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# An Analytical Observational Study to Determine the Antibacterial and Antioxidant Properties of Dashapushpam, Ten Medicinal Plants

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***Authors' contributions***

*This work was carried out in collaboration between both authors. Both PKP designed the study and the protocol. Author KS wrote the manuscript and manage the literature study. All authors read and approved the final manuscript.*

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## ABSTRACT

A collection of ten revered herbs known with their traditional and therapeutic significance is known as Dashapushpam in the Kerala state of India, and particularly utilized by Keralites on the onsets of the monsoons. The current research aims to showcase the antibacterial and antioxidant properties of these ten herbs. The antibacterial efficacy was assessed against Gram-negative *Escherichia coli* and Gram-positive *Bacillus subtilis* using the agar-gel diffusion method. All extracts exhibited varying degrees of activity against these test organisms, with their effectiveness compared to the standard antibiotic amoxicillin. Additionally, the study evaluated the antioxidant potential of the

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Dashapushpam herbs through the nitric oxide scavenging assay, revealing different levels of antioxidant activity among the plants. These findings support the potential use of these ten herbs in medicinal applications as anti-infective agents.

*Keywords: Dashapushpam; anti-bacterial; antioxidant; diffusion using agargel methods.*

## 1. INTRODUCTION

Microbial infections pose a constant threat to humanity, with their incidence and diversity having increased globally in recent years [1]. Prolonged use of synthetic antibiotics has lead to the development of resistance among infectious microorganisms [2]. This highlights the importance of developing natural drugs from plant extracts, which are considered safer and free from side effects due to their organic origins [3]. Dashapushpam comprises ten different herbs: *Cardiospermum halicacabum, Aerva lanata*, *Emilia sonchifolia, Ipomoea sepiaria*, *Biophytum sensitivum*, *Curculigo orchioides*, *Vernonia cinerea, Cynodon dactylon*, *Eclipta alba and Evolvulus alsinoides* [4]. These herbs possess various medicinal properties, including anti-inflammatory, anti-cancer, immunomodulatory, anti-helminthic, anti-tumor, hepatoprotective, anti-diarrheal, and anti-diabetic [5]. This study aims to explore the antibacterial and antioxidant activities of these plant extracts against both gram-positive and gram-negative bacteria, with antioxidant activity assessed using the nitric oxide scavenging assay [6]. The studies was carried out at the research lab in malappuram district, Kerala.

## 2. THE PHYTOCHEMICAL CONSTITUENTS OF DASHAPUSHPAM

a. *Aerva lanata* – It include alkaloids, flavonoids, methyl grevillate, lupeol, lupeol acetate benzoic acid, β-sitosteryl acetate and tannic acid [7].

b. *Eclipta alba* - wedololactone, demethylwedelolac- tone, desmethyl-wedelolactone-7 glucoside, ecliptal, β-amyrin, luteolin-7-O- glucoside, hentriacontanol, heptacosanol, stigmas-terol [8]

c. *Biophytum sensitivum* - flavonoids, saponins, tannins, terpenes, steroids, amino acids, essential oil, polysaccharides and pectin [9].

d. *Cardiospermum halicacabum* - palmitic acid, oleic acid, stearic acid, linoleic acid, and eicosenoic acid [10].

e. *Curculigo orchioides* - alkaloid lycorine, sterols including sitosterol, sapogenin, and flavone glycoside 5,7- dimethoxy glucopyranoside. Flavonoids include 5,7-dimethoxy glucopyranoside, and fatty acids such as linolenic, palmitic, behenic, arachidic, and oleic acids [11].

