



Phytochemical screening of ethanolic extract of leaves and stems of *Cucubita pepo* Linn.

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Abstract

The plants contain several chemicals known as phytochemical constituents. Preliminary phytochemical screening was performed on ethanolic extract of *Cucurbita pepo* Linn. by standard methods. The phytochemical screening revealed the presence of reducing sugars, alkaloids, flavonoids, tannins, gums, glycosides and terpenoids. The findings of the study concluded that the leaves and stems extracts may have potential bioactive substances that may be used to discover new and most potent drugs.

Keywords: phytochemical screening, *cucurbita pepo* Linn. ethanolic extracts, leaves and stems

Introduction

Cucurbita pepo Linn. an edible herbs of Bangladesh belongs to the family cucurbitaceae commonly known as Misti Cumra, Pumpkin, Vegetable marrow, Crookneck squash, Vegetable spaghetti, Crookneck squash, Crookneck squash, Spaghetti melon, Squash. The plants are typically 1-2.5 feet high, 2-3 feet wide, and have yellow flowers [5]. The leaves of *cucurbita pepo* Linn. simple and Foliolate and sessile or subsessile. Stem is striate, sparsely pubescent and with capitate glandular hairs; tendrils present. Traditionally the pumpkin seeds are used in treating parasites and worms, bladder infections. Leaves are used as a painkiller, a treatment for nausea, and a boost to haemoglobin content of the blood [6]. The herbs are also used in traditional folk medicine to treat cold and alleviate ache [7]. Upon sufficient literature survey on the different plants (*Citrullus lanatus*, *Momordica charantia*, *Cucumis sativus*) of Cucurbitaceae family it is found that the seed, leaves and stem was used for research work. In case of *Cucurbita pepo* several researcher found active component from the seed and pulp of *Cucurbita pepo* Linn. that have antidepressant, antiulcerant, hypoglycemic, hypolipidemic and cytotoxic effects. But the plant's leaves and stems was not much scientifically explored. That's why to screen it bioactive compound through chemical test ethanolic extract of leaves and stem was used.

Materials and Method

Collection of plants

For this present investigation the plant *Cucurbita pepo* Linn. (Family: Cucurbitaceae) was collected from Khulna, Bangladesh on October, 2013 and was identified by experts at Bangladesh National Herbarium, Mirpur, Dhaka.

Preparation of ethanolic extract

The collected plant parts were separated from undesirable materials or plants or plant parts and were washed with water. They were sun-dried for one week. The plant parts were grinded into coarse powder with the help of a suitable grinder (Capacitor start motor, Wuhu motor factory, China). The

powder was stored in an airtight container and kept in a cool, dark and dry place until analysis commenced. The leaves and stems were extracted by cold extraction method. 250gm grinded powder was soaked in 1000 ml of 98% of ethanol in a glass container for fifteen days accompanying regular shaking and stirring. The extract was separated from the plant debris by filtration by a piece of clean, white cotton material & it was done two times. The filtrate (Ethanol extract) obtained was evaporated under the air of ceiling fan and in a water bath until dried. After drying the filtrate, dried adhesive powdered mass was obtained. This powdered mass was designated as crude extract.

Reagents of Chemical Group Tests [4, 5]

Mayer's Reagent

1.36 g mercuric iodide in 60 mL of water was mixed with a solution containing 5 g of potassium iodide in 20 mL of water.

Dragendroff's Reagent

1.7 g basic bismuth nitrate and 20 g tartaric acid were dissolved in 80 mL water. This solution was mixed with a solution containing 16 gm potassium iodide and 40 mL water.

Fehling's Solution A

34.64 g copper sulphate was dissolved in a mixture of 0.50 mL of sulfuric acid and sufficient water to produce 500 mL.

Fehling's Solution B

176 g sodium potassium tartarate and 77 g of sodium hydroxide were dissolved in sufficient water to produce 500 mL. Equal volume of above solution were mixed at the time of use.

Benedict's reagent

1.73 g cupric sulphate, 1.73 g sodium citrate and 10 g anhydrous sodium carbonate were dissolved in water and the volume was made up to 100 mL with water.

Molish Reagent

2.5g of pure α -naphthol was dissolved in 25 mL of ethanol.

Liebermann-Burchard Reagent

5 mL acetic anhydride was carefully mixed under cooling with 5 mL concentrated sulfuric acid. This mixture was added cautiously to 50 mL absolute ethanol with cooling.

