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Formulation of Anti-Diabetic Polyherbal Suspension

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ABSTRACT:
Diabetes mellitus is a chronic disorder characterized by hyperglycemia and altered metabolism of carbohydrates, lipids and proteins, [5]. It is one of the common metabolic disorders affecting around 2.8% of the world's population and is anticipated to exceed 5.4% by the year 2025, [5]. Hebal medicines have long been esteemed as a source of medicine; therefore , they have become an integral part ofmodern , high-tech medicine. In view of these aspects, the present study was focusedmainly on the preparation of polyherbal antidiabetic suspension. Herbal medicine is the oldest form of healthcare known to humanity.We make herbal antidiabetic suspension by using the extract of Garlic (Allium sativum), Neem (Azadirachta indica), Cinnamon (Cinnamomum zeylanicum) and Fenugreek (Trigonella foenum-graecum). All this herbs show antidiabetic activity. Herbs had been used by all culture all over history.The goal of this study was to assess and create a herbal antidiabetic suspension made up of a variety of medicinal plants thought to possess antidiabetic characteristics.

KEYWORDS: Herbal suspension, Polyherbal preparation, Anti-diabetic activity, Allium sativum, Azadirachta indica, Cinnamomum zeylanicum, Trigonella foenum-graecum.

I. INTRODUCTION:

Diabetes Mellitus is a metabolic disorder associated by impairment in the metabolism of carbohydrate, fat and proteins which was recognized by insufficient insulin secretion or rising resistance to its action. It is one of the most common endocrine metabolic disorder which has caused significant morbidity and mortality due to microvascular and macrovascular complications. According to International Diabetes Federation (IDF) estimates, 80% of the world diabetic population will be from low to middle income countries in 2030, [1]. As per IDF 2011 report, China, India, and the United States of America have a diabetic population of 90.0, 61.3, and 23.7 million, which may be increased up to 129.7, 101.2, and 29.3 million respectively, by the year 2030, [1]. Herbal medicines are treated as traditional medicines since they were extensively used in the traditional system of medicine like Ayurveda, Siddha, Unani. Herbal medicines have no side effects or less side effects. So the polyherbal preparation is developed. In this polyherbal preparation, we used herbs which show anti-diabetic activity. Such as Azadirachta indica, Allium sativum, Cinnamomum zeylanicum, and Trigonella foenum-graceum. These all are used in combination to gain maximum benefit from their combined potential to reduce the side effects of one another.

Objectives
- To reduce the symptoms of hyperglycemia.
- To reduce the risk of long term complications of diabetes.
- To highlight the benefits & key ingredients of polyherbal anti-diabetic suspension.
- To demonstrate the proper usage and its effectiveness in diabetes control.
- Polyherbal formulations enhance the therapeutic action & reduce the concentrations of single herbs, thereby reducing adverse events.

Causes
- Obesity
- Smoking
- Genetic factor
- Stress
- High blood pressure
- Unhealthy food
- Autoimmune diseases
- Hormonal imbalances

Signs& Symptoms
- Polyuria (increasedurination)
- Polydipsia (increasedthirst)
- Polyphagia (increasedhunger)
- Losing weight without trying
- Blurred vision
- Fatigue

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- Getting a lot of infections
- Slow healing of wounds
- Feeling tired & weak
- Itchy skin

Types: The new classification proposed by WHO in 1980 and 1985 were identified four types ofdiabetes mellitus.

1. Type 1: Insulin dependent diabetes mellitus
2. Type 2: Non-insulin dependent diabetes mellitus
3. Type 3: Maturity Onset Diabetes of the Young (MODY)
4. Type 4: Gestational Diabetes Mellitus (GDM)

Herbs

Herbs include crude plant materials such as leaves, flowers, fruits, seed, stem, wood, bark, roots, rhizomes or other plant parts, which may be entire fragmented or powdered, [5].

Herbal preparations

Herbal preparations are the basis for finished herbal products and may include communicated or powdered herbal materials, or extracts, tinctures and fatty oils of herbal materials, [5].

Herbal dosage forms

Herbal dosage forms are the physical form (liquid, solid, semi-solid) of herbal products produced from herbs, with or without excipients in a particular formulation such as tablets, syrup, suspension, ointments, [5].

Preparation of herbal materials for processing

In general, for processes such as extraction , fractionation, purification and fermentation, [5]**.**

Extraction

Extraction is a process in which soluble plant chemical constituents including those which have therapeutic activity are separated from insoluble plant metabolites and cellular matrix, by the use of selective solvents, [5]. The purpose of extraction of herbal material is to eliminate unwanted materials and to concentrate other chemical constituents in a soluble form, [5].

