistant nature, treatment with conventional antibiotics does not assure desired clinical outcomes. Therefore, there is a need to find new compounds and/or alternative methods to get arsenal against the pathogen. Combination therapies using conventional antibiotics and phytochemicals fulfill both requirements [1]. A variety of essential oils have been screened for their antimicrobial activity [15]. The antimicrobial activity of plant-derived essential oils is the basis of many applications, especially in food preservation, aromatherapy and medicine. This present study demonstrates the efficacy of phenolic-phytochemical extract combinations from pine, coconut and oatmeal colloidal solution against multidrug resistant organisms such as *Staphylococcus aureus (ATCC 43300), Pseudomonas aeruginosa (ATCC 27853) and Candida albicans (ATCC 10231).* The efficacy of the phytochemical combinations against these microbes were measured by estimating the inhibitory zone as produced by antibiotic sensitivity disc diffusion method on Mueller-Hinton agar. These results can be used as a support in doing further quantitative analysis in prevention and treatment of the common infectious diseases that is caused by these organisms [4].

Keywords: *phytochemicals, antibiotic, nosocomial, drug resistance, staphylococcus, pseudomonas and candida*

Cite This Article: Sambasivarao Yaragalla, Surya k krishnanbabu, Jaisankar L Gopinath, Buvan G Gengadharan, Thanagam Brightline MK, and Harnell Madhurin, "Antimicrobial Properties of Phenolic Extracts against Nosocomial Drug Resistant Organisms." *American Journal of Microbiological Research*, vol. 5, no. 4 (2017): 91-93. doi: 10.12691/ajmr-5-4-4.

1. Introduction

Nosocomial multi Drug resistant organisms are one of the most common causes of infection in hospitals [1]. It has been nicknamed 'superbug' due to its multi-drug resistance to most of the contemporary antibiotics [6]. At this point, the use of drug combinations rather than single drugs provide better clinical outcomes, as the use of single agent is highly associated with occurrence of resistance [7]. Many reports suggest that the use of drug combinations against multi-drug resistant bacterial pathogens have better efficacy compared to monotherapy [8]. The use of western antibiotics, however, has encountered adaptive resistance over time, even in combinations [9,10]. Alternative compounds and secondary metabolites derived from plants or insects offer a rich source as antimicrobial agents [11]. Plants are a rich source of useful secondary metabolites that forms the plant defense mechanism against pathogenic invaders [11]. These include tannins, flavonoids, alkaloids, terpenoids and polyphenols. They have effective antibacterial properties against both Gram positive and Gram negative bacteria [11]. Therefore, their potential use in combinations with antibiotics can help to potentiate the activity of the western drugs, resulting in increased efficacy.

Antibiotics with different mechanism of actions and that are sensitive and resistance against Staphylococcus aureus, pseudomonas aeruginosa and candida albicans were chosen for this study. Tetracycline and streptomycin (Protein synthesis inhibitor drugs), Vancomycin (Cell wall inhibitory drug) and Amphotericin (cell membrane inhibitory drug) were used in combination with one phytochemical against three Staphylococcus aureus, pseudomonas aeruginosa and candida albicans strains. The results were assessed by antibiotic sensitivity disc diffusion method and Gram stain.

2. Materials and Methods

2.1. Bacteria and Growth Conditions

Three microbial species were employed as test organisms which include *Pseudomonas aeruginosa, Staphylococcus aureus and Candida albicans* which is obtained from St.Jude's Hospital, Saint Lucia. The bacteria were grown in Mueller-Hinton Agar (MH). Inocula were prepared by adding an overnight culture of the organism in MH broth to obtain an OD600 0.1. The cells were incubated to grow until they obtain the McFarland standard 0.5 (approximately 105-108 CFU/ml).

2.2. Test Plants and Their Extraction

Phenolic extract of pine, coconut and oatmeal colloidal solution was collected from the organic heritage, V-fort St. Lucia for the study. The content of total phenolic was carried out based on the absorbance values of the various extract solutions, reacted with Folin–Ciocalteu reagent and compared with the standard solutions [14].

2.3. Antibiotics & Culture

All antibiotics, Culture media and chemicals were obtained from Carolina Biological Ltd. (USA).

2.4. Microscopical Identification of the Isolated Bacteria and Fungi

Microbial Isolate were stained by lacto phenol cotton blue and Gram stain. Microbes Identified by the presence of:

Fungal hyphae (branched filaments)

Color

Shape and arrangement.

