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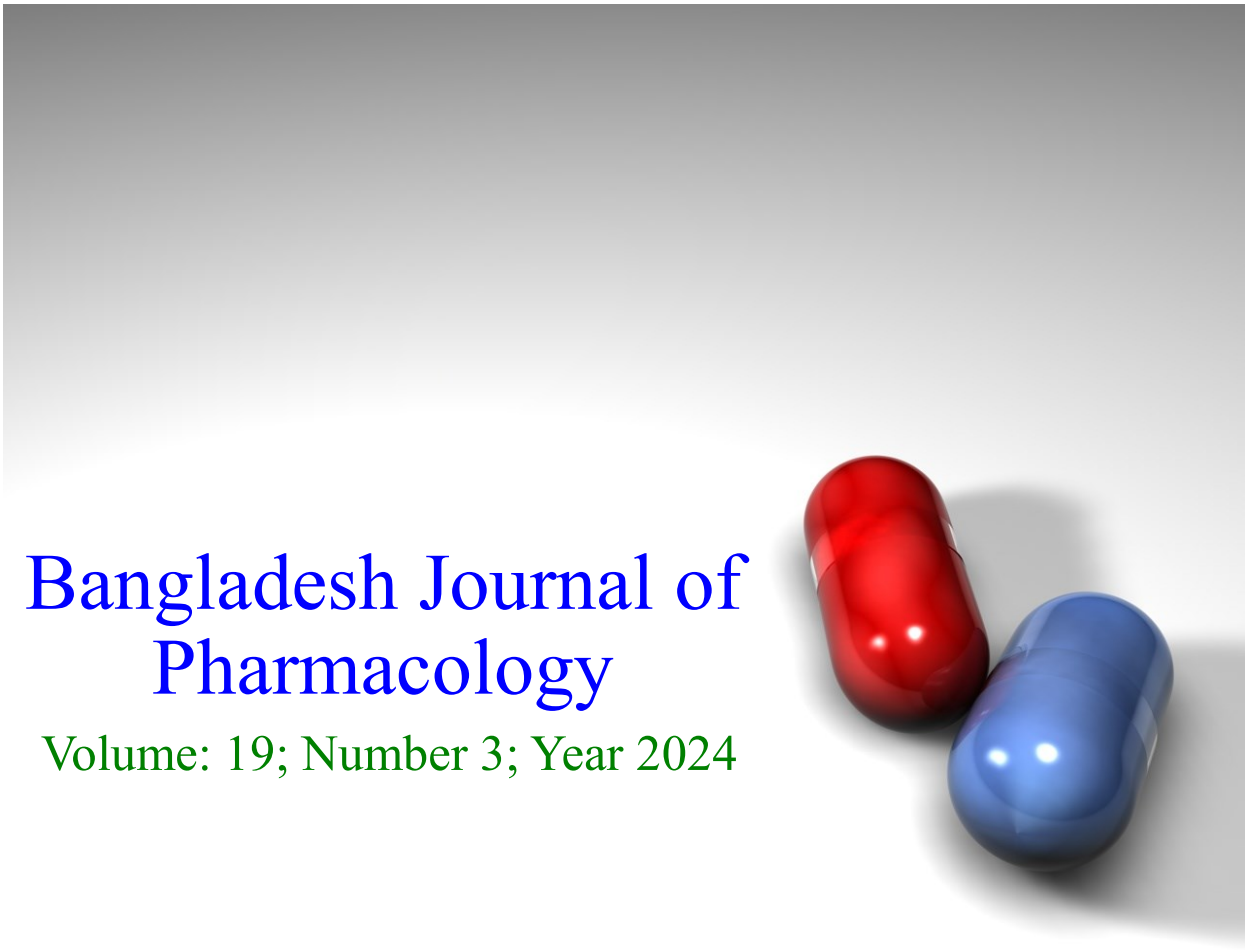

