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CHARACTERIZATION AND ANALYSIS OF BIOACTIVE OF *DIGERA ARVENSIS* BY FTIR

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ABSTRACT: The present study is focused on exploring the chemical components of the leaves, stem, root, and seeds of potential weed *Digera arvensis*. The plant has high nutritional value and therapeutic significance with prospective pharmacological activities. The various parts of the plants, i.e., leaves, stems, roots and seeds, were extracted using polar solvent ethanol. The preliminary phytochemical investigation of ethanolic extract of the plant parts showed the presence of primary and secondary metabolites like carbohydrates, reducing sugars, proteins, polyphenols, glycosides, alkaloids, tannins, flavonoids and terpenoids. The various spectroscopic methods reveal the presence of phytoconstituents, while the FTIR spectroscopic method was carried out on the ethanolic extracts of all the parts of *Digera arvensis* the confirming the presence of functional groups of the phytoconstituents. The FTIR analysis confirmed the presence of O-H, C-O, N-H, C-C, C-H, CO-O-CO and CH₃ functional groups of the polyphenolic compounds.

INTRODUCTION: *Digera arvensis* is an annual herb; it is growing up to 20-70 cm tall. It belongs to the family Amaranthaceae. It is a potential weed growing in the wastelands. It has been used for thousands of years traditionally in many areas as food. The leaves and shoots, which are preferably young, are used as vegetables locally ¹. It is a popularly cooked vegetable among the tribes of Kenya in the coastal region. In India, the curries are made using leaves, or the entire plant is boiled in water and used as food. They are medicinally used internally against digestive system disorders.

The seeds and flowers are used to treat urinary disorders, coolant, astringent ^{2,3}. *Digera arvensis* is native to Northeast Tropical Africa, Ethiopia and East Tropical Africa, and Western Asia, especially in India's eastern and northern provinces. In India, it is widespread in Andhra Pradesh, Rajasthan, and Maharashtra ⁴. It is commonly seen after rains.

Digera arvensis is known by different names in the world. It is commonly known as false amaranth. In India, specifically in Hindi or Bengali is typically called chanchali, lahsuva or latmahuria, in Kannada language chenchalisoppu, kankalisoppu or gorajepalle, in Punjabi leswa or tandla and in Telugu called as goraji playa or chenchalicet, getan or kunjara in Marathi, toyaKeeri in Tamil, aranya, aranyavastuka, kunanjara, or kuranjara in Sanskrit language ⁴. The botanical features of the plants are that the plants' size varies from 20cm to 70cm. Leaves are entire deltoid-ovate, blade 3-7 cm long,

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1.8-3.5 cm wide, acute or acuminate apex, leaf stalks are long up to 5 cm, the base is narrowed and the tip pointed. The stem of *Digera arvensis* are found simple or branched from the base, nearly hairless. Flowers are borne on slender spike-like racemes, which can be as large as 14 cm long. The stamens have lollipop hairs and therefore attract a variety of pollinators, especially flies, but the flowers are also capable of auto-pollination and the wind or rain transports the seeds. Flowers are white mixed with pink to carmine or red. Fruits are subglobose, slightly compressed, 2-2.5 mm, bluntly ribbed along each side, surmounted by a thick rim. Various plant extract fractions indicated the presence of alkaloids, flavonoids, terpenoids,

saponins, coumarins, tannins, cardiac glycosides, and anthraquinones⁵. The extracts of *Digera muricata* has many reported pharmacological activities like nephroprotective agents in carbon tetrachloride-induced nephrotoxicity⁶, antioxidant⁷, antimicrobial^{8, 9}, antidiabetic^{10, 11}, hepatoprotective¹², analgesic¹³. The literature review disclosed the presence of the active principles of the *Digera arvensis*. Hence, the present work was undertaken to systematically investigate and identify the phytochemical bioactives of the ethanolic extract of leaves, stem, and root and seed of *Digera muricata* using FTIR as the analytical tool.



FIG. 1: DIGERA ARVENSIS

MATERIALS AND METHODS: The research was carried out in R R College of Pharmacy, Rajiv Gandhi University of Health Sciences, Karnataka, Bangalore -560090.

1.1. Collection of Plant Material: The Plant material was collected from the Chitradurga district of Karnataka, India, during the winter season in the cultivation fields. Plant parts were allowed to dry immediately after picking. The plant specimen was identified by an expert, Dr. Noorunnisa Begum S. Associate Professor, Centre for Conservation of Natural Resources, Transdisciplinary University, Bangalore, Karnataka, India. The authentication numbers are FRLHT Acc. No. 5584, 5585, 5586, 5587, and 5588.

