

## Research Article

## Open Access

# In Vitro Antiglycation Activity of Some Medicinal Plants Used in Diabetes Mellitus

Perera PRD<sup>1\*</sup>, Sagarika Ekanayaka S<sup>2</sup> and Ranaweera KKDS<sup>1</sup>

<sup>1</sup>Department of Food Science and Technology, Faculty of Applied Sciences, University of Sri Jayewardenepura, Sri Lanka

<sup>2</sup>Department of Biochemistry, Faculty of Medical Sciences, University of Sri Jayewardenepura, Sri Lanka

### Abstract

Non enzymatic glycation is the major cause of spontaneous damage to proteins leading to various complications due to formation of non-reversible Advanced Glycation End Products (AGEs) and oxidative stress. Medicinal plants having both antiglycation and antioxidant activity may be good therapeutic potential in the treatment of diabetic mellitus. The present study was undertaken to investigate the antiglycation effects of *Cassia auriculata* – flowers and *Pillanthus embilica* – Fruit which are being used in the treatment of diabetes mellitus in Ayurvedha medicine. The water extracts of the commercial sample and the other three samples of *C. auriculata* flower had significant anti-glycation activity as 109.8 µg/mL, 218.3 µg/mL, 250 µg/mL and 202.3 µg/mL and *Pillanthus embilica* – Fruit had 37.4 µg/mL, 82.2 µg/mL, 74.7 µg/mL and 74.1 µg/mL respectively. Arbutin was used as the positive control and showed 65µg/mL.

**Keywords:** Diabetes mellitus; *Cassia auriculata*; Bovian serum albumin; Antiglycation activit; *Phyllanthus emblica*

### Background/Justification

Diabetes mellitus which was once considered a disease of the developed world has now become a worldwide diseases affecting both developed and developing countries. Non enzymatic glycation is the major cause of spontaneous damage to proteins leading to various complications due to formation of non-reversible Advanced Glycation End Products (AGEs) and oxidative stress. More than 400 traditional plants for diabetes mellitus have been recorded but very few have received scientific and medical evaluation for their efficacy. Medicinal plants having both antiglycation and antioxidant activity may be good therapeutic potential in the treatment of diabetic mellitus.

### Objective

Determine the antiglycation activity of *Cassia auriculata* – flowers and *Pillanthus embilica* – Fruit which are being used in the treatment of diabetes mellitus in Ayurvedha medicine.

### Materials and Methods

#### Collection of samples

Plant materials were collected from three different areas in Sri Lanka where given species is grown. *Cassia auriculata* flowers were collected from Anuradhapura, Katharagama and Mathugama and *Phyllanthus embilica* fruit from Anuradhapura, Bibile and Balangoda areas in Sri Lanka. Commercially available dried sample of each plant material was collected from traditional herbal market in Colombo, Sri Lanka and identification of samples was carried out by a Botanist at Bandaranayke Memorial Ayurvedha Research Institute at Nawinna.

#### Preparation of plant materials

Fresh plant parts collected from different areas were air dried for 24 hours in room temperature and dried in a dehydrator at (Leader) at 50°C for 24 hrs and powdered using a domestic grinder to obtain fine particles and stored in air tight containers in a refrigerator.

#### Preparation of water extract

Water extracts of dried and milled powdered samples were prepared

according to the traditional method practiced in Ayurveda to prepare 'Kasaya'. As 60 g of the powdered samples were simmerly boiled in 960 ml to obtain the decoction of 240 ml. This water extract was filtered through a fine silk cloth. The filtrate was freeze dried and the powdered samples were kept at -4°C in a cold room in air tight containers.

### Determination of Antiglycation activity

Antiglycation activity was determined using the Bovian Serum Albumin assay [1] with slight modification. In all experiments the final reaction volume was 1.0 ml and carried out in 1.5 ml Eppendrof tubes. Bovian serum albumin 500 µl (1 mg/ml concentration) was incubated with glucose 400 µl (500 mM final concentration) and 100 µl sample, 100 µl phosphate buffer saline instead of the sample, as the sample control and 100 µl Arbutin as the reference standard. Negative control was carried out at the same time with Bovian serum albumin 500 µl (1mg/ml concentration), 400 µl phosphate buffer saline and 100 µl sample incubating under same conditions. The reaction was allowed to proceed at 60°C for 24 hours thereafter, stopped the reaction by adding 10 of 100% (W/V) trichloroacetic acid (TCA). The TCA added mixture was kept at 4°C for 10 minutes and centrifuged 4 minutes at 13000 rpm. The precipitate was redissolved with alkaline PBS (pH 10) and quantified for the relative amount of glycated BSA based on fluoresce intensity by Fluorescent Microplate Reader (Spectra Max Gemini EM). The excitation and emission wavelength used were at 370 nm and 440 nm respectively. Each sample was carried out in five concentrations and in triplicate. Percentage of inhibition was calculated using the formula given below and the sample concentration required for the 50% of inhibition was calculated using minitab 14.

