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PHYTOCHEMICAL INVESTIGATION AND ANTIMICROBIAL ACTIVITY OF *Cichorium intybus* L.

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ABSTRACT

Medicinal plants are assumes greater importance in the primary health care of individuals and communities in many developing countries. Plants are believed to be much safer and proved elixir in the treatment of various ailments. In the present study was planned to conduct Phytochemical investigation and Antimicrobial activity of *Cichorium intybus* L. The results of this study clearly indicate that the preliminary phytochemical analysis of *Cichorium intybus* leaves revealed presence of of flavonoids, phenolics, steroids, tannin, saponins, glycosides, steroids, alkaloids while protein and anthroquinones were absent. The results reveal that extract of *Cichorium intybus* leaves were significantly effective against both bacteria *E. coli*, *St. aureus* and fungi *C. albicans*. The leaves of *Cichorium intybus* are a newly discovered potential source of natural antimicrobial compounds. The synergistic effect of plant extract against resistant bacteria and fungi leads to new choices for the treatment of infectious diseases. Overall, the Cichorium *intybus* leaves are a rich source of phytochemicals and antimicrobial activity that can be important in infectious disease prevention.

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INTRODUCTION

Plants have basic nutritional importance by their content of protein, carbohydrate, fats and oils minerals, vitamins and water responsible for growth and development in man and animals. Phytochemical simply means plant chemicals. Phytol is the Greek word for plant. Phytochemicals are classified as primary or secondary constituents, depending on their role in plant metabolism. Primary metabolism is important for growth and development of plants include the common sugars, aminoacids, proteins, purines and pyrimidines of nucleic acids, chlrophyll's etc. Secondary metabolism in a plant plays a major role in the survival of the plant in its environment. Attractions of pollinators, natural defense system against predators and diseases, etc., are examples of the roles of secondary metabolites. The secondary metabolites formed also are an important trait for our food plants (taste, colour, scent, etc.) and ornamental plants. Moreover, numerous plant secondary metabolites such as flavonoids, alkaloids, tannins, saponins, steroids, anthocyanins, terpenoids, rotenoids etc. have found commercial application as drug, dye, flavour, fragrance, insecticide, etc. Such fine chemicals are extracted and purified from plant materials. Plant produces these chemicals to protect itself but recent research demonstrates that many phytochemicals can protect humans against diseases including cancer, cardiovascular, arthritis, diabetic, etc. (Satyavati, 1982). The aim of the study to investigate the Phytochemical investigation and Antimicrobial activity of *Cichorium intybus* L..

MATERIALS AND METHODS

Plant materials:

The fully mature *Cichorium intybus* leaves were collected in December 2015 from Sundaraperumal Kovil, Thanjavur district, Tamil Nadu, India.

Preparation of alcoholic extract:

The leaf of *Cichorium intybus* was first washed well and dust was removed from the leaves. Leaf was washed several times with distilled water to remove the traces of impurities from the leaf. The leaves were dried at room temperature and coarsely powdered. The powder was extracted with 70% methanol for 24 hours. A semi solid extract was obtained after complete elimination of alcohol under reduced pressure. The extract was stored in refrigerator until used.

Phytochemical screening

Chemical tests were carried out on the alcoholic extract and on the powdered specimens using standard procedures to identify the constituents as described by Sofowara (1993), Trease and Evans (1989) and Harborne (1973).

Antimicrobial assay

Antibiogram was done by disc diffusion method (NCCLS, 1993; Awoyinka *et al*., 2007) using plant extracts. Petriplates were prepared by pouring 30 ml of NA /PDA medium for bacteria/fungi. The test organism was inoculated on solidified agar plate with the help of micropipette and spread and allowed to dry for 10 mints. The surfaces of media were inoculated with bacteria/fungi from a broth culture. A sterile cotton swab is dipped into a standardized bacterial/ fungi test suspension and used to evenly inoculate the entire surface of the Nutrient agar/PDA plate. Briefly, inoculums containing *Escherichia coli, Staphylococcus aureus* specie of bacteria were spread on Nutrient agar plates for bacteria and *Candida albicans was* spread on potato dextrose agar for fungus strains. Using sterile forceps, the sterile filter papers (6 mm diameter) containing the crude extracts (50µl, 100 µl and 150 µl) were laid down on the surface of inoculated agar plate. The plates were incubated at 37°C for 24 h for the bacteria and at room temperature (30±1) for 24-48 hr. for yeasts strains. Each sample was tested in triplicate.

