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(SRI-VIPRA)

SRI-VIPRA

Project Report of 2023: SVP-2337

Protective role of *Curcuma longa* extract supplementation in STZ induced diabetic rats

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SRIVIPRA PROJECT 2023

Title: Protective role of *Curcuma longa* extract supplementation in STZ induced diabetic rats

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List of students under the SRIVIPRA Project:

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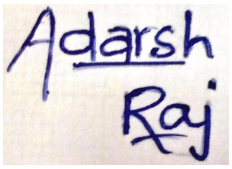
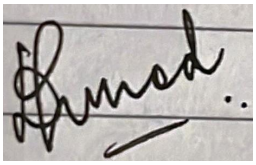
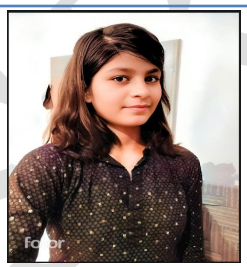
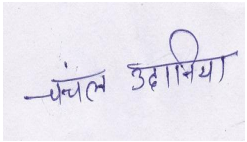
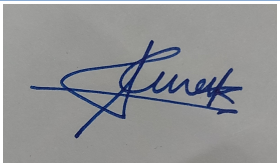
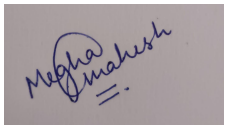
1		Adarsh Raj	1121121	B.Sc., Life Sciences	
2		Devyanshi Vinod	1321038	B.Sc., Biological Sciences	
3		Chanchal Udania	1121120	B.Sc., Life Sciences	
4		Aakanksha Chak	1121085	B.Sc., Life Sciences	

					
5		Mohammed Abaan Kurikkal MP	1321020	B.Sc., Biological Sciences	
6		Megha Mahesh	1121139	B.Sc., Life Sciences	

Signature of Mentor

Certificate of Originality

This is to certify that the aforementioned students from Sri Venkateswara

College have participated in the summer project **SVP-2337** titled "**Protective role of *Curcuma longa* extract supplementation in STZ induced diabetic rats"**. The participants have carried out the research project work under my guidance and supervision from 15th June, 2023 to 15th September, 2023. The work carried out is original and carried out in an online/ offline / hybrid mode.

Signature of Mentor

Acknowledgements

We wish to express our heartfelt gratitude to our mentor and guide **Dr. Obaiah Jamakala,** Assistant Professor, Department of Zoology, Sri Venkateswara College, University of Delhi, New Delhi for his supervision, support, creative ideas, masterly guidance offering valuable assistance at various stages of our progress, critical evaluation

and comments, patient understanding, continuous encouragement and a vision to pursue new ideas and unexplored territories with freedom during our project work.

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We take this opportunity to express our indebted gratefulness to all those who helped directly and indirectly in this eventful journey of our project work, it's our pleasure to acknowledge the help of all individuals.

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ABBREVATIONS

DM	-	Diabetes mellitus
NCD	-	Non-communicable disease
IDDM	-	Insulin dependent diabetes mellitus

NIDDM	-	Non-insulin-dependent diabetes mellitus
IDF	-	International Diabetes Federation
RIA	-	Radio Immune Assay
GDM	-	Gestational diabetes mellitus
MODY	-	Maturity onset diabetes of youth
STZ	-	Streptozotocin
RBC	-	Red blood corpuscle
WBC	-	White blood corpuscle
Hb	-	Haemoglobin
ATP	-	Adenosine triphosphate
SD	-	Standard Deviation
WHO	-	World Health Organization

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INTRODUCTION:

Diabetes mellitus (DM), commonly known as diabetes is a disorder of carbohydrate metabolism characterized by high blood sugar level (hyperglycemia) and high level of sugar in urine (glycosuria). Diabetes is a non – communicable, metabolic and lifestyle disorder in which blood glucose, also referred to as blood sugar, becomes too high (Saeedi *et al.,* 2019;

Mohammed and Tajuddeen, 2022; Sun *et al.,* 2022). It can be kept well under control and reasonably managed with proper care though it cannot be cured once it occurs but can be prevented.

Diabetes mellitus is a chronic metabolic disorder that prevents the body to utilize glucose completely or partially. It is characterized by raised glucose concentration in the blood and alterations in carbohydrate, protein and fat metabolism. Diabetes mellitus is a group of metabolic diseases characterized by high blood sugar levels, which result from defects in insulin secretion, or action, or both.

Diabetes mellitus, commonly referred to as diabetes which was first identified as a disease associated with "Sweet urine" and excessive muscle loss in the ancient world, Elevated levels of blood glucose lead to spillage of glucose in to the wine, hence the term sweet urine, normally, blood glucose levels are tightly controlled by insulin, a hormone produced by the pancreas. Insulin lowers the blood glucose level. Insulin is released from the pancreas to normalize the glucose level. In patients with diabetes, the absence or insufficient production of insulin causes hyperglycemia.

This can be due to failure in the formation of insulin or liberation or action (Papadakis, 2002). Since insulin is produced by the β-cells of the islets of langerhans, any alterations in the number of functioning cells will decrease the amount of insulin synthesis. Many diabetics can produce sufficient insulin but some stimulus to the islets tissue is needed for its secretion.

Diabetes is on increase in India. The multicentre ICMR study showed a prevalence of 2.5 percent in the urban and 1.8 percent in the rural population above the age of 15 years. The prevalence of Diabetes worldwide was found to be around 171 millions in the year 2000 and 366 millions in the year 2030, each year a further 7 million people develops diabetes, conducted a study on "Urban rural differences in prevalence of self-report Diabetes in India-The WHO - ICMR Indian non-communicable disease (NCD) Risk factor surveillance". Recent Indians; however, there are very few studies comparing urban, peri-urban and rural prevalence rates of diabetes and their risk factors al the national level.

This study is a part of the national non-communicable diseases (NCD) risk factor surveillance conducted in different geographical locations in India, This nation-wide NCD risk

factor surveillance study showed that the prevalence of self reported Diabetes is higher in urban, intermediate in Peri-urban and lowest in rural areas.

World Wide Prevalence:

Diabetes mellitus is a major public health problem worldwide. It is a long considered disease of minor significance to world health which is now taking its place as one of the major threats to human health in the 21st century. The past two decades have seen as explosive diabetes worldwide. Pronounced changes in the human environment, human behavior, lifestyle and accompanied globalization have resulted in escalating rates of both obesity and diabetes. It has been estimated that more than 33 millions of people in India are sufferers of diabetes (2000). This number is expected to increase to 57.2 million by 2025 as reported by King *et al.,* 1998. Initially considered a disease of the Western world, diabetes mellitus is now a global pandemic that affects approximately 536.6 million people worldwide and is predicted to rise to 643 million people by 2030 and 783.2 million people by 2045 (Saeedi *et al.,* 2019; Lin *et al.,* 2020; Sun *et al.,* 2022). In 2021 the International Diabetes Federation(IDF) estimated that Africa had a diabetes mellitus prevalence of 23.6 million people (4.5%) and projected an increase of up to 54.9 million people (5.2%) in 2045 (Sun *et al.,* 2022). World Health Organization (WHO) indicates that diabetes mellitus is one of the major killers of our time, with people in South East Asia and Western Pacific being at greater risk.

The incidence of IDDM is high in North European countries such as Finland (boys 36.9 and girls 31.6 per 100,000 per year), Sweden (24.2/22), Norway (23.3/20.7). Among the other racial groups such as blacks, Native Americans and Asians, the disease is less common whereas NIDDM is more prevalent in many parts of Asia and Pacific and in certain ethnic groups like Pima Indians, Norwegian, Mexicans and Americans. Outside the Europe the incidence of IDDM is less (about 6 per 100,000) among non whites compared to whites (about 29 per 100, 000). The IDDM epidemiology from many areas and populations in Asia, Africa and America are yet to be described. The risk of developing the disease by the siblings and of IDDM patients is 5-10% compared with about 0.5% in general population.

