



## Original Research Article

### Phytochemical Screening of the Leaves of *Stevia rebaudiana*, Bertoni

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#### ABSTRACT

##### Keywords

Bioactive compounds, *Stevia rebaudiana*, alkaloids, tannins.

Nature has store house of remedies to cure all ailments of mankind. From the ancient ages the plants have been used for medicinal uses and other useful proposes to humans. The present investigation was carried out to estimate the phytochemical constituents present in *Stevia rebaudiana* Bertoni. The results showed the presence of bioactive constituents of alkaloids, tannins, flavonoids, glycosides, saponins, quinone and triterpenes. Catechins, coumarins and xanthoproteins were not detected. More research work is recommended on the plant leaves for isolation and characterization of bioactive compounds.

## Introduction

*Stevia rebaudiana* Bertoni is a versatile herb with incredible sweetness that is gaining very high popularity amongst all type of sweetener users as most ideal substitute for sugar. It produces sweet steviol glycosides. It is a high demanding antidiabetic medicinal plant belonging to Asteraceae family.

It is perennial and endemic, medicinal herb (Sivaram and Mugundan, 2003). It is also called as honey plant due to its sweetness. Thus the present study is a preliminary attempt to identify some of the phytochemicals of the selected plant. Hopefully this will lead to new information on this plant application and new perspective on the potential use of *Stevia*.

## Materials and Methods

### Collection of plant materials:

*Stevia rebaudiana* plant were obtained from Thiruvananthapuram, Kerala and was grown under protective conditions. The flora of presidency of Madras (Gamble,1935) and the Flora of Tamil Nadu Carnatic (Matthew,1983) were used for identification and authentication of the plants. The leaves are collected and washed thoroughly in running tap water and rinsed in distilled water and dried under shade. After drying, the leaves were powdered using mixer grinder and then kept in well closed container.

### **Preparation of phytochemical extracts**

This powder was extracted in the soxhlet using ethanol and ethyl acetate and subjected to qualitative phytochemical screening for the identification of various chemical constituents using the method described by Trease and Evans (1987) and by Harbone (1973). The plant extract were screened for the presence of secondary metabolites such as alkaloids, flavonoids, tannins, saponins, triterpenes, glycosides, catechin, coumarin, quinone and xanthoprotein.

**Test for tannins:** About 0.5g of the dried powder was boiled in 20 ml of water in a test tube and then filtered. A few drops of 0.1% ferric chloride was added and was observed for brownish green or a blue black coloration.

**Test for alkaloids:** One milliliter of aqueous extract was stirred and placed in 1% aqueous hydrochloric acid on a steam bath. Then, 1 ml of the filtrate was treated with Dragendorff's and Mayer's reagent. Turbidity or precipitation with this reagent was considered as evidence for the presence of alkaloids.

**Test for glycosides:** 0.5g extract of sample was dissolved in 1 ml water and then aqueous sodium hydroxide was added. Formation of yellow color indicated the presence of glycosides.

**Test for saponins:** 0.5 g extract were dissolved in 10ml of distilled water in a test tube was stoppered with a cork and shaken vigorously for about 30 seconds. The test tube was allowed to stand in a vertical position and observed over a 30 minutes period of time. If a honey comb froth above the surface of liquid persists after 30 minutes the sample is suspected to contain saponin.

**Test for triterpenes:** Ten milliliter aqueous extract was placed in a small beaker and evaporated to dryness. The residue was dissolved in 0.5 ml each of acetic anhydride and chloroform.

The solution was transferred into a dry test tube and concentrated sulphuric acid was added. Brownish red or violet rings at the zone of the contact with the supernatant and green or violet coloration denoted the presence of sterols and triterpenes

**Test for Flavonoid (Shindo's test):** To the test solution, a few magnesium turnings and a few drops of concentrated hydrochloric acid were added and boiled for five minutes. Appearance of red or orange red colour indicates the presence of flavonoids.

### **Test for Catechin**

To the test solution, a few drops of Ehrlich reagent and concentrated hydrochloric acid were added. Appearance of pink colour indicates the presence of catechin.

### **Test for Coumarin**

To 2 ml of the test solution, a few drops of alcoholic sodium hydroxide were added. Appearance of yellow colour indicates the presence of coumarin.

### **Test for Quinone**

The test solution was treated with a few drops of concentrated sulphuric acid or aqueous sodium hydroxide solution. Colour formation indicates the presence of quinone compound.

### **Test for Xanthoprotein**

To the test solution, a few drops of concentrated nitric acid and few ml of ammonia were added. Appearance of a red

precipitate indicates the presence of xanthoprotein.

## Result and Discussion

The preliminary phytochemical analysis of the leaf extract revealed the presence of alkaloids, tannins flavonoids, glycosides, saponins, quinone and triterpenes (Table 1 and 2). The most abundant compounds in the ethanol leaf extract were the glycosides. Alkaloids and tannins, were also seen in higher amounts but lesser than glycosides. Flavonoids and glycosides were seen in moderate levels, and triterpenes and saponins were seen in least amounts. The test for catechins, coumarins, quinones and xanthoproteins showed negative result.

In the ethyl acetate leaf extract the most abundant compounds were the glycosides and tannins. Alkaloids and flavonoids, were also seen in higher amounts but lesser than glycosides. Flavonoids and triterpenoids were seen in moderate levels, and quinine is seen in least amount. The test for catechins,

coumarins, saponins and xanthoproteins showed negative result. The presence of these secondary metabolites suggests that the plant might be of industrial and medicinal importance. Several reports say that the compounds possess remarkable antitumor, antidiabetic and antioxidant activity (Gupta and Sharma, 2006; Kaur and Kaopoor, 2002 and Ray and Hussan, 2002).

These results give a picture that the plant has quite a number of chemical constituents, which may be responsible for the many pharmacological actions. They were known to show medicinal activity as well as exhibiting physiological activity (Sofowara, 1993). The presence of these phytochemicals in the investigated medicinal plant would be responsible for the antimicrobial activity of the plant too. This finding supports the traditional knowledge in selecting the most active medicinal plants to use in traditional medicine practices in the future. Further work is needed to isolate active principle from the plant and to carry out pharmaceutical studies.

**Table.1** Phytochemical constituents of *Stevia rebaudiana* leaf with ethanol extract

Phytochemical constituents	Results*
Alkaloids	+++
Flavonoids	++
Tannins	+++
Glycosides	+++
Catechin	-
Coumarins	-
Saponins	+
Quinone	-
Triterpenes	++
Xanthoproteins	-

**Table.2** Phytochemical constituents of *Stevia rebaudiana* leaf with ethyl acetate extract

Phytochemical constituents	Results*
Alkaloids	++
Flavonoids	++
Tannins	+++
Glycosides	+++
Catechin	-
Coumarins	-
Saponins	-
Quinone	+
Triterpenes	++
Xanthoproteins	-

+++ Strongly present; ++ Present;  
+ Weakly present; - Absent

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