EVALUATION OF INVITRO ANTI-DIABETIC ACTIVITY OF AREVA LANATA WHOLE PLANT EXTRACT

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ABSTRACT

Objective: The aim of the present study is to evaluate the in vitro anti-diabetic activity of Areva lanata whole plant extract by inhibiting α-amylase and α-glucosidase enzymes.

Methods: Different solvent extracts of Areva lanata were prepared in which the aqueous extract showed high enzyme inhibition when compared to other solvents. In order to prevent oxidative stress-induced diabetes, Areva lanata whole plant extract is used which inhibits α-amylase and α-glucosidase enzymes in a dose dependent manner.

Results: The phytochemical screening for successive solvent extraction of Areva lanata confirmed the presence several bioactive compounds like alkaloids, flavonoids (quercetin), tannins, carotenoids, lycopene, carbohydrates, terpenoids, saponins, cardiac glycosides and phenols. And it showed a significant anti-hyperglycemic activity.

Conclusion: whole plant extract of Areva lanata contains dose dependent anti-hyperglycemic activity.

Key words: Diabetes, Areva lanata, Anti-hyperglycemic activity, solvent extraction.

INTRODUCTION

Globally, as of 2014, an estimated 285 million people had diabetes, with type2 making up about 90% of the cases. The increase in incidence in developing countries follows the trend of urbanization and lifestyle changes, perhaps most importantly a western-style diet. Diabetes mellitus occurs when the pancreas doesn’t make enough or many hormone insulin, or when the insulin produced doesn’t work effectively. In diabetes, this causes the level of glucose in the blood to be too high are destroyed, causing severe lack of insulin. As the disease progresses tissue/vascular damage ensures leading to severe diabetic complications such as retinopathy, neuropathy, cardiovascular complications and ulceration [1].

Areva lanata is a plant that contains alkaloids [3], flavonoids [4], steroids, cardiac glycosides, cetosteryl palmitate, carotenoids [5], tannins, glycoside like kaempferol -3- rhamnogalactoside and kaempferol -13- (6-p-coumaryl) -O- glucose along with alkaloids saponins and sugars like sucrose, galactose, fructose and some minerals [2].

The plant extract is used to treat nasal bleeding, cough, scorpion stings, fractures, spermatorrhoea, anti-diabetic, anti-fungal, anti-microbial, antioxidant, nephroprotective, hypolipidemic, cytotoxic effect.

MATERIALS AND METHODS

The chemicals like petroleum ether, ethanol, ethyl acetate, aqueous, chloroform, 3, 5, di nitro salicylic
Acid, sodium acetate buffer, sodium potassium tartarate were used which are of analytical grade.

**Solvent extraction [6]:**

The whole plant of *Areva lanata* was collected and was washed, cleaned, dried, powdered in a grinder-mixer and a coarse powder was obtained which is passed through a 40-mesh sieve. Crude plant extract have been prepared by soxhlet extraction method. About 20gm of powdered plant material was uniformly packed into a thimble and extracted with 250ml of different solvents separately. The air dried coarse powder of the whole plant of *Areva lanata* was extracted successively with organic solvents of increasing polarity like petroleum ether, chloroform, ethyl acetate, and ethanol. The process of extraction continued for 24 hrs. The extract was taken in a beaker and heated at 30-40°C till the solvent gets evaporated. Dried extract is refrigerated at 4°C. The extract was subjected to various qualitative analytical tests like test for carbohydrates, test for alkaloids, flavonoids, proteins, amino acids, steroids, tannins, saponins and triterpinoids.

### RESULTS

**Preliminary phytochemical screening of plant**

The results of phytochemical screening are shown in Table no. 1

**In vitro methods**

**In vitro anti-diabetic activity by alpha-amylase inhibition**

The aqueous, ethanol, ethyl acetate, chloroform and petroleum ether extracts of *A. lanata* were found to possess dose dependent increase in percentage inhibitory activity against alpha amylase enzyme. In present findings, high polar solvent (aqueous & ethanol) extract showed increase in percentage inhibition of amylase. At a concentration of 50µg/ml of different solvent extract with increasing polarity showed a percentage alpha glucosidase enzyme inhibition 8.72, 12.43, 15.84, 23.71, 41.67%. Similarly for 250µg/ml plant extract 25.32, 32.43, 34.32, 64.48 & 73.32% in dose-dependent manner as shown in Table no. 2.

| S.No. | Phytochemicals      | Petroleum ether | Chloroform | Ethyl acetate | Ethanol | aqueous |
|-------|---------------------|-----------------|------------|--------------|---------|---------|
| 1     | Alkaloids           | -               | +          | -            | +       | +       |
| 2     | Flavonoids          | +               | +          | +            | +       | +       |
| 3     | Tannins             | -               | +          | -            | +       | +       |
| 4     | Proteins            | -               | -          | +            | -       | +       |
| 5     | Phenols             | -               | -          | -            | +       | +       |
| 6     | Carbohydrates       | +               | +          | -            | +       | +       |
| 7     | Cardiac glycosides  | +               | -          | +            | -       | +       |
| 8     | Saponins            | -               | -          | -            | +       | +       |
| 9     | Terpenoids          | +               | +          | +            | +       | +       |
| 10    | Phyto sterols       | +               | +          | +            | -       | -       |

