



Antibacterial and Antifungal Activities and Phytochemical Profile from Different Extracts *Ofricinus communis* L., And *Datura metel* L. Against Selected Pathogens

S. G. Antony Godson*, P.Nandini, S. Sowmiya, R. Abinaya, J. Eniyavaan, And S. Nagul

PG and Research Department of Biotechnology, Hindusthan arts and science college, Nava India, Coimbatore, Tamilnadu, India.

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Corresponding Author Email: dr.s.g.antonygodson@gmail.com

Abstract

The plant extracts are effective in treating pathological effects in our body has given us lavishing interest to look for more inside of it in its molecular level. In the present study, *Ricinus communis* and *Datura metel* are the two weed plants are employed for the extraction of phytochemicals and their effects on two human bacterial pathogens such as *Escherichia coli*, and *Bacillus subtilis*, and two fungal pathogens such as *Aspergillus niger*, and *Aspergillus flavus* assessed. Obviously, a weed implies for their negative effect on environment however, by the way considering a part of them, apparently, as beneficial is what the hypothesis of the study is and hence the disposal weed biomass. In the extraction process, extractants play an important role in making the plant juice more viable and efficient. The methanol/chloroform, and aqueous based extractant are used in the extraction process out of which methanol/chloroform-based extraction has yielded more productions of phytochemicals and have exhibited antimicrobial and antifungal effects as well. The M/C extract of *R. communis* showed the maximum antagonistic activity against *E. coli* species than *B. subtilis* followed by *D. metel*-based extract against the pathogens. The aqueous extracts of the weeds plants against given pathogens also much pronounced for inhibitory activity in most plates as that of *R. communis* with some exceptions that stand significant differences as well. However, the use of these weed plants extracts have proven to have rich in antimicrobial properties and other phytochemical bio-compound which can be of great significance in therapeutic treatments.

Keywords

Methanolic/chloroform and Aqueous extracts, *Ricinus communis*, *Datura metel*, antibacterial and antifungal activity, weed.

INTRODUCTION

Biologically active compounds have been successfully isolated from plant extract upon synthesis of drugs for various disease treatments. Such an isolated plant compounds should be optimized for their constituents to be assessed; for which it requires optimized methods by the way of its handiness in its efficacy for their environmental and economical affordability of processing the raw plant material ^(2,3). Having emerged as a nature's endowment which further processed and made them available via various techniques and methods to analyse their deliberate constituents for contemporary suitability, applicability in various ailments, and further to ensure upon sustainability in view of side effects ⁽¹⁾. The preparation of therapeutic agents from the plant crude extracts since a time immemorial but however, indefinite usage is not guaranteed. Inclusion of drastic increase of the population would make us resort not to abide nature's law but to find remedy in view of utilizing them at the expense of plants meanwhile to propagate them in the lab when nature does not support its matrix further. Folkloric medicine, undeniably, is what people relied upon since many times before any discovery of synthetic compounds being into existence which unarguably been employed for various ailments.

An estimated 3,00,000 – 4,00,000 is number of higher plant species exists around the globe. Of all the new chemical entities in drug formulation, 1073 are approved as drug between the year 1981 till 2010, about 36 % are purely synthetic compounds while, many of them are prepared from medicinal plants; out of which many are capable of becoming lead chemical compounds employed as a template in order to synthesis drugs and further assures in the designing of novel compounds with its enhanced biological attributes against many diseases. Hence, the proven pharmacological effects of plant following which their selection and screening of essential constituents in the drug development has come into effect. Moreover, World Health Organization has given its nod in various instances to promote these novel entities emerged out of plant extracts and forecasted its essentiality in disease management, and its role in future cancer medicine ⁽³⁾. Moreover, bio-compounds which acts as ligand and bind onto the protein target of the concerned pathogen, cancerous cell in such away as could exert the action ⁽²⁾. The modern era has arrived with many tools; which are instrumentations and

