Original Article

A review on preclinical studies conducted with Homeopathic medicine Cephalandra indica as an anti-hyperglycemic agent

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Abstract

Background Diabetes Mellitus is a metabolic disorder that affects the secretion of insulin from pancreas leading to hyperglycemia. If uncontrolled leads to complications triggered by free radical formed after oxidative stress. Homeopathic medicine Cephalandra Indica has shown antidiabetic activity in various potencies performed on preclinical studies on diabetic rat model. The present review highlights the pharmacological profile of homeopathic preparations from Cephalandra Indica on preclinical studies and calculating the probable human equivalent dosage from preclinical studies for the pilot studies. Methods Articles published between January 1988 and December 2018 were included in this review. Databases like PubMed – Medline, Google scholar were used for collecting the articles. Keywords like ‘Homeopathy’ or ‘Homoeopathy’, ‘in vitro’, ‘in vivo’ and ‘Cephalandra Indica’ were used. SABEH criteria was implemented for assessing the methodology quality of articles. Results Seven full text articles were included in review which had six in vivo studies and one in vitro study. This review article provided the scientific validation of high diluted homeopathic medicines pharmacological activity of C. indica and probable mechanism of action confirmed through preclinical studies. Conversion of dosage from animal model to human dosage for pilot studies has been hypothetically proposed. Conclusion Homeopathic medicine C. indica has a therapeutic and safety profile with no toxicity observed in preclinical studies. The proposed hypothesis of conversion of dosage needs to be validated for further studies.

Keywords: Cephalandra Indica, diabetes mellitus, homeopathy, preclinical studies, SABEH.

Introduction

Diabetes Mellitus (DM) is a metabolic disorder which leads to hyperglycemia with disproportion of protein, carbohydrate and fat metabolism resulting in depletion of production of insulin from pancreas (defects in insulin secretion), or when insulin lacks sensitivity within body (defect in insulin action), or both (1). Prolonged state of hyperglycemia leads to development of diabetic complications through oxidative stress and accompanied by chronic inflammation which are life threatening (2,3). Oxidative stress causes the generation of free radicals which leads to changes in glucose oxidation, non-enzymatic glycation of proteins etc. Free radicals affects cellular organelles and lipid peroxidation, providing damage to system of the enzyme and forming the major pathogenesis of diabetes and its complications (4).
Alternative therapies like homeopathy have been widely used for the treatment of DM. The market for homeopathic medicine is expected to grow at a Compound Annual Growth Rate (CAGR) of 12.5% (5). Homeopathic drugs are included in Drugs and Cosmetics Act, 1940 in Indian government regulatory agency, so it becomes mandatory to generate therapeutic profile, mechanism of action, toxicity and safety of these drugs included under scheduled ‘Y’ of Drug Controller General of India. Pre-clinical trials therefore play a major role before administering drug in healthy human volunteers for drug proving (6).

Extensive review study has been performed on crude extract of Cephalandra Indica (C. indica) in terms of pharmacognosy, phytochemical constituents and pharmacological properties (7-10). As such there had been no any previous review conducted on homoeopathic medicine C. indica action and its potencies on diabetes.

C. indica commonly known as little gourd belongs to family Cucurbitaceae (11). C. indica has antihyperglycemic profile in Ayurvedic system of medicine (12). Fresh juice from the roots of C. indica is used to control diabetes. Tincture prepared from leaves of C. indica is administered for treatment of gonorrhea. C. indica also has antispasmodic activity and also found to be useful in various skin disorders (13-15). Phytoconstituents majorly found in aerial parts of C. indica are Cephalandrol, Heptacosane, Bsitosterol, Tritriacontane, alkaloids and Cephalandrine (alpha and beta) (16). C. indica fruits constitutes Apo-6- lycopenal, B-carotene, Lycopene, Taraxerol, Taraxerone, and (24R)-24-ethylcholest-5 - en -3b- ol glucoside and Cryptoxanthin (17-19) The present review highlights the pharmacological profile of homeopathic preparations C. indica on preclinical studies and calculating the probable human equivalent dosage (HED) from preclinical studies for the pilot studies.

