



PHYTOCHEMICAL INVESTIGATION OF ASPARAGUS RACEMOSUS (WHOLE PLANT) AND CALENDULA OFFICINALIS (FLOWER)

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ABSTRACT:

Asparagus racemosus is an essential medicinal plant known as a 'rasayana' that promotes overall well-being by boosting cellular vitality and resistance. It has traditionally been used in India as a tonic, fever, tumors, and internal pain. *Calendula officinalis* Linn is a medicinal plant that has been used for the treatment of a variety of diseases. The plant's flowers have a wide spectrum of biological effects, some of which are very promising for future development. The essential oil of the plant flowers has been tested for its anti-inflammatory, antifungal, antitumor, and anticancer activities. The methanol extract of the plants flowers showed the presence of active metabolites such as Alkaloids, flavonoids, proteins, saponins, glycosides and phenols. The total phenolic (TPC) and total flavonoid (TFC) assays were performed. In the acute toxicity study, no signs of toxicity were found up to the dose of 2000 mg/kg body weight. It is advisable that the persons who have an established allergy to the Asteraceae (daisy) family should use it with caution.

KEYWORDS:

ASPARAGUS RACEMOSUS, CALENDULA OFFICINALIS, PHYTOCHEMICAL INVESTIGATION, SOXHLET EXTRACTION METHOD, TOTAL PHENOLIC CONTENT, TOTAL FLAVONOID CONTENT.

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INTRODUCTION

Herbal medicine, as defined by the World Health Organization (WHO), is a practice involving herbs, herbal materials, preparations, and finished products containing active substances derived from plants, other plant materials, or mixtures.^[1] Herbal medications are made up of active substances, plant parts, or plant materials, as well as excipients such as dilutions, solvents, or preservatives.^[2] Phytochemicals, such as saponins, flavonoids, glycosides, tannins, alkaloids, and terpenoids, have been scientifically confirmed to provide health benefits. Ayurvedic botanicals play an essential role in wound healing, as they boost repair mechanisms and preserve beauty. Over 70% of wound healing pharmaceutical medicines are plant-based, with 20% being mineral-based.^[3] Plant-based products are used in first aid as antiseptic coagulants and wound wash. Over 13,000 plants have been researched over the past five years, demonstrating the enormous potential of medicinal plants in diverse traditional systems.^{[4][5][6]}

ASPARAGUS RACEMOSUS:

Asparagus racemosus is an essential medicinal plant known as a 'rasayana' that promotes overall well-being by boosting cellular vitality and resistance. It has traditionally been used in India as a tonic, fever, tumors, and internal pain. It contains a wide variety of chemicals, including steroidal saponins, alkaloids, flavonoids,

dihydrophenanthrene derivatives, furan derivatives, and volatile compounds. In immunocompromised animals, supplementation increased cell-mediated immune response and potentiated antibody synthesis.^{[7][8][9]}

CALENDULA OFFICINALIS:

Calendula officinalis Linn is a medicinal plant that has been used for the treatment of a variety of diseases. The plant's flowers have a wide spectrum of biological effects, some of which are very promising for future development. The methanol extract of the flowers of the plant has been tested for its anti-inflammatory, antifungal, antitumor, and anticancer activities. The essential oil of the plants flowers has been found to be active against various bacteria, yeasts, fungi, and fungal strains. In vitro, calendulose F6'-O-n-butyl ester has shown potent in vitro inhibition of leukemia (MOLT-4 and RPMI 8226), colon cancer (HCC-2998), and melanoma (LOXIMVI, SK-MEL-5 and UACC62) cell lines with GI50 values of 0.77-0.99 mole, except for ovarian cancer (AK-1 and UO-31) and breast cancer (NCI/ADR-RES), and LACE has been shown to cause allergy in 9 patients out of 443 (2.03 %) when assessed by patch testing method (Reider et al., 2001). In conclusion, *C. officinalis* flowers are a promising source of natural products with pharmacological and therapeutic applications. It is advisable that the persons who have an established allergy to the Asteraceae (daisy) family should

use it with caution.^{[10][11][12]}

MATERIALS AND METHOD USED:

COLLECTION AND AUTHENTICATION OF PLANT MATERIAL:

Collection and authentication of medicinal plants *Asparagus racemosus* and *calendula officinalis* to confirm identity and purity.