techniques, which led us to synthesis new class of synthetic compounds which has attracted more researcher in the recent years. This has engrossed the researchers to exploit further in support of meeting the requirement triggered by the growing populations ⁽⁴⁾. Chemical synthesis of drug amidst the huge expectations has also emerged with a lot limitation; which are mainly of poor adsorption, low biological availability, and severe side effects as well. Hence, it is important to look for alternate source to substitute the existing synthetic compounds to retract once again to ascertain nature's way to make it become a therapeutic agent which consistently, and sustainably help cure diseases across the globe. Obviously, plant-based therapeutics has gained momentum in past few decades with its own acceleration in pace. The one common attributes among plants are to secrete secondary metabolites which are reportedly acting against microbes as microbicides and pesticides etc. Moreover, the extract of weed plant could substitute synthetic antimicrobial agents as it is having highly disastrous effects; whereas, the former not only fights against the disease causing pathogens but also it is free from any side effects as a result of prescribed medications. In due course of industrial era has seen predominant development of Genetically Modified Organisms (GMO) as well, which ended up another degrading phenomenon to nature because of the emergence of resistance organisms to the drugs, hence it is imperative to find appropriate solutions in such a way that could assist humanity to help survive and maintain the livelihood. So, it is high time for the scientific forums to decipher any possible mechanisms that could change the effects of the changing environment along the path which nature might adopts. Due to the prevalence of various side effects and building-up immunity by GMO against synthetic drugs, extracts of bio-compounds of plants are reported to be more effective alternate to them. Progressing further into unravelling the existing complexity in the conventional process, the system needs hybrid methods which promise to give rise more suitability, and sustainability. The thought of combining conventional and non-conventional techniques in an archetypal fashion implies the value added features incorporated to become viable techniques pertaining system, which further ended up a tremendous development in the field of drug discovery ^(1,2). Different extractants which is involved in

extraction process is a prominent step which assures the increase in the yield as well as offers support in process efficiency. Requiring in addition for the standardization of product of herbs for their medicinal value in order of making retain quintessential soluble constituents and to let out of those not in aid of the provided solvents. Re-insisting the reliability of the methods and scale-up further to substantiate the proof-of-concept to deliver yet more and simultaneously to optimize various parameters are the most critical process towards the product intended to accomplish from ⁽²⁾.

Weed as source of medicine:

Ricinus communis L.,

R. communis is a source of medicine in folklore perspectives, which is a fast-growing plant, grows the height of about 6m, and widespread in its distribution across Tamilnadu and India. Castor bean is the common name of the plant and has been an important oilseed crop cultivar since 6000 years ago. Carlos Linnaeus who named the plant and classified based their characteristic features. It is believed to have origin from Tropical Africa. It belongs to Euphorbiaceae family, comprising of about 6300 species ^(2,3). *R. communis* has been acting as a therapeutic agent for about 400 years and employed as herbal medicine for various ailments, disorders and infections. All parts of the plant especially, leaves, bark, roots have been used for medicinal intentions. The plant seed, with vary in size, shape, color, is used as laxative for about 2500 years in Greece and Rome ^(4,5). The oil from the plant has various applications in Ayurveda medicine. The plant also does possess various phytochemicals; especially, lectins has been used for treating tumour. The leaves, bark, and root of the plant have been used to repel mosquito, aphid, whitefly; leaves are believed to increase the yield of milk in cattle which has been reckoned as feed as well, also reported to relieve flatulence in infants ^(3,4,6). Moreover, juice from the leaves could alleviate poisoning effect of narcotics like opium. It is a powerful purgative, used as remedy for toothache. Moreover, the reported work on *R. communis* as feed for earthworm in vermicomposting has channelized an avenue to utilize further as vermicompost, though it is toxic for animals ⁽²⁾.

Datura metel L.,

D. metel is classified under solanaceae family are widespread throughout the world, includes 85 genera

and 2800 species. It is perennial herbaceous plants can reach to a height of about 1.5 m; their leaves are simple, alternate and dark green. Flowers of the plant are large, solitary and trumpet-shaped with sweet fragrance. A hermaphrodite plant, its pollination takes place by insects. The leaves and parts of the plant are reported to have many phytochemicals, which are believed to treat diarrhoea and skin diseases, catarrh, epilepsy, insanity, and hysteria etc. The extracts of plant have a greater potential as antimicrobial and antifungal agent.

