

## **Hypoglycemic Activity of *Cucumis sativus* Extract on Alloxan-induced Diabetic Sprague–Dawley Rats: A Pilot Study**

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### **Abstract**

*Today, incidence of diabetes mellitus rapidly elevates worldwide and yields a momentous impact to the society, making it one of the most prevalent and life threatening diseases among adults over the age of 50. Diabetes mellitus is due to an impaired production of insulin by the pancreas leading to uncontrolled elevation of glucose in the body. Usual treatment includes oral hypoglycemic agent and the use of insulin. Moreover, continuous usage of the synthetic antidiabetic drugs causes side effects and toxicity. Therefore, seeking natural and non-toxic antidiabetic drugs is needed for diabetic treatment. Medicinal plants play a vital role in the development of potent therapeutic agents. Present investigation was undertaken to investigate the ability of *Cucumis sativus* ethanolic extract as a hypoglycemic agent on Sprague-Dawley rats randomly divided into five groups of five induced to diabetes using alloxan (120 mg/kg body weight intraperitoneal). Treatment was done by oral administration of extract at doses of 1 mL, 2.5 mL and 5 mL. The efficacy of the plant extract was compared with intraperitoneal injection of insulin (0.1 ml), a well-known hypoglycemic drug. The hypoglycemic activity of the plant extract was studied by measuring the blood glucose levels at intervals of 0, 1, 2, 3, 4, and 5 hours. Results of the study revealed that the highest hypoglycemic activity resides on treatment with 1 mL *Cucumis sativus* extract wherein the blood glucose levels of rats nearly normalize. This suggests that *Cucumis sativus* extract possesses antidiabetic effect on alloxan-induced diabetic Sprague-Dawley rat and therefore can be used as an alternative therapy in treating diabetes mellitus.*

*Key words: diabetes mellitus, *Cucumis sativus*, hypoglycemic activity, alloxan, Sprague-Dawley rats*

## **INTRODUCTION**

Hypoglycemic activity of a plant extract is its ability to lower high blood glucose levels. Hyperglycemia is the major symptom caused by diabetes mellitus wherein there is excess sugar in the circulation (high blood sugar levels); this condition happens when the body has too little insulin or when the body cannot use insulin properly (Saha et al., 2011).

Diabetes mellitus is a chronic metabolic disorder that has caused significant morbidity and mortality due to various untreated acute and chronic substantial damage to the excretory, cardiovascular and nervous systems (Saha et al., 2011). This disease is caused by a defect in the cellular uptake of glucose primarily due to either cellular resistance to insulin or reduced insulin secretion that will affect the carbohydrate, lipid, and protein metabolism (Mahendran et al., 2014). It has no known permanent treatment and is extremely prevalent worldwide (Ediriweera & Ratnasooriya, 2009). Based on the Diabetes Atlas (2013) of the International Diabetes Federation (IDF), there are approximately 382,000,000 people worldwide who have diabetes with the greatest prevalence rate between ages of 40 and 59 years old. Out of 382,000,000, there are around 3,250,000 covered by the Philippines aging from 20 up to 79 making the Philippines ranked 15 in the world diabetes prevalence (IDF, 2012; IDF, 2013). In fact, every year, there are approximately 22,345 people who die due to diabetes making it as the eighth leading cause of mortality in the Philippines (DOH, 2009).

Conventional treatment of diabetes mellitus includes lifestyle modification and the use of insulin and/or oral hypoglycemic drugs. These pharmacologic agents target increased insulin secretion, increased sensitivity to insulin, and decreased hepatic glucose production. However, the use of these agents has certain drawbacks. For insulin, such drawbacks include short shelf life, requirement of constant refrigeration, ineffectiveness of oral administration and in the event of excess dosage-fatal hypoglycemia. While the use of oral hypoglycemic agents is associated, there were wide side effects like tendency to gain weight (Tanko et al., 2008).

Though several hypoglycemic agent have been synthesized for the treatment of diabetes, many of these synthetic drugs have numerous serious adverse effects (May et al., 2002). Hence, it is a challenge to the medical

system to synthesize a more effective drug for the management of hyperglycemia that is relatively low cost and that have minimal adverse effects (Sun, 2008).

In the modern drug discovery, phytochemicals from natural resources open new ways for the treatment of various diseases including diabetes due to their comparatively low side-effects and low cost (Li, 2011).

Currently, the importance of Cucurbitaceae has been markedly recognized in empirical control of diabetes (Balaraman et al., 2010). Cucurbitaceae is a plant family consisting of 125 genera and 960 species which include crops like cucumber, luffas, melons, and squashes that are cultivated primarily on subtropical and tropical countries. This family is considered to be one of the most important families of plants with potent hypoglycemic effects (Bnouham et al., 2006). Species from this family were reported to cause remarkable hypoglycemic activity, this includes *Momordica charantia* (bitter melon) (Chauhan et al., 2010), *Cucumis melo* (melon) (Gill et al., 2011; Gill et al., 2010), and *Citrullus colocynthis* (bitter melon) (Marzouka et al., 2010).

