

Studies on Phytochemical, Nutritional Analysis and Screening of in Vitro Biological activities of Meliadubia Leaf Extract

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Abstract:

Meliadubia also called as Maha neem or forest neem is a high value medicinal plant of India, Srilanka, Malaysia, Australia and Angola. The present study was conducted to determine the phytochemical properties, proximate, physicochemical, elemental and antimicrobial activity of the crude extracts of Melia dubia. The extracts were prepared by using Hexane, Chloroform, Ethylacetate, and Ethanol and Aqueous solvents. Qualitative and quantitative screening of the bioactive principles and proximate analysis were carried out using the standard methods. The phytochemical screening indicated the presence of alkaloids, tannins, carbohydrates, phlobatannin, saponins, polyphenols, flavonoids, steroids, terpenoids, triterpenoids, glycosides and proteins. The proximate and Physicochemical analysis values of leaf on dry matter basis contains moisture (73.72%), ash (99%), crude protein (7.25mg), crude lipid (5.27%), vitamin-B (40.80%), Acid insoluble ash (22.68%), Water soluble ash (36.65%), Alcohol soluble extractive (31.80%), Water insoluble extractive (19.65%) and vitamin-C (0.28%). Quantitative analysis of the total Alkaloids ($132 \pm 9.24\text{mg/g}$), Phenols (106.66 ± 7.46), Tannins (46.60 ± 3.26), Saponin ($144 \pm 10.08\text{mg/g}$), Terpenoids ($92 \pm 6.44\text{mg/g}$), and flavonoids was $40 \pm 2.80\text{mg/g}$. The results of mineral composition clearly indicate that Meliadubia leaves contain rich source of mineral such as calcium, magnesium, potassium, iron, manganese and zinc. The leaf extract of Meliadubia solvated by ethanol and methanol extracts were subjected to test their biological activities by few in vitro tests like anti-bacterial activity and anti-fungal activity. They showed the spectrum of inhibition on *E.coli* and *Staphylococcus aureus* for alkaloids and *E.coli* and *K.pneumonia* for flavonoids. Similarly the extracts were found to be more effective against the fungal strains such as *A. flavus* for flavonoids and *A. Niger* for alkaloids. The study revealed the leaves of Melia dubia to be a potential source of nutrition, minerals and useful drugs for human body.

Index Terms: Melia dubia, Phytochemical screening, Proximate analysis, Elemental analysis, Physicochemical analysis, In vitro test.



1 INTRODUCTION :

Herbal medicine and various types of plant-based therapeutic/prophylactic products have been available for centuries and applied in the treatments of diseases throughout history. Worldwide, phytomedicine and herbal medicine are culturally accepted and ubiquitously practiced [1]. The word medicinal plant often leads to the thought of some miraculous and supernatural cures. In India, medicinal plants have played a significant role in the development of our ancient material medica [2]. The increasing price of modern medicine and prevalence of diseases have resulted in the demand for discovery of less expensive and potent drugs. Plants of medicinal characteristics are one of such source. In the present investigation *Meliadubia* have been selected due to their medicinal importance. *Melia dubia* belonging to the family Meliaceae has shown great potential for the best management in terms of secondary plant chemistry [3]. *M. dubia* is also called as a Maha neem or forest neem which is fastest growing tree species of India, Sri Lanka, Malaysia to Australia and Angola. It is found in deciduous forests from

plains to 750m above sea level. It is popularly known as *Melia azedarach* Linn. *Melia dubia* flowers between March-April and fruits from April [4]. It is an indigenous species of tree to India, south east, Asia, Australia where it has been cultivated as a source of fire wood. The wood having good demand from plywood industries"[5,6]". A survey of the literature revealed that *Meliadubia* has lot of Pharmacological activities like Anti Bacterial and fungal activity"(7,8), Anti inflammatory activity (9), Anti oxidant activity(10), Hepatoprotective activity(11) Analgesic activity(12), Anti diabetic activity (13,14), Antifeedant activity (15) Anti-urolithiatic activity(16), Larvicidal activity(17,18), Ovicidal & biopesticidal activity (19). Murugesan et al had reported that the phytochemical components of *Melia dubia* (Cav) showed the presence of unsaturated fatty acids and Phytochemical compounds such as Linolenic acid, Palmitic acid, Caryophyllene, Humulene, Aromadendrene, Probuco, Germacrene-D, Phthalic acid 6-ethyl-3-octyl, Butylated hydroxy toluene. The profile of the *Melia dubia* described in Table-1.