Fungi and bacteria were identified by comparing micrographic characteristic of fungi to stander microbiology text book.

2.5. Antibacterial Assay

Antibacterial activity of the extracts and antibiotics were tested using the agar diffusion Kirby bauer method on Muller Hinton agar with negative control. The plates were incubated at 37°C for 24 hours and the zones of inhibition measured.

2.6. Antifungal Assay

Antifungal activity of the extracts and antibiotics were tested using the agar diffusion Kirby bauer method on Sabourauds Dextrose agar with negative. The plates were incubated at 28°C for 24-72 hours and the zones of inhibition measured.

3. Results

The combination of extract, antibiotics and bacterial samples was assessed by Disc diffusion method in department of microbiology using Muller Hinton agar media to see the antibacterial activity and sabouraud's dextrose agar (SDA) media for antifungal activity. The results obtained are presented in Table 1 for qualitative analysis of Anti-bacterial and antifungal activity.

Given is antibacterial activity in Figure 1, Figure 2 illustrates a representative plate showing the antibacterial activity of phenolic extracts from pine, coconut and oatmeal colloidal solution that produced zones of inhibition against staphylococcus aureus and no inhibitory zone for pseudomonas and candida albicans.

Table 1.

S. No	Anti-bacterial activity of		
	staphylococcus aureus	pseudomonas aeruginosa	Candida albicans
Negative control	No Inhibitory zone		
Tetacyclin (20ug)	No Inhibitory zone	No Inhibitory zone	-
Streptomycin (15 ug)	Resistance	No Inhibitory zone	-
Vancomycin (10 ug)	5mm	-	-
Phenolic Extract	9mm	No Inhibitory zone	No Inhibitory zone
Amphotericin 30 ug	-	-	No Inhibitory zone
Ticarcillin 15 ug	-	6mm	-
Methicillin 20 ug	No Inhibitory zone	-	-

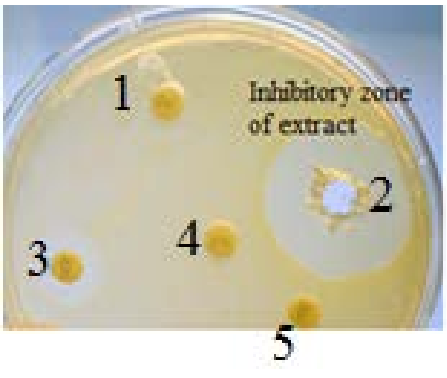

Figure 1. 1. Penicillin- Resistance; 2. Phenolic extract-Sensitive; 3. Vancomycine-Sensitive; 4. Methicillin-Resistance; 5. Tetracycline-Resistance

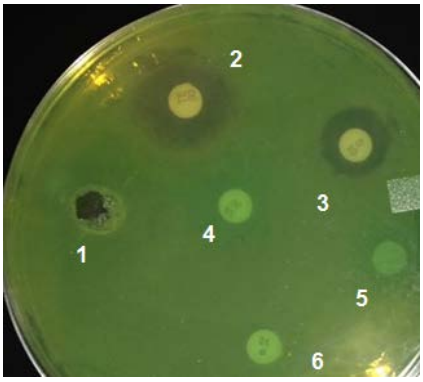

Figure 2. 1. Phenolic extract- Resistance or Growth seen; 2. Ticarcillin – Sensitive; 3. Streptomycin – Sensitive; 4. Tetracycline – Resistance; 5. Neg. control–Growth seen; 6. Penicillin – Resistance

4. Discussion

Phytochemicals is a fundamental requirement and pharmaceutical drug products. According to WHO, phytochemicals need to be standardized with safety guideline before releasing into market [12]. In this study, Phenolic extract was used which are easily available in St. Lucia and other tropical countries. The results were presented in Table 1. Further work is warranted to isolate and characterize the active principles available in the phenolic extracts essential oils. It is quite sure that such components could be useful in developing drugs [13]. This study shows that there is a significant bacteriostatic activity against Staphylococcus aureus, but did not have any effect on Pseudomonas aeruginosa and Candida albicans, that is evident by the lack of inhibitory zone in their respective culture Medias. It is evident that this phytochemical can exhibit bacteriostatic effects against Staphylococcus aureus. Furthermore work should be done and characterize its active principles and it could possibly developed as a new drug that can kill or prevent the drug resistant Staphylococcus aureus.