*Cucumis sativus* (cucumber) is an edible vine that bears cylindrical fruits; it is a widely distributed plant around the world particularly in Asia, Africa and South America (Minaiyan et al., 2011). Since *Cucumis sativus* fruit is an edible plant, the thought of its toxic effect can be spelled out, though it is proven that its stems and leaves possess cytotoxic effect. Furthermore, this plant is readily available and cost-effective that is why it can be considered as a substitute anti-diabetic drug (Das et al., 2012). However, there is still no direct scientific report of *Cucumis sativus* for its anti-diabetic constitutions.

In order to determine *Cucumis sativus* extract's efficacy in lowering blood glucose levels, it is tested on different groups of laboratory animals including alloxan-induced diabetic Sprague-Dawley rats. Sprague-Dawley rats are commonly used strains of rats in experimental research due to their metabolic characteristics which are closer to the human and their ability to replicate many symptoms of human conditions (Cortés-Ortiz et al., 2014; Hubert et al., 2000).

Alloxan has been proven to induce diabetes mellitus in experimental

animals due to the selective destruction of insulin-producing pancreatic beta-islets. Henceforth, alloxan-induced models appear to be the experimental animals that yield the most reliable results (Rohila & Ali, 2012).

*Cucumis sativus* was used in the study because the researchers hypothesized that it possesses hypoglycemic effect due to the fact that it is a member of the Cucurbitaceae family, a family considered to be one of the most important plant families that has antidiabetic activity. Likewise, the present study was designed to find an alternative solution in treating diabetes mellitus using a natural treatment with less side effects.

### **Objectives of the Study**

The main objective of the present study is to investigate the ability of *Cucumis sativus* ethanolic extract as a hypoglycemic agent on alloxan-induced diabetic white male Sprague-Dawley rats. Furthermore, this study aimed to determine the blood glucose level of different groups of rats.

## **MATERIALS AND METHODS**

### **Research Design**

The present study used experimental method to identify the existence of hypoglycemic property of *Cucumis sativus* ethanolic extract. The extract was tested on different groups of rats that includes a normal group (negative control), isophane insulin treated alloxan-induced diabetic group (positive control), 1 mL/kg *Cucumis sativus* extract treated diabetic rats group, 2.5 mL/kg *Cucumis sativus* extract treated diabetic rats group, and 5 mL/kg *Cucumis sativus* extract treated diabetic rats group. Methods of other related researches were adopted and modified and were used in this study.

### **Plant Materials**

The fruits of *Cucumis sativus* were purchased from the local market in Calamba City, Laguna on September 2015. The plants were identified and authenticated at the Museum of National History, University of the Philippines – Los Baños (UPLB).

### **Chemicals**

Alloxan was purchased from Sigma-Aldrich Co., Singapore. All other

chemicals that were used in the study came from the laboratory of LPU – St. Cabrini College of Allied Medicine and other chemicals were purchased commercially. The researchers ensured that all were of analytical grade, chemical compounds of a known high standard of purity.

### **Preparation of Plant Extracts**

Authenticated plant materials were brought to the Forest Products Research and Development Institute (FPRDI) located at UPLB to obtain *Cucumis sativus* ethanolic extract. The fruits were blended with 80% ethanol and were macerated for 72 hours. Afterwards, the extract was filtered, a total of 4000 mL of plant extract was obtained. Using a rotary vacuum evaporator, 595 ml of extract with a 14% concentration was achieved. The crude plant extract was then transferred to an amber bottle and was kept in dessicator for 10 days until use.

### **Maintenance of Animal and Approval of Protocol**

Adult male Sprague-Dawley rats (98–188 g) were procured from the Food and Drug Administration (FDA). Animal facility is not yet available at the university; hence, the researchers sought the assistance of an animal laboratory at Department of Science and Technology (DOST). Rats were transported from FDA to DOST using an air-conditioned vehicle. Animals were housed in colony cages (five rats per cage) and were identified by tail tattoos, kept under standard conditions [maintained in an air-conditioned room (24± 2°C) with relative humidity of 45-55% under a 12 hour light/dark cycle] and have free access to food and water *ad libitum*. In order for the animals to adapt to the new environment, all experiments were carried out for 24 hours after their accommodation (acclimatization) for seven (7) days to allow the rats to adjust to the new environment and to overcome stress possibly incurred during transit. The researchers were not allowed to conduct certain procedures on laboratory animals. The veterinarians from DOST conduct the different experimental procedures including the induction of diabetes to laboratory animals using alloxan, blood collection, euthanization of rats using CO<sub>2</sub>, and the housing of the animals in their laboratory animal house. The ethical considerations for the use of laboratory animal were secured from the Institutional Animal Care and Use Committee (IACUC).