1.2. Preparation of the Extract: The whole plants were collected, and leaves, stems, roots and seeds

were separated. The separated parts were shade dried for a week grounded into powder using the electric mill. Passed through sieve no. 44. The coarse powder of the various parts was placed into the different maceration chambers, and the menstruum was poured into the drug material and covered completely, kept for 3 days, and shaken periodically. The extract is collected on the last day and filtered. The ethanolic extracts were concentrated under reduced pressure below 40 °C further used for phytochemical screening^{14, 15, 16}.

1.3: Preliminary Phytochemical Screening: The preliminary phytochemical testing of the *Digera arvensis* ethanolic extracts was performed as per the standard reported methods to detect the various classes of phytoconstituents such as carbohydrates, reducing sugar, alkaloids, glycosides, phenolic

compounds, flavonoids, proteins, saponins, lipids, steroids and tannins to ensure the presence of the chemical constituents^{17, 18}.

I. Test for Carbohydrates:

Molisch's test: To 2-3ml of extract, a few drops of α -naphthol solution in alcohol were added, shaken, and concentrated sulphuric acid was added from the side of the test tube. It was observed for the violet ring at the junction of two liquids.

II. Tests for Glycosides:

Keller Kiliani test: Alcoholic extract of the drug is mixed with an equal volume of water and 0.5 ml of lead acetate solution. Shaken and filtered to the filtrate, add an equal volume of chloroform.

The chloroform layer is evaporated to dryness, and the residue is dissolved in 3 ml of Glacial acetic acid. To this ferric chloride solution was added, and sulphuric acid was added; this showed the bluish-green color.

III. Test for Sterols:

Liebermann-Burchard's, reaction: Mixed 2ml of extract with chloroform. Added 1-2 ml of acetic anhydride and 2 drops of concentrated sulphuric acid from the side of the test tube. Observed the first red then blue and finally green.

IV. Test for Sugars:

Fehling's, test: 1ml of Fehling A and 1ml of Fehling B solutions was mixed and boiled for 1 min. equal volume of test solution was added and heated in boiling water bath for 5-10 min and observed for a yellow and then brick-red precipitate.

Benedict's test: Equal volume of Benedict's reagent and test solution in test tube were mixed. Heated in boiling water bath for 5min. solution may appear green-yellow or red depending on the amount of reducing sugar present in the test solution.

V. Test for Amino Acids:

Ninhydrin test: 3ml of test solution and 3 drops of ninhydrin were heated in boiling water bath for 10 min observed for purple or bluish color.

VI. Test for Proteins:

Millon's, test: Mixed 3ml of test solution with 5ml of Millon's reagent, white precipitate obtained.

Precipitate warmed turns brick red, or precipitate dissolves given red solution.

VII. Test for Flavonoids: Too small quantity of residue, added lead acetate solution observed for yellow-colored precipitate. To the test solution, added few drops of ferric chloride solution were observed for intense green.

VIII. Test for Alkaloids:

Mayer's test: 2-3 ml of filtrate with a few drops of Mayer's reagent was observed for precipitate.

Hager's test: 2-3 ml of filtrate with few drops Hager's reagent was observed for yellow precipitate.

ix. Tests for Tannins: 1 ml of Tannin solution is added to 1% Gelatin solution containing 10% sodium chloride forming precipitation.

1.4. Procedure for the FTIR²⁰: The various parts of the *Digera arvensis* were macerated for 24 hr at room temperature, with a magnetic stirrer using ethanol as the solvent. The ethanolic extract is then filtered using Whatman filter paper. The filtrate was evaporated to dryness below 40 °C. The samples collected were given to IISc, Bangalore, for the analysis of FTIR. The functional group analysis of the ethanolic extract of *Digera arvensis* was carried out using the instrument Thermo Fisher Scientific; the model used is Nicolet iS50. The source used is IR using XT-KBr as a beam splitter; detection is done through the detector, DTGS KBr. The extracts were scanned in the wavelength range of 4000 cm^{-1} - 400 cm^{-1} with a resolution of 4 and optical velocity of 0.4747, and characteristic peaks of the functional groups were detected, and the FTIR peak values were recorded.

