Research Article

IN VITRO ANTIMICROBIAL ACTIVITY, NUTRITIONAL PROFILE AND PHYTOCHEMICAL SCREENING OF WILD EDIBLE FRUIT OF GARHWAL HIMALAYA (*FICUS AURICULATA*)

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ABSTRACT

The various fractions of *Ficus auriculata* were screened for in vitro Antibacterial, Antifungal activity, Nutritional Evaluation and Phytochemical Screening. The plant edible fruit content nutrients such as crude protein 5.32%, carbohydrates 27.09%, crude fiber 16.96% and ash content 3.7% and minerals as calcium, magnesium, potassium and phosphorus (1.35, 0.90, 2.11 and 0.28 mg/100gm) respectively. The ethanolic fruit extracts of *Ficus auriculata* showed significant activity 14±1mm, 13±1mm and 12±1mm against *Shigella flexneri, Escherichia coli* and *Staphylococcus epidermidis* against food poisoning bacteria, and phytochemical screening for the presence of glycosides, flavonoids, phenols, resin and tannins. However, alkaloids were absent. This analysis revealed that, the fruits contained higher value of fat, protein, fiber and minerals as compared to the cultivated fruits with apple and mango. *Ficus auriculata* fruits contain sufficient amount of nutrients, required per day by a person. Consumption of fruits may promote general health and well-being as well as reduce the risk of chronic diseases. These findings confirm that the *Ficus auriculata* may be potential source for the formulation of nutraceuticals or natural foods.

Keywords: Antibacterial, Antifungal, Nutritional value and Phytochemical screening.

INTRODUCTION

India has been known to be rich repository of medicinal plants. The forest in India is the principal repository of large number of medicinal and aromatic plants, which are largely collected as raw materials for manufacture of drugs and medicinal products. Traditional herbal medicine possesses greater significance in Uttarakhand Himalaya hence interest in herbal medicine has gradually increased in recent years[1]. The Garhwal Himalaya region of Uttarakhand is highly enriched with its vegetation including wild edible fruits due to its varied eco-geographical and eco-climatic conditions. Local inhabitants to play a significant role as supplementary food consume wild fruits. They cover a wide range of pesticides, insecticides, fertilizers and other poisons. Plant species have long been the principal ingredients of traditional medicine[2] and their use dates back to the beginning of human civilization[3]. *Ficus auriculata* is a huge tropical, deciduous and evergreen tree with more than 800 species. Bark, root, leaves, fruit and latex of this plant are frequently used for the treatment of various illnesses. *Ficus auriculata* produces a unique fruit which is actually an inverted flower. Ficus species are rich source of polyphenolic compounds, flavonoids which are responsible for strong antioxidant properties that help in prevention and therapy of various oxidative stress related diseases such as neurodegenerative and hepatic diseases. Herbal medicine has clearly recognizable therapeutic effects[4] as any medium, provided the original work is properly cited. Leaves are crushed and the paste is applied on the wounds. They are also used in diarrhea and dysentery. Stem bark juice is effective for diarrhea, cuts and wounds. Roasted figs are taken for diarrhea and dysentery. Root latex is used in mumps, cholera, diarrhea and vomiting. *Ficus auriculata* is a very tasty fruit. It is very much liked by all. The fig is a very juicy fruit Work should be taken on standardizing the techniques for making various products, such as squash, jam and jelly from this fruit. It contains B-sitosterol, friendelin and epifriedlanal isolation from egyption plant.

MATERIALS AND METHODS

Plant Material

The fresh parts of fruit of *Ficus auriculata* were collected from adjoining area of Langasu city (Dist- Chamoli Uttarakhand) in the month of July - August. The plant was authenticated by botanist Dr. R. D. Guar, Department of Botany; H. N. B. G. U. Srinagar Garhwal.

Preparation of plant Extract

The plant material was separated into its selected parts (leaf, bark and fruit) air dried ground to moderately fine powder and Soxhlet extracted with increasing polarity solvent (Petroleum ether, chloroform, ethyl acetate, acetone, methanolic, ethanolic and water)[5]. Each extract was evaporated to dryness under reduce pressure using rotary evaporator. The coarse powder of fruit bark and root was subjected to successive hot continuous extraction with various solvent each time before extracting with next solvent the powdered material will be air dried (weight of crude extract 100gm). The various concentrated extracts were stored in air tight container for further studies.

