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VARIOUS EXTRACTION AND PHYTOCHEMICAL INVESTIGATION OF FOLK MEDICINAL PLANT OF *ANDROGRAPHIS PANICULATA*

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ABSTRACT

Traditional medicine is the knowledge, skills and skills inherent in various cultures that are used to maintain health and prevent, diagnose, ameliorate, or treat physical and mental ailments, is practice. Traditional Chinese Medicine (TCM) is an important example of how ancient accumulated knowledge is being applied to today's holistic approach to medicine. Andrographis (*Andrographis paniculate*) is a plant native to South Asian countries. Andrographis leaves and stems may stimulate the immune system. It may also prevent the influenza virus from attaching to cells in the body. Andrographis is commonly used for common colds, osteoarthritis, throat and tonsil infections and an intestinal disorder called ulcerative colitis. It is also used for many other conditions, but there is not enough scientific evidence for its other uses. Weak preliminary evidence suggests that *Andrographis paniculata* has anti-inflammatory, urinary tract anti-inflammatory and anti-allergic effects, may regulate blood sugar levels and reduce jaundice. Preliminary phytochemical studies have revealed the presence of alkaloids, phenols, flavonoids, tannins, terpenoids and saponins.

KEYWORDS

Andrographis paniculata, Alkaloids, Phenols, Flavonoids, Tannins, Terpenoids and Saponins.

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INTRODUCTION

Preparing medicinal plants for experimental purposes is the first step and key to achieving high-quality research results. This includes extraction of bioactive components and determination of quality and quantity before proceeding with the desired biological test. The main objective of this study was to evaluate different methods used for medicinal plant production and screening in routine research[1-2]. Extracts, bioactive fractions, or compounds obtained

from medicinal plants are used for a variety of purposes, but the techniques for their preparation are generally the same, regardless of the intended biological test[3-6]. Critical steps to obtain high-quality bioactive molecules are the selection of appropriate solvents, extraction methods, phytochemical screening methods, fractionation methods, and identification techniques. The details and exact roadmap of these methods will depend entirely on the study design. When extracting medicinal plants, usually polar solvents (e.g. water, alcohol), medium polar solvents (e.g. isopropanol, acetone, dichloromethane) and non-polar solvents (e.g. n-hexane, benzene, ether, chloroform, DMSO) isused[7-9]. Generally, extraction methods include maceration, digestion, decoction, infusion, infusion, Soxhlet extraction, surface extraction, ultrasound-assisted and microwave-assisted extraction. *Andrographis paniculata* this plant grows as an upright herb to 30to110cm (12 to 43inches) in height in moist, shady locations. The slender stems are dark green, square in cross section, with longitudinal grooves and wings at the corners[10-12]. The lanceolate leaves have glabrous petioles up to 8cm (3.1 in) long and 2.5cm (0.98 in) long. The small flowers are pink, solitary, and arranged in loosely spreading panicles or panicles[13]. The fruit is a capsule approximately 2cm long and several millimeters wide. It contains many yellow-brown seeds. The seeds are square, coarse and glabrous[14-16]. Subramaniam *et al*, (2015) stated that the extract improved recovery from CCl-induced liver injury in albino rats. Suzuki *et al*, (2016) used methanol extracts from leaves obtained by partitioning in EtOAc followed by silica gelchromatography. Banerjee *et al*, (2017) used various organic solvents to extract A. paniculata against Pseudomonasaeruginosa infection. Shaikh *et al*, (2019) Degreasing with hexane and fractionation with chloroform and methanol. The aim of this study is to describe different extraction methods and different polar and non-polar solvents used for the extraction of active plant components and to compare simple and efficient extraction methods for further studies.

MATERIAL AND METHODS

Plant collection

Arial parts of *Andrographis paniculate* it's collected from medicinal plant garden were growing in the Cheran College of Pharmacy, Coimbatore, Tamilnadu, India.