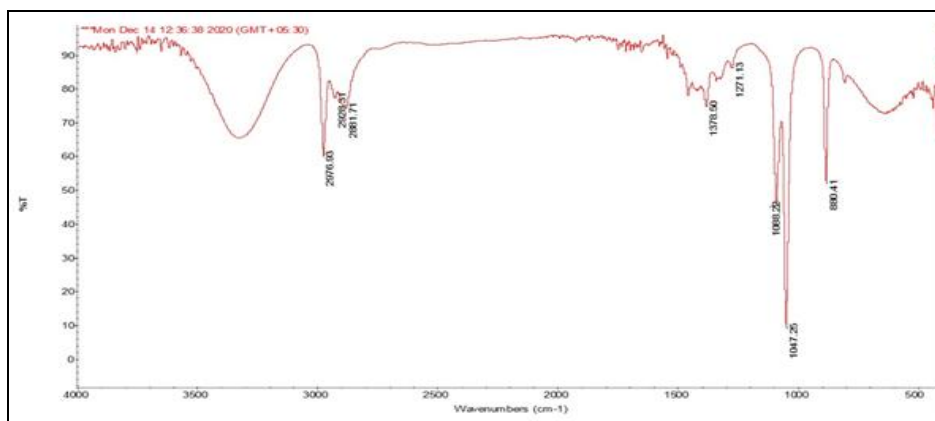
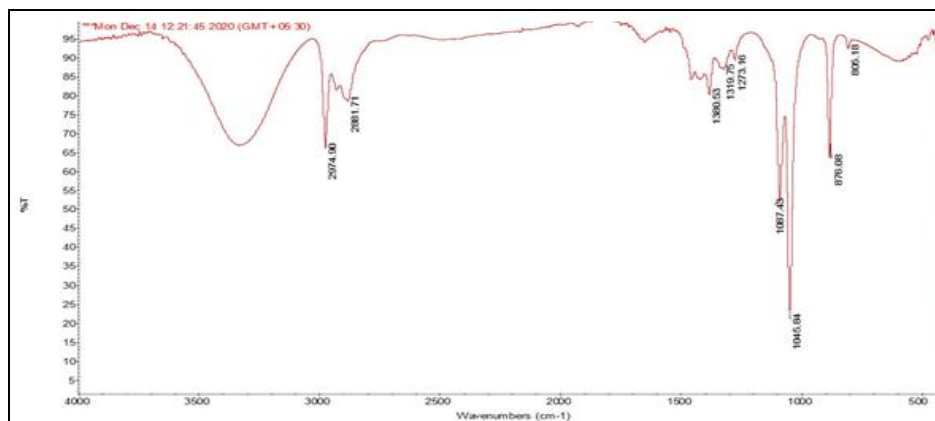
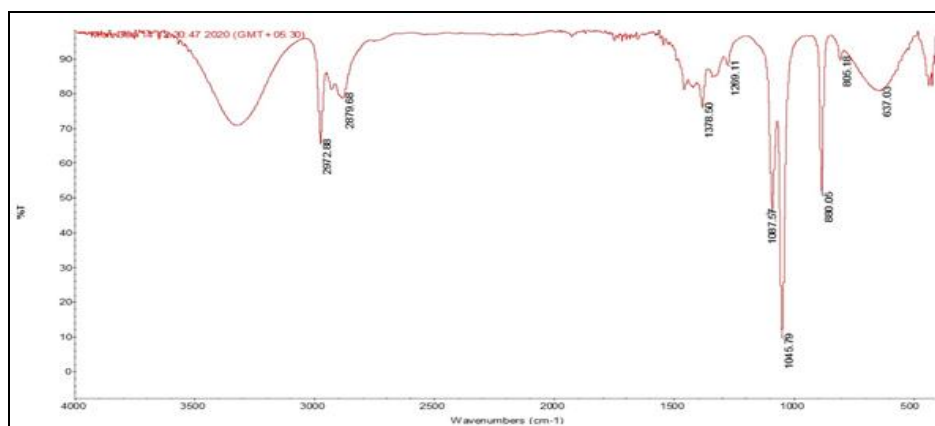
RESULTS AND DISCUSSION:

2.1 Preliminary Phytochemical Investigation: The phytochemical screening of leaf, stem, root, and seed extracts of *Digera arvensis* are summarized in **Table 1**.

The results indicated the presence of flavonoids, cardiac glycosides, alkaloids, sterols, tannins, phenols in the leaf, stem, root, and seed extracts. The carbohydrates, reducing sugars, and proteins were also found to be present in the extract.

