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Pharmacognostic Investigation, Anti-Helminthic Activity and Anti Microbial Activity of Rubus Ursinus

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ABSTRACT

To evaluate the physicochemical parameter. Anti-Helminthic activity, Anti-Fungal activity and Anti-bacterial activity by various extracts (petroleum ether, chloroform, acetone, methanol) against various gram positive and gram negative bacteria (E.coli, Staphylococcus aureus and Bacillus subtilis) using a standard as Amoxicillin, Anti-Fungal activity (Candida Albicans, Aspergillus) using Ketoconazole and Anti-Helminthic activity using a standard as Albendazole. The collected leaves of Rubus Ursinus were dried under shade for 10 days and then powdered into coarse particles in a mechanical grinder for further use, Extracted with petroleum ether, chloroform, acetone, methanol. Antihelmentic activity was evaluated on adult Pheritima Posthuma. Earthworms by subjecting to standard drug albendazole at a dose level of 10mg/ml and to the extracts of petroleum ether, chloroform, acetone, methanol at doses of 10mg/ml, 20mg/ml,25mg/ml 30mg/ml and 35mg/ml respectively. Anti-Fungal activity and Anti-bacterial activity was also performed against different bacteria by using disc diffusion method, the zones of inhibition of the extracts using standard as Amoxicillin and Ketoconazole were determined. The powder leaves of Rubus Ursinus plant was subjected to extraction with four solvents petroleum ether, chloroform, acetone, methanol. High yield was obtained by methanol extracts of Rubus Ursinus plants. The qualitative test for methanol extraction was given positive tests for alkaloids, proteins & aminoacids,Tannins,Glycosides,FlavonoidsSaponins,Carbohydrates,Steroids& terpenoids and Phenolic compounds The anti-helmintic study and anti- fungal study of BlackBerry leaves was observed by using the extract of methyl alcohol. The anti-bacterial study of various BlackBerry leaves extracts (petroleum ether, chloroform, acetone, methanol) were found to be effective against various gram positive and gram negative bacteria. The leave part of plant of RUBUS URSINUS was observed for the Pharmacognostic investigation and Potent Anti-helmentic activity and significant Anti-Microbial activity. . The purified components may have even more potency with respect to inhibition of microbes.

Key Words: RUBUS URSINUS, Amoxicillin, Ketoconazole, Albendazole

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INTRODUCTION

The **blackberry** is an edible fruit produced by many species in the *Rubus* genus in the Rosaceae family, hybrids among these species within the *Rubus* subgenus, and hybrids between the *Rubus* and *Idaeobatus* subgenera.

Medicinal plants have been used for centuries as remedies for human diseases because they contain components of therapeutic value. About 75-80% of the world population, mainly in the developing countries still use plant based medicines for primary health care[3]. Based on a large number of chemical and pharmacological research work, numerous bioactive compounds have been found in medicinal plants.

Useful in treatment of sore throats, gum inflammations, Mouth washes, cough remedy, , diarrhea and show anti oxidant property and astringent property.

MATERIALS AND METHOD

The plant RUBUS URSINUS Fresh leaves are collected, identified and authenticated by Mr.Salman Department of Botany , Charminar Nursery TalabKatta and voucher specimen of the plant were preserved at institute herbarium library.

Preparation of Plant Extract

Fresh plants leaves were collected, washed to remove adhered dirt, rinsed with distilled water, blotted and dried in shade. The shade-dried specimens were powdered in a mixer. Various leaf extracts of RUBUS URSINUS were prepared by successive solvent extraction by maceration. The extracts obtained were evaporated under reduced pressure and dried. Anti-bacterial activity by various extracts (petroleum ether, chloroform, acetone, methanol) against various gram positive and gram negative bacteria (E.coli, Staphylococcus aureus and Bacillus subtilis) using a standard as Amoxicillin, Anti-Fungal activity (Candida Albicans,Aspergillus) using Ketoconazole and Anti-Helminthic activity using a standard as Albendazole.

Organoleptic evaluation, transverse section, powder analysis was carried out and Physicochemical parameters i.e. moisture content, Total ash, Acid insoluble ash, Water soluble extractive value, Alcohol soluble extractive value determination were carried out as per WHO Guidelines.

RESULTS AND DISCUSSION

Pharmacognostic study

Organoleptic evaluation

The powder of black berry leaves was studied for their organoleptic characters and the results were shown below.