Table – I: Top countries for estimated number of people with Diabetes, 2000-2030

Ranking	2000	2030

	Country	People with Diabetes (Millions)	Country	People with Diabetes (Millions)
1	India	31.7	India	79.4
2	China	20.8	China	42.3
3	U.S.	17.7	U.S.	30.3
4	Indonesia	8.4	Indonesia	21. .3
5	Japan	6.8	Japan	8.9
6	Pakistan	5.2	Pakistan	11.9
7	Brazil	4.6	Brazil	11.3
8	Bangladesh	3.2	Bangladesh	11.1

Prevalence in India:

Diabetes has emerged as a major healthcare problem in India. According to the Diabetes Atlas published by the International Diabetes Federation (IDF), there were an estimated 40 million people with diabetes in India in 2007 and this number is predicted to rise almost to 70 million people by 2025. According to an estimate by the American Diabetes Association, there are at least 31.7 million diabetic patients in India and the number is expected to grow to 79.4 million by 2030. The countries with the largest number of diabetic people will be India, China and USA by 2030. It is estimated that every fifth person with diabetes will be an Indian. Due to these sheer numbers, the economic burden due to diabetes in India is amongst the highest in the world. The real burden of the disease is however due to its associated complications which lead to increased morbidity and mortality. WHO estimates that mortality from diabetes, heart disease and stroke costs about $210 billion in India in the year 2005. Much of the heart disease and stroke in these estimates was linked to diabetes. WHO estimates that diabetes, heart disease and stroke together will cost about $ 333.6 billion over the next 10 years in India alone.

Rapid urbanization and industrialization have produced advancement on the social and economic front in developing countries such as India which have resulted in dramatic lifestyle changes leading to lifestyle related diseases. The transition from a traditional to modern lifestyle, consumption of diets rich in fat and calories combined with a high level of mental stress has compounded the problem further. There are several studies from various parts of India which

revealed a rising trend in the prevalence of type-II diabetes in the urban areas. A National Urban Survey in 2000 observed that the prevalence of diabetes in urban India among adults was 12.1 %. Recent data has illustrated the impact of socio-economic transition occurring in rural India. The transition has occurred in the last 15 years and the prevalence of diabetes has risen from 2.4% to 6.4%.

Types of Diabetes Mellitus:

The widely accepted classification of diabetes mellitus was published by WHO in 1980 and in modified form in 1985. The 1980 and 1985 classifications of diabetes mellitus and allied categories of glucose intolerance included clinical classes and two statistical risk classes. In 1980, Expert Committee proposed two major classes of diabetes mellitus and named them, insulin dependent diabetes mellitus (IDDM) or Type-I, and non-insulin-dependent diabetes mellitus (NIDDM) or Type-II. The WHO classification of diabetes mellitus and allied categories of glucose intolerance includes number of clinical classes and two statistical risk classes. Among the different clinical classes, Type-I or IDDM or Juvenile or Childhood diabetes and Type-II or N1DDM are the two main groups.

Type-I Diabetes Mellitus:

Type-I diabetes, formerly known as IDDM is also known as childhood or juvenile diabetes. It is a heterogeneous and polygenic disorder with a number non-HLA loci (about 20) contributing to the disease susceptibility (Lernmark and Ott, 1998). The symptoms of IDDM are polydipsia, polyuria, polyphagia and ketoacidosis. It is estimated that incidence of type-I diabetes will be about 40% higher in the year 2010 than in 1997 (Okamo *et al.,* 1999). It has been estimated that yearly incidence of Type-I diabetes is 3.7 to 20 % in 100000 people. More than 700,000 Americans have been suffering with this type of diabetes. Type-I affects both children and adults but it was traditionally termed “juvenile diabetes” because it affects majority of children. The IDDM is due to complete autoimmune destruction of β-cells with maintains of the α (Glucagon secretion) and γ- (Smatostatin secreting) cells within Langerhans (Foulis *et al.,* 1986). The autoimmunity in IDDM can be evidenced by the presence of insulin autoantibodies to glutamic acid decarboxylase or 64 KDa protein in the circulation. Type-I Diabetes has less tendency to have other family members affected with diabetes than type-II. In the first large

family study of diabetes, less than 4% of parents and 6% of siblings of a person with diabetes also had diabetes. In studies with identical twins less than 50% of the siblings of a person with diabetes also had diabetes versus almost 100% of siblings of people with type-II Diabetes. Children of type-I diabetes fathers are more likely to develop type-I autoimmune diabetes than children of type-I diabetic mothers. Type-I Diabetes must be treated with insulin shots. Some new insulin pumps are being developed and tested now.

Type-II Diabetes Mellitus:

Type-II diabetes is also called adult onset diabetes as the onset usually takes place after the age of 35. Type-II diabetes is most prevalent in the world, and 90 to 95 % of the population has type-II diabetes (Polonsky *et al.,* 1996). Diabetes is the third major leading cause of deaths in the world after the deaths resulting from heart attacks and cancer. NIDDM or type-II diabetes mellitus, which was previously called maturity onset diabetes of youth (MODY) is the most common form of diabetes, often presents with less intense hyperglycemic systems or weight loss than in IDDM, and some times with none at all. The function of pancreatic β cells in NIDDM has been studied extensively since the discovery of Insulin Radio Immune Assay (RIA) by Yallow and Berson in 1960. Their studies produced astonishing results that the insulin levels in NIDDM subjects were higher or equal to non-diabetic controls. This finding led to the conclusion that NIDDM is not caused by insulin deficiency, but by ability to lower glucose levels effectively, an abnormally termed insulin resistance. Insulin resistance and pancreatic β cell dysfunction are the two prominent causes of type-II diabetes; it has been proposed that both of these complications have roots in mitochondrial defects (Lowell and Shulman, 2005). Recent findings have suggested that inherited defects in mitochondrial oxidative phosphorylation activity might play a key role in the development of IR (Insulin Resistance). Sreekumar and Nair (2007) have studied the interrelation between skeletal muscle mitochondrial changes resulting in reduced oxidative phosphorylation and insulin resistance in type-II diabetes, the ability to produce insulin does not disappear completely. But the body becomes increasingly resistant to insulin, so tablets are needed to balance this. The tablets do not contain insulin. But act by increasing the sensitivity to it, or by increasing there release of insulin from the pancreas. It is rare for insulin injections to be necessary in the early stages of type-II diabetes. Although type-II diabetes is also called NIDDM, many people need treatment with insulin at a later stage in the

same way as people with type-I diabetes. An increasing number of reports from North America, Japan, the UK, and other parts of the industrialized world indicate that overweight teenagers are now beginning to develop type-II diabetes. This appears to be more common in girls than in boys.

The characteristic features which can be used to distinguish IDDM from NIDDM are 1. A history of sudden onset of hyperglycemia 2. Marked recent weight loss. 3. Spontaneous sustained ketosis or ketonuria and 4. Presence of Markers of autoimmune activity in circulation.

Gestational Diabetes:

Gestational diabetes mellitus (GDM) also involves a combination of inadequate insulin secretion and responsiveness, resembling type-II diabetes in several respects. It develops during pregnancy and may improve or disappear after delivery. Even though it may be transient, GDM may damage the health of the fetus or mother, and about 20%-50% of women with GDM develop type-II diabetes later in life. GDM occurs in about 2%-5% of all pregnancies. It is temporary and fully treatable but, if untreated, may cause problems with the pregnancy, including macrosomia (high birth weight), fetal malformation and congenital heart disease. It requires careful medical supervision during the pregnancy.

Modern scientific pharmacologists concentrated to a large extent on curative or system regulating medicines. Preventive medicine has not made much progress except for the handful of vaccines and basic capsules. In this context it is essential for us to turn to Ayurveda as a holistic science, it tends to consider various factors which help human beings to achieve healthy living with no side effects by using herbal medicines. Approximately four billion people in developing countries depend on herbal traditional medicine for the treatment of metabolic diseases such as diabetes mellitus because of the presence of a wide range of bioactive phytochemical compounds in plants (Ekor, 2014; Choudhury *et al.,* 2018). Plant-based traditional medicines are considered to be cheap and readily available to the majority of the rural population (Mugumbate *et al.,* 2018). By conducting large number of research work, numerous traditional medicines have been found for diabetes.

Ayurvedic Approach:

Ayurveda, an Indian traditional medicine provides an effective solution for this possible risk. It is effective because allopathic approaches are found to produce more side effects. Ayurveda otherwise called herbal therapy is steadily increasing its popularity even in the western world today. This 5,000 year old system of medicine recommends a combination of lifestyle management (which includes diet, exercise and meditation), and treatment with specific herbs and minerals to cure various diseases. The botanicals in the Ayurveda have been proven to be safe and effective, through several hundred to several thousand years of use.

