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Antidiabetic activity of cow urine and it's preparations: an overview

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Abstract

In Ayurveda, cow urine (Gomutra) occupies a unique place and has been recognized as water of life or "Amrita". In Sushruta Samhita, it has been described as the most effective substance of animal origin. In India, drinking of cow urine has been practiced for thousands of years. Kamdhenu or Indian cow (Bos indicus) is worshipped as sacred animal by Hindus. The reason for worshipping is the tremendous therapeutic values of cow derived products like cow milk, cow milk curd, cow butter (ghee), cow urine, cow dung and a combination formulation Panchgavya. The use of these products has been well defined in ancient Ayurvedic texts like Charak samhita, Shushruta samhita, Brahad-Wagbhatt etc. Cow ghee showed anticancer and hepatoprotective potential by altering the enzymatic activities whereas cow urine acts by an unknown mechanism.The effect of cow urine formulation (Gomutra ark, GoA) on experimental alloxan-induced diabetes in rats was studied. Wistar albino rats of either sex weighing 200-250 g were used. The biochemical parameters observed were blood sugar, vitamin C and malondialdehyde(MDA) release. GoA significantly lowers blood glucose in diabetic rats although the observed effect was found to be less than standard antidiabetic, glibenclamide. It is suggested that GoA might have a significant protective effect against alloxan-induced type I Diabetes Mellitus. GoA contains volatile fatty acids like acetic acid 2 propenyl ester, acetic acid methyl ester, 2 2 3 trichloro propionic acid, Butanoic acid-3methyl, propyl ester, 1H indol-3-acetate, acetic acid phenyl ester, quinoline, which act as an antioxidant. The antioxidant potential might be contributing for the antihyperglycemic effect, by preventing formation of the free radicals which cause damage to the beta cells of pancreas. There are so many claims regarding the use of cow urine. Out of these the most important claim is regarding its antidiabetic and antioxidant activity, but only few scientific literatures are available to support this claim.

Keywords: Gomutra ark, Antidiabetic Activity, Cow Urine Preparations, Treatment of Diabetes

Introduction

Cow products (Panchagavya) are getting a lot of attention these days, be it cow dung, cow urine, milk, ghee or curd. Cow urine especially, is promoted as an alternative medicine in India for numerous diseases including cancer, diabetes and tuberculosis. A lot of research work is going on these traditional Indian methods

for curing life threatening diseases and their results also approves the age old medicine.Diabetes is a condition in which the body does not produce enough, or properly respond to insulin, a hormone produced in the pancreas. Insulin enables cells to absorb glucose in order to turn it into energy. In diabetes, the body can't use its own insulin as well as it should doesn't make enough insulin, and sometimes both. This causes glucose to accumulate in the blood, often leading to various complications.

The American Diabetes Association reported in 2009 that there are 23.6 million children and adults in the United States—7.8% of the population, who have diabetes. While an estimated 17.9 million in the US alone have been diagnosed with diabetes, nearly one in four (5.7 million) diabetics are unaware that they have the disease. In 2000, according to the World Health Organization, at least 17.9 million people worldwide suffer from diabetes, or 2.8% of the population. Its incidence is increasing rapidly, and it is estimated that by the year 2030, this number will almost double. Diabetes mellitus occurs throughout the world, but is more common (especially type 2) in the more developed countries. The greatest increase in prevalence is, however, expected to occur in Asia and Africa, where most patients will probably be found by 2030. The increase in incidence of diabetes in developing countries follows the trend of urbanization and lifestyle changes, perhaps most importantly a "Western-style" diet. This has suggested an environmental (i.e., dietary) effect, but there is little understanding of the mechanism(s) at present, though there is much speculation, some of it most compellingly presented. For at least 20 years, diabetes rates in North America have been increasing substantially. In 2008 there were about 24 million people with diabetes in the United States alone, from those 5.7 million people remain undiagnosed. Other 57 million people are estimated to have pre-diabetes. [1] Diabetes mellitus is a metabolic disorder characterized by hyperglycemia and alternation in carbohydrates, fat and protein metabolism associated with absolute or relative deficiencies in insulin secretion or insulin action. It is considered as one of the five leading causes of death in the world. About 150 million or 1.3% people are suffering from diabetes worldwide which is almost five times more than the estimates 10 years ago and this may double by the year 2030. Different types of oral hypoglycemic agents are available along with insulin for the treatment of diabetes mellitus but their long-term use produces undesirable side effects such as skin rashes, transient leucopenia, thrombocytopenia, severe hypoglycemia, and increase chances of cardiovascular death of unknown mechanism. [1] According to the American Diabetes Association, approximately 18.3% (8.6 million) of Americans age 60 and older have diabetes. Diabetes mellitus prevalence increases with age, and the numbers of older persons with diabetes are expected to grow as the elderly population increases in number. The National Health and Nutrition Examination Survey (NHANES III) demonstrated that, in the population over 65 years old, 18% to 20% have diabetes, with 40% having either diabetes or its precursor form of impaired glucose tolerance. Indigenous populations in first world countries have a higher prevalence and increasing incidence of diabetes than their corresponding non-indigenous populations. In Australia the age-standardised prevalence of self-reported diabetes in Indigenous Australians is almost 4 times that of non-indigenous Australians. Preventative community health programs such as Sugar

Man (diabetes education) are showing some success in tackling this problem.[1]

Types of diabetes:

- Type 1: Insulin-dependent diabetes mellitus (IDDM), juvenile onset diabetes mellitus: There is β cell destruction in pancreatic islets majority of cases are autoimmune (type 1A). Antibodies that destroy β cells are detectable in blood, but some are idiopathic (type 1B). In all type 1 cases circulating insulin levels are low or very low, and patients are more prone to ketosis. This type is less common and has a low degree of genetic predisposition results from the body's failure to produce insulin. It is estimated that 5-10% of persons who are diagnosed with diabetes have type 1diabetes. Presently almost all persons with type 1 diabetes must take insulin injections.
- Type 2: Non insulin-dependent diabetes mellitus (NIDDM), maturity onset diabetes mellitus: There is no loss or moderate reduction in β cell mass; insulin in circulation is low, normal or even high, no anti β-cell antibody is demonstrable; has a high degree of genetic predisposition; generally has a late onset (past middle age). Over 90% cases are type 2 diabetes mellitus(DM). Causes may be: Type 2 Results from Insulin resistance, a condition in which cells fail to use insulin properly, sometimes combined with relative insulin deficiency. Many people designated to develop type 2 diabetes spend many years in a state of Pre-diabetes: termed as a condition that occurs when a person's blood glucose levels are higher than normal but not high enough for a diagnosis of type 2 diabetes. As of 2009 there are 57 million Americans who have pre-diabetes.
  - Abnormality in gluco-receptor of β cells so that they respond at higher glucose concentration or relative β cell deficiency.
  - Reduced sensitivity of peripheral tissues to insulin: reduction in number of insulin receptors, 'down regulation' of insulin receptors. Many hypertensives are hyperinsulinaemic, but normoglycaemic; exhibit insulin resistance associated with dyslipidaemia (metabolic syndrome). Hyperinsulinaemia *per se* has been implicated in causing angiopathy.
  - Excess of hyperglycaemic hormones (glucagon, etc.)/obesity: cause relative insulin deficiency—the β cells lag behind.
  - Gestational diabetes: Pregnant women who have never had diabetes before but who have high blood sugar (glucose) levels during pregnancy are said to have gestational diabetes. Gestational diabetes affects about 4% of all pregnant women. It may precede development of type 2 (or rarely type 1).
  - Many other forms of diabetes mellitus are categorized separately from these. Examples include congenital diabetes due to genetic defects of insulin secretion, cystic fibrosis-related diabetes, steroid diabetes induced by high doses of glucocorticoids, and several forms of monogenic diabetes.[1,2]

All forms of diabetes have been treatable since insulin became medically available in 1921, but there is no cure for the common types except a pancreas transplant, although gestational diabetes usually resolves after delivery. Diabetes and its treatments can cause many complications. Acute complications including hypoglycemia, diabetic ketoacidosis, or nonketotic hyperosmolar coma may occur if the disease is not

adequately controlled. Serious long-term complications include cardiovascular disease, chronic renal failure, retinal damage, which can lead to blindness, several types of nerve damage, and microvascular damage, which may cause erectile dysfunction and poor wound healing. Poor healing of wounds, particularly of the feet, can lead to gangrene, and possibly to amputation. Adequate treatment of diabetes, as well as increased emphasis on blood pressure control and lifestyle factors such as not smoking and maintaining a healthy body weight, may improve the risk profile of most of the chronic complications. In the developed world, diabetes is the most significant cause of adult blindness in the non-elderly and the leading cause of non-traumatic amputation in adults, and diabetic nephropathy is the main illness requiring renal dialysis in the United States**.**[2]

The classical symptoms of diabetes are polyuria, polydipsia, weight loss, fatigability and increased thirst and consequent increased fluid intake. Symptoms may develop quite rapidly (weeks or months) in type 1 diabetes, particularly in children. However, in type 2 diabetes symptoms usually develop much more slowly and may be subtle or completely absent. Type 1 diabetes may also cause a rapid yet significant weight loss (despite normal or even increased eating) and irreducible mental fatigue. All of these symptoms except weight loss can also manifest in type 2 diabetes in patients whose diabetes is poorly controlled, although unexplained weight loss may be experienced at the onset of the disease. Final diagnosis is made by measuring the blood glucose concentration.[1,3]

When the glucose concentration in the blood is raised beyond its renal threshold, reabsorption of glucose in the proximal renal tubule is incomplete, and part of the glucose remains in the urine (glycosuria). This increases the osmotic pressure of the urine and inhibits reabsorption of water by the kidney, resulting in increased urine production (polyuria) and increased fluid loss. Lost blood volume will be replaced osmotically from water held in body cells and other body compartments, causing dehydration and increased thirst.[3]

Prolonged high blood glucose causes glucose absorption, which leads to changes in the shape of the lenses of the eyes, resulting in vision changes; sustained sensible glucose control usually returns the lens to its original shape. Blurred vision is a common complaint leading to a diabetes diagnosis; type 1 should always be suspected in cases of rapid vision change, whereas with type 2 change is generally more gradual, but should still be suspected**.**[3]

Insulin is released into the blood by beta cells (β-cells), found in the Islets of Langerhans in the pancreas, in response to rising levels of blood glucose, typically after eating. Insulin production is more or less constant within the beta cells, irrespective of blood glucose levels. It is stored within vacuoles pending release, via exocytosis, which is primarily triggered by food, chiefly food containing absorbable glucose. The chief trigger is a rise in blood glucose levels after eating.[4]

Insulin is the principal hormone that regulates uptake of glucose from the blood into most cells (primarily muscle and fat cells, but not central nervous system cells). Therefore deficiency of insulin or the insensitivity of its receptors plays a central role in all forms of diabetes mellitus. [4]

Most of the carbohydrates in food are converted within a few hours to the monosaccharide glucose, the principal carbohydrate found in blood and used by the body as fuel. The most significant exceptions are fructose, most disaccharides (except sucrose and in some

people lactose), and all more complex polysaccharides, with the outstanding exception of starch. [4]

Treatment of Diabetes:

Insulin

Insulin was discovered in 1921 by Banting and Best who demonstrated the hypoglycaemic action of an extract of pancreas prepared after degeneration of the exocrine part due to ligation of pancreatic duct. It was first obtained in pure crystalline form in 1926 and the chemical structure was fully worked out in 1956 by Sanger. Insulin is a two chain polypeptide having 51 amino acids and MW about 6000. The A-chain has 21 while B-chain has 30 amino acids.

Pork insulin is more homologous to human insulin than is beef insulin. The A and B chains are held together by two disulfide bonds.

Insulin is synthesized in the β cells of pancreatic islets as a single chain peptide Preproinsulin (110 A A) from which 24 AAs are first removed to produce Proinsulin. The connecting or 'C' peptide (35 AA) is split off by proteolysis in Golgi apparatus; both insulin and C peptide are stored in granules within the cell. The C peptide is secreted in the blood along with insulin.