Materials

Petroleum ether, Hexane, Chloroform, Ethyl acetate, Methanol, Distilled water, Hydrogen peroxide, Griess reagent, DPPH (2, 2 -diphenyl-1-picrylhydrazyl), Hydrochloric acid, Sulphuric acid, Alpha-naphthol, Copper sulphate, Sodium hydroxide, Barfoed's solution, Benedict's solution, Potassium mercuric iodide, Potassium bismuth iodide, Iodine, Potassium iodide, Picric acid, Con.HNO3, NH4OH, Millon's reagent, Ninhydrin, Biuret reagent, Ammonia, 95% Ethanol, lead acetate, Potassium hydroxide, Phenolphthalein, Lead acetate, Ferric chloride, Agar-Agar, Potassium dihydrogen phosphate, Calcium carbonate, All the chemical purchased from Ranchem, Pure Chemicals in AR Grade. Rotary vaccum.

Extraction

There are number of extraction techniques used to extract crude drugs, among these here we utilized some few methods like viz., Distillation Method, Maceration Method, Percolation Method, Soxhlet extraction Method.

Soxhlet extraction Method

A dried and blended crude drug is placed in SOX thimble. Solvent is heated under reflux. Condensation and extraction with "fresh" solvent. Solutes are transferred from the extraction chamber into the reservoir. Continuous repetition of the extraction. Exhaustive extraction is complete.

Maceration Method

Petroleum ether extraction

The Dried whole plant of *Andrographis paniculate* (1000gm) was extracted by macerating with 2.0 litter of Petroleum ether for 10 days at room temperature in a dark cabinet. After 10 days, the extracts were collected by filtration, the marc was separated. Solvent extract was evaporated to dryness using rotary vacuum evaporator and weighed.

Hexane extraction

The residue of petroleum ether extraction was dried at sun shade in dark place. After dried residue were extracted by macerating with 2.0 litter of hexane for 10 days at room temperature in a dark cabinet. After 10 days, the extracts were collected by filtration, the marc was separated. Solvent extract was evaporated to dryness using rotary vacuum evaporator and weighed.

Chloroform extraction

The residue of hexane extraction was dried at sun shade in dark place. After dried residue were extracted by macerating with 2.0 litter of chloroform for 10 days at room temperature in a dark cabinet. After 10 days, the extracts were collected by filtration, the marc was separated. Solvent extract was evaporated to dryness using rotary vacuum evaporator and weighed.

Ethyl acetate extraction

The residue of chloroform extraction was dried at sun shade in dark place. After dried residue were extracted by macerating with 2.0 litter of ethyl acetate for 10 days at room temperature in a dark cabinet. After 10 days, the extracts were collected by filtration, the marc was separated. Solvent extract was evaporated to dryness using rotary vacuum evaporator and weighed.

Methanol extraction

The residue of ethyl acetate extraction was dried at sun shade in dark place. After dried residue were extracted by macerating with 2.0 liter of methanol for 10 days at room temperature in a dark cabinet. After 10 days, the extracts were collected by filtration, the marc was separated. Solvent extract was evaporated to dryness using rotary vacuum evaporator and weighed.

Distilled water extraction

The residue of methanol extraction was dried at sun shade in dark place. After dried residue were extracted by macerating with 2.0 liter of distilled water for 10 days at room temperature in a dark cabinet. After 10 days, the extracts were collected by filtration, the marc was separated. Solvent extract was evaporated to dryness using rotary vacuum evaporator and weighed.

RESULTS AND DISCUSSION

Preliminary phytochemical screening

The Arial part of plants extract of *Andrographis paniculate* were examined preliminary phytochemical screening of the entire test conducted and the result was showed in Table No.1.

Determination of total ash

In a tarred platinum or silica dish, approximately 2grams of air-dried crude drug were accurately weighed before being incinerated at a temperature not exceeding 4500 degrees Celsius until carbon was removed, after which it was cooled and weighed. The drug was air-dried when the percentage of ash was calculated. Results are showed on Table No.2.

