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Phytochemical Estimation (Qualitative Estimation) of Leafy Extract of *Asparagus racemosus*

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ABSTRACT

Nature is a preeminent source of structures of high phytochemical diversity, each phytochemical possess enormous and interesting biological activities and therapeutic properties. Plants which having a medicinal value are a rich source of bioactive compounds or bionutrients. Studies on phytochemical nature of medicinal plants elucidated that during the past 2– 3 decades have shown that these phytochemicals have an important role in preventing various chronic diseases like cancer, diabetes and heart diseases. The major classes of phytochemicals with disease-preventing functions are dietary fibre, antioxidants, anticancer, detoxifying agents, immunity-potentiating agents and neuropharmacological agents. Each class of these functional agents consists of a wide range of chemicals with differing efficiencey. There is, however, much scope for further systematic study in screening of medicinal plants for elucidation of phytochemicals and assessing their potentialand ability in protecting against different types of diseases and having therapeutic value (Saxena 2013)[i]

Key Words- Medicinal plants, Phytochemicals, Therapeutic, Screening, Bionutrients

INTRODUCTION

Phytochemicals word derived from the Greek word phyto, meaning is biologically active, naturally occurring chemical compounds found in plants, which provide health benefits for humans (Hasler 1999)[ii]. They protect plants from disease causing pathogens and damage and also contribute to the plant's color, aroma and flavor and they protect plant cells from environmental hazards such as pollution, stress, drought, UV exposure (Mathai 2000)[iii]. Recently, it is clearly elucidated that they have major roles in the protection of human health, when their dietary intake is significant. Nearly 4,000 phytochemicals have been cataloged and discovered and are classified on the basis of protective function, physical characteristics and chemical characteristics.

Phytochemicals accumulate in vegetative and reproductive parts of the plants, such as in the roots, stems, leaves, flowers, fruits or seeds and having biological properties such as antioxidant activity, antimicrobial effect, modulation of detoxification enzymes, stimulation of the immune system, decrease of platelet aggregation and modulation of hormone metabolism and anticancerous property. (Panche 2016)[iv]

Classification of Phytochemicals

Phytochemicals are classified as primary or secondary constituents, depending on their role in plant metabolism. Primary metabolites includes the sugars, amino acids, proteins, nucleic acids, chlorophyll's etc. Secondary metabolites consists of alkaloids, terpenes, flavonoids, lignans, plant steroids, curcumines, saponins, phenolics, flavonoids and glucosides.(Saxena 2013)[v]

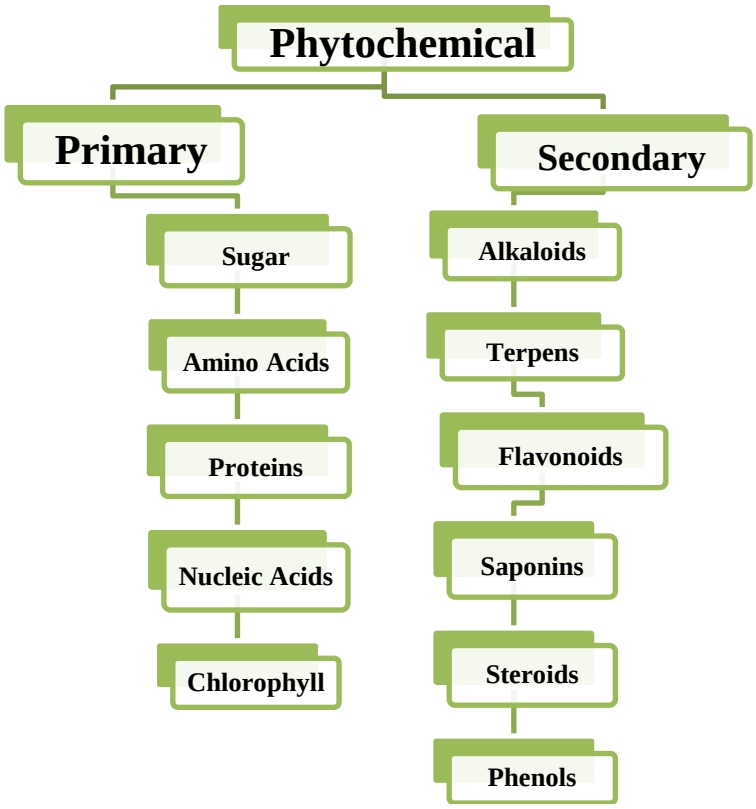


Fig 1: Ray diagram showing metabolites

Material and Methodology

Medicinal plant for Phytochemical estimation (Qualitative analysis)