Media

Nutrient broth, Nutrient agar, Muller Hinton agar, Malt extract broth and Sabouraud dextrose agar, Alcohol, Hydrochloric acid, alcohol, and sulphuric acid, Distilled

water etc all product of Himedia Laboratories Mumbai (India) were used in this study.

Bacterial Strains

Ten bacterial strains were used namely *Escherichia coli, Klebsiella pneumoniae, Enterobacter gergoviae, salmonella entericatyphim, shigella flexneri, Staphyloccus aureus, staphyloccus epidermidis, streptococcus pyogenes,* and *Bacillus cereus*. The bacterial strains were supplied by the Microbial Type Culture Collection and Gene Bank, Institute of Microbial Technology, Chandigarh, India (Customer no. 3921).

Fungal Strains

Three fungal strains were used namely *Candida albicans, Aspergillus flavus* and *Aspergillus parasiticus*. The fungal strains were supplied by the Microbial Type Culture Collection and Gene Bank, Institute of Microbial Technology, Chandigarh, India.

Antibacterial assay

The disc diffusion assay methods were used to determine the growth inhibition of bacteria by plant extracts[6, 7]. Diluted bacterial culture (100µl) was spread over nutrient agar plates with a sterile glass L-rod. 10mg/ml and 50mg/ml of the each extracts were applied to each filter paper disc (Whatman No. 1, 5 mm diam.) and allowed to dry before being placed on the agar plate. Each extract was tested in triplicate (3 discs/ plate) and the plates were inoculated at 37°C for 24 h. After incubation, the diameter of inhibition zones was measured with a caliper.

Antifungal assay

The antifungal activity was tested by disc diffusion method[8, 9]. The Sabouraud dextrose agar plates were each similarly seeded with each fungal strain The 24 hrs. both culture of each bacterium and 7 days inoculated fungus culture were used to seed sterile Sabouraud dextrose agar at 45°C respectively and fungal plates were incubated at 25-28°C for 7 days after which diameter of zones of inhibition were measured. Each disc filled with extract.

Nutritional & Mineral assay

The edible portion of fruits was analyzed for moisture, ash, fat and Fiber as per method[10]. Total nitrogen was analyzed by micro-kjeldhal method[11] and for crude protein the value was multiplied by 6.25. Total carbohydrates were obtained by subtracting the value moisture, crude protein, crude fat crude fiber and ash from 100%[12]. The total energy value equal to addition of fat, protein and sugars calorie, each gram of fat give 9 kcal, protein and sugar give 4 kcal energy. The minerals analyzed were Potassium using atomic absorption spectrophotometer, Calcium and Phosphorus by flame photometer. Ascorbic acid in fruits was estimated[13].

Phytochemical analysis

The qualitative phytochemical properties of the dried powdered sample were determined using standard methods[14].

Table 1: Antibacterial activity of ten bacterial strains against *Ficus auriculata* plant fruit extract. Disc size, 5 Mm, Inhibitory zone size ±1 Mm, Mm means (millimetres) and – indicate (NIZ) No inhibitory zone

Bacterial Name		Petroleum ether Extract		Chloroform Extract		Ethyl acetate Extract		Acetone Extract		Ethanol Extract		Water Extract	
		Concentration (Mg/ml)											
Genus /Species/Subspe.	MTCC (Code)	10	50	10	50	10	50	10	50	10	50	10	50
Bacillus cereus	1272	-	-	-	-	-	7	6	8	7	9	9	11
Escherichia coli	729	-	6	-	7	8	10	-	9	9	11	-	9
Enterobacter gergoviae	621	-	-	-	-	-	9	7	9	9	12	-	7
Klebsiella pneumonia	432	-	-	-	8	7	9	8	10	8	10	7	10
Salmonella entericatyphim	98	-	-	-	-	-	8	-	8	7	9	-	8
Shigella flexneri	1457	-	6	-	-	8	10	-	9	10	14	9	12
Staphyloccus aureus	902	-	7	-	7	7	11	8	10	7	9	8	10
Staphyloccus epidermidis	435	-	-	-	-	7	9	-	8	9	12	-	8
Streptococcus pyogenes	1925	-	-	-	6	-	8	-	9	-	9	7	9
Escherichia coli	443	-	8	-	9	8	9	-	-	10	13	-	8

