



## Anti-diabetic Activity and Microbial Quality of Ready-to-Serve *Momordica charantia* (MC) Drink

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**Abstract** - Diabetes mellitus has a high prevalence in Malaysia and is expected to rise in the future due to lifestyle changes. Thus, consumers are turning to alternative methods in the prevention and cure of the disease. The *Momordica charantia* has been studied for its anti-diabetic activity both in vitro and in vivo and is shown to be effective in inhibiting glucose absorption. Therefore, the MC is used as a main ingredient in the development of health beverages to offer alternatives for patients or the health conscious. The microbial quality of the product is examined to ensure the safety of the product and to find methods to enhance its shelf life.

**Keywords:**  $\alpha$ -glucosidase inhibitory activity, anti-diabetic activity, microbial quality, *Momordica charantia* (MC), ready-to-serve drink, shelf life

### Introduction

The prevalence of diabetes mellitus is estimated to increase in many countries around the world due to changing lifestyles. In 2000, the prevalence was estimated to be 2.8% for all age groups worldwide and was expected to increase to 4.4% in 2030, which brings the total number of 171 million in 2000 to 366 million in 2030 (Wild, et al., 2004; Han et al., 2009). A more recent study by Shaw, Sicree and Zimmet (2010) had estimated the numbers to be higher from 6.4% which affects 285 million people in 2010 to 7.7% and 439 million people by 2030. Also in this study, Malaysia was listed as one of the top 10 countries for diabetes prevalence which was 11.6% in 2010 and is expected to reach 13.8% by 2030. According to a local study, it was expected that the prevalence of Type 2 diabetes in Malaysia would exceed the world's prevalence rate due to changes in demographic patterns, increased job opportunities and improved economy and changes in dietary habit (Zaini, 2000). According to the Clinical Practice Guidelines by the Ministry of Health Malaysia (2009), 48% of patients above 30 years old are unaware they have diabetes. Management of the disease should start as early as detected to enable a person to maintain a healthy lifestyle. The diet plays an important role in the treatment of diabetes while medication is only given to certain patients.

Many patients are now turning to alternative treatment besides medication and functional food is one of the things that people these days are seeking for. It is also a better approach to the prevention of diabetes as it is easily accessible and affordable. Studies also show that up to 30% of patients with diabetes mellitus was estimated to use alternative medicine (Ryan, Pick & Marceau, 2001). Habicht (2011) mentioned that the bitter melon is being used in traditional medicine to treat diabetes due to its ability to lower blood glucose levels. *Momordica charantia* (MC), which is also known as the bitter melon or bitter melon is well known for the treatment of diabetes-related conditions. It grows in tropical and subtropical areas of Asia, Africa and the Caribbean. Various studies had been carried out

in-vitro and in-vivo of the anti-diabetic effects of the MC and health products are developed using the MC as the main ingredient.

### **In-Vitro Studies**

In-vitro studies of anti-diabetic activity are carried out using the alpha ( $\alpha$ ) glucosidase inhibitory activity screening.  $\alpha$  -glucosidase inhibitors in the small intestine suppresses postprandial hyperglycemia by inhibiting the digestion of carbohydrates (Matsuura et al., 2002). The  $\alpha$  -glucosidase inhibitory activity assay performed by Mun'im et al. (2013) on the ethanolic extracts of MC showed that the fruits extracts showed no inhibitory activity, whereby the  $IC_{50}$  (ppm) is 1861.99. Contrary to this study, polypeptide-k and oil isolated from the MC seed demonstrated blood glucose level-reducing activity where its inhibition% is highest at 53.55% at concentration 2 mg/mL (Ahmad et al., 2012). Methanolic extracts of the seeds also exhibited  $\alpha$  -glucosidase inhibitory activity (Matsuura et al., 2002).

In cell culture studies, it was found that MC juice had a form of protection towards apoptosis in rat insulinoma (RIN) cells and may help in recovering or regenerating destroyed pancreatic  $\beta$ -cells (Krawinkel & Keding, 2006; Sitasawad, Shewade & Bhonde, 2000).

### **In-Vivo Studies**

The in-vivo studies of anti-diabetic activity of the MC showed more prominent prove of its anti-diabetic activity as demonstrated by Viridi et al. (2003) Diabetic induced rats divided into three categories were fed with MC extracts of water, methanol and chloroform at a dosage of 20 mg/kg body weight. There was a significant reduction in blood glucose for the methanolic extract and constant activity for the water extract after a duration of four weeks. However, there was no antihyperglycemic activity for the chloroform extract.

In the investigation of suppressing gluconeogenesis, the ethanolic extract of MC inhibited hepatic gluconeogenesis enzymes fructose-1, 6-biphosphatase and glucose-6-phosphate (Chowdhury et al., 2012). The seeds extracts orally fed to streptozotocin (STZ) induced diabetic rats showed a decrease in plasma glucose, thio-barbituric acid-reactive substances, lipid-hydroperoxides, alpha-tocopherol, at the same time increased level of ascorbic acid, reduced glutathione and insulin (Sathishsekar and Subramanian, 2005). 1, 10 and 50% juice concentration of MC orally fed to mice did not show any activity in normal mice but STZ-induced diabetic mice showed significant lowering of hyperglycemia when fed with MC juice of 50% for 5 days compared to unfed diabetic mice (Sitasawad, Shewade & Bhonde, 2000).

