



ANTIOXIDANT AND ANTIDIABETIC POTENTIAL OF *THEOBROMA CACAO* SEED

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Abstract

Diabetes mellitus is the common form of chronic diseases people have to search an alternative way of medications for the treatment due to high cost. Though there is no cure for diabetes, several types of treatments are available for healing the effect of this disease but currently nutritional approaches are being explored. Cocoa fruit the dried and fully fermented fatty bean is a source of chocolates is considered as a part of a healthy diet possesses antidiabetic activity. The present study was carried out to determine the phytochemical, antioxidant and antidiabetic activity of cocoa seed powder. Phytochemicals such as alkaloids, flavonoid, steroids and saponin were confirmed through the preliminary screening test. The antioxidant activity of cocoa seed powder showed increased potential effect and antidiabetic activity of 65.75%. The enriched sample product of cocoa seed help in minimizing nutritional deficiencies enhanced the anti-diabetic activity.

Key words: Cocoa fruit, *Diabetes mellitus*, Phytochemical, Antioxidant, Antidiabetic activity

Introduction

Diabetes mellitus is a chronic metabolic disorder which is characterized due to continual damage in β cells of pancreas and insulin insufficiency at the peripheral tissues (Martin *et al.*, 2016). Insulin hormone is produced by β cells of pancreas if it is not produce properly hyperglycemia is persisted. The long term hyperglycemia is correlated to elevating dyslipidemia reactive oxygen species production and reducing

antioxidants. Despite the numerous medical treatments for diabetes, the usage of medicinal plants is used to manage diabetes without any side effects. Thus inquiring for a new form of medicinal compounds is important to overcome diabetic problems (Vishwakarma *et al.*, 2010).

Generally phytomedicine obtained from plant extracts are being used to treat large variety of clinical diseases (Karyano *et al.*, 2002). The medicinal plants contain a great variety of naturally occurring compounds such as polyphenols like flavonoid, glycosides and terpenoid are effective for treating *Diabetes mellitus* (Cetto and Wiedenfiend, 2001). The increased level of glucose in blood leading to oxidative stress may produce free radicals which damage the pancreas cells. The oxidation reaction inhibited by antioxidant rich compounds is having the power to reduce the risk of Diabetic complications (Ojewole, 2005).

The cocoa tree is one of the most world's popular food products of chocolate an ancient name "kakawa" which translates into "Food of the Gods" (Pucciarelli and Grivetti, 2008). *Theobroma cacao* is an ever green plant belongs to the family Malvaceae belongs to the genus Theobroma, part of the family Sterculiaceae (Ukpanukpong *et al.*, 2019). The present investigation was carried out to analyze the phytochemical, antioxidant and anti-diabetic potentiality of cocoa fruit seed.

Materials and Methods

Collection & Processing of Sample

The Cocoa fruit samples collected from Kulasekharam were broken and the seeds were dried in sun light for two to three days, after three days the dried seeds were roasted for 10 mts. Finally, the roasted seeds were powdered by using mixer grinder and stored in an air tight container for further studies.

Phytochemical Analysis

Preliminary qualitative analysis was carried out by using the standard procedure (Harbone, 1978).

Antioxidant Activity - DPPH radical scavenging activity

The free radical scavenging activity of the fractions was measured *in vitro* by 2, 2'- diphenyl-1-picrylhydrazyl (DPPH) assay according to the standard method (Williams *et al.*, 1995).

Antidiabetic Proteinase Activity

The different concentrations of sample such as 62.5µg/ml - 500µg/ml from a stock concentration of 10mg/ml were used for the study. The reaction mixture (2ml) containing 0.06mg trypsin, 1ml 20Mm TrisHCl buffer (pH 7.4) and 1ml test sample of different concentrations. The mixture was incubated at 37°C for 5 minutes. Then 1ml of 0.8% (w/v) casein was added. 2ml of 70% per chloric acid was added to terminate the reaction. Cloudy suspension was centrifuged at 3000rpm for 10mts. The absorbance was measured using UV-Visible Spectrophotometer at 200nm (SL119, Systronics) against buffer as blank.

Result & Discussion

Phytochemical Analysis

The phytochemical analysis of cocoa seed powder extract showed the presence of alkaloids, flavonoid, steroids, phenols, saponin and reducing sugar whereas aqueous extract showed the presence of flavonoid, tannins and saponin (Table 1). The health promoting properties of cocoa were linked to their phenol compounds mainly flavonoid and a significant amount of procyanidin monomers namely catechin, epicatechin and tetradecamer also exhibited the powerful antioxidant and antidiabetic effects (Adamson *et al.*, 1999; Rios *et al.*, 2003 and Kelm *et al.*, 2006).

