https://doi.org/ 10.33472/AFJBS.6.10.2024.387-392

African Journal of Biological Sciences

Journal homepage: http://www.afjbs.com

Research Paper Open Access

In Vitro Evaluation of the Antidiabetic and Antimitotic Potential of *Dracaena trifasciata* Leaves Extract

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Article History
Volume 6,Issue 10, Feb 2024
Received:17 Feb 2024
Accepted : 01 Apr 2024

doi: 10.33472/AFJBS.6.10.2024.387-392

Background

The present study aimed to investigate the in vitro anti-diabetic and antimitotic activity of leaves extract of *Dracaena trifasciata*. As it was reported that *Dracaena trifasciata* contain flavonoids and other phytoconstituents, an attempt was made to determine its anti-diabetic and antimitotic potential.

Methods

The *in-vitro* α–amylase inhibition and antimitotic activity of the leaves extract of *Dracaena trifasciata* were evaluated by using 3,5-dinitrosalicylic acid (DNSA) and Allium cepa root meristematic cells.

Results

The result of α–amylase enzyme inhibition activity was found in a dose-dependent way, the strongest activity was shown by 25mg/ml hydroalcoholic extract 32.48±0.76%. The antimitotic activity was screened by using *Allium cepa* root meristematic cells. Methotrexate (0.1 mg/mL) was used as a standard. It is found that all concentration of extracts showed antimitotic activity, but 100mg/ml posses' maximum antimitotic potential. Extract shows 61.88% mitotic index at 100mg/ml concentration.

Keywords: *Dracaena trifasciata*, Flavonoids, polyphenols, Antidiabetic, Antimitotic

Introduction:

Diabetes mellitus presents as a severe and enduring collection of metabolic irregularities, marked by continual high levels of glucose in the blood. These anomalies stem from disruptions in carbohydrate, fat, and protein processing, which can result from both genetic predispositions and external factors. These disruptions may manifest as either insufficient insulin production or inadequate response to the insulin generated (Okur ME *et al*., 2017, Wadkar K *et al*., 2008). The antidiabetic activity of medicinal plants is due to the presence phenolic compounds Flavonoids, Terpenoids, Alkaloids, Glycosides, Steroid, Peptides, Lipids and other constituents (Demissew S *et al*., 2011, Grover J *et al*., 2002, Vuksan V *et al*., 2005). Flavonoids interfere with cyclin-dependent cell cycle regulation and interact with drug transport (Halliwel B. 2007). Ahmed *et al*. (2015) stated that flavonoids inhibit the proliferation of cell lines and demonstrated strong cytotoxicity towards colon cancer cells.

Dracaena trifasciata is a species of flowering plant in the family Asparagaceae, native to tropical West Africa and Asia. It is most commonly known as the snake plant (Takawira-Nyenya R. 2021). Thu ZM et al. (2020) reported presence of homoisoflavonoids, named trifasciatine A and (-)-(3*R*)-trifasciatine B from extract of *D. trifasciata.* The present study aimed to investigate the *in-vitro* anti-diabetic activity and anti-mitotic activity of leaves extract of *Dracaena trifasciata*.

Materials and method:

Dracaena trifasciata leaves was collected from Ashtang Ayurvedic College, Indore. The botanical identification and authentication of the plant materials were performed by Mrs. S. Mishra (botanist) and the voucher specimen was deposited in Herbarium of Biology Department, Govt. SN College, Knw (M.P.). The leaves were air dried at room temperature and milled with the aid of grinding machine.

Preparation of leaves extract:

Plant materials (100 g) were soaked in 450 mL of 50% hydroalcoholic solvent (Ethanol: water). This solvent system placed for 48 h at room temperature. After 48 h, solvents were filtered through Whatman filter paper no.1 separately in a beaker. The filtrate was dried in a rotary evaporator at 55 °C to obtain the concentrated yield of extracts (De Mesquita *et al*.2007).

Determination of total phenolic compounds and flavonoids content:

The Folin-Ciocalteau method was employed to ascertain the total phenolic compound contents. In this process, 0.5 ml of various dilutions of the extract samples were combined with 2.5 ml of 0.2 N Folin-Ciocalteau reagent (obtained from Sigma–Aldrich) for a duration of 5 minutes. Subsequently, 2.0 ml of 75 g/l sodium carbonate solution was added. After an incubation period of 2 hours at room temperature, the absorbance of the reaction was measured at 760 nm. The outcomes were quantified in terms of gallic acid equivalents. Additionally, total flavonoids were assessed utilizing the method developed by Ordonez et al., 2006. To 0.5 ml of the sample, 0.5 ml of 2% AlCl3 ethanol solution was introduced.