Different techniques are used for extraction such as maceration, infusion, percolation- including hot continuous (Soxhlet) extraction- and decoction, [5].

Methods of extraction

1. **Maceration:** Maceration is a simple extraction technique that involves soaking plant material in a solvent at room temperature. The solvent can be water, oil or alcohol.

How it works:

-Prepare the plant material by cutting it into coarse pieces or grinding it into a powder

-Place the plant material in an airtight container

-Add the solvent to the container

-Agitate the container occasionally

-Let the material soak in the solvent for at least three days.

2. **Infusion:** Infusion refers to an extraction procedure in which boiling water is poured on the herb or herbal material to produce a dilute liquid preparation. Typically, the herb or herbal material is allowed to stand for some time usually 5-20 minutes, [5].

3. **Decoction:** Decoction is the most common method for making herbal preparations in various traditional medicine contexts. It involves boiling the herbal material in water, during time the chemical constituents are dissolved or extracted into the hot liquid. This procedure is suitable for extracting soluble and heat stable active constituents of the herb or herbal material, [5].

4. **Percolation:**Percolation is the procedure in which the solvent is allowed to continuously flow through the herbal material in a percolator. Typically, the properly comminuted herbal material is moistened with an appropriate amount of solvent and allowed to stand for a few hours before being packed into the percolator. Additional solvent is added to totally wet the comminuted herbal material for some time. The bottom end of the percolator is then opened, with fresh solvent being replenished from the top of the percolator to maintain a steady flow of solvent through the bed of herbal material. The flow rate of the liquid is controlled by adjusting the valve of the outlet. The extraction liquid is collected from the bottom outlet of the percolator. When the process is completed, the marc may be pressed and all liquids pooled to obtain the percolate. In addition to the solvent used for the extraction, the flow rate and the temperature, influence the extraction yields and they have to be carefully controlled. Percolation is often used for an exhaustive

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extraction of the herbs and is applicable to both initial and large scale extraction, [5].

5. **Supercritical fluid extraction:**It is a modern technique making use of the solvating property of a fluid in its supercritical state (carbon dioxide is the most common supercritical solvent) to dissolve the chemical constituents in herbal materials, [5].

Suspension

A suspension is a coarse dispersion in which internal phase (therapeutically active ingredient) is dispersed uniformly throughout the external phase. The internal phase consisting of insoluble solid particles having a range of size (0.5 to 5 microns) which is maintained uniformly through out the suspending vehicle with aid of single or combination of suspending agent. The external phase (suspending medium) is generally aqueous in some instance, may be an organic or oily liquid for non oral use.

- The reasons for the formulation of a suspension:

1. When the drug is insoluble in the delivery vehicle.
2. To mask the bitter taste of the drug.
3. To increase the drug stability.
4. To achieve controlled/sustained drug release.

Types of suspension

- Based on general classes:

1. Oral suspension
2. Externally applied suspension
3. Parenteral suspension

- Based on proportion of solid particles:

1. Dilute suspension
2. Concentrated suspension

- Based on electrokinetic nature of solid particles:

1. Flocculated suspension
2. Deflocculated suspension

- Based on size of solid particles:

1. Colloidal suspensions (< 1 micron)
2. Coarse suspensions (> 1 micron)
3. Nano suspensions (10 ng)

II. MATERIALS & METHODS

The ingredients which are used in the preparation of Polyherbal anti-diabetic suspension are listed below.

Sr. No.	Name of Herb	Parts used	Scientific Name	Family	Anti-diabetic Phytoconstituents
1	Garlic	Bulbs	Allium Sativum	Amaryllidaceae	Allicin
2	Fenugreek	Seeds	Trigonella Foenum Graecum	Fabaceae	Trigonelline
3	Neem	Leaves	Azadirachta Indica	Meliaceae	Azadirachtin
4	Cinnamon	Barks	Cinnamomum Zeylanicum	Lauraceae	Cinnamaldehyde

Fig. 1: Allium Sativum (Garlic)

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Allium sativum commonly known as garlic and belonging to family Alliaceae, [4]. It contains allicin, apigenin, s- allyl cysteine sulfoxide, which are responsible for hypoglycemic activity, [4]. Garlic possesses many therapeutic benefits other than hypoglycemic as it exhibits anti-coagulant, antioxidant, antimicrobial, hypocholesterolaemic and hypotensive activity, [4]. It's a perennial herb that's grown all throughout the India, [3]. It is widely used as a food additive, [3]. In comparison to sucrose controls, it helps to decrease in fasting blood sugar. The oral treatment of garlic extract significantly lowers blood glucose. Therefore it is regarded as a good option for future diabetes mellitus research, according to the researchers, [3].