### Induction of Diabetes to Test Animals

The overnight fasted rats were induced to diabetes mellitus by a single dose of alloxan (120 mg/kg) dissolved in normal saline solution and was injected intraperitoneally (Stanely, Prince & Menon, 2000; Madhavan et al., 2008; Venkatesh et al., 2010). Diabetes was certified in the rats by measuring fasting blood glucose two days following alloxan injection (Ragavan & Krishnakumari, 2006; Rajagopal & Sasikala, 2008). Only test animals with glucose levels over 200 mg/dl were considered diabetic and included in the experiment (Sharma et al., 2010).

### Categorization

A 5-hour short term study was chosen to perform to check the effect of *Cucumis sativus* on alloxan-diabetic rats. Animals received extract once after recognition of their diabetic condition. Twenty five (25) laboratory rats were randomly divided into five groups (I - V) of five rats (n=5) (20 surviving diabetic rats and 5 normal rats) (Eseyin et al., 2010). The administration of drug or control was followed as indicated in the table.

**Table 1. Categorization of the Sprague-Dawley Rats**

Group	Administration
I	Negative control rats (received 5mL/kg of normal saline solution orally)
II	Positive control rats (alloxan-induced diabetic rats treated with Isophane Insulin [reference drug]0.10 mL/kg i.p.)
III	Diabetic rats treated with 1mL/kg <i>Cucumis sativus</i> extract orally
IV	Diabetic rats treated with 2.5 mL/kg <i>Cucumis sativus</i> extract orally
V	Diabetic rats treated with 5 mL/kg <i>Cucumis sativus</i> extract orally

### Blood Sampling and Chemical Analysis

Blood samples were collected by cutting the tail-tip of the rats. The fasting blood glucose levels and body weight of the animals were determined at 0, 1<sup>st</sup>, 2<sup>nd</sup>, 3<sup>rd</sup>, 4<sup>th</sup>, and 5<sup>th</sup> hour. Determination of blood glucose levels was done by an enzymatic glucose-oxidase principle using ONE TOUCH Basic (Lifescan, Switzerland, Europe) instrument and results were reported as mg/dL (Sedigheh et al., 2011 and Tanko et al., 2008). At the termination of the

experiment, all rats used in the study were humanely euthanized using CO<sub>2</sub> and were disposed properly.

### **Data Analysis**

Analysis of data was performed using the SPSS statistical software. All the data were expressed as mean  $\pm$  standard deviation (SD). The obtained results were analyzed by analysis of variance (ANOVA) with multiple comparisons versus control group and  $p$ -Values  $<0.05$  were considered statistically significant.

## **RESULTS AND DISCUSSION**

### **Results**

After injection of alloxan intraperitoneally, the drug successfully elevated the fasting blood glucose level of the rats resulting to induction of diabetes. Alloxan-induced diabetic rats exhibited polyphagia and polydipsia associated with decrease in endogenous insulin and hyperglycemia. A baseline was established by measuring the fasting blood glucose level prior to administration of alloxan that served as the initial basis of the experiment. The effect of three different doses (1mL, 2.5mL and 5mL) of *Cucumis sativus* extract on blood glucose levels of alloxan-induced Sprague-Dawley rats were presented in Table 2. The hypoglycemic activity of *Cucumis sativus* was assessed by measuring the blood glucose of rats at 1<sup>st</sup>, 2<sup>nd</sup>, 3<sup>rd</sup>, 4<sup>th</sup>, and 5<sup>th</sup> hour after confirmation of the presence of diabetes to rats due to alloxan. As shown in Table 2, the blood glucose levels of diabetic rats increased dramatically after injection of alloxan. However, the results reveal that there is no statistically significant decrease in the blood glucose levels, nevertheless, it can be observed that glucose levels in blood is decreasing gradually especially on the reference drug, insulin, as indicated in Table 3.

**Table 2. Effect of *Cucumis sativus* extract on the Blood Glucose Levels of Alloxan-induced Diabetic Sprague-Dawley Rats**

Treatment	Blood Glucose Levels, mg/dL						
	Baseline	After Alloxan	1 <sup>st</sup> hour	2 <sup>nd</sup> hour	3 <sup>rd</sup> hour	4 <sup>th</sup> hour	5 <sup>th</sup> hour
<b>NSS (Negative Control)</b>	73.67*	418.67	535.20	529.00	527.80	520.00	534.40
<b>Insulin (Positive Control)</b>	82.50*	581.80	504.20	403.00	395.40	379.40	370.20
<b>1 mL <i>Cucumissativus</i> extracts</b>	82.83*	487.60	472.40	460.40	461.40	456.20	466.40
<b>2.5 mL <i>Cucumissativus</i> extracts</b>	78.67*	563.20	593.80	>600.00	>600.00	>600.00	>600.00
<b>5 mL <i>Cucumissativus</i> extracts</b>	87.67*	592.25	591.40	585.60	587.80	577.00	582.80