Table-1: Profile of the medicinal plant – Meliadubia Cav

| PROFILE OF THE PLANT-MELIA DUBIA Cav | | |
|--------------------------------------|----------------|-------------------|
| 1 | Kingdom | Plantae |
| 2 | Class | Magnoliopsida |
| 3 | Order | Sapindales |
| 4 | Family | Meliaceae |
| 5 | Genus | Melia |
| 6 | Species | Melia dubia |
| 7 | General name | Melia dubia |
| 8 | English name | Melia composite |
| 9 | Botanical name | Melia dubia cav |
| 10 | Tamil name | Malai vembu |
| 11 | Hindi name | Kala khajur |
| 12 | Marati name | Kuriaput |
| 13 | Guajarati name | Kadukajar |
| 14 | Telungu name | Munnatikaraks |
| 15 | Kannadam name | Hebbevti karibvam |
| 16 | Malayalam name | Malavembu |
| 17 | Oriya name | Batra |

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2 MATERIALS AND METHODS:

2.1 SAMPLE COLLECTION:

The leaves of *Melia dubia* were collected from in and around areas in Trichy district and used for chemical and preliminary analysis. The taxonomic identification of the plant was confirmed by the Rapinat Herbarium, St. Joseph's College (Autonomous) Trichy.

2.2 EXTRACTION:

Fifty grams of the shade-dried and powdered *Meliadubia* leaves were packed in a wide-mouthed bottle. The powdered material is soaked in different solvent namely Hexane, Chloroform, Ethylacetate, Ethanol and water in a bottle. The bottle is closed air-tight and allowed to stand for three days, undisturbed. After three days, the different extracts were collected and were concentrated by distillation process and the concentrated extracts were taken for the phytochemical screening.

2.3 QUALITATIVE ANALYSIS OF PRIMARY AND SECONDARY METABOLITES:

The preliminary Qualitative phytochemical studies were investigated for the detection of different constituents such as alkaloids, flavonoids, tannins, phenolic compounds, steroids, terpenoids by adopting the procedure described by Harborne, (1980), Obadoni and Ochuko (2002), Allen *et al.* (1973), and Schanderl, (1970) respectively.etc [20].

2.4 QUANTITATIVE DETERMINATION OF MELIADUBIA:

1. Determination of total phenols by spectrophotometric method:

Meliadubia powder (2g) was boiled with 50 ml of ether for the extraction of the phenolic component for 15 min. 5 ml of the extract was pipetted into a 50 ml flask, then 10 ml of distilled water was added. 2 ml of ammonium hydroxide solution and 5 ml of concentrated amyl alcohol were also added. The samples were made up to mark and left to

react for 30 min for colour development. This was measured at 505 nm.

2. Determination of Alkaloid by the method of Harborne (1973):

5 g of the *Melia dubia* powder was weighed into a 250 ml beaker and 200 ml of 10% acetic acid in ethanol was added and covered and allowed to stand for 4 h. This was filtered and the extract was concentrated on a water bath to one-quarter of the original volume. Concentrated ammonium hydroxide was added drop wise to the extract until the precipitation was complete. The whole solution was allowed to settle and the precipitate was collected and washed with dilute ammonium hydroxide and then filtered. The residue is the alkaloid, which was dried and weighed.

3. Determination of Tannin by method of Van-Burden and Robinson (1981):

500 mg of the *Meliadubia* powder was weighed into a 50 ml plastic bottle. 50 ml of distilled water was added and shaken for 1 h in a mechanical shaker. This was filtered into a 50 ml volumetric flask and made up to the mark. Then 5 ml of the filtrate was pipetted out into a test tube and mixed with 2 ml of 0.1 M ferric chloride in 0.1N HCl and 0.008 M potassium ferrocyanide. Standard Tannic Acid solutions of range 20-100 mg were treated similarly to the above sample. The absorbance was measured at 120 nm within 10 min.

