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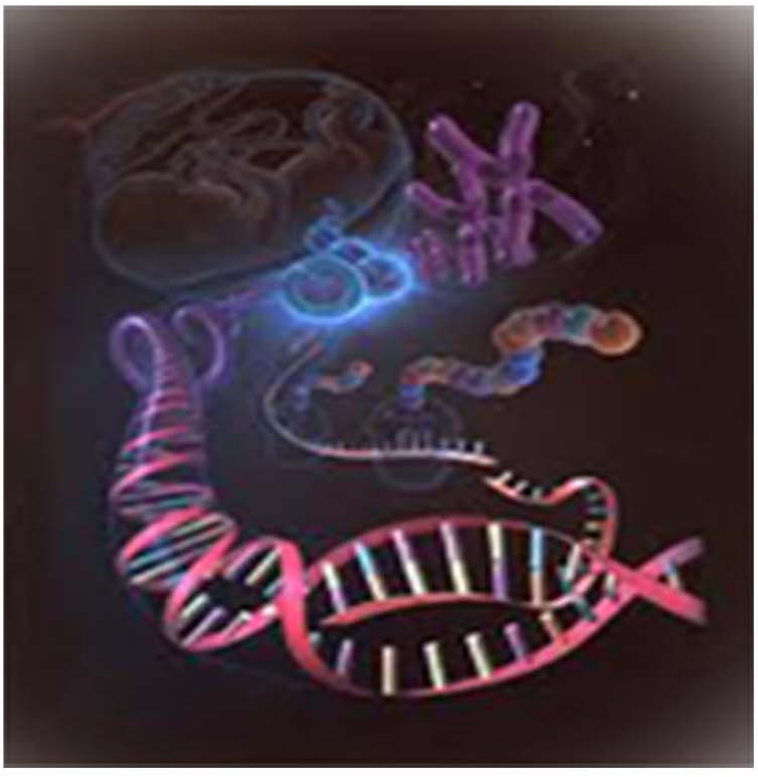

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Research Paper

EFFECT OF INDIVIDUAL AND COMBINATION OF HERBAL EXTRACTS ON GLUCOSE TOLERANCE, IN EUGLYCEMIC RATS

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Objective: To assess the glucose tolerance of alcoholic plant root extract of *Clitoria ternatea* Linn and *Salacia chinensis* Wight, individually and in combination. **Methods**: The 20 wistar rats each weighing around 150g were randomly divided in to 4 groups, each group containing 5 rats. Normal control group was fed with 2g/kg body weight glucose alone and treatment groups were administered with 100mg/kg body weight extract of *Clitoria ternatea* Linn and *Salacia chinensis* Wight separately and in combination, all these administrations were done orally by using oral gastric tube which is followed by the administration of 2 g/kg body weight of sugar. Later blood sugar was measured after every 30min till the end of 90 min duration. The results were summarized by mean +/- standard deviation of blood sugar levels at regular intervals of time, one way Analysis of variance (ANOVA) was used to compare the mean blood sugar level of 4 groups which is followed by Schiff's multiple comparison test. **Results:** Both the drugs have shown increased glucose tolerance, but when they were administered in combination the results have shown increased tolerance of glucose in rats at the end of 90 min when compared with the extracts administered individually. **Conclusion:** The herbs *Clitoria ternatea* Linn and *Salacia chinensis* Wight have the capacity to increase the glucose tolerance but their combined administration is showing relatively a higher degree of tolerance against their administration individually, this effect could be due to cellular level interaction of number of phytochemicals to increase the post prandial glucose tolerance.

Keywords: Herbal extracts, Glucose tolerance, *Clitoria ternatea* Linn, *Salacia chinensis* Wight, Euglycemia, Prediabetic, Streptozotocin.

INTRODUCTION

Diabetes Mellitus is a chronic disorder of carbohydrate, fat, and protein metabolism, due to this condition relative or absolute deficiency of insulin secretory response. Which translates into impaired carbohydrate (glucose) utility resulting into hyperglycemia and glucose intolerance, which represents a heterogeneous group of disorders that have hyperglycemia as a common feature.

