IDENTIFICATION OF ANTI-DIABETIC ACTIVITY OF STRYCHNOUS NUXVOMICA ROOTS IN INVIVO CONDITIONS

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ABSTRACT

Strychnous nuxvomica is a medicinal plant with many chemical constituents like strychnine, brucine, indole alkaloids etc. This study was design to investigate the effect of aqueous methanol extract of nuxvomica roots, on various biological parameters in diabetes induced rabbits. The effect of crude nuxvomica root and its organic extracts on blood glucose level of rabbits has been studied. It was found that methanol extract had some hypoglycemic effect on the induced hyperglycemia in the experimental animals.

KEYWORDS: Strychnous nuxvomica, Methanol extract, Diabetes, Auto analyzer

INTRODUCTION

The therapy of diabetes has changed markedly in the past 20-25years. 2 million cases of diabetes diagnosed approximately 22% are controlled by diet alone, about 33% by insulin and about 45% by oral hypoglycemic agents (Burger,1970).Except for insulin, none of the current therapeutic agents was available in 1957.

DIABETES MELLITUS: It is a group of metabolic diseases in which a person has high blood sugar, either because the body doesn't produce enough insulin, or because cells do not respond to the insulin that is produced. This high blood sugar produces the classical symptoms of polyuria, polydipsia and polyphagia.

Types of Diabetes:

1. **Type 1- Diabetes:** Results from the body's failure to produce insulin, and presently requires the person to inject insulin, also referred to as insulin-dependent diabetes mellitus, IDDM for short and juvenile diabetes.
2. **Type 2-Diabetes:** Results from insulin resistance, a condition in which cells fails to use insulin properly, sometimes combined with an absolute insulin deficiency. (Referred as non-insulin dependent diabetes mellitus).

3. **Gestational Diabetes:** when pregnant women, who have never had diabetes before, have as high blood glucose level during pregnancy. It may precede development of type 2 DM.

Other forms of diabetes mellitus include congenital diabetes, which is due to genetic defects of insulin secretion, cystic fibrosis-related diabetes, steroid diabetes induced by high doses of gluco -corticoids, and several forms of monogenic diabetes

Strychnous nuxvomica is a medium sized tree with a short, crooked, thick trunk, the wood is white hard, close grained, durable and the root very bitter. The seeds have the shape of flattened disks densely covered with closely appressed satiny hairs, radiating from the center of the flattened sides and giving to the seeds of characteristic sheen, very hard. Nuxvomica contains the alkaloids, strychnine and brucine, traces of strychnine, glucosideloganin, about 3% fatty matter, caffeotannic acid and trace of copper.

Strychnine is a highly toxic (LD_{50} = c. 16 mg/kg in rats, 1–2 mg/kg orally in humans), colorless crystalline alkaloid used as a pesticide, particularly for killing small vertebrates such as birds and rodents. Strychnine causes muscular convulsions and eventually death through asphyxia or sheer exhaustion. Most common source is from the seeds of the Strychnous nuxvomica tree.

Strychnine was the first alkaloid to be identified in plants of the genus Strychnous, Family Loganiaceae. Strychnous, named by Carl Linnaeus in 1753, genus of trees and climbing shrubs of the gentian order. The genus contains 196 various species and is distributed throughout the warm regions of Asia (58 species), America (64 species), and Africa (75 species). The seeds and bark of many plants in this genus contain the powerful poison strychnine.

Strychnine was first discovered by French chemists Joseph Bienaimé Caventou and Pierre-Joseph Pelletier in 1818 in the Saint-Ignatius' bean. In some Strychnous plants a 9, 10-dimethoxy derivative of strychnine, the alkaloid Brucine, is also present. Brucine is not as poisonous as Strychnine. Historic records indicate that the strychnine alkaloid had been used to

kill dogs, cats, and birds in Europe as far back as 1640. The structure of strychnine was first determined in 1946 by Sir Robert Robinson and in 1954 this alkaloid was synthesized in a laboratory by Robert B. Woodward. This is one of the most famous syntheses in the history of organic chemistry. Both chemists won the Nobel Prize (Robinson in 1947 and Woodward in 1965.