Given these perspectives, the present study was conducted to evaluate unique ability of the extraction methods and analyse the bioactive compounds present in *R. communis* and *D. metel* and their potency on test organisms has been addressed.

MATERIALS AND METHODS:

Plant material:

The sampling was done based on the standard techniques and collection of the prescribed plant parts; such as stem, leaves, and root of *R. communis* and *D. metel* was done from in and around and vicinity territory of Hindusthan Arts and Science college, Navakarai, Coimbatore, India. The plant materials were properly washed with tap water, chopped into tiny pieces and finally air dried in shade for some days; which is then later finely ground to powder using electric grinder ^(2,5,6).

Extraction process:

40 grams of plant materials such as leaves, stem, roots have been prepared in the manner of drying as described above. There are two types are extraction process carried out: First is that; methanol: chloroform (M/C) based extraction, which is 1:1 proportion of M/C was prepared for 80 ml by maceration in 7 days. In the same manner for Aqueous extracts, 20 grams of powder was boiled in distilled water (Aq. extract) for about 5 hours ^(2,4). The obtained filtrate following filtration of the concerned plant parts using Whatman filter paper 1 and centrifuged at 10,000 rpm for 15 min thus prepared extract was further concentrated in rotary evaporator at 50 °C. The filtrate is then stored at 4 °C for future use ^(6,7).

Drug and reagents required:

Amphicillin and Griseofulvin was the standard drugs used in the present study as antibacterial and antifungal agents respectively. They were purchased from Precision Scientific co. Coimbatore. The

experiment requiring other chemical analytical reagents for the full-fledged assessment was made available from PG and Research Department of Biotechnology, Hindusthan Arts and Science college, Coimbatore.

Quantitative phytochemical analysis:

Determination of total phenolics in extracts:

The quantification of total phenolic content from different extracts was assessed Spectrophotometrically by Folin–Ciocalteu method. For this, the different extracts in distilled water was prepared for 1mg per ml, in which 0.5 ml of folin-ciocalteu reagent was added with extract quantity of 0.5 ml subsequently, the final volume was made up to 8.5 ml with distilled water. 1.5 ml of sodium carbonate (20 %) was added following the extract which was kept for 10 min at room temperature ^(4,7). The tubes were taken to water bath and kept for 20 min at 40 °C. As the transfer of heat could exert reactions in the solution mixture led to the development of blue colour and thus formed intensity can be ascertained at 755 nm employing UV-visible spectrophotometer (Varian, CARY-300 Bio). The blank was prepared using distilled water. Following the calibration, the quantity of total phenolic content was estimated and was expressed as Gallic acid milligram per gram by interpreting Beer-Lambert's law^(3,8).

Qualitative phytochemical analysis:

Six tubes were arranged and filled with extract prepared from two plants was carried out by employing standard procedure in order identify the following phytochemicals ^(4,6,8).

Alkaloids:

For alkaloid identification, 4 ml of 1 % HCL used with 0.25 ml of plant extract and later it was filtered at slight warm of heat. For one ml of filtrate, six drops of Mayor's reagents and Dragendorff reagent was added one after another. The appearance of greenish or orange precipitate indicates the present alkaloids

Saponins (frothing test):

0.5 ml of the extract was heated to boil in 5 ml of distilled water to produce froth following the vigorous shaking of test tube.

Anthraquinones:

0.5 ml of the extract was boiled with 3 ml of 1 % HCl and was filtered at room temperature. Add 2 ml of benzene and shake well during the process, the occurrence of removal of benzene layer and subsequent addition of few drop of 10 % NH₄OH

caused the formation of pink, violet or red color indicating the presence of anthraquinones.

Terpenoids (Liebermann-Burchard Reaction):

0.5 ml of the extract was added with 2 ml of chloroform and filtered out the residues. Add equal volume of acetic acid along with a drop of concentrated H₂SO₄ for the appearance of blue green ring indicating the presence of terpenoids.

Flavonoids:

0.5 ml of extract was dissolved in petroleum ether to get a defatted residue which was further dissolved in 20 ml of 80 % of ethanol and was filtered out to remove the residues. Take 3 ml of filtrate mixed with 4 ml of 1 % KOH to bring out the appearance of dark yellow color indicates the present of flavonoids.