EXTRACTION OF PLANT BY SOXHLET EXTRACTION METHOD:

Extraction of *Asparagus racemosus* and *calendula officinalis* by soxhlet extraction method, dried residue stored in air-tight container.

PHYTOCHEMICAL INVESTIGATION

The tests used to detect the presence of *Asparagus racemosus* and *calendula officinalis* extracts. These tests include Molisch's Test, Fehling's Test, Benedict's Test, Barfoe's Test, Hager's Test, Froth Test, Triterpenoids and Steroids Test, Libermann-Burchard Test, Salkowski Test, Tannin and Flavonoid Compounds Test, Ferric Chloride Test, Gelatin Test, and Lead Acetate Test. Molisch's Test indicates the presence of carbohydrates, Fehling's Test indicates the presence of reducing sugar, Benedict's Test indicates the presence of alkaloids, Wagner's Test indicates the presence of alkaloids, Mayer's Test indicates the presence of saponins, Libermann-Burchard Test indicates the presence of triterpenoids and steroids,

Salkowski Test indicates the presence of steroids, Ferric Chloride Test indicates the presence of tannins, Gelatin Test indicates the presence of phenolic compounds, and Lead Acetate Test indicates the presence.^[13]

TOTAL PHENOLIC CONTENT AND TOTAL FLAVONOID CONTENT:

The total phenolic content of *Asparagus racemosus* and *calendula officinalis* extracts was determined using the Folin-Ciocalteu Assay. Calibration curves were composed using standard solutions of Gallic Acid Equivalent (GAE) mg/gm. The total flavonoid content was determined using the Aluminium chloride method. Absorbance of the mixture was estimated at 510 nm using UV spectrophotometer. Calibration curves were composed using standard solutions of Rutin Equivalent (RE) mg/gm. The results were expressed as mg Rutin equivalent per gram dry extract weight.^{[14][15]}

RESULT:

PERCENT AGEYIELD

In phytochemical extraction the percentage yield is very crucial in order to determine the standard efficiency of extraction for a specific plant, various sections of the same plant or different solvents used. The yield of extracts received from the *Asparagus racemosus* and *calendula officinalis* is shown in Table: 5

TABLE 1: PERCENTAGE YIELD OF CRUDE EXTRACTS OF ASPARAGUS RACEMOSUS AND CALENDULA OFFICINALIS EXTRACT

S.No	Plant name	Solvent	Theoretical Weight	Yield(gm)	%yield
1	<i>Asparagus racemosus</i>	Pet ether	299	1.49	0.49%
2		Methanol	310	5.22	1.68%

S.No	Plant name	Solvent	Theoretical Weight	Yield(gm)	%yield
1	<i>Calendula Officinalis</i>	Pet ether	298	1.31	0.43%
2		Methanol	322	5.09	1.58%

PRELIMINARY PHYTOCHEMICAL STUDY:

TABLE 2: PHYTOCHEMICAL TESTING OF ASPARAGUS RACEMOSUS

S.No.	Experiment	Presence or absence of phytochemical test	
		Pet.Ether extract	Methanolic extract
1.	Alkaloids		
1.1	Dragendroff's test	Absent	Present
1.2	Mayer's reagent test	Absent	Present
1.3	Wagner's reagent test	Absent	Present

1.3	Hager's reagent test	Absent	Present
2.	Glycoside		
2.1	Borntrager test	Present	Present
2.2	Legal's test	Present	Present
2.3	Killer-Killiani test	Present	Present
3.	Carbohy drates		
3.1	Molish's test	Absent	Absent
3.2	Fehling's test	Absent	Absent
3.3	Benedict's test	Absent	Absent
3.4	Barfoed's test	Absent	Absent
4.	Proteinsand Amino Acids		
4.1	Biuret test	Absent	Present
5.	Flavonoids		
5.1	Alkaline reagent test	Absent	Present
5.2	Lead Acetate test	Absent	Present
6.	Tanninand Phenolic Compounds		
6.1	Ferric Chloride test	Absent	Present
7.	Saponin		
7.1	Foam test	Absent	Present
8.	Test for Triterpenoids and Steroids		
8.1	Salkowski's test	Absent	Absent
8.2	Libbermann-Burchard's test	Absent	Absent

