**THE ANTI HYPERGLYCERMIC: EFFECT OF MOMORDICA CHARANTIA ON BLOOD GLUCOSE LEVEL OF ALBINO WISTAR RATS**

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 **ABSTRACT**

 The study was carried out to investigate the effect of aqueous and petroleum ether extract of momordica charantia on blood glucose level of albino rats.The extraction was done using petroleum ether and water .Thirty two albino rats were grouped into eight (A ,B,C,D,E,F,G,H). Comprising of four in each group .Diabetes was induced with a single Dose of 100mg/kg body weight of alloxan solution .After induction of alloxan ,group A and B were orally administered with 100 and 200mg/kg body weight of aqueous momordica charantia extract respectively .group C,D and E were administered with 100 ,200,300mg /kg body weight of petroleum ether extract of momordica charantia ,group F received 70mg/kg body weight of Glibenclamide ,group G received distilled water and feed and was not induced and was not induced alloxan while group H was induced with Alloxan but not treated with the extract nor synthetic drug.The glucose concentration of all the groups treated with alloxan was significantly higher than the untreated one. After treatment the Momordica charantia showed the antihypergylcermic effect on the blood glucose concentration compared to those with glibenaclamide.

Keywords: Diabetes mellitus, Alloxan monohydrate, Glibenclamide, Momordica charantia.

 LIST OF TABLES

Table 1 Grouping of the animals 20

Table 2 Determination of the blood glucose level 22

Table 3 Determination of the average body weight 24

3.2.3 Collection of animal 19

3.2.4 Experimental animal model 20

3.2.5 Collection of Blood Samples 20

3.3.6 Principle 20

CHAPTER FOUR

4.0 Results 22

CHAPTER FIVE

5.0 Discussion and Conclusion 26

5.1 Discussion 26

5.2 Conclusion 26

References

 **CHAPTER ONE**

 **INTRODUCTION**

**1.0 BACKGROUND OF THE STUDY**

The term Diabetes and Mellitus are from GREEK. Diabetes denotes Sweet It is thought that the Greeks named it so due to the excessive amount 0f urine produced by diabetic patients attracted flies and bees. The traditional way of diagnosing diabetes mellitus in ancient Chinese was by observing whether ants are attracted to a person’s urine themselves, a scene occasionally depicted in Gothic beliefs (Patlak, 2002).

Diabetes Mellitus is a combination heterogeneous disorder commonly presenting with episodes of hyperglycemia and glucose intolerance, as a result of lack of insulin, defective insulin action or both (Sicree at al., 2006). Such complication arise due to derangement in the regulatory system for storage and mobilization of metabolic fuels including the catabolism and anabolism of carbohydrate, lipids and proteins emanating from defective insulin secretion, insulin action or both (Shilliote 1988; Votey and Peter 2004). Classification of diabetes mellitus viz; type 1 diabetes, type 2 diabetes, gestation diabetes and other specific types(Sicree *et al.,* 2006).Type 1 diabetes is said to account for only minority of the total burden of diabetes in a population although it is the major type of diabetes in young age of groups at majority of well to do countries. The incidence of type 1 is increasing in both rich and poor countries. Further a shift towards type 1 diabetes occurring in children at earlier ages is imminent (Sicree *et al.,* 2006). 85% to 95% of all diabetes in high in countries are of type 2 accounting for an even higher dominance in developing countries ,It is currently common and serious health concern globally .Insulin is a polypeptide hormone synthesized in humans and other mammals within the beta cells of the islets of langerhans of the pancrease .The islets of langerhans form the endocrine part of the pancrease accounting for 20% of the total mass of the pancrease with beta cells constuting 60-80% of all the cells of islets of the langerhans (Anon 2004). Insulin inhibits a multitude of effects in mammalian tissue with liver tissue and adipose tissue being the most important target organs for insulin action .The basic synthesis of carbohydrate ,protein ,lipids ,and nucleic acids .The effect of insulin action carbohydrate metabolism include stimulation of glucose transport across muscle and adipocyte cell membrane ,regulation of hepatic glycogen synthesis and inhibition of glycogenolysis and gluconeogenesis (Pero,2006) .The end result of this action is reduction in glucose concentration with regard to protein metabolism ,insulin promotes transfer of amino acid across the membrane .stimulates protein synthesis and inhibits proteolysis ,,incorporation of fatty acids from circul;ating triglyceride and lipid synthesis are synthesized are stimulated by insulin ,lipolysis is inhibited ,Insulin contributed to nucleic acid synthesis by stimulating the formation of ATP,DNA,RNA ,(Cahill,1971).