Tannins:

0.25 ml of extract was heated to boil in 10 ml of distilled water and filtered out the residues.

1 % FeCl₃ was used to get the appearance of brownish green or blue-black indicates the presence of tannins.

Assay for antibacterial and anti-fungal activities:

Determination of zone of inhibition:

Anti-bacterial, and antifungal activities were carried out for M/C and Aq. extracts prepared from *R. communis* and *D. metal* under in-vitro lab condition against two pathogenic bacteria of each one gram positive and gram negative type, and two pathogenic fungi by using agar disk diffusion method. The purified extracts were rendered to dissolve in dimethyl sulfoxide and filtered using sintered glass filter and later it was stored at 4°C^(1,4,8). The zone of inhibition from the provided plating inoculated with pathogenic bacteria, and fungi and positive control of standard antibacterial (Amphicillin) and antifungal (Griseofulvin) were charged with samples for comparison study. Extracts prepared from *R. communis* and *D. metal* plant materials were employed to screen for their antibacterial and antifungal activities against two bacterial species (*E. coli* and *B. subtilis*), and two fungal species (*A. niger* and *A. flavus*). There were three dilutions sets prepared: Which are having different concentration series, such as 25, 100, and 250 µg/ml; since it is expected to demonstrate less or nil antibacterial/antifungal activity below 25 µg/ml of concentration, it is presumed that, the suggested dilution might help to ascertain interpret on comparison with standard. Inoculums of both bacteria and fungi were gathered by picking the distinct

colonies which is around 1mm in size from the 24 hours old culture grown on Luria-Bertani and Nutrient broth and Sabourauds Dextrose agar of *B. subtilis* and *E. coli* and fungi species respectively. Collected colonies of bacteria and fungi are suspended in 5 ml sterile 0.85 % saline so as to adjust the turbidity of the respective bacterial and fungal suspension to 0.5 McFarland reference range. Muller Hintonagar, and modified Muller Hinton agar with 2 % glucose are prepared and seeded with cultures to carryout susceptibility test for bacteria and fungi respectively. Deposit the disks within 24 mm distance apart and load the prescribed concentration of plant extracts in the given spot containing disks along with positive control i.e. respective standard drugs ^(6,7). Experiments which run in duplicates along with control were presented in similar manner. The zone of inhibition triggered by test organisms were evaluated after 18 to 24 hours at 37°C

in bacteria plates whereas in fungal plates, it was after 48 to 96 hours at 28°C. Soon after that, inhibitory zones on agar surface around the disk were measured in such a way as to manifest sensitivity of microorganism to the effects of bioactive components of the plant extracts by measuring the diameter of inhibitory zones. The zone of inhibition which are confined less than 8 mm in diameter were considered not-active-zone against test organism ⁽⁶⁾.

Statistical Analysis:

The experimental data represented as Mean \pm SD, while one-way analysis of variance (ANOVA) followed by Dunnett' test, using SPSS software of 10 versions in order to make a multiple comparison, as well as the data sets were assessed for the significant level from calculated values ($P < 0.05$)^(3,4).

Table 1. Different classes of phytochemicals in different extracts prepared from *R. communis* and *D. metal*

SN	Name of medicinal plant	Sampled plant parts	Total phenolics (mg/g GAE)		Terpenoids		Tannins		Saponins		flavono id		Alkaloids		Anthraquinones	
			M/C \pm STD	Aq \pm STD	M/C	Aq	M/C	Aq	M/C	Aq	M/C	Aq	M/C	Aq	M/C	Aq
	<i>R. communis</i>	stem	11.4 \pm 2.2	07.5 \pm 2.1	+	-	+	+	+	-	+	+	+	+	+	+
		leaves	10.7 \pm 1.6	6.8 \pm 1.5	+	-	+	+	+	+	+	+	+	+	+	+
		root	11.3 \pm 1.6	7.9 \pm 1.4	+	+	+	+	+	+	+	+	+	-	+	-
	<i>D. metal</i>	stem	9.5 \pm 2.4	6.4 \pm 1.7	-	-	+	-	+	+	+	+	+	+	-	-
		leaves	7.8 \pm 1.6	6.6 \pm 2.2	-	-	+	+	+	-	+	+	+	+	-	-
		root	8.4 \pm 2.3	5.6 \pm 2.5	-	-	+	-	+	+	+	+	+	-	-	-

