

PHYTOCHEMICAL INVESTIGATIONS AND PHARMACOLOGICAL SCREENING OF *XANTHOSOMA SAGITTIFOLIUM* (L.) LEAF EXTRACT

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Abstract

Xanthosoma Sagittifolium is a common herb consumed in Bangladesh for its nutritional and traditional medicinal values. The current study was carried out to evaluate the presence of phytochemicals and to screen out antioxidant and antibacterial potentialities of *Xanthosoma Sagittifolium* (L.) ethanolic leaf extract. Phytochemical screening of extract ensured the presence of carbohydrates, tannins, flavonoids, steroids and alkaloids. Antioxidant activity was evaluated using DPPH free radical scavenging assay where ascorbic acid used as standard. The leaf extract showed free radical scavenging capacity which is an indication that this extract may have antioxidant ingredient. Antimicrobial sensitivity of ethanolic extract was measured by disk diffusion method with Mueller-Hinton agar media using kanamycin as standard. The extract showed mild activity against four of the tested bacterial species such as *Bacillus subtilis*, *Staphylococcus aureus*, *Escherichia coli*, *Salmonella enterica*, *Vibrio cholera* and *Shigella dysenteriae*.

Keywords: *Xanthosoma sagittifolium*, phytochemical, antioxidant, antibacterial activity

Introduction

The plant kingdom is a rich source of potential drugs and awareness in recent years has been heightened towards the importance of medicinal plants. Drugs that are obtained from plants are easily attainable, cheap, safe and seldom have side effects [1]. *Xanthosoma sagittifolium*, commonly known as Cocoyam, is one of the common root and tuber crops consumed by over 400 million people around the world as a nutriment. It is considered as one of the six most important root and tuber crops [2]. It has many common names including callalo, yautia, tannia, malanga, cocoyam and new cocoyam which are used to refer to the domesticated species of *X. sagittifolium* [3]. In Bangladesh, the common name of cocoyam is Mukhi kochu and varieties of cocoyam is found in Bangladesh but among those some are edible and some are very much wild and they have their distinct acidity [4,5]. It is an herb of Araceae family which is native to tropical America and found in several countries of Asia and Africa. Various part of the plant are used in traditional medicine to prevent and treat various diseases. High level of calcium oxalate in leaves of *X. sagittifolium* are known to help in preventing osteoporosis [6, 7]. However, *X. sagittifolium* corm is rich in sodium, potassium and phosphorus which helps to maintain a high potassium -sodium ratio in blood. Thus, the plant parts are beneficial for the patient with high blood pressure. The leaves has reported expectorant and astringent properties. The decoction from the corm peel is used in folk medicine for its antidiarrhoeal activity. *In vitro* analgesic and anti-inflammatory activity of the herb were also determined in a study [8]. *X. sagittifolium* has prominent antifungal activity and a study has shown that *X. sagittifolium* may also have antitumor activity which can be used in anticancer therapy [9]. Studies have shown that easily digestible starch can be found in cocoyam and can also provide tangible amounts of protein, vitamin C, thiamine, riboflavine and niacin [10]. According to the suggestion of many researchers, consumption of parts *X. sagittifolium* may lower the risk of colon cancer due to its potential antioxidant effect. There is also recognition of *X. sagittifolium* inflorescent as a good source of micro and macro phytochemicals that have nutritional and therapeutic properties [11].

Anti-hyperglycemic activity of corm has also been reported in alloxan induced diabetic albino rats [12]. In our present study, the antibacterial activity of ethanolic leaves extract of *X. sagittifolium* was evaluated against some of the bacterial species as no available data on such study were found.

Materials and methods

Collection and identification of plant material

Plant samples, *Xanthosoma sagittifolium* was collected from Rajbari, Dhaka in November 2016. The plant were identified and confirmed by the Bangladesh National Herbarium, Mirpur, Dhaka with the DACB accession number of 43241.

Preparation of extract

The shade dried leaves was grinded into fine powder which had been made completely devoid of any water content. Then about 150gm of powdered leave material was macerated into sufficient ethanol for 10 days. After filtration through Whatman filter paper, the filtrate was concentrated at 40°C with a rotary evaporation. The 3.56gm concentrated extract was then preserved in refrigerator.