Materials and methods

Research manuscript published between January 1988 and December 2018 were identified for further review. Search database included manuscript from Pubmed-Medline, Google scholar with keywords 'Homeopathy' or 'Homoeopathy', 'in vitro', 'in vivo' and ‘Cephalandra Indica’. Manuscript selected was in English language which was available in full text. The selection period was restricted to last 30 years as there has been rapid improvement in methodology all through this period. Score for assessment of biological experiment on homoeopathy (SABEH) was designed and modified for assessment of methodological quality which had following criteria: a) Defined Objectives b) Control c) Blinding of measurement of outcomes d) Randomization e) Consistency f) Experiment standardization g) Statistical analysis h) Result (20).

Results

SABEH criteria was designed in order to overcome the quality assessment of a research manuscript. Seven original full text research papers were identified by the above mentioned keywords. Out of which 6 in vivo studies and 1 in vitro study were identified. The details are mentioned in Table 1 and Table 2 represents the data of C. indica dosage administering.

C. indica MT (Mother Tincture), 6C and 30C potency was widely used in rat model experiments. Statistical parameter one way ANOVA and student t-test were found repeatedly in use in research manuscript. C. indica was measuredly tested on antihyperglycemic, antioxidant, advance glycation end products, lipid and cholesterol.
Table 1 – Summary of preclinical studies conducted on homeopathic medicine *C. indica*.

