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## EVALUATION OF ANTIDIABETIC AND ANTIOXIDANT ACTIVITY OF LEAF EXTRACT AND SOLVENT FRACTIONS OF VISCUM ARTICULATUM IN MICE

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| Article History                                                                                                                                        | Abstract                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                         |
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| Received: 22-06-2026<br>Revised: 06-07-2026<br>Accepted: 17-08-2026                                                                                    | Flavonoids are widely recognized for their antioxidant properties and have received considerable attention for their potential role in the prevention and management of diabetes mellitus. Their biological activities are strongly influenced by their chemical structures and mechanisms of action. The present study was undertaken to evaluate the antioxidant and antidiabetic potential of the fruit of <i>Viscum articulatum</i> . The antioxidant activity was investigated using dot blot analysis, 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical-scavenging assay, 2,2'-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid) (ABTS) radical-scavenging assay, ferric reducing antioxidant power (FRAP) assay, and xanthine oxidase inhibition (XOI) assay. The antidiabetic activity of aqueous and alcoholic extracts of <i>V. articulatum</i> was evaluated at doses of 20 and 30 mg/kg body weight using an alloxan-induced diabetic rat model. Acute oral toxicity studies demonstrated that the extracts were well tolerated and produced no observable toxic effects at doses up to 2000 mg/kg body weight. Diabetes was induced by a single intraperitoneal administration of alloxan monohydrate following fasting. Fasting blood glucose levels were monitored on the 7th, 14th, and 21st days, while oral glucose tolerance was assessed on the 8th, 15th, and 22nd days. Glibenclamide was used as the standard antidiabetic drug. Both aqueous and alcoholic extracts produced a significant reduction in fasting blood glucose levels and improved oral glucose tolerance in diabetic rats, with the observed effects being comparable to those of glibenclamide. The extracts also demonstrated hypoglycemic activity in normal rats and considerable antioxidant potential. The findings suggest that <i>V. articulatum</i> fruit may exert antidiabetic effects through improvement of glycemic control and possible protection or regeneration of pancreatic insulin-secreting $\beta$ -cells. Overall, the results support the traditional medicinal use of <i>V. articulatum</i> and indicate its potential as a natural source of antioxidant and antidiabetic agents. |
| <b>Keywords:</b> <i>Viscum articulatum</i> ; antidiabetic activity; antioxidant activity; hypoglycemic activity; alloxan-induced diabetes; flavonoids. |                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                  |
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### INTRODUCTION

Medicinal plants have been used for thousands of years in traditional medicine and remain an important source of bioactive compounds and modern therapeutic agents. Advances in extraction techniques, including ultrasound-assisted and supercritical fluid extraction, along with analytical methods such as HPLC and LC/MS, have improved the identification and characterization of phytochemicals [1].

Free radicals are highly reactive molecules containing one or more unpaired electrons. Reactive oxygen species (ROS) and reactive nitrogen species (RNS) include both radical and non-radical oxidants. Important examples include hydroxyl radical, superoxide, nitric oxide, hydrogen peroxide, peroxynitrite and hypochlorous acid. They are generated through enzymatic and non-enzymatic cellular processes, including mitochondrial respiration, phagocytosis, prostaglandin synthesis and cytochrome P450 activity [2].

At low or moderate concentrations, ROS and RNS have beneficial physiological functions. They participate in cellular signaling, immune defense, regulation of blood flow and destruction of invading microorganisms. However, excessive

production causes an imbalance between free-radical formation and antioxidant defense, resulting in **oxidative stress** [3-4].

Oxidative stress can damage lipids, proteins and DNA through processes such as lipid peroxidation and oxidative DNA modification. Consequently, it is associated with several chronic and degenerative conditions, including cancer, cardiovascular and neurological diseases, pulmonary disorders, rheumatoid arthritis, renal disease and ocular disorders. Antioxidants act as free-radical scavengers and help prevent or repair oxidative damage [5-].