f. *Cynodon dactylon* - β- sitosterol, β-carotene, vitamin C, palmitic acid, triterpenoids, arundoin, friedelin, selenium, alkaloids- ergonovine and ergonovinine, Ferulic, syringic, p- coumaric, vanilic, p hydroxybenzoic and o-hyroxyphenyl acetic acids, Cyanogenic hyperoside, Cyanogenic glucoside- triglochinin, furfural, furfural alcohol,phenyl acetaldehyde, acetic acid, phytol, β- ionone; mono and oligosaccharides, lignin [12]

g. *Emilia sonchifolia* - beta-sitosterol, stigmasterol, palmitic acid and honey acid [13]

h. *Ipomoea sepiaria* - alkaloids, saponins, phenols and resin. [14]

i. *Vernonia cinerea* - vernolide-A and vernolide-B, β-amyrin, lupeol and their acetates; and β-sitosterol, stigmasterol, α-spinasterol and phenolic resin [15]

j. *Evolvulus alsinoides* - steroids, reducing sugars, alkaloids, phenolic compounds, saponins, tannins, flavonoids, amino acids, terpenoids and cardioglycosides [16]

## 3. MATERIALS AND METHODS

### 3.1 Anti-Bacterial Assay

#### 3.1.1 Collection of plant materials

The ten important herbs that constitute Dashapushpam were gathered from the different locations, mainly within the district called Malappuram of Kerala state in India. These collected plants were then thoroughly given a tap water wash. After that the moisture was removed

by drying under the shade (temperature between 30°c to 45°c). Then these plants were grinded and converted into a powder form and stored for further use. The plant powder was stored in air tight containers and kept at room temperature. The whole plants are used for the study.

#### 3.1.2 Preparation of the aqueous extracts from the ten herbs

Each of the plants was first cleaned by washing under the tap water, then dried under the shade and powdered separately. A 50 g portion of the sample powder is then mixed with of distilled water (300 ml) and heated for fifteen minutes by mixing continuously. This mixture is then allowed to cool to room temperature for 24 hours. Following this, this sample solution is filtered by using the filter paper (Whatman filter paper No.1with pore size 11µm ) with the aid of a vacuum pump [17]. This filtrate is then concentrated at 40°C and continued until the solvents had completely evaporated, and the resulting sample obtained after concentration was then dissolved using the sterile distilled water [17].

#### 3.1.3 Micro-organisms used for the anti - bacterial assay

The Gram negative bacterium used is *Escherichia coli* and *Bacillus subtilis* was the *Gram* positive organism used in this assay.

#### 3.1.4 Procedure

The antibacterial potential of the each plant extract is assessed using the agargel diffusion technique. Plates with nutrient agar, type 1 (20 ml) were prepared. Both bacterial suspensions 1ml each was inoculated on to the solidified nutrient agar plates. Each Petridish contains three wells of 6 mm in diameter cut out in it. The plant extract was added in to the first well, the positive control (amoxicillin, 30 µg/ml), and the negative control (sterile distilled water) was added to the second and third well respectively. Specifically, 20 µl of 0.5 g/ml of the plant extract is added to one plate containing Gram-positive bacteria and another plate containing the Gram-negative bacteria. These inoculated nutrient agar plates is kept in incubator at 37°C for two to three days. Zones of inhibition were observed around the wells containing the sample and the positive control, and the diameters of these zones were measured [17].

### 3.2 Antioxidant Assay by Nitric Oxide Scavenging Activity

#### 3.2.1 Preparation of plant extract

The leaves of the ten herbs are taken for the study.20 mg of plant sample is dissolved in 2mL of methanol by crushing in a motor and pestle and incubated overnight. The extract was then serially diluted with distilled water to make concentrations of 250$\mu$L (2.5 mg), 500$\mu$L (5mg) and 1000 $\mu$L (10 mg). The same reaction mixture without the extract but the equivalent amount of methanol was served as the positive control [18].

