

ANTIBACTERIAL AND ANTIFUNGAL STUDIES ON THE BIOSOOT OF COMMON WEEDS

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Abstract

Introduction: Biosoot is defined as the smoke or soot generated when burning the biomaterials which may either be of plant or of animal origin. This airborne soot is mostly considered as a negative sign as it may cause different respiratory ailment when they become airborne. However, the impact of this airborne soot depends upon characters like their size and their composition. Due to their chemical or organic compounds associated with the biosoot, their biological activity may differ.

Aim: In this study an attempt was made to study the antimicrobial potency of the biosoot of few of the common weeds, *Calotropis gigantea*, *Lantana camara* and *Parthenium hysterophorus*. **Methods:** The shade dried stem of the plants were burnt to the formation of soot and the soot deposited on the surface of tiles exposed were collected. The methanolic extract of the biosoot were evaluated against the bacteria, *Bacillus subtilis*, *Enterococcus faecalis*, *Salmonella typhi* and *Pseudomonas aeruginosa* and the fungi, *Aspergillus flavus* and *Aspergillus niger* using well diffusion method. **Results:** The soot of *Lantana camara* showed high antibacterial activity against *Enterococcus faecalis* and *Bacillus subtilis*, *Calotropis gigantea* against *Salmonella typhi* and the soot of *Parthenium hysterophorus* recorded high antibacterial activity against the bacteria, *Pseudomonas aeruginosa*. Similarly the biosoot of *Calotropis gigantea* showed better antifungal activity against the fungus, *Aspergillus flavus* and *Parthenium hysterophorus* against the fungus, *Aspergillus niger*. **Conclusion:** The study aptly demonstrates the utilization of biosoot as an antimicrobial agent when they are in general considered as waste and hazardous. Further, the biosoot of these plants can be potentially utilized in pharmaceutical and cosmeceutical industries after proper evaluation of their toxicity.

Keywords: Biosoot, Antimicrobial, *Calotropis gigantea*, *Lantana camara*, *Parthenium hysterophorus*, Pharmaceuticals, Cosmeceuticals.

INTRODUCTION

Soot is made up of fine black particles (Chuang et al., 2011). They may be prepared in a controlled environment for their usage in printing, painting and automobile industries. They may also found to emanate due to coal, charred wood, while cooking, oil lamps, fireplaces, wood stoves, furnaces and in general by combustion of fossil based fuels (Glacer et al., 2005; Scheeper and Bos, 1992; Kamboures et al., 2013). However, Biosoot is defined as a smoke or soot emanating by burning any biological material which may either be of plant origin or animal origin (Udayaprakash and Bhuvanewari, 2019). Soot in general is considered as waste by products which are derived from combustion of organic materials (Long et al., 2013). It is reported that polyaromatic hydrocarbons associated with the soot are considered as a cancer causing agent (Cain et al., 2010). Further, it is learnt that the environmental heterogenous conditions like oxidation reactions in the environment changes the chemical composition of soot (Browne et al., 2014). The components of soot are found to cause premature

human mortality (Goto, 2014). Thus, the soot in general is considered as waste and as a negative sign causing different respiratory ailment when they become airborne. However, the impact of this airborne soot depends upon characters like their size and their composition. Due to their chemical or organic compounds associated with the biosoot, their biological activity may differ.

It is known that the microorganisms are continually developing resistance towards many antibiotics resulting in formation of different multidrug resistant strains. They show resistance towards popular antibiotics posing bigger challenge for the scientists to find novel antibiotics. Thus, developing newer method of development of antibiotics, identifying novel antibiotics are identifying different sources of antibiotics are need of the hour. In that sense, many researchers work towards different nanoparticles against bacteria and fungi (Arokiyaraj et al., 2014). Over the past few years, carbon nanoparticles were also evaluated for their antimicrobial potency (Dong et al., 2012; Varghese et al., 2013). In this work the biosoot prepared from the stem of few common weeds, *Calotropis gigantea*, *Lantana camara* and *Parthenium hysterophorus* were studied for their antimicrobial property against the bacteria, *Bacillus subtilis*, *Enterococcus faecalis*, *Salmonella typhi* and *Pseudomonas aeruginosa* and the fungi, *Aspergillus flavus* and *Aspergillus niger*. It is the first report on the antimicrobial property of a Biosoot is reported.