Antioxidants are molecules that prevent or slow oxidation and protect cells from damage caused by free radicals. They neutralize free-radical intermediates and help maintain the balance between reactive species and antioxidant defenses. Endogenous antioxidants include enzymes such as superoxide dismutase, catalase and glutathione peroxidase, while important nonenzymatic antioxidants include vitamins C and E, glutathione and melatonin. A deficiency or imbalance in antioxidant defenses has been associated with disorders such

as cancer, inflammation, atherosclerosis, cardiovascular disease and diabetes [6-7].

Several medicinal plants possess antioxidant potential due to phytoconstituents such as polyphenols, flavonoids, alkaloids, tannins and terpenoids. Plants including *Asparagus racemosus*, *Acacia arabica*, *Terminalia chebula*, *Eucalyptus globulus* and *Citrus limon* have demonstrated free-radical scavenging or lipid-peroxidation-inhibitory activities in experimental studies. Diabetes mellitus is a metabolic disorder characterized by hyperglycemia resulting from inadequate insulin secretion, impaired insulin action or both. It is broadly classified into type 1 and type 2 diabetes. Type 1 results primarily from autoimmune destruction of pancreatic  $\beta$ -cells, whereas type 2 involves insulin resistance together with progressive  $\beta$ -cell dysfunction. Common manifestations include polyuria, polydipsia, polyphagia, fatigue, weight changes, blurred vision, infections and delayed wound healing [8-].

Management includes insulin and oral hypoglycemic agents. However, medicinal plants have attracted considerable interest as potential complementary sources of antidiabetic agents. Plants such as *Momordica charantia*, *Trigonella foenum-graecum*, *Gymnema sylvestre*, *Allium sativum* and *Aloe vera* have been investigated for potential hypoglycemic effects [9].

## AIM, OBJECTIVES, NEED FOR THE STUDY AND METHODOLOGY

### Aim and Objectives

The study aims to evaluate the **in-vitro antioxidant activity and in-vivo antidiabetic activity** of methanolic/aqueous and alcoholic extracts of *Viscum articulatum* using suitable experimental models in albino Wistar rats. The major objectives include collection and authentication of the plant material, preparation of extracts, preliminary phytochemical screening, acute oral toxicity evaluation, assessment of antioxidant activity by different *in-vitro* methods, and evaluation of antidiabetic activity in normal and alloxan-induced diabetic rats.

### Need for the Study

Medicinal plants have historically served as important sources of therapeutic agents and continue to provide accessible healthcare, particularly in developing and rural populations. Although *Viscum articulatum* has been traditionally used and has demonstrated several pharmacological properties, the available literature indicates comparatively limited investigation of its antioxidant and antidiabetic potential. Therefore, the present study was undertaken to investigate the antioxidant activity of its aqueous and alcoholic extracts and their antidiabetic effects using experimental animal models.

### Plan of Work

The study involved collection, authentication, shade drying and powdering of *Viscum articulatum*, followed by extraction and preliminary phytochemical evaluation. Acute oral toxicity was assessed according to **OECD guideline 425**, using doses up to 2000 mg/kg. Antioxidant activity was evaluated using **FRAP, metal-chelating, DPPH, superoxide radical-scavenging and hydrogen-peroxide-scavenging assays**. Quercetin and BHT were used as reference antioxidants [10-11].

## Experimental Antidiabetic Study

Aqueous and alcoholic extracts were suspended in water containing 3% Tween-80 and administered orally at **20 and 30 mg/kg**. Glibenclamide served as the standard drug. Hypoglycemic activity and oral glucose tolerance were assessed in normal rats. Diabetes was induced in Wistar rats using **alloxan (120 mg/kg, i.p.)**, and animals with fasting blood glucose above 200 mg/dL were selected for further study. Blood glucose levels were monitored at specified intervals during acute and subacute treatment, and OGTT was performed periodically.