According to WHO(1994),this problem has been aggravated by rapid cultural and social dynamics ,ageing population,increasing urbanization ,dietary changes ,reduced physical activity and other unhealthy lifestyle lifestyle and behavioural patterns .

 Diabetes mellitus and lesser form of glucose intolerance ,particularly impaired glucose tolerance ,can now be found in almost every population in the world and epidermological evidence suggest that without effective prevention and control programs ,diabetes would likelform of glucose intolerance ,particularly impaired glucose tolerance ,can now be found in almost every population in the world and epidermological evidence suggest that without effective prevention and control programs ,diabetes would likely continue globally (WHO 1994).

In 2010,about 285 million people in the age group 20-79 were envisaged to have diabetes world wide ,about 70% of whom ,Further ,by 2030,the number of people with ICT is projected to increase to 472 million or 8.4% of the adult population (Sicree et al .,2006).

Then debilitating effects of diabetes mellitus include various organ failure ,progressive metabolic complication such as retinopathy ,nephropathy and neuropathy (Piero,2006).

Diabetes are accompanied by risk of cardiovascular ,peripherial vascular and cerebrovascular diseases.

Natural plants have been valuable source of mineral agent with proven potential of treating infectious disease and with lesser side effect compared to s ynthetic drugs agent (2002).its fruits has a distinguishing bitter taste which is more pronounced as it ripens ,hence the name bitter melon or bitter gourd .Momordica charantia member of cucubitabcae family ,is known as bitter melon ,bitter gourd ,and balsam pear.it grows in tropical area of Amazon ,East African and Asian countries and is usually used traditionally as food and medicine .

Pharmacological and medicinal properties of bitter gourd have been studied by various researchers that include ,anticarcinogenic ,antioxidative ,antitumor ,immune modulating activities etc.Invitro studies reveal that the bitter gourd proteins (A and B monorcharin) have been inhibitory effect against human immune virus (HIV).Leaf extracts have demonstrated broad spectrum and antimicrobial activities against Ecoli staphylococcus,pseudomonas,sallmonelas,streptococcus{30}.furthermore,it has been refferd to posses anagelsic and abortificient properties.Biochemical and animal model have produced abundant data and hypothesis accounting for the antidiabetic effect of momordica charantia.

Biochemical and Animal model experiment have produced abundant data and hypothesis accounting for antidiabetic effects of momordica charantia.the work is aimed at evaluating the effect of petr0leum ether extractof momordica .on total scrum glucose in alloxan iinduced albino wistar rats

1.1 RESEARCH AND OBJECTIVE

To investigate the antihyperglycermic propertie of Momordia charantia.

 CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Taxonomic Hierachy of Momordica charantia

Kingdom plantae

Subkingdom Viridiplantae

Infrankingdom streptophyta

Super division Embryophyta

Division Tracheophyta

Subdivision Spermatophyta

Class Magnoliopsida

Super order Rosanne

Order Curcubitacae

Family Cucurbitacace

Genus Momordica

Species Momordica charantia

Common names \_Balsam pear,Bitter guard,Bitter cucumber,Garilla.

2.2 PLANT DESCRIPTION

Momordica charantia aslso known or bitter gourd or melon ;this tropical vine is tender perennial .Bitter melon grows in tropical areas including parts of the east Africa,Asian ,the Caribbean and South American where it is used as afood and as well as medicine ,the leaves and fruit have both been used to make teas and beer or to make soup in western countries .Bitter melon is being studied in the support treatment of diabetes and psoriases .