#### 3.2.2 Procedure

The initiation of the reaction was done by adding 2.0ml of sodium nitroprusside (100 mM) of ACS Grade and 0.5 ml of PBS (phosphate-buffered saline, pH 7.4) to 0.5 ml of leaf extracts (50 mg). This mixture is then incubated at 25°C for 30 minutes. Following this, 0.5ml of Griess reagent ACS Grade (composed of 1%sulfanilamide, 2%$H_3PO_4$, and 0.1% naphthylethylene diamine dihydrochloride) was added and the mixture was again incubated for another 30 minutes. Control tubes were prepared without using the leaf extracts. The absorbance is then measured at 546 nm against a reagent blank using a spectrophotometer [18]

The assay was calculated as [18]:

$$\text{Radical scavenging activity} = \frac{\text{Control OD} - \text{Test OD} * 100}{\text{Control OD}}$$

## 4. RESULTS AND DISCUSSION

The anti-microbial assay results assessed by the agargel diffusion method are summarized in Table 1. All the plants constituting the Dashapushpam exhibited varying degrees of anti-bacterial activity against *Escherichia coli* and the *Bacillus subtilis*. The positive control, amoxicillin, produced inhibition zones with diameters of 1.6 cm for *E. coli* and 1.7 cm for *B. subtilis*, while the negative control (distilled water) showed no zones of inhibition. In the case of the individual plant extracts, *Ipomoea sepiaria* demonstrated the highest antibacterial activity, with inhibition zones measuring 1.5 cm in diameter for Gram-negative bacteria and 1.3 cm for Gram-positive bacteria. Conversely, *Evolvulus alsinoides* exhibited the least activity, with inhibition zones of 0.9cm and 0.8cm for Gram-negative and Gram-positive bacteria,

respectively. The antibacterial activity of the other plant extracts fell within these ranges.

The results of the antioxidant assay are presented in Table 2. The ten herbs displayed varying levels of antioxidant activity in the nitric oxide radical scavenging assay. *Aerva lanata* showed the highest antioxidant activity, while *Cardiospermum halicacabum* had the lowest values across all tested concentrations. The remaining plants demonstrated antioxidant activities within this range.

**Table 1. Antibacterial test results**

| Sl no | Name of Plant | Scientific Name | Bacteria | Diameter of Inhibition Zone | | |
|---|---|---|---|---|---|---|
| | | | | Sample (cm) | Positive Control (cm) | Negative Control (cm) |
| 1 | Cheroola | *Aerva laneta* | Gram -ve | 1.4 | 1.6 | 0 |
| | | | Gram +ve | 1 | 1.7 | 0 |
| 2 | Mukkutty | *Biophytum sensitivum* | Gram -ve | 1.1 | 1.6 | 0 |
| | | | Gram +ve | 1.1 | 1.7 | 0 |
| 3 | Valliyuzhinja | *Cardiospermum halicabum* | Gram -ve | 1 | 1.6 | 0 |
| | | | Gram +ve | 1.2 | 1.7 | 0 |
| 4 | Nilappana | *Curculigo orchoid* | Gram -ve | 1 | 1.6 | 0 |
| | | | Gram +ve | 1 | 1.7 | 0 |
| 5 | Karuka | *Cynodon dactlyon* | Gram -ve | 1.1 | 1.6 | 0 |
| | | | Gram +ve | 0.9 | 1.7 | 0 |
| 6 | Kayyunyam | *Eclipta alba* | Gram -ve | 1 | 1.6 | 0 |
| | | | Gram +ve | 1.3 | 1.7 | 0 |
| 7 | Muyal cheviyan | *Emilia sonchifolia* | Gram -ve | 1.3 | 1.6 | 0 |
| | | | Gram +ve | 1.2 | 1.7 | 0 |
| 8 | Vishnu kranthi | *Evolvulus alsinoides* | Gram -ve | 0.9 | 1.6 | 0 |
| | | | Gram +ve | 0.8 | 1.7 | 0 |
| 9 | Thiruthali | *Ipomea sepiaria* | Gram -ve | 1.5 | 1.6 | 0 |
| | | | Gram +ve | 1.3 | 1.7 | 0 |
| 10 | Puvam kurunnel | *Vernonia cineirea* | Gram -ve | 1.1 | 1.6 | 0 |
| | | | Gram +ve | 1.2 | 1.7 | 0 |