+ present; - absent; **M/C**= Methano chloroform; **Aq**= Aqueous; **STD**=Standard deviation; * = significant ($P < 0.05$)

Table 2 Antibacterial and anti-fungal activities against the test organisms as a function of mean diameters (mm) of zones of inhibition

SN	Plant name	Concentration of sample (µg/disc)	Amphicillin, (Standard)	Antibacterial activity				Griseofulvin (Standard)	Antifungal activity			
				M/C extracts		Aq. extracts			M/C extracts		Aq. extracts	
				<i>Bacillus subtilis</i>	<i>E. coli</i>	<i>Bacillus subtilis</i>	<i>E. coli</i>		<i>A. niger</i>	<i>A. flavus</i>	<i>A. niger</i>	<i>A. flavus</i>
1	<i>R. communis</i>	25	8.5±3.2	12.5±2.2	15±1.4	11.51±2.1	12.8±2.6	11.2±1.3	13.9±1.8	14.4±1.8	11.5±1.3	11.8±1.6
		100	12.1± 1.4	15.1 ± 1.2	17 ± 2.5	13.1 ± 1.2	14 ± 1.5	16±2.3	17.7 ± 1.5	19.0 ± 1.4	14.7 ± 1.5	16.9 ± 1.4
		250	15.8± 1.6	20.1 ± 1.2	21 ± 2.5	19.1 ± 1.2	20 ± 1.5	18.9±1.6	20.5±1.7	21.5±2.3	19.5±1.5	20±2.8
2	<i>D. metal</i>	25	6.8±1.4	11±1.7	13±1.7	10±2.6	12±1.4	8.5±3.2	11±2.4	13.8±1.4	10.2±1.4	12±3.1
		100	14.5±1.3	15±2.1	16±1.4	13±2.1	14±2.9	15.7±1.7	19.9±2.7	20.8±1.8	13±2.8	14.6±2.3
		250	17.4±2.1	19±2.3	20±1.4	18±1.9	18±1.3	20.4±1.4	24.3±2.3	25.4±1.8	16.4±2.3	17.6±1.8

M/C= Methano chloroform; **Aq**= Aqueous; **STD**=Standard deviation; * = significant ($P < 0.05$)

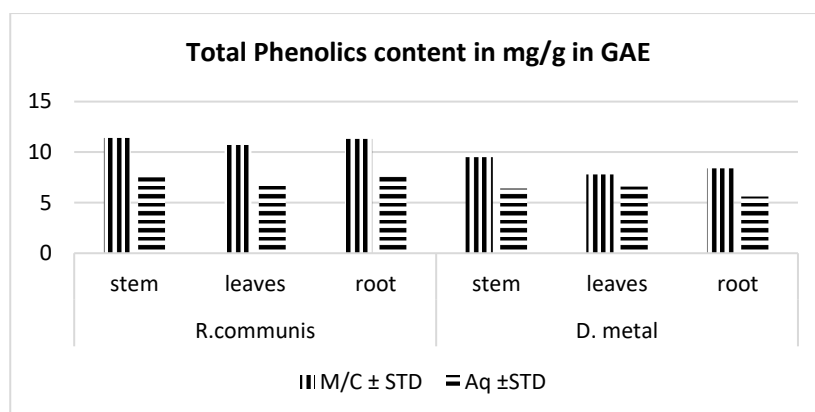


Figure 1. Total phenolic content in extracts prepared from *R. communis* and *D. metal*

