

Phytochemical Screening of Ethanolic Leaf Extracts of Citrus limon and Cassia auriculata

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ABSTRACT

Medicinal plants are widely recognized for their therapeutic value due to the presence of biologically active compounds known as phytochemicals. The present study aimed to investigate the qualitative phytochemical composition of ethanolic leaf extracts of Citrus limon and Cassia auriculata. Fresh leaves were collected, shade dried, powdered, and extracted using ethanol through Soxhlet extraction. Preliminary phytochemical screening was performed using standard qualitative methods to detect the presence of major secondary metabolites including alkaloids, flavonoids, steroids, tannins, carbohydrates, and proteins. The results revealed that both Citrus limon and Cassia auriculata contained alkaloids, flavonoids, steroids, and tannins. Carbohydrates were detected only in Cassia auriculata, while proteins were absent in both extracts. The findings indicate that Cassia auriculata possesses a comparatively richer phytochemical profile. These phytochemical constituents support the medicinal importance of both plant species and provide a basis for future pharmacological and phytochemical investigations.

Keywords: Phytochemical screening, Ethanolic extract, Citrus limon, Cassia auriculata, Secondary metabolites

1. INTRODUCTION

Medicinal plants have played a significant role in traditional and modern healthcare systems due to their therapeutic properties and natural origin. Plant-derived compounds continue to contribute substantially to pharmaceutical development because of their effectiveness and relatively lower side effects.

Phytochemicals are naturally occurring secondary metabolites synthesized by plants for protection and survival. These compounds exhibit numerous biological activities and are widely investigated for their medicinal potential. Important phytochemicals such as alkaloids, flavonoids, tannins, steroids, and carbohydrates have demonstrated antioxidant, anti-inflammatory, and antimicrobial activities.

Citrus limon (family: Rutaceae) is an economically and medicinally important species known for its phytochemical richness including flavonoids, limonoids, and essential oils. Similarly, Cassia auriculata (family: Fabaceae) is traditionally used in herbal medicine and contains various biologically active constituents.

Phytochemical screening is an important preliminary analytical technique used to identify the presence of bioactive compounds in plant extracts. Therefore, the

present study was undertaken to evaluate the phytochemical composition of ethanolic leaf extracts of Citrus limon and Cassia auriculata.

2. MATERIALS AND METHODS

2.1 Collection of Plant Materials

Fresh leaves of Citrus limon and Cassia auriculata were collected from local areas of Thoothukudi district, Tamil Nadu. The collected leaves were washed thoroughly to remove impurities.

2.2 Preparation of Ethanolic Extract

Leaves were shade dried for 7–10 days and powdered using a mechanical grinder. Approximately 20–30 g of powdered sample was extracted using ethanol by Soxhlet extraction for 6–8 hours. The extract was filtered and concentrated under reduced temperature and stored at 4°C.

2.3 Qualitative Phytochemical Screening

Standard procedures were employed for preliminary phytochemical analysis.

Alkaloids (Mayer's Test)

Extract was treated with Mayer's reagent. Yellow precipitate indicated positive results.

Flavonoids Test

Extract was treated with magnesium ribbon and concentrated hydrochloric acid. Development of pink or red colour confirmed flavonoids.

Protein Test (Biuret Test)

Extract was treated with sodium hydroxide and copper sulfate solution. Violet coloration indicated proteins.

Tannins Test

Extract filtrate was treated with ferric chloride solution. Formation of bluish-black coloration indicated tannins.

Steroids Test

Extract was treated with chloroform and sulfuric acid. Colour change indicated steroid presence.

Carbohydrates Test (Benedict's test)

Formation of orange-red precipitate confirmed carbohydrates.

Picture 1.1. Preliminary phytochemical screening of Ethanolic leaf extracts of *C. limon*



Picture 1.2. Preliminary phytochemical screening of Ethanolic leaf extracts of *C. auriculata*



3. RESULTS

3.1 Qualitative Phytochemical Screening of Ethanolic Leaf Extracts of *Citrus limon* and *Cassia auriculata*

The qualitative phytochemical analysis of ethanolic leaf extracts of *Citrus limon* and *Cassia auriculata* was performed to identify the presence or absence of major secondary metabolites. The phytochemical screening included carbohydrates, alkaloids, flavonoids, steroids,

tannins, and proteins using standard qualitative procedures.

The results obtained from the preliminary phytochemical investigation demonstrated that both plant extracts contained several important bioactive compounds. The occurrence of these phytochemicals confirms the medicinal significance of the selected plants and indicates their potential value for future phytochemical and pharmacological studies.

Table 1. Qualitative Phytochemical Constituents Present in Ethanolic Leaf Extracts

S.No	Phytochemical Constituents	Citrus limon	Cassia auriculata
1	Carbohydrates	–	+
2	Alkaloids	+	+
3	Flavonoids	+	+
4	Steroids	+	+
5	Tannins	+	+
6	Proteins	–	–

(+) Presence

(–) Absence

3.2 Phytochemical Profile of Citrus limon

The ethanolic leaf extract of Citrus limon exhibited the presence of four major phytochemical groups including alkaloids, flavonoids, steroids, and tannins. However, carbohydrates and proteins were not detected.

The positive result obtained for alkaloids indicates that Citrus limon contains nitrogen-containing secondary metabolites, which are considered important plant compounds. The presence of flavonoids suggests that the extract possesses valuable phenolic constituents. Steroids detected in the extract indicate the occurrence of structurally active plant compounds that contribute to physiological functions.