**Table 2. Antioxidant Assay-Nitric oxide radical scavenging activity**

| Sl no | Name of Plant | Scientific Name | Percentage of Inhibition | | |
|---|---|---|---|---|---|
| | | | Concentration (250µl) | Concentration (500µl) | Concentration (1000µl) |
| 1 | Cheroola | *Aerva laneta* | 75.4 | 84.5 | 95.3 |
| 2 | Mukkutty | *Biophytum sensitivum* | 20.3 | 26.7 | 32.4 |
| 3 | Valliyuzhinja | *Cardiospermum halicabum* | 2.1 | 3.2 | 5.1 |
| 4 | Nilappana | *Curculigo orchoid* | 65 | 79 | 83.2 |
| 5 | Karuka | *Cynodon dactlyon* | 15 | 18.7 | 26.5 |
| 6 | Kayyunyam | *Eclipta alba* | 42 | 76.1 | 92 |
| 7 | Muyal cheviyan | *Emilia sonchifolia* | 65.2 | 77.4 | 85 |
| 8 | Vishnu kranthi | *Evolvulus alsinoides* | 9 | 12.1 | 19 |
| 9 | Thiruthali | *Ipomea sepiaria* | 9.6 | 15.2 | 20.6 |
| 10 | Puvam kurunnel | *Vernonia cineirea* | 11.6 | 26 | 34.5 |