RESULTS AND DISCUSSION

Plants have basic nutritional importance by their content of protein, carbohydrate, fats and oils minerals, vitamins and water responsible for growth and development in man and animals. Secondary metabolism in a plant plays a major role in the survival of the plant in its environment. Attractions of pollinators, natural defense system against predators and diseases, etc., are examples of the roles of secondary metabolites (Sofowara, 1993).

Plants are believed to be much safer and proved elixir in the treatment of various ailments. Plants synthesize an array of chemical compounds that are not involved in their primary metabolism. These 'secondary compounds' instead serve a variety of ecological functions, ultimately to enhance the plants survival during stress. In addition these compounds may be responsible for the beneficial effects of fruits and vegetables on an array of health related measures. Medicinal plants are assumes greater importance in the primary health care of individuals and communities in many developing countries. There has been an increase of demand in international trade because of very effective, cheaply available, supposedly have no side effects and used as alternative to allopathic medicines (Liu, 2003).

In the present study was carried out on the *Cichorium intybus* leaves revealed the presence of medicinally active constituents. The phytochemical characters of the *Cichorium intybus* leaves investigated and summarized in Table-1 and fig- 2. The phytochemical screening *Cichorium intybus* leaves showed that the presence of flavonoid, Polyphenol, tannin, saponin, glycoside, alkaloids, carbohydrate, terpenoid, tri terpenoid and anthroquinone while steroid, phlobatannin and protein and were absent.

Falodun *et al*. (2006) reported the occurrence of flavonoids, saponins, diterpenes and phorbol estersin in the aqueous and methanol extracts of *Euphorbia heterophylla*. Raghavendra *et al*. (2006) examined the powdered leaf material of different solvent of *Oxalis corniculata* and reported the presence of phenols, glycosides, carbohydrates, phytosterols and tannins. Awoyinka *et al*. (2007) extracted eight bioactive compounds from dry leaf of *Cnidoscolus aconitifolius* using water and ethanol. Different extracts of *Semecarpus anacardium* were analysed by Mohanta *et al*. (2007) for its phytochemical properties.

Onwukaeme *et al*. (2007) detected reducing sugars, phenols, tannins and flavonoids in *Pycanthus angolensis*. Uma Devi *et al*. (2007) carried out the phytochemical analysis in *Achyranthes bidentata*. The methanol and acetone extracts of 14 plants belonging to different families were evaluated for phytochemical analysis and this study revealed the presence of tannins, cardiac glycosides, steroids and saponins (Vaghasiya and Chanda, 2007). Ayoola *et al*. (2008) investigated the phytochemical components of four medicinal plants used for the treatment of malaria in Southwestern Nigeria. *Ichnocarpus frutescens* leaf, stem and root were investigated (Mishra *et al*., (2009) for its phytochemical and phytochemical properties.

Quantitative analysis

Quantitative analysis revealed that the plant has polyphenol, alkaloids, tannin and saponin. Rich amount of total phenol (207.21mg/gm), tannin (59.70mg/gm), alkaloids (80.54mg/gm), saponin (60.24 mg/gm) and flavonoid (120.10mg/gm) was presented (Table 2). The above phytoconstituents were tested as per the standard methods. This is because of the pharmacological activity of this plant is used to trace the particular compound.

Antimicrobial activity

Nature has bestowed on us a very rich botanical wealth and a large number of diverse types of plants grow in different parts of the country. Emergence of pathogenic microorganisms that are resistant/multi-resistant to major class of antibiotics has increased in recent years due to indiscriminate use of synthetic antimicrobial drugs. In addition, high cost and adverse side effects are commonly associated with popular synthetic antibiotics, such as hypersensitivity, allergic reactions, and immunosuppressant and are major burning global issues in treating infectious diseases (Karaman *et*

al., 2003). This situation forced scientists to search for new antimicrobial substances with plant origin.