4. Determination of Saponin by the method of Obadoni and Ochuko (2001):

20 g of *Meliadubia* powder were put into a conical flask and 100 ml of 20% aqueous ethanol were added. The samples were heated over a hot water bath for 4 h with continuous stirring at about 55°C. The mixture was filtered and the residue re-extracted with another 200 ml of 20% ethanol. The combined extracts were reduced to 40 ml over water bath at about 90 °C. The concentrate was transferred into a 250 ml separation funnel and 20 ml of diethyl ether was added and shaken vigorously. The aqueous layer was recovered while the ether layer was discarded. The purification process was repeated. 60 ml of n-butanol was added. The combined n-butanol extracts were washed twice with 10 ml of 5%

aqueous sodium chloride. The remaining solution was heated in a water bath. After evaporation the samples were dried in the oven to a constant.

5. Determination of Flavonoid by the method of Bohm and Kocipai-Abyazan (1994) :

10 g of the *Meliadubia* powder was extracted repeatedly with 100 ml of 80% aqueous methanol at room temperature. The whole solution was filtered. The filtrate was later transferred into a crucible and evaporated into dryness over a water bath and weighed to a constant weight.

6. Estimation of total terpenoid content by standard method (Ferguson, 1956):

1 g of *Meliadubia* powder was taken separately and soaked in alcohol (50ml) for 24 hrs. Then filtered, the filtrate was extracted with petroleum ether (40ml) for 2 hours. This filtrate was transferred into a crucible and evaporated into dryness over a water bath and weighed to a constant weight [21-24].

2.5 ESTIMATION OF PROXIMATE ANALYSIS:

1. Estimation of water content:

A known amount of fresh leaves of *Melia dubia* was taken in pre-weighed porcelain crucible. The sample was kept in an oven at 110°C-120° C for about 2 hours and weighs the crucible. The amount of water was determined by the weight difference.

2. Estimation of Vitamins, Proteins and Lipids:

The primary metabolites like proteins, vitamins were analyzed by standard method using the aqueous extract of melia dubia leaves [25, 26].

2.6 ESTIMATION OF ASH VALUES (OR) PHYSIOCHEMICAL PROPERTIES:

The total ash, acid insoluble ash and water-soluble ash values were determined from air-dried samples using the procedure described in the IP [27].

1. Total ash value:

A known amount of dried sample was taken in a pre-weighed porcelain crucible. Burn the dry sample in the burner till the sample become ash. Weight the ash with crucible. The difference

between the ash with crucible and the sample weight will give the amount of ash [28].

2. Acid insoluble ash:

Ash obtained from the total ash was boiled with 25ml of 2N HCl for a few minutes and Filtered. The filter paper was transferred into a silica crucible. Incinerated at 450°C in a muffle furnace until free from carbon. The crucible was cooled and weighed. Percentage of acid insoluble ash was calculated with reference to air-dried substance.

3. Water soluble ash:

Ash obtained from the total ash was boiled with 25 ml of distilled water for a few minutes. And filtered through an ash less filter paper. The filter paper was transferred into a tarred silica crucible. Incinerated at 450°C in a muffle furnace until free from carbon. The crucible was cooled and weighed. Percentage of water-soluble ash was calculated with reference to air-dried substance.

4. Determination of extractive values:

Extracting values are useful for determining of crude drugs and it gives an idea about the nature of the chemical constituents present. The solvent used for the extraction should be in position to dissolve appropriate quantities of desired substances [28, 29].

5. Determination of alcohol soluble extractive value:

About 5gms of air dried coarse powdered drug was weighed. And macerated with 100ml of 90%alcohol in a closed flask for 24 hours, shaking frequently during the first 6 hrs & these allowed standing for 18 hrs. There after it was filtered rapidly taking precautions against loss of the solvent.25 ml of the filtrate was evaporated to dryness in a tarred flat bottomed swallowed dish, dried at 105°C and weighed. The percentage of the alcohol soluble extractive values was calculated with reference to the air-dried drug.