Fig. 2: Trigonella Foenum Graecum (Fenugreek)

Trigonella foenum graecum is commonly known as Fenugreek and Methi, is a member of family Fabaceae, [4]. In its natural state, fenugreek is found in the Mediterranean area as well as in India and China, [6]. It is cultivated mainly as a forage plant. Both the seeds and the leaves of fenugreek appear in literature as an ingredient of food and as medicine, [6]. The seeds of fenugreek contain mucous polysaccharides-galactomannans (25-45%), proteins (43.8%, mainly tryptophan and lysine), free amino acids (mainly 4-hydroxyisoleucine and histidine), fats (7.9%), steroid saponins, alkaloid-trigonelline, coumarins, flavonoids, sterols, lecithin and choline as well as nicotinic acid (formed from the breakdown of trigonelline during roasting) and minerals, [6]. This plant also showantioxidant activity. Fenugreek is considered to be one of the Indian plant species exhibiting anti-diabetic activity, [6].

Fig. 3: Azadirachta Indica (Neem)

Azadirachta indica is commonly called as neem belonging to the family meliaceae, [4]. Neem is the common name for this plant. It is tropical and semi-tropical tree native to India, Burma, Bangladesh, SriLanka, Malaysia and Pakistan, [3]. The powdered part, aqueous extract and alcoholic extract of A. indica show significant hypoglycemic activity, [4].Azadirachta indica has been used in ayurvedic medical tradition as a treatment for diabetes mellitus, its in-vivo antidiabetic activity was seen in streptozotocin-nicotinamide induced diabetes mice. Apart from having anti-diabetic activity, this plant also has antibacterial, antifungal, antimalarial, antifertility, anti-inflammatory, antiarthritic, antiulcer, hepatoprotective and antioxidant effects. These properties may be due to the presence of phytochemical constituents like alkaloids, tannins, coumarin, proteins, stigmasterol, flavonoids, polyphenols and saponins, [4].

Fig. 4: Cinnamomum Zeylanicum (Cinnamon)

Cinnamon is an evergreen tree belongs to the Lauraceae family, which grows from 20 to 30 feet; the leaves are dark green on top and lighter green underneath.Cinnamon is generally used in the aroma and industries due to its smell, which can be combined into diverse varieties of foodstuffs, perfumes and medicinal products. The main constituents of cinnamon are cinnamaldehyde,

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cinnamate, cinnamic acid and numerous essential oils. It also contains procyanidins tannins, mucilage, and a bit amounts of coumarin. Historically, it has also been known for its antibacterial, antifungal, and carminative properties. The antioxidative and antibacterial activity of an extract derived from cinnamon has been demonstrated in recent years. Antidiabetic activity was analyzed in the studies on diabetic rats. It was observed that administering 200 mg of ethanolic extract of Cinnamomum zeylanicum per kg of body weight to animals once a week for 4 weeks had a hypoglycaemic effect. In this study, blood glucose (from 257.0 to 122.9 mg/dL after 4 weeks) and glycosylated hemoglobin levels were reduced. The Cinnamon methylhydroxychalcone polymer (MHCP) from cinnamon functions as a mimetic for insulin in 3T3-L1 adipocytes, [2]. Therefore MHCP may be useful in the treatment of TypeII Diabetes Mellitus, [2].

Excipient profile

The excipients which are used in the preparation of Polyherbal anti-diabeticsuspension are listed below:

Sr. No.	Excipients	Uses
1	Sodium Carboxymethyl Cellulose	Suspending agent
2	Methyl paraben	Preservative
3	Sodium citrate	Buffering agent
4	Water	Solvent/Vehicle

Identification tests for anti-diabetic phytoconstituents

- **For Allicin:**

Sr. No.	Test	Observation	Inference
1	Lead acetate test Add few drops of lead acetate solution to the garlic extract	Development of black precipitate	A black precipitate indicates the presence of allicin
2	Sodium nitroprusside test Add few drops of sodium nitroprusside solution to the garlic extract followed by a drop of sodium hydroxide	Development of purple colour	Purple colour indicates the presence of allicin