TABLE 1: PHYTOCHEMICAL ANALYSIS OF LEAF, STEM, ROOT, AND SEED EXTRACTS OF *DIGERA ARVENSIS*

Phytochemical group	Ethanollic extract of leaf <i>Digera arvensis</i>	Ethanollic extract of stem <i>Digera arvensis</i>	extract of root <i>Digera arvensis</i>	extract of seed <i>Digera arvensis</i>
Flavonoids	+	+	+	+
Alkaloids	+	+	+	+
Glycosides	+	+	+	+
Sterols	-	-	-	-
Saponins	+	+	+	+
Tannins	+	+	+	+
Carbohydrates	+	+	+	+
Reducing Sugars	+	+	+	+
Protein	+	+	+	+
Gums	-	-	-	-
Fat	-	-	-	-

**FIG. 2: FT-IR SPECTRUM OF ETHANOLIC EXTRAC OF LEAVES OF *DIGERA ARVENSIS*****FIG. 3: FT-IR SPECTRUM OF ETHANOLIC EXTRAC OF ROOT OF *DIGERA ARVENSIS*****FIG. 4: FT-IR SPECTRUM OF ETHANOLIC EXTRAC OF SEED OF *DIGERA ARVENSIS***

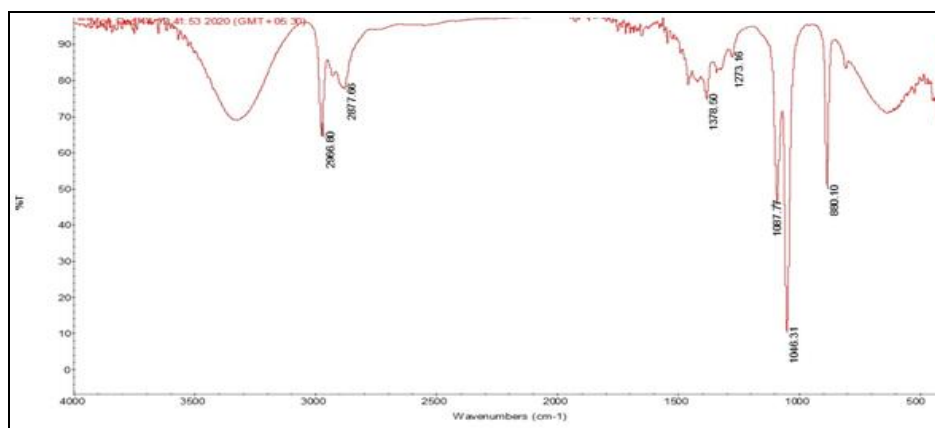


FIG. 5: FT-IR SPECTRUM OF ETHANOLIC EXTRACT OF STEM OF *DIGERA ARVENSIS*

TABLE 2: FTIR AND MAJOR PEAKS ASSESSMENT OF LEAF, STEM, ROOT, AND SEED EXTRACT

Vibration frequency [cm ⁻¹]	Assignment of leaf extract	Vibration frequency [cm ⁻¹]	Assignment of stem extract	Vibration frequency [cm ⁻¹]	Assignment of root extract	Vibrational frequency [cm ⁻¹]	Assignment of seed extract
3343	OH stretch	3343	OH stretch		OH stretch	3343	OH stretch
2976	CH stretch			3343		2972	CH stretch alane
				2974	CH stretch alane		
2928	CH stretch alane	2966	CH stretch alane	2881	OH benzyl alcohol	2879	CH stretch alane
				1380	CH bending alcohol		
2881	NH stretching (amine salt)	1378	OH bending alcohol	1319	S=O stretching sulfonic	1378	CH bending alcohol
1378	OH bending alcohol		CO stretching alkyl aryl ether	1273	CO stretching alkyl aryl ether	1269	CN stretching aromatic amine stretching
1271	CO stretching	1273		1087	CO stretching aliphatic ether	1087	CO stretching aliphatic ether
1088	CO stretching aliphatic ether	1087	CO stretching aliphatic ether	1045	CO-O-CO anhydride stretching	1045	CO-O-CO anhydride stretching
1047	CO-CO-CO stretching	1046	CO-CO-CO stretching	876	CH bending 1, 3 di substituted	880	CH bending 1, 2, 4, tri substituted
880	CH bending 1, 2, 4, tri substituted	880	CH bending 1, 2, 4, tri substituted	805	CH bending 1, 4, di substituted	805	CH bending 1, 4, disubstituted