Test Method for phytochemicals [6, 7, 8, 9]

Testing of different chemical groups present in extract represents the preliminary phytochemical studies. In each test 5% (w/v) solution of extract in ethanol was taken unless otherwise mentioned in individual test. The chemical group test, which are performed as follows

Tests for reducing sugar

Benedict's test

0.5 mL of aqueous extract of the plant material was taken in a test tube. 5 mL of Benedict's solution was added to the test tube, boiled for 5 minutes and allowed to cool spontaneously. A red color precipitate of cuprous oxide indicates the presence of a reducing sugar.

Fehling's Test (Standard Test)

2 mL of an aqueous extract of the plant material was added to 1 mL of a mixture of equal volumes of Fehling's solutions A and B and boiled for few minutes. A red or brick red color precipitate formation indicates the presence of a reducing sugar.

Tests for combined reducing sugar

1ml of aqueous extract of plant material was boiled with 2 ml of dilute hydrochloric acid for 5 minutes, then cooled and neutralized with sodium hydroxide solution and then Fehling's test was performed as described above. A red or brick red color precipitate formation indicates the presence of a combined reducing sugar.

Tests for tannins

Ferric Chloride Test

5 mL solution of the extract was taken in a test tube. Then 1 mL of 5% Ferric chloride solution was added. Greenish black precipitate indicates the presence of tannins.

Potassium dichromate test

5 mL solution of the extract was taken in a test tube. Then 1 mL of 10% Potassium dichromate solution was added. A yellow precipitate indicates the presence of tannins.

Test for flavonoids

5 mL of dilute ammonia solution was added to a portion of the aqueous filtrate of plant extract followed by addition of concentrated H_2SO_4 . A yellow coloration observed in each extract indicates presence of flavonoids. The yellow coloration disappears on standing.

In another method, 0.2 g extract was dissolved in dilute sodium hydroxide and then neutralized with dilute hydrochloric acid. Formation of yellow color and disappearance of color indicate the presence of flavonoids.

Test for Saponins

1 mL solution of the extract was diluted with distilled water to 20 mL and shaken in a graduated cylinder for 15 minutes. No layer of foam indicates the absence of saponins.

Test for carbohydrates / gums

5 mL solution of the extract was taken and then Molish's reagent and sulphuric acid are added. Red violet ring produced at the junction of two liquids indicates the presence of gums and carbohydrate.

Test for Steroids

Liebermann-Burchard test for cholesterol

1ml solution of chloroform extract was taken and then added 2 mL Liebermann-Burchard reagent. No greenish color found.

Another method was performed while 1 mL solution of chloroform extract was taken and then added 1ml sulphuric acid. No red color found which indicates the absence of steroids.

Test for alkaloids

Mayer's test

2 mL solution of the extract and 5 ml of dilute hydrochloric acid (1%) were taken in a test tube. Then 1 mL of Mayer's reagent was added. A white or creamy white color precipitate indicates the presence of alkaloids.

Dragendroff's test

2 mL solution of the extract and 5 ml of dilute hydrochloric acid (1%) were taken in a test tube. Then 1 mL of Dragendroff's reagent was added. Orange brown precipitate indicates as the presence of alkaloids.

Test for Glycosides

A small amount of an alcoholic extract was taken in 1ml of water. A few drops of aqueous NaOH were added. A yellow color indicates the presence of glycosides.

Tests for proteins-xanthoprotein

To 1 mL of extract, few drops of nitric acid were added by the sides of the test tube and yellow color was not formed. This indicates the absence of xanthoprotein.

Tests for terpenoids (Salkowski test)

To 0.5 g of the extract, 2 mL of chloroform was added; Conc. H_2SO_4 (3 mL) is carefully added to form a layer. A reddish brown coloration at the interface indicates the presence of terpenoids.

Results and Discussion

The experimental findings from the study showed that the ethanol extract of the leaves & stems of *Cucurbita pepo* Linn. may have the following phytochemical compounds – reducing sugars, alkaloids, flavonoids, tannins, gums, glycosides and terpenoids.

The medicinal value of plant lies in the bioactive phytochemical constituents of the plant and which shows various physiological effects on human body. So through phytochemical screening one could detect the various

important compounds which could be used as the base of modern drugs for curing various diseases.

It is reported that the polyphenolic compounds, as like phenolic acids, flavonoids and tannins, commonly found in different plants and exert multiple biological response ^[10].

Conclusion

Among these constituents' phenolic compounds, flavonoids, tannins and alkaloids are the most valuable for therapeutic activity. So identification of the nature of the compounds is essential to evaluate the biological activity of the extract.

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