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## Phytochemical Constituents and Antioxidant Activity of Various Extracts of Corn Silk (Zea mays. L)

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#### ABSTRACT

Corn silk is a collection of the stigmas (fine, soft, yellowish threads) from the female flowers of the maize plant. Corn silk has been used traditionally as diuretic, antilithiasic, uricosuric and for curing cystitis, gout, kidney stones, nephritis and prostatitis. Phytochemical constituents, free radical scavenging activity and total antioxidant activity of various extracts of corn silk were carried out in the study. Phytochemicals were extracted from corn silk using various solvents such as benzene, chloroform, ethanol, ethyl acetate, methanol and petroleum ether. Screening of phytochemicals showed positive results for the presence of flavonoids, alkaloids, phenols, steroids, glycosides, carbohydrates, terpenoids and tannins. Phytochemicals were extracted best in methanol. Free radical scavenging activity was determined using DPPH assay as DPPH is a stable antioxidant. Methanolic extract was found to have maximum DPPH scavenging activity and total antioxidant activity. These activities may be due to the presence of flavonoids, alkaloids, phenols, steroids, glycosides and tannins in corn silk.

Key words: Phytochemicals, corn silk, solvents, antioxidant activity, free radical scavenging activity.

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#### INTRODUCTION

Plants have been used for centuries as remedy for human diseases because they contain components of therapeutic values. Maize (Zea mays.L) is the third most planted food crop and one of the major energy sources among the people of the semiarid tropics [1].Zea mays L., also known as maize, Indian corn or corn is a cereal that is one of the most important edible grains in the world. In addition to the grains, leaves, corn silks, stalk and inflorescence of the maize plant are used for the treatment of several ailments. Corn silks are scientifically referred to as Maydis stigma or Zea mays as they reflect the soft, fibre-like growth which accompanies the ear of the corn [13]. This yellowish thread-like strands or tassels called stigmas are found inside the husks of corn. They are relatively (4-8 inches) long with a mild sweetish taste. Corn silk has been used as diuretic, antilithiasic, uricosuric, and antiseptic. It is used for the treatment of edema as well as for cystitis, gout, kidney stones, nephritis, and prostatitis.

Phytochemicals are plant chemicals. Phytochemicals are defined as bioactive nonnutrient plant compounds in fruits, vegetables, grains, and other plant foods that have been linked to reducing the risk of major chronic diseases [12]. It is estimated that 5000 individual phytochemicals have been identified in fruits, vegetables, and grains. They are otherwise called as the secondary metabolites. The phytochemicals vary in distribution within the plant parts, as well as in their occurrence within plant species [2].

Free radicals are associated with various physiological and pathological events such as inflammation, aging, mutagenicity and carcinogenicity. Simply defined, the term free radicals refer to any chemical species (capable of independent existence) possessing one or more unpaired electrons, an unpaired electron being one that is alone in an orbital. Free radicals, especially the oxygen radical, superoxide, when formed could lead to the formation of other radicals [17].Oxidative stress, an excessive production of reactive oxygen species (ROS) outstripping antioxidant defense mechanism, has been implicated in the pathophysiological conditions that affect the cardiovascular system, and can cause severe damage to biological macromolecules and dysregulation of normal metabolism [19].Antioxidants are free-radical scavengers which provide protection to living organisms from damage caused by uncontrolled production of reactive oxygen species and subsequent lipid peroxidation, protein damage and DNA strand breaking. Therefore, there is a need for isolation and characterization of natural antioxidants having less or no side effects, for use in foods or medicines to replace synthetic antioxidant [15].

The present work deals with the preliminary phytochemical investigation of various extracts (benzene, chloroform, ethanol, ethyl acetate, methanol and petroleum ether) of corn silk, to identify the major group of phytochemicals which impart the medicinal property to the plant. Free radical scavenging activity using DPPH assay and total antioxidant of various extracts of corn silk were also analyzed.



#### MATERIAL AND METHODS

#### Plant material

Fresh sweet corns were collected from local market in Coimbatore. Corn silk was removed from them, shade dried and stored at room temperature for further analysis.

#### Preliminary phytochemical screening of corn silk

Fresh sample of corn silk was used to screen the presence of phytochemicals. For this, five grams of the corn silk was weighed, mashed and homogenized with 50ml of alcohol, acid (1% HCl) and water separately. These were boiled for one hour, cooled, filtered and used for the analysis of phytochemicals. The extract was analyzed for the presence of phytochemicals such as flavonoids, phenols, anthocyanins, tannins, saponins, steroids, alkaloids and terpenoids using standard procedure [10].