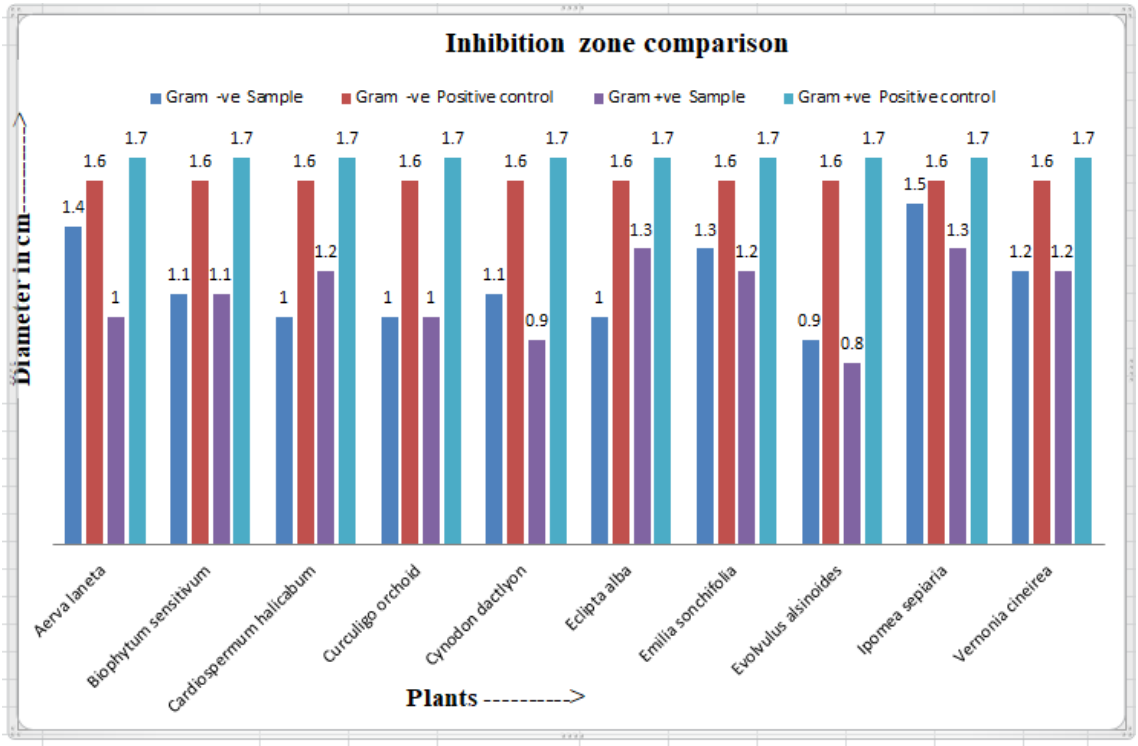


**Fig. 1. Inhibition zone comparison**

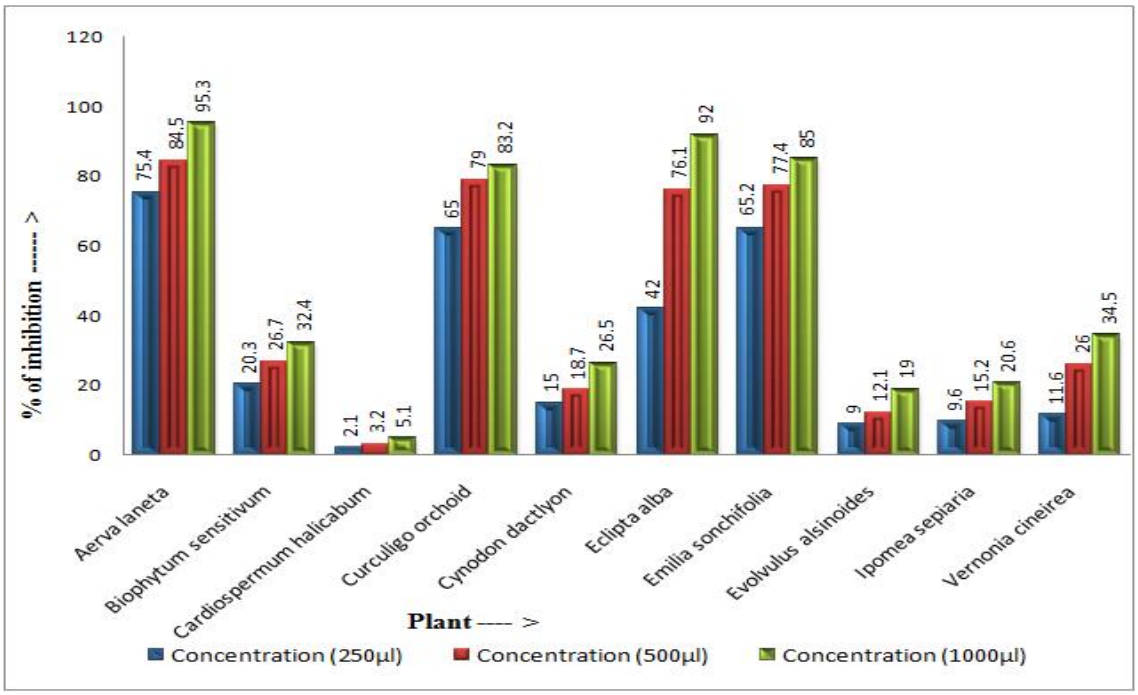


**Fig. 2. Percentage of inhibition**

## 5. CONCLUSION

The results indicate that the ten plant extracts possess significant antibacterial properties. For gram-negative organisms, the extracts demonstrated activity comparable to the positive control antibiotic, amoxicillin. Future studies are necessary to fully explore the potential of these

ten herbs. Investigations including Phytochemical Studies are essential to identify the specific compounds responsible for their anti-microbial potential. The present study supports the usage of Dashapushpam in various pharmaceutical products and underscores the potential of herbal extract as a strong therapeutic agent against the pathogenic microorganisms. Further investigations discovering a natural remedy for the bacterial infections would be a significant advancement for modern medicine.

**DISCLAIMER (ARTIFICIAL INTELLIGENCE)**

We hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during writing or editing of manuscripts.

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## COMPETING INTERESTS

Authors have declared that no competing interests exist.