**\*Corresponding author:** Perera PRD, Department of Food Science and Technology, Faculty of Applied Sciences, University of Sri Jayewardenepura, Sri Lanka, Tel: 011 94 714439319; E-mail: [rupika.perera@yahoo.com](mailto:rupika.perera@yahoo.com)

**Received** October 16, 2013; **Accepted** November 27, 2013; **Published** December 02, 2013

**Citation:** Perera PRD, Sagarika Ekanayaka S, Ranaweera KKDS (2013) *In Vitro* Antiglycation Activity of Some Medicinal Plants Used in Diabetes Mellitus. Med Aromat Plants 2: 143. doi: [10.4172/2167-0412.1000143](https://doi.org/10.4172/2167-0412.1000143)

**Copyright:** © 2013 Perera PRD, et al. This is an open-access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

$\% \text{ of inhibition} = \frac{\text{OD blank} - (\text{OD sample} - \text{OD sample negative})}{\text{OD blank}} \times 100$

## Results

Antiglycation activity is inversely proportional to the sample concentration. Commercial sample showed the highest activity comparing to the other samples and it showed higher activity than the positive control Arbutin (Figure 1).

Commercial sample of *Cassia auriculata* flowers showed high antiglycation activity than the samples freshly collected and dried under laboratory conditions. Arbutin is the positive control (Figure 2).

## Discussion

The presence of Gallic acid, Catechol, Ellagic acid, Phloroglucinol, Indol acetic acid, Vitamin C,  $\beta$  carotene, superoxide dismutase enzyme

have been reported in the *P. emblica* fruit by Kalra [2], Singh et al. [3]. The phytochemical screening of aqueous and water-soluble fraction of ethanol extract of *C. auriculata* flowers revealed the presence of flavonoids, phenolic acids, sterols, triterpenoids, alkaloids, tannins and anthocyan by Hakkim et al. [4] in their study. High antioxidant activity of the water extracts of the *P. emblica* fruit and *C. auriculata* flowers has been reported by the authors in a previous study should be due to the availability of these known antioxidant compounds. As oxidation plays an important role in the formation of Advanced Glycation End Products by converting Amadori products to reactive Intermediate carbonyl compounds, the antioxidant potential of the water extract of Emblica fruit would be involved in retarding the formation of AGEs in diabetes mellitus.

Hansen et al. [5] reported in their study as the *P. emblica* fruit containing high moisture content and sugar, during the drying process fermentation occurs and increases the free phenolic acids due to hydrolyzing of the bound phenolic compounds. This process may occur during the shade drying process than oven drying due to temperature fluctuation and slow drying.

The water extracts of commercial sample of *Phyllanthus emblica* and *Cassia auriculata* shade dried under natural environment showed high antiglycation activity than the samples dried under laboratory conditions. Effect on drying methods on total phenolic contents were studied, and they found air dried samples contains higher Total phenolic content than oven dried samples [6]. Thermal degradation of compounds may occur during hot air drying. Effect of the drying method could be affected for the variation of the antiglycation potential of the above samples. Maturity levels of the collected samples and the geographical variation also may contribute to the variation of glycation activity among the samples.

## Conclusion

The commercial samples of *Cassia auriculata* flowers and *Phyllanthus emblica* fruits available in the traditional herbal market showed high antiglycation activity than the samples collected from other areas and dried under laboratory conditions. Among the two herbal plants *Phyllanthus emblica* fruits showed high activity.

## Outcome

In the previous studies of the authors, it is reported that all samples of *C. auriculata* flower and *Pillanthus embilica* – Fruit contained high phenolic contents and high antioxidant activities. The flower of *Cassia auriculata* and the fruit of *Pillanthus embilica* have a good therapeutic potential for diabetes mellitus.