The detection of tannins confirms the existence of polyphenolic compounds in the leaf extract. Since carbohydrates were absent, the extract appears to contain comparatively lower amounts of primary metabolic constituents under ethanolic extraction conditions. Proteins were also absent, indicating that ethanol extraction mainly recovered secondary metabolites.

Overall, Citrus limon demonstrated a moderate phytochemical profile with four positively detected phytochemical classes.

3.3 Phytochemical Profile of Cassia auriculata

The ethanolic leaf extract of Cassia auriculata demonstrated a broader phytochemical composition compared with Citrus limon. The extract showed positive results for carbohydrates, alkaloids, flavonoids, steroids, and tannins, whereas proteins remained absent.

The presence of carbohydrates in Cassia auriculata indicates additional chemical diversity within the extract. Similar to Citrus limon, alkaloids and flavonoids were strongly detected, indicating the presence of medicinally valuable secondary metabolites.

Steroids were observed in the ethanolic extract, suggesting effective extraction of biologically active compounds. Tannins were also identified, indicating the occurrence of polyphenolic substances that contribute to the phytochemical richness of the plant.

The absence of proteins suggests that ethanol selectively extracted secondary metabolites while excluding larger protein molecules.

Overall, Cassia auriculata demonstrated a comparatively richer phytochemical composition with five positive phytochemical constituents.

3.4 Comparative Evaluation of Phytochemical Constituents

A comparison of the phytochemical profiles showed that both plant species shared several common constituents including alkaloids, flavonoids, steroids, and tannins. These common phytochemicals indicate similarities in their chemical composition and medicinal relevance.

However, a notable difference was observed in the detection of carbohydrates, which were identified only in Cassia auriculata. This additional constituent suggests that Cassia auriculata possesses greater phytochemical diversity than Citrus limon.

Among the six phytochemical groups screened, Cassia auriculata showed five positive responses, whereas Citrus limon showed four positive responses. Proteins were absent in both extracts.

The results of the present investigation indicate that ethanolic extraction is an effective method for recovering important phytochemical constituents from medicinal plant leaves. The observed phytochemical profile provides scientific evidence supporting the traditional importance of both plant species and highlights their potential for further phytochemical characterization.

4. DISCUSSION

The present study was carried out to evaluate the phytochemical constituents present in the ethanolic leaf

extracts of *Citrus limon* and *Cassia auriculata*. Phytochemical screening is considered an important preliminary step in medicinal plant research because it helps to identify biologically active compounds responsible for therapeutic properties.

In this study, ethanolic extracts of both plant leaves were subjected to qualitative phytochemical analysis for the detection of carbohydrates, alkaloids, flavonoids, steroids, tannins, and proteins. The results showed that both *Citrus limon* and *Cassia auriculata* contained several important secondary metabolites, indicating their medicinal significance.

The results revealed that alkaloids were present in both plant extracts. Alkaloids are naturally occurring nitrogen-containing compounds widely known for their pharmacological importance. These compounds play an important role in plant defense mechanisms and are often associated with biological activities. The presence of alkaloids in both plants indicates their potential medicinal value and supports their traditional use in herbal remedies.

Flavonoids were also detected in both *Citrus limon* and *Cassia auriculata*. Flavonoids are among the most important plant secondary metabolites because of their protective and therapeutic roles. They are commonly reported to possess antioxidant properties and contribute significantly to plant survival under environmental stress conditions. The presence of flavonoids suggests that both plants may serve as valuable natural sources of phytochemicals.

Steroids were identified in both ethanolic extracts. Plant steroids are recognized as important compounds involved in physiological regulation and biological activities. Their presence indicates that the extraction method was effective in recovering bioactive constituents from plant tissues. Steroids are commonly detected in medicinal plants and contribute to their pharmacological importance.

Tannins were also present in both extracts. Tannins are naturally occurring polyphenolic compounds that are widely distributed in medicinal plants. They are responsible for many biological activities and contribute to the medicinal quality of plant materials. Detection of tannins in both species further supports their traditional medicinal applications.

One of the important observations in this study was the presence of carbohydrates only in *Cassia auriculata* and

their absence in *Citrus limon*. This difference indicates variation in phytochemical composition between the two plant species. The presence of carbohydrates may reflect differences in metabolism, storage compounds, and extraction efficiency.

Proteins were not detected in either plant extract. This may be due to the extraction procedure and the selective ability of ethanol to dissolve secondary metabolites rather than larger protein molecules. The absence of proteins suggests that the major constituents extracted were phytochemical compounds rather than primary metabolites.

Overall, the phytochemical profile obtained in this study demonstrates that both *Citrus limon* and *Cassia auriculata* are rich in medicinally important secondary metabolites. However, *Cassia auriculata* showed a comparatively broader phytochemical composition due to the additional presence of carbohydrates. Therefore, among the two plants studied, *Cassia auriculata* exhibited a richer phytochemical profile.

The findings of this study support previous reports that medicinal plants are valuable sources of natural phytochemicals and emphasize the importance of phytochemical screening in identifying plant-derived bioactive compounds for future scientific and pharmaceutical studies.

5. CONCLUSION

The present study confirmed the occurrence of major phytochemical constituents in ethanolic leaf extracts of *Citrus limon* and *Cassia auriculata*. Both plants showed positive results for alkaloids, flavonoids, steroids, and tannins, while proteins were absent. Carbohydrates were detected only in *Cassia auriculata*.

These findings support the medicinal relevance of both plants and indicate their potential application in future phytochemical and pharmaceutical studies.

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