INJ INTERNATIONAL NEUROUROLOGY JOURNAL

Anti-Diabetic Activity of Bitter Gourd Seed and Lemon Peel in Streptozocin Induced Diabetic Rats

Ms Roshini Khan, Dr.Vinay Jain, Dr Saloni Jain
Department of Pharmacology, Shri Ram College of Pharmacy Banmore, Morena, Madhya Pradesh, India
Department of Pharmacognosy, Shri Ram College of Pharmacy Banmore, Morena, Madhya Pradesh, India
Department of Pharmacology, Shri Ram College of Pharmacy Banmore, Morena, Madhya Pradesh, India

Abstract

The most well-known endocrine problem, diabetes mellitus affects hundreds of millions of individuals worldwide and claims more than a million lives each year. This high death rate emphasises the necessity of conducting thorough research on anti-diabetic medications. This review investigates the anti-diabetic properties of lemon peel and bitter gourd seeds' phytochemical composition. According to studies, the plant may be useful in the management or treatment of diabetes mellitus since it includes various phytochemicals with hypoglycemic properties, such as bitter gourd seeds and lemon peel. The physiological and biochemical underpinnings of the anti-diabetic properties of lemon peel and bitter gourd seeds are also discussed. Bitter gourd seeds and lemon peels have anti-diabetic properties that manifest through the inhibition of NF-κβ and MAPKs in pancreatic cells, which in turn promotes the breakdown of fats and carbohydrates, increases the absorption of fats, produces insulin, reduces insulin resistance, activates the AMPK pathway, and inhibits the enzymes involved in glucose metabolism.

KEYWORDS: Streptozocin, extraction, radicals, alkaloids, insulin, wistar rat, M. charantia.

Introduction

The first known mention of diabetes symptoms was in 1552 B.C., when Hesy-Ra, an Egyptian physician, documented frequent urination as a symptom of a mysterious disease that also caused emaciation. Also, around this time, ancient healers noted that ants seemed to be attracted to the urine of people who had this disease. Diabetes Mellitus is a complex chronic disease which has been associated with high amount of blood glucose level or hyperglycemia that occurs from the deficiency of insulin secretion, action, or both of these conditions. Insulin is a hormone created in the pancreas that helps transport (glucose) from the circulatory system in to the cells so they can separate it and use it for fuel. The chronic metabolic imbalance associated with the disease put patient at high risk of micro and macrovascular complications, which if not treated properly, leads to frequent urination, hospitalization, including the risk for cardiovascular disease. The clinical diagnosis of diabetes is reliant on either one of the four-plasma glucose (PG) criteria:

- Fasting plasma glucose (FPG) (>126mg/dl),
- Random PG (200 mg/dl) with classic signs and symptoms of hyperglycemia, or
- Hemoglobin level.6.5%.
- 2 h PG during a 75-g oral glucose tolerance test (OGTT) (>200 mg/dl).

Type 2 diabetes mellitus (T2DM) can occur with other medical conditions such as gestational diabetes occurring during the 2nd or 3rd trimester of pregnancy or pancreatic diseases associated with cystic fibrosis. T2DM can also be induced, by use of glucocorticoids or use of highly active antiretroviral agents like protease inhibitor in HIV positive patients.

T2DM is a common and increasing disease and a major public health concern across the world. The diabetes federation (international) estimates that there are approximately 387 million people diagnosed with diabetes across the world. Diabetes can be reduced if the standard care as well as patient compliance and participation is implemented. Patient with T2DM may develop the complications of diabetes ketoacidosis (DKA)

Etiological characterization of diabetes mellitus

TYPES	CAUSES
Type-I (Insulin Dependent Diabetes Mellitus) A. Autoimmune Idiopathic	Caused by damaging injury of pancreatic β cell, typically prompting outright insulin inadequacy.