Bitter melon is also known by the name karela and balsam pear .it grows in tropical areas including part of East Africa ,Asia ,the Caribbean,and South America ,where it is used food as well as medicine ,it is a green cucumber shaped fruit with gourd like bumps all over it .It looks like an ugly light green cucumber .The fruit should be firm like cucumber and its taste very bitter although the seeds,leaves ,and vines of bitter melon have all been used ,the fruit is the safest and most prevalent part of the plant used medicinally .The plants are monocious –The flowers are upto 1/4inches (3cm)across and have 5 out fled eggs shaped petals .the females flowers are followed by warty,egg shaped, oblong green ,ripening to yellow and then orange fruit with a tapering tip .when fully ripe ,the orange fruit ripe ,the orange fruit split open into three three curling segment to reveal black seed ,enclosed in soft bright red ,pulp,.The leaves are green hairly,alternate and deeply palmately based with five coarsely toothed lobes .The stem have warring tendril and slender,green and five coarsely toothed lobes .The stems have warring tendril and are slender ,green and hairy bitter melons.

Momordica charantia grows under a very wide range of conditions throughout the tropic and subtropics .Its rapid growth and maturation allow it to colonize any area where there is sufficient soil term moisture .it grows from sea level to 1300m (Tjitrosoedirijo,1990)and in areas with annual rainfall as low as 480mm.minimum average annual temperatures may be as low as 12.5degree calcium though the plant is unlikely to thrive in such cold areas .it grows in soils with PH ranging from 4.3 to 8.7(Holm et al .;1997) Mormordica charantia is a fast growing vine and quickly lovers the supporting vegetation or structure .In general ,the specie can be found growing in coastal areas ,along Greeks and rivers ,forest edges and distributed sites (Hall et al;2012) for optimum growth .Momordica charantia needs a strong support 1-4m tail ,however ,it will also grow as a matted ground cover .genetics the chromosome number reported for M. charantia is 2n-22(Holm et al ,1999).

Momordica charantia is a monocious species and reproductive activity starts when plants are 30 to 33 days old (days after germination).flowering and fruiting can occur throughout the year .flowering starts with flower about 30days after germination while the female flower appears 10days later.flowering may continue for upto 6months .flowers open early in the morning before daylight with anthesis at about down .Anthers dehisce about two hours before anthesis and optimum viability of pollen and receptivity of the stigma are attained at anthesis flowers are pollinated by bees and other insects (Holm et al,;1997,PROTA 2014).

Momordica grows best in wet areas from sea level to 100m in elevation ,precipitation ranging from 480mm to 4100mm and temperature between 125degree celcius and 25degree celcius it prefers deep drained sandy highly organic matter content ,water retaining capacity and with the PH ranging from 4.3 to 8.7.

Most of the insect and pathogens that affect cucurbit crops in the tropics also affect momordica when it grows as a crop and presumably when it occurs as a weed .There is a very large amount of literature on insect and pathogen affecting momordica as a crop especially in india .pests of a particular note include root knot nematodes (Me loidogunesp)(paruth et al .;1995)epilachunabettles(Ahmed and khatun,mosaic poty virus (fulamoto et al.1996).

2.2.1 stems

Stems prostrate five edges ,dark green,hairy,section of the main vine buds movement strong and can branch out lateral and bilateral vine section bud also could grow outside lateral vine create a more luxuriant vine leaf system is one of the melon specie which has more lateral vineand slim .in addition ,there are section outside auxillary buds and trendrils solitary.

2.2.2.1 Leaf

Bitter gourd cotyledon unearthed ,generally do not carry out photosynthesis .primary leaves one pair ,opposite ,shield shape,green ,after the leaf is alternate ,palmately lobed ,green abaxially pale green ,veins radia ,with five radial veins ,leaf length 16-18cm and width 18-24cm ,petiole length a -10dm ,yellow –green.

2.2.3 Flower

Flowers monocious ,plants generally generate male flower ,female later ,solitary,Male calyx bell shaped ,separated 5green ,petal yellow ;0.2cm ,green ,stamen 3,separation with five Athens ,the s-shaped bend near one another joint .morning flowering most flowering.

22.4 Fruit

Bitter gourd fruit are berries that have many irregular surfaces tubercels fruit shape fusiform ,short,round hammer shaped oblong hammer shape etc..,color green white and white yellow when ripen.Ripening fruit too easy to easy to crack ,exposing the blood red flesh .wrapped seeds .The fruit is edible when harvested green and cooked ,the taste is bitter.

2.2.5 Seed

Bitter melon seeds are large, flat tortoise shell like, yellow thick seed coat the surface pattern each fruit contain seeds from 20 to 30 weights 150-180g per 1000 seeds. Acre planted with 200 to 300g.