*The data represent the mean (n = 5).*

*Statistical Method: One way Analysis of Variance (ANOVA) followed by Dennett's multiple comparison test. When baseline is compared to after alloxan, values are statistically significant at \*P<0.005. When after alloxan is compared to 1<sup>st</sup>, 2<sup>nd</sup>, 3<sup>rd</sup>, 4<sup>th</sup>, and 5<sup>th</sup> hour, values are statistically not significant.*



**Table 3. Effect of the Oral Administration of Different Treatment on the Blood Glucose Level of Alloxan-induced Diabetic Rats**

Group	Treatment	Rat No.	BLOOD GLUCOSE LEVELS, mg/dL						
			Before Alloxan	After Alloxan	1st hour	2nd hour	3rd hour	4th hour	5th hour
1	NSS (Negative Control)	1	64	333	381	381	368	376	375
		2	101	529	554	563	583	569	>600
		3	64	349	541	501	488	455	497
		4	66	>600	>600	>600	>600	>600	>600
		5	64	>600	>600	>600	>600	>600	>600
2	Insulin (Positive Control)	1	98	>600	>600	>600	>600	>600	>600
		2	67	>600	>600	>600	>600	>600	>600
		3	69	>600	>600	>600	>600	>600	>600
		4	96	>600	564	146	140	62	32
		5	85	509	157	69	37	35	<20
3	1 mL Cucumber extract	1	77	>600	>600	>600	>600	>600	>600
		2	97	>600	>600	>600	>600	>600	>600
		3	74	239	236	217	209	208	208
		4	68	399	326	285	298	273	324
		5	88	>600	>600	>600	>600	>600	>600
4	2.5 mL Cucumber extract	1	81	>600	>600	>600	>600	>600	>600
		2	78	>600	>600	>600	>600	>600	>600
		3	81	>600	>600	>600	>600	>600	>600
		4	63	499	569	>600	>600	>600	>600
		5	93	517	>600	>600	>600	>600	>600
5	5 mL Cucumber extract	1	93	594	>600	>600	>600	>600	>600
		2	88	>600	>600	>600	>600	>600	>600

*Multiple comparative analyses between doses indicate no significant differences.*

The data demonstrate that treatment with *Cucumis sativus* extract to diabetic rats decreased elevated blood sugar level. The effect was more pronounced in case of the insulin (positive control) on rat number 5 (i.e., from >600mg/dL after alloxan injection to 32mg/dL on the 5<sup>th</sup> hour). While in the case of the negative control, values fluctuated (i.e., from 53mg/dL, values raised to 583mg/dL and decreased to 569mg/dL). Furthermore, there is a noticeable reduction of the blood glucose levels on the 3<sup>rd</sup> rat in Group 3, wherein the blood glucose levels nearly normalize. This implies that the effect of the extract was found to be effective against hyperglycemia. Besides, it is very remarkable that there are values that are outside the normal value. Extract formulation at dose 2.5mL did not show a proportional improvement in the fasting blood glucose as compared to 1mL suggesting that an increase concentration of the active ingredients is not always proportionally beneficial. This is with agreement to the studies on Wistar rats by Stanely, Prince & Menon (2000).

## **Discussion**

Several synthetic drugs have been industrialized for the treatment of diabetes. However, these drugs have limits in terms of efficacy and side effects. Therefore, there is much interest in discovering natural treatments without negative side effects in diabetic patients. Plant materials seem to be the solution.

Medicinal plants are widely used by the populations of underdeveloped countries as an alternative therapy. This study was therefore designed to investigate the hypoglycemic property of *Cucumis sativus* on alloxan-diabetic rats. Alloxan-induced hyperglycemia has been described as a suitable experimental model to study the activity of hypoglycemic agents because it selectively destroyed the pancreatic insulin secreting  $\beta$ -cells, leaving less active cell resulting in a diabetic state (Szkudelski, 2001).

Five groups consisting of five rats were used. The glycemc change in blood glucose levels of diabetic rats was measured at different time intervals after oral administration of *Cucumis sativus* extract at the doses of 1mL, 2.5mL, and 5mL as presented in Table 2. In all the groups of rats, it can be

seen that after induction to diabetes using alloxan, rats' blood glucose level tremendously increased due to the destruction of pancreatic cells and subsequent release of insulin. After measuring the blood glucose level, test samples were administered orally and blood glucose levels were then measured at 1<sup>st</sup>, 2<sup>nd</sup>, 3<sup>rd</sup>, 4<sup>th</sup>, and 5<sup>th</sup> hour. It is notable that blood glucose levels of all groups gradually decreases up to the 4<sup>th</sup> hour and then increased again at the 5<sup>th</sup> hour, insulin group as the exception which continually decreases up to the 5<sup>th</sup> hour. Hence, it can be assumed that the therapeutic effect of *Cucumis sativus* starts to diminish on the 4<sup>th</sup> hour that is why an additional dose may be given to increase the therapeutic level of the first dose; this diminished effect may be primarily due to the absorption and distribution processes that occur in response to oral administration of the drug followed by elimination. While the principal reason why insulin's effect is rapid is due to its route of administration which is intraperitoneal injection.

