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HYPOGLYCEMIC EFFECT OF PSIDIUM GUAJAVA AND AZADIRACHTA INDICA LEAVES POWDER EXTRACT IN ALLOXAN INDUCED MICE

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ABSTRACT

Traditional plant treatment for Diabetes has shown a surging treatment in the last few decades. This study focuses on herbal Anti Diabetic formulations of two different plants and also mixed group is used in the Treatment of Diabetes. The Efficacy of the herbal plant aqueous extract of Psidium guajava , Azadirachta indica and mixed group (equal amount of Psidium guajava and Azadirachta indica) has been tested on Alloxan induced Diabetic Mice. Three extract fed mice groups were compared to the diabetic mice, for the Hypoglycemic effect. The aqueous plant extract of Psidium guajava dose level of 200 mg/kg body weight , produced a significant decrease in the blood glucose level, when compared to the other two groups.

KEY WORDS: Diabetic mellitus, Plant Extract, Alternative medicine, Glucometer.

INTRODUCTION

Diabetic mellitus is an irreversible disease that is characterized by chronic hyperglycaemia caused by dysfunctional lipid, carbohydrate, and protein metabolism (Newman D.J., Cragg, *et al* 2002). The incidence of type 2 diabetes mellitus (T2DM) is more severe compared to that of type 1 diabetes mellitus (T1DM) (M.Kumar, *et al* 2021*)*. The T2DM prevalence is about 90%, affecting approximately 460 million people worldwide, with statistics predicting an increase to over 700 million in 25 years (A. Sharma, *et al* 2016*).* T1DM results from an autoimmune destruction of beta cells that produce insulin. It is one of the most prevalent paediatric illnesses, with an annual incidence rate of 2% to 5% (Amat-ur-Rasool. H, *et al* 2020). T2DM is associated with reduced glucose tolerance due to insulin resistance (Mannino.G, *et al* 2020). Many pharmaceutical medications are utilized to treat diabetes; however, plant-based remedies are often thought to be less toxic and devoid of adverse effects. However, a complex drug molecule causes lower drug absorption, limiting a medicine's bioavailability.

Plants have been used for medicinal purposes long before prehistoric period. Ancient Unani manuscripts Egyptian papyrus and Chinese writings described the use of herbs. Evidence exist that Unani Hakims, Indian Vadis and European and Mediterranean cultures were using herbs for over 4000 years as medicine. Medicinal plants contain phytoconstituents, some showing mild-to-potent antihypoglycaemic activity. Bioactive compounds isolated from plants may lower blood glucose levels by improving beta cell function, promoting glucose reabsorption, reducing insulin resistance, or regulating glucagon-like peptide-1 homeostasis (Unuofin JO,2020). Accessibility is one of the main reasons herbal medicines are widely used in the treatment of diabetes, especially in rural communities (Arya.V, *et al* 2011). Hence, the anti diabetic potential of a wide repertoire of medicinal plants has been widely reviewed in the last few decades (Salehi B,*etal* 2019). Medicinal plants are in high demand right now and their adaptation is expanding due to their higher availability and lower toxicity to humans (Lewicki S, *et al* 2014). Several diseases have been cured using a variety of plant preparations in folk medicine since ancient times and, presently, cosmetic, pharmaceutical, and nutraceutical industries are paying more attention to plant preparations and pure phytochemicals.

As a result, the present project aims to evaluate the antidiabetic efficacy of *Psidium guajava* and *Azadirachta Indica.*

MEDICINAL IMPORTANCE OF PSIDIUM GUAJAVA:

Psidium guajava (guava) is well known tropic tree which is abundantly grown for fruit. Many countries have a long history of using guava for medicinal purposes. This plant finds applications for the treatment of diarrhoea, dysentery, gastroenteritis, hypertension, diabetes, caries and pain relief and for improvement in locomotors coordination. Its leaf's extract is being used as a medicine in cough, diarrhoea, and oral ulcers and in some swollen gums wound. Its fruit is rich in vitamins A, C, iron, phosphorus and calcium and minerals.

MEDICINAL IMPORTANCE OF AZADIRACHTA INDICA


Whole plant is extensively used in Ayurvedic System Medicine for various skin disorders and diabetes. Nature has served this plant with various organic Compounds that are used as insecticides and pesticides. Neem is a Major herb with anti fungal, anti bacterial ,anti diabetic, anti-viral and anti-Helminthic Properties in Ayurveda.