2.3DISTRIBUTION OF MOMORDICA CHARANTIA

Momordica charantia is widely distributed throughout tropical and subtropical regions on all containers ,it appears to be native to the African and Australian continents .Buts its actual origin has been obscured by its spread as a food crop .currently it can found cultivated and neutralized in north central and south American the west.

2.4MEDICINAL PROPERTIES OF MOMORDICA CHARANTIA

Pharmacological and medicinal properties of bitter gourd have been studied by various researchers that include antidiabetic ,antinecerogenic ,antimutagenic , antioxidative ,antitumor ,immune modulating activities etc .Invitro studies reveal that the bitter gourd proteins (A and B monocharin)have inhibitory effect against human immune virus (HIV). Leaf extracts have demonstratated broad –spectrum and antimicrobial activities against coli staphylococcus, pseudomonas, salmonella, streptobacillus and streptobacillus etc.it has been referred to possess antipolyptic ,,analgesic,and abortificient.

2.4.1 Anti-Tumour properties

Some researchers have found that bitter melon fruit contained anticarcinogenic or chemo protective agent .Several in vivo studies have demonstrated the antimogorous activity of the entire plant of bitter gourd .In one study a water extract blocked the growth of rat prostrate carcinoma ;another study reported that a hot water extract of the entire of the entire plant contributed to development of mammary tumours in mice .numerous invitro studies have also demonstrated anticancerous and antileukemic activity of bitter gourd against numerous cells lines including liver cancer ,human leukaemia ,mlanana and solid sacronom,antidiabetic .The other means showing that bitter gourd is an effective immunomodulator .However, one clinical trial found very limited evidence that bitter melon might improve immune cells function in people with cancer ,but this needs to be verified and amplified on the research .

4.2 Anti-inflammatory properties

Wild species bitter gourd (WBG) is considered to be more potent in disease prevention than cultivable species. A study on the anti-inflammatory effect of wild species of bitter gourd (WBG) on lipopolysaccharide (LPS) stimulated RAW264.7 macrophages suggested that WBG is beneficial for reducing LPS-induced inflammatory responses by modulating NF-KB activation . In this study ,among the hot water 95% ethanol and acetate extract of wild species of bitter gourd (WBG) the ethanol extracts .Compared showed a peak reduction of (LPS)-induced nitric oxide(NO) and prostaglandins E2(PGE2)production and inducible oxide synapse (iNOS) and pro inter leukin -1 beta expression .

LPS-induced cyclooxygenase -2-expression was not affected by WBG extract. Compared to WBG extracts .Compared to WBG ,the extracts of normal bitter gourd showed a lesser inhibition on LPS –induced events electrophoetic mobility shift assay further sowed that both the hot water and ethanol extract of WBG inhibited NF-KB activation .Although information is lacking on bioactive component of WBG ,the phinotic compound potent of each significantly parallel is anti-inflammatory activity or ability (37) .

2.5 DIABETES MELLITUS

**D**iabetes for the whole world is not an epidemic anymore but has turned into pandemic (Lal et al.; 2009). According to the world health organization (WHO) projections .The prevalence of diabetes is likely to increase by 35% in the year 2025(Asaduzman et al.; 2010).Indian has a high prevalence of diabetes and the numbers are increasing at an alarming rate .it is expected to increase from 40.6 million in 2006 to 79.4 million by 2030(Mehtaet et al.; 2009).

2.5.1 DEFINITIONS OF DIABETES MELLITUS

Diabetes mellitus is a complex chronic disease, it is a condition characterized by elevation of glucose in the blood .Insulin a hormone produced by pancreas controls the blood storage of glucose .In diabetes there may be decrease in insulin produced by the pancreas which can lead to abnormalities in protein Carbohydrate and fats protein.

The resulting hyperglycermia may lead to acute metabolic complications including ketoacidosis and in the longterm contribute to macrovascular complications (Smeltzer and Bare,1992).

2.5.2 CLASSIFICATION OF DIABETES MELLITUS

2.5.3 TYPE1 DIABETES

Type1 diabetes mellitus is characterised by loss of the insulin –producing cells of the islet of langerhans in the pancreas, leading to insulin deficiency. This type can be further classified as immune –mediated or idiopathic. The majority of type1 diabetes is of the immune mediated nature in which beta cells loss is a T-cell-mediated autoimmune attack.(Rother and K.I. April ,2007). Type1 insulin dependent diabetes is present in 5-10% of all diabetics ,but is increasing in adolescent minority groups (Malese et al.,2007) The prevalence of type1 diabetes increases with age and the overall incidence of the disease may be increasing (Lewder and silverstein ,2005).