Ayurvedic physicians have treated diabetes for thousands of years using a combination of regulated lifestyle and herbal formulations. The following paragraphs summarizing the description of diabetes mellitus by two ancient Indian physicians are excerpted here from a fairly recent publication on Ayurveda.

About the one transmitted genetically, he (Sushruta) says *"a person would be diabetic if his father and grandfather are diabetic*". In fact, he mentions that such type of person is clinically diabetic. The genetically transmitted entity of insulin dependent diabetes mellitus is well known today. The characteristics of diabetes of dietary origin are described to be exactly opposite, which also fit in with the features of Type-II mentioned in modern medicine. Charaka too agrees with the genetic origin of diabetes and adds that this type is more difficult to cure. The ancient physicians have written factors predisposing to diabetes mellitus, and these stand confirmed even today. The factors described are lack of exercise, sedentary habits, sleeping during day time and eating excessively, particularly sweet and fatty substances. These individuals lack enthusiasm, are overweight, obese and have excessive appetite.

The physicians also prescribed specific herbal formulations for the treatment of diabetes. In recent times, the safety and efficacy of these herbs have been validated by laboratory experiments and clinical trials. Since olden days, plants are used to treat many ailments. India has about 45,000 plant species and several thousands have been claimed to possess medicinal properties. A large variety of compounds obtained from several plant families were found to hypoglycemic effect. The glycosides, glycans, certain triterpenes, various types of sulfide molecules, polysaccharides, oils, vitamins, alkaloids, saponins, glycoproteins, peptides, amino acids and proteins isolated from various plant families showed beneficial effects in reducing the

blood sugar (Rahman *et al.,* 1989). Many Indian medicinal plants are reported to be useful in diabetes (Kirithikar *et al.,* 1975) (Nadkarni, 1976; Nadkarni, 1997; Sadak Basha *et al.,* 2010a; 2010b; Guru Sekhar *et al.,* 2010). Medicinal plants used to treat hypoglycemic or hyperglycemic conditions are of considerable interest for ethno-botanical community as they are recognized to contain valuable medicinal properties in different parts of the plant and a number of plants have shown varying degree of hypoglycemic and anti-hyperglycemic activity. The active principles of many plant species are isolated for direct use as drugs, lead compounds or pharmacological agents. Several species of medicinal plants are used in the treatment of diabetes mellitus. Traditional plant medicines or herbal formulations might offer a natural key to unlock diabetic complications.

Antioxidants play an important role to protect against damage by reactive oxygen species and their role in diabetes has been evaluated. Many plant extracts and products were shown to possess significant antioxidant activity. In the present study *Curcuma longa* plant was selected for evaluation of their antioxidant potential mediated anti-diabetic activity.

***Curcuma longa*:**

Taxonomy:

Kingdom : Plantae
Division : Magnoliophyta
Class : Liliopsida
Order : Zingiberales
Family : Zingiberaceae
Genus : *Curcuma*
Species : *longa*

Turmeric (*Curcuma longa*) is a rhizomatous herbaceous perennial plant of the ginger family, Zingiberaceae which is native to tropical South Asia. It is often pronounced as turmeric. Its active ingredient is curcumin and it has an earthy, bitter, peppery flavor and a mustardy smell.

In Ayurvedic medicine, turmeric is thought to have many medicinal properties and in India many people use it as a readily available antiseptic for cuts, burns and bruises. Practitioners of Ayurvedic medicine say that it has fluoride which is thought to be essential for teeth. It is also

used as an antibacterial agent. It is taken in some Asian countries as a dietary supplement, which allegedly helps with stomach problems and other ailments. It is popular as a tea in Okinawa, Japan. The active ingredient in turmeric is exploding. U.S. National Institutes of Health had four clinical trials to study curcumin treatment for pancreatic cancer, multiple myeloma, Alzheimer's, and colorectal cancer. Curcumin has been used for thousands of years as a safe anti-inflammatory agent in a variety of ailments as part of Indian traditional medicine". A recent study involving mice has shown that turmeric slows the spread of breast cancer into lungs and other body parts. Turmeric also enhances the effect of taxol in reducing metastasis of breast cancer.

Researchers had discovered that turmeric-treated mice were less susceptible to developing type-II diabetes, based on their blood glucose levels, and glucose and insulin tolerance tests. They also discovered that turmeric-fed obese mice showed significantly reduced inflammation in fat tissue and liver compared to controls. They speculated that curcumin in the turmeric lessens insulin resistance and prevents type-II diabetes in these mouse models by dampening the inflammatory response provoked by obesity. Curcumin and its analogues have a variety of physiological and pharmacological activities such as anti-inflammatory, anti-carcinogenic and antioxidant properties (Osawa *et al.,* 1995; Sreejayan *et al.,* 1997; Sadak Basha *et al.,* 2010a; 2010b; Guru Sekahar *et al.,* 2010).

AIMS:

- To evaluate the beneficial effects and Protective role of Plant Extract of *Curcuma longa* against Streptozotocin induced Diabetes.
- To determine the role of plant extract on hematological and serum biochemical parameters.

MATERIAL AND METHODS:

Procurement and Maintenance of Animals:

Healthy female albino wistar rats (180±20g) were procured from Sri Venkateswara Enterprises, Bangalore, Karnataka, India (Reg. No: 237/99/CPCSEA). Animals were maintained in the animal house of Sri Venkateswara University, Dept of Zoology, Tirupati. Rats were kept in sterilized polypropylene cages lined with paddy husk (18"x10"x8"). The animals were

maintained under a regulated 12 h light: 12 h dark scheduled at 24±1ºC and relative humidity of 55±15%. Rats were provided standard rat chow (Sai Durga Feeds and Foods, Bangalore, India) and water *ad libitum*. The protocol and animal use has been approved by the Institutional Animal Ethics Committee, Sri Venkateswara University, Tirupati, Andhra Pradesh, India.

Procurement of chemicals:

All the chemicals used in the present study were Analar Grade (AR) and were obtained from Sigma (St. Louis, MO, USA), Fisher (Pitrsburg, PA, USA), Merck (Mumbai, India), Ranbaxy (NEW Delhi, India), Qualigens (Mumbai, India) scientific companies.

For the present work Barnstead Thermoline water purification plant was used for Nano pure water, Kubota KR 200000T centrifuge for centrifugation of the homogenates and Hitachi UV -2000 Spectrophotometer for measuring the optical density values were used for high –quality results.

Streptozotocin (STZ):

Systematic (IUPAC) name: - 2-deoxy-2-({[methyl (nitroso) amino] carbonyl} amino)-β-D-glucopyranose

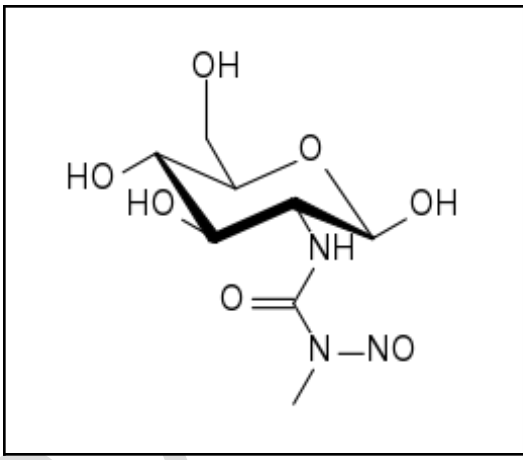
Identifiers

CAS number : 18883-66-4

ATC code : L01AD04

PubChem : 29327

Drug Bank : APRD00209



Chemical data

Formula : **C**8**H**15**N**3**O**7

Mol. Mass : 265.221 g/mol

SMILES molecules and PubChem

Properties: **Streptozotocin (C8H15N3O7)**

Streptozotocin is a mixture of α- and β-stereoisomers. It occurs as pale yellow or off-white crystals, powder, or platelets, while the research grade may be off-white to tan solid. It is very soluble in water, ketones, and lower alcohols, slightly soluble in polar organic solvents, and insoluble in monopolar organic solvents. The pure compound is sensitive to humidity and light. Streptozotocin decomposes to diazomethane in alkaline solutions at 0°C. When heated to decomposition, it emits toxic fumes of nitrogen oxides (IARC 1978, HSDB 2001).