| SR. NO | AUTHORS NAME | SPECIES/CULTURE | POTENCY | VARIABLES | RESULT | CONTROL | STATICALS | REFERENCE | YEAR |
|--------|---------------|-----------------|---------|-----------|--------|---------|-----------|-----------|------|
| 1      | D.P. Rastogi et al. | Albino rats | C. indica MT (25µl/100 gbw) 50 µl/100 gbw 75µl/100 gbw 100 µl/100 gbw | 1. Blood Sugar Level (fasting in mg/dl) 2. Beta cell count per islet area in cross section (mm² x 350) 3.ED 50 - ED100 | In vitro study suggested neuro-endocrinological mechanism of *C. indica* MT in stabilizing in blood sugar level. Anti-hyperglycemic Activity established by *C. indica* MT | - Physiological normal saline (0.9% w/v) - 41% alcohol v/v - Alloxan - Insuline 40unit/ml - Gilbenclamide (Daonil) - Tolbutamide (Rastinon) | Mean ± Standard error mean / students t-test | 21 | 1988 |
| 2      | Sathish Sampath et al. | Wistar strain albino Rats | S. jambolanum MT (20 µl/100 grm) S. jambolanum 6C (20 µl) S. jambolanum 30 (20µL) C. indica MT (20 µl/100 gbw) C. indica 6C (20 µl / gbw) C. indica 30C (20µL/ gbw) | 1. Fasting Blood Glucose 2. Radioumnoassay insulin (IR,Akt,GLUT4) 3. Lipid profile 4. mRNA expression analysis 5. Protein isolation 6. Western blot analysis | S. jambolanum and *C. indica* exhibit antidiabetic effect by favoring glucose uptake and oxidation in skeletal muscles through improvement in said variables. | Plane control Metformin (50 mg/kg) | One way analysis of variance (ANOVA) duncan multiple range test | 22 | 2013 |
| 3      | Arindam Pal et al | Mouse fibroblast 3T3 cells Male Wistar albino rats | C. indica MT (6C, 24X, 12C and 30C) (75 µL /100 g) | - Determination of blood glucose and cholesterol level - Liver and pancreatic biopsy - Glucose uptake assay | The present study clearly indicates a significant antidiabetic effect of *C. indica* and lends support for its usage | - Septozotocine - Placebo control 40% ethanol - Gilbenclamide | Student’s t-test | 23 | 2013 |
| Id | Authors | Species | Potencies | Phenolics Identification Methods | Phenolics Extracted as a Homoeopathic Medicine | Study Design | Statistical Test | Year |
|----|---------|---------|-----------|----------------------------------|-----------------------------------------------|-------------|----------------|------|
| 4  | Subasri Muthuviveganandavel et al | *C. indica* MT | Blood Serum glucose - protein - cholesterol - reduces glutathione - triglyceride - lipid peroxidase - asparates aminotransferase - alanine aminotransferase - alkaline phosphates | This study shows some positive role of *C. indica* (MT) for decreasing blood serum sugar and cholesterol level | Sham Control Placebo Control (40% ethanol) | Student's t-test | 24 | 2014 |
| 5  | Rashmi Santosh Tupe et al | Human Erythrocytes | Albumin Glycation: 1. Determination of glycation markers: Estimation of fructosamine, Estimation of protein carbonyls, Estimation of protein bound sugar 2. Determination of structure modification: Estimation of free thiol, Estimation of free amino group 3. Determination of phytochemical Total phenolic Contains | | Positive control glycated Samples | Mean Value standard deviation ANOVA | 25 | 2015 |
| 6. | Lalit Kishore et al. | Male Wistar rats | *C. indica* (MT 6C and 30C) 2ml/kg | Determination flavonoids Determination of glycosides Cellular Study: 1. Erythrocytes with glycated BSA samples 2. Hemolysis Test 3. Intracellular anti-oxidant activity of erythrocytes 1. Body weight, blood glucose and serum insulin estimation 2. Behavioral assessment a. Assessment of thermal hyperalgesia - Tail-immersion (hot water) test - Hot-plate test b. Assessment of mechanical hyperalgesia c. Assessment of allodynia d. Motor nerve conduction velocity. e. Biochemical analysis. - Estimation of superoxide dismutase, - Estimation of reduced Glutathione | homeopathic potencies of *C. indica* have protective effect against diabetic neuropathy (DN) via inhibition of oxidative stress and AGE's | - Streptozotocine - Nikotinamide | Mean ± SEM and one-way Analysis of variance (ANOVA) was used for statistical analysis. ANOVA was followed by Tukey’s as post hoc multiple comparison Test. | 26 | 2017 |
| 1. Nitrite estimation.  
2. Aged estimation in sciatic nerve | C. indica MT  
C. indica 6C  
C. indica 30C | In vitro Antioxidants activity:  
1. Determination of total phenol content.  
2. Total antioxidant capacity  
2-diphenyl-1-picrylhydrazyl scavenging activity:  
1. Hydrogen peroxide scavenging activity  
2. Reducing power assay  
3. Nitric oxide scavenging activity  
4. Superoxide radical scavenging activity  
5. Antiglycation activity  
6. Erythrocyte sorbitol accumulation inhibition  
7. Aldose reductase enzyme inhibition | Defensive effects of homoeopathic preparation of C. indica may suggest its use for the attenuation of oxidative stress and Diabetic complications. | Parameter describe individually on different variables | Mean + standard deviation | 27 | 2018 |

7. Kishore L. et al  
-Male Wistar rats  
- Erythrocytes | C. indica MT  
C. indica 6C  
C. indica 30C | In vitro Antioxidants activity:  
1. Determination of total phenol content.  
2. Total antioxidant capacity  
2-diphenyl-1-picrylhydrazyl scavenging activity:  
1. Hydrogen peroxide scavenging activity  
2. Reducing power assay  
3. Nitric oxide scavenging activity  
4. Superoxide radical scavenging activity  
5. Antiglycation activity  
6. Erythrocyte sorbitol accumulation inhibition  
7. Aldose reductase enzyme inhibition | Defensive effects of homoeopathic preparation of C. indica may suggest its use for the attenuation of oxidative stress and Diabetic complications. | Parameter describe individually on different variables | Mean + standard deviation | 27 | 2018 |
Table 2 – Score for assessment of biological experiment on homeopathy (SABEH) included studies.