Determination of water insoluble ASH

The total ash was boiled for 5.0 min, with 25.0ml of water. The insoluble matter was collected in a gooch crucible or an ash less filter paper. It was washed with hot water and ignited for 15min, at a temperature not exceeding 4500C. The weight of the insoluble matter was subtracted from the weight of the ash, the difference in the weight of the ash, represent the water soluble ash. The percentage of water soluble ash calculated with reference to the air dried drug.

Determination of acid insolulbe ASH

The was boiled with 25ml of 2M HCL for 15min. the insoluble matter was collected in a Gooch crucible or an ash less filter paper. It was washed with hot water and ignited; it was then cooled in a desiccators and weighed. The percentage of water soluble ash calculated with reference to the air dried drug. Results are showed on Table No.2.

Alcohol soluble extractive

Took 5grams of the powder were macerated for 24 hours in a closed flask with 100 milliliters of the specified strength of alcohol, shaking frequently for 6 hours and left to stand for 18 hours. It was quickly filtered to prevent alcohol loss, and 25 milliliters of the filtrate were weighed after being evaporated to dryness at 105 degrees Celsius. With regard to the drug that had been air dried, the percentage of water-soluble extractive was gathered.

Water soluble extractive

In a closed flask, 5g of the powder was macerated with 100ml of water for 24 hours, shaking frequently

for 6 hours, and allowed to stand for 18 hours. It was quickly filtered to prevent alcohol loss, and 25 milliliters of the filtrate were weighed after being evaporated to dryness at 105 degrees Celsius. With regard to the drug that had been air dried, the percentage of water-soluble extractive was gathered.

Loss on drying

The percentage of weight lost after drying is referred to as the loss on drying procedure. To be used in the determination, a glass weighing bottle with a stopper that had been dried for 30 minutes in the same conditions was weighed. The bottle and its contents were precisely weighed after the sample was placed inside and covered. The sample was evenly distributed up to a maximum depth of 10mm. The stopper was removed before the loaded bottle was placed in the oven's drying chamber. At a temperature of 110 degrees Celsius in hot air, the sample was dried to a constant weight. Results are showed on Table No.2.

Table No.1: Preliminary phytochemical screening (preliminary-chemical test)

S.No	Test	Result
1	Test for carbohydraes Test for starch	(+) (+)
2	Tests for proteins and aminoacids	(+)
3	Test for alkaloids	(-)
4	Test for flavonoids	(+)
5	Test for tannins	(+)
6	Test for phytosterols	(+)
7	Test for saponons	(+)
8	Test for glycosides	(+)
9	Test for quinones	(-)
10	Test for anthocyanins	(-)

Presence = (+); Absent= (-)

Table No.2: Results of physiochemical evaluation

S.No	Physiochemical Parameter	Experiments		
		I (% w/w)	II (% w/w)	Average (% w/w)
1	Total ash	18.0	17.0	17.5
2	Water insoluble ash	8.8	8.6	8.7
3	Acid insoluble Ash	17.2	16.8	17.0
4	Loss on drying	23.0	20.0	21.5

CONCLUSION

For this study were extraction carried out with where's polar and nonpolar solvents, among this Petroleum ether, Ethyl acetate, Hexane, Chloroform were produced more extraction value. From the preliminary phytochemical study, the primary active constituent of Alkaloides, Flavonoides, Saponin, Terpenoide, Tannin, Glycosides, Phytosterol and Proteins, obtained from the arialpart extraction of *Andrographis paniculate*. The entire part of *Andrographis paniculate* is traditionally used for common cold, diarrhoea, fever due to several infective cause, jaundice, male infertility, musculoskeletal disorders, autoimmune diseases and inflammation.

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CONFLICT OF INTEREST

The entire author's declared as no conflict of interests.

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