Preparation of *Curcuma longa* extract:

The fine powder of *Curcuma longa* rhizome was purchased (AGMARK symbol) in Tirupati. The powder is extracted by cold percolation with 95% ethanol for 24h. The extract was recovered and 95% ethanol was further added to the plant material and the extraction was continued. The process was repeated three times. The three extractions were pooled together, combined, filtered and the filtrate was concentrated to dryness under reduced pressure in rotary evaporator. The resulting ethanol extract was air-dried. Finally light yellow powdery, crude ethanol extract of *Curcuma longa* was obtained. Without any further purification the plant crude ethanol extract was used in the study. Dose equivalent to 250mg kg/body was calculated and suspended in 2% v/v Tween 80 solution for the experiment (Sadak Basha *et al.,* 2010a).

Induction of Diabetes:

Streptozotocin (STZ, 2-deoxy-2-({[methyl (nitroso) amino] carbonyl} amino)-β-Dglucopyranose) frequently used dosage is 40mg/kg BW (Ganada *et al.,* 1993) Single injection of STZ given intravenously or intraperitonially to the adult rats to induce diabetes.

After fasting for 18hrs, rats were injected intraperitonially with a single dose of 40mg STZ (Sigma, St. Louis, Mo., USA) freshly dissolved in 0.1 M cold sodium citrate buffer, (pH 4.5). After injection, they had a free access to food and water were given 5% glucose solution to drink overnight to counter hypoglycemic shock. Diabetes in rats was identified by moderate polydipsia and marked polyuria. From the second day onwards fasting blood samples were collected from the rats by tail vein and blood glucose was measured by Accu chek Sensor comfort glucometer (Manufacture-Johnson and Johnson) to know the induction of diabetes. If the blood glucose levels were more than 300mg/dL, insulin (IIU Protamine Zinc Insulin) is given to the diabetic rats for diabetic condition for one week. After one week the rats with hyperglycemia (blood glucose level 250mg/dL) were selected and used for the experiment (Radha Madhavi *et al.,* 2012).

Grouping of animals:

Group -1 : Normal Control rats.

Group- 2 : Diabetic rats (Streptozotocin)

Group -3 : Diabetic rats treated with 250 mg/Kg b.w. of *Curcuma longa*.

The blood samples were collected after completion of treatment i.e. on 22nd day of the treatment. The blood was used for the hematological parameters and separated serum was used for the serum biochemical parameters.

Estimation of Blood glucose:

Estimation of Blood glucose levels was carried out by using Accu Chek glucometer (Sensor Comfort).

Body Weight Changes:

Body weights of all groups of (eight) rats were recorded before and after treatments. The body weights of all groups were recorded at an interval of one week till the completion of the experiential period (21 days).

Haematology:

Blood samples were collected at the end of experimentation period immediately after sacrifice the blood was collected from the jugular vein and the blood was allowed into a

graduated centrifuge tubes containing 10% EDTA, a common anticoagulant used for routine hematological work. The blood parameters like total erythrocytic count, total leucocyte count, hemoglobin and hematocrit (PCV) were estimated by using standard procedures as per Jain (1993).

Red blood corpuscle (RBC) count:

RBC count was estimated by using Biosystems haematology analyzer. RBC count was expressed in million of RBC/Cu.mm.

Estimation of Haemoglobin concentration (Hb):

Hb concentration was estimated by Sahali's method. Hb concentration was expressed in grams per cent.

White blood corpuscle (WBC) count:

WBC count was estimated by using Biosystems haematology analyzer. The WBC count was expressed in number of 10^3 WBC/Cu.mm.

Biochemical Parameters:

With out adding anticoagulant, the blood was collected into separate tubes and subject for centrifugation and serum collected was used for biochemical analysis. The parameters such as of glucose, total proteins, albumin, globulin, total cholesterol, creatinine, blood urea nitrogen, and bilirubin were estimated by using diagnostic kits supplied by SD fine, Ranbaxy, span diagnostics Ltd., India, and the procedures mentioned in the kit.

Estimation of Total Proteins:

Serum total proteins were estimated by Biuret Method (Tietz, 1996) using the kit purchased from Excel Diagnostics Pvt. Ltd. Total proteins levels were expressed as g/dl.

Estimation of Albumin:

Serum albumin was estimated by BCG Dye Binding Method (Tietz, 1996) using the kit purchased from Excel Diagnostics Pvt. Ltd. Albumin levels were expressed as g/dl.

Estimation of Globulin:

Serum globulin was estimated by BCG Dye Binding Method (Tietz, 1996) using the kit purchased from Excel Diagnostics Pvt. Ltd. Albumin levels were expressed as g/dl.

Estimation of Urea:

Serum urea was estimated by DAM Method (Fearon, 1939) using the kit purchased from Excel Diagnostics Pvt. Ltd. Albumin levels were expressed as g/dl.

Estimation of serum Total Cholesterol:

Serum cholesterol was estimated by Nader *et al.,* (2001) using the kits purchased from Excel Diagnostics Pvt. Ltd. Total cholesterol levels were expressed as mg/dl.

Estimation of serum Creatinine:

Serum creatinine was estimated by Jaffe's Method (Bowers, 1980) using the kits purchased from Excel Diagnostics Pvt. Ltd. Creatinine level in serum was expressed as mg/dl.

Statistical analysis:

All assays were did in six replicates from each group. The data was analyzed using ANOVA programme to know the differences between means of each experimental group and its age matched controls. Differences were did under the conditions following zero order kinetics.

RESULTS AND DISCUSSION:

Blood Glucose:

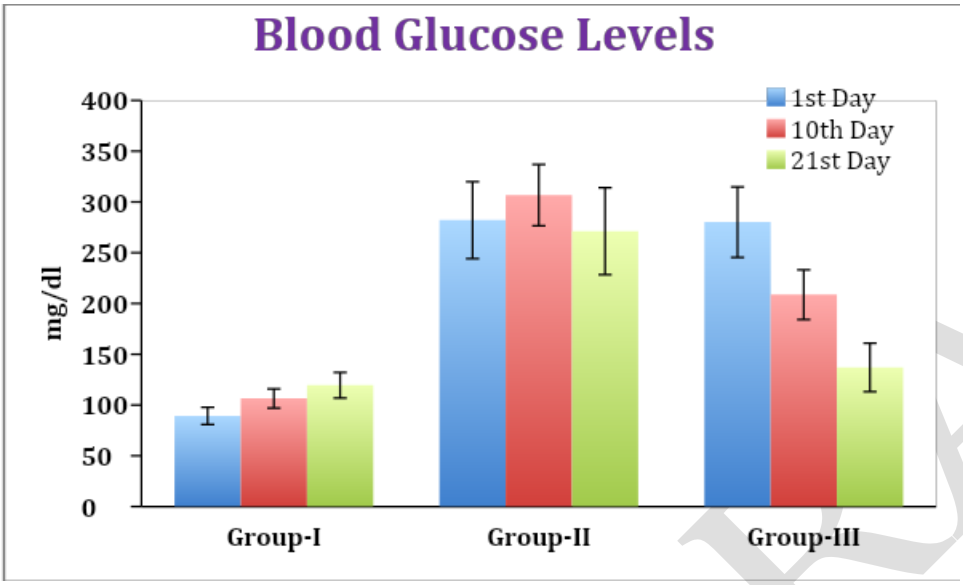
Blood glucose levels were measured using glucometer (Accu Chek) in control, diabetic, diabetic treated with *Curcuma longa* extract groups before and after treatment. In group II, the blood glucose levels were significantly increased after induction with STZ when compared with control. Blood glucose levels were significantly decreased in the group III, where the rats were subjected to *Curcuma longa* extract. The various blood glucose values of alterations are as shown in Table-1 and Figure-1.