Type-II (Non-Insulin Dependent Diabetes Mellitus)	Insulin secretion is defective in these patients and insufficient to compensate for insulin resistance.
Type-III (Due to other explicit system or pancreatic ailment). A. Those in which explicit transformations have been distinguished as reason for hereditary weakness. B. Those related with different maladies or conditions norm of pancreatic β-cell work	• Genetic variation • Disease of exocrine pancreas. • Endocrine sickness. • Infections. • Various hereditary disorders. Liver sickness
Type IV Gestational diabetes mellitus	Glucose bigotry

Signs and Symptoms of Type-2 Diabetes Milletus

- Frequent urination (Frequent bed-wetting in children who have been toilet trained)
- Excessive Thirst
- Excessive Hunger
- Weakness and fatigue
- Drowsiness
- Irritability
- Blurred vision or any change in sight
- Tingling or numbness in legs, feet or fingers
- Slow healing of cuts
- Frequent skin or vaginal infections or itchy skin

Types of tests required for diagnosis of DM

Types of tests	Normal (mg/dL)	Prediabetes (mg/dL)	Diabetes (mg/dL)
Fasting glucose test	Less than 100	100-125	126 or higher
Random glucose test	Less than 140	140-199	200 or higher
A1c test	Less than 5.7%	5.7-6.4%	6.5% or higher
Oral glucose tolerance test (OGTT]	Less than 140	140-199	200 or higher

Diabetes mellitus is among the most common disorder in developed and developing countries, and the disease is increasing rapidly in most parts of the world. It has been estimated that up to one-third of patients with diabetes mellitus use some form of complementary and alternative medicine. One plant that has received the most attention for its anti-diabetic properties is bitter melon, Momordica charantia (M. charantia), commonly referred to as bitter gourd, karela and balsam pear. Its fruit is also used for the treatment of diabetes and related conditions amongst the indigenous populations of Asia, South America, India and East Africa. Abundant pre-clinical studies have documented in the anti-diabetic and hypoglycaemic effects of M. charantia through various postulated mechanisms.

Bitter gourds require a very hot and humid climate to grow well. Plant bitter gourd seeds in a location that gets 6 to 8 hours of sunlight every day. Seeds will take 8 to 10 days to germinate in soil temperatures ranging from 15 to 20°C.17

ACTIVE INGREDIENT IN BITTER GOURD:

Main constituents in bitter gourd which have antidiabetic effects-

- ❖ Triterpene
- ❖ Proteid
- ❖ Steroid
- ❖ Lipid
- ❖ Phenolic Compound
- ❖ Polypeptide-P
- ❖ Charantin (Steroidal Glycoside)
- ❖ Vicine

- ➢ Mainly we used these two ingredients Polypeptide-P, and Charantin.

POLYPEPTIDE-P:

- ➢ A hypoglycemic peptide, Polypeptide-p, has been isolated from fruit, seeds, and tissue of Momordica charantia Linn (bitter gourd). Amino acid analysis indicates a minimum molecular weight of approximately 11,000 (166 residues). Polypeptide-p is a very effective hypoglycemic agent when administered subcutaneously to gerbils, langurs, and humans.

CHARANTIN:

- ➢ Charantin is a chemical substance obtained from the Asian bitter melon (Momordica charantia), reputed to be responsible for the hypoglycaemic properties of those plants. It was identified by Lolitkar and Rao in 1960.
- ➢ Charantin, a natural cucurbitane type triterpenoid, has been reported to have beneficial pharmacological functions such as anticancer, antidiabetic, and antibacterial activities.

Taxonomical classification of Momordica charantia

Kingdom	Plantae
Clade	Rosids
Order	Cucurbitales
Family	Cucurbitaceae
Genus	Momordica
Species	M. Charantia
Binomial Name	Momordica Charantia

3.1.7 MEDICINAL USE:

Its fruits and pulp are used in treating-

- Asthma,
- Constipation,
- Colic,
- Diabetes,
- Cough,
- Fever (Malaria),
- Gout,
- Helminthiases,
- Leprosy,
- Inflammation,
- Skin Diseases,
- Ulcer, And
- Wound.

Lemon is a flowering plant of the rutaceae family. Approximately 140 genes and 1300 species are present in the genus citrus The lemon has a number of different species and has Lemon is traditionally used as an additive to soothe the sore throats of our foods. The lemon's alkaloid features are also popular. Cross extracts from several citrus parts have shown anti-cancer and antibacterial features against bacterial strains of clinically significant importance. It is mainly used to decrease blood pressure, mental health, respiratory problems, arthritis and rheumatism. the kidney stones are prevented. The origins of the Lemon are not known although it is believed that the first lemons were cultivated in Assam (in north-eastern India), northern Burma or China. Lemons are mostly used for their juice, peel and oil. The oil is present in the peel, juice sacs and seeds and is used as flavouring agents in drinks, food, pharmaceutical and other produces. The peel is used in the production of brined peel, pectin and flavonoids.