Regulation of insulin secretion

Under basal condition ~1U insulin is secreted per hour by human pancreas. Much larger quantity is secreted after every meal. Secretion of insulin from β cells is regulated by chemical, hormonal and neural mechanisms.

Chemical: The β cells have a glucose sensing mechanism dependent on entry of glucose into β cells (through the aegis of a glucose transporter GLUT2) and its phosphorylation by glucokinase. Glucose entry and activation of the glucoceptor indirectly inhibits the ATP-sensitive K^+ channel resulting in partial depolarization of the β cells. This increases intracellular Ca^{2+} availability (due to increased influx, decreased efflux and release from intracellular stores) exocytotic release of insulin storing granules. Other nutrients that can evoke insulin release are—amino acids, fatty acids and ketone bodies, but glucose is the principal regulator and it stimulates synthesis of insulin as well. Glucose induces a brief pulse of insulin output within 2 min (first phase) followed by a delayed but more sustained second phase of insulin release.

Glucose and other nutrients are more effective in invoking insulin release when given orally than i.v. They generate chemical signals 'incretins' from the gut which act on β

cells in the pancreas to cause anticipatory release-of insulin. The incretins involved are glucagon-like peptide-1 (GLP-1), glucose-dependent insulin inotropic polypeptide (GIF), vasoactive intestinal peptide (VIP), pancreozymin-cholecystokinin, etc.; but different incretins may mediate signal from different nutrient. Glucagon and some of these peptides enhance insulin release by increasing cAMP formation in the β cells.

Hormonal: A number of hormones, e.g. growth hormone, corticosteroids, thyroxine modify insulin release in response to glucose. PGE has been shown to inhibit insulin release. More important are the intra-islet paracrine interactions between the hormones produced by different types of islet cells. The β cells constitute the core of the islets and are the most abundant cell type. The αcells, comprising 25% of the islet cell mass, surround the core and secrete glucagon. The delta cells (5-10%) elaborating somatostatin are interspersed between the α cells. There are some PP (or F) cells (pancreatic polypeptide containing) also.

- Somatostatin inhibits release of both insulin and glucagon.
- Glucagon evokes release of insulin as well as somatostatin.
- Insulin inhibits glucagon secretion.

The three hormones released from closely situated cells influence each other's secretion and appear to provide fine tuning of their output in response to metabolic needs.

Neural: The islets are richly supplied by sympathetic and vagal nerves.

- Adrenergic β_2 receptor activation decreases insulin release (predominant) by inhibiting β cell adenylyl cyclase.
- Cholinergic—muscarinic activation by acytylcholine or vagal stimulation causes insulin secretion through IP_3/DAG-increased intracellular Ca^{2+} in the β cells.

These neural influences appear to govern both basal as well as evoked insulin secretion, because the respective blocking agents have effects opposite to that mentioned above. The primary central site of regulation of insulin secretion is in the hypothalamus: stimulation of ventrolateral nuclei evokes insulin release, whereas stimulation of ventromedial nuclei has the opposite effect. [4]

Actions of Insulin

The overall effects of insulin are to favour storage of fuel. The actions of insulin and the results of its deficiency can be summarized as:

1. Insulin facilitates glucose transport across cell membrane; skeletal muscle and fat are highly sensitive. The availability of glucose intracellularly is the limiting factor for its utilization in these and some other tissues. However, glucose entry in liver brain, RBC, WBC and renal medullary cells is largely independent of insulin. Ketoacidosis interferes with glucose utilization by brain diabetic coma. Muscular activity induces glucose entry in muscle cells without the need for insulin As such, exercise has insulin sparing effect.
2. The first step in intracellular utilization of glucose is its phosphorylation to form glucose – 6 – phosphate. This is enhanced by insulin through increased production of glucokinase. Insulin facilitates glycogen synthesis from glucose in liver, muscle and fat by stimulating the enzyme glycogen synthase.
3. Insulin inhibits gluconeogenesis (from protein, FFA and glycerol) in liver by gene mediated decreased synthesis of phosphoenol pyruvate carboxykinase. In insulin deficiency, proteins and amino acids are funneled from peripheral tissues to liver where these are converted to carbohydrate and Urea.
4. Insulin inhibits lipolysis in adipose tissue and favours triglyceride synthesis. In diabetes increased amount of fat in broken down due to unchecked action of lipolytic hormones (glucagons, Adr, thyroxine, etc.) increased free fatty acid.
5. Insulin enhances transcription of vascular endothelial lipoprotein lipase and thus increases clearance of VLDL and chylomicrons.
6. Insulin facilitates amino acid entry and their synthesis into proteins, as well as inhibits protein break down in muscle and other cells.

Adverse reactions of insulin

1. Hypoglycaemia: This is the most frequent and potentially the most serious reaction. It is commonly seen in patients of 'labile' diabetes in whom insulin requirement fluctuates unpredic-tably. Hypoglycaemia can occur in any diabetic following inadvertent injection of large doses, by missing a meal or by performing vigorous exercise. The symptoms, can be divided into those due to counter-regulatory

sympathetic stimulation— sweating, anxiety, palpitation, tremor; and those due to deprivation of the brain of its essential nutrient glucose (neuroglucopenic symptoms) — dizziness, headache, behavioural changes, visual disturbances, hunger, fatigue, weakness, muscular incoordination and sometimes fall in BP. Generally, the reflex sympathetic symptoms occur before the neuroglucopenic, but the warning symptoms of hypoglycaemia differ from patient to patient and also depend on the rate of fall in blood glucose level. After long-term treatment about 30% patients lose adrenergic symptoms. Diabetic neuropathy can abolish the autonomic symptoms. Hypoglycaemic unawareness tends to develop in patients who experience frequent episodes of hypoglycaemia. Finally, when blood glucose falls further (to < 40 mg/dl) mental confusion, abnormal behaviour, seizures and coma occur. Irreversible neurological deficits are the sequelae of prolonged hypoglycaemia.