Table 2: Fungal activity of three fungal strains against *Ficus auriculata* plant fruit extract. Disc size, 5 Mm, Inhibitory zone size ±1 Mm, Mm means (millimetres) and – indicate (NIZ) No inhibitory zone

Fungal Name		Petroleum ether Extract		Chloroform Extract		Ethyl acetate Extract		Acetone Extract		Ethanol Extract		Water Extract	
		Concentration (Mg/ml)											
Genus /Species/Subspe.	MTCC (Code)	10	50	10	50	10	50	10	50	10	50	10	50
Candida albicans	3017	-	-	-	-	-	7	-	8	-	7	-	8
Aspergillus flavus	2798	-	7	-	-	-	-	-	-	-	9	-	7
Aspergillus parasiticus	2796	-	9	-	-	-	-	-	7	-	8	-	8

RESULTS AND DISCUSSION

Plants are important source of potentially bioactive constituents for the development of new chemotherapeutic agents. The first step towards this goal is the in vitro antimicrobial activity assay. The results of antibacterial, antifungal, nutritional value and phytochemical screening activity, tables 1-5 and figures 1-3, reveals that antibacterial, antifungal, nutritional, and phytochemical screening activity of fruit of *Ficus auriculata* was evaluated against ten bacterial and three fungal pathogenic strains.

Table 3: Nutritional value of *Ficus auriculata* fruit

Nutrients	Value
Moisture (%)	46.64 ± 0.15
Ash (%)	3.7 ± 0.08
Total protein	5.32 ± 0.04
Crude fat (%)	0.65 ± 0.25
Crude fibre (%)	16.96 ± 0.09
Soluble carbohydrates	27.09± 0.11
Organic matter	96.30± 0.16
Energy value	135.51± 0.09
Vit.C	0.09± 0.04
Mg/100gm nitrogen	0.85 ± 0.07
Ca mg/100gm	1.35 ± 0.10
Mg mg/100gm	0.90± 0.06
K mg/100gm	2.11± 0.08
P mg/100gm	0.28 ± 0.04

Table 4: Qualitative estimation of *Ficus auriculata* fruit and leaf phytochemical screening

Test	*Ficus auriculata* fruit	*Ficus auriculata* leaf
Carbohydrates/ glycosides		
(1) Molish test	(+)	(-)
(2) Fehling test	(+)	(-)
(3) Benedict test	(+)	(+)
Alkaloid		
(1) Mayer's test	(+)	(-)
(2) Dragondroff test	(-)	(-)
Flavonoid	(+)	(+)
Saponins	(+)	(+)
Tannins		
(1) Pyrogoll & catechol	(+)	(+)
(2) Gallic acid	(-)	(-)
Unsaturated sterol/triterpenes		
(1) Liebermann Burchard test	(+)	(-)
(2) Salkowiskis test	(+)	(-)
Resin	(+)	(-)

Antibacterial and antifungal activity

Ficus auriculata ethanolic fruit extract significant activity 14±1mm, 13±1mm and 12±1mm against *Shigella flexneri, Escherichia coli* and *Staphylococcus epidermidis* for food poisoning bacteria, the order of the species based on total antibacterial activity is as follows: *Shigella flexneri* > *Escherichia coli* > *Staphylococcus epidermidis*.

Nutritional value

The level of nutrients such as crude protein 5.32%, carbohydrates 27.09%, crude fiber 16.96% and ash content 3.7% and minerals as calcium, magnesium, potassium and phosphorus (1.35, 0.90, 2.11 and 0.28mg/100gm) respectively.

Table 5: Qualitative estimation of *Ficus auriculata* fruit amino acid screening

Amino acid test	*Ficus auriculata* fruit
L- Hydroxy proline	(+)
DL Serine	(-)
DL Iso-leucine	(+)
DL Valine	(+)
DL-2-Aminobutyric acid	(+)
L-Ornithin	(-)
L-Cystein hydroxyl	(+)
DL-Nor-leucine	(-)
DL-Tryptopham	(+)
DL-Alanine	(+)
L-Glutamic acid	(-)
Glycine	(-)
L –Proline	(-)
L- Arginine	(+)
DL – Aspartic acid	(+)
L –Cystein hydroxychloride	(+)
L- Histidine	(-)
L – Leucine	(+)
L –Lysine monochloride	(+)
DL – Methionine	(-)
DL – β-Phenyl alanine	(-)
DL – Threonine	(+)
L – Tyrosine	(+)
3-C-3-4Dihydroxy phenyl	(-)