- **For Trigonelline:**

Sr. No.	Test	Observation	Inference
1	Dragendorff's test Add 1 ml of dragendorff's reagent to the 2 ml of fenugreek extract	An orange-red precipitate is formed	An orange-red precipitate indicates the presence of trigonelline
2	Mayer's test Add few drops of mayer's reagent to the fenugreek extract	Formation of creamy white precipitate	A creamy white precipitate indicates the presence of trigonelline
3	Hager's test Add few drops of hager's reagent to the 2 ml of fenugreek extract	Formation of yellow precipitate	A yellow precipitate indicates the presence of trigonelline

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➢ **For Cinnamaldehyde:**

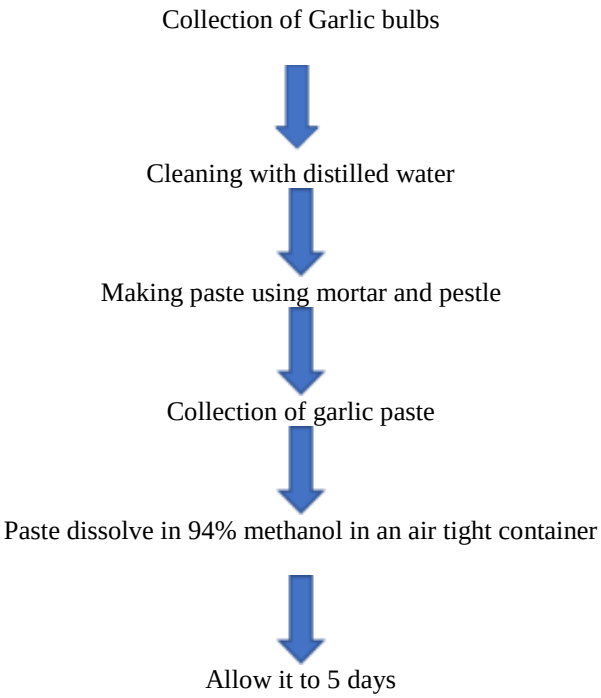
Sr. No.	Test	Observation	Inference
1	Tollen's test Add 2 ml of Tollen's reagent in a test tube containing cinnamaldehyde. Then place the test tube in a water bath set at 35^0 C for 5 minutes.	Formation of silver mirror on the inside of the test tube	A silver mirror around the test tube indicates the presence of cinnamaldehyde
2	2,4-Dinitrophenylhydrazine (2,4-DNP) test Add 2,4-DNP reagent to the extract.	Development of red precipitate	A red precipitate indicates the presence of cinnamaldehyde
3	Schiff's test Add 2 ml of Schiff's reagent to the extract.	Development of pink colour	A pink colour indicates the presence of cinnamaldehyde

➢ **For Azadirachtin:**

Sr. No.	Test	Observation	Inference
1	Liebermann-Burchard test Add acetic anhydride and sulfuric acid to the extract	Development of green colour	A green colour indicates the presence of azadirachtin
2	Salkowski test Add chloroform and sulfuric acid to the extract	Development of reddish-brown colour	A reddish-brown colour indicates the presence of azadirachtin

Extraction of Herbal Ingredients

1. Garlic :



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Filter and collect the extract

2. Fenugreek:

Collection of Fenugreek seeds

Cleaning with distilled water

Making powder using mortar and pestle

Collection of powder

Powder dissolve in 94% methanol in an air tight container

Allow it to 5 days

Filter and collect the extract

3. Neem:

Collection of Neem leaves

Cleaning with distilled water

Making powder using mortar and pestle

Collection of powder

Powder dissolve in 94% methanol in an air tight container

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Allow it to 5 days

Filter and collect the extract

4. Cinnamon:

Collection ofCinnamon bark

Cleaning with distilled water

Making powder using mortar and pestle

Collection of powder

Powder dissolve in 94% methanol in an air tight container

Allow it to 5 days

Filter and collect the extract

METHOD OF PREPARATION OF POLYHERBAL SUSPENSION:

Step 1: Suspensions are prepared by grinding the insoluble materials in the mortar to a smooth paste with a vehicle containing the wetting agent.

Step 2: All soluble ingredients are dissolved in same portion of the vehicle and added to the smooth paste to step 1 to get slurry.

Step 3: The slurry is transformed to a graduated cylinder, the mortar is rinsed with successive portion of the vehicle.

Step 4: Add the vehicle containing the suspending agent.

Step 5: Make up the dispersion to the final volume. In this way, the suspension is prepared.

FORMULATION TABLE: Formulation of 100 ml anti-diabetic polyherbal suspension.