Following a 1-hour incubation at room temperature, the absorbance was recorded at 420 nm. Total flavonoid contents were determined by referencing a calibration curve constructed with quercetin

α -Amylase inhibition activity

The α-amylase inhibition assay was conducted following the 3,5-dinitrosalicylic acid (DNSA) method as outlined by Wickramaratne *et al*. (2016). The hydroalcoholic extract, ranging in concentrations from 5, 10, 15, 20 and 25mg/mL, derived from Dracaena trifasciata leaves, was dissolved in a buffer solution (0.02 M Na2HPO4/NaH2PO4, 0.006 M NaCl) adjusted to pH 6.9. To initiate the assay, 200 µL of α-amylase solution (2 units/mL) was combined with 200 µL of the extract and incubated for 10 minutes at 30°C. Subsequently, 200 µL of starch solution (1% in water, w/v) was added to each tube and incubated for 3 minutes. The reaction was halted by adding 200 µL of DNSA reagent (comprising 12 g of sodium potassium tartrate tetrahydrate, 8.0 mL of 2 M NaOH, and 20 mL of 96 mM 3,5-dinitrosalicylic acid solution) followed by boiling for 10 minutes in a water bath set at 85°C. After cooling to room temperature, the mixture was diluted with 5 mL of distilled water, and the absorbance was measured at 540 nm using a UV-Visible spectrophotometer. A blank with 100% enzyme activity was prepared by substituting the plant extract with 200 µL of the buffer. Additionally, a control reaction was set up using acarbose, following the same procedure as with the plant extract. The inhibition of α-amylase was expressed as percentage of inhibition and was calculated by the following equation:

Inhibition (%) =[(Ac− Acb) − (As − Asb)/(Ac − Acb)] × 100,

Where,

Ac-=absorbance of control; Acb=absorbance of control blank; As=absorbance of sample;

Asb=absorbance of sample blank. The % α-amylase inhibition was plotted against the extract concentration and the IC50 values were obtained from the graph.

Antimitotic assay

A modified technique outlined by Fiskesjo (1985) was employed to assess the antimitotic activity using Allium cepa roots. Allium cepa bulbs weighing approximately 40 ± 10 grams were allowed to germinate in water for 72 hours at room temperature under dark conditions. Bulbs exhibiting roots grown to approximately 3 cm were chosen for subsequent experimentation. These onion roots were placed in beakers filled with water, methotrexate (0.1 mg/mL), and various concentrations of plant extracts 10, 20, 30, 40, 50 and 100mg/ml for duration of 24 hours. Water was utilized for dilution purposes, serving as the control, while methotrexate was employed as the standard for the study. Following the 24-hour exposure period, the number of dividing and non-dividing cells was recorded, and the mitotic index was calculated using the following formula:

$$\text{Mitotic Index} = \frac{\text{No. of dividing cell}}{\text{Total no. of cells}} \text{ X } 100$$

Results and discussion

Total phenol and flavonoids contents:

The concentration of total phenol compounds in the leaves was determined using the Folin-Ciocalteu method and expressed in terms of gallic acid equivalents, utilizing a standard curve equation (y = 0.0064x) with a correlation coefficient (r^2) of 0.989. The total phenolic content was measured to be 138.3 ± 5.5 mg gallic acid equivalent per gram of extract powder. Similarly, the total flavonoid content in the leaves was quantified as 29.1 ± 0.8 mg quercetin equivalent per gram of extract powder, employing a standard curve equation (y = 0.0066x + 0.0131) with an r^2 value of 0.996.

In-vitro α-Amylase Inhibition Activity

In this study, leaves extract were evaluated for their possible α-amylase inhibitory activities alongside acarbose as a positive control. The α-amylase inhibitory activities and IC50 values of the acarbose and leaves extract are summarized in Table 1. Results showed the inhibitory activities of extracts and standard, IC50 values were found to be 50.42 and 7.5 for leaves extract and acarbose respectively. Concentration dependent inhibition was observed for various concentrations of the tested extracts and the standard. Among the extracts, the hydroalcoholic extract of 25mg/mL concentration showed the highest alpha–amylase enzyme inhibition activity of 32.48±0.76%**.** The standard positive control Acarbose showed an IC50 value of 96.22±0.78% inhibition at 25mg/mL)

Table 1: α-Amylase inhibitory activities of the extracts and standard

Concentration	Percentage inhibition	
	Extract	Acarbose
5mg/ml	18.52±0.84	46.82±0.68
10mg/ml	22.96±0.78	54.16±0.64
15mg/ml	26.36±0.82	66.32±0.58
20mg/ml	29.12±0.88	75.45±0.66
25mg/ml	32.48±0.76	96.22±0.78