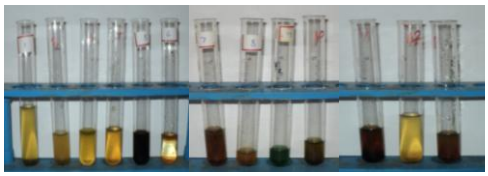
Plant extract of *Cichorium intybus* was screened against *Escherichia coli* and *Staphylococcus aureus* species of bacteria and *Candida albicans* species of fungi were evaluated using the standard agar disc diffusion method. The disc diffusion method is used to detect the antimicrobial activity of plant extract. The solidified Nutrient agar plates were swapped with the test organism and the samples were impregnated. After the incubation the zone was measured. The antimicrobial activity of plant extracts was detected by the indication of zone around the disc. The *in vitro* antimicrobial activity of the *Candida albicans* leaves extract against these bacteria and fungi were qualitatively assessed by the presence of inhibition zones represented in the photographic Fig 3. The inhibitory activities in culture media of the *Candida albicans* reported in Table 3 were comparable with standard antimicrobiotic viz. chloromphenical and fluconazole.

Table: 1 Phytochemical screening of *Cichorium intybus* leaf extract

S.No	Phytochemical analysis	Coloration	Results
1	Tannin	Brownish green	+
2	Phlobatannin	-	-
3	Saponin	Emulsion	+
4	Flavonoid	yellow	+
5	Steroid	blue	-
6	Terpenoid	Two layers	+
7	Triterpenoid	Reddish violet	+
8	Alkaloids	White precipitate	+
9	Carbohydrate	Red precipitate	+
10	Protein	Violet	-
11	Anthroquinone	Rose pink	+
12	Polyphenol	Bule green	+
13	Glycoside	Brown ring	+

(+) Presence (-) Absence

Fig: 2 Phytochemical screening of *Cichorium intybus* leaf extract



1. Tannin, 2. Phlobatannins, 3.Saponin, 4. Flavonoids, 5. Steroids, 6.Terepenoids, 7.Triterpenoids, 8. Alkaloids, 9.Carbohydrate, 10. Protein, 11.Anthroquinone, 12. Polyphonol and 13.Glycoside

Table: 2 Quantitative analysis of *Cichorium intybus* leaf extract

S.No	Name of the Test	Result (mg/gm)
1.	Polyphenol	180.21
2.	Flavonoid	120.10
3.	Alkaloid	80.54
4.	Saponin	60.24
5.	Tannin	59.70
6.	Terpenoids	60.00

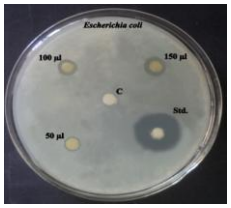
Table: 3 Antimicrobial activity of *Cichorium intybus* leaf extract

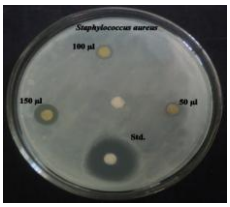
Microbial Organism	50µl	100 µl	150 µl	Standard	Control
Escherichia coli (mm)	2.40±0.16	3.02±0.21	6.74±0.47	11.56±0.80	0
Staphylococcus aureus (mm)	2.17±0.08	2.93±0.20	4.36±0.30	11.18±0.78	0
Candida albicans (mm)	1.02±0.07	2.14±0.14	4.22±0.29	10.36±0.72	0

Values were expressed as Mean ± SD.
Bacterial standard - Chloromphenical
Fungal standard - Fluconazole

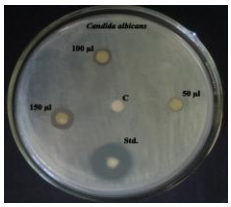
Fig: 3 Antimicrobial activity of *Cichorium intybus* leaf extract

Escherichia coli ***Staphylococcus aureus***





Candida albicans



CONCLUSSION

The results of this study clearly indicate that the preliminary phytochemical analysis of *Cichorium intybus* leaves revealed presence of of flavonoids, phenolics, steroids, tannin, saponins, glycosides, steroids, alkaloids while protein and anthroquinones were absent. The results reveal that extract of *Cichorium intybus* leaves were significantly effective against both bacteria *E. coli*, *St.*

aureus and fungi *C. albicans*. The leaves of *Cichorium intybus* are a newly discovered potential source of natural antimicrobial compounds. The synergistic effect of plant extract against resistant bacteria and fungi leads to new choices for the treatment of infectious diseases.

Overall, the Cichorium *intybus* leaves are a rich source of phytochemicals and antimicrobial activity that can be important in infectious disease prevention.

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