TABLE 3: PHYTOCHEMICAL TESTING OF CALENDULA OFFICINALIS

S.No.	Experiment	Presence or absence of phytochemical test	
		Pet. Ether extract	Methanolic extract
1.	Alkaloids		
1.1	Dragendroff's test	Absent	Absent
1.2	Mayer's reagent test	Absent	Absent
1.3	Wagner's reagent test	Absent	Absent
1.3	Hager's reagent test	Absent	Absent
2.	Glycoside		
2.1	Borntrager test	Absent	Absent
2.2	Legal's test	Absent	Absent
2.3	Killer-Killiani test	Absent	Absent
3.	Carbohydrates		
3.1	Molish's test	Present	Present

3.2	Fehling's test	Present	Present
3.3	Benedict's test	Present	Present
3.4	Barfoed's test	Present	Present
4.	Proteins and Amino Acids		
4.1	Biuret test	Absent	Absent
5.	Flavonoids		
5.1	Alkaline reagent test	Absent	Present
5.2	Lead Acetate test	Absent	Present
6.	Tannin and Phenolic Compounds		
6.1	Ferric Chloride test	Absent	Present
7.	Saponin		
7.1	Foam test	Absent	Absent
8.	Test for Triterpenoids and Steroids		
8.1	Salkowski's test	Absent	Present
8.2	Libbermann-Burchard's test	Absent	Present

QUANTITATIVE ANALYSIS PRELIMINARY:

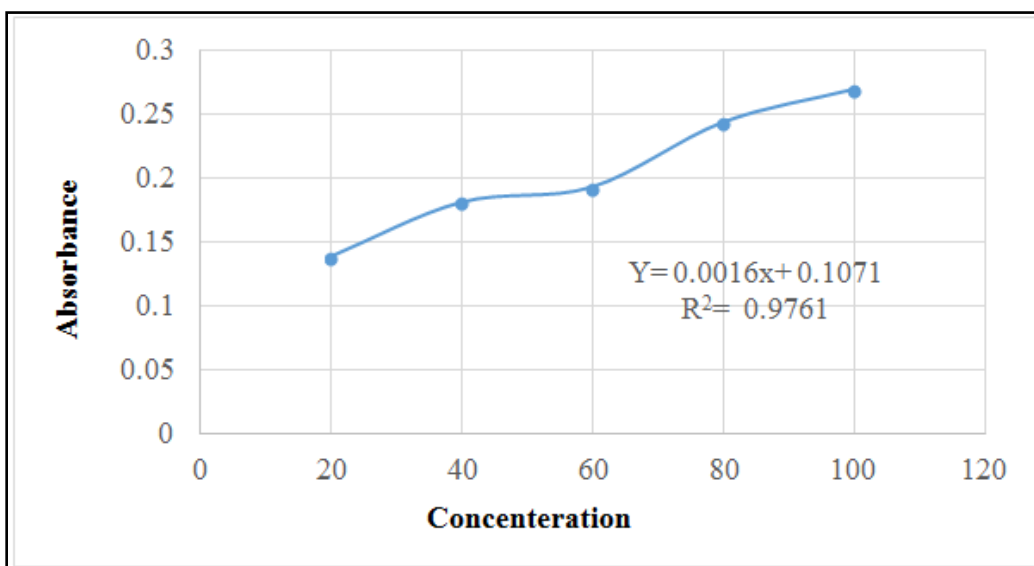
Phytochemical testing of crude extracts confirmed the presence of phenolics and flavonoids in plant material. To

estimate their amount total phenolic (TPC) and total flavonoid content (TFC) assays were performed.

TOTAL PHENOLIC CONTENT (TPC) ESTIMATION

TABLE 4: STANDARD TABLE FOR GALLIC ACID

S.No.	Concentration(µg/ml)	Absorbance
1.	20	0.138
2.	40	0.181
3.	60	0.193
4.	80	0.244
5.	100	0.270



