Alpha-amylase Inhibitory Activity of Sitopaladi Churna: 
An Ayurvedic Formula

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Abstract

Sitopaladi Churna is an ayurvedic formula, traditionally used as anti-inflammatory, carminative, sedative, expectorant, anti-infective, and various respiratory disorders. The aim of the present study is to screening for alpha amylase (α-amylase) inhibition activity of aqueous extract of Sitopaladi Churna. In vitro α-amylase inhibition activity of Sitopaladi Churna was screened by the 3,5-dinitrosalicylic acid method. Sitopaladi Churna showed potent (α-amylase) inhibitory activity with an IC50 - 46.38 µg/ml. Thus, Sitopaladi Churna may consider as a remedy for diabetes and other insulin resistance-related diseases; however, animal and human studies are needed to confirm this activity.

Keywords: Sitopaladi Churna, Ayurvedic Formula, Diabetes, Alpha Amylase, Dinitrosalicylic Acid

1. INTRODUCTION

Amylase inhibitors are also known as starch blockers because they contain substances that prevent dietary starches from being absorbed by the body. Starches are complex carbohydrates that cannot be absorbed unless they are the first broken down by the digestive enzyme amylase and other secondary enzymes [1].

Salivary and pancreatic amylases catalyze the hydrolysis of glycosidic linkages in starch and other related polysaccharides, their inhibition have been theorized to have beneficial therapeutic effects by reducing carbohydrate-induced hyperglycemia and hyperinsulinemia [2]. Early studies of commercially available alpha-amylase (α-amylase) inhibitory preparations failed to decrease starch digestion in humans perhaps because of insufficient anti-amylase activity. More recent research utilizing purified amylase inhibitors have demonstrated that these nutrients can rapidly inactivate amylase in human intestinal lumen in a dose-dependent manner and prevent postprandial

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Received: 07 Jan 2015 Revised: 19 Mar 2015 Accepted: 05 Apr 2015


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rises in glucose and insulin [3]. Essentially, it allows the carbohydrates to pass through the system possibly with less caloric intake.

Sitopaladi Churna is an ayurvedic formulation, traditionally used as anti-inflammatory, carminative, sedative, expectorant, anti-infective, and various respiratory disorders. It also increases the appetite, helps digestion, and provides strength to the body. It is recommended for seasonal coughs and colds, as it is a very good expectorant [4]. The present study is to investigate α-amylase inhibitory activity of this ayurvedic formula by in vitro method.

2. METHODS

2.1 Preparation of Sitopaladi Churna

Sitopaladi Churna contains vamaslochana (inner white part of Bombusa arundinacia), Lavanga Tvak (bark of Cinnamomum zeylanicum), Ela (fruit of Elettaria cardamomum), Pippali (fruit of Piper longum), and Sitophila (Sugar candy). The ingredients were procured from Yuca Enterprises, Mumbai, Maharashtra, India and were made into fine powder, mixed and stored. Sitopaladi Churna was extracted with distilled water by soxhlet apparatus, and extract was dried in vacuum and used for in vitro α-amylase inhibitory activity.

2.2 In Vitro α-Amylase Inhibition Assay [3, 5-Dinitrosalicylic Acid (DNSA) Method]

The inhibition assay was performed according to Miller [5] using DNSA method. Varied concentration of dried aqueous extract of Sitopaladi Churna in 500 µl were added to 500 µl of 0.02 M sodium phosphate buffer (pH = 6.9 containing 6 mM sodium chloride) containing 0.04 units of α-amylase solution and were incubated at 37°C for 10 minutes, followed by addition of 500 µl of a 1% of starch solution in 0.02 M sodium phosphate buffer (pH = 6.9). The reaction was stopped with 1.0 ml of DNSA reagent. The test tubes were then incubated in a boiling bath water for 5 minutes and cooled in room temperature. The reaction mixture was then diluted after adding 10 ml of distilled water, and absorbance was measured at 540 nm. The control samples were also prepared accordingly without any plant extracts and were compared with the test samples containing various concentrations of the plant extracts prepared with different solvents. The results were expressed as % inhibition calculated using the formula:

\[
\text{% inhibition} = \frac{A_b (\text{control}) - A_b (\text{Test})}{A_b (\text{control})} \times 100
\]

2.3 Statistical Analysis

IC₅₀ values were calculated by origin Microcol 6.0 software (OriginLab Corporation).

3. RESULTS AND DISCUSSION

The results of α-amylase inhibition activity are shown in figure 1. In the present study, it was observed that all the concentrations (25-1000 µg) of aqueous extract of Sitopaladi Churna have shown potent α-amylase inhibition activity. The activity reveals concentration dependent nature of Sitopaladi Churna with α-amylase inhibition activity with IC₅₀ value of 46.38 µg/ml.

![Figure 1. Alpha-amylase inhibitory activity of different concentrations of Sitopaladi Churna](http://jtim.tums.ac.ir)

Drugs that inhibit carbohydrate hydrolyzing enzymes have been demonstrated to decrease postprandial hyperglycemia and improve impaired glucose metabolism without promoting insulin secretion in non-insulin dependent diabetes mellitus patients. Natural health products of vegetable origin...
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were clearly indicated as a promising avenue for the prevention of chronic diseases [6].

Postprandial hyperglycemia is one of the risk factors associated with type 2 diabetes mellitus. Digestion of dietary starch α-amylase plays a significant role in elevated blood glucose thus inhibition of amylase enzyme is very useful tool in the management of hyperglycemia [7].

Although the acute effects of α-amylase inhibitors may appear to have therapeutic benefit in patients suffering from diabetes mellitus, obesity, and other insulin resistance-related diseases. Chronic administration in animal models has been shown to induce adverse effects including deleterious histological changes to the pancreas. Because it is unclear if these dietary antinutrients can elicit similar deleterious changes in the pancreatic structure and function of humans [8] the presence of α-amylase inhibitors in human foodstuffs are generally undesirable.

*B. arundinacea* roots were used in bleeding gums, painful joints, skin eruptions, urinary infections, constipation, leprosy, and fever [9], [10]. *B. arundinacea* has been reported to possess anti-inflammatory, antiulcer [11], antioxidant [12], and antifertility activity [13]. *C. zeylanicum* bark has been traditionally used in uterine hemorrhage, stomachache, as an astringent and an antiseptic [14]. The scientific studies have confirmed its immunomodulatory activity. It has been also reported to possess remarkable pharmacological effects in the treatment of hyperglycemia [15], [16].

*E. cardamomum*, fruit is a sweet spice and employed as a medicinal flavoring agent it has been reported to possess antioxidant, anti-inflammatory, appetite stimulant, and carminative [17], [18], [19], [20]. *P. longum* fruit is highly valued in ayurveda for treating several disorders mostly related to indigestion, fever, asthma, and cough [21]. *P. longum* has been reported to possess potent antihyperglycemic and inhibit lipid peroxidation in an alloxan-induced diabetic rats [22]. Thus, Sitopaladi Churna may consider as a remedy for diabetes and other insulin resistance-related diseases; however, animal and human studies are needed to confirm this activity.

6. CONFLICT OF INTERESTS
Authors have no conflict of interests.

7. ACKNOWLEDGMENTS
The authors are thankful to K.V. Naveen Kiran, Chairman of Sri K.V. College of Pharmacy, for providing facilities to carry out this work.

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