MATERIALS AND METHOD:

EXPERIMENTAL ANIMALS:

This study used Male white albino mice aged (6-8 weeks) weighing 25 to 30 g. The mice were acclimatized with free access to food and water. Nine mice were randomly divided into 3 groups, each group contains 3 animals (n=3).

EXPERIMENTAL INDUCTION OF DIABETES:

The animals were acclimatized for one week, before inducing Alloxan .Diabetes was induced in the mice by oral administration of alloxan (120 mg/kg) body weight. Alloxan was first weighed individually for each animal, according to its weight, and solubilized with water. Blood glucose level was measured using a glucometer. Diabetic mice having blood glucose level above 100 mg/dL were selected for this study. All the animals (albino mice) used in this study developed diabetes mellitus after alloxan administration .

EXPERINMENTAL DESIGN:

The animals were divided into 3 groups (3 mice in each group)

Group I: Psidium guajava group

Alloxan (120 mg/kg body weight) and Psidium guajava leaf extract.

Group II: Azadirachta indica group

Alloxan (120 mg/kg body weight) and Azadirachta indica leaf extract.

Group III: Mixed group

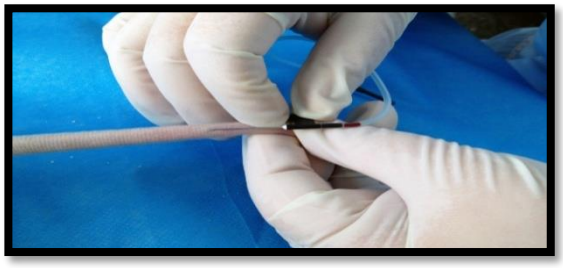
Alloxan (120 mg/kg body weight) and (Psidium gujava and Azadirachta indica) in equal amount.

S.NO	GROUPS	GROUP NAME	ALLOXAN DOSE (MG/KG)	PLANT EXTRACT DOSE (MG/KG)
1	Group 1	Psidium guajava	120 (mg/kg)	200 (mg/kg)
2	Group 2	Azadirachta indica	120 (mg/kg)	200(mg/kg)
3	Group 3	Mixed (Guajava and indica)	120 (mg/kg)	200(mg/kg) (Equal amount)

In each group (n=3), one animal kept as control mice (no alloxan and no treatment) free access for food pellets and water ,2nd animal is Diabetic group (Alloxan induced) without treatment and 3rd animal is extract fed group (alloxan induced and herbal treatment) will be given for 8 days.

COLLECTION OF BLOOD SAMPLE:

Blood sample was collected from the mice by tail vein puncture. In the un anesthetized animals, lateral or dorsal veins are dilated by rubbing with xylol and then cleaning the part with spirit. The tail was grasped between the thumb and index finger and a needle (25 to 27 gauge and 0.5-1 inch long) is introduced near the distal portion of tail with bevel up. The drop of blood accumulated at the tip of tail after puncture was collected at the tip of Glucometer strip and reading was noted down.



ESTIMATION OF BLOOD GLUCOSE LEVEL

On the 1st, 3rd, 6thand 10thdays of treatment, the blood glucose level was measured by the “Rupturing tail vein technique” using a one-touch glucometer.

RESULTS AND DISCUSSION:

The results of the hypoglycemic study conducted were recorded in the Tables (1-3)

TABLE NO: 1 PSIDIUM GUAJAVA GROUP

DAYS	Control mice (animal no:1)	Diabetic Mice (animal no: 2)	Extract fedmice (animal no :3)
1st day	72	87	70
3rd day	73	135	143
6th day	73	220	120
10th day	75	337	73

Units: Glucose values are expressed as mg/dl n=3 in each group. Comparison made between normal control diabetic and extract fed (treated)

At the first day, before induction of Alloxan, normal blood glucose levels were checked for all animals, then Alloxan (120 mg/kg body weight) is induced. After 48 hours of Alloxan induction, during the 3rd day again blood glucose levels were measured to check the increased blood glucose levels. From the third day itself herbal treatment started for the extract fed group Aqueous plant extract of *Psidium guajava* was given till the tenth day. Then blood glucose level was determined in the 6th and 10th day.