There were distinct elements to the essential oil of the leaves and peel of the citrus limon. In both essential oils, limonene is the primary element, leaf oil was recognized with β-pine, myrcene, neral, geranial, neryl acetate, geranyl and β-caryophyllene. Peel oil had μ-terpinine, β-pinene, myrcene. In lemon, there are certain flavonoids, like hesperidoside, limocitrine, in Spanish lemon pericarp. Lemon flowers contain Citric acid, ascorbic acid, and caffeic acid. Caffeine is present in both, flowers and lemon tree leaves. Lemons contain quantities of distinct chemicals are supposed to be of some health advantages. Lemons are extremely important for human health and are high in vitamin C (ascorbic acid). A citrus juice of 100 millilitres includes about 50 milligram vitamin C and about five grams of citric acid. (55% of the daily value advised). It also includes Na, K, Ca, Cu, Fe, Mg, Zn and P. Mineral products Iron, copper, zinc and manganese are vital for the nutritional sector and are commonly used in health, environmental science and health. K was at its highest (8600±0.028 mg/100 g) concentration. Ca is concentrated 8452.5±0.050 mg/100 g in lemon peel samples. The bone formation is Ca accountable. Ca controls several cell procedures and plays significant structural functions in living organisms.

Taxonomical classification of citrus limon [lemon]

Kingdom	Plantae
Division	Magnoliophyta
Class	Magnoliopsida
Subclass	Rosidae
Family	Rutaceae
Order	Sapindales
Genus	Citrus
Subgenus	Amygdalus
Species	Limon

Materal And Methods

Streptozocin drug was obtained from Mittal Chemical Industries, Agra, U.P.
Selection and collection: The bitter gourd and lemons were collected from the local vegetable market of Dholpur, Rajasthan.

Drying: Herbal products to remove the water/moisture content from it, by natural process i.e., under shed of sunlight.

Authentication: Identified and Authentication by Dr. Rishi Saraswat, Department of Pharmacy and Agriculture, University of Technology, Jaipur (303903) Rajasthan India and Date of Authentication 22/09/2023.

250g of each of bitter gourd seeds and lemon peels were dried, ground into a coarse powder to increase surface area, and extracted with 60–65% ethanol using various Soxhlet extractors. The Soxhlet extraction is widely utilised in the extraction of plant metabolites due to its perceived ease of usage. With this procedure, almost all initial and bulk extraction can be completed. The primary benefit of the Soxhlet apparatus, commonly referred to as the hot continuous extraction technique, is full extraction with the least amount of solvent needed. A round bottom flask, syphon tube, distillation channel, condenser, cooling water intake, cooling water exit, heat source, and thimble make up the Soxhlet extractor system. This procedure involves drying, powdering, and grinding the sample into tiny particles before placing it in a porous bag or "thimble" made of sturdy filter paper and placing it in the Soxhlet apparatus's thimble chamber. Using a heating source such as a heating mantle, the extraction solvent is placed in a round-bottom flask and heated. The solvent in the bottom flask vaporises in the condenser due to the heat, causing the sample thimble to drip back. The extraction chamber is made to overflow and drip

ISSN:2093-4777 | E-ISSN:2093-6931
DOI: 10.5123/inj.2024.1.inj56
Vol. 28 Iss. 1 (2024)

back down into the boiling flask when the solvent around the sample reaches a predetermined level. The procedure is shown by the clear solution in the syphon tube. When the liquid content reaches the syphon arm, the liquid contents are dumped into the bottom flask once more. The procedure ought to take roughly sixteen hours to complete. After that, bitter gourd seeds and extract from lemon peel were compressed until they were dry, resulting in a solid that was both dark green and brown. The yield for bitter gourd is 17.7 g (7.08%) and lemon is 21.4 g (8.56%). Next, the extracts were ground into a powder at lower pressure. This system's benefit is that just one batch of solvent is recycled, as opposed to several portions of warm solvent being passed through the sample. Since prolonged heating may cause compound degradation, this approach cannot be applied to thermolabile compounds.