MATERIAL AND METHOD

Plant Collection

The plants, i.e. common weeds, *Lantana camara* (Verbenaceae), *Calotropis gigantea* (Apocynaceae) and *Parthenium hysterophorus* (Asteraceae) were collected from the wastelands spread over Chennai, the state capital of Tamil Nadu, India. The plants are found to grow as a weed in most of the parts of Chennai and also found throughout the state of Tamil Nadu.

Soot Collection:

The stem of the plants were collected and shade dried. The shade dried stem are examined for their crispiness when tried to break. Such air dried stem are burnt openly (combustion) and the soot developed were exposed to porcelain tiles. The deposited soot were collected and stored in glass vials for further usage. A gram of biosoot was dissolved in 10ml of methanol and allowed under continuous stirring for 48 hours. The extract was filtered and used to evaluate antibacterial and antifungal studied.

Antimicrobial Property

The antimicrobial potency of the soot collected from different plant species were subjected to antimicrobial studies. The studies on antibacterial potency against the bacterial standards of *Bacillus subtilis* (MTCC 121), *Enterococcus faecalis* (MTCC 1320), *Salmonella typhi* (MTCC 531) and *Pseudomonas aeruginosa* (MTCC 424) procured from Microbial Type Culture Collection and Gene Bank, Chandigarh, India and antifungal studies against the fungal strains of *Aspergillus niger* (MLCT 0021) and *Aspergillus flavus* (MLCT 0022) belonging to Marina Labs, Chennai were conducted.

Evaluation of Antibacterial Potency

The stock cultures were maintained on slants of nutrient agar in 4°C. Active cultures for screening their susceptibility were prepared by transferring loop full of cells from stock cultures to test tubes containing Mueller Hilton Broth and were incubated at 37°C for 24 hours. Well diffusion method was used to screen the antibacterial activity. For assay, Mueller Hinton Agar (Beef Extract - 2g; Acid Hydrolysate of Casein - 17.5g; Starch - 1.5g; Agar - 17g. was used and Final pH was maintained at 7.3 ± 0.1) (MHA) was used. Onto the sterile MHA plates 0.1mL of the saline suspension of individual cultures were swabbed uniformly. The well of 5mm diameter was bored on the MHA plates using a sterile, stainless steel cork borer. Different concentrations of the the methanolic extracts (25µg, 50µg and 75 µg) were loaded onto the well. The plates were incubated at 37°C

for 24 hours. After incubation period, the diameter of inhibition zones formed around the well was measured in millimeter. These studies were performed in duplicates for all the bacterial samples. Streptomycin (25mg/well) and was used as positive control.

Evaluation of Antifungal Potency

The assay is carried out by well diffusion method. Sabouraud's Dextrose Agar (SDA) were used to study antifungal activity. The plates containing SDA were spread with the spores of specific culture, i.e. *Aspergillus niger* (MLCT 0021) and *Aspergillus flavus* (MLCT0022) using a swab and wells were bored in each plate at the diameter of 5mm each. The wells were filled with the extracts of 25µg, 50µg and 75 µg concentration/well. The plates with fungal cultures were incubated at 28°C for 72 h for evaluation. The development of the zone of inhibition was recorded. Ketoconazole (25mg/well) was used as positive control for fungal species. The assays were performed in duplicate.

RESULTS

Antibacterial Activity

The antibacterial studies using the soot samples of *Calotropis gigantea*, *Lantana camara* and *Parthenium hysterophorus* were conducted against the following bacteria, i.e. *Bacillus subtilis* (MTCC 121), *Enterococcus faecalis* (MTCC 1320), *Salmonella typhi* (MTCC 531) and *Pseudomonas aeruginosa* (MTCC 424). The well diffusion test was conducted using Mueller Hinton Agar with the concentration of 25 µg, 50 µg and 75 µg/well. Maximum zone of inhibition was recorded for the soot sample of *Parthenium hysterophorus* against the bacteria *Pseudomonas aeruginosa*. The minimum zone of inhibition was recorded against the *Bacillus subtilis* for the soot sample of *Calotropis gigantea*. The soot of *Lantana camara* showed high antibacterial activity against *Enterococcus faecalis* and *Bacillus subtilis*, *Calotropis gigantea* against *Salmonella typhi* and the soot of *Parthenium hysterophorus* recorded high antibacterial activity against the bacteria, *Pseudomonas aeruginosa*. The zone of inhibition recorded for each soot sample against individual bacteria is provided in Table 1.

Table 1. Zone of inhibition recorded for the biosoot of different plants against the bacteria studied.