Phytochemical screening

Freshly prepared *X. sagittifolium* plant extracts were subjected to different qualitative tests like Molisch's test for carbohydrate; Fehling's test for reducing sugars; alkaloid test by using Mayer's, Dragendorff's and Wagner's reagents; frothing test for saponin; FeCl₃ test for tannin; alkali test for flavonoids; Salkowski's test for triterpenoids; Baljet test for finding glycosides.

Scavenging of 2, 2-Diphenyl-1- picrylhydrazyl (DPPH)

Antioxidant activity of *X. sagittifolium* was tested using DPPH (2,2-Diphenyl-1- picrylhydrazyl) free radical scavenging assay method. Different concentrations (1-512 µg /mL) of the extract were added to 3 mL of a 0.004% w/v solution of DPPH. The absorbance was then determined at 517 nm. Then % inhibitions were plotted against log concentration and IC₅₀ was calculated from the graph. The experiment was performed in triplicate and average absorption was noted for each concentration.

Tested bacterial strains and reagents

Three gram positive bacteria (*Bacillus subtilis*, *Staphylococcus aureus*) and four gram negative bacteria (*Escherichia coli*, *Salmonella enterica*, *Vibrio cholera*, *Shigella dysenteriae*) were used for

measuring the antimicrobial sensitivity. The organisms were verified by gram staining and sub culturing in appropriate selective media on before testing the plant extract.

Preparation of sterile disc:

In this method, measured amount of the test samples are dissolved in definite volumes of solvent to give solutions of known concentration ($\mu\text{g/ml}$). Then sterile blank discs (BBL, Cockville, USA) are impregnated with known amount of test substances using micropipette and dried. Standard antibiotic discs are used as positive control and the blank discs that adsorb only solvent are used as negative control.

Antimicrobial assay

For screening of antimicrobial activity by disk diffusion method, the discs were placed in petridishes (120 mm in diameter) containing a nutrient agar media sowed with the test microorganisms using sterile transfer loop. Kanamycin ($30\mu\text{g/disc}$) standard disc was used as the reference standard. The plates are then kept at 4°C for facilitating maximum diffusion. The test material diffuses from the discs to the surrounding medium. The plates are then kept in an incubator (37°C) for 12-18 hour to allow the growth of the microorganisms. The antibacterial activity of the test agent is recorded by measuring the diameter of the zone of inhibition in term of millimeter. [13]

Results

Phytochemical screening

The leaf extract of *X. sagittifolium* was subjected to various phytochemical tests. Table 1 showed different phytochemical test with results which were subjected to the extract.

Antioxidant Activity

Table 2 revealed the antioxidant activity of the extract by determining standard DPPH scavenging capacity. Inhibition of free radical by DPPH in percent (%) was calculated by the following equation:

$$\% = 1 - (A \text{ sample}/A \text{ blank}) \times 100\%$$

Where A blank is the absorbance of the control reaction (containing all reagents except the test material).

Antimicrobial assay

Some antibacterial activity of *X. sagittifolium* extract was identified against various bacterial strains which

have been showed in Table 3. Here zone of inhibition was calculated in mm.

Discussion:

According to the phytochemical tests result it can be suggested that *X. sagittifolium* leaf extract has carbohydrate, steroids, alkaloids and flavonoids. Test for tannins, proteins, saponins and glycoside gave negative result for the extract. The presence of various phytochemicals like flavonoids supported the result observed against DPPH scavenging activity of the extract. Standard an Plant extract were taken in each test tube by using following concentration:- $500\mu\text{g/ml}$, $250\mu\text{g/ml}$, $125\mu\text{g/ml}$, $62.5\mu\text{g/ml}$, $31.23\mu\text{g/ml}$, $15.63\mu\text{g/ml}$, $7.81\mu\text{g/ml}$, $3.91\mu\text{g/ml}$, $1.95\mu\text{g/ml}$ and $0.97\mu\text{g/ml}$. The comparison on the graph between the extract and standard ascorbic acid stated that *X. sagittifolium* extract could be as potential as standard. As standard antibacterial agent Kannamycin and $250\mu\text{g/ml}$ and $500\mu\text{g/ml}$ concentration for the extract were used. Zone of inhibition of the extract at concentration $250\mu\text{g/ml}$ against *Salmonella enterica* was very low compared to the standard while the $500\mu\text{g/ml}$ concentration had zone of inhibition 30mm and standard was above 40mm . Highest zone of inhibition of the extract was observed against *Escherichia coli* at $500\mu\text{g/ml}$ concentration which was around 47mm while standard had around 52mm . Very low zone of inhibition was observed for *Vibrio cholera* and *Shigella dysenteriae* at both concentrations.