#### Soxhlet extraction of the plant sample

The shade dried corn silk was ground into coarse powder. Dried corn silk powder was successively extracted with different solvents such as petroleum ether, benzene, chloroform, ethyl acetate, methanol and ethanol with their increasing order of polarity by soxhlation for 6-12 hours. For the extraction, 20g of dried powdered sample was used with 200ml of the solvent. Then the extract obtained were collected separately and kept for further analysis. The qualitative phytochemical tests of various extracts of corn silk were carried out using standard procedure [10].

## Assay of free radical scavenging activity (DPPH activity)

The stable 1, 1-diphenyl-2-picryl hydrazyl radical (DPPH) was used for the determination of free radical scavenging activity of the various extracts. Different concentrations (10-100 $\mu$ g) of each of the extract of corn silk were added with an equal volume of methanolic DPPH solution (0.5mM) and incubated at 37<sup>o</sup>C for 30 min. DPPH solution with methanol was used as positive control and methanol acted as negative control. When DPPH reacts with antioxidant, DPPH was reduced and the colour changed from deep violet to light yellow. This was measured at 517 nm. [15].The percentage antioxidant activity was calculated by the following formula

Absorbance of the sample% scavenging activity =------- X 100Absorbance of the controlAbsorbance of the control

## Estimation of total antioxidant activity in corn silk

The phosphomolybdenum method was used to evaluate the total antioxidant activity of various extracts of corn silk. Antioxidants can reduce Mo (IV) to Mo (V) and the green

October – December 2011 RJPBCS Volume 2 Issue 4 Page No. 988



phosphate / Mo (V) compounds, which have an absorption peak at 695 nm, were generated subsequently [6], 0.1 ml of the sample was mixed with 1.0 ml of the reagent solution (0.6 M Sulphuric acid, 28 mM sodium phosphate and 4 mM ammonium molybdate). Reaction mixture was incubated at 95° C for 90 min in boiling water bath and cooled to room temperature. Absorbance of all mixtures the was measured at 695 nm against blank in UV spectrophotometer before and after the incubation. The blank solution contained 1.0 ml of reagent solution and the appropriate volume of the same solvent used for the sample and it was incubated under same conditions as rest of the sample. Total antioxidant activity was expressed as the number of equivalents of ascorbic acid in milligrams per gram of extract.

Total antioxidant activity = 100  $[1 - (A_o - A_t) / (A_o^o - A_t^o)]$ 

Where  $A_o$  is the OD of the sample at time  $t_o$  minutes and  $A_t$  is the time of the sample at time t = 90minutes.  $A_0^o$  and  $A_t^o$  represent the OD of the control at time t = 0 minutes and t = 90 minutes respectively.

#### **RESULTS AND DISCUSSION**

#### Phytochemical analysis

The preliminary phytochemical screening showed the presence of phytoconstituents such as alkaloids, amino acids, carbohydrates, phenolic compounds, terpenoids, steroids, proteins and tannins. Table 1 shows the results of phytochemical analysis of various extracts of corn silk.

| Inference |   |   |   |  |  |
|-----------|---|---|---|--|--|
| В         | С   | Е   | EA  | М  | PE   |
| -         | -   | +   | -   | +  | -  |
| +         | +   | +   | +   | +  | -  |
| -         | -   | +   | -   | +  | +  |
| +         | +   | +   | +   | +  | +  |
| +         | +   | +   | +   | +  | +  |
| +         | +   | +   | +   | +  | +  |
| -         | -   | -   | -   | -  | +  |
| +         | -   | +   | +   | +  | +  |
| +         | +   | +   | +   | +  | +  |
| +         | +   | +   | +   | +  | +  |
| +         | +   | +   | +   | +  | +  |
| -         | -   | -   | -   | +  | -  |
|           | B<br>-<br>+<br>+<br>+<br>+<br>+<br>+<br>+<br>+<br>+<br>+<br>+ | B C   - -   + +   - -   + +   + +   + +   + +   + +   + +   + +   + +   + +   + +   + +   + +   + + | B C E   - - +   + + +   - - +   + + +   + + +   + + +   + + +   + + +   + + +   + + +   + + +   + + +   + + +   + + +   + + + | B C E EA   - - + -   + + + +   - - + +   + + + +   + + + +   + + + +   + + + +   + + + +   + + + +   + + + +   + + + +   + + + +   + + + + | B C E EA M   - - + - +   + + + + +   - - + + +   - - + + +   + + + + +   + + + + +   + + + + +   + + + + +   - - - - -   + + + + +   + + + + +   + + + + +   + + + + +   + + + + +   + + + + +   + + + + + |

Table 1: Phytochemical Constituents of Various Extracts of Corn Silk

B Benzene; C Chloroform; E Ethanol; EA Ethyl acetate; M Methanol; PE Petroleum ether

October – December

2011 RJPBCS

Volume 2 Issue 4



The phytochemical analysis of the benzene, chloroform, ethanol, ethyl acetate, methanol and petroleum ether extracts of corn silk showed positive results for the presence of flavonoids, alkaloids, phenols, steroids, glycosides, carbohydrates, aminoacids, terpenoids and tannins. Methanolic extract of corn silk gave the maximum extraction of phytochemicals than any other extracts. Methanolic extract was followed by ethanolic extract for the presence of phytochemical constituents.