Treatment: Glucose must be given orally or i.v. (for severe cases)—reverses the symptoms rapidly. Glucagon 0.5-1 mg i.v. or adrenaline 0.2 mg s.c. (less desirable) may be given as an expedient measure in patients who are not able to take sugar orally and injectable glucose is not available.

2. Local reactions**:** Swelling, erythema and stinging sometimes occur at site of injection especially in the begin-ning. Lipodystrophy occurs at injection sites after long usage. This is not seen with newer preparations—which may even facilitate reversal of lipoatrophy when injected at the same sites.

3. Allergy : This is infrequent; is due to contaminating proteins; very rare with human/highly purified insulins. Urticaria, angioedema and anaphylaxis are the manifestations.

4. Edema : Some patients develop short-lived dependent edema (due to Na^+ retention) when insulin therapy is started. **[4]**

Antidiabetic activity

The effect of cow urine formulation (Gomutra ark, GoA) on experimental alloxan-induced diabetes in rats was studied. Wistar albino rats of either sex weighing 200-250 g were used. The biochemical parameters observed were blood sugar, vitamin C and malondialdehyde(MDA) release. GoA significantly lowers blood glucose in diabetic rats although the observed effect was found to be less than standard antidiabetic, glibenclamide. It is suggested that GoA might have a significant protective effect against alloxan-induced type I Diabetes Mellitus. GoA contains volatile fatty acids like acetic acid 2 propenyl ester, acetic acid methyl ester, 2 2 3 trichloro propionic acid, Butanoic acid-3methyl, propyl ester, 1H indol-3-acetate, acetic acid phenyl ester, quinoline, which act as an antioxidant. The antioxidant potential might be contributing for the antihyperglycemic effect, by preventing formation of the free radicals which cause damage to the beta cells of pancreas. It significantly lowers the level of malondialdehyde and vitamin C in diabetic rats. No toxicity was observed even when cow urine was given 32 times of the study dose in acute toxicity and no significant change were observed when it was used chronically, suggesting that cow urine is having a very high therapeutic index. The findings of the study supported the traditional use of cow urine in diabetes and have a high therapeutic index and safety profile for chronic use 15. In a study of use of cow urine distillate in diabetes rats, the diabetes was induced by administration of streptozotocin (50 mg/kg bw., i.p) dissolved in citrate buffer (0.1 M, pH 4.5). The anti diabetic effect of the

(three different doses) and a standard drug Glibenclamide (0.25 mg/kg, p.o) was studied in these diabetic rats. The parameters employed in the study included assessment of fasting blood glucose levels, serum lipid profiles, liver glycogen levels and initial and final changes in body weight. The cow urine distillate produced a significant ($P<0.05$) reduction of the elevated blood glucose, serum cholesterol and serum triglycerides level when compared with the diabetic control. The diabetic rats treated with cow urine distillate also showed a significant increase in HDL levels and gain in body weight when compared with the diabetic control

Oral Hypoglycaemic Drugs

These drugs lower blood glucose levels and are effective orally. The main draw back of insulin is-it must be given by injection. Orally active drugs have always been searched.

(A) ***Sulfonylureas***

First generation

Tolbutamide
Chlorpropamide

Second generation

Glibenclamide
(Glyburide)
Glipizide
Gliclazide
Glimepiride

(B) ***Biguanides***

Metformin

(C) ***Meglitinide / Phenyl Alanine Analogues***

Repaglinide, Nateglinide

(D) ***Thiazolidinediones***

Rosiglitazone, Pioglitazone

(E)α-*Glucosidase Inhibitors*

Acarbose, Miglitol

Mechanisms, main actions and adverse effects

Sulfonylureas provoke a brisk release of insulin from pancreas. They act on the so called sulfonylurea receptors (SURI) on the pancreatic β cell membrane-cause depolarization by reducing conductance of ATP sensitive K^+ channels. This enhances Ca^{2+} influx sensitive K^+ channels. This enhances Ca^{2+} influx degranulation. The rate of insulin secretion at any glucose concentration is increased. In type 2 glucose or meals is delayed and subdued. The sulfonyfureas primarily augment the 2^{nd} phase insulin secretion with little effect on the 1^{st} phase. That they do not cause hypoglycemia in pancreatectomised animals and in type 1 diabetics (presence of at least 30% functions β cells is essential for their action) confirms their indirect action through pancreas.

Interactions

Drugs that enhance sulfonylurea action (may precipitate hypoglycamia) are-

(a) Displace from protein binding Phenylbuta-zone, sulfinpyrazone, salicylates, sulfonamides, PAS.

(b) Inhibit metabolism/excretion : Cimetidine, sulfo namides, warfarin, chloramphenicol, acute alcohol intake (also synergises by causing hypoglycaemia)

(c) Synergise with or prolong pharmacodynamic action : Salicylates, propranolol (cardioselective β, blockers less liable), sympatholytic antihyper-tensives, lithium, theophylline, alcohol (by inhibiting gluconeogeesis).

Adverse effects:- Incidence of adverse effects is quite low (3-7%)

1. Hypoglycaemia: It is the commonest problem, may occasionally be severe and rarely fatal. It is more common in elderly liver and kidney disease patients and when potentiating drugs are added chlorpropamide is a frequent culprit due to

its long action. Tolbutamide carries lowest risk due to its low potency and short duration of action lower incidence is also reported with glipizide, glibenclamide, glimepiride.

Treatment to give glucose, may be for a few days because hypoglycaemia may recur.

2. Nonspecific side effects Nausea, vomiting flatulence, or constipation headacine paresthesias and weight gain.

3. Hypersensitivity Rashes, photosensitivity, purpura, transient leucopenia, rearely agranulo-cytosis.

Chlorpropamide in addition causes cholestatic jaundice, dilutional hyponatermia (sensitizes the kidney to ADH action), intolerance to alcohol in predisposed subject (flushing and a disulfirm like reaction); other sulfonylureas are less prone to this interaction.

Tolbutamide reduces iodide uptake by thyroid but hypothyroidism does not occur.

Safety of sulfony lureas during pregnancy is not established-change over to insulin. They are secreted in milk : should not be given to nursing methers.