Strychnine poisoning in animals occurs usually from ingestion of baits designed for use against gophers, moles and coyotes. Since 1990 in the United States most baits containing strychnine have been replaced with zinc-phosphide baits. In the Netherlands rodenticides with strychnine are forbidden. Strychnine toxicity in rats is dependent on sex. It is more toxic to females than to males when administered via subcutaneous injection or intraperitoneal injection. Differences are due to higher rates of metabolism by male rat liver microsomes. Dogs and Cats are more susceptible among the domestic animals, pigs are believed to be as susceptible as dogs, and horses are able to tolerate relatively large amounts of strychnine. Birds affected by strychnine poisoning exhibit wing droop, salivation, tremors, muscle tenseness and convulsions. Death occurs as a result of respiratory arrest. The clinical signs of strychnine poisoning relate to its effects on the central nervous system. The first clinical signs of poisoning include nervousness, restlessness, twitching of the muscles, and stiffness of the neck. As the poisoning progresses, the muscular twitching becomes more pronounced and convulsions suddenly appear in all the skeletal muscles. The limbs are extended and the neck is curved to opisthotonus. The pupils are widely dilated. As death approaches, the convulsions follow one another with increased rapidity, severity, and duration. Death results from asphyxia due to prolonged paralysis of the respiratory muscles. Following the ingestion of strychnine, symptoms of poisoning usually appear within 15 to 60 min. The LD_{50}values for strychnine in animals are listed as shown in the table below.

Organism	Route	LD_{50} (mg/kg)
Mouse	Intraperitoneal	0.98
Mouse	Intravenous	0.41
Mouse	Oral	2.0
Mouse	Parenteral	1.06
Mouse	Subcutaneous	0.47
Rabbit	Intravenous	0.4
Rabbit	Oral	0.6
Rat	Oral	16.0
Rat	Oral	2.35

Table: 01The LD_{50} values for strychnine drug dose in different animals

EXPERIMENTAL

2.1 COLLECTION OF PODS:

Strychnous nuxvomica roots collected from nuxvomica plant situated in Nallamalla forest in Andhra Pradesh .The roots were shade dried and powdered for extraction purpose.

2.2 PREPARATION OF EXTRACT:

Aqueous methanolic extract of nuxvomica roots is prepared by using soxhlet apparatus for 8 hours by using the mixture of Methanol. Then the extract was dried with the help of Rotavapor.

2.3 SELECTION OF ANIMAL:

Male New Zealand type, white color with red eyes which is weighed about 2-2.8kg was used. None of the rabbits had any clinically evident infections.

All animals are housed at the animal house in stainless steel case under standard laboratory condition (12 hrs light and 12 hrs dark. 21±2˚C, and relative humidity 55%, green fodder and water availability). The animals received human care.

2.4 EXPERIMENTAL PROCEDURE:

1) Six healthy rabbits were selected in each group and made to fast overnight for 12 hours.
2) Then the Methanol extract of nuxvomica root of 0.6mg/Kg [5] quantity was given according to the weight of each rabbit by oral root in in-vivo conditions.
3) Experiment was conducted for a time period of 24 hours.
4) Then blood samples were collected from marginal-ear vein of each rabbit in the veils. (Given in Table: 02)
5) Then each veil was subject to centrifuge for about 5 – 10 minutes at 3000rpm thereby the blood and plasma was separated.
6) Plasma Glucose levels were found out by using Auto analyzer machine

FIG : 2 CENTRIFUGE MACHINE CLINICAL

FIG 3: AUTO ANALYZER (COBAS U 411)