It is well known fact that hyperglycemia is causing multiple life threatening complications

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like cardiovascular diseases, renal disease, cerebrovascular disorders, hypertension, neuropathy, etc. in both Insulin Dependent Diabetes Mellitus (IDDM) and Non Insulin Dependent Diabetes Mellitus (NIDDM). Among the number of life threatening problems hyperglycemia in its long course affects the peripheral and the central nervous system. In many regions of the world the disease prevention through the holistic approach is highly appreciated by the principles of Ayurveda where it implies equally on the prevention and cure of number of prevailing ailments (Etuk, 2012; Pulok *et al*., 2011).

Ethnobotony is a field which deals with the scientific study of relationship that exists between the people and plant where the hundreds of herbs have been used since centuries for the human benefit (Sharma *et al*., 2010). The knowledge of utility of herbs were realized through constant practice, timely improved human civilization has started their one way of documentation regarding the understanding the disease etiology, pathology, treatment and prognosis in a rational manner, later which are mentioned in the classical literature of Ayurvedic medicine.

The experimental trials generally recommend to start with simple tests which should provide healthy reproducible, affordable, applicable results. The phytochemicals are recommended to start with crude extracts given orally or intraperitoneally (Verspohl , 2002). The herb shankapushpi (*Clitoria ternatea* Linn) belongs to Group of Madhya rasayana (brain tonic) drugs which were used to treat number of cognitive functional disorders as mentioned in the Texts of Ayurveda. This herb was called by different names like Shankapushpi, Aparagita, etc. Its flower resembles the conch shell hence the name shankapushpi.

It grows through out India in hedges and thick forest and also cultivated in gardens, this is a normal perennial twinning herb which has terete stems and branches, leaves are compound, impairipinnate, leaflets are 5-7 in number, available in blue and white flowered variety. According to the scholars of Ayurvedic treatises like Bhavaprakasha, Dhanvantari nigantu (dictionary) the roots, leaves and seeds are having pharmacological utility, particularly the roots carry medicinal value as an intellect promoting ophthalmic, laxative, aphrodisiac, and as tonic used in vitiated conditions of body. Other different parts of shankapushpi plant was used to prove its properties like antihyperglycemic, cognitive function supporter, antidiarrhoeal, etc. (Upwat *et al*., 2010). There are number of preparations mentioned in the classical texts containing the herb shankapushpi as an important ingredient (Kirtikar and Basu, 1999).

The Saptha chakra (*Salacia chinensis* Wight) it is a woody shrub with blackish branches; stem is pale yellow colored with yellowish flowers, it is found in Southern Orissa, Kerala, Malabar, and regions from Mumbai to Coorg , coastal areas and near the river banks and forests. It is also called by different names like Swarna moola, Saptharangi, etc. Classical texts like Sahasrayoga, Yogamrutam and Chikitsa manjari have quoted extensively about the use of this herb in the management of diabetes. Its root bark is light brownish colored and on the cross section the root shows seven circles, hence the name 'Sapta Chakra" (Sastry, 2005).

As per the modern botanical classification the herb shankapushpi is identified as *Clitoria ternatea* Linn. belongs to the family Fabaceae. It contains compounds like kaempferol-3-O-rhamnosyl,1-6 glycoside and kaempferol-3-O-

rhamnosyl,1-6 galactoside, kaempferol 3-O-glucoside, kamempferol-3-O-rutionoside, kaempferol-3-O neohesperidoside and clitorin. As per the modern botanical classification the herb *Salacia chinensis* Wight, belongs to family Celastraceae, contains compounds like triterpenes, Q,T,U assigned as friedelan-1,3,dione bearing 24-al, and 24-ol and 24 oic groups respectively (Rastogi and Mehrotra, 1980-1984).