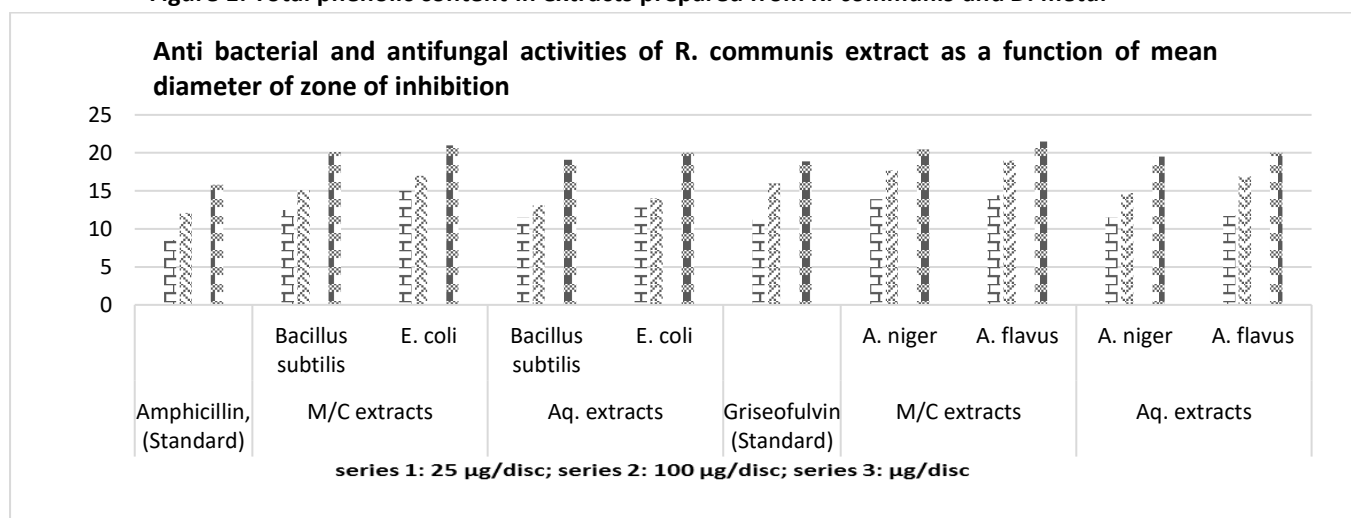


Figure 2. Antibacterial and anti-fungal activities of *R. communis* extract against the test organisms as a function of mean diameters (mm) of zones of inhibition

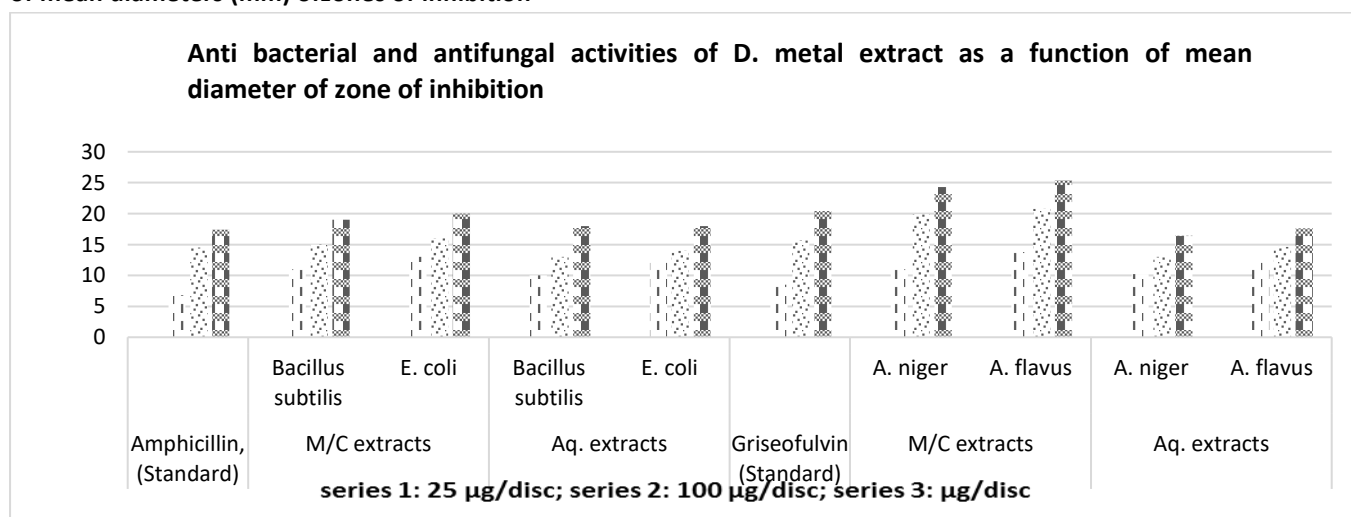


Figure 3. Antibacterial and anti-fungal activities of *D. metal* plant extract against the test organisms as a function of mean diameters (mm) of zones of inhibition