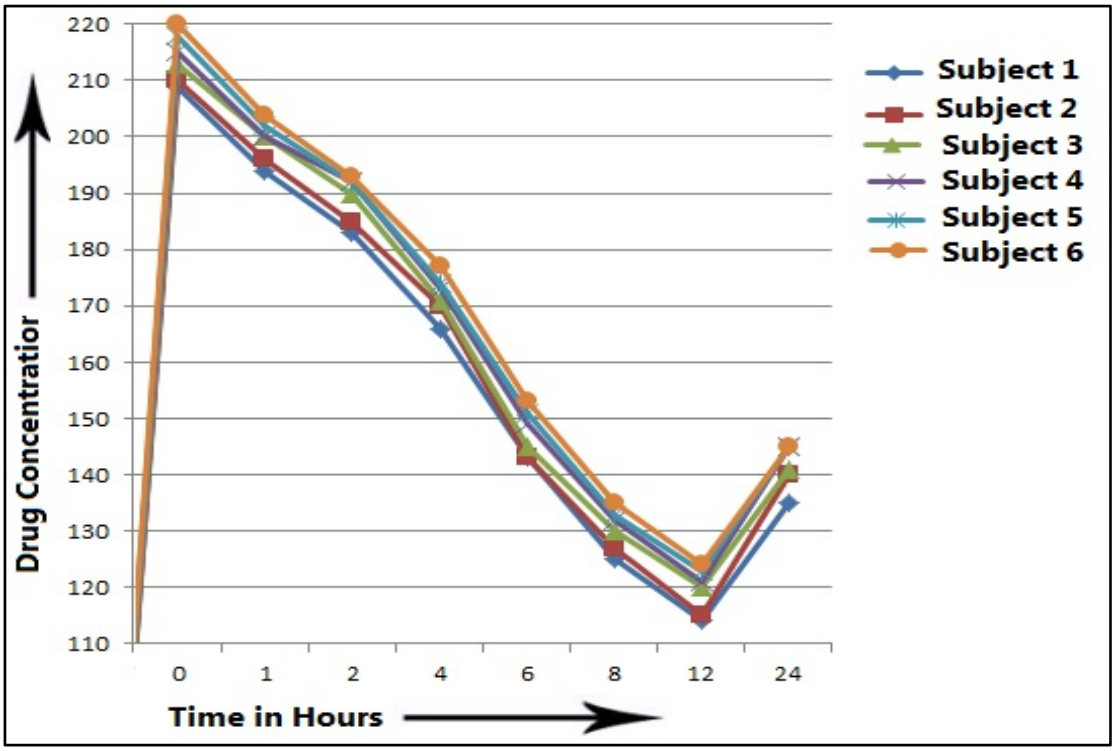
RESULT AND DISCUSSION

Table – 2

S.NO	Time In hours	Amount of Glucose present in mg/dl					
		I	II	III	IV	V	VI
1	0	209	210	213	215	218	220
2	1	194	196	200	200	202	204
3	2	183	185	190	192	192	193
4	4	166	170	171	173	174	177
5	6	143	143	145	149	151	153
6	8	125	127	130	132	133	135
7	12	114	115	120	121	123	124
8	24	135	140	141	145	145	145

Amount of Glucose levels found in different Rabbits (Treated)

Graph: 01



Amount of Glucose levels found in different Rabbits (Treated)

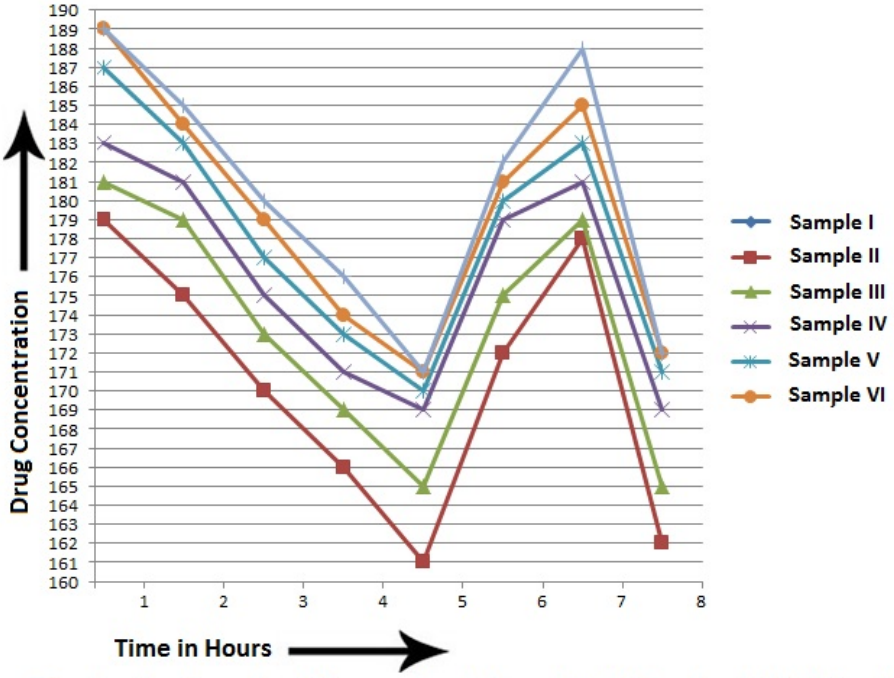
Standard:

Table – 3

S.NO	Time In hours	Amount of Glucose present in mg/dl					
		I	II	III	IV	V	VI
1	0	179	181	183	187	189	189
2	1	175	179	181	183	184	185
3	2	170	173	175	177	179	180
4	4	166	169	171	173	174	176
5	6	161	165	169	170	171	171
6	8	172	175	179	180	181	182
7	12	178	179	181	183	185	188
8	24	162	165	169	171	172	172

Amount of Glucose levels found in different Rabbits (Standard)

Graph: 2

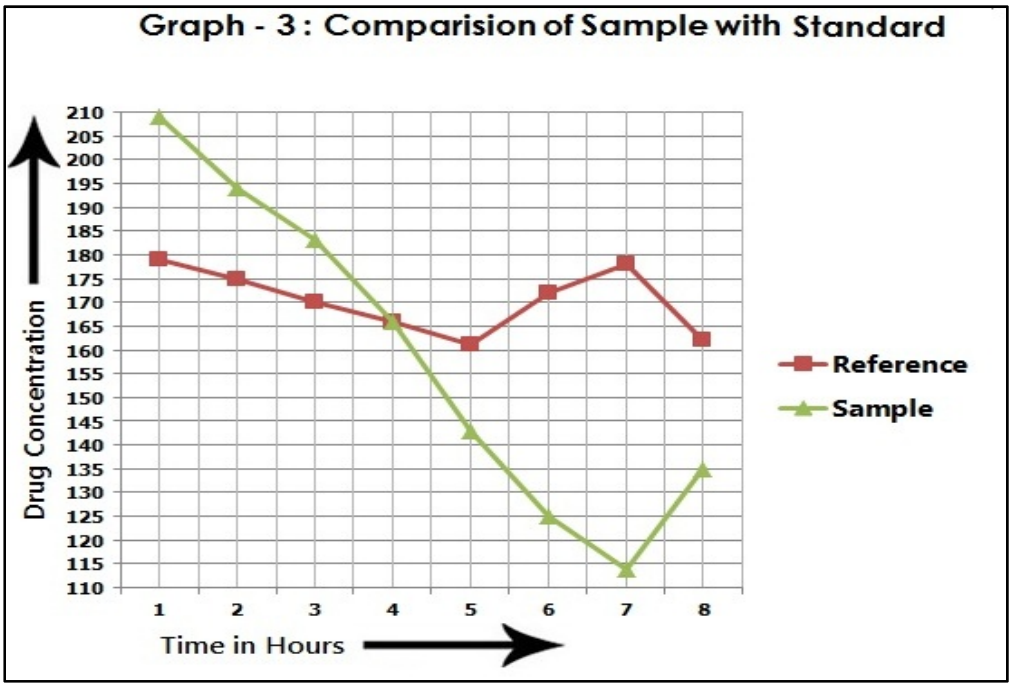


Graph – 2 : Amount of Glucose levels found in different rabbits (Standard)

Table – 4

S.No	Time in hours	Reference	Sample
1	0	179	209
2	1	175	194
3	2	170	183
4	4	166	166
5	6	161	143
6	8	172	125
7	12	178	114
8	24	162	135

Comparison of sample with standard



This study was aimed to find out the Anti-diabetic effect of Strychnous nuxvomica root. The hypoglycemic activity of methanol extract of nuxvomica root was determined in two groups of rabbits.