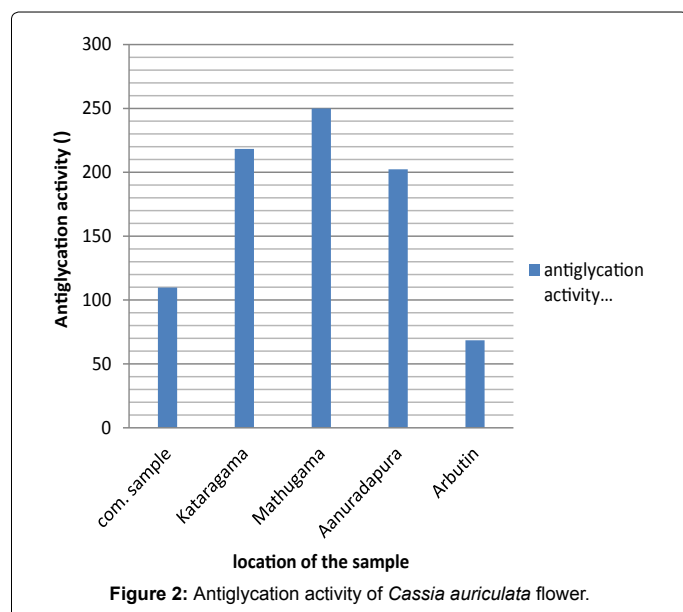
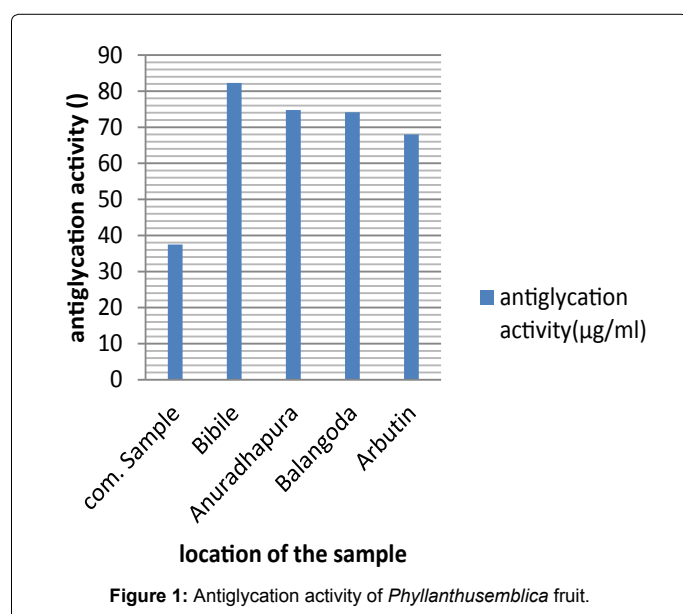
## Acknowledgement

Acknowledge the financial support given by the University Grant (Grant o.ASP/08/RE/2008/09) of University of Sri Jayewardenepura.

We are also thankful to Dr. Sirimal Premakumara, Director, Industrial Technology Institute, Sri Lanka and Mr. P. Ranasinghe of Herbal Technology Division, Industrial Technology Institute for providing facilities during the research work.

## References

1. Matsuura N, Aradate T, Sasaki C, Kojima H, Ohara M, et al. (2002). Screening system for the Maillard reaction inhibitor from natural product extracts. *J Health Sci* 48: 520-526.
2. Kalra CL (1989) The chemistry and technology of amla (*Phyllanthus emblica*)-A resume. *Journal of Indian Food Packer* 67-68.
3. Singh BP, Singh IP, Kumar KA (1987) Physiochemical composition of different cultivars of aonla. *Journal of Indian Food packer*.



4. Hakkim FL, Girija S, Kumar SR, Jalaludeen MD (2007) Effect of aqueous and ethanol extracts of *Cassia auriculata* L. flowers on diabetes using alloxan induced diabetic rats. Int J Diabetes Metabolism 15: 100-106.
5. Hansen JES, Larsen VA, Bog-Hansen TC (1984) The microheterogeneity of  $\alpha_1$ -acid glycoprotein in inflammatory lung disease, cancer of the lung and normal health. Clinica Chimica Acta 138: 41-47.
6. Hajimedipoor H, Adib N, Khanavi M, Mobi M, Amin GR, et al. (2012) Comparative study on the effect of different methods of drying on phenolics content and antioxidant activity of some edible plants. IJPSR 3: 3712-3716.