RESULTS AND DISCUSSION:

The antibacterial and antifungal activities of plant extracts in *R. communis* and *D. metal* was evaluated upon their zone of inhibition against the bacterial

strain such as *B. subtilis* and *E. coli*, and fungal strains such as *A. niger* and *A. flavus* in comparison with standard drugs such as antibacterial drug of Ampicillin and antifungal drug of Griseofulvin are

presented in table 2 and figure 2, and 3. Total phenolic content was maximum in the extract prepared from *R. communis* in M/C extract has shown maximum of 11.4 GAE mg/g from stem extract followed by root extract which is 11.3 GAE mg/g over the total phenolic content observed from the extract from *D. metal*. In all cases, Aq. extract have shown less yield in total phenolic content in both instances of plant extracts; which is due to the influence of the less effect of water during extraction however, the result might change with increasing exposure time with extractant (Table 1 and Figure 1)^(3,5). Among the phytochemicals analysed, Terpenes, Tannins, Saponins, Flavonoid, Alkaloid, Anthraquinones were essentially found to be present in *R. communis* plant extract. However, of the plant parts employed for extracting phytochemicals, stem does not have terpenoid and saponins and leaves without terpenoid; whereas, alkaloid and anthraquinones were not extracted from root part of the in the case of Aq. extracts of *R. communis* plant extracts (Table 1)^(5,7). With phytochemicals such as Tannins, Saponins, Flavonoid, and Alkaloid duly present in *D. metal*; Terpenes, and Anthraquinones did have an utter absence; while, Tannins from stem and root, Saponins from leaves, and Alkaloids from root were not noticed in Aq. extract of the plant. Presence of these bioactive compound has given strong indication upon the potential effect such as anti-inflammatory effect. for e.g. Wounds and skin disease^(3,4). The reported work on Tannins and other secondary metabolites from *R. communis* and *D. metal* have served to maximum extend to arrest bleeding and to treat wounds as traditional medicine^(3,5,7). The plant extracts presented varying magnitude of antibacterial potential due to different chemical compositions (Table 2). In general M/C extracts inhibited bacterial and fungal growth more effectively as compared to aq. extracts. The linear increase pattern was observed as the concentration of extract increases the antibacterial and antifungal activities in terms of their zone of inhibition as a function of mean diameter of the zone also increases (Table 2). Comparing the provided drugs against the extracts have given drastic effect on inhibition of bacteria and fungi; in case of *R. communis* extract, M/C extract was observably effective in all concentrations than Aq. Extract of *R. communis*: This is because of M/C extract has yielded relatively good decoction / active compound which caused more efficacy than Aq. counterpart (Figure 2). *E. coli* was