For the evaluation of hypoglycemic activity, glucose tolerance tests were performed. The difference in the tests with and without the administration of methanol extract nuxvomica root can easily be compared by plotting the respective tolerance curves.

CONCLUSION

According to this study the results showed that methanol extract of nuxvomica root contain some constituents which can be helpful in lowering the blood glucose level. Based on the complete study and results, it was proved that root of Strychnous nuxvomica plants have Anti diabetic activity

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REFERENCES:

1) S.Tripathi, Y. Emmanuel Prakash, KatragaddaAneela, Golusu. Balakrishna, DevathiSumanKumar, MusunuriRavikumar identification of antidiabatic activity strychnosnux-vomica roots, 2011, international journal of science innovations and discoveries, volume 1, issue3.
2) INCHEM: Chemical Safety Information from Intergovernmental Organizations: Strychnine. http://www.inchem.org/documents/pims/chemical/pim507.htm
3) Pelletier and Caventou (1818) “Note sur un nouvelalkalai” (Note on a new alkali), Annales de Chimieet de Physique, vol. 8, pages 323-324. See also: Pelletier and Caventou (1819) “Mémoiresurunnouvelalcali vegetal (la strychnine) trouvédans la feve de Saint-Ignace, la noixvomique, etc.” (Memoir on a new vegetable alkali (strychnine) found in the St. Ignatius bean, the nux vomica, etc), Annales de Chimie et de Physique, vol. 10, pages 142 - 176.
4) Gupta, R.C., Handbook of Toxicology of Chemical Warfare Agents, Elsevier Inc, 2009, Chapter 14
5) RTECS (Registry of Toxic Effects of Chemical Substances) (1935)
6) Longo, V.G.; Silvestrini, B.; Bovet, D. (1959). "An investigation of convulsant properties of the 5,7-diphenyl-1,3-diazadamantan- 6-ol (IS 1757)". J. Pharmacol. Exp. Ther. 126 (1): 41–9. PMID 13642285.http://jpet.aspetjournals.org/content/126/1/41.abstract
7) Setnikar, I; Murmann, W; Magistretti, MJ; Da Re, P (1960). "Amino-methylchromones, brain stem stimulants and pentobarbital antagonists". The Journal of pharmacology and experimental therapeutics 128: 176–81.
8) Haas, H (1960). "On 3-piperidino-1-phenyl-1-bicycloheptenyl-1-propanol (Akineton). 2". Archives internationales de pharmacodynamie et de therapie 128: 204–38..
9) Prasad, CR; Patnaik, GK; Gupta, RC; Anand, N; Dhawan, BN (1981). "Central nervous system stimulant activity of n-(delta 3-chromene-3-carbonyl)-4 iminopyridine (compound 69/224)". Indian journal of experimental biology 19 (11): 1075–6.
10) Zapata-Ortiz, V; CASTRO DE LA MATA R; Barantes-Campos, R (1961). "The anticonvulsive action of cocaine". Arzneimittel-Forschung 11: 657–62. PMID 13787891.
11) Sandberg, F; Kristianson, K (1970). "A comparative study of the convulsant effects of strychnos alkaloids". ActapharmaceuticaSuecica 7 (4): 329–36
12) Spector, W.S. (1956). Handbook of Toxicology, Vol. 1, p. 286. W.B. Saunders Company, Philadelphia.

13) Ward, Justus C.; Crabtree, D. Glen (2006). "Strychnine X. Comparative accuracies of stomach tube and intraperitoneal injection methods of bioassay". Journal of the American Pharmaceutical Association 31 (4): 113–115. doi:10.1002/jps.3030310406.
14) Maqsood Ahmad et al., Antidiabetic and Hypolipidemic effect of aqueous methanolic extract of Acacia Nilotica Pods in Alloxan – induced diabetic rabbits, Scand. J. Lab. Anim. Sci. 2008 Vol 35 No. 1.