The experimental data were expressed as **mean  $\pm$  SEM** and analyzed using **one-way ANOVA followed by the stated post-hoc t-test comparison**, with statistical significance considered at  $P < 0.05$  and  $P < 0.001$ . Overall, the methodology was designed to determine whether *Viscum articulatum* extracts possess significant antioxidant and glucose-lowering activities and to identify their potential as sources of pharmacologically active phytoconstituents.

## RESULTS AND DISCUSSION

### Phytochemical Screening

Preliminary phytochemical screening of aqueous and alcoholic extracts of *Viscum articulatum* demonstrated the presence of several biologically important phytoconstituents. Alkaloids, gums, flavonoids, reducing sugars and tannins were detected in the aqueous extract, whereas glycosides, flavonoids, reducing sugars and tannins were detected in the alcoholic extract. Steroids and saponins were not detected in either extract. The presence of flavonoids, tannins and other phenolic constituents may contribute to the antioxidant and antidiabetic effects observed in subsequent experiments.

Table 01: Preliminary phytochemical screening of *Viscum articulatum* extracts

| Phytoconstituents | Aqueous extract | Alcoholic extract |
|-------------------|-----------------|-------------------|
| Alkaloids         | +               | -                 |
| Glycosides        | -               | +                 |
| Steroids          | -               | -                 |
| Gums              | +               | -                 |
| Flavonoids        | +               | +                 |
| Saponins          | -               | -                 |
| Reducing sugars   | +               | +                 |
| Tannins           | +               | +                 |

+ = Present; - = Absent.

### Antioxidant Activity

The antioxidant potential of *Viscum articulatum* was evaluated using FRAP, metal-chelating, DPPH, superoxide and hydrogen-peroxide scavenging assays. Since antioxidant activity involves multiple mechanisms, assessment using several complementary models provides a broader evaluation of the activity.

The extracts demonstrated ferric-reducing activity, with the alcoholic extract showing greater activity than the aqueous extract. The FRAP values were  $1.90 \pm 0.026$  and  $2.65 \pm 0.014$  for aqueous and alcoholic extracts, respectively. Although the extracts were less active than quercetin and BHT, the results indicated appreciable electron-donating capacity [12].

In the metal-chelating assay, aqueous and alcoholic extracts demonstrated chelating activities of 22.10% and 32.12%, respectively, at the tested concentration. The alcoholic extract showed comparatively greater activity, although EDTA and reference antioxidants exhibited stronger chelating effects. Both extracts exhibited concentration-dependent DPPH radical-scavenging activity. The aqueous extract showed an IC<sub>50</sub> of 1.95 ± 0.052, while the alcoholic extract showed 2.63 ± 0.000. The findings indicate that *V. articulatum* contains constituents capable of donating hydrogen or electrons to neutralize free radicals [13].

Superoxide radical-scavenging activity was also observed in both extracts. The aqueous and alcoholic extracts showed IC<sub>50</sub> values of 0.36 ± 0.002 and 0.320 ± 0.006, respectively. Similarly, both extracts demonstrated hydrogen-peroxide scavenging activity, with the aqueous extract showing greater activity than the alcoholic extract under the reported experimental conditions.

Table 02: Antioxidant activity of *Viscum articulatum* extracts

| Assay                                    | Standard                 | Aqueous extract | Alcoholic extract |
|------------------------------------------|--------------------------|-----------------|-------------------|
| FRAP                                     | Quercetin: 15.12 ± 0.015 | 1.90 ± 0.026    | 2.65 ± 0.014      |
| Metal chelation                          | EDTA: 7.12               | 22.10           | 32.12             |
| DPPH                                     | Quercetin: 4.10 ± 0.024  | 1.95 ± 0.052    | 2.63 ± 0.000      |
| Superoxide                               | Quercetin: 0.016 ± 0.005 | 0.36 ± 0.002    | 0.320 ± 0.006     |
| H <sub>2</sub> O <sub>2</sub> scavenging | Quercetin: 0.050 ± 0.006 | 0.075 ± 0.010   | 0.552 ± 0.014     |

Values are presented as reported in the source data; units should be interpreted according to the respective assay.