Table-1: Showing Blood glucose levels in the control and experimental animals

Days	Group-I	Group-II	Group-III
1st Day	89.33 ±2.38	282.16 ±3.92	280.16 ±4.62
10th Day	106.50 ±2.35	306.66 ±3.32	208.67 ±2.41
21st Day	119.52 ±2.56	271.33 ±4.78	137.16 ±3.92

- Values are mean ± S.D. of 6 individual rats

Figure-1: Showing Blood glucose levels in control and experimental animals



- Values are mean ± S.D. of 6 individual rats

Discussion:

Most of the body cells use the sugar called glucose as their major source of energy. Glucose molecules are broken down within cells in order to produce adenosine triphosphate (ATP) molecules, energy-rich molecules that power numerous cellular processes. Glucose molecules are delivered to cells by the circulating blood and therefore, to ensure a constant supply of glucose to cells, it is essential that blood glucose levels be maintained at relatively constant levels. Level constancy is accomplished primarily through negative feedback systems, which ensure that blood glucose concentration is maintained within the normal range of 70 to 110 mg/dl. The levels of glucose in the blood are monitored by the cells in the pancreas. If the blood glucose level falls to dangerous levels (as in very heavy exercise or lack of food for extended periods), the Alpha cells of the pancreas release glucagon, a hormone which alerts the liver to increase blood glucose levels and converts stored glycogen into glucose (Glycogenesis). Thus glucose is released into the blood stream, increasing blood sugar levels. There are several other causes for an increase in blood sugar levels. Among them diabetic stress due to the accumulation of reactive oxygen spices is a major cause.

In the present study blood glucose levels were maintained at normal levels in control rats. A significant increase in glucose levels found in STZ treated rats could be due to the distruction of pancreatic beta-cells by STZ induced oxidative stress. The elevation of glucose in STZ treated rats was due to an oxidative stress produced in the pancreas, due to a single strand break in

pancreatic islets DNA (Omamto and Uchigata, 1981). In experimental diabetes, enzymes of glucose and fatty acid metabolism are markedly altered; hence blood glucose levels were increased (Gottried and Rosenberg, 1973; Sochar *et al.,* 1985). An increased hyperglycemia has been reported to induce oxidative stress due to glycation of proteins and accumulation of polyols (Low *et al.,* 1997). One of the consequences of hyperglycemia is increased metabolism of glucose by sorbitol pathway. Besides this, other path ways, such as fatty acid and cholesterol biosynthesis favor hyperglycemia (Vijay kumar *et al.,* 2006). Hyperglycemia is currently considered to be primarily responsible for the auto-oxidative glycosylation, formation of hydro peroxides and free radicals, in particular the hydroxyl radical and low density lipoprotein oxidation (Hunt *et al.,* 1990).

The action of STZ in the pancreas is preceded by its rapid uptake by the B cells (Weaver *et al.,* 1978a, Boquist *et al.,* 1985). Rapid uptake by insulin-secreting cells has been proposed to be one of the important features determining STZ diabetogenicity. Another aspect concerns the formation of reactive oxygen species (Heikkila *et al.,* 1976). A similar uptake of STZ also takes place in the liver. However, the liver and other tissues are more resistant to reactive oxygen species in comparison to pancreatic β cells and this resistance protects them against STZ toxicity (Malaisse *et al.,* 1982; Tiedge *et al.,* 1997). The formation of reactive oxygen species is preceded by STZ reduction. In beta cells of the pancreas its reduction occurs in the presence of different reducing agents. Since STZ exhibits a high affinity to the SH-containing cellular compounds, reduced glutathione (GSH), cysteine and protein-bound sulfhydryl groups (including SH containing enzymes) are very susceptible to its action (Lenzen and Munday, 1991). However, other reducing agents such as ascorbate may also participate in this reduction (Zhang *et al.,* 1992). Lenzen *et al.,* (1987) proposed that one of the SH-containing compounds essential for proper glucose-induced insulin secretion is glucokinase (EC 2.7.1.2), being very vulnerable to STZ. STZ reacts with two -SH groups in the sugarbinding side of glucokinase resulting in the formation of the disulfide bond and inactivation of the enzyme. Glucose can protect glucokinase against the inactivation hindering the access of alloxan to the -SH groups of the enzyme (Lenzen *et al.,* 1987, 1988, Lenzen and Mirzaie-Petri, 1991).

In case of rats which were subjected to both STZ and plant extracts, the decrease in blood glucose was due to the hypoglycemic activity of the extracts. Changes of blood glucose levels in the group III where diabetic rats were treated with plant extract is due to the flavonoid and

triterpenoid compounds in them. A number of investigations had reported that 6-gingerol, tannins, polyphenolic compound, flavonoids, triterpenoids posses analgesic, hypoglycemic and other pharmacological actions in various experimental animal models (Jiang *et al.,* 2006; Ojwole, 2006).The plant favorably affected glycolytic, gluoneogenic, and liogenic enzymes to restore glucose homeostasis in STZ-induced diabetic rats (Raju *et al.,* 2001). The administration of *Curcuma longa* powder to diabetic animals has been shown to lower blood glucose levels and partially restore the activities of key enzymes of carbohydrate and lipid metabolism close to normal values in animal model systems (Vats *et al.,* 2003; Raju *et al.,* 2001). Oxidant induced alterations in the glucose utilizing system during diabetic manifestation is partially reversed by the administration of herbal extracts (Methanol extracts (75%) of *Aegle marmelos, Momordica charantia, Trigonella foenum-graecum, curcuma longa, Eclipta prostrata, Salacia oblonga, Coriandrum sativum, Vernonia anthelmintica* and *Murraya koenigii*) having antioxidant activity (Sabu, 2003). Various reports demonstrated that the *Curcuma longa* have hypoglycemic, hypocholserolemic and hyperinsulinomic effects on type 1 and type 2 diabetes mellitus patients and experimental diabetic animals (Khosla *et al.,* 1995 Puri *et al.,* 1995; Stark and Madar, 1993; Duke, 1992). Oral administration of extract from *Curcuma longa* lowers blood glucose and attenuates STZ-induced hyperlipidemia in diabetic rabbits. (Sarah Nwozo, *et al.,* 2009). *Curcuma longa* rhizomes he been reported to possesses active constituents showing blood glucose lowering activity in STZ induced diabetic rats (Shnkar *et al.,* 1980). Curumin has been shown to lower blood glucose levels in typ-2 diabetic KK-ay mice (Nishiyama *et al.,* 2005) and STZ treated rats (Mahesh *et al.,* 2005). The administration of an aqueous extract of turmeric and abromine powder resulted in a significant reduction in blood glucose.

Body Weights:

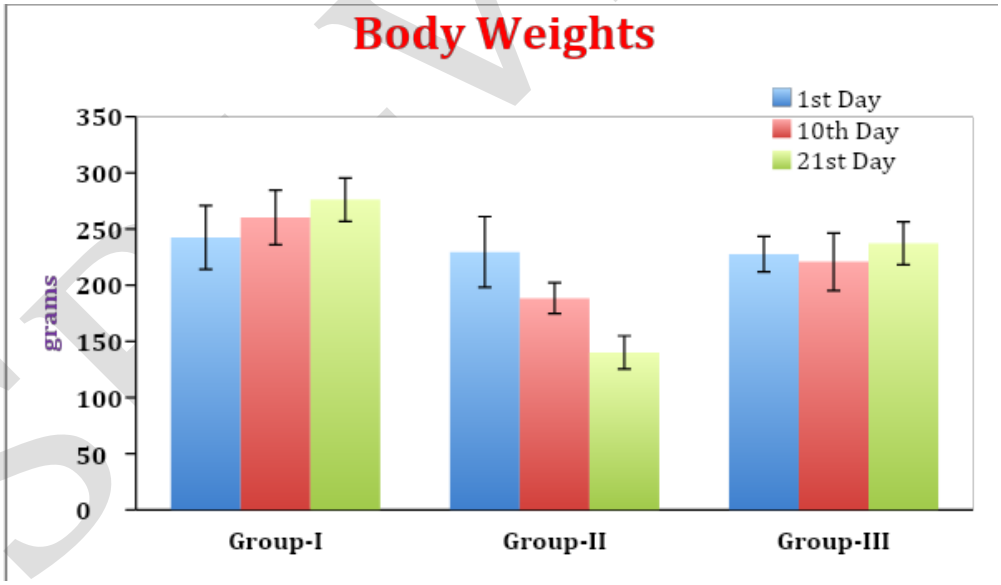
Body weights of rats were measured using a digital balance at an interval of 10 days during the experimental period. The initial average weight of animals was in the range of 180 ± 20g. In group II, the body weights were significantly decreased after induction with STZ when compared with the control rats. In the group III, the body weights were significant increased when compared with the diabetic (group II) rats. The changes of body weights are as shown in Table-2 and figure-2.