2.2 Fourier Transforms Infrared (FT-IR) Spectroscopy:

The chemical nature of the functional groups could be determined by analyzing the types of vibrational study of the existing bands on the spectrum obtained by infrared absorption spectra. The vibrational modes of the characteristic structure are given. The bands and stretch of the spectra are compared to the data reported for similar compounds. The FT-IR spectrums obtained from the ethanolic extract of the leaf is shown in Fig. 1. The components of the leaf extract are determined by the bands observed at 3343, 2976, 2928, 2881, 1378, 1271, 1088, 1047,

880 cm⁻¹. The FT-IR spectrums obtained from the ethanolic extract of the stem are shown in Fig. 2. The bands determine the components of the stem extract observed at 3343, 2966, 2877, 1378, 1273, 1087, 1046, 880 cm⁻¹. The FT-IR spectrums obtained from the ethanolic extract of the root are shown in Fig. 3. The components of the root extract are determined by the bands observed at 3343, 2974, 2881, 1380, 1319, 1273, 1087, 1045, 876, 805 cm⁻¹. The FT-IR spectrums obtained from the ethanolic extract of seed are shown in Fig. 4. The components of the seed extract are determined by the bands observed at 3340, 2972, 2879, 1378,

1269, 1087, 1045, 880, 805 cm^{-1} . The *D. arvensis* ethanolic extract of leaf spectra showed a strong absorption band at 3343 cm^{-1} , which may be the OH stretching vibration of polyphenols and alcohol. The peak at 2976, 2928, 2881 cm^{-1} are attributed to CH stretching, peak at 1378 cm^{-1} is due to OH bending alcohol, peak at 1271 cm^{-1} is due to C-O stretching alkyl aryl ether, peaks at 1088 cm^{-1} are characteristic to C-O stretching aliphatic ether, 1047 cm^{-1} peak may be due to CO-O-CO anhydride and peak at 880 cm^{-1} is due to C-H bending 1,2,4 tri-substituted. The *D. arvensis* ethanolic extract of stem spectra showed a strong absorption band at 3343 cm^{-1} , which may be the OH stretching vibration of polyphenols and alcohol. The peak at 2966, 2877 cm^{-1} are attributed to CH stretching, peak at 1378 cm^{-1} is due to OH bending alcohol, peak at 1273 cm^{-1} is due to C-O stretching alkyl aryl ether, peaks at 1087 cm^{-1} are characteristic to C-O stretching aliphatic ether, 1046 cm^{-1} peak may be due to CO-O-CO anhydride and peak at 880 cm^{-1} is due to C-H bending 1, 2, 4 tri-substituted.

The *D. arvensis* ethanolic extract of root spectra showed a strong absorption band at 3343 cm^{-1} , which may be the OH stretching vibration of polyphenols and alcohol. The peak at 2974, 2881 cm^{-1} are attributed to CH stretching, peak at 1380 cm^{-1} is due to CH bending alkane, peak at 1319 cm^{-1} is characteristic to S=O stretch sulfone, peak at 1273 cm^{-1} is due to C-O stretching alkyl aryl ether, peaks at 1087 cm^{-1} are characteristic to C-O stretching aliphatic ether, 1045 cm^{-1} peak may be due to CO-O-CO anhydride and peak at 876 cm^{-1} is due to C-H bending 1,3 disubstituted. The *D. arvensis* ethanolic extract of seed spectra showed a strong absorption band at 3343 cm^{-1} , maybe OH stretching vibration of polyphenols and alcohol. The peak at 2972, 2879 cm^{-1} are attributed to CH stretching, peak at 1378 cm^{-1} is due to CH bending alcohol, peak at 1269 cm^{-1} is characteristic to C-N stretching of aromatic amine, peaks at 1087 cm^{-1} are characteristic to C-O stretching aliphatic ether, 1045 cm^{-1} peak may be due to CO-O-CO anhydride and peak at 880 cm^{-1} is due to C-H bending 1,3 di substituted, peak at 805 is due to 1, 3 disubstituted.

CONCLUSION: The natural source proved to have fewer adverse effects and curative effects on the diseases effectively from the past. In this study,

the ethanolic extracts of all parts of *D. arvensis* were observed to contain many phytochemicals. Henceforth the weed can be highly valuable & which would contribute to the medicinal usage for the remedy of various human ailments. It is anticipated that the present investigation would prompt an investigation of more potent values of *D. arvensis*. Further, there is a requirement to isolate the active molecule and evaluate the pharmacological activities of the isolated component.

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CONFLICTS OF INTEREST: The authors declare that they do not have any conflict of interest.

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