Wistar rats of either sex (200-250 g) were obtained from the central animals of Shriram College of Pharmacy, Banmore and were maintained in polypropylene cages on rodent pellet condition of controlled temperature (22±2°C) and acclimatized to 12/12 h light/dark cycle. Free access to food and water were allowed until 2 hours before the experiment. The care and maintenance of the animals were as per approved guidelines of the "Committee for the purpose of control and supervision of experiment on animals (CPCSEA)". Food and water were provided 2 hours after the experiment. All experiment on animals were conducted according to the guidelines of establishment's ethical committee on animal experimentation, the project proposal no. SRCP/IAEC/70/20-21

Induction of Diabetes

After fasting for 12 h, Diabetes was induced with a single dose of Streptozocin (STZ) (60mg /kg body wt., i.p) 2% solution dissolved in 0.9% Nacl. After the injection, they were free access to feed and water glucose solution to counter the hypoglycaemic shock. The development of diabetes was confirmed after 72 hours. The rats having fasting blood glucose level more than 200mg/dl have been selected for the experimentation.

Drug Treatment

Different drug treatment was given to different groups of rats after the induction of diabetes. The treatment includes:

i. Bitter gourd seed extract+ Lemon peel extract (200 mg/kg and 400 mg/kg body wt.; orally)
ii. Glibenclamide (0.5 mg/kg body wt.; peritoneal cavity)

7.9. Grouping of Animals

The animals were divided into 5 groups ,6 in each, total no. of animals =30
Group I: Normal control (vehicle only i.e., 0.9 % NaCl).
Group II: Diabetes control (STZ induced 60mg/kg; i.p).
Group III: Diabetes induced+ standard drug (Glibenclamide 0.5mg/kg; peritoneal cavity)
Group IV: Diabetes induced+ Bitter gourd+ Lemon extracts (200mg/kg; p.o)
Group V: Diabetes induced+ Bitter gourd+ Lemon extracts (400mg/kg; p.o)

Preparation of doses

The extracts dissolved or suspended in distilled water; its pH brought to 7.0.
Administration of doses
Herbal extracts are administered in a single by gavages using stomach tube.

Evaluation Parameters

i. BLOOD GLUCOSE

The treatment was started from the same day except normal control and diabetic control groups for a period of 21 days orally. During this period, animals in groups had free access to standard diet and water. Blood glucose levels were estimated on 0,3rd,7th,14th and 21st day of the treatment. Blood withdrawn from the tail vein and glucose levels were estimated using glucometer strip and glucometer. (Jyoti scientific laboratories).

ii. BODY WEIGHT

The body weight of rats was recorded on 0,3rd,7th,14th and 21st day of the treatment with the help of electronic balance.

Statistical Analyses

The data obtained from the different studies and the biochemical estimation is expressed as mean± SEM for each group. After this, the statistical analysis was carried out using was carried out using one way analysis of variance (ANOVA) followed by Dennett's test using SPSS 16.0 window version. Values $p > 0.05$ were considered non-significant, $p<0.05$ as significant.

Results And Observations

RESULTS

Table: Phytochemical Testing of Herbal Extracts.

S. NO.	Test	Extract (ethanol)	
		BITTER GOURD (SEED)	LEMON (PEEL)
1.	**Alkaloids**		
	Mayer's test	+	+
	Dragendroff's test	+	+
2.	**Carbohydrates**		
	Benedict's test	+	_
	Fehling's test	+	+
3.	**Proteins**		
	Biuret test	+	+
	Millon's test	+	+
4.	**Amino acids**		
	Ninhydrin test	+	+
	Tyrosine test	+	_
5.	**Glycosides**		
	Borntrager's test	+	+
6.	**Flavonoids**		
	Lead acetate test	+	+
7.	**Phytosterols**		
	Salkowski test	+	+
8.	**Fats and oils**		
	Solubility test	-	_
	Stain test	+	_
9.	**Phenolics and tannins**		
	Acetic acid test	+	+
10.	**Volatile oils**		
	Solubility test	-	-

(+) Indicates positive result, (–) Indicates negative result.

Table: Effect of ethanolic extract of bitter gourd seeds and lemon peel in different animal groups on blood glucose level.