Calotropis gigantea				
Extract (µg)	Enterococcus faecalis	Salmonella typhi	Bacillus subtilis	Pseudomonas aeruginosa
25µg	14mm	17mm	8mm	8mm
50µg	15mm	20mm	9mm	12mm
75µg	16mm	20mm	9mm	20mm
Lantana camara				
25µg	14mm	18mm	8mm	16mm
50µg	17mm	20mm	20mm	17mm
75µg	18mm	20mm	20mm	21mm
Parthenium hysterophorus				
25µg	10mm	10mm	13mm	20mm
50µg	14mm	10mm	18mm	23mm

75µg	15mm	17mm	18mm	24mm
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Antifungal Activity

The antibacterial studies using the soot samples of *Calotropis gigantea*, *Lantana camara* and *Parthenium hysterophorus* were conducted against the following bacteria, i.e. *Aspergillus niger* and *Aspergillus flavus*. The well diffusion test was conducted using Sabouraud's Dextrose Agar with the concentration of 25 µg, 50 µg and 75 µg/well. Maximum zone of inhibition was recorded for the soot sample of *Parthenium hysterophorus* against both the fungus, *Aspergillus niger* and *A. flavus*. Similarly the biosoot of *Calotropis gigantea* showed better antifungal activity against the fungus, *Aspergillus flavus* and *Parthenium hysterophorus* against the fungus, *Aspergillus niger*. The zone of inhibition recorded for each soot sample against individual fungus is provided in Table 2.

Table 2. Zone of inhibition recorded for the biosoot of different plants against the fungi studied.

Calotropis gigantea		
Extract (µg)	Extract (µg)	Extract (µg)
25µg	25µg	25µg
50µg	50µg	50µg
75µg	75µg	75µg
Lantana camara		
25µg	25µg	25µg
50µg	50µg	50µg
75µg	75µg	75µg
Parthenium hysterophorus		
25µg	25µg	25µg
50µg	50µg	50µg
75µg	75µg	75µg

DISCUSSION

Soot represents the by product which is unwanted due to uncontrolled incomplete combustion or pyrolysis of carbon containing materials (IARC, 1985). They are derived from different materials which includes, burning of paper, rubber, plastic etc. However, biosoot are defined as soot developed from biological material which is one among the major aim of this study. In general soot are found to consist of organic compounds, resins and majorly carbon. The toxicity of soot is attributed to the chemical composition associated with the core carbon material. They are also found to release in atmosphere thus carrying the toxic materials to longer distance (WHO, 2009). The soot in general which are neglected for their allergic reaction causing respiratory problem and other ailments, the study provides the bioactivity of biosoot and brings its importance in different application. The antibacterial and antifungal activity is attributed to the associated compounds which are of biological origin of source, i.e. the plant material itself. The antibacterial study on *Lantana camara* and *Parthenium hysterophorus* was already reported (Prakash et al., 2012).

This study is first of its kind to evaluate the antifungal and antibacterial impact of biosoot of different plant species which were evaluated individually. The study reveals that the soot of *Parthenium hysterophorus* possess maximum antibacterial and antifungal activity. Thus, the study may pave a way for newer field to evaluate the biosoot against the bacteria causing dental carries, wound and other surface infections. Further, it can also be added as an agent of cosmeceutical materials like face pack, soaps and detergents. Biosoot functioning as an airborne nanomaterial, impact on human beings as an agent of asphyxiation, the ability to cause renal and neural

diseases, their impact in plant and agriculture blocking respiratory and photosynthesis of plants thus affecting ecology and food chain and their role as a nanocarrier functioning as even a war material is highlighted (Udayaprakash et al., 2022). Thus, application of biosoot in pharmaceutical and cosmeceutical industry is recommended after proper evaluation of its toxicity.

CONCLUSION

In general the soot are neglected due to their health impact. In this the biosoot of common weeds, *Calotropis gigantea*, *Lantana camara* and *Parthenium hysterophorus* were evaluated for their antibacterial and antifungal activity. The study aptly demonstrates their ability as successful antimicrobial agents. The usage of biosoot in pharmaceutical and cosmeceutical industry is recommended after proper evaluation of their toxicity. This is the first report evaluating the biosoot for their antimicrobial property.

CONFLICT OF INTEREST

Authors declare that there is no conflict of interest.