MATERIALS AND METHODS

Healthy 20 wistar rats each weighing around 150g of either sex were procured as per the resolution code JNMC/Institutional animal ethical committee (IAEC) 2/1/2008 and divided them randomly into 4 groups, each group containing 5 rats (Xang and Tan, 2000).

The well grown plant *Clitoria ternatea* Linn (white flowered) roots were collected from the college campus garden of J. N. Medical College, Belgaum and the well grown *Salacia chinensis* Wight roots were collected from Western Ghats forest near Sirsi taluk of Uttarakannada district, Karnataka state, India. Both the plants were authenticated by the scientist and Botanist Dr. Harsha Hegde, from Indian Council of Medical Research (ICMR) Regional Centre Belgaum, India. *Clitoria ternatea* Linn and *Salacia chinensis* Wight roots were separately shade dried and cut into small pieces, made into coarse powder by putting into pulverizer, by using the soxhlet extraction procedure. First the powder was defatted by using the petroleum ether; later the same powder was used for extraction with absolute alcohol. The extract was dried by using the hot water evaporation bath and later it was collected in a clean container covered with lid.

Grouping and Treatment

The 20 rats each weighing 150 g were randomly selected under 4 groups, each group was containing 5 rats. After the confirmation of their Fasting Blood Sugar in the overnight fasted rats, the Group I normal control (NC) fed with glucose alone, simultaneously the remaining groups II, III, IV were fed with herbal extracts individually and in combination which is followed by the administration of glucose 2 gm/kg body weight at the end of the 30 min. In all the groups these drugs were administered once which is followed by monitoring the glucose levels at regular intervals.

Group II was fed with 100mg/kg body weight of alcoholic Extract of *Clitoria ternatea* Linn alone (Lawlor, 1997). Group III was fed with 100mg/kg body weight of alcoholic extract of *Salacia chinensis* Wight alone, Group IV was fed with the 100mg/kg body weight of each above mentioned both the herbal extracts in combination (Rai *et al*., 2001; and Sahayam *et al*., 2011).

Observations

The blood sugar was checked regularly at 30 min of interval, up to 90 min of duration by using Glucometer device Optium Xceed belongs of Abbott diabetes care Ltd. (Kumar *et al*., 2007).

Statistical Analysis

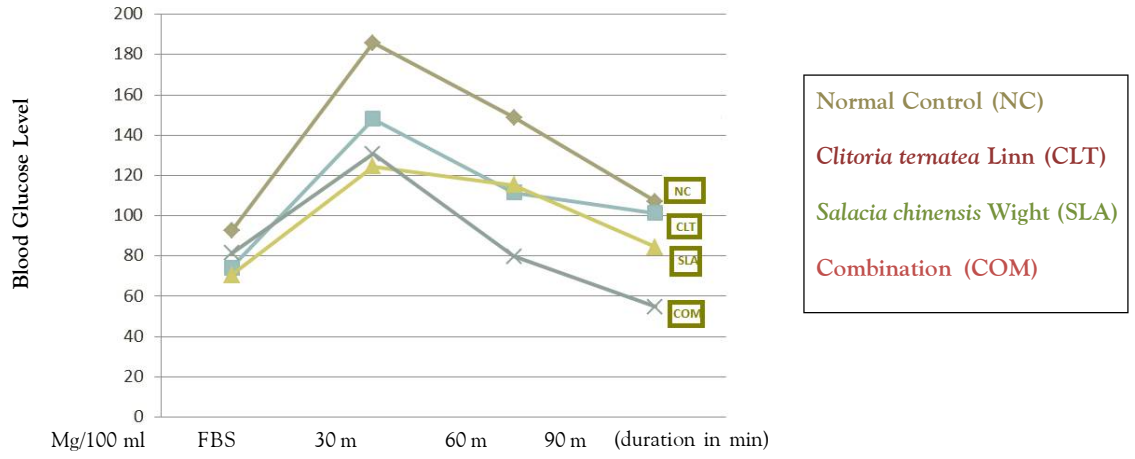
The results were summarized by mean ± standard deviation of blood sugar levels at different intervals of time, one way ANOVA was used to compare the mean blood sugar level of 4 groups (Table 1). For multiple comparison of mean deference, Schiff's test was used $P<0.05$ is considered to be statistically significant.