Table-2: Showing Body Weight levels in the control and experimental animals

Days	Group-I	Group-II	Group-III
1st Day	242.53 ±2.81	229.66 ±2.38	227.64 ±1.73
10th Day	260.43 ±2.31	188.42 ±1.76	220.88 ±2.57
21st Day	276.17 ±1.28	140.19 ±1.71	237.33 ±1.02

- Values are mean ± S.D. of 6 individual rats

Figure-2: Showing Body weights levels in control and experimental animals



- Values are mean ± S.D. of 6 individual rats

Discussion:

Body weight is determined by energy intake on one hand and energy expenditure on the other. Imbalance between energy intake and expenditure results in a change in body weight. Organisms expend energy to perform daily work required for survival, such as finding food or evading predators. Metabolic efficiency refers to the amount of energy an organism has to exert to perform a given amount of work.

Metabolic efficiency varies among different species of organisms and among different individuals within a species. An individual with high metabolic efficiency will expend less energy to perform a specific task, such as climbing a set of stairs, than an individual with low metabolic efficiency. Compared with an individual with low metabolic efficiency, an individual with high metabolic efficiency is better able to preserve body weight during negative daily energy balance (expenditure exceeding intake), but likely to gain more weight during positive energy balance (intake exceeding expenditure). The ability of an organism to minimize reduction in body weight during long periods of starvation is likely associated with its survival. As a result, millions of years of evolution may have favored organisms with high metabolic efficiency. (Neel *et al.,* 1962, Knowler *et al.,* 1983; Ravussin and Bogardus, *et al.,* 1990; Sharma *et al.,* 1998).

A constellation of clinical studies has established the close link between obesity and type 2 diabetes. (Must *et al.,* 1999; Hu *et al.,* 2001; Mokdad *et al.,* 2001) This correlation, however, is not perfect; many diabetic patients are not obese, and many obese individuals are perfectly responsive to insulin. Regardless of whether a causal relationship exists between obesity and the body's response to insulin, beneficial effects of weight loss on the metabolic parameters of many diabetic patients are well documented. (Olefsky *et al.,* 1974; Pi-Sunyer, 1993) Thus, it is not surprising that a combination of weight loss and exercise is an effective treatment for many diabetic patients (Klein *et al.,* 2004).

In the present study, STZ induced diabetic rats showed decreased level of body weights. The decrease in body weight in diabetic rats clearly shows a loss or degradation of structural proteins. Weight loss which is one of the clinical features of diabetes mellitus may be due to the degeneration of the adipocytes and muscle tissues to make up for the energy lost from the body due to frequent urination and over conversion of glycogen to glucose. Weight loss is a very serious issue in the management of diabetes mellitus (Reno and Leland, 1999).

Due to diabetes the structural proteins are known to contribute for the body weight (Rajkumar and Govindarajulu, 1991). STZ induced diabetes is characterized by a severe loss in body weight (Chen and Lanuzzo, 1982). The control diabetic animals showed a significant decrease in body weight compared with normal rats (Al Amin, 2006). Changes in body weight in adult and non adult diabetic rats varied. Since the non adult diabetic rats are in the growing age, diabetic loss of weight is not seen in them and they even show a slight weight gain. In adult rats, however diabetes is an accompanied by loss of weight (Akbarzadeh *et al.,* 2007). Weight loss during diabetes is mainly related to urinary glucose excretion because cells become to use glucose. Another factor could be also the osmotic diuresis resulting in hyper osmotic dehydration (Kaplan *et al.,* 1982).

In the case of diabetic rats treated with *Curcuma long* extract (group III) increased levels in body weights were observed. They showed almost same response as that of control rats. This shows that *Curcuma long* plant extract apposes degeneration of the adipocytes and muscle tissues which occurs during diabetic stress in order to make up for the energy lost from the body due to frequent urination and over conversion of glycogen to glucose.

Haematological Parameters:

Results:

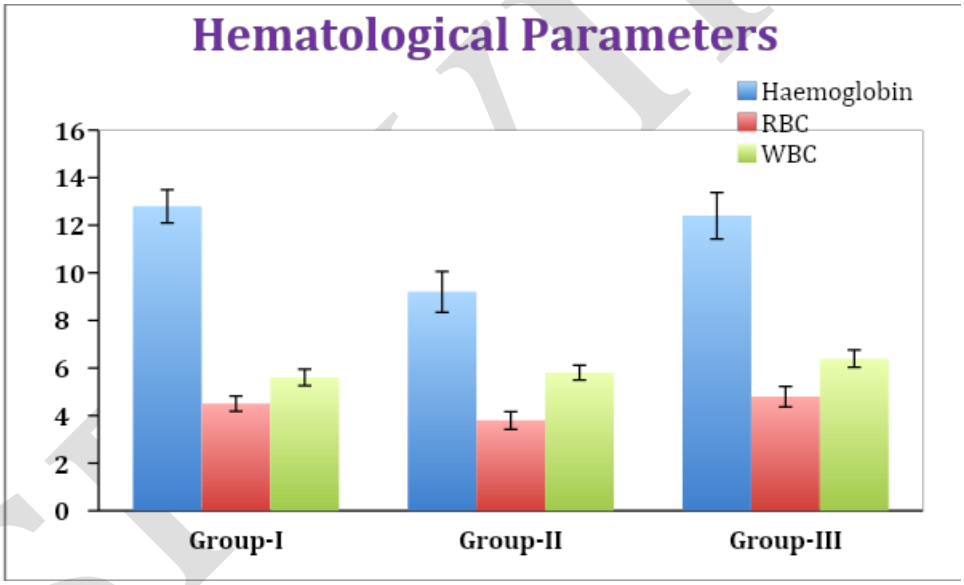
Significant decreased levels of haemoglobin and RBC observed during diabetes when compared with corresponding control group. But WBC was slightly increased in diabetes rats. Administration of *Curcuma longa* extract tended to bring the values to near to normal range and the effect was more pronounced in the group of rats treated with plant extract.

Table-3: Showing Blood Parameters levels in the control and experimental animals

Parameters	Group I	Group II	Group III
Haemoglobin gm/dl	12.8 ± 0.7	9.2 ± 0.86	12.4 ± 0.98
RBC millions/µl	4.5 ± 0.32	3.8 ± 0.37	4.8 ± 0.43
WBC cells/µl	5600 ±1.23	5800 ± 1.03	6400 ± 1.24

- Values are mean ± S.D. of 6 individual rats

Figure-3: Showing Blood Parameters levels in the control and experimental animals



- Values are mean ± S.D. of 6 individual rats

Discussion:

In diabetic rat haemoglobin (Hb) levels were found to be low when compared to normal rats, as the Hb synthesis might also be depressed. Thus *Curcuma longa* treated rats showed improved levels of Hb because of its glucose lowering effect. The various proteins including

haemoglobin undergo an enzymatic glycation in diabetes. Glycosylated haemoglobin was found to be increased in diabetes condition and the amount of increase is directly proportional to that of fasting blood glucose level (Sheela & Augusti, 1992; Mude Ravi Naik *et al.,* 2010). Lowered levels of haemoglobin were observed in diabetic rats which might be due to the increased formation of HbA1c. Hyperglycemia is the clinical hallmark of poorly controlled diabetes, which is known to cause glycation, and also known as nonenzymatic glycosylation. HbA1c was found to increase in patients with diabetes mellitus and the increase was directly proportional to the fasting blood glucose levels (Alberti, 1982). Previous studies reported that the active components present in *Curcuma longa* were effective in raising the haemoglobin levels in rats.

The link between chronic diseases and anemia is well characterized (Weiss and Goodnough, 2005). The occurrence of anaemia in diabetes mellitus has been reported due to the increased non-enzymatic glycosylation of RBC membrane proteins, which correlates with hyperglycemia (Oyedemi *et al.,* 2011). Oxidation of these proteins and hyperglycaemia in diabetes mellitus causes an increase in the production of lipid peroxides that lead to haemolysis of RBC (Arun and Ramesh, 2002). The major pathological consequences of free radical induced membrane lipid peroxidation include increased membrane rigidity, decreased cellular deformability, reduced erythrocyte survival, and lipid fluidity (Kolanjiappan *et al.,* 2002). In this study, the RBC membrane lipid peroxide levels in diabetic rats were not measured. The reversal effect shown by the *Curcuma longa* were effective in reduce the RBC levels in rats.