| S.NO | AUTHOR | OBJECTIVES (1) | CONTROLS (1) | BLINDING (1) | RANDOMIZATION (1) | CONSISTENCY (1) | EXPERIMENT STANDARDIZATION (1,1) | STATISTICS (1) | RESULT INTERPRETATION (1) | SABEH (9) | REFERENCES |
|------|--------|----------------|--------------|--------------|-------------------|----------------|----------------------------------|----------------|----------------------------|----------|------------|
| 1    | D.P. Rastogi et.al. | 1 | 1 | 0 | 0 | 1 | 1,1 | 1 | 1 | 7 | 21 |
| 2    | Sathish Sampath et.al. | 1 | 1 | 0 | 0 | 1 | 1,1 | 1 | 1 | 7 | 22 |
| 3    | Arindam Pal et.al | 1 | 1 | 0 | 0 | 1 | 1,1 | 1 | 1 | 7 | 23 |
| 4    | Subbari Muthuviveganandavel et.al | 1 | 1 | 0 | 0 | 1 | 0 | 1 | 1 | 5 | 24 |
| 5    | Rashmi Santosh Tupe et.al | 1 | 1 | 0 | 0 | 1 | 1,1 | 1 | 1 | 7 | 25 |
| 6    | Lalit Kishore et.al. | 1 | 1 | 0 | 0 | 1 | 1,1 | 1 | 1 | 7 | 26 |
| 7    | Kishore L. et.al | 1 | 1 | 0 | 0 | 1 | 1,1 | 1 | 1 | 7 | 27 |
Discussion

Homeopathic medicines in these recent times are tested on various preclinical studies. Earlier research conducted on this high dilution medicine showed efficacy in various preclinical studies. Similarly, homeopathic preparation of *C. indica* in its various potencies had been used in preclinical studies for exploring its antioxidant properties with antihyperglycemic activity. The present review highlights the formulation of probable dosage required for testing homeopathic medicine in diabetic animal model studies and provides information on pharmacological activity of homeopathic preparation *C. indica* in its various potencies.

The quality of research manuscript was assessed by SABEH criteria modified through previous conducted systematic review. The SABEH criteria include details (Objectives, Controls, Blinding, Randomization, Consistency, Experiment Standardization, Statistical analysis, Results interpretation.), each representing score of 1, which gives sum of 9. Paper having SABEH score higher than 5 had been included in the review study (20).

According to the study conducted by D.P. Rastogi et al., *C. indica* Mother Tincture when administered in alloxan-induced diabetic rats showed a significant control of blood sugar level within fourteen to twenty days after withdrawal of the drug. Regeneration of pancreatic beta cells was confirmed through histopathological studies which lowered the blood sugar level through the hypothalamo-hypophysial-pancreatic axis, which indirectly release inhibitory factors from hypothalamic neurons, inhibiting the secretion of growth hormone and triggering insulin secretion from beta cells (21).

Sathish Sampath et al. conducted the molecular study identifying the antidiabetic effects of homeopathic preparations of *S. jambolanum* and *C. indica* in their mother tincture, 6C and 30C through skeletal muscle of rats with high fat and fructose-induced type-2 diabetes mellitus. *S. jambolanum* and *C. indica* in their mother tincture, 6C and 30C were tested on parameters like serum insulin and insulin signaling molecules in the skeletal muscle (gastrocnemius), insulin receptor (IR), v-akt murine thymoma viral oncogene homolog (Akt), p-Aktser473 and glucose transporter-4 (GLUT4) protein expression, fasting blood glucose and lipid profile. Homeopathic preparations of *S. jambolanum* and *C. indica*, showed antidiabetic effects in ultramolecular dilutions by improving insulin action via activation of insulin signaling molecules in skeletal muscle of type-2 diabetic rats (22).