S.NO.	Groups	0 day	After 3days (mg/dL)	After	After	After
	Treatment/ Dose	(mg/dL)		7days (mg/dL)	14days (mg/dL)	21days (mg/dL)

DOI: 10.5123/inj.2024.1.inj56
ISSN:2093-4777 | E-ISSN:2093-6931
Vol. 28 Iss. 1 (2024)

I	Normal control	96.32±	95.71±	96.08±	96.54±	98.92±
		1.09	0.97	1.31	1.25	0.98
II	Diabetic control	264.18±	281.06±	271.12±	219.37±	209.14±
	(STZ induced 60mg/kg)	2.67	1.97	2.55	2.31	1.98
III	Standard drug	256.13±	200.12±	152.43±	118.74± 2.88	98.16±
	(Glibenclamide 0.5 mg/kg)	3.28	3.02	2.96		3.07
IV	Bitter gourd + Lemon	263.16±	247.74±	208.17±	176.51±	130.36±
	(200mg/kg)	2.89	2.25	3.09	3.25	2.95
V	Bitter gourd + Lemon	261.49±	221.12±	160.75±	130.50± 2.87	110.69± 3.08
	(400mg/kg)	3.85	3.56	2.61		

Streptozocin (60 mg/kg) was administered; i.p in sterile saline, single dose 7 days before the administration of different ethanolic extracts.

Standard drug Glibenclamide (0.5mg/kg body wt. by peritoneal cavity) once a day

Ethanolic extracts of bitter gourd and lemon were administered orally once a day, in a single dose daily seven days after confirmation of Hyperglycaemia.

N=6 (no. of animals in each group)

Statically significance test was done by one way ANOVA followed by Dunnett's test using SPSS 16.0 window version.

*p<0.05 compared to disease control group.

Observations

DOI: 10.5123/inj.2024.1.inj56
ISSN:2093-4777 | E-ISSN:2093-6931
Vol. 28 Iss. 1 (2024)

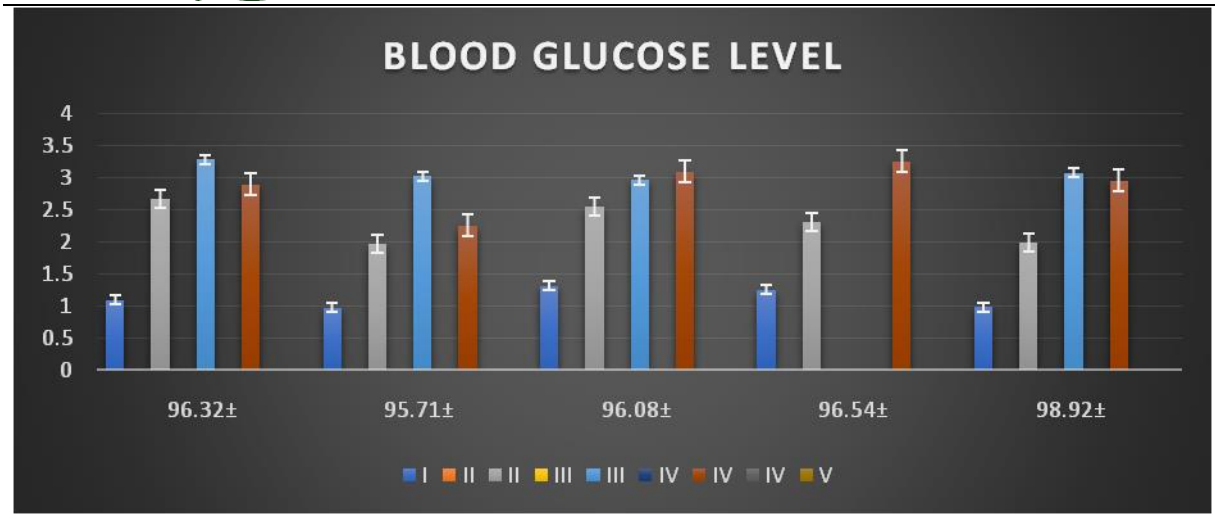

Figure: Effect of ethanolic extract of bitter gourd seeds and lemon peel in different animal groups on blood glucose level.

Statically significance test was done by one way ANOVA followed by Dunnett's test.

*p<0.05 compared to disease control group. Control value for Body weight -228.86±2.76 All values are MEAN± SEM of 6 animals per group