Genetic predisposition ,autoimmunity and viral infection are the main etiological factors complicated in the pathogenesis of type1 diabetes .(Muuthukrishman et al ,.2007).Type 1 diabetes which may develop at any age is a chronic autoimmune disease characterised by irreversible autoimmune destruction of the insulin secreting beta cells of the islet in the pancreas .There is hepatic over production of glucose by glycogenolysis and gluconeogenesis and decrease cellular uptake of glucose from the circulation.Type1 diabetes requires lifelong treatment with exogenous insulin for survival .(Mehra et al .,2007).

2.5.4 TYPE2 DIABETES MELLITUS

Type 2 diabetes is the most common form of the disease .accounting for about 90 to 95% of diagnosed disease of diabetes .In type2 diabetes the body does not produce enough insulin or the cells ignore the insulin(Badyaland Kaur,2008).The incidence of type2 diabetes increasing among all age groups including adolescence ,among whom type2 diabetes was formerly rare(Metzger ,2008).

Type2 diabetes occurs usually on individuals over 40yearsof age and dramatically increases as a result of changes in human behaviour and increased body mass index (Elmer et al., 2004).The increasing portion of the ageing population ,consumption of carorierich diet ,obesity, and the sedentary life style has led to a tremendous increase in the number of diabetes worldwide .(Elshe nawy and Abdeiby ,2006).this form of diabetes previously referred to insulin resistance and usually have relative(rather than absolute )insulin deficiency .At least initially and often throughout their life time .,these individual do not need insulin treatment to survive .most patient with this form of diabetes obese and obesity itself causes some degree of insulin resistance .this form of diabetes frequently usually go undiagnosed for many years (American diabetes association ,2010).impair intrauterine growth and nutrition results in the programming of system that regulate insulin sensitivity ,Insulin secretion and energy storage and utilization throughout the lifetime of the individual (Metzger 2006)the familiar predisposition of type2 diabetes is mediated by both genetic and intrauterine environmental factors (Seshial et al.,2008)

 2.5.5 GESTATION AND DIABETES MELLITUS

The onset of gestational diabetes mellitus is during pregnancy usually secreted by the placenta which inhibits the action of insulin , it occurs in about 2-5% 0f all pregnancies about the 30-40% of patients with question diabetes mellitus will develop type2 diabetes within 5-10 years (especially if obese). Impaired glucose tolerance and statistical risk groups are example of gestational diabetes . statistical risk groups are individuals o f greater risk than the general population of developing diabetes and the risk factors include immediate family members with disease and presence of is lets cell anti bodies (Royle and waish, 1992:597 smeltzerand Bare 1992:1022) .

2.5.6 CAUSES OF DIABETES MELLITUS

The causes of types diabetes mellitus are described and include genetic factor immunologic factor environmental factor and of infectious agents.

GENETIC FACTOR

People do not inherit type1 diabetes mellitus itself but they inherit a genetic predisposition towards developing type1 diabetes mellitus .The genetic tendency have been found in people found in people with certain HLA. HLA is a cluster of gene responsible for transplantation of antigens and other immune processes .The risk of developing type1 diabetes is increased in three to five times in people which one of these two HLA types Rayland Walsh 1992 :596;Smeltzland Bare 1992:1022.

IMMUNOLOGIC FACTORS

People with diabetes have an autoimmune response is abnormal response in which antibodies are directed against normal tissue of the body, responding to tissue as if they are foreign. Auto-antibodies against islet cells against endogenous (internal) insulin have been detected in people at the time of diagnosis and even several years prior to the development of clinical sign of type1diabetes.

2.5.7SIGNS AND SYMPTOMS OF DIABETES MELLITUS

The classic symptoms of untreated diabetes is weight loss and polyuria (frequent urination ).polypsdia(increased thirst ).(cookie et al .,2008)symptoms may develop rapidly weeks or months ) in type1 diabetes while they are usually develop much more slowly and maybe subtle in type2 diabetes .