6. Determination of water soluble extractive value:

About 5gm of air-dried powdered drug was taken & macerated with 100 ml of chloroform water in a

closed flask for 24 hrs shaking frequently during the first 6 hrs and then allowed to stand for 18 hrs. Thereafter, it was filtered rapidly taking precautions against loss of the solvent. 25 ml of the filtrate was evaporated to dryness in a tarred flat bottomed shallow dish, dried at 105°C and weighed. The percentage of the water soluble extractive value was calculated with reference to the air-dried drug.

2.7. MINERAL ANALYSIS:

1. Estimation of Minerals [30-36] :

Ash was estimated using Gravimetric method by ASTM (1988). Organic Carbon estimated by the method of Walkey and Black (1934). Total Nitrogen estimated by the method of Micro Kjeldhal (Bremner, 1965). Sulfur was measured with Gravimetric method (Ali Ehyaei, 1997). Total Phosphorous by Pemberton (1927) method. Total Potassium, Total Sodium, Total Calcium, Total Magnesium were analyzed by Stanford and English method (1949). Total Zinc, Total Copper, Total Iron, Total Manganese, Total Boron, Total Molybdenum were analyzed by using Atomic Absorption Spectroscopy (Solar-AAS2-UK made).

2.8. ASSAY OF ANTIMICROBIAL ACTIVITY:

1. Microbial inoculum preparation:

The nutrient broth was prepared, and then the identified bacterial colonies were inoculated into the broth culture and used for antimicrobial activity.

2. Kirby Bauer agar disc diffusion assay:

The nutrient agar medium was prepared and sterilized by autoclaving at 121°C 15 lbs pressure for 15 minutes then aseptically poured the medium into the sterile petriplates and allowed to solidify, The Bacterial broth culture was swabbed on each petriplates using a sterile buds. Then wells were made by well cutter. The organic solvent extracts of leaves were added to each well aseptically. This procedure was repeated for each Petri plates then the petriplates were incubated at 37°C for 24 hrs.

After incubation the plates were observed for the zone of inhibition.

RESULT AND DISCUSSION:

2.3. Phytochemical screening of *Meliadubia*:

The phytochemical screening revealed that the presence of alkaloids, carbohydrates, proteins, phenolic compounds and flavonoids in all the fractions except in hexane and the results listed in the table-2. But a positive result was found only with Ethanolic and Ethylacetate fraction for phlobatannin. The result of phytochemical screening shows that most of the phytoconstituent present in ethanol and chloroform extract compare to other extract.

Table-2: Phytochemical screening of the plant *Meliadubia*:

| S.No. | Phytochemicals | S1 | S2 | S3 | S4 | S5 |
|-------|----------------|----|----|----|----|----|
| 1 | Tannin | - | + | + | + | + |
| 2 | Phlobatannin | - | - | - | + | - |
| 3 | Saponin | - | + | + | + | + |
| 4 | Flavonoids | - | + | + | + | + |
| 5 | Steroids | + | + | - | + | - |
| 6 | Terpenoids | - | + | + | + | - |
| 7 | Triterpenoids | - | - | - | + | - |
| 8 | Alkaloid | - | + | + | + | + |
| 9 | Carbohydrates | + | + | + | + | + |
| 10 | Polyphenols | - | - | - | + | + |
| 11 | Glycosides | - | + | - | - | + |
| 12 | Anthrocyanin | - | - | - | - | - |
| 13 | Proteins | + | + | + | + | + |

S1 – Hexane, S2 – Chloroform, S3 - Ethyl acetate, S4 – Ethanol, S5 – Water.

(+) – Presence of phytoconstituent, (-) - Absence of phytoconstituent

2.4. QUANTITATIVE ANALYSIS OF *Melia dubia* LEAVES:

Ethanol extract of leaves of *Meliadubia* were subjected to quantitative estimation of alkaloids, flavonoid, saponins, tannins, terpenoids and

Phenols revealed the presence of significantly high concentration of saponin and flavonoid compounds and significantly low concentration of alkaloid compare to all other secondary metabolites. The results are shown in the table- 3.