Asparagus racemosus

Systematic Position of *Asparagus racemosus*

Kingdom- Plantae
Division- Angiosperms
Class- Monocotyledons
Order- Asparagales
Family- Liliaceae
Genus- *Asparagus*
Species- *racemosus*

Fig 2: Plant of *Asparagus*

Morphology of *Asparagus racemosus*

Asparagus racemosus is broadly distributed around various parts of the world. *Asparagus racemosus* is a climber having stems up to 4 m long. Its roots are both fibrous and tuberous. *Asparagus* commonly known as Shatavari has small pine-needle like phylloclades (photosynthetic branches) that are uniform and shiny green. In July, it produces minute, white flowers on short, spiky stems, and in September it fruits, producing blackish-purple, globular berries. (Alok et al 2013)[vi] It has an adventitious root system with tuberous roots that measure about one metre in length, tapering at both ends, with roughly a hundred on each plant. The roots are 30-100 cm in length, 1-2 cm in thickness and yellowish-cream in colour. The roots contain long needle shaped structure known as pith which is meant for the conduction of water.

Collection of Test Material

Fresh leaves of *Asparagus* plant is collected from medicinal garden of Sophia Girls' College when the vegetative growth of plant favourable. The plant materials were taxonomically identified The plant materials were shade dried until all the water molecules evaporated and plants became well dried for grinding. After drying, the plant materials were ground well using mechanical blender into fine powder and transferred into airtight containers with proper labeling for future use.

Preparation of Plant Extract

Preparation of plant leafy extract with the help of Soxhlet apparatus.

The solvent (250 ml of ethanol) is added to a round bottom flask, which is attached to a Soxhlet extractor and condenser on an isomantle. The crushed plant material is loaded into the thimble, which is placed inside the Soxhlet extractor. The side arm is lagged with glass wool. The solvent is heated using the isomantle and will begin to evaporate, moving through the apparatus to the condenser. The condensate then drips into the reservoir containing the thimble. Once the level of solvent reaches the siphon it pours back into the flask and the cycle begins again then extract was kept in refrigerator when not in use.

Qualitative Phytochemical Analysis

The leafy extracts of *Asparagus racemosus* were subjected to different chemical tests for the detection of phytoconstituents such as carbohydrates, glycosides, alkaloids, proteins, amino acids, tannins, phenolics, saponins, flavonoids, triterpenoids, steroids, fixed oils, gums and mucilages.

Table 1 : Test for Phytochemical Analysis of *Asparagus*

S.No	Phytoconsituents	Test for Phytoconsituents
1.	Test for carbohydrates	**Benedict's test**- Take 1ml of a sample solution is placed in a test tube and two ml of Benedict's reagent (a solution of sodium citrate and sodium carbonate mixed with a solution of copper sulfate) is added. The odbtained solution is then heat in a boiling water bath for three minutes. **Fehling's test**- The test material mixed with Fehling's A and B, heated and then examined **Molisch test** - In a test tube containing sample, 2 ml of distilled water and 2 drops of freshly prepared 20% alcoholic solution of alpha naphthol were added and mixed well and then add 2ml of concentrated sulphuric acid along the side of the test tube was added.
2.	Tests for alkaloids	**Dragendroff's test**- In prepared test sample add few drops of potassium bismuth iodide solution **Wagner's test**- The test material was mixed with Wagner's reagent and examined **Mayer's test** - The test material was mixed with little amount of dilute hydrochloric acid and Mayer's reagent and examined
3.	Tests for proteins and amino acids	**Biuret test-**In test tube add 1 ml of sample solution then add 5 to 8 drops of copper sulphate solution (10%) was added. **Xanthoprotein test**- To 1 ml of test solution, add 1ml of concentrated HNO_3Mix and heat (In case of protein solution, initially white precipitate appears due to denaturation of protein, which turns yellow on heating).Add 1 ml of 40% NaOH.
4	Tests for tannins and phenolics	2-3 drops of ferric chloride to 1ml of extract