## REFERENCES

1. Natarajan D, Shivakumar MS, Srinivasan. Antibacterial activity of leaf extract of Biophytum sensitivum", Journal of Pharmaceutical Sciences and Research. 2010;2(11):717.
2. Mandal, Bitasta, Madan, Swati. *Aerva lanata: A Blessing of Mother Nature",* Journal of Pharmacognosy and Phytochemistry. 2015;5:92-101.
3. Arun Raj Gr, Shailaja U, Rao Prasanna N, Ajayan S, The therapeutic potential of ten sacred plants (dashapushpam) of kerala state of southern India. Journal of Ayurveda and Holistic Medicine. 2013;1(3).
4. Payal C, Gurlaganjeet K, Davinder K, Gagan S, Amit C, et al. A review on phytochemistry and biological activities of aerva. Med Aromat Plants. 2015;4 :187.
   DOI: 10.4172/2167-0412.1000187
5. Syed Atif Raza, Shahzad Hussain, Humayun Riaz and Sidra Mahmood, Review of beneficial and remedial aspects of Cardiospermum halicacabum L., Afr. J. Pharm. Pharmacol. 2013;7 (48):3026-3033
6. Anbarasu R, Selvan G, Baskar S, Raja V. Pharmacological potential of silver nanoparticles (AgNPs) derived from *Evolvulus alsinoides*. International Journal of Recent Research and Applied Studies. 2016;36(5):30-38.
7. Goyal M, Pareek A, Nagori BP, Sasmal D. *Aerva lanata: A review on phytochemistry and pharmacological aspects.* Pharmacogn Rev. 2011;5(10): 195-8.
   DOI: 10.4103/0973-7847.91120. PMID: 22279378; PMCID: PMC3263055.
8. Jadhav, Varsh M. Thorat, Salaskar VJ. Chemical composition, pharmacological activities of Eclipta alba., Journal of Pharmacy Research; 2009.
9. Pawar, Anil, Vyawahare, Phytochemical and pharmacological profile of biophytum sensitivum (L) DC., International Journal of Pharmacy and Pharmaceutical Sciences. 2014;18-22.
10. Jeyadevi R, Sivasudha T, Ilavarasi A, Thajuddin N. *"Chemical Constituents and Antimicrobial Activity of Indian Green Leafy Vegetable Cardiospermum halicacabum.* Indian J Microbiol. 2013;53 (2):208-13.
    DOI: 10.1007/s12088-012-0333-4 Epub 2012 Nov 10. PMID: 24426110; PMCID: PMC3626954.
11. Pallishree Bhukta, Santosh Kumar Ranajit, Pratap Kumar Sahu, Deepankar Rath, Phytochemistry and pharmacology of Curculigo orchioides Gaertn: A review, Journal of Applied Pharmaceutical Science. 2023;13(10):083-091.
    Available:http://www.japsonline.com
    DOI: 10.7324/JAPS.2023.135164 ISSN 2231-3354
12. Asthana A. Kumar, A. Dora, J. Gangwar, Smriti. *Pharmacological perspectives of Cynodon dactylon*., Research Journal of Pharmaceutical, Biological and Chemical Sciences. 2012; 1135-1147.
13. Gao JJ et al. Chemical constituents of Emilia sonchifolia L. DC. China Journal of Chinese Materia Medica. 1993;182:102-3 127 .
14. Prasanth B, Aleykutty NA, J. Harindran. *pharmacognostic studies on leaves and stems of ipomoea sepiaria roxb,* International Journal of Pharmaceutical

Sciences and Research. 2018; :3938-3943,
Doi: 10.13040/ijpsr.0975-8232.9(9).
15. Sunita Verma, Phytochemical and pharmacological investigation of Vernonia cinerea: Asteraceae. The Pharma Innovation Journal. 2018;7(6):519-521.
16. Gomathi D, Kalaiselvi M, Ravikumar G, Sophia D, Gopalakrishnan VK, Uma C. Secondary metabolite credentials of *Evolvulus alsinoides* by high performance thin layer chromatography (HPTLC). Journal of Biomed Res. 2012(4):295-302.
DOI: 10.7555/JBR.26.20110128. Epub.
17. Parekh J, Chanda S. *In-vitro Antimicrobial Activities of Extracts of Launaea procumbens Roxb. (Labiateae), Vitis vinifera L. (Vitaceae) and Cyperus rotundus L. (Cyperaceae)*. African Journal of Biomedical Research. 2006;9(2):89-93.
18. Darley-Usmar, Wiseman V, Halliwell HB. Nitric oxide and oxygen radicals a question of balance. FEBS Lett. 1995 309:131-135.



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