When performed using butanol-soluble fraction (saponin fraction) of MC and its water-soluble fraction, inhibitory activity against disaccharides the water-soluble fraction showed 5 times more inhibition rate at  $IC_{50}$  5.5 mg/ml compared to the butanol-soluble fraction at  $IC_{50}$  1.0 mg/ml (Oishi et al., 2007). In an experiment investigating glucose reduction level hourly, it was showed that despite an initial increase in fasting blood glucose (FBG) in the first hour, a significant reduction was observed at the second hour using a methanolic extract of MC. Both concentration of 125mg/kg and 375mg/kg reached the same maximum percentage reduction after 12 hours (Nkambo, Anyama & Onegi, 2013).

### **Development of MC in Drinks**

Satkar, Kulthe and Chalke (2013) prepared a ready-to-serve MC beverage using the MC juice extracted from a pulper and filtered through muslin cloth. Cane sugar and citric acid was added to adjust the total soluble solids (TSS) and acidity. The beverage was stored for 90 days at room temperature and refrigerated temperature. They had concluded that the TSS had increased during the storage period while the acidity had decreased which may be due to chemical interactions and enzymatic activity. The sensory score decreased over the storage period because of changes in colour, flavour and taste.

Another development of beverage using the MC by Din et al. (2011) used different concentrations of the MC and adding of artificial sweetener. The beverage was stored for 6 weeks and was analysed for its pH, colour, ascorbic acid and sensory properties. Similar to the previous study, the pH of the beverage increased during the storage period and there was a decline in its sensory attributes due to the increased pH and colour changes caused by possible Maillard reaction. There was also a significant loss in ascorbic acid during storage which decrease is most probably due to storage temperature, oxidative enzymes and atmospheric oxygen in head space.

Dietetic soft drinks were also subsequently being formulated using its crude polysaccharides as done by Aziz et al. (2011) Different amount of MC slices were added with sweeteners, citric acid, water and was boiled before filtration and homogenisation to obtain the beverage. It was determined that for this formulation 7.5% concentration of MC was well accepted but further studies are needed to study its effectiveness towards its anti-diabetic properties. The shelf life of the beverage was not being evaluated in this study.

### **Microbial Quality**

The microbial quality of drinks are evaluated in order to ensure safe consumption and to evaluate its shelf life. The MC plant itself contains antibacterial activity which is a possible contributor to the extended shelf life in the development of the MC drink. When tested with major foodborne pathogens such as *E. coli*, *Staphylococcus*, *Pseudomonas*, *Salmonella* and *Streptobacillus*, the water, ethanol and methanol extracts of the MC pulp and skin were found to be contain antibacterial activity against the pathogens (Saeed & Tariq, 2005).

Further microbiological analysis on drinks are necessary in ensuring the drink is free from contamination. Methods that are commonly used in microbiological analyses of drinks include the Total Plate Count (TPC), Most Probable Number (MPN) for coliform determination, *E. coli* confirmation and enumeration of yeast and moulds. TPC is considered as an important factor in accessing quality and safety of food in determining its total viable cell count (Solis et al., 2009). Coliform determination and confirmation for *E. coli* depicts the sanitary level of water, which is an important factor in drinks. Yeast spoilage is among the main spoilage factors for drinks due to high water activity ( $A_w$ ) (Battey, Duffy & Schaffner, 2002).

### **Toxicity**

In animal studies the MC is demonstrated to be safe when ingested in low doses in a duration of 2 months, but toxicity and death had been reported when administered in high doses (Grover & Yadav, 2004). Several adverse effects of overconsumption of the MC are being reported, whereby children are reported to be sick after consuming the MC, and hypoglycemic coma and convulsions are observed also in children after drinking MC tea (Lucas et al., 2010). Headaches are also reported to happen after ingesting the MC seeds (Krawinkel & Keding, 2006). The MC had also been shown to have abortifacient activity traditionally and in experiment. Also, the fruit and seeds are found to have greater toxicity than other parts of the plant (Grover & Yadav, 2004). Marles and Farnsworth (1997) suggested that the toxicity of the MC in its seeds and outer rind acted as protein synthesis inhibitors in the intestinal cells, but the observations are not linked to clinical signs or symptoms.

### **Conclusion**

Alternative methods besides medical solutions need to be implemented in countries with high prevalence of diabetes mellitus. The MC which have anti-diabetic activity can be used to develop into health products for consumers to help cope with their condition. Products such as beverages need to have more solid results of its efficacy in helping to lower blood glucose levels. The microbiological quality of the beverage has to be examined to ensure the safety of the product and at the same time to find methods to prolong the shelf life of the product. Studies also need to be done to explore the toxicity of the MC to prevent from leading to conditions due to over consumption.

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