Furthermore, other important matters should be taken into consideration that may be the reason of the occurrence of the test results. Even though the result is the same and changing of the trend cannot be seen precisely, it does not mean that the results are not significant. The researchers postulate that the sensitivity and specificity of the instrument used in measuring blood glucose levels, which is the glucometer, directly or indirectly affect the test results. First, the glucometer currently available at the marketplace was designed to measure blood glucose levels ranging from 20 to 600 mg/dL and any value outside this range cannot be detected. The researchers believe that there are changes that in reality do occur but cannot be evaluated properly due to this reason. Rats may have a blood glucose level of 700 mg/dL, 660 mg/dL, 640 mg/dL, 620 mg/dL, and 605mg/dL on the 1st, 2nd, 3rd, 4th, and 5th hour respectively; considering the fact that it is outside the measuring capacity of the machine, these values cannot be measured properly, the same goes with the test results that occur on Insulin group wherein values are too low. Usually, glucometers are designed to measure human blood glucose levels ranging from 20 to 600 mg/dL, as previously mentioned, because any deviation outside this range is extremely alarming due to its effect on the body such as tachycardia, weakness, and dizziness for any values lower than 20 mg/dL and blurry vision, increased thirst, and need to urinate more often for any values higher than 600 mg/dL. Generally, blood glucose levels outside this range pose a life-threatening risk and abrupt medication is needed because this may lead to immediate death due to

hypoglycemic and/or hyperglycemic shock. At present, there is no device that is specifically used to measure blood glucose levels of rats.

The researchers also found out that the hypoglycemic property of *Cucumis sativus* is due to the presence of compounds like flavonoids such as Quercetin 3-L-rhamnoside, Quercetin 3-rutinoside, Feruloyl glucose, Saponarin 4'-O-glycoside and terpenes stimulate secretion or possess an insulin like-effect (Abu Reidah, 2013). They reduce hyperglycemia caused by alloxan in diabetic rats by proliferation of pancreatic  $\beta$ -cells leading to their and secretion of more insulin. The flavonoids present in *Cucumis sativus* may also be acting similarly thereby decreasing the high blood glucose levels of alloxan-diabetic rats. Moreover, the extract might insulin-like effect on peripheral tissues either by promoting glucose uptake and metabolism or inhibiting hepatic gluconeogenesis. Therefore, in the present investigation, substantial hypoglycemic activity was observed. Maximum reduction in glucose levels was observed in Group 3 at a dose of 1mL.

## CONCLUSIONS AND RECOMMENDATIONS

The ability of the *Cucumis sativus* extract to significantly decrease the blood glucose levels in diabetic rats support antidiabetic activity in rats. On the basis of this evidences obtained in laboratory animal study, the researchers concluded that the *Cucumis sativus* extract possesses hypoglycemic properties which suggests the presence of biologically active components such as terpenoids, alkaloids, flavonoids, and phenolics have shown antidiabetic potential through the insulinomimetic activity of the plant extract which may be worth further investigation and elucidation. Statistical analysis of the experimental data revealed that different volume of *Cucumis sativus* ethanolic extract is not significant at  $<0.05$  level of significance, however, data show that 1mL of the extract almost normalize the blood glucose level of rats. Overall, this research study presents *Cucumis sativus* extract as a new formulation for achieving an antidiabetic activity. Hence, it might help in preventing diabetic complications and may serve as a good alternative in the present armamentarium of antidiabetic drugs.

It is recommended that instruments that are species specific and can measure a wide range of blood glucose levels should be used, if there is any. Likewise, as much as possible, procure rats that are near to or resides at the testing area so that any rat related interferences may be prevented.

Furthermore, the experiment span or period should be increased. In this way, the longtime effect of the extract on the rats can be observed. Likewise, time extension might be needed to determine if the extract can be considered as safe supplementary therapy for a long-term and effective management of diabetic patients. Histopathological analysis of rat's liver and pancreas may be conducted to know the cytological effect of the extract. In addition, more parameters should be studied such as insulin enhancement, HbA<sub>1c</sub> level, and free radical production, to elucidate the mechanism of action of the active constituents of garlic.