Biguanides

Two biguanide antidiabetics, phenformin and metformin were introduced in the 150s. Because of higher risk of lactic acidosis, phenformin was withdrawn in many countries and has been banned in India since 2003.

They differ markedly from sulfonylureas : cause little or no hypoglycaemia in nondiabetic subjects, and even in diabetics episodes of hypoglycaemia due to methformin are rare. They do not reported to improve β cells. Metformin is reported to improve lipid profile as well in type 2 diabetics.

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Herbal Treatment of Diabetes

According to ayurveda, diabetes is a metabolic kapha type of disorder in which diminished functioning of agni leads to a tendency toward high blood sugar. (Ayurveda recognizes 24 forms of the disease commonly classified under Prameha - 4 are due to Vata dosha, 6 are due to Pitta dosha, and 10 are caused by Kapha dosha. The main causes of these diseases are fat, urine, and Kapha buildups due to foods, liquids, lifestyle and others.

Ayurvedic practitioners attack diabetes using a multiprong approach. First, they address diet modification, eliminating sugar and simple carbohydrates, and emphasizing complex carbohydrates. Protein is limited, since excessive intake can damage the kidneys. Fat is also limited because there is often a deficiency of pancreatic enzymes, making fat digestion difficult. Since many diabetics have autoantibodies, a cleansing program is instituted. Panchakarma is typically used for this purpose. This begins with herbal massages and an herbal steam sauna,

followed by fasting to cleanse the body. This is followed by an herbal purge for the liver, pancreas and spleen. Colon therapy is next, first to cleanse the digestive tract and then to reconstitute the system.

Ayurvedic practitioners also use several herbal preparations for diabetics. Exercise is another cornerstone of ayurvedic treatment of diabetes. Yoga and breathing exercises are traditionally used.

The most important herbs for all doshas are shilajit, gudmar turmeric, neem, amalaki, guggul, and arjuna. Turmeric with aloe vera gel (1 to 3 gms./.035 to .1 oz) is best used during the early stages of diabetes for regulating pancreas and liver functions.

Enumeration of Antidiabetic plants

1. Abrus precatorius L. (Fabaceae). Local Name: Kundumani. The plant is a climber commonly known as Wild Liquorice and found through the plains of India. Leaf of this plant is mixed with the leaves of Andrographis paniculata, Gymnema sylvestre and seeds of Syzygium cumini. The mixture is shade dried and ground into powder and taken orally along with cow's milk. Dosage: About 50 ml of mixture is taken twice a day before food for 120 days.

2. Andrographis lineata Wallich ex Nees (Acanthaceae). Local Name: Siriya nangai. The plant is annual herb found in the hedgerows throughout the plains in India and commonly cultivated in gardens. Leaf is shade dried, powdered and taken orally along with cow's or goat's milk. Dosage: 2 teaspoon of powder is taken twice a day after food for 2-3 months.

3. Andrographis paniculata (Burm.f.) Wall, ex Nees (Acanthaceae). Local Name: Periya nangai. The plant is annual herb (Commonly known as King of Bitters) found in the hedgerows throughout the plains in India and cultivated in gardens. Leaf is shade dried, powdered and mixed with boiled rice and cow's milk and taken orally. Dosage: 50 ml of mixture is taken thrice a day after food for 120 days.

4. Canthium parviflorum Lam. (Rubiaceae). Local Name: Sakkarai kovaimaram. A shrubby and woody plant found throughout the Western Ghats. Shade dried leaf powder is mixed with cup of water or goat's or cow's milk or boiled rice and taken orally. Dosage: One or two teaspoon is taken early in the morning regularly until cure.

5. Costus speciosus (Koenig.) J. E. Smith (Costaceae). Local Name: Kostak-kilangu. A tuberous fleshy herb, plentifully found in north India and in the Western Ghats the plant is seen in hilly areas. Fresh rhizome is ground into a paste and taken orally. Dosage: 20-25 gm is taken thrice a day after food for 2 months.

6. Gymnema sylvestre (Retz.) R. Br. ex Schultes (Asclepiadaceae). Local Name: Siru kurinjan. A climbing shrub commonly found in the plains of central and southern India. Dried leaves are grounded and the fine powder thus obtained is taken orally along with milk. Dosage: About 50 ml is taken twice a day after food for 120 days to treat diabetes.

7. Memecylon umbellatum Burm.(Family - Melastomataceae). Local Name: Sakkarai vaembu. A bushy small tree found in the hilly areas of Western Ghats. Shade dried leaf powder is mixed with cup of water and boiled rice and kept overnight and taken orally. Dosage: One teaspoon is taken early in the morning for forty days or until cure.

8. Momordica charantia L. (Cucurbitaceae). Local Name: Kaattu pagar-kai. The plant is commonly known as Bitter guard and has many varieties. The plant is climbing shrub and generally cultivated everywhere in India. Unripe fruits are taken orally along with food.

Dosage: 2-3 fresh unripe fruits are taken at any time per day for 3 months.

9. Syzygium cumini (L.) Skeels. (Myrtaceae). Local Name: Naaval maram. The plant is large tree and commonly known as Jambolan or Black Plum found throughout the plains. Juice extracted from the leaf is mixed with honey or cow's milk and fresh fruits are taken orally. Dosage: 2 teaspoon of juice is taken twice a day after food for 3 months. It is one of the significant antidiabetic plant and it has long been reported for its use in many pharmacological activities mainly diabetes. During the last four decades, numerous folk medicine and scientific reports on the antidiabetic effects of this plant have been cited in the literature. Clinical and experimental studies suggest that, different parts of the plant especially fruits, seeds and stem bark possess promising activity against diabetes mellitus. S. cumini exerts a dual effect namely a combination of mechanism of action of sulfonylurea and biguanids and may bring about its hypoglycaemic action through stimulation of surviving β cells of islets of langerhans to release more insulin.