TABLE NO: 2 AZADIRACHTA INDICA GROUP

DAYS	Control group (animal no:1)	Diabetic Control Mice (animal no: 2)	Extract fed (animal no :3)
1st day	72	71	80
3rd day	74	150	165

6th day	**74**	**287**	**134**
10th day	**75**	**398**	**93**

Units: Glucose values are expressed as mg/dl n=3 in each group. Comparison made between normal control diabetic and extract fed (treated)

At the first day, before induction of Alloxan, normal blood glucose levels were checked for all animals ,then Alloxan (120 mg/kg body weight) is induced .After 48 hours of Alloxan induction ,during the 3rd day again blood glucose levels were measured to check the increased blood glucose levels. From the third day itself herbal treatment started for the extract fed group Aqueous plant extract of Azadirachta indica was given till the tenth day. Then blood glucose level was determined in the 6th and 10th day.

TABLE NO: 3 MIXED GROUP

DAYS	Control group (animal no:1)	Diabetic Control Mice (animal no: 2)	Extract fed (animal no :3)
1st day	**65**	**71**	**69**
3rd day	**65**	**174**	**183**
6th day	**68**	**220**	**159**
10th day	**69**	**376**	**98**

Units: Glucose values are expressed as mg/dl n=3 in each group. Comparison made between normal control diabetic and extract fed (treated)

At the first day, before induction of Alloxan , normal blood glucose levels were checked for all animals ,then Alloxan (120 mg/kg body weight) is induced .After 48 hours of Alloxan induction ,during the 3rd day again blood glucose levels were measured to check the increased blood glucose levels .From the third day itself herbal treatment started for the extract fed group Aqueous plant mixed extract of Psidium guajava and Azadirachta indica was given in equal amount till the tenth day. Then blood glucose level was determined in the 6th and 10th day.

DISCUSSION

The result obtained from the above experimental models clearly envisages that the leaf extract of the plant guajava and Azadirachta indica has enough potency to reduce the blood glucose level in mice. The results demonstrate that the extracts of guajava and Azadirachta indica leaves showed anti -hyperglycemic effect in diabetic model mice when the extracts were fed . In the Psidium guajava group, blood glucose level was steadily increased in the control group (C) at 3rd, 6th, 10thdays of intervals.

In (Ef) group the blood glucose level reduced consistently at 6th and 10 th days of intervals when compared to the control group. At the 10th day, the blood glucose level of Ef mice was recorded almost equal to the normal value. In diabetic mice, a drastic increase in the blood glucose level was recorded when compared to that of normal mice. In the Ef mice, the blood glucose level reduced consistently at 6thand 10th days intervals when compared that of control mice. Similar reduction in the blood glucose level was obtained in the mice fed with leaves of Azadirachta indica and mixed group.

Although hypoglycemic property of Guajava, Azadirachta indica leaves and both on normal and alloxan diabetic animals has been reported, most of these studies were conducted only on days basis. Sepha and bose treated diabetic mice with guajava and Azadirachta indica leaves powder. Brahmchari and Augusti concluded that leaves of herbal plants contained some alcohol soluble orally effective hypoglycemic principle. Tested the pulp of its leaves powder on normal mice, alloxan diabetic mice. Kedar and Chakrabarti tested guajava and Azadirachta indicaleaves powder on alloxan diabetic mice and reported that the blood sugar level and glucose tolerance test became normal within few days. (Sharma R.R *et al 2021*.)

One important finding of the present study is that the blood sugar level which once dropped to normal level after extract feeding did not increase again after discontinuation of extract feeding. This finding is in accordance with

the observation of Sepha and Bose on diabetic mice. It can be concluded that the effect of aqueous extract of guajava and Azadirachta indica appears to be curative rather than palliative.

In the present study, we have found that most of the biologically active substances were present in the aqueous extracts of guajava and Azadirachta indica seed. The possible mechanism by which leaves brings about a decrease in blood sugar level.

CONCLUSION:

Azadirachta indica and mixed group showed a little significant when compared to the Psidium guajava extract. Each extract was reported safe and were conducted separately by using Psidium guajava and Azadirachta indica. However the combined extract of Psidium guajava and Azadirachta indica has a moderate hypoglycemic effect, but the single extract of Psidium guajava had better hypoglycemic effect than the other two plants extract**.**

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