GRAPH 1: REPRESENT STANDARD CURVE OF GALLIC ACID

TOTAL PHENOLIC CONTENT IN EXTRACT

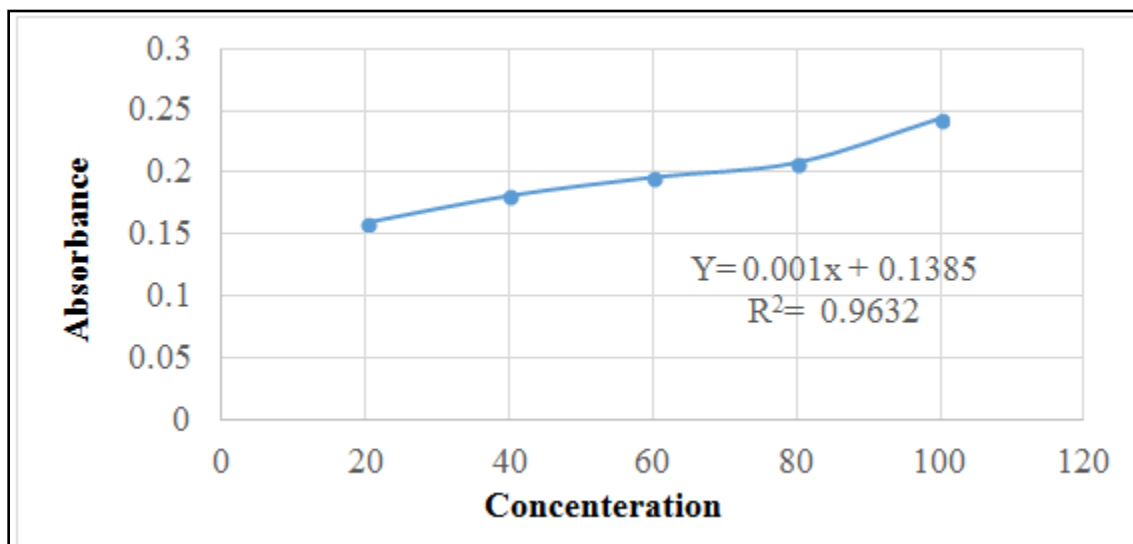
TABLE 5: TOTAL PHENOLIC CONTENT IN EXTRACTS

Extracts	Total phenolic content (mg/gm equivalent to gallic acid)	
	<i>Asparagus racemosus</i>	<i>Calendula officinalis</i>
Absorbance(mean ± SD)	0.189±0.08	0.199±0.10
TPC	84	92

TOTAL FLAVONOIDS CONTENT (TFC) ESTIMATION

TABLE 6: STANDARD TABLE FOR RUTIN

S.No.	Concentration (µg/ml)	Absorbance
1.	20	0.159
2.	40	0.181
3.	60	0.196
4.	80	0.208
5.	100	0.244



GRAPH 2: REPRESENT STANDARD CURVE OF GALLIC ACID

TOTAL FLAVONOID CONTENT IN EXTRACT

TABLE 7: TOTAL FLAVONOID CONTENT IN EXTRACTS

Extracts	Total flavonoids content (mg/gm equivalent torutin)	
	<i>Asparagus racemosus</i>	<i>Calendula officinalis</i>
Absorbance(mean ± SD)	0.160±0.009	0.180±0.011
TFC	22	42

DISCUSSION:

Qualitative phytochemical screening of *Asparagus racemosus* is showed the presence of active metabolites such as Alkaloids, flavonoids, proteins, saponins, glycosides and phenols is presented and *Calendula officinalis* showed the presence of active metabolites such as flavonoids, steroids, carbohydrates, phenols, proteins and amino acid.

Quantitative phytochemical assay was performed by calculating total phenolic content (TPC) and total flavonoid content (TFC). The TPC was computed using gallic acid as the standard, while the TFC was computed using Rutin as the standard. In the acute toxicity study, nosigns of toxicity were found up to the dose of 2000 mg/kg body weight. Hence 1/10th and 1/5thdosesi.e.500mg/kg have been

fixed for study.

CONCLUSION:

The study analyzed the phytochemical properties of *Asparagus racemosus* and *calendula officinalis* extracts using various tests. The results showed that the extracts contained carbohydrates, alkaloids, saponins, triterpenoids, steroids, tannins, phenolic compounds, and lead acetate. The total phenolic content was determined using the Folin-Ciocalteu Assay, while the total flavonoid content was determined using the Aluminium chloride method. The percentage yield was crucial in determining the efficiency of extraction for specific plants or solvents. The results were expressed as mg Rutin equivalent per gram dry extract weight.

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CONFLICT OF INTEREST:

The authors declare that they have no conflict of interest regarding the research, data analysis and publication of this paper. No financial or personal relationships with other people or organizations that could influence the work have influenced this research.

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