10. Wattakaka volubilis (L.f.) Stapf. (Asclepiadaceae). Local Name: Perun-kurinjan. The plant is a fleshy and very large climber found throughout the plains with papery leaves. Leaf powder is taken orally along with cow's milk. Dosage: 50-75 ml of mixture is taken twice a day after food for 90 days. [6]

Cow Urine Therapy

Cow is a mobile dispensary. It is the treasure of medicines. The cow urine therapy is capable of curing several curable and incurable diseases. The holy texts, like Atharva Veda, Charak Samhita, Rajni Ghuntu, Vridhabhagabhatt, Amritasagar, Bhavprakash, Sushrut Samhita contain beautiful description about these things. Cow Urine Treatment and Research Center, Indore has conducted a lot of research in the past few years on patients directly and claimed that it is capable of curing diabetes, blood pressure, asthma, psoriasis, eczema, heart attack, blockage in arteries, fits, cancer, AIDS, piles, prostrate, arthritis, migraine, thyroid, ulcer, acidity, constipation, gynecological problems, ear and nose problems, abortion and several other diseases.[7] Cow urine has a unique place in Ayurveda and has been described in "sushrita samhita" and a ashtanga sangraha to be the most effective substance/secrection of animal origin with innumerable therapeutic value. It has been recognized as water of life or amrita. This kind of alternative treatment as panchgavya therapy or cowpathy has been reported to be beneficial even for dreaded disease like cancer, AIDS, and diabetes. Improvement has been shown or reported with those suffering from flu, allergies, colds, rheumatoids arthritis, bacterial/viral infection, tuberculosis, chicken pox, hepatitis, leucorrhuea, leprosy, ulcer, heart disease, asthma, skin infection, aging, chemical intoxication. Through extensive research studies of cow urine distillated fraction popularly know as ark has been identified as bioenhancer of the activity of commonly used antibiotic, antifungal and anticancer drug. Cow urine enhances the immunocompetence and improve general health of an individual prevent the free radicals formation and act as anti-aging factor reduce apoptosis in lymphocytes and help them to survive and efficiently repair the damaged DNA and this is effective for cancer therapy. [8] The analysis of cow urine has shown that it contains nitrogen, sulphur, phosphate, sodium, manganese, carbolic acid, iron, silicon, chlorine, magnesium, malic, citric, tartric and succinic acid, calcium salts, Vitamin A, B, C, D, E, minerals, lactose,

enzymes, creatinine, hormones and gold. A person falls ill when there is deficiency or excess of the substances inside the body. The cow urine contains those substances, which are present in the human body. Therefore consumption of cow urine maintains the balance of these substances and cures incurable diseases. [8]

Rat pancreatic islets exposed to interleukin-lβ (IL-lβ) in the presence of succinic acid monomethyl ester (SAM) have a higher insulin release in response to glucose and higher glucose oxidation rates, as compared to islets exposed to IL-1β alone. These beneficial effects of SAM were not accompanied by any decrease in IL-lβ-induced nitric oxide (NO) production nor inhibition of aconitase activity. Moreover, SAM did not increase biosynthesis of glutamate decarboxylase. SAM apparently improves β-cell function. [9]

The monoethyl, monopropyl and monoisopropyl esters of succinic acid, administered intravenously at the dose of 2 |mili mol/g body weight, were found to increase the insulinotropic action of gliquidone (0.2 nmol/g body weight) in anaesthetized rats. The monoisopropyl ester of succinic acid also doubled the hypoglycemic action of gliquidone. These findings indicate that it is possible to design esters of succinic acid that are devoid of the risk of generating methanol by intracellular hydrolysis, and yet susceptible to increase both the insulinotropic and hypoglycemic responses to antidiabetic agents. [10]

Cow urine has a unique place in Ayurveda and has been described in 'Sushrita Samhita' and 'Ashtanga Sangraha' to be the most effective substance/secretion of animal origin with innumerable therapeutic values. It has been recognized as water of life or "Amrita’. Various products of cow urine have been suggested as a successful remedy against more than 100 diseases from fever to cance

Cow urine Preparations

(1) Fresh cow urine : Fresh cow urine is first urine collected in the morning.

(2) Distillate cow urine (gau arc) : Cow urine is boiled in an earthen pot or iron pot to which a vapour condensing device is attached. The vapour through tube is collected in a pot put over cold water. So the vapour gets condensed here . The water is changed often to maintain it cool. The tube through which vapour passes should be transparent so that movement of vapour is visible. If smoke is visible then reduce the flame. The properties of this condensed cow urine are not as good as fermented cow urine, as the residue remains in the pot and some component evaporate as gases. But being clean and odour free it is liked more. Benefit is achieved by using for longer period. It can be given to children and ladies easily. If honey is mixed while taking then it is more effective. 12 millilitres of ark (distilled cow urine) mixed with honey should be taken after food.

(3) Residue of cow urine (ganavati) :

Use deep iron pan. Go on boiling cow urine till it becomes concentrated and salts remain. This has to be done just as sugarcane juice is concentrated and jaggery is formed. When the cow urine is concentrated remove it from fire and let it cool. From one kilo cow urine 50 gm concentrate is available. Scratch it from pan and make round tablets of the size of gram.

To keep tablets non-sticky; burn some dried good quality cow dung cakes to ashes and filter ash using a thin cloth piece and keep the tablets in the ash. To make colour of attractive. The tablets should be kept in the cow dung ash powder only. The cow dung power acts as absorbent and heat isolator. The tablets now could be put

in plastic bags out. It is just to keep tablets free from moisture. [11]

Chemical description of cow urine as per modern concepts and cure of diseases accordingly.

Table 1: Chemical contents of cow urine and cure of diseases as per them.