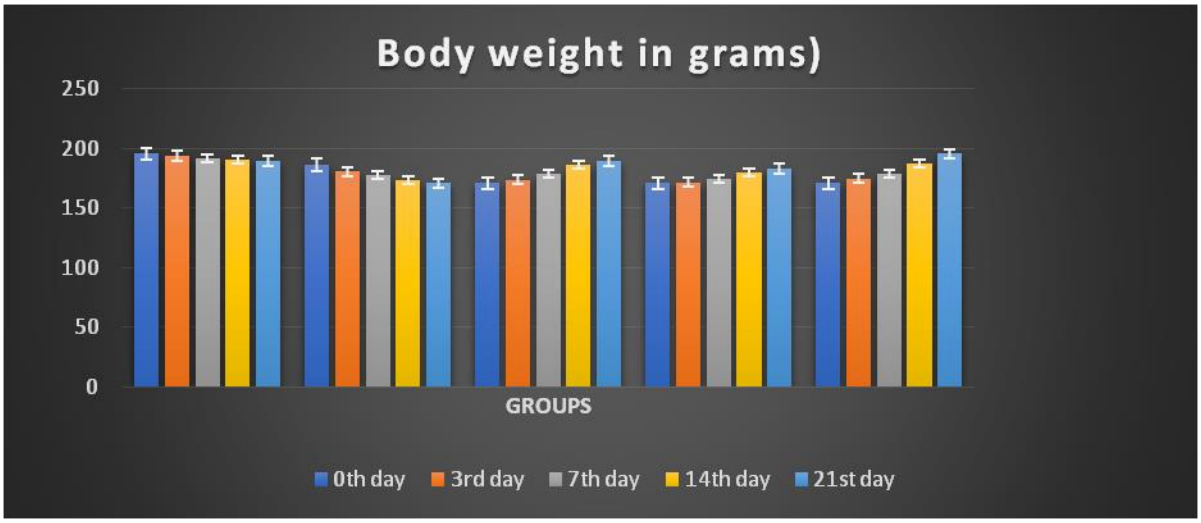

Figure: Effect of ethanolic extract of bitter gourd seeds and lemon peel in different animal groups on body weight.

Statically significance test was done by one way ANOVA followed by Dunnett's test *p<0.05 compared to disease control group

Conclusion

This study assessed the anti-diabetic properties of extracts from lemon peels and almond seeds. Numerous studies came to the conclusion that both herbal medications are effective in treating diabetes. In order to investigate the combined effects of both herbal medication extractions on type-II Diabetes milletus, we took combination dosages and compared the outcomes with a standard group. Our findings demonstrated the exceptional effectiveness of this test chemical combination against diabetes and its related high blood glucose adverse effects. Additionally, the animals' body weight grew during the course of their treatment with the test substances.