Prolonged high blood glucose can cause glucose absorption in the lens of the eye which leads to changes in its shape .blurred vision is a common complaint leading to diabetes diagnosis .A number of skin rashes that occur in diabetes are collectively known as diabetic dermadromes (Cookie ,et al,2008).

2.5.8PREVENTION 0F DIABETES MELLITUS

Diabetes mellitus risk can be reduced by increased physical activities ,proper breast feeding and moderate supply of vitamin D during early stage of life (stuebe et al .,2008).

American diabetes association recommends maintaining a healthy weight ,getting at least two and half hour of rest per week and eating sufficient fibre.

2.5.8TREATMENT AND MANAGEMENT OF DIABETES

DIET: Diet therapy is the cornerstone of treatment in diabetes especially for patient with type2 diabetes .it is difficult to maintain dietary control for a long time but dietary control is important and necessary (shabidder et al 2006).

PLANT COMPOUND AS ANTIDIABETIC AGENTS: the use of plant by man for treatment of disease is an age long practise .Diabetes was also known in ancient times and some medicinal plant have been used for its control in traditional medicine (Muckhejee et al .,2006)The oral antihyperglycermic agent currently used in clinical trials have characteristic profile of serious side effect .

 **CHAPTER THREE**

3.0MATERIALS AND METHOD

3.1 MATERIALS

Water bath (griffen )

spectrophotometer

Reagent bottles

Nose mask

Weighing balance

Cage(locally made )

Electric grinder

Conical flask

Stirrer

**T**est tube

Syringe

Nose mask

Spatula

Muslin cloth

Beaker

Pipettes

Reagent bottles

Electric grinder

Hand gloves

CHEMICAL AND REAGENTS

Petroleum ether

Alloxan

Glibenclamide

3.2 METHODS

3.2.1 COLLECTION AND IDENTIFICATION

Fresh leaves of momordica charantia were collected from p&t garden and was authenticated by Mr.Donatus a taxonomist in University of Nigeria Nsukka .the leaves were dried at room temperature for 14days after which they were blended and weighed and dried in an air tight container.

3.2.2PREPARATION AND EXTRACTION OF SAMPLES

200g of granulated leaf was weighed and soaked in 100ml of petroleum ether for 8hrs and was stirred intermittently every 4hrs .The samples was filtered and filtrate was obtained and was dried at room temperature .the percentage yield was gotten after drying them and was kept in an airtight container .

3.2.2 COLLECTION OF ANIMAL

The animals were obtained from the University of Nigeria Nsukka in Enugu state and were kept in the biological science animal house of Godfrey Okoye University.

3.2.3COLLECTION OF BLOOD SAMPLES

Blood samples were collected by a snip cut on the tail of the vein and blood sugar level was measured with Accu –check glucometer.

3.2.4PRINCIPLE

When blood is dropped on the yellow square spot on the test strip inserted inside the glucometer, glucose in the blood react with the chemical reagent on the strip .Glucometer test strip is base on the double sequential enzyme reaction in which an enzyme reaction glucose oxidase converts glucose hydrogen peroxide and glucoronic acid while perioxidase oxidizes the dye in the test strip to produce a colour .the blood glucose level in mg/dl will be displayed on the screen after 20seconds.

 **CHAPTER FIVE**

**DISSCUSION AND CONCLUSION**

**5.1 DISCUSSION**

In the experiment Alloxan administration was shown to induce hyperglycermia in albino rats .The induction of hyperglycermia on administration of alloxan in the presence and absence of glucose has been attributed to cytotoxic action of reactive oxygen species with a simultaneous massive increase in cytosolic calcium concentration (Szkudelski, 2001 ) it exerts diabetogenic action when it is administered intravenously or subcutaneously (William ,1963).Furthermore ,oral administration of petroleum ether of momordica charantia (karela )to Alloxan induced diabetic rats lowered their blood glucose level by suppressing gluconeogenesis by inhibition of the key hepatic gluconeogenic enzyme glucose -6-phosphatase and fructose 1,6,bisphosphate dehydrogenase and stimulating glucose oxidation by pentose phosphate pathway through activation of glucose 6 phosphate dehydrogenase the rate limiting enzyme .

5.2CONCLUSION

Momordica charantia extract appears to have active principle which elicited antihyperglycermic effect.