The overall antioxidant findings suggest that *V. articulatum* extracts possess multiple free-radical-scavenging and reducing mechanisms. The activity may be associated with flavonoids, tannins and other phenolic constituents detected during phytochemical screening [14].

## ANTIDIABETIC ACTIVITY

### Acute Toxicity

Acute oral toxicity evaluation indicated that the alcoholic extract of *V. articulatum* was well tolerated up to 2000 mg/kg, with no mortality or apparent toxic behavioral or neurological effects during the observation period. This finding supported the selection of lower doses for subsequent pharmacological evaluation.

### Hypoglycemic Activity in Normal Rats

Repeated administration of aqueous and alcoholic extracts produced reductions in fasting blood glucose levels in normal rats. The effect became more evident with repeated treatment over 7, 14 and 21 days. Glibenclamide produced the expected hypoglycemic effect and served as the reference treatment. Among the extracts, the alcoholic extract at 20 mg/kg produced marked reduction in fasting blood glucose, while the 30 mg/kg alcoholic extract also demonstrated substantial activity. These findings suggest that *V. articulatum* extracts possess hypoglycemic potential [15].

Table 03: Effect of *Viscum articulatum* extracts on fasting blood glucose in normal rats

| Treatment      | Dose (mg/kg) | Day 7        | Day 14       | Day 21       |
|----------------|--------------|--------------|--------------|--------------|
| Normal control | —            | 86.10 ± 1.50 | 76.10 ± 1.10 | 70.12 ± 1.05 |
| Glibenclamide  | 10           | 76.25 ± 2.20 | 72.10 ± 3.56 | 69.25 ± 1.41 |
| AQVA           | 20           | 84.10 ± 2.14 | 82.14 ± 5.10 | 78.38 ± 3.15 |
| AQVA           | 30           | 82.15 ± 3.26 | 75.10 ± 2.12 | 73.15 ± 2.26 |
| ALVA           | 20           | 71.20 ± 1.12 | 65.25 ± 1.52 | 62.19 ± 2.50 |
| ALVA           | 30           | 80.41 ± 2.25 | 76.14 ± 2.91 | 72.14 ± 1.50 |

Values are mean ± SEM, n = 6.

### Oral Glucose Tolerance Test

Both aqueous and alcoholic extracts suppressed the increase in blood glucose following glucose loading in normal rats. The effect was observed during the 0.5–2 h observation period and became more evident following repeated treatment. Glibenclamide produced a comparatively pronounced glucose-lowering response [16].

### Antidiabetic Activity in Alloxan-Induced Diabetic Rats

Administration of alloxan produced marked hyperglycemia, with fasting blood glucose levels increasing to approximately 250–266 mg/dL after induction. Treatment with *V. articulatum* extracts subsequently reduced fasting blood glucose levels during the 21-day treatment period.

The alcoholic extract at 30 mg/kg produced the greatest reduction among the tested plant extracts, with blood glucose decreasing from 212.10 ± 12.05 mg/dL on day 7 to 160.14 ± 12.99 mg/dL on day 21. The alcoholic extract at 20 mg/kg also produced a substantial reduction. Aqueous extracts demonstrated comparatively lower activity under the reported conditions [18].