Zea mays husk has analgesic and anti-inflammatory effects that are due to the presence of tannins and polyphenolic constituents [3]. Among cereals, only maize has high amount of carotenoids, tocopherols and oil content compared with other major food crops such as rice and wheat [7].

Studies on Zea mays pollen reported that flavonol glycosides of quercetin, isohamnetin and kaempferol were present in Zea mays pollen. Among this the most prominent type of flavonols was diglycosides of quercetin and isohamnetin [5].

#### Free radical scavenging activity

The stable free radical scavenging activity by DPPH method is an easy, rapid and sensitive way to survey the antioxidant activity of a specific plant extract [8].

Figure 1 indicates the percentage of free radicals scavenging activity in various extractions with different concentrations (10-100µg) of corn silk.

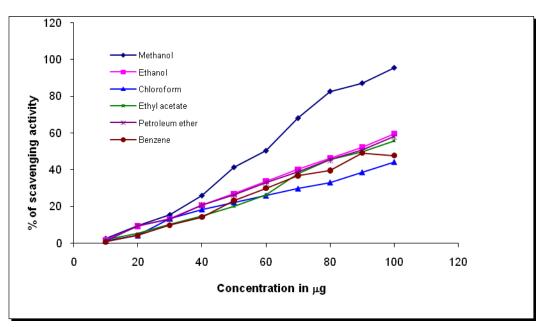


Figure 1: DPPH Radical Scavenging Activity Of Various Extracts of Corn Silk

In this study percentage inhibition of free radicals were carried out with different extractions of corn silk. Among the above extracts the methanolic extract of corn silk with 100



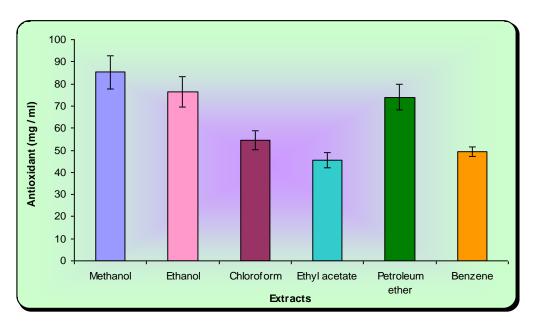
 $\mu$ g concentration gives higher percentage (95.6%) of free radicals scavenging activity than the other extracts. Low percentage (44.2%) of free radical scavenging activity was noted in chloroform extract of the corn silk.

Comparative studies on the crude ethanol, petroleum ether, acetic ether, N-butanol and water extraction of corn silk reported higher percentage of scavenging activity in petroleum ether and ethanol [12].

The free radical scavenging activity increases with increase in concentration. At a concentration of  $10\mu$ g/ml, methanolic extract obtained 2.5% of free radical scavenging activity and got increased to 95.6% in  $100\mu$ g/ml of concentration.

#### Total antioxidant activity of corn silk

Plants have good antioxidant ability and are safer than the synthetic antioxidants [16]. Secondary metabolite from medicinal plants function as small molecular weight antioxidant, but their particular mechanism of action are variable, and depends both on the structure and environment [14].Figure 2 shows the total antioxidant activity present in various organic extracts of corn silk.



#### Figure 2: Total Antioxidant Activity of Various Extracts of Corn Silk

Methanolic extract of corn silk exhibited the strongest antioxidant activity (85.2 mg/ml) among all the other extracts, while ethyl acetate extract yielded the lowest (45.5 mg/ml). The total antioxidant activity may be attributed to the presence of Phenolic and flavonoids constituents in all the fractions [11].



Studies on Egyptian corn silk showed that the upper parts of corn silk (dark brown part exposed to air) was found to have highest total antioxidant activity and DPPH scavenging activity then the lower parts (light yellow parts, not exposed to air) [9].

Higher activity was observed for total antioxidant in methanolic extract and it has also been reported that solvents used for extraction have dramatic effect on the chemical species [4].

The results indicate that corn silk is rich in phytochemical, which may be responsible for its medicinal property. The antioxidant activity of corn silk may be due the presence of flavonoids and tannins. Among the extracts methanolic extract showed the maximum activities for DPPH and total antioxidant activity. Further studies are under progress in our laboratory for the isolation of the active compounds.

## CONCLUSION

From the study, it may be concluded that corn silk is a rich source of phytochemical and has antioxidant property. Phytochemical were extracted best in methanol among the solvents and methanolic extract showed the maximum DPPH and total antioxidant activities.

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October – December 2011 RJPBCS Volume 2 Issue 4 Page No. 992



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