Table 1. Phytochemical Analysis of Cocoa seed Powder

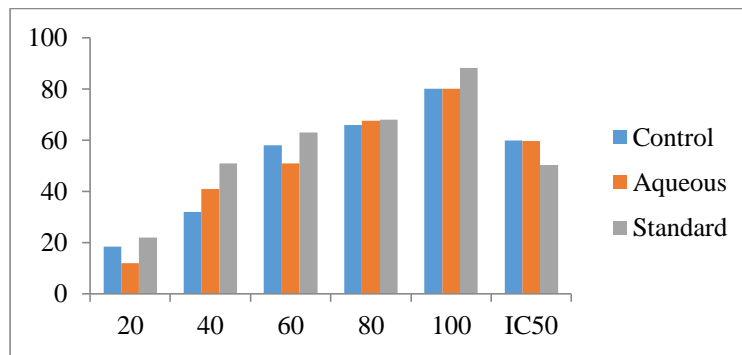
Phytochemicals	Control	Aqueous Extract
Alkaloids	+	-
Flavonoid	+	+
Steroids	+	-
Tannins	-	+
Phenols	+	-
Saponin	+	+
Anthroquinone	-	-
Reducing Sugar	+	-
Terpenoid	-	-

+ Presence - Absence

Antioxidant Activity

Antioxidant activity of Cocoa Seed powder and aqueous extract were investigated based on their DPPH radical scavenging activity and their variations were noticed (Fig 1). Inhibition concentrations (IC₅₀) of Cocoa Seed powder showed high percentage inhibition activity and slightly variations appeared in aqueous extract showed good inhibition activity when compared to standard ascorbic acid. *T. cacao* contain beta sitosterol and phytosterol has been rich antioxidants be utilized as an anti-diabetic, anti-hyperglycemic activity (Ceri *et al.*, 2019, Oracz *et al.*, 2014, Labib *et al.*, 2013, Endrini *et al.*, 2015 and Balamurugan *et al.*, 2011). For comparison, the most potent isolate obtained from cocoa pod husk using ethyl acetate extract yielded an EC₅₀ value of 16.4-53.3 µg/ml (Umri *et al.*, 2019).

Figure 1: Inhibition of DPPH scavenging Assay

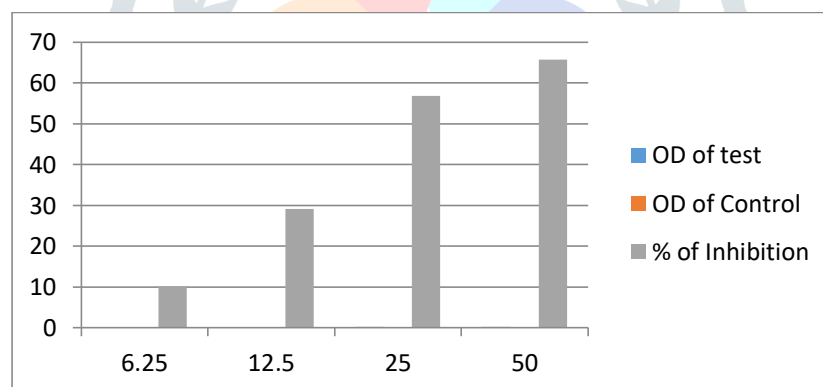


Values are expressed as mean \pm SEM

Anti-diabetic Inhibition of Proteinase Activity

The antidiabetic activity of Cocoa seed powder assessed by inhibition of proteinase assay showed high potential of 65.75 at 50 mg/ml concentration (Fig: 2). Tomaru *et al.*, (2007) reported that the Cocoa seed powder has the ability to prevent the development of diabetes is genetically inherited in diabetic rats. The earlier experimental evidence in obese-diabetic mice suggested that the cocoa prevents hyperglycemia (Corti *et al.*, 2009). The consumption of dark chocolate and cocoa improves glucose metabolism including insulin resistance and sensitivity (Grassi *et al.*, 2005).

Figure 2: Proteinase Inhibition Activity of Cocoa seed Powder



Values are expressed as mean \pm SEM

Conclusion

The present investigation concluded the phytochemical constituents of cocoa seed act as a promising herbal fruit especially for Diabetes because of its effectiveness and antioxidant potential. Further studies on therapeutic food products have to develop using cocoa fruit seed and it can be supplemented to diabetic patients to add health benefits.

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