Color – Greenish Taste – Mucilaginous

Odour – Characteristic

Powder analysis

The rubus ursinus leaves powder has shown the presence of following plant tissue systems under microscope:

Starch grains : Oval shaped

Vascular tissue : Xylem and Phloem

Trichomes : Unicellular covering trichomes along with epidermal cells

Stomata : Dumble shaped

Fibers : Lignified

Calcium oxalate crystals: Prismatic type

Physicochemical parameters of Rubus Ursinus leaves

Sample	Moisture Content %	Total Ash %	Acid Soluble Ash %	Water Soluble ash %	Water soluble extractive value %	Alcohol soluble extractive value %
Leaves	5.05	7.8	3.3	4.5	3.5	4.75

EXTRACTION OF PLANT MATERIAL

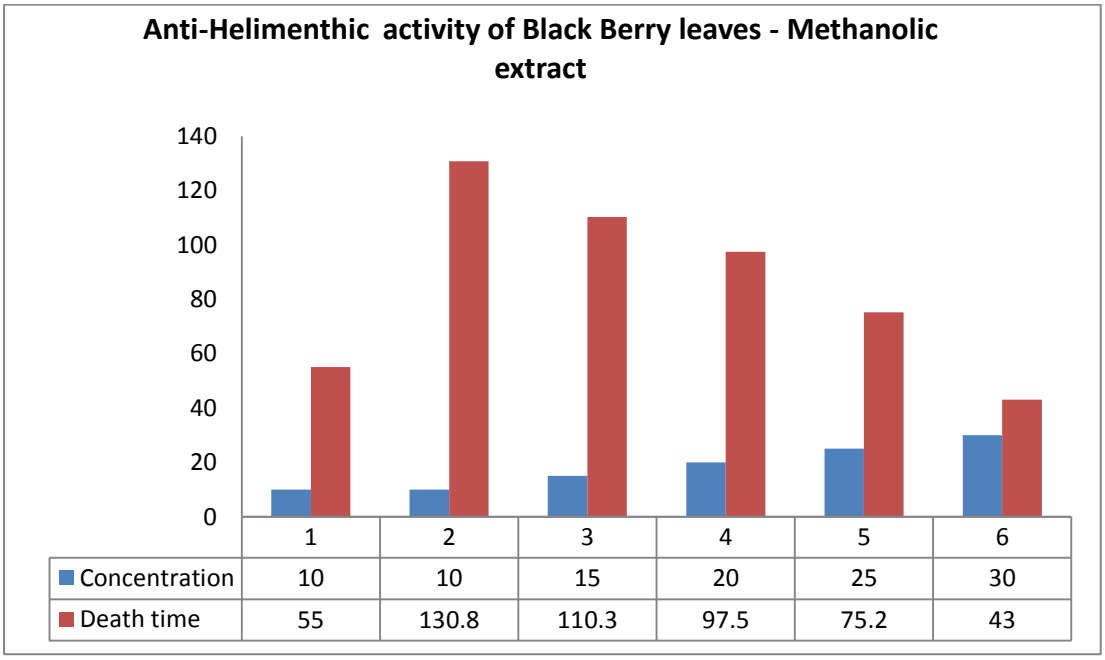
Percentage yield of Rubus Ursinus leaves

S.No	Method of extraction	Solvent	Physical form	Percentage yield
1.	Maceration	Petroleum ether Extract	Resinous	1.204
2.	Maceration	Methanol Extract	Powder	3.853
3 .	Maceration	Acetone Extract	Resinous	1.674
4.	Maceration	Chloroform Extract	Resinous	2.148

Anthelmintic Property

Groups	Treatment	Concentration Used(mg/ml)	Time Taken For Paralysis(min)(X=S.D)	Time Taken For Death(min)(X=S.D)
1	Vehicle Normal Saline	-	-	-
2	Standard (Albendazole)	10	1.9+0.25	39.6+0.2
3	EE 1	10	83.7+0.5	115.21+0.5
4	EE 2	15	7.4 +0.5	105.3+4
6	EE 3	20	61+0.2	92+0.1
7	EE 4	25	41+0.3	62.2
8	EE 5	30	41+0.3	62.2+0.1