5. Conclusion

In conclusion, this study have provided informative data about the antimicrobial resistance to tested plant extracts towards Staphylococcus aureus, pseudomonas aeruginosa and candida albicans strains. Furthermore research is much needed for this phytochemical combination to be developed into a drug that can be used against community and nosocomial associated infections caused by multidrug resistant organisms. The millenarian use of these plants in folk medicine suggests that they represent an economic and safe alternative to treat infectious diseases.

Acknowledgements

We are sincerely thankful for Dr. Gurumurthy from Spartan health Sciences University to his efforts to evaluate results, Institutional Review Board approval and completion of this work in the department of Microbiology. Also thankful to Dr. Rajeev Pandey for CHARCOAS Board approval at Spartan university.

References

[1] Bhone Myint Kyaw, Shuchi arora, and Chu Sing Lim. 2012. Bactericidal antibiotic-phytochemical combinations against methicillin resistant Staphylococcus aureus, Jul-Sep; 43(3): 938-945.

[2] Aimé Gabriel Fankam, Jules-Roger Kuiate and Victor Kuete. 2017. Antibacterial and antibiotic resistance modulatory activities of leaves and bark extracts of Recinodindron heudelotii (Euphorbiaceae) against multidrug-resistant Gram-negative bacteria, *BMC Complementary and Alternative Medicine*BMC series – open, inclusive and trusted, 17: 168.

[3] Biswajit Chakraborty, Anupam Nath, Himadri Saikia, Mahuya Sengupta. 2014. Bactericidal activity of selected medicinal plants against multidrug resistant bacterial strains from clinical isolates, Asian Pac J Trop Med, 7(Suppl 1): S435-S441.

[4] Pandey R, Sambasivarao Y, Gurumurthy. 2013. Antibacterial Activity of Medicinal Plants against Pathogens from Extracts of Achyranthes Aspera. Med Aromat Plants 2: 135.

[5] Mohtar M, Johari SA, Li AR, Isa MM, Mustafa S, Ali AM, Basri DF. 2009. Inhibitory and resistance-modifying potential of plant-based alkaloids against methicillin-resistant Staphylococcus aureus (MRSA), Curr Microbiol. Aug; 59(2): 181-6.

[6] Foster, T.J. 2004. The *Staphylococcus aureus* "superbug". *J. Clin. Invest.* 114 (12), 1693-1696.

[7] Yamaoka T. 2007. The bactericidal effects of anti-MRSA agents with rifampicin and sulfamethoxazole-trimethoprim against intracellular phagocytized MRSA, *J Infect Chemother*. Jun; 13(3): 141-6.

[8] Cottarel G, Wierzbowski J. 2007. Combination drugs, an emerging option for antibacterial therapy, *Trends Biotechnol.* Dec; 25(12): 547-55.

[9] Entenza JM, Giddey M, Vouillamoz J, Moreillon P. 2010. In vitro prevention of the emergence of daptomycin resistance in Staphylococcus aureus and enterococci following combination with amoxicillin/clavulanic acid or ampicillin, *Int J Antimicrob Agents.* May; 35(5): 451-6.

[10] Mouton JW. 1999. Combination therapy as a tool to prevent emergence of bacterial resistance, *Infection.* 27 Suppl 2: S24-8.

[11] Cowan MM. 1999. Plant products as antimicrobial agents, *Clin Microbiol Rev*. Oct; 12(4):564-82.

[12] Owoyale, J. A., G. A. Olatunji, and S. O. Oguntoye. 2005. "Antifungal and antibacterial activities of an alcoholic extract of Senna alata leaves." *Journal of Applied Sciences and Environmental Management*, 9.3: 105-107.

[13] Geetha K, Narayanan KR, Murugesan AG, 2010. Antimicrobial efficiency of *Achyranthes aspera* L. *a*gainst selected pathogenic organisms. J Biosci Res 1: 187-190.

[14] Singelton, V.R., Orthifer, R., Lamuela-Raventos, R.M., 1999. Analysis of total phenols and other oxidation substrates and antioxidants by means of Folin–Ciocalteu reagent. Methods in Enzymology 299, 152-178.

[15] Cantrell CL, Fischer NH, Urbatsch L, McGuire MS, Franzblau SG. 1998. Antimycobacterial crude plant extracts from South, Central, and North America. Phytomedicine 5(2): 137-145.