The study conducted by Arindam Pal et al. investigated the effects of *C. indica* MT and potencies in streptozotocin (STZ)-induced diabetic Wistar rats. Parameters like body weight, cholesterol level, blood glucose level, and beta-cells of pancreatic islets of Langerhans were measured, with this mouse fibroblast (3T3) cell line were used to study the Glucose uptake mechanism. *C. indica* in MT and various potency showed regeneration of beta-cells in the pancreas which further significantly reduced the blood glucose level with regain of body weight by reducing uptake of glucose confirmed in 3T3 cell line (23).

According to the study conducted by Subasri Muthuviveganandavel et al., *C. indica* MT was studied on parameters like cholesterol, protein, glucose and triglycerides via blood serum of male albino rat. *C. indica* MT showed low-dose acute effect on the above mentioned parameters (24).

Rashmi Santosh Tupe et.al had worked on *S. jambolanum* and *C. indica* (MT, 30C, 200C) respectively in evaluating cellular protection ability in human erythrocytes via glycation induced structural modifications in vitro. Parameters of glycation (carbonyls, bound sugar, fructosamines), structural
modifications (thiol group and free amino) were tested. Phytochemical characterization (total phenolic, flavonoids and glycosides contents) was also performed. The homeopathic preparations especially of *S. jambolanum* in human erythrocyte in vitro prevented glycation induced albumin modifications and subsequent toxicity (25).

Lalit Kishore et al. had conducted research on DN in Wistar rats via streptozotocin induced experimental diabetes model which evaluated the effect of homeopathic preparation of *C. indica* (family Cucurbitaceae) MT, 6C and 30C potencies. Parameters for DN were studied through thermal (tail immersion and hot plate method) and mechanical hyperalgesia (randell-sellito analgesiometer test), motor nerve conduction velocity (MNCV), alldynia (von frey hair test) and oxidative-nitrosative stress. Tissue antioxidant enzymes (TBARS, GSH, and SOD) levels were also measured to assess the oxidative stress with levels of advanced glycation end products (AGEs) in sciatic nerve along with nitrite estimation. *C. indica* significantly reduced oxidative stress and AGESs level in sciatic nerve with protective effect against DN (26).

Another study conducted by Lalit Kishore et al. was designed to evaluate action of *C. indica* (MT, 6C and 30C) against oxidative stress. The antioxidant activity was confirmed via 2,2-diphenyl-1-picrylhydrazyl radical, hydrogen peroxide, nitric oxide and superoxide radical scavenging activity. Phyto-constituents like total phenol content, gallic acid, ascorbic acid were measured respectively within potencies. Homoeopathic preparations of *C. indica* (MT, 6C and 30C) showed protective effect against oxidative stress (27).

**Future perspective** – (Dose escalation protocol, conversion of animal model to human dosage)

Homeopathic system is purely on the concept of drug proving performed on healthy volunteers. Drug action of homeopathic medicine is interpreted in the form of subjective analysis of perception felt by volunteer. The dosage parameter used in proving is still an empirical source which needs to be standardized for safety of human consumption. Although the dilutions of homeopathic medicines are above the Avogadro’s equation which ensure a less chance of observing adverse event as compared to those of conventional ones. Preclinical studies might be a essential guide for predicting dose of first in human or initial phase studies or in drug proving trial, which might be used as standard protocol on the basis of NOAEL. Conversion model bases on body surface area and inter species relation through conversion factor equation. The above mentioned studies performed with *C. indica* confirm no toxicity after administration of MT, potency when compared with standard control and vehicle control. Further studies with parenteral route of administration might be a new outlook for administration of homeopathic medicines and comparing its efficacy with the oral intake in animal model experiments of *C. indica*.

**Conclusion**

Homeopathic medicine *C. indica* has a therapeutic and safety profile with no toxicity observed in preclinical studies. The proposed hypothesis of conversion of dosage needs to validate in first phase of diabetic trial.

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Conflict of Interest

Authors Declares no any conflict of interest.

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