Anthelmintic Property of Methanol extract of Rubus Ursinus



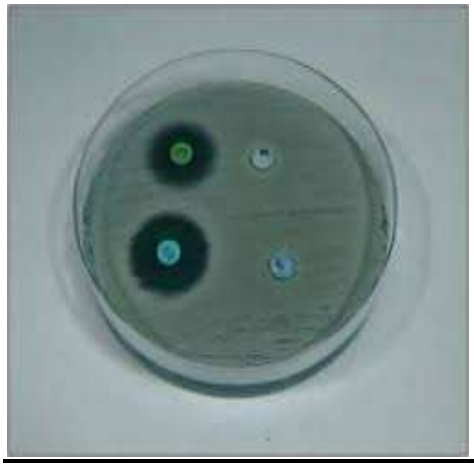
Methanolic Extract of Black Berry

Antifungal Activity of Leaf Extract

S.No	Name of the organism	Diameter of zone of inhibition(mm) Standard Ketaconozole 10ug/ml	Test MEAI 300 ug/ml
1	Candida Albicans	35	30
2	Aspergillus Specius	35	30

ANTI FUNGAL ACTIVITY

Zone Of Inhibition



Aspergillus species

Candida Albicans

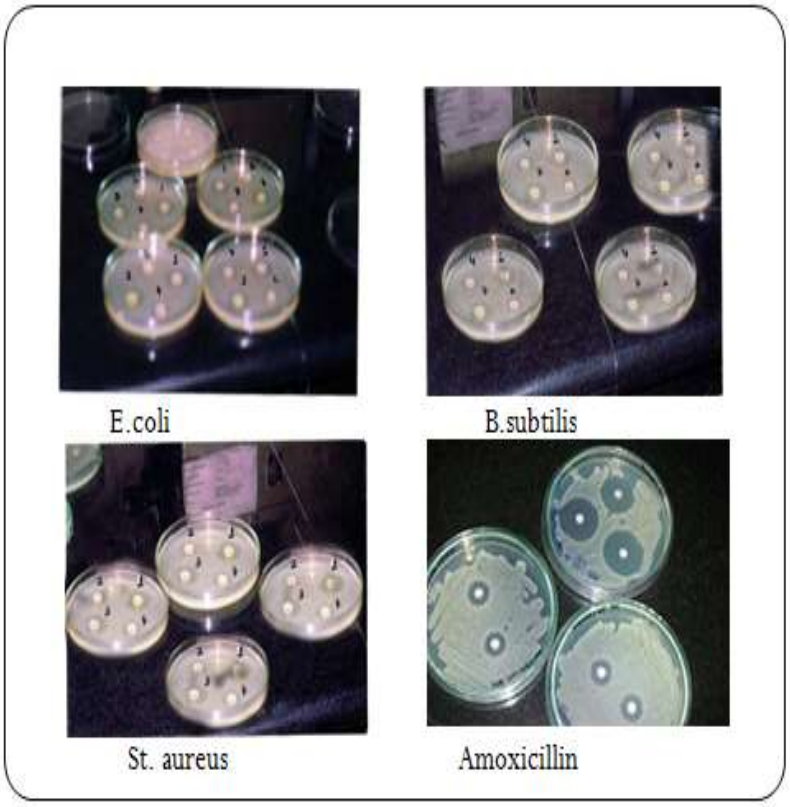
Ketaconazole

Anti Bacterial Activity Of Rubus Ursinus (Leaf Extract)