Lastly, further studies in detail are warranted to explore its active ingredients responsible for the beneficial action and the mechanisms involved. Controlled clinical trials are also strongly needed to confirm the hypoglycemic effects in human subjects. These are our focus for future studies.

## REFERENCES

- Abu Reidah, Ibrahim M. (2013). HPLC—ESI-Q-TOF-MS for a comprehensive characterization of bioactive phenolic compounds in cucumber whole fruit extract. Retrieved January 10, 2015 from [https://www.researchgate.net/publication/216863705\\_HPLC-ESI-Q-TOFMS\\_for\\_a\\_comprehensive\\_characterization\\_of\\_bioactive\\_phenolic\\_compounds\\_in\\_cucumber\\_whole\\_fruit\\_extract](https://www.researchgate.net/publication/216863705_HPLC-ESI-Q-TOFMS_for_a_comprehensive_characterization_of_bioactive_phenolic_compounds_in_cucumber_whole_fruit_extract).
- Balaraman, A., Singh, J., Dash, S., &Maity, K. (2010). Antihyperglycemic and hypolipidemic effects of *Melothriamaderaspatana* and *Cocciniaindica* in streptozotocin induced diabetes in rats. *Saudi Pharmaceutical Journal*, 18, 173-178. Retrieved January 10, 2015 from [http://www.researchgate.net/profile/Ashok\\_Kumar\\_Balaraman/publication/n/233937455\\_sPJ\\_FINAL/links/0912f50d29\\_a942648d000000.pdf](http://www.researchgate.net/profile/Ashok_Kumar_Balaraman/publication/n/233937455_sPJ_FINAL/links/0912f50d29_a942648d000000.pdf).
- Bnouham, M., Ziyat, A., Mekhfi, H., Tahri, A., Legssyer, A. (2006). Medicinal plants with potential antidiabetic activity – a review of ten years of herbal medicine research (1990–2000). *International Journal Diabetes and Metabolism*, 14, 1–25. Retrieved January 10, 2015 from [http://ijod.uaeu.ac.ae/iss\\_1401/a.pdf?q=decreased](http://ijod.uaeu.ac.ae/iss_1401/a.pdf?q=decreased).
- Cortés-Ortiz, M., Leal-Galicia, P., Chávez-Álvarez, B.E., del Carmen Cárdenas- Aguayo, M., Meraz-Ríos, M.A. (2014). Effect of cholesterol enriched or fatty- acid diets on cholesterol and lipid levels in young

- wistar rats. *Advances in Bioscience and Biotechnology*, 5, 846-852. Retrieved January 10, 2015 from <http://www.scirp.org/journal/PaperDownload.aspx?paperID=50223>.
- Department of Health (DOH) of the Philippines. Leading cause of mortality. Retrieved January 10, 2015 from <http://www.doh.gov.ph/node/198.html>.
- Das, J., Chowdhury, A., Biswas, K.S., Karmakar, K.U., Sharif, R.S., Raihan, S.Z., et al. (2012). Cytotoxicity and antifungal activities of ethanolic and chloroform extracts of *Cucumissativus*Linn (Cucurbitaceae) leaves and stems. *Research Journal of Phytochemistry*. 6 (1), 25 – 30. Retrieved January 10, 2015 from <http://scialert.net/qredirect.php?doi=rjphyto.2012.25.30&linkid=pdf>.
- Ediriweera, E. &Ratnasooriya, W.D. (2009). A review on herbs used in treatment of diabetes mellitus by Sri Lankan ayurvedic and traditional physicians. *Ayu Journal*, 30 (4). Retrieved January 10, 2015 from [http://www.ayujournal.org/temp/Ayu304373-3312444\\_005512.pdf](http://www.ayujournal.org/temp/Ayu304373-3312444_005512.pdf).
- Eseyin, O., Ebong, P., Eyong, E., Awofisayo, O., &Agboke, A. (2010). Effect of *Telfairiaoccidentalis* on oral glucose tolerance in rats. *African Journal of Pharmacy and Pharmacology*, 4 (6). 368 – 372. Retrieved January 10, 2015 from [http://www.academicjournals.org/article/article1380889497\\_Eseyin%20et%20al.pdf](http://www.academicjournals.org/article/article1380889497_Eseyin%20et%20al.pdf).
- Gill, N.S., Bajwa, J., Dhiman, K., Sharma, P., Sood, S., Sharma, P.D., et al. (2011). Evaluation of therapeutic potential of traditionally consumed *Cucumismelo* seeds. *Asian Journal Plant Sci*. 10 (1), 89 – 91. Retrieved at January 10, 2015 from <http://docsdrive.com/pdfs/ansinet/ajps/2011/86-91.pdf>.
- Gill, N.S., Sharma, P., Bajwa, J., Dhiman, K., Sood, S., Sharma, P.D., & Bali M., (2010). Study on *Cucumismelo* var. *utilissimus* seeds for the therapeutic. *J. Plant Sci*. 5, 248 – 255. Retrieved January 10, 2015 from <http://scialert.net/abstract/?doi=jps.2010.248.255>.
- Hubert, M.F., Laroque, P., Gillet, J.P., & Keenan, K. (2000). The effects of diet, *ad libitum* feeding, and moderate and severe dietary restriction on body weight, survival, clinical pathology parameters, and cause of death in control Sprague-Dawley rats. *Toxicological Sciences*