+ Present - absent

### Table no. 2: In vitro alpha-amylase enzyme inhibiton

| S.no. | Conc (µg/ml) | Pet. Ether  | Chloroform | Ethyl acetate | Ethanol  | Aqueous  | Standard drug |
|-------|-------------|-------------|------------|--------------|----------|----------|---------------|
| 1     | 50          | 8.72±1.15   | 12.43±1.42 | 15.84±1.96   | 18.34±1.24 | 23.71±1.32 | 41.67±1.76   |
| 2     | 100         | 12.66±1.52  | 17.35±0.77 | 21.36±1.52   | 24.67±2.51 | 36.34±1.48 | 54.32±3.98   |
| 3     | 150         | 18.65±2.51  | 22.17±1.78 | 26.04±1.01   | 35.72±1.59 | 44.76±2.97 | 67.45±3.97   |
| 4     | 200         | 21.34±1.57  | 26.69±0.87 | 31.66±1.16   | 46.12±1.78 | 59.42±1.72 | 80.24±2.08   |
| 5     | 250         | 25.32±1.15  | 32.43±1.82 | 34.32±1.98   | 64.48±2.03 | 73.33±1.67 | 91.18±2.16   |

Values were given as mean ±SD and showed significant results compared to standard drug.
In vitro anti-diabetic activity by alpha-glucosidase inhibition

The aqueous, ethanol, ethyl acetate, chloroform and petroleum ether extracts of *A. lanata* were found to possess dose dependent increase in percentage inhibitory activity against alpha amylase enzyme. In present findings, high polar solvent (aqueous & ethanol) extract showed increase in percentage inhibition of glucosidase. At a concentration of 50µg/ml of different solvent extract with increasing polarity showed a percentage alpha glucosidase enzyme inhibition 10.92, 13.42, 17.67, 20.78 & 24.64%. Similarly for 250µg/ml plant extract 27.36, 34.78, 34.94, 43.42 & 47.34% in dose-dependent manner as shown in Table no. 3.

### Table no. 3: Invitro alpha glucosidase enzyme inhibition

| S.no. | Conc (µg/ml) | Pet. Ether | Chloroform | Ethyl acetate | Ethanol | Aqueous | Standard drug |
|-------|-------------|------------|------------|---------------|---------|---------|---------------|
| 1     | 50          | 10.92 ±1.94 | 13.42± 1.72 | 17.67± 1.47   | 20.78± 1.98 | 24.64± 1.13 | 35.73± 2.32   |
| 2     | 100         | 14.72± 1.75 | 18.37± 0.78 | 23.42± 1.36   | 29.84± 1.67 | 38.57± 2.04 | 40.64± 2.11   |
| 3     | 150         | 20.64 ±1.62 | 24.24± 1.48 | 27.06± 1.84   | 38.35± 1.73 | 40.66± 1.62 | 42.36± 2.23   |
| 4     | 200         | 24.23± 1.65 | 28.52± 1.92 | 33.72± 1.34   | 41.43± 2.43 | 42.23± 1.71 | 49.33± 2.08   |
| 5     | 250         | 27.36± 1.27 | 34.78± 1.52 | 34.94± 2.56   | 43.42± 1.75 | 47.34± 1.91 | 56.34± 2.01   |

Values were given as mean ±SD and showed significant results compared to standard drug.

### DISCUSSION

The presence of several bioactive compounds like alkaloids, flavonoids (quercetin), tannins, carotenoids, lycopene, carbohydrates, terpenoids, saponins, cardiac glycosides, and phenols which could be responsible for the versatile medicinal properties of this plant was extracted. The consequences of oxidative stress can promote the development of complications of diabetes mellitus [8]. Any of these secondary metabolites, singly or in combination with others could be responsible for the anti-diabetic activity of the plant. Different extracts of *Areva lanata* are efficient in inhibiting α-amylase in vitro in dose dependent manner and showed significant anti-hyperglycemic activity. Among the different solvent extracts, the aqueous extract showed high enzyme inhibition than other solvent extracts. Flavonoids [9] from the plant used have high antioxidant property and it decreases oxidative stress induced diabetes. The solvent extracts of the plant efficiently inhibits α-glucosidase and α-amylase enzymes in dose-dependent manner.

### CONCLUSION

The findings from the present study concludes that different solvent extracts with increasing polarity of whole plant of *Areva lanata* do possess significant anti-hyperglycemic property in dose-dependent manner which provides the novel class of therapeutics in the management of diabetes and the results also justify the utility of *Areva lanata* by the local traditional practitioners in the treatment of diabetes however, further pharmacological and toxicological studies at the molecular and clinical levels are still warranted to confirm the true anti diabetic potential of *Areva lanata*.

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