Table 3: Quantitative determination of *Meliadubia*

| S.No | Name of the chemical constituents | Results (mg/gm) |
|------|-----------------------------------|-----------------|
| 1 | Phenols | 106.66 ± 7.46 |
| 2 | Flavonoids | 40 ± 2.80 |
| 3 | Saponins | 144 ± 10.08 |
| 4 | Tannins | 46.60 ± 3.26 |
| 5 | Alkaloids | 132 ± 9.24 |
| 6 | Terpenoids | 92 ± 6.44 |

Values are expressed as Mean ± SD for triplicates

2.5. PROXIMATE ANALYSIS OF *MELIADUBIA* LEAVES EXTRACT:

The proximate analysis values of leaf on dry matter basis in the present study were found to be moisture (73.72%), ash (99%), crude protein

(7.25mg), crude lipid (5.27%), vitamin-B (40.80%) and vitamin-C (0.28%). Proximate analysis result shown in the table-4.

Table-4: Proximate analysis of *Melia Dubia*

| S.No. | Constituents | <i>Meliadubia</i> composition |
|-------|---------------|-------------------------------|
| 1 | Moisture | 73.22% |
| 2 | Crude Protein | 7.25mg |
| 3 | Lipid | 5.27% |
| 4 | Vitamin-B | 40.8% |
| 5 | Vitamin-C | 0.28% |

2.6. Estimation of Ash values:

The result of estimation of ash values shows that the amount of ash (99%), acid insoluble ash

(22.68%), Alcohol soluble extractive (31.8%), Water soluble ash (36.65) and Water insoluble extractive (19.65%) present in the 5g leaves of *Meliadubia* as the results shown in the table-5.

Table-5: Ash value analysis of Melia Dubia

| S.NO | Ash Values | Amounts in Compositions |
|------|----------------------------|-------------------------|
| 1 | Ash | 99% |
| 2 | Acid insoluble ash | 22.68% |
| 3 | Water soluble ash | 36.65% |
| 4 | Alcohol soluble extractive | 31.8% |
| 5 | Water insoluble extractive | 19.65% |

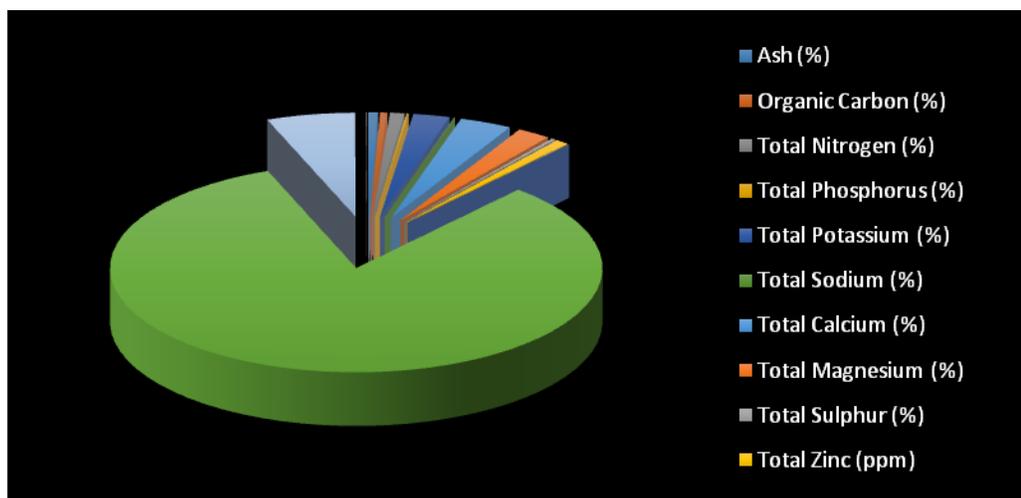
2.7. Estimation of minerals:

The results of mineral composition clearly indicate that *Meliadubia* leaves contain rich source of mineral elements. This becomes important when the usefulness of such mineral like phosphorus, calcium, magnesium, potassium, iron and zinc in the body considered. The zinc content could mean that the plants can play a valuable role in the management of diabetes, which results from insulin malfunctioning. Zinc is essential for the production of insulin, a hormone and carbonic anhydrase, an enzyme in the body. Iron is a component of hemoglobin. It helps oxygen transport. Iron together with hemoglobin and ferrodizin plays important role in man’s metabolism. However, the lower sodium content of *Meliadubia* might be an added advantage due to the direct relationship of sodium intake with hypertension in human, as the results shown in the table-6 and figure-1 shows that the graphical pie representation of mineral compositions.