Peripheral WBC count has been shown to be associated with insulin resistance, type 2 diabetes (Ohshita *et al.,* 2004), coronary artery disease (Lee *et al.,* 2001), stroke (Lee *et al.,* 2001), and diabetes micro- and macrovascular complications (Tong *et al.,* 2004). Peripheral blood leukocytes are composed of polymorphonuclear cells, including monocytes as well as lymphocytes. Polymorpho- and mononuclear leukocytes can be activated by advanced glycation end products (Pertynska-Marczewska *et al.,* 2004), oxidative stress (Shurtz-Swirski *et al.,* 2004; Radha Madhavi *et al.,* 2010), angiotensin II (Lee *et al.,* 2004b), and cytokines (Scherberich, 2003) in a state of hyperglycemia. Leukocytes may be activated through the release of cytokines, such as TNF-α (Shanmugam *et al.,* 2003), transforming growth factor-1 (Korpinen *et al.,* 2001), superoxide (Kedziora-Kornatowska, 1999), nuclear factor κB (NF-κB) (Hofmann *et al.,* 1998), monocyte chemoattractant protein 1, interleukin-1β, and others (Shanmugam *et al.,* 2003) to participate in the pathogenesis of diabetic micro- and macrovascular complications. The profile

of the WBC count reflects the balance between the rate of granulocyte production and that of WBC. Kozlov et al. (1995) reported that diabetes in mice was accompanied by moderate neutrophilic leukocytosis and prolonged circulation times of neutrophils and monocytes, and a shortened circulation time of lymphocytes, which increases the susceptibility to infection. The raised leukocyte count may also reflect low-grade inflammation. The active components present in *Curcuma longa* decreases the WBC count.

Serum Biochemical Parameters:

Results:

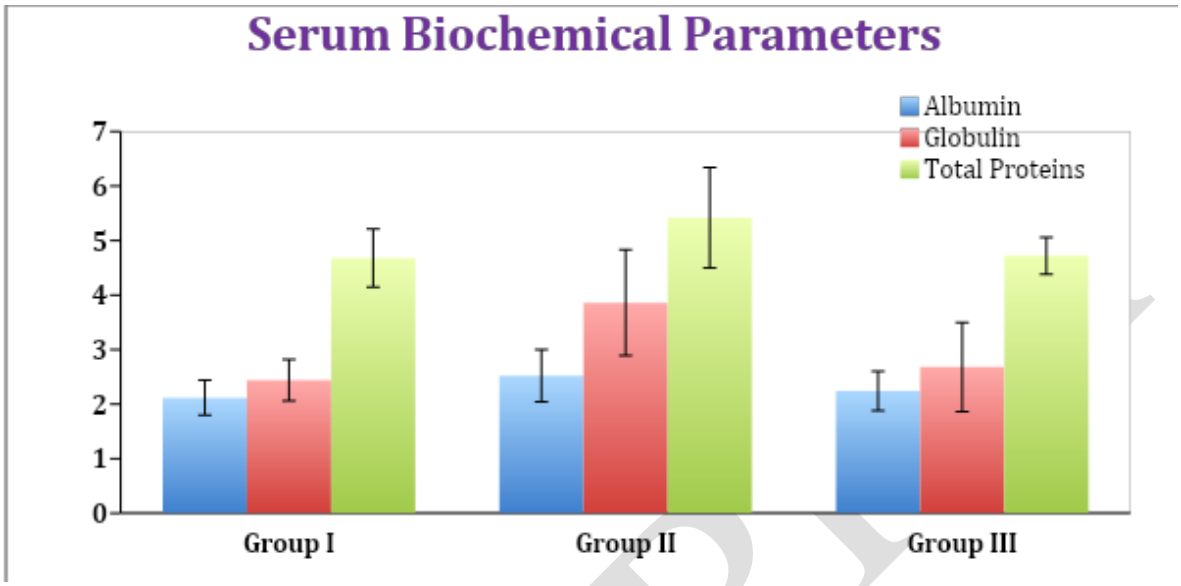
A significant increase in serum total proteins (5.42), albumin (2.52) and globulin (3.86) was recorded in diabetic untreated rats when compared to the normal control rats (Group-I). *Curcuma longa* extract treated diabetic rats showed significant decrease in serum total proteins (4.72), albumin (2.24) and globulin (2.68) levels compared to the diabetic rats and also reached the nearly levels of the control rats.

Table-4: Showing Serum Biochemical Parameters levels in the control and experimental animals

Parameters	Group I	Group II	Group III
Total Proteins g/dl	4.68 ± 2.53	5.42 ± 2.92	4.72 ± 0.34
Albumin g/dl	2.12 ± 0.32	2.52 ±2.48	2.24 ± 2.36
Globulin g/dl	2.44 ± 0.38	3.86 ± 2.97	2.68 ± 2.82

- Values are mean ± S.D. of 6 individual rats

Figure-4: Showing Serum Biochemical Parameters levels in the control and experimental animals



- Values are mean ± S.D. of 6 individual rats

Discussion:

Hyperlipidemia is a known complication of diabetes mellitus and coexists with it and is characterized by increased levels of cholesterol and also changes in lipoprotein patterns (Hafern, 1991; Bagdade *et al.,* 1974). Interest in the study of plasma lipids in diabetes arises from the widely acknowledged higher incidence of atherosclerotic disease which is a major cause of premature death in diabetic patients whether it is type-I or type-II (Betteridge, 1989).

In the present investigation, results show a significant in plasma albumin, globulin, total protein levels in diabetic rats which are in agreement with many earlier reports. These alterations in diabetes are due to enhanced catabolism of proteins (Babu and Srinivasan, 1997; Willatgamuwa *et al.,* 1998; Mude Ravi Naik *et al.,* 2010). It is well known that in insulin deficiency (diabetes) decreased protein synthesis and increased protein degradation lead to release of amino acids which are directed for gluconeogenesis. Lowered albumin and globulin in diabetic rats might be due to increased degradation and/or decreased production and/or increased urinary excretion of these substrates. Microalbuminuria in STZ-diabetic rats and humans is well documented with increased albumin excretion range (AER) (Berg *et al.,* 1997) and formation of

advanced glycation and products (AGEs) leading to kidney damage and diabetic glomerulopathy (Bangstad *et al.,* 1993). *Curcuma longa* supplementation appears to have rectified this abnormality in diabetic rats as evidenced by significantly elevated serum albumin levels in rats receiving *Curcuma longa* observed that the risk of progression to over proteinuria can be reduced by improved glycemic control. In the present study also, the glycemic control exerted by *Curcuma longa* might have contributed to the restored plasma albumin levels. Moreover, supplementation of *Curcuma longa* might have induced protein synthesis by effective utilization of the available amino acids and also by reducing protein catabolism and/or by regulating certain signal transduction mechanisms and enzymes. Phytochemicals of *Curcuma longa* extract appear to have mitigated the metabolic abnormalities and restored the urea and creatinine levels.

Insulin is the principal regulatory hormone involved in the tight regulation of fuel metabolism. In response to blood glucose levels, it is secreted by the β-cell of the pancreas and exerts its effects by binding to cell surface receptors that are present on virtually all cell types and tissues (Cheng *et al.,* 2002). In the present study, normal rats treated with *Curcuma longa* extract showed normal levels of insulin while diabetic rats had shown very low levels of insulin as a consequence of pancreatic β-cell damage indicating low pancreatic β-cell activity followed by Streptozotocin.