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Letter to the Editor

Antibacterial effect of *Dracaena indivisa* leaf extracts

Dear Editor,

Antibiotic overuse is harmful to the environment, ecosystems, and human well-being. Additionally, it stimulates the emergence of drug-resistant bacteria, a major worldwide concern that is rapidly growing more intense in both hospital and community settings and creating emerging challenges for healthcare providers in terms of morbidity and mortality. To manage resistant bacteria, it is imperative to find and create alternate strategies. One promising strategy is the rational localization of bioactive phytochemicals with antibacterial activity (Masoumian and Zandi, 2017). The antibacterial effect of Dracaena plant species such as *D. colorama* (Sundar et al., 2020), *D. victoria* (Sundar et al., 2020), *D. mahatma* (Saranya et al., 2018) and *D. marginata* (Shiny et al., 2012) have been reported. The current study focused on the investigation of the antibacterial effect of leaf extracts from *D. indivisa* against bacterial pathogens.

Healthy plant parts free of disease were gathered from the Vellore district and rinsed with clean water. These plant parts, likely leaves, were then dried in the air, and ground into a fine powder using a blender. Roughly 10 grams of this powder was soaked in various solvents (around 100 mL each). These soaking solvents included petroleum ether, acetone, methanol, and chloroform. The mixtures were shaken constantly for two days at 37°C and a specific speed (120 rpm). Next, the liquids were separated from the plant material using filter paper. A rotatory evaporator was used to remove the remaining solvents. The concentrated substances left behind were then dissolved in another liquid (DMSO) for further tests.

Qualitative phytochemical screening was carried out to determine the presence or absence of saponins, tannins, anthraquinone glycosides phenols, flavonoids, and terpenoids using standard procedures (Devi et al., 2012). The study assessed the *in vitro* antagonistic efficacy of crude extracts against *Staphylococcus aureus*, *Listeria monocytogenes*, *Klebsilla pneumoniae*, *Salmonella typhi,* and *Pseudomonas aeruginosa* using a well diffusion assay. Briefly, bacterial suspensions were prepared in nutrient broth, standardized, and uniformly spread onto Muller-Hinton agar plates. Wells (6 mm diameter) were created in the agar, and varying concentrations (50, 75, and 100 µg/mL) of the prepared extracts were introduced. Following incubation at 37°C, the diameters of zones of inhibition surrounding the wells were measured. DMSO served as a negative control, while ciprofloxacin antibiotic was a positive control (Santos et al., 2015).

The broth micro-dilution technique was used to determine the minimum inhibitory concentration of the extracts that showed significant antibacterial activity. A 96-well plate was added with approximately 100 µL of Muller-Hinton broth. Seven wells were prepared using a two-fold serial dilution process after 100 µL of the crude extracts were added to the first well at a concentration of 1 mg/mL from the stock solution. The wells' concentrations vary between 50 and 0.78 µg/mL subsequently. After adding 5 µL of bacterial culture, the final two wells with Muller-Hinton broth served as a positive control, while the well-containing bacteria and MHB served as a negative control. For 12 to 18 hours, the plate was incubated at 37°C. Post incubation the culture growth was visually detected. The lowest concentration that inhibits the visible growth of bacteria was measured as MIC value (Santos et al., 2015).

A Perkin Elmer Clarus-680 fitted with a Clarus 600 mass spectrometer and a capillary column (30 m, 0.25 mm ID, 250 µm film thickness) was used to conduct the GC-MS analysis. The initial oven temperature was

Table I

MIC of the crude extracts of *D. indivisa* (µg/mL)

Extracts	*L. monocytogenes*	*P. aeruginosa*	*S. typhi*	*K. pneumoniae*	*S. aureus*
Petroleum ether	12.5	6.25	12.5	6.25	6.25
Acetone	6.25	12.5	3.12	3.12	12.5
Chloroform	1.56	3.12	1.56	1.56	3.12
Methanol	12.5	25	12.5	3.12	25