References

1. Singh B, Singh JP, Kaur A, Singh N. Phenolic composition, antioxidant potential and health benefits of citrus peel. Food Research International. 2020 Jun 1;132:109114.
2. Liu N, Li X, Zhao P, Zhang X, Qiao O, Huang L, Guo L, Gao W. A review of chemical constituents and health-promoting effects of citrus peels. Food Chemistry. 2021 Dec 15;365:130585.
3. Naim M, Amjad FM, Sultana S, Islam SN, Hossain MA, Begum R, Rashid MA, Amran MS. Comparative study of antidiabetic activity of hexane-extract of lemon peel (Limon citrus) and glimepiride in alloxan-induced diabetic rats. Bangladesh Pharm. J. 2012 Nov 12;15(2):131-4.
4. Adeleke AA, Zamisa SJ, Islam MS, Olofinsan K, Salau VF, Mocktar C, Omondi B. Quinoline functionalized schiff base silver (I) complexes: interactions with biomolecules and in vitro cytotoxicity, antioxidant and antimicrobial activities. Molecules. 2021 Feb 24;26(5):1205.
5. Oboh G, Ademosun AO. Shaddock peels (Citrus maxima) phenolic extracts inhibit α-amylase, α-glucosidase and angiotensin I-converting enzyme activities: A nutraceutical approach to diabetes management. Diabetes & Metabolic Syndrome: Clinical Research & Reviews. 2011 Jul 1;5(3):148-52.
6. Lim SM, Goh YM, Mohtarrudin N, Loh SP. Germinated brown rice ameliorates obesity in high-fat diet induced obese rats. BMC complementary and alternative medicine. 2016 Dec;16(1):1-1.
7. Parmar HS, Kar A. Antidiabetic potential of Citrus sinensis and Punica granatum peel extracts in alloxan treated male mice. Biofactors. 2007 Jan 1;31(1):17-24.
8. Alu'datt MH, Rababah T, Alhamad MN, Al-Mahasneh MA, Almajwal A, Gammoh S, Ereifej K, Johargy A, Alli I. A review of phenolic compounds in oil-bearing plants: Distribution, identification and occurrence of phenolic compounds. Food chemistry. 2017 Mar 1;218:99-106.
9. Akhila S, Bindu AR, Bindu K, Aleykutty NA. Comparative evaluation of extracts of Citrus limon burm peel for antioxidant activity. Journal of Young Pharmacists. 2009;1(2):136-40.
10. Miyake Y, Yamamoto K, Tsujihara N, Osawa T. Protective effects of lemon flavonoids on oxidative stress in diabetic rats. Lipids. 1998 Jul;33(7):689-95.
11. KunduSen S, Gupta M, Mazumder UK, Haldar PK, Saha P, Bala A. Antitumor activity of Citrus maxima (Burm.) Merr. leaves in ehrlich's ascites carcinoma cell-treated mice. International Scholarly Research Notices. 2011;2011.
12. Loizzo MR, Pugliese A, Bonesi M, Tenuta MC, Menichini F, Xiao J, Tundis R. Edible flowers: A rich source of phytochemicals with antioxidant and hypoglycemic properties. Journal of agricultural and food chemistry. 2016 Mar 30;64(12):2467-74.
13. Ernawita, Wahyuono RA, Hesse J, Hipler UC, Elsner P, Boehm V. In vitro lipophilic antioxidant capacity, antidiabetic and antibacterial activity of citrus fruits extracts from Aceh, Indonesia. Antioxidants. 2017 Feb 3;6(1):11.
14. Rafique S, Hassan SM, Mughal SS, Hassan SK, Shabbir N, Perveiz S, Mushtaq M, Farman M. Biological attributes of lemon: A review. Journal of Addiction Medicine and Therapeutic Science. 2020 May 22;6(1):030-4.
15. Albano F, Vecchio E, Renna M, Iaccino E, Mimmi S, Caiazza C, Arcucci A, Avagliano A, Pagliara V, Donato G, Palmieri C. Insights into thymus development and viral thymic infections. Viruses. 2019 Sep 9;11(9):836.
16. Dixit Y, Kar A. Protective role of three vegetable peels in alloxan induced diabetes mellitus in male mice. Plant Foods for Human Nutrition. 2010 Sep;65:284-9.
17. Choi GN, Kim JH, Kwak JH, Jeong CH, Jeong HR, Lee U, Heo HJ. Effect of quercetin on learning and memory performance in ICR mice under neurotoxic trimethyltin exposure. Food Chemistry. 2012 May 15;132(2):1019-24.
18. Murhekar MV, Bhatnagar T, Thangaraj JW, Saravanakumar V, Kumar MS, Selvaraju S, Rade K, Kumar CG, Sabarinathan R, Turuk A, Asthana S. SARS-CoV-2 seroprevalence among the general population and healthcare workers in India, December 2020–January 2021. International Journal of Infectious Diseases. 2021 Jul 1;108:145-55.
19. Hotez PJ, Bottazzi ME, Strych U, Chang LY, Lim YA, Goodenow MM, AbuBakar S. Neglected tropical diseases among the Association of Southeast Asian Nations (ASEAN): overview and update. PLoS neglected tropical diseases. 2015 Apr 16;9(4):e0003575.
20. Oyelere SF, Ajayi OH, Ayoade TE, Pereira GB, Owoyemi BC, Ilesanmi AO, Akinyemi OA. A detailed review on the phytochemical profiles and anti-diabetic mechanisms of Momordica charantia. Heliyon. 2022 Apr 1;8(4).
21. Salehi B, Ezzat SM, Fokou PV, Albayrak S, Vlaisavljevic S, Sharifi-Rad M, Bhatt ID, Sharifi-Rad M, Belwal T, Ayatollahi SA, Kobarfard F. Athyrium plants-review on phytopharmacy properties. Journal of traditional and complementary medicine. 2019 Jul 1;9(3):201-5.

22. Joseph B, Jini D. Antidiabetic effects of Momordica charantia (bitter melon) and its medicinal potency. Asian pacific journal of tropical disease. 2013 Apr 1;3(2):93-102.
23. Fuangchan A, Sonthisombat P, Seubnukarn T, Chanouan R, Chotchaisuwat P, Sirigulsatien V, Ingkaninan K, Plianbangchang P, Haines ST. Hypoglycemic effect of bitter melon compared with metformin in newly diagnosed type 2 diabetes patients. Journal of ethnopharmacology. 2011 Mar 24;134(2):422-8.
24. Ooi CP, Yassin Z, Hamid TA. Momordica charantia for type 2 diabetes mellitus. Cochrane database of systematic reviews. 2012(8).