S. No.	Name of chemical	Effect of chemical on diseases
1.	Nitrogen (N_2)	Removes blood abnormalities and (toxins, Natural stimulant of urinary track, activates kidneys and it is diuretic.
2.	Sulphur (S)	Supports motion in large intestines. Cleanses blood.
3.	Ammonia (NH_3)	Stabilise bile, mucous and air of body. Stabilises blood formation.
4.	Copper (Cu)	Controls built up of unwanted fats
5.	Iron (Fe)	Maintains balance and helps in production of red blood cells & haemoglobin. Stabilises working I power.
6.	Urea ($CO(NH_2)_2$)	Affects urine formation and removal, Germicidal.
7.	UricAcid	Removes heart swelling or inflammation. It is diuretic therefore destroys toxins.
8.	Phosphate (P)	Helps in removing stones from urinary track.
9.	Sodium (Na)	Purifies blood. Antacid
10.	Potassium (K)	Cures hereditary rheumatism, Increases appetite. Removes; muscular weakness and laziness.
11.	Manganese (Mn)	Germicidal, stops growth of germs, protects decay due to gangrene.
12.	Carbolic acid (HCOOH)	Germicidal, stops growth of germs and decay due to gangren
13.	Calcium (Ca)	Blood purifier, bone stregthener, germicidal, Rakta skandak
14.	Salt (NaCl)	Sanyas vishamta decreases acidic contents of blood, germicidal
15	Vitamins A,B,C,D,E	Vitamin B is active ingredient for energetic life and saves from nervousness and thirst, strengthens bones and reproductive ingredient for energetic life and saves from nervousness and thirst, strengthens bones and reproductive power.
16.	Other Minerals	Increase immunity
17.	Lactose ($C_6H_{12}O_6$)	Gives satisfaction. Strengthens Mouth, strengths heart, removes thirst and nervousness.
18.	Enzymes	Make healthy digestive juices, increase immunity
19.	Water (H_2O)	lit is life giver. Maintains fluidity of blood, maintains body temperature
20.	Hipuric acid	Removed toxins through urine
21.	Creatinin	Germicide
22.	Aurum Hydroxide (AuOH)	it is germicidal and increases immunity power. AuOH is highly antibiotic and anti-toxic

The study by Khan and Vinoy (2005) illustrated the antitoxic and bioenhancing role of Kamdhenu Ark (cow's urine distillate) in fertility rate of male mice affected by cadmium chloride toxicity. [12]

Achliya et al (2003) investigated the Hepatoprotective activity of Panchagavya Ghrita (PG) against CCl_4 induced hepatotoxicity in albino rats.

The administration of PG markedly prevented CCl_4 induced elevation of levels

of serum GPT, GOT, ACP and ALP. Histopathology study of liver exhibited almost normal architecture, as compared to control group. [13]

It merely shows that if used in the right combination with complex modern drugs, the distillate of cow urine makes the drugs more potent and works as a bioenhancer or increases body's ability to use the drug and hence increasing bioavailability of many drugs. [14]

Mashelkar, R. A (2007) revealed that the combination having cow urine distillate might also help reduce the inputs (and the resultant costs) of manufacturing drugs like Taxol, an anti-cancer agent derived from the bark of the Pacific Yew tree. [15]

Antigenotoxic / Ameliorative effect of Ark in human polymorphonuclear leukocytes was conducted by Dr. Datta, S. Saravana Devi et al. The ark and redistilled ark was found to possess total antioxidant status of around 2.6mmol contributed mainly by volatile fatty acids (1500mg/litre). These fatty acids and other antioxidants might be responsible for the observed ameliorative effect. [16]

A.K. Maheshwari, A.K.Gupta, et. al, demonstrated that Cow Urine is having antiseptic properties. The urine implicated wounds were found less infected and healing time was also less when compared to antiseptic cream. Administration of fresh urine orally has added effect on wound healing due to immunological properties. [17]

The study conducted by A.K. Singh, et. al and they concluded free radicals attack the nearest stable molecule and steal the electron. This is a chain reaction of destruction. They can attack enzymes, fat, proteins, etc and causes DNA to mutate. This in turn may predispose for- Parkinson's Disease, Alzhimor's Disease, Cancer, Sclerosis, Stroke, Stress, Fibrosis, Cataract, Macular Degeneration, Aging and Cow urine prevents the free radicals formation. [18]

The study conducted by Dipanwita Datta, the Arka could protect mitomyncin C induced chromosomal aberration. The Arka has Volatile acids about 39mg/ltrs and these Volatile Acids are Antioxidants, which show the ameliorating effect on DNA and protect DNA damage. [19]

K Darma, et. al, concluded that the cow Urine Therapy is suggested to posses potent Anticancer abilities. Cow urine efficiently repairs the damaged DNA. Damage of DNA by chemicals is the major cause for Cancer. This property reduces the spread of malignant cancers and helps fighting tumor. [20]

The study conducted by Sangeeta Mehta, et. al and the study shows that Sahiwal cow's milk is closer to human mother's milk because of its significantly lower soluble phosphate level when compared to H.F or Buffaloe. [21]

Girish S. Achilya, et.al, concluded that the "Unmadanashak Ghrita" is a Ayurvedic formulation containing Ferula Narthex, Gardenia Gummifera, Ellataria cardamom, Bacopa monneri, Cowsghee (76%) Unmadanashak Ghrita has CNS depressant and anticonvulsant activity. [22]

The study conducted by Sarman Singh and the study shows that Leishmaniasis (Kala azar) is a highly endemic disease in Indian Sub continent and the cow urine shows strong growth inhibitory action. [23]

C.V. Gore et.al demonstrated that 4 gms of Ashtamangal Ghrit was given every day for 4 months to the students. Academic performance and Intelligence test was performed. The result showed favorable effect on the intelligence of students. [24]

Antidiabetic activity of Cow Urine is also conducting by Researchers of pharmacology and toxicology department, Veterinary College, Hebbal, India.

Succinic acid is the physiologically occurring substrate of succiinate

dehydrogenase in mammals that play a role in cellular respiration and energy metabolism.

Boden G. Front. Biosci, demonstrated that free fattly acids induce insulin resistance in human in a dose dependent fashion. D169-175 (1998); Boden G., Diabetes 46(1) : 3-10 (1997). Lowring of plasma free fatty acid levels is accordingly effective in the treatment of insulin resistance in a mammal.

It now been found that administration of and effective amount of succinic acid or salt thereof to insulin resistant mammals is effective therapy for treating of insulin resistance. Lowering of plasma free fatty acid levels accompanies a lowering of pathologically elevated insulin and glucose levels that reflects and improving in insulin.