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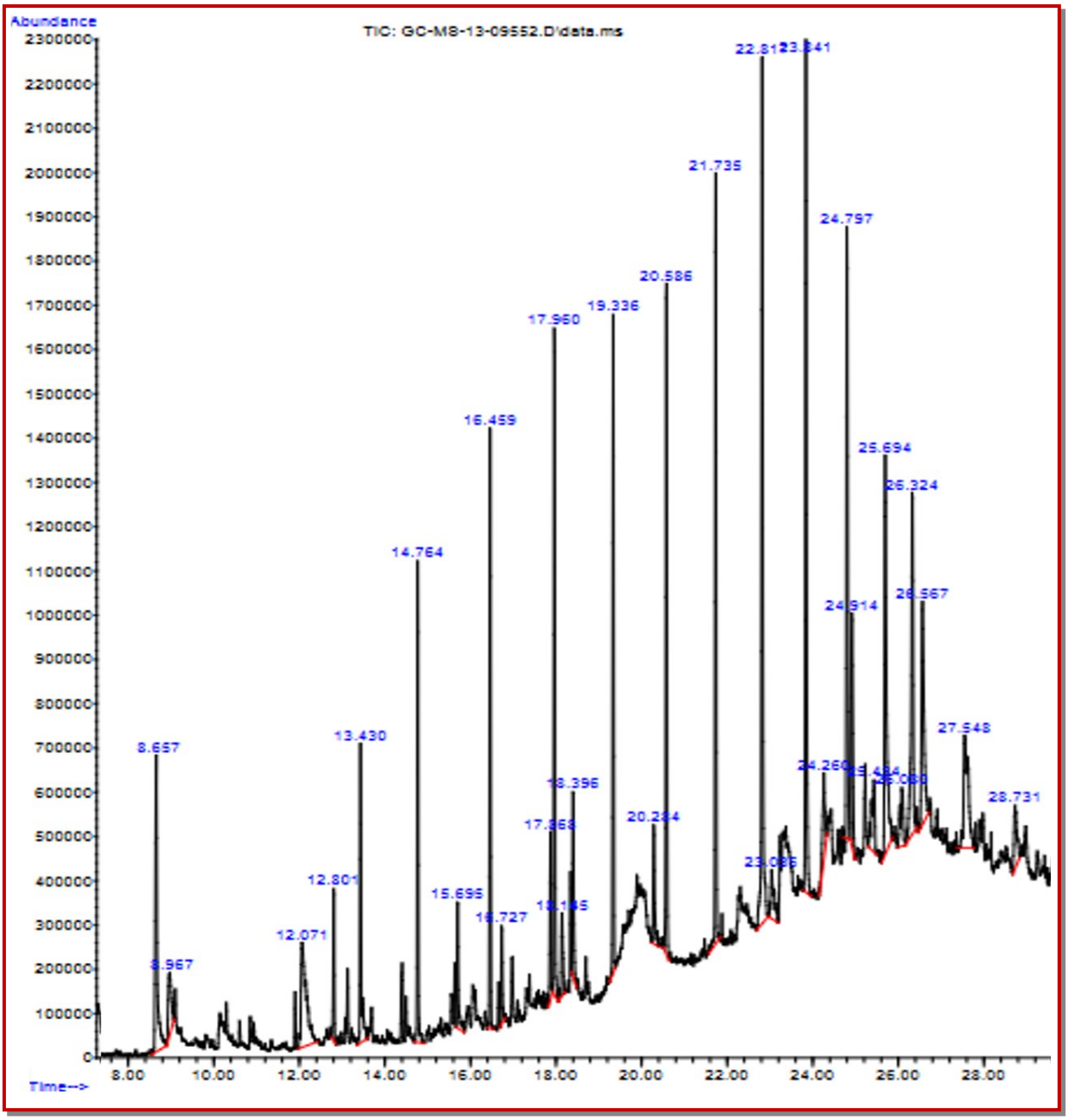

Figure 1: GC-MS analysis of *D. indivisa* chloroform extract

maintained at 60°C for 2 min, then increased to 300°C at a rate of 10°C/min for 6 min. The flow of helium was maintained at 1 mL/min. Temperatures of 240°C were chosen for the mass transfer line and the source. The entire procedure took 25 min to complete. The chemical combinations were compared to the mass spectral profiles in the NIST collection (2008) using Turbo mass software (version 5.4.2) for spectrum analysis (Sundar et al., 2020).