5	Test for flavonoids	**Shinoda Test**- To the test sample add few piece of magnesium turnings and then add conc.HCl was added drop wise
6	Test for triterpenoids	Dissolving two or three granules of tin metal in 2 ml thionyl chloride solution and then, add 1 ml of the extract into the test tube.
7	Tests for steroids	**Libermann Burchard test**- The sample was treated with few drops of acetic anhydride, boiled and cooled. Conc.sulphuric acid was added from the sides of the test tube. **Salkowski test** - The sample was treated with few drops of conc. sulphuric acid
8	Test for saponins	1 ml of extract which is diluted with 20 ml distilled water and then shaken in a graduated cylinder for 15 minutes.
9	Tests for glycosides	**Keller Kiliani Test**- In 10 ml of test material a solution of glacial acetic acid (4.0 ml) with 1 drop of 2.0% $FeCl_3$ mixture was mixed and then 1 ml conc H_2SO_4.
10	Test for gums	Hydrolyzing the 1 ml of extract using dil. HCl (3ml). Then Fehling's solution is added drop by drop
11	Test for mucilages	Treating 1 ml of extract with 2 ml of ruthenium red solution

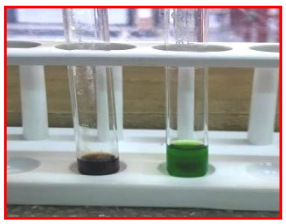
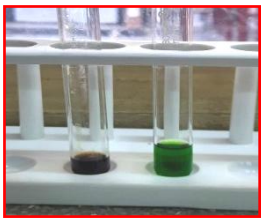
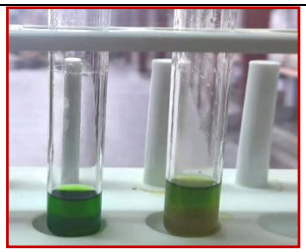
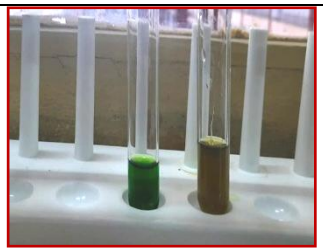
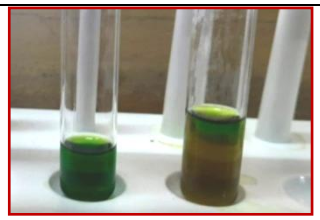
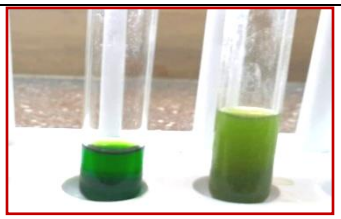
RESULT

Table 2: Result of Phytochemical Analysis of *Asparagus*

S.No	**Phytoconstituents**	**Observation**
1.	**Test for carbohydrates** - Benedict's test - Fehling Test	 No formation of a reddish precipitate No Appearance of red colouration
2.	**Tests for alkaloids** - Dragendroff's test - Wagner Test	Formation of red colouration Red Colour ppt formation
3.	**Tests for proteins and amino acids** - Biuret Test - Xanthoprotein Test	No ppt formation No ppt formation
4	**Tests for tannins and phenolics**	Formation of a yellow or brownish black colour
5	**Test for flavonoids** - NaOH Test - H_2SO_4 Test	Formation of Yellow Orange Colour Formation of Orange Colour

6	**Test for triterpenoids**	Formation of a pink colour
7	**Tests for steroids** Libermann Burchard Test Salkowski test	Brown ring is formed at the junction of two layers and upper layer turns green Formation of red colour at lower layer
8	**Test for saponins**	No Formation of Foam Layer
9	**Tests for glycosides** Keller Kiliani Test-	A brown ring formed between the layers
10	**Test for gums**	No Formation of red colour
11	**Test for mucilages**	Formation of red colour

Table 3: Observation of Phytochemical Analysis

S.No	Phytochemical	Observation	
1.	**Alkaloid Test** • **Dragendoff Test** • **Wagner Test**	 **Dragendoff Test**	 **Wagner Test**
2.	**Flavonoids Test** • **NaOH Test** • **H_2SO_4 Test**	 **NaOH Test**	 **H_2SO_4 Test**
3.	**Tannins** • **Ferric chloride test** • **Lead acetate test**	 **Ferric Chloride Test**	 **Lead Acetate Test**

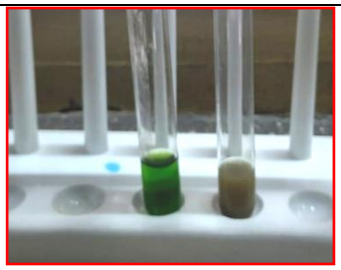
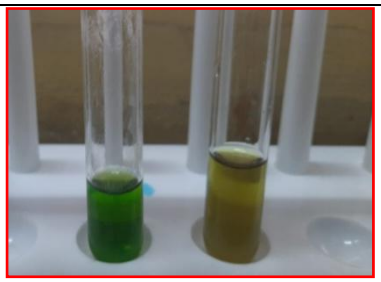
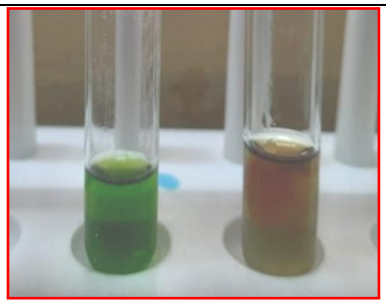
4.	**Triterpenoids**	
5.	**Steroids**	
6.	**Mucilages**	