58. 195 – 207. Retrieved January 10, 2015 from <http://www.ncbi.nlm.nih.gov/pubmed/11053556>.
- International Diabetes Federation (IDF), (2012). Diabetes Atlas News, 5th Edition of Diabetes Atlas. Retrieved January 10, 2015 from [https://www.idf.org/sites/default/files/21991\\_diabAtlas\\_5thEd.pdf](https://www.idf.org/sites/default/files/21991_diabAtlas_5thEd.pdf).
- Li, Y.G., Ji, D.F., Zhong, S., Lv, Z.Q., Lin, T.B., Chen, S., et al. (2011). Hybrid of 1-deoxynojirimycin and polysaccharide from mulberry leaves treat diabetes mellitus by activating PDX-1/insulin-1 signaling pathway and regulating the expression of glucokinase, phosphoenolpyruvatecarboxykinase and glucose-6-phosphatase in alloxan-induced diabetic mice. *J. Ethnopharmacol.* 134, 961 – 970. Retrieved at January 10, 2015 from <http://www.ncbi.nlm.nih.gov/pubmed/21333726>.
- Madhavan, V., Nagar, J.C., Murali, A., Mythreyi, R., Yoganarasimhan, S.N. (2008). Antihyperglycemic activity of alcohol and aqueous extracts of *Pandanufascicularis* Lam. roots in alloxan induced diabetic rats. *Pharmacologyonline*, 3, 529-536. Retrieved January 10, 2015 from [http://pharmacologyonline.silae.it/files/archives/2008/vol3/051\\_Madhavan.pdf](http://pharmacologyonline.silae.it/files/archives/2008/vol3/051_Madhavan.pdf).
- Mahendran, G., Manoj, M., Muruges, E., Sathish Kumar, R., Shanmughavel, P, Rajendra Prasad, K.J., et al. (2014). In vivo anti-diabetic, antioxidant and molecular docking studies of 1, 2, 8-trihydroxy-6-methoxy xanthone and 1, 2-dihydroxy-6-methoxyxanthone-8-O- $\beta$ -D-xylopyranosyl isolated from *Swertiacorymbosa*. *Phytomedicine*. 21, 1237–1248. Retrieved January 10, 2015 from [https://www.researchgate.net/profile/Ganesh\\_Mahendran/publication/264900156\\_In\\_vivo\\_antidiabetic\\_antioxidant\\_and\\_molecular\\_docking\\_studies\\_of\\_1\\_2\\_8-trihydroxy-6-methoxy\\_xanthone\\_and\\_1\\_2-dihydroxy-6-methoxyxanthone-8-O-Dxylopyranosyl\\_isolated\\_from\\_Swertia\\_corymbosa/links/54d256b30cf28e069723e64f.pdf](https://www.researchgate.net/profile/Ganesh_Mahendran/publication/264900156_In_vivo_antidiabetic_antioxidant_and_molecular_docking_studies_of_1_2_8-trihydroxy-6-methoxy_xanthone_and_1_2-dihydroxy-6-methoxyxanthone-8-O-Dxylopyranosyl_isolated_from_Swertia_corymbosa/links/54d256b30cf28e069723e64f.pdf)
- Marzouka, B., Marzoukb, Z., Halouib, E., Feninab, N., Bouraouic, A., & Aounia, M. (2010). Screening of analgesic and anti-inflammatory activities of *Citrulluscolocynthis* from Sounther Tunisia. *J. Ethnopharmacol.* 128, 15 – 19. Retrieved January 10, 2015 from <http://www.ncbi.nlm.nih.gov/pubmed/19962436>.
- May, L.D., Lefkowitz, J.H., Kram, M.T., Rubin, D.E. (2002). Mixed