Qualitative phytochemical screening of the leaf extract discovered the existence of phenols, flavonoids, alkaloids, anthraquinone glycosides and terpenoids in all four extracts. The plant extracts studied exhibited varying levels of inhibitory effects against bacterial strains, as measured by the diameter of the growth inhibition zones. The results, showed that all tested bacteria were susceptible to the extracts, with significant differences ($p < 0.05$) in the mean diameters of the inhibition zones. The chloroform crude extract had the highest inhibitory effect against *K. pneumoniae* and *L. monocytogenes*, with a zone of inhibition measuring 23 and 21 mm with 1.56 MIC µg/mL respectively. Followed by methanol extract against *K. pneumoniae* (20 mm) MIC 3.12 µg/mL and *S. aureus* (18.8 mm) MIC 25 µg/mL. The minimum inhibitory concentration of the extracts

ranged between 3.12-1.56 µg/mL for chloroform crude extract (Table I).

The GC-MS analysis of chloroform extract revealed the presence of 30 compounds 2,4-di-tert-butylphenol (RT 13.43), n-hexadecanoic acid (RT 18.39), octadecanoic acid (RT 20.28), benzeneacetamide (RT 12.07), cyclooctasiloxane, hexadecamethyl- (RT 14.76), cyclononasiloxane, octadecamethyl- (RT 16.45), 2,5-dihydroxybenzoic acid, 3TMS derivative (RT 21.73) were the antibacterial compounds present in the chloroform extract which has been reported (Figure 1).

Previous study reported that octadecanoic acid from hydroponics root ethyl acetate extract of *Trigonella foneum graecum* (Sudharsan et al., 2011). *Trichaptum biforme* extract has antagonistic effect on *B. subtillis* and *S. aureus* in which the GC-MS analysis determines that n-hexadecanoic acid key component for its antibacterial property (Yakhlef et al., 2020). Cyclooctasiloxane, octadecamethyl- from ethanol extracts of *Sedum pallidum* exhibited antibacterial activity (Dahpour et al., 2012). 2, 3-dihydroxybenzoic acid was isolated from the fruit extract of *Flacourtia inermis* showed activity against *E. coli, P. aeruginosa, S. aureus* and *K. pneumonia* (George et al., 2011). 2,4-Di-tert-butylphenol (2,4-DTBP) purified from *Streptomyces* sp. KCA1 from *Phyllanthus niruri* exhibited antagonism *E. coli and S. aureus* (Seenivasan et al., 2022). 3TMS derivative is a bioactive component present in *Moringa oleifera* fruit (Shunmugapriya et al., 2017)

D. colorama leaf ethyl acetate extract showed the highest zone of inhibition against *P. aeruginosa* of about 16 mm at 100 µg/mL concentration (Sundar et al., 2020). Leaf extract of *D. victoria* revealed substantial antagonism against *E.coli* with 22 mm ZOI at 10 mg/mL concentration (Saranya et al., 2018).

The present investigation suggests that the chloroform extract from *D. indivisa* holds significant promise for antibacterial capabilities.

Limitations of this study are to validate analytical chemistry research on the purification and identification of bioactive secondary metabolites, as well as their mechanisms in inhibiting the growth of microorganisms.

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Ethical issue: The bacterial cultures used for current research, does not require an ethical approval.

Conflict of interest: The authors declare no competing interests.

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