Conclusion:

Based on the current study, it can be resolved that *X. sagittifolium* extract possesses antioxidant and antibacterial activity. Therefore, the use of different parts of the plants in folk medicine are justified. For the identification and isolation of the responsible bioactive compounds for the pharmacological activity of *X. sagittifolium* leaf extract further studies are required to be conducted.

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Table 1: Result of phytochemical screening of *X. sagittifolium* extract

Chemical group	Name of test	Leaf extract of <i>X. sagittifolium</i>
Carbohydrate	Molisch Test	+
	Fehling's Test	+
Proteins	Biurets's Test	-
Saponins	Frothing Test	-
Flavonoids	Test for Flavonoids	+
Steroids	Salkowski test	++
Alkaloids	Mayer's Test	+
	Dragendroff's Test	++
	Hager's Test	+
Glycoside	Baljet's test	-
	Liebermann's test	-
	Borntrager's test	-

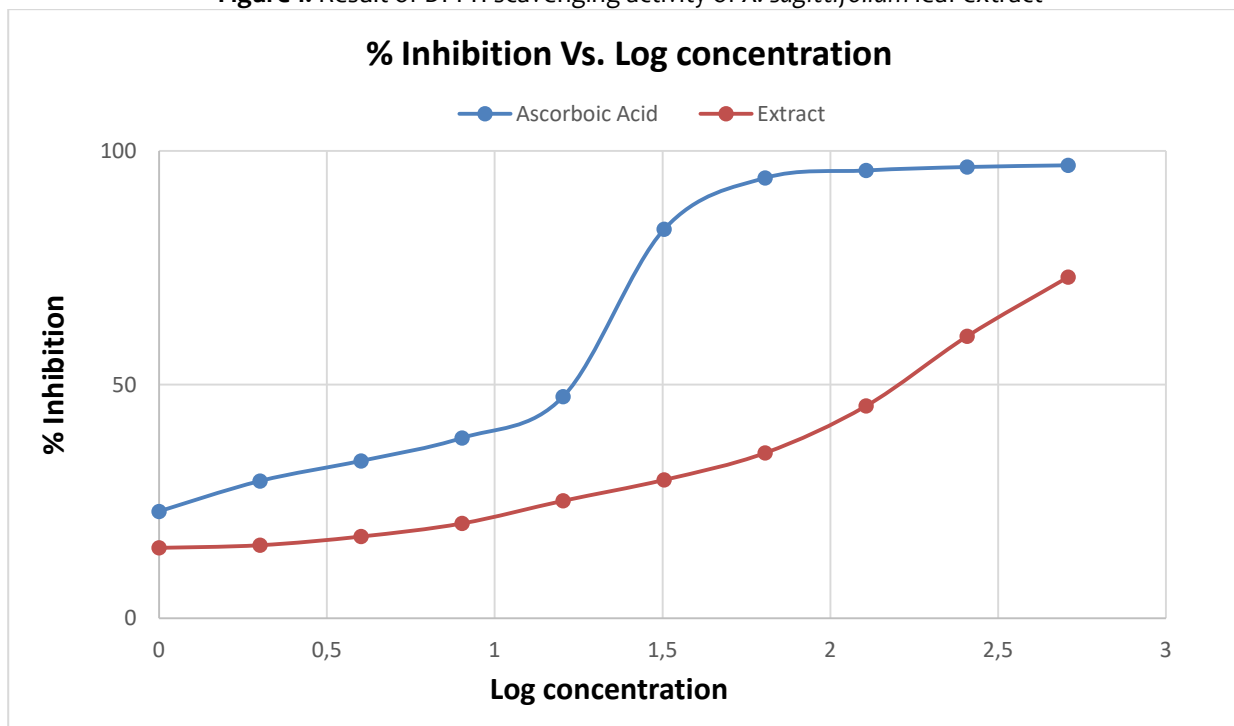
Figure 1: Result of DPPH scavenging activity of *X. sagittifolium* leaf extract

Figure 2: Result of antibacterial activity of *X. sagittifolium* leaf extract by disk diffusion method