- hepatocellular cholestatic liver injury after pioglitazone therapy. *Ann. Intern. Med.* 136, 449 – 452. Retrieved January 10, 2015 from <http://www.ncbi.nlm.nih.gov/pubmed/11900497>.
- Minaiyan, M., Zolfaghari, B., and Kamal, A. (2011). Effect of hydroalcoholic and buthanolic extract of *Cucumissativus* seeds on blood glucose level of normal and streptozotocin-induced diabetic rats. *Iran Journal Basic Med. Sci.* 14, 436 – 442. Retrieved January 10, 2015 from <http://www.ncbi.nlm.nih.gov/pmc/articles/PMC3586845/>.
- Ragavan, B., Krishnakumari, S. (2006). Antidiabetic effect of *T. arjuna* bark extract in alloxan induced diabetic rats. *Ind. J. Clin. Biochem.* 21, 123-128. Retrieved January 10, 2015 from <http://www.ncbi.nlm.nih.gov/pmc/articles/PMC3453977/>.
- Rajagopal, K., & Sasikala, K. (2008). Antihyperglycaemic and antihyperlipidaemic effects of *Nymphaea stellata* in alloxan-induced diabetic rats. *Singapore Med. J.*, 49: 137-141. Retrieved January 10, 2015 from <http://www.ncbi.nlm.nih.gov/pubmed/18301841>.
- Rohila, A. and Ali, S. Alloxan Induced Diabetes: Mechanisms and Effects (2012). *International Journal of Research in Pharmaceutical and Biomedical Sciences*. Vol. 3 (2), 819 – 823. Retrieved January 10, 2015 from <http://www.ijrbsonline.com/files/59-3290.pdf>.
- Saha, P., Mazumder, U.K., Haldar, P.K., KunduSen, S., Naskar, S. (2011). Antihyperglycemic activity of *Lagenariasiceraria* aerial parts on streptozotocin induced diabetes in rats. Retrieved January 10, 2015 from <http://www.idb.hr/diabetologia/11no2-3.pdf>.
- Sedigheh, A., Jamal, M.S., Mahbubeh, S., Somayeh, K., Mahmoud, R., Azadeh, A., et al. (2011). Hypoglycaemic and hypolipidemic effects of pumpkin (*Cucurbita pepo* L.) on alloxan-induced diabetic rats. *African Journal of Pharmacy and Pharmacology*. 5(23), 2620 – 2626. Retrieved January 10, 2015 from [http://www.academicjournals.org/article/article1380895192\\_Sedigheh%20et%20al%20%2012.pdf](http://www.academicjournals.org/article/article1380895192_Sedigheh%20et%20al%20%2012.pdf).
- Sharma, U., Sahu, R., Roy, A., Golwala, D. (2010). In vivo antidiabetic and antioxidant potential of *Stephania hernandifolia* in streptozotocin-induced diabetic rats. *J. Young Pharm.* 2, 255 – 260. Retrieved January 10, 2015
- Sharma, R. & Arya, V. (2011). A review on fruits having anti-diabetic potential.



- Journal of Chemical and Pharmaceutical Research*. 3 (2), 204 – 212. Retrieved January 10, 2015 from <http://jocpr.com/vol3-iss2-2011/JCPR-2011-3-2-204-212.pdf>.
- Stanely, P., Prince, M., & Menon, V.P. (2000). Hypoglycaemic and other related actions of *Tinosporacordifolia* roots in alloxan-induced diabetic rats. *Journal of Ethnopharmacology*. 70, 9 – 15. Retrieved January 10, 2015 from <http://www.ncbi.nlm.nih.gov/pubmed/10720784>.
- Sun, J.E., Ao, Z.H., Lu, Z.M., Xu, H.Y., Zhang, X.M., Dou, W.F., et al. (2008). Antihyperglycemic and antilipid peroxidative effects of dry matter of culture broth of *Inonotus obliquus* in submerged culture on normal and alloxan-diabetes mice. *J. Ethnopharmacol.* 118, 7 – 13. Retrieved January 10, 2015 from <http://www.ncbi.nlm.nih.gov/pubmed/18434051>.
- Szkudelski, T. (2001). The Mechanism of alloxan and streptozotocin action in B cells of the rat pancreas. *Physiol. Research*. 50, 536-546. Retrieved January 10, 2015 from [http://www.tigis.cz/images/stories/DMEV/2011/03/04\\_soltesova\\_Z.pdf](http://www.tigis.cz/images/stories/DMEV/2011/03/04_soltesova_Z.pdf).
- Tanko, Y., Yerima, M., Mahdi, M.A., Yaro, A.H., Musa, K.Y., and Mohammed A. (2008). Hypoglycemic activity of methanolic stem bark of *Adansonia digitata* extract on blood glucose levels of streptozocin-induced diabetic wistar rats. *International Journal of Applied Research in Natural Products*. Vol. 1(2), 32 – 36. Retrieved January 10, 2015 from <http://www.ijarnp.org/index.php/ijarnp/article/view/70/0>.
- Venkatesh, S., Reddy, B.M., Reddy, G.D., Mullangi, R., Lakshman, M. (2010). Antihyperglycemic and hypolipidemic effects of *Helicteresisora* roots in alloxan-induced diabetic rats: a possible mechanism of action. *J. Nat. Med.*, 64(3), 295-304. Retrieved January 10, 2015 from <http://www.ncbi.nlm.nih.gov/pubmed/20238178>.