found to be more sensitive in terms of their repelling action against the plant extracts since, Gram negative bacteria is reportedly having a thin peptidoglycan layer against the presence of extra layer in the case of Gram positive bacteria, which could have caused the former to permit antibiotic content more easily than the later into the protective layer through their pores^(3,4). On the other hand, the plates inoculated with fungal species that is *A. flavus* has seen more inhibitory zone compared to *A. niger* irrespective any extraction methods and plant species concerned, which indicates the attributes of the fungal species by means of their disease infestation capacity against one another. The results show that the extracts of *R. communis* were found to be more effective than *D. metal* in both instances of M/C as well as Aq. extracts in both bacterial and fungal species experimented to an extent of 1-2mm diameter. The bacterial zone inhibition is relatively less in diameter than the plates inoculated with fungal strains which is because of manifesting attributes of bacteria upon virulence over fungi is much pronounced in the given circumstances. The presence of flavonoids and other phytochemicals in *R. communis* and *D. metal* have shown wide of range antifungal properties and antibacterial properties as in other reports^(2,3,4). Considering the preciseness over the inoculation of bacterial and fungal isolates and resultant zone inhibition by the influence of antibacterial and antifungal properties of the studied extracts caused the appreciable level of reproducibility from standard deviation between 1.2 to 2.9 mm in almost all cases; except in one case which was 3.1mm is maximum deviated reproducibility found to be occurred in aqueous extracts of *D. metal* by *A. flavus* triggered zone of inhibition (Table 2). One-way ANOVA results revealed that there were significant differences noted between the extracts prepared from *R. communis* and *D. metal* plants in two different extractions method; which is M/C and Aq. extracts, in terms of their total phenolic concentrations, and their inhibitory effect on infectious agents between two pathogenic bacteria such as *B. subtilis* and *E. coli*, and two pathogenic fungi such as *A. niger* and *A. flavus* and among the microorganisms as well with regard to their antibacterial, and antifungal activities respectively with that of their corresponding standard drugs in three different sample concentrations series as a function of mean diameter of zone of inhibition presented in table 1 and table 2. However, in one case,

a significant difference of around 7mm observed between zone of inhibition of M/C and Aq. extracts in *A. flavus* plates; notwithstanding that, an average of one to two-millimetre zone inhibition is noted in almost all cases (Table 2). It is observed from the experimental data that, Aq. extract can work as par to the performance of M/C extract by means of anti-bacterial and anti-fungal activities. The Aq. extract of *D. metal* has demonstrated a significant level differences between Griseofulvin and sample extracts in terms of their inhibitory activity in almost all cases to the magnitude of 5mm diameter, however in one case, the disks with 100, and 250 µg/disc concentration of standard exhibited more efficacy to an extent of 1 to 4 mm against the bioactive compound from the aqueous extracts of the plant (Figure 3). It is to be understood based on the observation that as not in synthetic antibiotics which are mainly prepared from synthetic sources via reproducible manufacturing methods and procedures; on the other hand, the herbal medicine which are the source of plant origin; likely to be putrefiable and subject to degradation and deterioration. Further, the storage of this material does not last longer due to humidity, temperature and light. Still, the extracts which showed inhibitory activities against skin and wound disease-causing pathogens are supported with similar results achieved by other authors^(3,5,8). The efficacy of plant extracts provides a scientific basis of understanding and thus corroborates their traditional applications in the treatment of skin and wound diseases associated with these bacteria and fungi. Hence, the plant extracts employed in this experiment is considered as excellent as it is proved to have broad spectrum of inhibitory effect.

SUMMARY AND CONCLUSION:

The presence of biologically active compounds occurs usually at low concentration due to its origin and attributes of the plants. However, the techniques involved in order to obtain maximum yield with less changes in functional attributes also warranted^(3,4). Number of studies reported several methods of extraction of bioactive compounds in order to ascertain the efficiency of output in various plants extract. In this study, we have put forth two different plants extract with two different extraction techniques which are anew in its own stand upon the productivity in terms of their qualitative and quantitative

assessment of phytochemicals and thus, it is substantiating the need for plant based phytochemicals in lieu of synthetic chemicals as well as it is a proof-of-concept that, *R. communis* and *D. metal* based plant extracts are effective in anti-bacterial and anti-fungal activities as well as in their qualitative and quantitative assessment which are in par with other plant based phytochemicals, and its activities. These results indicate that the extraction by traditional method using M/C extract, and Aqueous extract are one among the viable technique which is substantiating further by experimental data. The M/C and Aq. extracts of *R. communis* and *D. metal* were found to be active on most of the clinically isolated bacteria and fungi compared to standard drugs. The present study assures the claimed uses of leaves in the traditional practices of medicine from the weed plants to treat various infectious disease caused by the bacteria and fungi. Nevertheless, more studies are required to ascertain their molecular level working mechanism and structural elucidation of the crude extracts as the antibacterial and antifungal agents. However, industrial application would require a scale-up studies, mathematical modelling via computer simulation and green extraction method might aid for the process efficiency and process optimization towards sustainable yield.

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