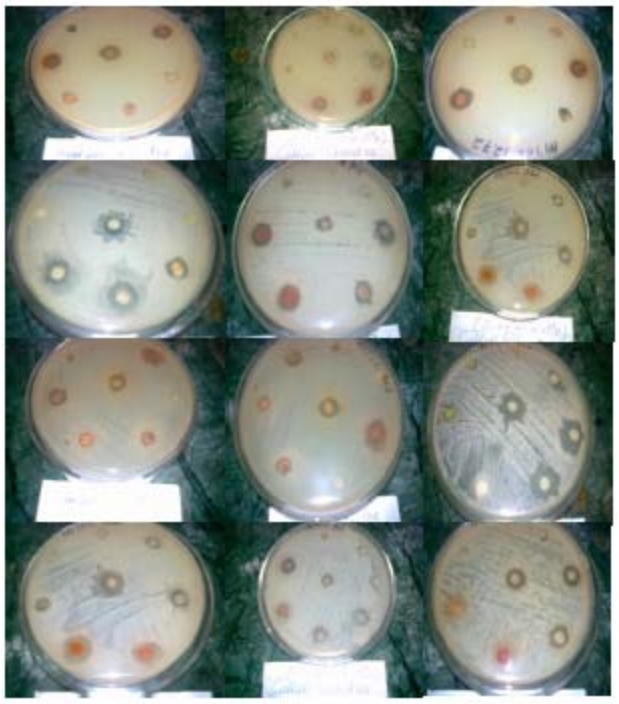

Figure 1: Antibacterial and antifungal activity of ten bacterial strains & three fungal strains against *Ficus auriculata* plant fruit extract.

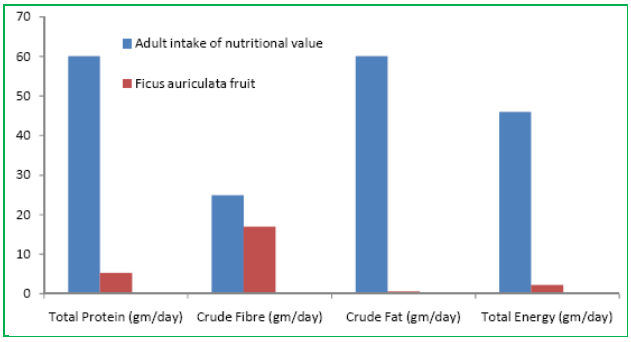

Figure 2: Comparison of per day intake of nutrients by Adults with the nutrients present in the fruits of *Ficus auriculata*.

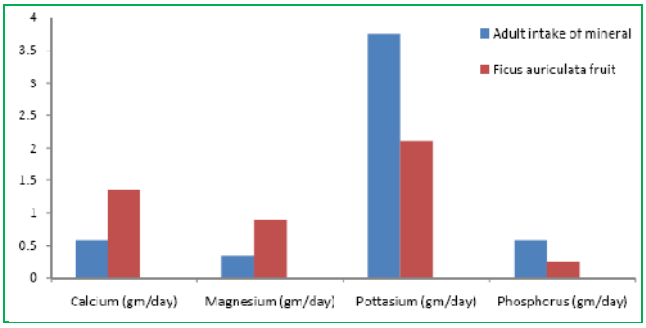

Figure 3: Comparison of per day intake of minerals by Adults with the mineral present in the fruits of *Ficus auriculata*.

Phytochemical screening

The phytochemical screening for the presence of glycosides, flavonoids, phenols, resin and tannins. However alkaloids were absent. This analysis revealed that, the fruits contained higher value of fat, protein, fiber and minerals as compared to the cultivated fruits with apple and 200 gm fruits contain sufficient amount of nutrients, required per day by a person.

CONCLUSION

The in vitro antimicrobial studies present *F. auriculata* to have considerable efficacy against various pathogenic bacteria. The study provides a scientific basis for the use of the plant as folk medicine. The fruit of the plant is a good source of essential nutrients including minerals, carbohydrates, proteins and lipids. However, more advanced pharmacological and clinical studies would be required to investigate in vivo mechanism of nutraceuticals effects of this important wild plant.

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