There was significant difference in the mean FBS of all the 4 groups, the difference was found in mean FBS of all the 4 groups, a significant difference was found between group 1 with group 2 and 3, other groups did not differ significantly. At first 30 min interval there were significant differences in the mean

Table 1: Blood Sugar Level in mg/100 ml (Mean ± SD)

Groups	FBS	At 30 min	At 30 min	At 90 min
	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD
1	92.6 ± 6.58	185.8 ± 9.57	148.8 ± 18.83	107 ± 12.04
2	74.2 ± 6.68	148 ± 26.3	111.4 ± 26.29	101.4 ± 13.27
3	70.6 ± 5.41	124.4 ± 15.34	115.2 ± 14.014	84.4 ± 16.89
4	81.4 ± 4.39	130.6 ± 24.35	79.8 ± 16.57	54.8 ± 14.66
F3,16	13.780	7.547	8.393	13.393
P	< .001	<0.002	<.001	<.001

Figure 1: Blood Sugar Levels at Regular Intervals of 30 min Duration in all Experimental Groups



blood sugar levels of all the 4 groups and the significant difference in the mean value of group 1 with group 3 and 4 were found.

There was significant difference in mean sugar level at 60 min of time with the group 1, which differed significantly with group 4 but others did not differ much. At the 90 min, group 4 differed significantly with group 1, 2, 3 but others did not differ much. In summary the major difference was found between the groups 4 with other 1, 2, 3 groups in this study.

DISCUSSION

The change in quality and quantity of food habits of present generation has drastically changed when compared to past. Sedentary work and lack of physical exercise is adversely influenced by our food habits. The routine use of adulterated, modernized, highly refined food articles are becoming the strong reasons for number of ailments. Consumption of inevitable excess of carbohydrates are leading to excess of circulating blood sugar load on body in its long run showing

its impact on the endocrine part of pancreatic cells leading to gradual inefficiency in handling the glucose load in the body by influencing Prediabetic changes in the body. Childhood obesity is also becoming an important precipitating factor for diabetes in young individuals and the same in adults can lead to number of ailments from simple neuropathy to nephropathy. It has been debated during the last decades whether asymptomatic, unrecognized diabetes or even a lesser degree of hyperglycemia in random samples indicating the increased risk of Cardiovascular Disease (CVD) and death; however investigator who studied the association between hyperglycemia and the development of diabetic complications focus on fasting glucose levels (Tuomilehto, 2002). In diabetic studies it is generally recommended to start with simple tests which should provide highly effective reproducible results and it should not be much time consuming. The euglycemic animals are used for testing potential oral hypoglycemic agents; this is still a valid screening method which is often used in addition to diabetic animal models (Verspohl, 2002). Large interventional studies have shown that achieving and maintaining near normal glycemic levels can reduce the risk of micro vascular and macro vascular complications in type 2 diabetes. The impact of post prandial blood sugar control is equally important to prevent its long course complications (Parkin and Neil Brooks, 2002). The strict control over the blood sugar level possibly by changing our life style and food habits, along with the rational use of cost effective and easily available herbal supplement is the essential need of time. The WHO (world health organization) Expert committee recommended and also high lightened the investigation of antihyperglycemic agents from plant origin which are used in traditional medicine for the treatment of diabetes mellitus. The numbers of anti hyperglycemic agents from the plant origin are used in traditional medicine are showing their beneficial effects. In this experimental study on glucose tolerance particularly we have chosen the drugs called saptharangi (*Salacia chinensis* Wight) which is used in the diabetes as an important antihyperglycemic agent, and another drug shankapushpi (*Clitoria ternatea* Linn) which belong to Medhya rasayana, to support the brain cells by counteracting the adverse impact of glucose.