Table 4: Result of Qualitative Estimation

S.No	Phytochemical	Result
1	**Test for carbohydrates**	--
2	**Tests for alkaloids**	++
3	**Tests for proteins and amino acids**	- -
4	**Tests for tannins and phenolics**	++
5	**Test for flavonoids**	++
6	**Test for triterpenoids**	++
7	**Tests for steroids**	++
8	**Test for saponins**	++
9	**Tests for glycosides**	-
10	**Test for gums**	-
11	**Test for mucilages**	++

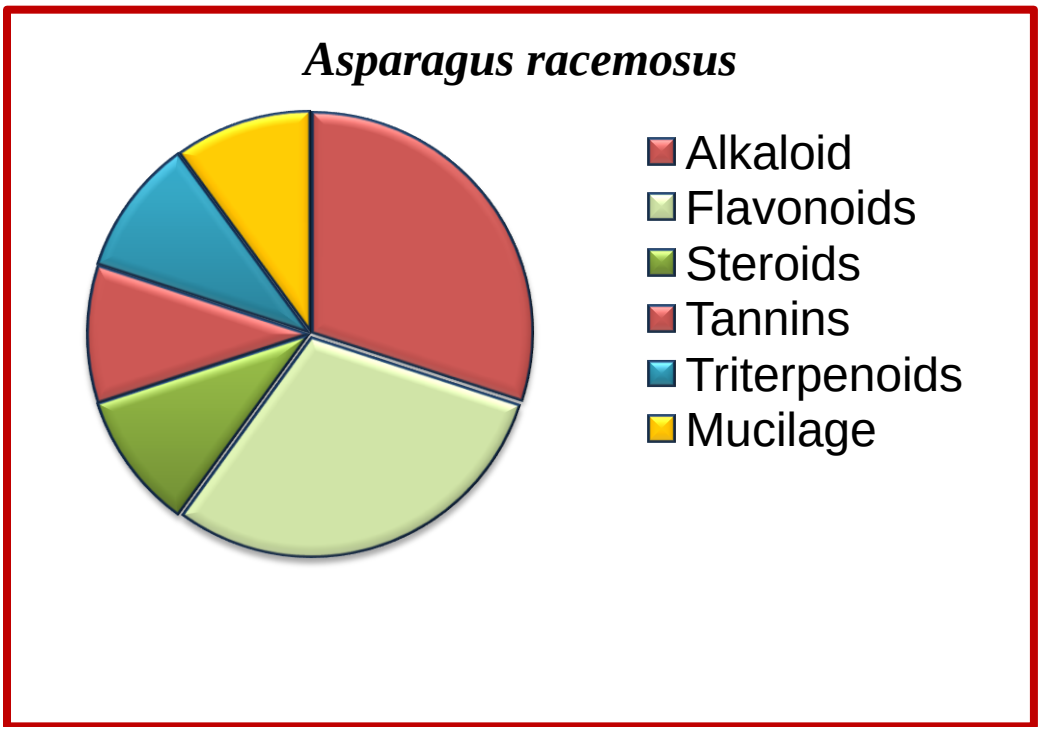


Fig 3: **Pie diagram showing phytochemicals in *Asparagus***

DISCUSSION

The study of *Asparagus* plant revealed that the presence of phytochemicals considered as active medicinal chemical constituents and phytochemical constituents which are estimated from test material having presence of tannins, flavonoids, alkaloids, steroids, triterpenoids and mucilage. The results were summarized in Table 1& 2. In our studies it was investigated that alkaloids and flavonoids are present in rich amount and mainly responsible for medicinal properties. Alkaloids and flavonoids possess a variety of biological properties, namely, being antioxidants, immunostimulants, antihepatotoxic, antibacterial, useful in diabetic retinopathy, anticarcinogenic, antidiarrheal, antiulcerogenic, antioxytocic, and reproductive agents and also known to be antimicrobial to inhibit mould and to protect plants from insects. They may be considered as defense system and have been included in a large group of protective molecules found in plants named phytoanticipins or phytoprotectants.

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