Sr. No.	Ingredients	Role	F1	F2	F3
1	Allicin	API (anti-diabetic)	5 ml	5 ml	5 ml
2	Trigonelline	API (anti-diabetic)	5 ml	5 ml	5 ml
3	Azadirachtin	API (anti-diabetic)	5 ml	5 ml	5 ml

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4	Cinnamaldehyde	API (anti-diabetic)	5 ml	5 ml	5 ml
5	Sodium Carboxymethyl Cellulose	Suspending agent	1.5 gm	2 gm	1.8 gm
6	Methyl Paraben	Preservative	0.18 gm	0.30 gm	0.25 gm
7	Sodium Citrate	Buffering agent	0.3 gm	0.4 gm	0.2 gm
8	Water	Vehicle/Solvent	Q.S.	Q.S.	Q.S.

EVALUATION PARAMETERS:

1. **Colour:** The colour of the suspension is determined by observing the 10 ml of final suspension with our naked eyes.

2. **Odour:** The odour of the suspension is determined by smelling the 10 ml of final suspension.

3. **Taste:** The taste of the suspension is determined by taking the little amount of final suspension on taste bud of tongue.

4. **pH determination:** The pH of the suspension can be determined using pH meter and noted.

5. **Determination of viscosity:** The viscosity of the suspension can be determined at room temperature using OSWALD's viscometer.

6. **Flow rate:** The time required for suspension sample to flow through a pipette can be determined the apparent viscosity of the suspension and can be calculated using the following equation:

   Flow rate = Volume of pipette (ml) / Flow time

7. **Sedimentation volume:**The suspension formulation (50 ml) is poured separately into 100 ml measuring cylinders and sedimentation volume was noted after 1,2,3, and 7 days. Triplicate results were obtained for each formulation. Sedimentation volume is calculated according the equation:

$F = V_u / V_o$

Where, F = sedimentation volume
V_u= ultimate height of the sediment
V_o= initial height of the total suspension

8. **Crystal growth:**The stability of suspension will also reduce because of crystal growth, whichusually occurs from temperature variation during storage and form broad particle sizedistribution. Crystal formulation can be determined at 4^o C, room temperature and 47^o C.

Sr. No.	Evaluation Parameter	F1	F2	F3
1	Colour	reddish brown	dark reddish brown	reddish brown
2	Odour	pleasant	pleasant	pleasant
3	Taste	pungent	pungent	pungent
4	pH	6.4	6.5	6.4
5	Viscosity	56.6 cP	59.2 cP	63.4 cP
6	Flow rate	5ml/3.80 sec	5ml/4.20 sec	5ml/3.60 sec
7	Sedimentation volume/ rate	1.8	2.1	1.6
8	Crystal growth	No	No	No

III. CONCLUSION:

Polyherbal anti-diabetic suspension made from extracts of Garlic (Allium Sativum), Neem (Azadirachta Indica), Cinnamon (Cinnamomum Zeylanicum), and Fenugreek (Trigonella Foenum-Graecum) have shown promising results in managing blood sugar levels. The combined effect of these herbs can lead to several potential outcomes. These herbs work synergistically to enhance the body's response to insulin, potentially reducing insulin resistance, a key factor in type 2 diabetes. These herbs can help to regulate cholesterol and triglyceride levels, reducing the risk of cardiovascular complications associated with

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diabetes. Neem contains bioactive compounds like flavonoids and glycosides that help to lower blood glucose levels by enhancing insulin sensitivity. Garlic contains allicin, which is responsible for anti-diabetic activity. It helps to decrease in fasting blood sugar levels. Cinnamon improves insulin function by increasing glucose uptake in cells, reducing fasting blood sugar levels. Fenugreek which is rich in soluble fiber and saponins that slow carbohydrate digestion and improves glucose metabolism. Fenugreek and cinnamon help to slow the absorption of carbohydrates, preventing rapid sugar spikes after meals. Diabetes is linked to oxidative stress and inflammation. Neem, Cinamon and Fenugreek possess strong antioxidant properties, which may protect pancreatic beta cells from damage. Fenugreek and cinnamon aid in appetite control and metabolism, which may help with weight management a crucial aspect of diabetes care.

The prepared polyherbal suspension subjected to various evaluation parameters and compared with standard anti-diabetic suspension. Herbal medicines have lesser side effects. The above study suggest positive results, but the efficacy depends on the dosage, formulation, and individual responses. Consultation with a healthcare provider is recommended before starting any herbal supplement, especially for individuals on anti-diabetic medications to prevent hypoglycemia.

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