This result is unexpected to Japanese Patent No. 61141417 describing that dicarboxylic acids succinic acid are useful as antidiabetics showing promoting action on insulin secretion.

MacDonald at al, demonstrated contrary to Japanese Patent No. 61171417 data that unesterified suiccinate, the compound of the present invention, did not stimulate insulin release in pancreatic islets but only esters of succinic acid are potent insulin secre-tagogues.

MacDonald, M.J. Fahien, L.A. Diabetes 37(7) : 997-99(1988), demonstrated that promotion of insulin secretion are useful in treating insulin dependent diabetic mammals with low or no insulin secretion, while insulin resistant mammals including non-insulin dependent diabetic mammals are needed in decreasing of elevated insulin levels rather than promotion of insulin secretion.

Discussion

Management o diabetes with the agents devoid of any side effects is still a challenge to the medical system. This has led to an increase in the demand for natural products with antihyperglycemic activity and fewer side effects. The cow urine preparations exhibited dose-dependent antidiabetic property. The antidiabetic effect of these preparations at the dose of 5 ml/kg is even slightly higher then gliclazide 10 mg/kg. After daily treatment with cow urine preparations and gliclazide led to a dose dependent fall in blood sugar levels. The reduction of hyperglycemia were significant ($p<0.01$) on 7^{th},14^{th} & 21^{st} days after treatment with the cow urine preparations, as compared with the day 0 observations.

The antihyperglycemic effects exhibited by 5 ml/kg of cow urine preparations was slightly higher than that of gliclazide 10 mg/kg.

Boden G. Front. Biosci, demonstrated for the first time induce insulin resistance in human in a dose dependent fashion. Lowring of plasma free fatty acid levels is accordingly effective in the treatment of insulin resistance in a mammal.

It has now been found that administration of and effective amount of succinic acid or salt thereof to insulin resistant mammals is effective therapy for treating of insulin resistance. Lowering of plasma free fatty acid levels accompanies a lowering of pathologically elevated insulin and glucose levels that reflects and improving in insulin sensitivity.[25]

Cow urine preparations contain succinic acid as observed in the study. Cow urine preparations were given a dose 5 ml/kg. The cow urine preparations as (1 ml) contain 0.39 mg i.e. 1.95 mg/kg. This amount of succinic acid show the antidiabetic activity in the study. The dose used of succinic acid in previous study is 5 mg/kg of body weight.

Lipids play an important role in the pathogenesis of diabetes mellitus. Hyperlipidemia is a recognized consequence of diabetes mellitus demonstrated by the elevated levels of

tissue cholesterol, phospholipids and free fatty acids. Diabetes-induced hyprtlipidemia is attributable to excess mobilization of fat from the adipose tissuee due to the under utilizations of glucose. The abnormal high concentration of serum lipids in diabetes is mainly due to the increase in the mobilization of free fatty acids from the peripheral depots since insulin inhibits the hormone sensitive lipase. On the other hand, glucagons, catecholamine, and other hormones enhance lipolysis. The level of serum lipids is usually raised in diabetes and such an elevation represents a risk factor for coronary heart disease. The levels of serum cholesterol and triglycerides were raised in diabetic rats but which lowered significantly with the treatment of cow urine preparations. It indicates that the cow urine preparations are more useful in the treatment of diabetes as it has hypolipidemic effect. Moreover, its hypolipidemic effect could represent a protective mechanism against the development of atherosclerosis, which is usually associated with diabetes. The levels of HDL cholesterol were significantly increased in the groups treated with cow urine preparations.

The levels of SGOT, SGPT and Serum creatinine were increased very were increased in diabetic rats as compared with normal control rats. Treatment with cow urine preparations reduced the SGOT, SGPT and Serum creatinine very significantly decreased (P<O.05), when compared with the diabetic control group. The findings indicate that these preparations may have hepatoprotective effects and could be effective therapy for both diabetes and hepatotoxicity.

The effects of the cow urine preparations on body weight in the alloxan-induced diabetic rats are shown in [Table 15]. The results of the body weight analysis indicate that the body weight of the untreated diabetic rats was found to be significantly (P<0.05) decreased when compared with the normal control group. The body weight was slightly increased in the normal control group compared to initial weight. Treatment with cow urine preparations and glyclazide prevented reduction in body weight and the weight was increased after the treatment. This shows that cow urine preparations increase body weight reduced due to diabetes and this may help to maintain normal body weight.

Conclusion

The cow urine preparations have significant hypoglycaemic effect in alloxan induced diabetic rats. Cow urine preparations also lowers hypertri-glyceridemia and hyperlipidemia in alloxan induced diabetic rats. There weresignificant improvement in the animals in lowering blood glucose level. The reduction of hyperglycemia were significant (P<O.01) on 7^{th}, 14^{th}, and 21^{th} days after treatment with the cow urine preparations, as compared with the 0 day observations. The reduction of hyperglycemia was significant comparable with the standard gliclazide. The levels of serum cholesterol and triglycerides were increased very significantly and the levels of HDL were decreased in diabetic rats as compared with normal control rats. Cholesterol and triglycerides level were decreased in the cow urine preparations treatd groups at the dose of 5ml /kg as compared with diabetic control group. HDL levels were increased in diabetic rats treated with cow urine preparations as compared with diabetic control group. Effect of cow urine preparations specially fresh cow urine have more significant reduction in lipid profile (TG, TC, LDL) & increase in HDL-Cholestrol than standard gliclazide.

The present work has detected that cow urine preparations were contained succnic

acid that administration of and effective amount of succinic acid or salt thereof to insulin resistant mammals is effective therapy for treating of insulin resistance.

The levels of SGOT, SGPT and Serum creatinine were increased in diabetic rats as compared with normal control rats. Treatment with cow urine preparations reduced the SGOT, SGPT and Serum creatinine very significantly decreased (P<O.01), when compared with the diabetic control group.

From findings of present study we can conclude that cow urine and its preparations could be effective in treating diabetes, arthrosclorsis, hyperlipidaemia, hepatic & renal toxicity.

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