The above mentioned plant extracts used in combination have shown much greater reduction in the postprandial blood sugar at the end of 90 min of time when it was compared with individually administered experimental groups (Kumar *et al*., 2007). This effect could be due to molecular level combined interaction of different phytochemicals present in our two different herbal different extracts. Herbs are well known, human friendly which are easily available and cost effective source to treat number of disorders with respect to their effective prevention and cure. Even today 60% of world populations rely upon their traditional medicine. The world's oldest medical sciences like Ayurveda and traditional Chinese medicine have mentioned number of herbs and herbal formulations to tackle this hyperglycemic condition *Salacia chinensis* Wight is one among them. The experimental drug trials with the isolated compounds like Mangiferin from the *Salacia chinensis* Wight in diabetic rats have shown significant decrease in blood sugar levels along with increased insulin content in the body, which support the rationality of its ethnobotonical usage (Sellamurthy *et al*., 2009; Melanie-Howes

and Peter J Houghton, 2003). The gross Study trials by using the aqueous extracts of *Clitoria ternatea* Linn leaf and flower in diabetic rats are also significantly decreased the levels of blood glucose and glycosylated hemoglobin. At the same time there was an increased serum insulin levels to nearly matching with normal control groups, which is suggestive of its pancreatic effect (Daisy and Rajathi, 2009). The probable action of herbs may be through their stimulatory effect on the endocrine part of pancreas which is manifested as increased glucose tolerance (Eliakim-Ikechukwu and Obri, 2009). Our main interest of this study is to assess how best these herbal extracts individually and in combination can effect the glucose tolerance. Our study is exclusively based on its ethnobotonical utility where the people in the past were dependent only on easily available, cheap and effective source available around them for number of ailments. This indicating that the conventional use of crude herbs in diabetes contains some effective and essential phytochemical constituents, which are need to be identified, isolated and trialed in advanced research set up to prove their mechanism of anti hyperglycemia. The Ayurvedic principles consider whole human body, mind and spirit along with therapeutic preventive or curative treatments. While dealing with the maintenance of health in healthy individual and treating the disease in a holistic way is well acceptable in the present generation.

Hyperglycemia however is not a simple medical issue, the blood glucose level tend to show diurnal and seasonal variations. Thus we have firm evidence that rapid load of post prandial glucose can be delayed. In the current era of evidence-based medicine this knowledge should be the strongest argument to test the glucose intolerance in the people especially those who tend to face the risk in future.

Here we have chosen the whole root extracts, their mechanism of action can be understood by further trials by isolating their phytochemical constituents. Irrespective of any predisposing factors for glucose intolerance, early adoption practice of herbal supplements as a part of our lifestyle accompanied with good diet and exercise certainly can reduce the long term consequences on endocrine part of pancreas by preventing the early exhaustion of islets, which is an ultimate change taking place in the chronic diabetics forcing the individual to become more dependent on the insulin therapy.

In western medicine we have a clear indication of drug action site in the body, but the utility of crude extract which is a combination of many known and unknown compounds which may influence the antihyperglycemic effect in a complex pattern. The herbs may act by reducing the glucose production from the liver tissue by preventing the glycogenolysis or it may prevent the glucose absorption from the intestines so that the post prandial glucose load can be substantially reduced or it may act as stimulant for the production of insulin secretion by supporting the tissue recovery changes in the pancreatic islet cells or it may help many insulin dependent tissues like skeletal muscles to utilize the glucose in a effective manner. To prove the antihyperglycemic effects which needs further trials to prove an effective natural alternate therapy.

COMPETING INTERESTS

Author declares that he do not have any competing interest.

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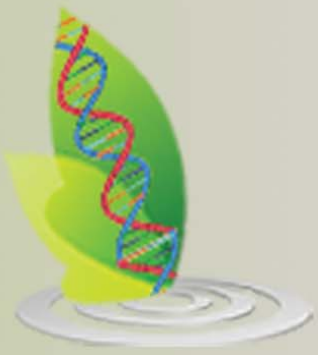

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