REFERENCES

- Arokiyaraj, S, Mariadhas Valan Arasu, Savariar Vincent, Nyayirukannaian Udaya Prakash, Seong Ho Choi, Young-Kyoon Oh, Ki Choon Choi, Kyoung Hoon Kim, Rapid green synthesis of silver nanoparticles from *Chrysanthemum indicum* L and its antibacterial and cytotoxic effects: an in vitro study. *Int J Nanomedicine*, (2014) 9:379-88. doi: 10.2147/IJN.S53546
- Browne EC, Franklin JP, Canagaratna MR, Massoli P, Kirchstetter TW, Worsnop DR, et al. Changes to the chemical composition of soot from heterogeneous oxidation reactions. *J Phys Chem A* (2015) 119(7):1154–63. doi:10.1021/jp511507d
- Cain JP, Gassman PL, Wang H, Laskin A. Micro-FTIR study of soot chemical composition-evidence of aliphatic hydrocarbons on nascent soot surfaces. *Phys Chem Chem Phys* (2010) 12(20):5206–18. doi:10.1039/b924344e
- Canagaratna MR, Onasch TB, Wood EC, Herndon SC, Jayne JT, Cross ES, et al. Evolution of vehicle exhaust particles in the atmosphere. *J Air Waste Manag Assoc* (2010) 60(10):1192–203. doi:10.3155/1047-3289.60.10.1192
- Chuang HC, Jones T, Chen Y, Bell J, Wenger J, BeruBe K. Characterisation of airborne particles and associated organic components produced from incense burning. *Anal Bioanal Chem* (2011) 401(10):3095–102. doi:10.1007/s00216-011-5209-7
- Dong L, Henderson A, Field C. Antimicrobial activity of single-walled carbon nanotubes suspended in different surfactants. *J Nanotechnol.*, (2012), 1–7.
- Glaser B, Dreyer A, Bock M, Fiedler S, Mehring M, Heitmann T. Source apportionment of organic pollutants of a highway-traffic-influenced urban area in Bayreuth (Germany) using biomarker and stable carbon isotope signatures. *Environ Sci Technol* (2005) 39(11):3911–7. doi:10.1021/es050002p
- Goto D. Modeling of black carbon in Asia using a global-to-regional seamless aerosol-transport model. *Environ Pollut* (2014) 195:330–5. doi:10.1016/j.envpol.2014.06.006
- International Agency for Research on Cancer (IARC). Polynuclear aromatic compounds, Part 4, bitumens, coal-tars and derived products, shale-oils and soots. *IARC Monogr. Eval. Carcinog. Risk Chem. Hum.* (1985), 35, 1–247.
- Kamboores MA, Hu S, Yu Y, Sandoval J, Rieger P, Huang SM, et al. Black carbon emissions in gasoline vehicle exhaust: a measurement and instrument comparison. *J Air Waste Manag Assoc* (2013) 63(8):886–901. doi:10.1080/10962247.2013.787130
- Long CM, Nascarella MA, Valberg PA. Carbon black vs. black carbon and other airborne materials containing elemental carbon: physical and chemical distinctions. *Environ Pollut* (2013) 181:271–86. doi:10.1016/j.envpol.2013.06.009
- Prakash, N.U., Bhuvanewari, S., Jahnvi, B., Abhinaya, K., Rajalin, A.G., Kumar, M.P., Sundaraman, G., Elumalai, K., Devipriya, S., Kannan, V. and Sriraman, V., 2012. A study on antibacterial activity of common weeds in northern districts of Tamil Nadu, India. *Research Journal of medicinal plant*, 6(4), pp.341-345.
- Scheepers PT, Bos RP. Combustion of diesel fuel from a toxicological perspective. I. Origin of incomplete combustion products. *Int Arch Occup Environ Health* (1992) 64(3):149–61. doi:10.1007/BF00380904
- Udayaprakash N.K and Bhuvanewari, S. Haze in Chennai: Biosoot cannot be viewed with haziness. *News letter, ENVIS*, (2019) 17(4),1-3.
- Udayaprakash, N.K., SundaraVenkata Prasad, R., Anantha Krishnan, G and Bhuvanewari, S. (2022). Airborne Nanoparticles as a war material, *Proceedings, International Symposium on War Studies*, Ankara, Turkey, 74-82.
- Varghese S, Kuriakose S, Jose S. Antimicrobial activity of carbon nanoparticles isolated from natural sources against pathogenic Gram-negative and Gram-positive bacteria. *J. Nanotechnol.*, (2013), 1–5.
- World Health Organization. *Global Health Risks: Mortality and Burden of Disease Attributable to Select Major Risks*; World Health Organization: Geneva, Switzerland, 2009.