Results:

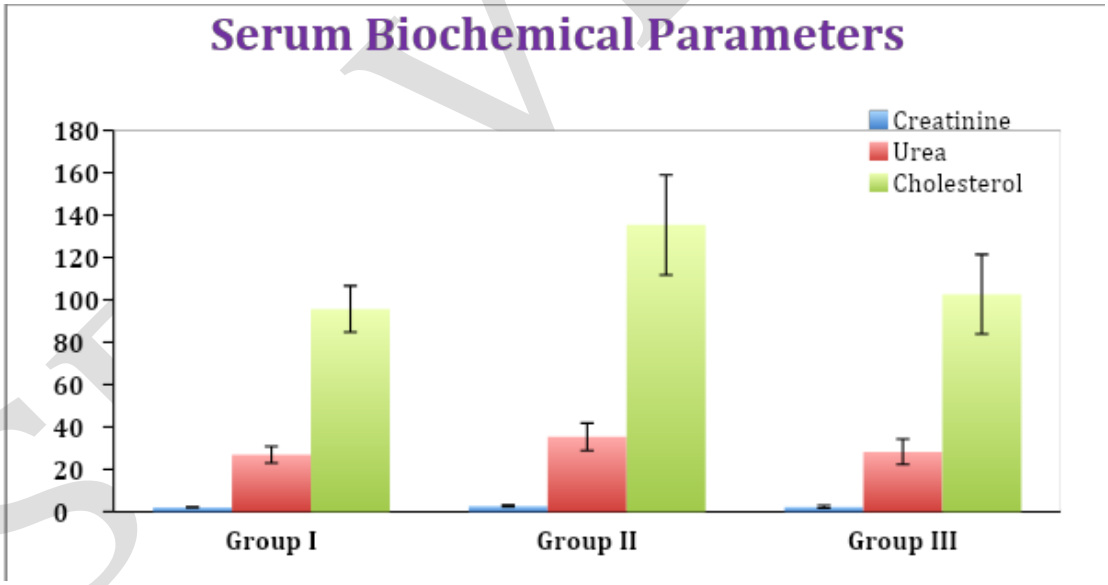
The serum biochemical parameter of control rats are Creatinine (2.16), Urea (26.945) and (95.74) was tabulated in Table-5. Increased levels of the creatinine (2.83), urea (35.36) and cholesterol (135.38) were recorded in the diabetic untreated rats (group-II). *Curcuma longa* treated diabetic rats showed significantly decreased levels of creatinine (2.34), urea (28.28) and cholesterol (102.68) when compared to the diabetic rats and also near to the normal rats (Table-5 and Figure-5).

Table-5: Showing Serum Biochemical Parameters levels in the control and experimental animals

Parameters	Group I	Group II	Group III
Creatinine mg/dl	2.16 ± 0.16	2.83 ± 0.42	2.34 ± 0.68
Urea mg/dl	26.94 ± 1.04	35.36 ± 1.96	28.28 ± 1.98
Cholesterol mg/dl	95.74 ± 1.92	135.38 ± 2.6	102.68 ± 2.04

- Values are mean ± S.D. of 6 individual rats

Figure-5: Showing Serum Biochemical Parameters levels in the control and experimental animals



- Values are mean ± S.D. of 6 individual rats

Discussion:

Hyperlipidemia is a known complication of diabetes mellitus and coexists with it and is characterized by increased levels of cholesterol and also changes in lipoprotein patterns (Hafern, 1991; Bagdade *et al.,* 1974). Interest in the study of plasma lipids in diabetes arises from the widely acknowledged higher incidence of atherosclerotic disease which is a major cause of premature death in diabetic patients whether it is type-I or type-II (Betteridge, 1989; Radha Madhavi *et al.,* 2012).

In the present investigation, results show a significant in urea and creatinine levels in diabetic rats which are in agreement with many earlier reports. These alterations in diabetes are due to enhanced catabolism of proteins (Babu and Srinivasan, 1997; Willatgamuwa *et al.,* 1998). It is well known that in insulin deficiency (diabetes) decreased protein synthesis and increased protein degradation lead to release of amino acids which are directed for gluconeogenesis. Due to increased catabolism of proteins and amino acids, hepatic ureagenesis and creatinine production are elevated in diabetic rats (Iyer *et al.,* 1996; Mude Ravi Naik *et al.,* 2010). As a consequence, increments in urea and creatinine levels occur in plasma (Ignacimuthu and Amalraj, 1998). Microalbuminuria in STZ-diabetic rats and humans is well documented with increased albumin excretion range (AER) (Berg *et al.,* 1997) and formation of advanced glycation and products (AGEs) leading to kidney damage and diabetic glomerulopathy (Bangstad *et al.,* 1993). *Curcuma longa* supplementation appears to have rectified this abnormality in diabetic rats as evidenced by significantly elevated serum albumin levels in rats receiving *Curcuma longa* observed that the risk of progression to over proteinuria can be reduced by improved glycemic control. In the present study also, the glycemic control exerted by *Curcuma longa* might have contributed to the restored plasma albumin levels. Moreover, supplementation of *Curcuma longa* might have induced protein synthesis by effective utilization of the available amino acids and also by reducing protein catabolism and/or by regulating certain signal transduction mechanisms and enzymes. Phytochemicals of *Curcuma longa* extract appear to have mitigated the metabolic abnormalities and restored the urea and creatinine levels.

Insulin is the principal regulatory hormone involved in the tight regulation of fuel metabolism. In response to blood glucose levels, it is secreted by the β-cell of the pancreas and exerts its effects by binding to cell surface receptors that are present on virtually all cell types

and tissues (Cheng *et al.,* 2002). In the present study, normal rats treated with *Curcuma longa* extract showed normal levels of insulin while diabetic rats had shown very low levels of insulin as a consequence of pancreatic β-cell damage indicating low pancreatic β-cell activity followed by Streptozotocin.

SUMMARY AND CONCLUSION

In present investigation, anti-diabetic properties of *Curcuma longa* in STZ induced diabetic rat hematological parameters and serum biochemical parameters were studied with the blood glucose and body weight levels. Wistar stain male albino rats of 3 months age were used in the present study. They were maintained in the animal house at 24±2° C, humidity of 45-64% with photoperiod of 12 hours light and 12 hours darkness. Regarding selection of age and grouping of animals as mentioned in "Material and methods" was taken in to consideration to select 3 months old rats as adult age in this experimental design for expected results. They were maintained in clean poly propylene cages and fed with standard rat pellet diet (Hindustan lever Ltd., Mumbai) and water *ad libitum*. The animals of same age group were divided in to 3 groups, each group consists of six animals and the division of groups is as follows.

Group -1 : Normal Control rats.

Group- 2 : Diabetic rats (Streptozotocin)

Group -3 : Diabetic rats treated with 250 mg/Kg b.w. of *Curcuma longa*.

The blood samples were collected after completion of treatment i.e. on 22nd day of the treatment. The blood was used for the hematological parameters and separated serum was used for the serum biochemical parameters.

The summary of the results from this study is presented as follows:

1. No significant blood glucose level changes were observed in control rats. In diabetic rats blood glucose levels were increased. *Curcuma longa* extract supplemented rats showed decreased levels of blood glucose. This may be due to the anti-diabetic compounds present in *Curcuma longa*.
2. We observed body weight changes in the current investigation in all experimental rats. In diabetic rats, the body weights were significantly decreased after induction of STZ. The decrease in body weight in diabetic rats clearly showed a loss or degradation of structural proteins. Weight loss which is one of the clinical features of diabetes mellitus may be due to the degeneration of the adipocytes and muscle tissues to make up for the energy loss from the body due to frequent urination and over conversion of glycogen to glucose. In the

Curcuma longa treated rats, body weights were gained near to control levels after treatment with *Curcuma longa* plant extract.

3. The blood parameters revealed significant alterations in all experimental groups. In group-II (Diabetic rats) the blood parameters such as Hemoglobin, RBC, WBC counts were significantly decreased which suggest the anemic condition in the body and increased count was observed in group-III (Diabetic + *Curcuma lona*) treated rats.
4. Increased levels were observed in Albumin, Globulin, Total proteins, Creatinine, Urea and Cholesterol in group-II (Diabetic) rats indicating its impact on Soluble proteins, disturbance on immune mechanism etc, whereas the same were decreased in group-III treated with *Curcuma longa* plant extract of diabetic rats.

Based on the findings of the present study envisages that treatment with selected dosage of *Curcuma longa* extract is beneficial in countering the alterations in various blood and serum biochemical parameters. This study drawn a conclusion, stating that *Curcuma longa* treatment to diabetic rats may be beneficial to improve the metabolic efficiency and thereby improve the health status. Thus *Curcuma longa* may be useful in the formulation of herbal drugs which can be used in the treatment of diabetes.

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