Table 6: Mineral composition of Meliadubia leaves:

| Sl.No | Name of the parameter | SAMPLE DETAILS |
|-------|------------------------|----------------|
| 1. | Ash (%) | 1.22 |
| 2. | Organic Carbon (%) | 0.89 |
| 3. | Total Nitrogen (%) | 1.82 |
| 4. | Total Phosphorus (%) | 0.26 |
| 5. | Total Potassium (%) | 4.59 |
| 6. | Total Sodium (%) | 0.11 |
| 7. | Total Calcium (%) | 6.59 |
| 8. | Total Magnesium (%) | 4.25 |
| 9. | Total Sulphur (%) | 0.52 |
| 10. | Total Zinc (ppm) | 1.69 |
| 11. | Total Copper (ppm) | 0.06 |
| 12. | Total Iron (ppm) | 159.32 |
| 13. | Total Manganese (ppm) | 11.20 |
| 14. | Total Boron (ppm) | 0.05 |
| 15. | Total Molybdenum (ppm) | 0.02 |

Picture-1: Mineral composition of *Meliadubia* in graphical pie representation



2.8. Antibacterial screening of *Meliadubia* leaves:

Antibacterial activity of the Ethanolic and Methanolic extract of *Meliadubia* was tested against four clinically important bacterial strains namely *Vibrio cholerae*, *Escherichia coli*, *staphylococcus aureus*

and *Klebsiella pneumoniae* following the procedure of sondi *et al.* which showed promising antibacterial activity against all the pathogens. The results of anti bacterial activity of methanolic and Ethanolic extracts of *Meliadubia* shows in the Table 7 & 8 and the image shown in the picture-2

Table – 7: Antibacterial activity of Methanolic extract of *Meliadubia* leaves:

| Pathogen used | Methanol Extract added and Zone of inhibition (mm/ml) | | | | |
|------------------------------|---|-------|-------|--------|---------|
| | 25 µl | 50 µl | 75 µl | 100 µl | Control |
| <i>Staphylococcus aureus</i> | 15 | 17 | 20 | 23 | 18 |
| <i>E.coli</i> | 18 | 22 | 28 | 30 | 13 |
| <i>Klebsiellapneumoniae</i> | 15 | 20 | 22 | 25 | 25 |
| <i>Vibrio cholera</i> | 12 | 17 | 20 | 22 | 18 |

Table – 8: Antibacterial activity of Ethanolic extract of *Meliadubia* leaves:

| Pathogen used | Ethanol Extract added and Zone of inhibition (mm/ml) | | | | |
|------------------------------|--|-------|-------|--------|---------|
| | 25 µl | 50 µl | 75 µl | 100 µl | Control |
| <i>Staphylococcus aureus</i> | 15 | 17 | 18 | 20 | 20 |
| <i>E.coli</i> | 16 | 18 | 22 | 32 | 15 |
| <i>Klebsiellapneumoniae</i> | 12 | 15 | 17 | 20 | 20 |
| <i>Vibrio cholera</i> | 15 | 17 | 20 | 23 | 25 |

2.8. Antifungal screening of *Meliadubia* leaves:

Antifungal activity of the ethanolic and methanolic extract of *Meliadubia* was tested against four clinically important fungal strains namely *Candida albicans*, *A.Niger*, *A.flavus* and *A.fumigatus* by following the procedure of sondiet *al.* which showed promising antifungal activity against all the pathogens . The results of anti fungal activity of methanolic and Ethanolic extracts of *Meliadubia* shows in the Table 9 & 10 and the image shown in the picture-2.