Table 04: Effect of *Viscum articulatum* extracts on fasting blood glucose in alloxan-induced diabetic rats

| Treatment        | Dose (mg/kg) | Day 7          | Day 14         | Day 21         |
|------------------|--------------|----------------|----------------|----------------|
| Normal control   | —            | 80.27 ± 1.10   | 83.05 ± 2.18   | 84.25 ± 2.75   |
| Diabetic control | 10           | 290.10 ± 13.10 | 272.05 ± 11.06 | 250.12 ± 10.24 |
| Glibenclamide    | 10           | 263.26 ± 10.14 | 244.20 ± 5.01  | 230.36 ± 25.12 |
| AQVA             | 20           | 362.05 ± 10.41 | 351.10 ± 11.60 | 326.52 ± 12.13 |
| AQVA             | 30           | 372.15 ± 19.81 | 360.26 ± 19.25 | 343.14 ± 19.01 |
| ALVA             | 20           | 281.52 ± 15.20 | 266.61 ± 15.81 | 221.25 ± 14.26 |
| ALVA             | 30           | 212.10 ± 12.05 | 190.02 ± 11.32 | 160.14 ± 12.99 |

Values are mean ± SEM, n = 6.

The results indicate that the alcoholic extracts, particularly the 30 mg/kg dose, produced a more pronounced

antihyperglycemic response than the aqueous extracts. The glucose-lowering activity may be associated with the flavonoids, alkaloids, tannins and other secondary metabolites identified during phytochemical screening. These constituents may influence glucose utilization, intestinal glucose absorption, oxidative stress and other pathways involved in glycemic regulation. However, the precise mechanism requires further investigation.

#### Oral Glucose Tolerance in Diabetic Rats

Both extracts demonstrated an ability to suppress the elevation of blood glucose following oral glucose administration in alloxan-induced diabetic rats. The response was observed on days 8, 15 and 22. The alcoholic extract at 20 mg/kg showed a marked reduction by day 22, whereas the aqueous extract at 20 mg/kg also demonstrated progressive improvement.

The present study demonstrates that *Viscum articulatum* possesses appreciable **antioxidant and antidiabetic potential**. Phytochemical screening revealed flavonoids and tannins in both extracts, together with alkaloids, glycosides, gums and reducing sugars. Such phytoconstituents may contribute to the observed pharmacological effects [19].

The antioxidant experiments demonstrated activity through several mechanisms, including electron donation, free-radical scavenging and metal-ion chelation. The concentration-dependent DPPH, superoxide and hydrogen-peroxide scavenging activities indicate the ability of the extracts to neutralize reactive species. Oxidative stress is closely associated with diabetes, and the antioxidant properties of the plant may therefore contribute to its observed glucose-lowering effects [20-22].

In normal rats, repeated administration of the extracts produced reductions in fasting blood glucose and improved glucose tolerance [23]. In alloxan-induced diabetic rats, both extracts demonstrated antihyperglycemic activity, with the alcoholic extract at 30 mg/kg producing the most pronounced reduction among the tested plant extracts [24]. The activity may potentially involve enhanced peripheral glucose utilization, reduced intestinal glucose absorption, modulation of endogenous glucose production or protection against oxidative damage [24]. These mechanisms, however, were not directly investigated in the present study and require confirmation through further mechanistic experiments.

#### CONCLUSION

The present investigation demonstrates that aqueous and alcoholic extracts of *Viscum articulatum* possess significant **in-vitro antioxidant and in-vivo antidiabetic activities** in the experimental models evaluated. The extracts contained several phytoconstituents, particularly flavonoids and tannins, which may contribute to their biological activities. The antioxidant assays demonstrated free-radical-scavenging, reducing and metal-chelating properties, while the animal studies demonstrated hypoglycemic, antihyperglycemic and glucose-tolerance-improving effects.

Among the tested preparations, the **alcoholic extract, particularly at 30 mg/kg, showed a comparatively stronger antidiabetic response** in alloxan-induced diabetic rats. The

findings provide experimental support for the potential pharmacological relevance of *V. articulatum* as a source of antioxidant and antidiabetic phytoconstituents. Nevertheless, further studies involving **standardization, isolation and characterization of active compounds, mechanistic investigations, longer-term toxicity studies and well-designed experimental models** are required before therapeutic application can be considered.

#### AUTHOR CONTRIBUTIONS

Both authors contributed to equally.

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#### CONFLICTS OF INTEREST

The authors declare no conflict of interest.

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