Values are expressed as Mean± SD for triplicates

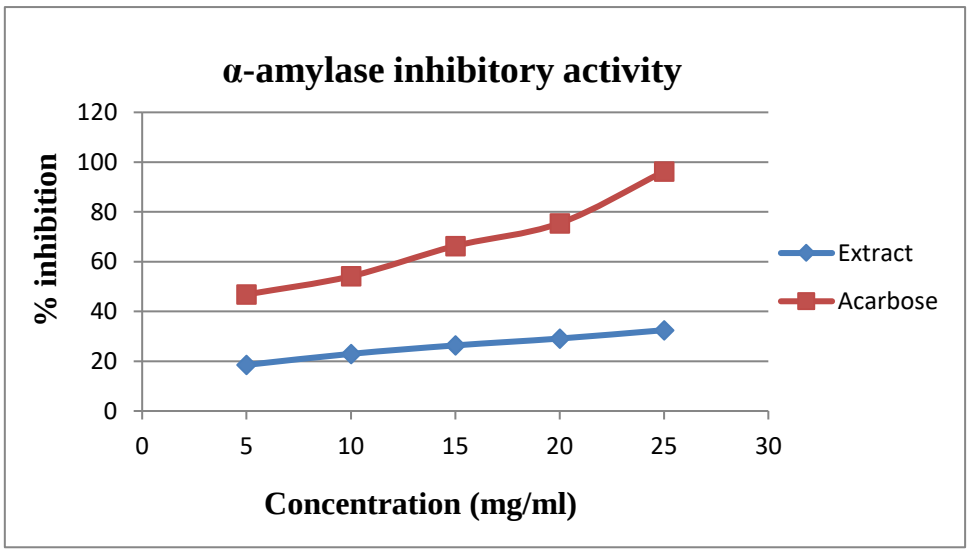

Figure 1: α-Amylase inhibitory activities of the extracts and standard

Antimitotic assay:

The results of antimitotic assay effect of *Dracaena trifasciata* hydroalcoholic extract on mitotic index (MI) of Allium cepa root tip cells are given in Table and Figure 2. Thus, all plants extracts displayed significant antimitotic activity which indicates its use as a potent antimitotic agent.

Table 2 Effect of *D. trifasciata* extract on mean root length mitotic index (MI) of Allium cepa root tip cells

Treatments		**Mean Root length (in mm)**	**Mitotic Index (MI) %**
Water		34.12±1.26	90.85
Methotrexate 0.1 mg/ml		16.44±1.28	14.52
Dracaena trifasciata Extract	**10mg/ml**	29.22±1.32	88.22
	20mg/ml	28.56±1.34	84.56
	30mg/ml	26.08±1.3\	79.64
	40mg/ml	22.32±1.48	75.45
	50mg/ml	20.18±1.44	72.34
	100mg/ml	19.86±1.32	61.88

Values are expressed as Mean± SD for triplicates

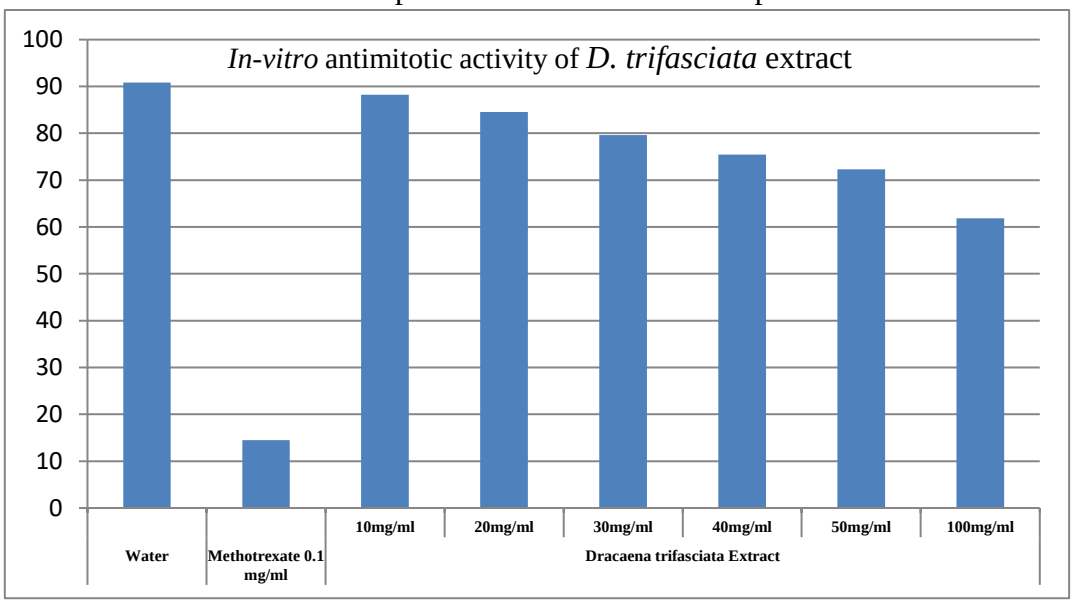

Figure 2: Effect of *D. trifasciata* extract on mitotic index (MI) of Allium cepa root tip cells

Conclucion

According to the result of the study on the leaf extracts of *D. trifasciata* exhibit α-amylase inhibitory activity with remarkable activity in the hydroalcoholic extract. Results for antimitotic activity for extract were comparable to the activity of methotrexate. Maximum numbers of non-dividing cells were observed. As a result of this cells arrest in mitosis and eventually die by apoptosis. Furthermore, this study has opened opportunities for future

research in searching for novel effective drugs for diabetics that possess anti-diabetic activity and for antimitotic or anticancer agent.

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