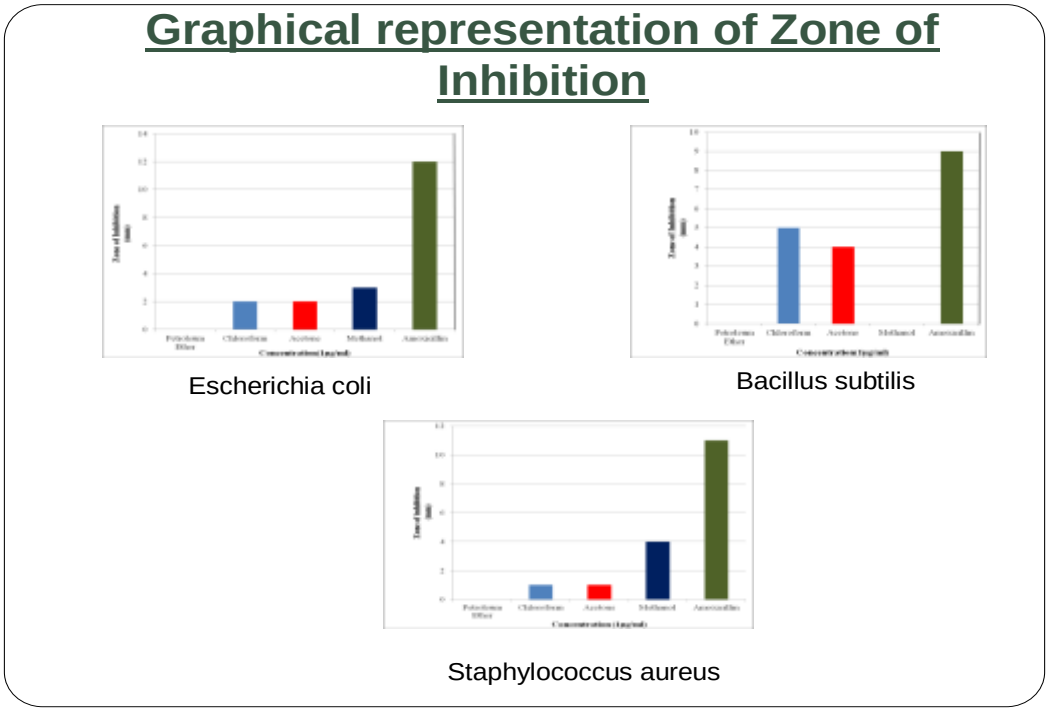
S.no	Micro organism	Formaldehyde (mm)	Chloroform (mm)	Acetic acid(mm)	Ethanol (mm)	Amoxicillin (mm)
1	E.coli	-	2	2	3	12
2	Bacillus subtilis	-	5	4	-	9
3	Streptococcus aureus	-	1	1	4	11

Zone of inhibition

S. No	Name of the organism	Diameter of zone of inhibition (mm)	
		Test300 ug/ml	Standard 10ug/ml
1	Escherichia coli	75	85
2	Bacillus subtilis	60	80
3	Streptococcus aureus	80	85



Zone Of Inhibition



Graphical representation of zone of inhibition

DISCUSSION:

In ethno medicinal practices, the medicinal healers use Black berry leaves to treat dysentery, diarrhea, whooping cough, colitis, labor pains, and toothaches.

In the present study, microscopic evaluation has been documented such as transverse section of Black berry leaves showed an upper epidermis with cuticle, unicellular trichomes, dumble shaped stomata, vascular bundles surrounded by sclerenchymatous tissue.

Macroscopic evaluation has also been documented. Powder analysis showed oval shaped starch grains, vascular bundles, unicellular trichomes, dumble shaped stomata, lignified fibers, and prismatic type calcium oxalate crystals.

The powdered drug was subjected to extraction with various solvents such as formaldehyde, Ethanol, Acetic acid, Chloroform by successive maceration based on polarity and the concentrated extracts (1µg/ml) were used for anti-bacterial assay.

All the extracts obtained from blackberry leaves powder showed mild to strong activity against most of the tested organisms. The results were compared with those of Amoxicillin as standard antibiotic. Of the four extracts, formaldehyde extract did not show any activity against gram positive and gram negative bacteria. Chloroform extract displayed excellent activity against gram positive bacteria Bacillus subtilis whereas acetone extract showed considerable activity against gram positive bacteria Bacillus subtilis. Methanolic extract showed moderate activity against both gram negative bacteria Escherichia coli and gram positive bacteria staphylococcus aureus. On over all consideration, formaldehyde was found inactive compared with other extracts.

The anti-helmintic study of BlackBerry leaves was observed by using the extract of methyl alcohol. When decreasing the concentration of methanol of RUBUS URSINUS, the death time of worms increases. The Methanol extract of RUBUS URSINUS, showed significant paralysis & also cause death of worms at low concentration as compared to standard reference of Albendazole.

The anti- fungal study of BlackBerry leaves was observed by using the extract of methyl alcohol. The methanol extract of RUBUS URSINUS, showed significant anti- fungal at low concentration as compared to standard reference of Ketoconazole.

CONCLUSION:

BlackBerry plant was selected for thesis presentation, the plant was identified based on literature survey, not much work has done on the leaves of this plant, so we selected leaves part for the project to investigate Photochemical and to evaluate Anti-helmentic activity and Anti-Microbial

activity. The leaves part of plant of RUBUS URSINUS was observed for the Pharmacognostic investigation and Anti-helmentic activity and Anti-Microbial activity.

Several phytoconstituents like flavonoids, phenolics and polyphenols, tannins, terpenoids, sesquiterpenes etc are effective anti-helmenthic, anti-fungal and anti-bacterial substances against wide range of micro-organisms. The purified components may have even more potency with respect to inhibition of microbes.

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