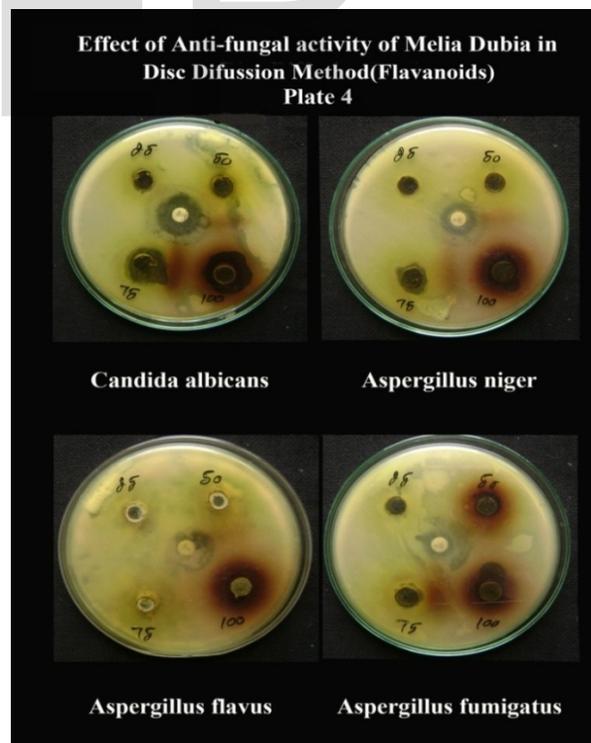
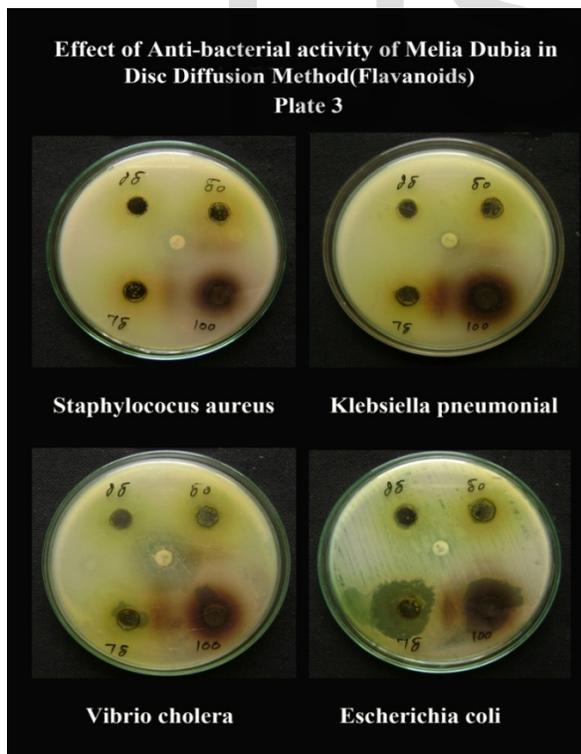
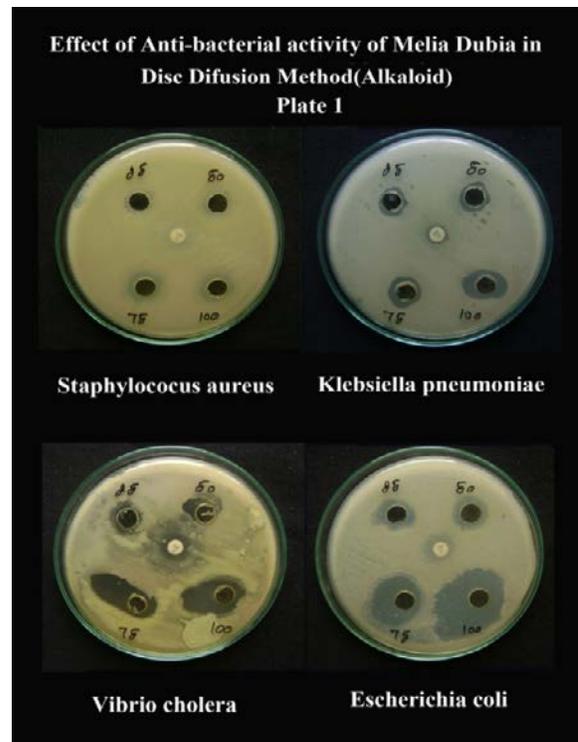
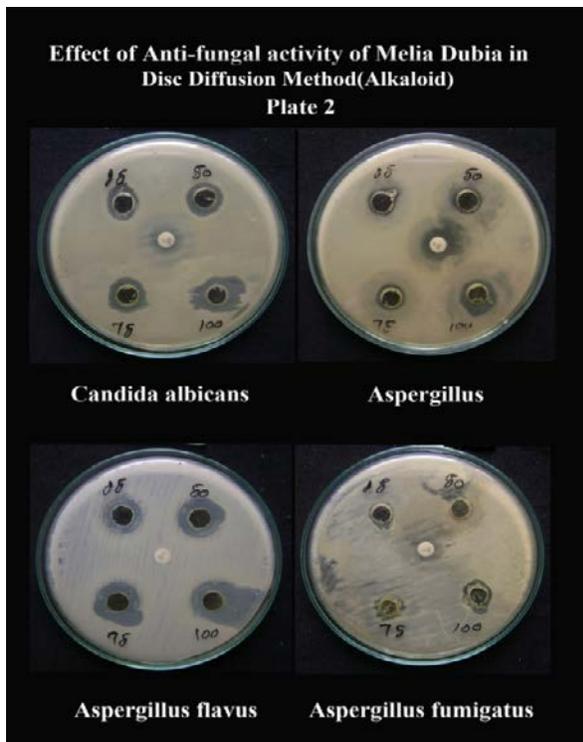
Table-9: Antifungal activity of Methanolic extract of *Meliadubia* leaves:

| Pathogens used | Methanol Extract added and Zone of inhibition (mm/ml) | | | | |
|-------------------------|---|-------|-------|-------|---------|
| | 25 µl | 50 µl | 75 µl | 100µl | Control |
| <i>Candida albicans</i> | 13 | 15 | 17 | 19 | 22 |
| <i>A.niger</i> | 15 | 20 | 25 | 29 | 25 |
| <i>A.flavus</i> | 12 | 15 | 18 | 20 | 22 |
| <i>A.fumigatus</i> | 12 | 14 | 15 | 20 | 20 |

Table-10: Antifungal activity of Ethanolic extract of *Meliadubia* leaves:

| Pathogens used | Ethanol Extract added and Zone of inhibition (mm/ml) | | | | |
|-------------------------|--|-------|-------|-------|---------|
| | 25 µl | 50 µl | 75 µl | 100µl | Control |
| <i>Candida albicans</i> | 12 | 15 | 18 | 20 | 25 |
| <i>A.niger</i> | 10 | 13 | 15 | 22 | 23 |
| <i>A.flavus</i> | 13 | 20 | 22 | 25 | 25 |
| <i>A.fumigatus</i> | 11 | 15 | 19 | 25 | 21 |

Images of Anti Bacterial and Fungal activities of Methanolic and Ethanolic Extracts of *Meliadubia* Leaves:



CONCLUSION:

With the increasing interest and so many promising drug candidates in the current development pipeline that are of natural origin, and with the lessening of technical drawbacks associated with natural product research, there are better opportunities to explore the biological activity of previously inaccessible sources of natural products. In addition, the increasing acceptance that the chemical diversity of natural products is well suited to provide the core scaffolds for future drugs will be further useful in the development of novel natural products and chemical libraries based on natural products in drug discovery campaigns.

Preliminary phytochemical analysis of the leaves indicates the presence of biologically active compounds such as simple phenols, terpenoids, flavonoids and anthraquinones which can account for antimicrobial activity of the above medicinal plant. The proximate and physicochemical analysis values of leaf on dry matter basis contain moisture, ash, crude protein, crude lipid, ash values, extractive values and vitamins by quantitatively. The results of mineral composition clearly indicate that *Meliadubia* leaves contain rich source of mineral elements. This becomes important when the usefulness of such mineral like phosphorus, calcium, magnesium, potassium, iron and zinc in the body considered.

Finally, one can conclude that from the results of the nutrient and mineral composition *Meliadubia* can be of immense use in phytomedicine and can be included in health care delivery system particularly in the developing economics. It can be concluded from this reports that the extracts from the leaves of *Meliadubia* revealed the presence of phytochemical will be supports antimicrobial agent.

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